

**Gasoline Blending Streams**  
**Category**  
**Robust Study Summaries**

# Gasoline Blending Streams Category Robust Study Summaries

Petroleum HPV Testing Group  
Consortium Registration #1100997  
March 14, 2014

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# Physical-Chemical SIDS



<b>Melting Point</b>	
<b>Test Substance - Melting Point</b>	
<b>Category Chemical:</b>	No CAS Number Provided
<b>Test Substance:</b>	No CAS Number Provided
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance in Gasoline Blending Streams Category See Category Analysis Document at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Read-Across
<b>Test Substance Result Type:</b>	Estimated
<b>Results - Melting Point</b>	
<b>Melting Indicator:</b>	Melts
<b>Melting Point Value/Range (Temperature):</b>	-138 - 13 °C
<b>Results Remarks:</b>	Melting point values estimated by EPI Suite(TM) for various hydrocarbon constituents of gasoline blending streams having carbon numbers from C4 to C12. Gasoline blending streams typically exist as liquids at ambient temperatures.
<b>Study/Method - Melting Point</b>	
<b>Key Study Sponsor Indicator:</b>	Weight of Evidence
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	MPBPWIN subroutine V1.40 in EPIWIN V 3.10
<b>GLP:</b>	
<b>Study Reference:</b>	REFERENCE: US EPA (2000) Estimation Programs Interface (EPI) Suite(TM). Washington, DC.
<b>Reliability/Data Quality - Melting Point</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	RELIABILITY: Estimated melting points were calculated using a validated computer model.





<b>Boiling Point</b>	
<b>Test Substance - Boiling Point</b>	
<b>Category Chemical:</b>	No CAS Number Provided
<b>Test Substance:</b>	No CAS Number Provided
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Read-Across
<b>Test Substance Result Type:</b>	
<b>Results - Boiling Point</b>	
<b>Boiling Indicator:</b>	
<b>Boiling Point Value/Range (Temperature):</b>	37 - 200 °C
<b>Results Remarks:</b>	Boiling point values from supplemental chemical CAS # 68290-81-5 (Antiknock gasoline; API sample PS-6) not included for read across BP range determination.
<b>Study/Method - Boiling Point</b>	
<b>Key Study Sponsor Indicator:</b>	Weight of Evidence
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	
<b>GLP:</b>	
<b>Study Reference:</b>	
<b>Reliability/Data Quality - Boiling Point</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	Studies used to determine BP range have reliabilities of 2 - Valid with Restrictions.



<b>Boiling Point</b>	
<b>Test Substance - Boiling Point</b>	
<b>Category Chemical:</b>	(64741-46-4) Naphtha, petroleum, light straight-run
<b>Test Substance:</b>	(64741-46-4) Naphtha, petroleum, light straight-run
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Naphthenic naphthas Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Boiling Point</b>	
<b>Boiling Indicator:</b>	
<b>Boiling Point Value/Range (Temperature):</b>	49 - 177 °C @ Pressure: 1013 hPa
<b>Results Remarks:</b>	<p>Decomposition: No</p> <p>The samples which were used by the API in its toxicity assessments for this Group were prepared by the fractionation of two types of crude oil, using a pilot plant still and separating cuts in a distillation range of 120 to 350°F (49 to 177°C). These figures represent a typical boiling range for light straight-run naphtha, CAS No. 64741-46-4. The standard oil industry method for determination of boiling range is ASTM D86.</p> <p>Sample API 81-08 [CAS # 64741-87-3] had an initial boiling point of 102 °F and a final boiling point of 238 °F by method ASTM D86 (equivalent to 39 and 114 °C respectively).</p>
<b>Study/Method - Boiling Point</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	ASTM D86
<b>GLP:</b>	No Data
<b>Study Reference:</b>	<p>American Society for Testing and Materials (ASTM), 1991 Annual Book of ASTM Standards. Section 5, Petroleum Products, Lubricants and Fossil Fuels, ASTM, Philadelphia, Pa., 1991.</p> <p>King, R.W. et al., Skin carcinogenicity potential of petroleum hydrocarbons. 1 -Separation and characterization of fractions for bioassay. In: Applied Toxicology of Petroleum Hydrocarbons, pp. 123-138, American Petroleum Institute Publication, API, Washington DC, 1984.</p>

American Petroleum Institute (1987)  
Comprehensive analytical analysis of API generic refinery  
streams

**Reliability/Data Quality - Boiling Point**

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Study was not conducted under GLP  
Study was conducted using a standard ASTM method



<b>Boiling Point</b>	
<b>Test Substance - Boiling Point</b>	
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Olefinic Naphthas; Sample API 83-20 Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Boiling Point</b>	
<b>Boiling Indicator:</b>	
<b>Boiling Point Value/Range (Temperature):</b>	37 - 168 °C
<b>Results Remarks:</b>	Sample API #83-20 [CAS #64741-55-5] had an initial boiling point of 99 °F and a final boiling point of 334 °F (equivalent to 37 °C and 168 °C, respectively).
<b>Study/Method - Boiling Point</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	1987
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	ASTM D86 This is the standard oil industry method for determination of boiling range.
<b>GLP:</b>	No Data
<b>Study Reference:</b>	American Petroleum Institute (1987) Comprehensive analytical analysis of API generic refinery streams
<b>Reliability/Data Quality - Boiling Point</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	No data available on GLP status of study Study was conducted using a standard ASTM method



<b>Boiling Point</b>	
<b>Test Substance - Boiling Point</b>	
<b>Category Chemical:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>AROMATIC NAPHTHAS; API 83-05            Substance is in the Gasoline Blending Streams Category.            See Category Analysis Document at  <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p> <p>Substance type: Petroleum product            Physical status: Liquid            Remark: Aromatic naphtha streams are obtained from the catalytic reforming of mainly n-alkane and cycloparaffinic feedstocks into aromatic and branched chain hydrocarbons. The hydrocarbons are mainly in the range C5 to C12. A typical aromatic naphtha is composed of the following hydrocarbon classes in the approximate proportions shown:            Content (volume %)            Paraffins 32            Olefins 0.5            Naphthenics 4            Aromatics 63.5            Full range catalytically reformed naphtha (CAS 68955-35-1) is a typical aromatic naphtha stream and the American Petroleum Institute (API, 1987) have characterized a specific sample (API 83-05) of a Full range catalytic reformed naphtha.</p>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Boiling Point</b>	
<b>Boiling Indicator:</b>	
<b>Boiling Point Value/Range (Temperature):</b>	58 - 200 °C
<b>Results Remarks:</b>	Sample API 83-05 [CAS # 68955-35-1] had an initial boiling point of 136 °F and a final boiling point of 392 °F (equivalent to 58 and 200 °C respectively).
<b>Study/Method - Boiling Point</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	<p>ASTM D86            This is the standard oil industry method for determination of boiling range.</p>
<b>GLP:</b>	No Data
<b>Study Reference:</b>	American Petroleum Institute (1987) Comprehensive analytical analysis of API generic refinery streams
<b>Reliability/Data Quality - Boiling Point</b>	

**Reliability:** Valid with Restrictions

**Reliability Remarks:** No data available on GLP status of study  
Study was conducted using a standard ASTM method



<b>Boiling Point</b>	
<b>Test Substance - Boiling Point</b>	
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Sample API 83-19 [CAS # 64741-66-8] Paraffinic Naphthas Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Boiling Point</b>	
<b>Boiling Indicator:</b>	
<b>Boiling Point Value/Range (Temperature):</b>	37 - 175 °C
<b>Results Remarks:</b>	Sample API 83-19 [CAS # 64741-66-8] had an initial boiling point of 98 °F and a final boiling point of 347 °F (equivalent to 37 and 175 °C respectively).
<b>Study/Method - Boiling Point</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	1987
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	
<b>GLP:</b>	No Data
<b>Study Reference:</b>	American Petroleum Institute (1987) Comprehensive analytical analysis of API generic refinery streams
<b>Reliability/Data Quality - Boiling Point</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	No data available on GLP status of study Study was conducted using a standard ASTM method



<b>Boiling Point</b>	
<b>Test Substance - Boiling Point</b>	
<b>Category Chemical:</b>	(64741-87-3) Naphtha, petroleum, sweetened
<b>Test Substance:</b>	(64741-87-3) Naphtha, petroleum, sweetened
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Naphthenic naphthas; sample API 81-08 Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Boiling Point</b>	
<b>Boiling Indicator:</b>	
<b>Boiling Point Value/Range (Temperature):</b>	39 - 114 °C
<b>Results Remarks:</b>	The standard oil industry method for determination of boiling range is ASTM D86.  Sample API 81-08 [CAS # 64741-87-3] had an initial boiling point of 102 °F and a final boiling point of 238 °F by method ASTM D86 (equivalent to 39 and 114 °C respectively).
<b>Study/Method - Boiling Point</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	ASTM D86 This is the standard oil industry method for determination of boiling range.
<b>GLP:</b>	No Data
<b>Study Reference:</b>	American Society for Testing and Materials (ASTM), 1991 Annual Book of ASTM Standards. Section 5, Petroleum Products, Lubricants and Fossil Fuels, ASTM, Philadelphia, Pa., 1991.  American Petroleum Institute (1987) Comprehensive analytical analysis of API generic refinery streams
<b>Reliability/Data Quality - Boiling Point</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	No data available on GLP status of study Study was conducted using a standard ASTM method





<b>Vapor Pressure</b>	
<b>Test Substance - Vapor Pressure</b>	
<b>Category Chemical:</b>	No CAS Number Provided
<b>Test Substance:</b>	No CAS Number Provided
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p> <p>All measured data used for read across are from: CONCAWE (1995) Physico-chemical characterization of gasoline samples. Study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.</p>
<b>Category Chemical Result Type:</b>	Read-Across
<b>Test Substance Result Type:</b>	
<b>Results - Vapor Pressure</b>	
<b>Vapor Pressure Value/Range (Pressure):</b>	1290 - 9150 hPa @ Temperature: 37.8 °C
<b>Results Remarks:</b>	<p>Vapor pressure measurements were reported for selected members of the gasoline blending streams category. The cited data reflect vapor pressure values measured following ASTM method D5191, which determines the total vapor pressure exerted in vacuum by air-containing, volatile, liquid petroleum products. Measurements indicate that a range of 1290 hPa to 9150 hPa may be considered typical vapor pressures for members of the gasoline blending streams category.</p>
<b>Study/Method - Vapor Pressure</b>	
<b>Key Study Sponsor Indicator:</b>	Weight of Evidence
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	ASTM D5191
<b>GLP:</b>	Yes
<b>Study Reference:</b>	<p>CONCAWE (1995) Physico-chemical characterization of gasoline samples. Study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.</p>
<b>Reliability/Data Quality - Vapor Pressure</b>	
<b>Reliability:</b>	Valid Without Restrictions

**Reliability Remarks:**

Measured data used to develop vapor pressure range for read across to untested category members were all classified as "(1) valid without restriction"



<b>Vapor Pressure</b>	
<b>Test Substance - Vapor Pressure</b>	
<b>Category Chemical:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>The sample was identified by CONCAWE as MRD-95-047, gasoline sample W94/812, CAS No. 64741-63-5, a light reformate.</p> <p>See: CONCAWE (1995) Physico-chemical characterization of gasoline samples, study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Vapor Pressure</b>	
<b>Vapor Pressure Value/Range (Pressure):</b>	= 5500 hPa @ Temperature: 37.8 °C
<b>Results Remarks:</b>	
<b>Study/Method - Vapor Pressure</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	ASTM D5191
<b>GLP:</b>	Yes
<b>Study Reference:</b>	See: CONCAWE (1995) Physico-chemical characterization of gasoline samples, study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995
<b>Reliability/Data Quality - Vapor Pressure</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	(1) valid without restriction



<b>Vapor Pressure</b>																																									
<b>Test Substance - Vapor Pressure</b>																																									
<b>Category Chemical:</b>	(64741-46-4) Naphtha, petroleum, light straight-run																																								
<b>Test Substance:</b>	(64741-46-4) Naphtha, petroleum, light straight-run																																								
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>LSRN (Low Naphthenic), CONCAWE sample CWE3            The sample was identified by CONCAWE as MRD-95-091, gasoline sample CWE3, CAS No. 64741-46-4, a light straight-run naphtha.</p> <p>Substance type: Petroleum product            Physical status: Liquid            Remark: The naphtha streams that are rich in naphthenes are obtained from the atmospheric distillation of crude oil. The streams contain saturated and aromatic hydrocarbons, mainly in the range C4 to C10.</p> <p>The naphthenic naphthas typically are composed of the following hydrocarbon classes:            Approx. Content (volume %)            Paraffins 72            Olefins &lt;0.1            Naphthenics 21            Aromatics 7</p> <p>Low naphthenic content            CONCAWE sample CWE3            CAS No. 64741-46-4            Density (g/ml @ 16°C) 0.6662            Sulfur (ppm) 83            Detailed hydrocarbon analysis (Method ASTM D 5134-92)</p> <table border="1"> <thead> <tr> <th></th> <th>Olefins</th> <th>Naphthenes</th> <th>Aromatics</th> <th>Paraffins</th> </tr> </thead> <tbody> <tr> <td>Total%</td> <td>1.04</td> <td>12.23</td> <td>3.27</td> <td>n- i- 48.19 34.02</td> </tr> <tr> <td>C4</td> <td>0.00</td> <td>0.00</td> <td>0.00</td> <td>0.006 0.000</td> </tr> <tr> <td>C5</td> <td>0.085</td> <td>4.047</td> <td>0.00</td> <td>31.91 8.228</td> </tr> <tr> <td>C6</td> <td>0.830</td> <td>6.696</td> <td>2.252</td> <td>16.139 23.917</td> </tr> <tr> <td>C7</td> <td>0.119</td> <td>1.056</td> <td>0.382</td> <td>0.647 1.241</td> </tr> <tr> <td>C8</td> <td>0.00</td> <td>0.303</td> <td>0.334</td> <td>0.263 0.324</td> </tr> <tr> <td>C9</td> <td>0.00</td> <td>0.165</td> <td>0.243</td> <td>0.162 0.178</td> </tr> </tbody> </table> <p>Substance is in the Gasoline Blending Streams Category.            See Category Analysis Document at  <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>		Olefins	Naphthenes	Aromatics	Paraffins	Total%	1.04	12.23	3.27	n- i- 48.19 34.02	C4	0.00	0.00	0.00	0.006 0.000	C5	0.085	4.047	0.00	31.91 8.228	C6	0.830	6.696	2.252	16.139 23.917	C7	0.119	1.056	0.382	0.647 1.241	C8	0.00	0.303	0.334	0.263 0.324	C9	0.00	0.165	0.243	0.162 0.178
	Olefins	Naphthenes	Aromatics	Paraffins																																					
Total%	1.04	12.23	3.27	n- i- 48.19 34.02																																					
C4	0.00	0.00	0.00	0.006 0.000																																					
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C9	0.00	0.165	0.243	0.162 0.178																																					
<b>Category Chemical Result Type:</b>	Measured																																								
<b>Test Substance Result Type:</b>	Measured																																								
<b>Results - Vapor Pressure</b>																																									
<b>Vapor Pressure Value/Range (Pressure):</b>	= 9150 hPa @ Temperature: 37.8 °C																																								
<b>Results Remarks:</b>																																									
<b>Study/Method - Vapor Pressure</b>																																									
<b>Key Study Sponsor Indicator:</b>	Key																																								
<b>Year Study Performed:</b>	1995																																								
<b>Method/Guideline Followed:</b>	Other																																								
	ASTM D5191																																								

<b>Method/Guideline and Test Condition Remarks:</b>	
<b>GLP:</b>	Yes
<b>Study Reference:</b>	CONCAWE (1995) Physico-chemical characterization of gasoline samples, study no. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.
<b>Reliability/Data Quality - Vapor Pressure</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	(1) valid without restriction



<b>Vapor Pressure</b>	
<b>Test Substance - Vapor Pressure</b>	
<b>Category Chemical:</b>	(64741-70-4) Naphtha, petroleum, isomerization
<b>Test Substance:</b>	(64741-70-4) Naphtha, petroleum, isomerization
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>The sample was identified by CONCAWE as MRD-95-045, gasoline sample W94/810, CAS No. 64741-70-4, isomerate naphtha.</p> <p>See: CONCAWE (1995) Physico-chemical characterization of gasoline samples. Study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Vapor Pressure</b>	
<b>Vapor Pressure Value/Range (Pressure):</b>	= 7860 hPa @ Temperature: 37.8 °C
<b>Results Remarks:</b>	
<b>Study/Method - Vapor Pressure</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	ASTM D5191
<b>GLP:</b>	Yes
<b>Study Reference:</b>	CONCAWE (1995) Physico-chemical characterization of gasoline samples. Study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.
<b>Reliability/Data Quality - Vapor Pressure</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	(1) valid without restriction



## Vapor Pressure

### Test Substance - Vapor Pressure

**Category Chemical:** (64741-46-4) Naphtha, petroleum, light straight-run

**Test Substance:** (64741-46-4) Naphtha, petroleum, light straight-run

**Test Substance Purity/Composition and Other Test Substance Comments:**

LSRN-Hi Naphthenic, CONCAWE sample W94/809  
The sample was identified by CONCAWE as MRD-95-044, gasoline sample W94/809, CAS No. 64741-46-4, a light straight-run naphtha.

Substance type: Petroleum product  
Physical status: Liquid  
Remark: The naphtha streams that are rich in naphthenes are obtained from the atmospheric distillation of crude oil. The streams contain saturated and aromatic hydrocarbons, mainly in the range C4 to C10. The naphthenic naphthas typically are composed of the following hydrocarbon classes:  
Approx. Content (volume %)  
Paraffins 72  
Olefins <0.1  
Naphthenics 21  
Aromatics 7  
High naphthenic content  
CONCAWE sample W94/809  
CAS No. 64741-46-4  
Density (g/ml @ 16°C) 0.7587  
Sulfur (ppm) <10  
Detailed hydrocarbon analysis (Method ASTM D 5134-92)

	Olefins		Naphthenes		Aromatics		Paraffins	
	n-	i-	n-	i-	n-	i-	n-	i-
Total%	2.18	33.92	17.26	18.88	26.83	0.00	0.141	0.059
C4	0.019	0.00	0.00	0.592	0.468	0.756	1.565	1.341
C5	0.090	0.138	0.00	5.218	3.811	0.663	10.265	5.218
C6	0.066	2.578	0.756	8.407	9.409	0.663	10.265	5.218
C7	0.663	10.265	5.218	3.762	8.834	0.074	11.036	9.044
C8	0.074	11.036	9.044	0.778	0.153	1.161	9.117	2.080
C9	1.161	9.117	2.080	0.778	0.103	0.103	0.778	0.153
C10	0.103	0.778	0.153	0.009	0.007	0.00	0.009	0.007
C11	0.00	0.009	0.007					

Substance is in the Gasoline Blending Streams Category.  
See Category Analysis Document at  
<http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

**Test Substance Result Type:** Measured

### Results - Vapor Pressure

**Vapor Pressure Value/Range (Pressure):** = 1290 hPa @ Temperature: 37.8 °C

**Results Remarks:**

### Study/Method - Vapor Pressure

**Key Study Sponsor Indicator:** Key

**Year Study Performed:** 1995

**Method/Guideline Followed:** Other

<b>Method/Guideline and Test Condition Remarks:</b>	ASTM D5191
<b>GLP:</b>	Yes
<b>Study Reference:</b>	CONCAWE (1995) Physico-chemical characterization of gasoline samples, study no. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.
<b>Reliability/Data Quality - Vapor Pressure</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	(1) valid without restriction





<b>Vapor Pressure</b>	
<b>Test Substance - Vapor Pressure</b>	
<b>Category Chemical:</b>	(68919-37-9) Naphtha, petroleum, full-range reformed
<b>Test Substance:</b>	(68919-37-9) Naphtha, petroleum, full-range reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>The sample was identified by CONCAWE as MRD-95-089, gasoline sample CWE1, CAS No. 68919-37-9, a reformat full range.</p> <p>See: CONCAWE (1995) Physico-chemical characterization of gasoline samples, study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Vapor Pressure</b>	
<b>Vapor Pressure Value/Range (Pressure):</b>	= 4630 hPa @ Temperature: 37.8 °C
<b>Results Remarks:</b>	
<b>Study/Method - Vapor Pressure</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	ASTM D5191
<b>GLP:</b>	Yes
<b>Study Reference:</b>	See: CONCAWE (1995) Physico-chemical characterization of gasoline samples, study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995
<b>Reliability/Data Quality - Vapor Pressure</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	(1) valid without restriction



<b>Vapor Pressure</b>	
<b>Test Substance - Vapor Pressure</b>	
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>The sample was identified by CONCAWE as MRD-95-090, gasoline sample CWE2, CAS No. 64741-55-5, a catalytically-cracked light naphtha.</p> <p>See: CONCAWE (1995) Physico-chemical characterization of gasoline samples, study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Vapor Pressure</b>	
<b>Vapor Pressure Value/Range (Pressure):</b>	= 5550 hPa @ Temperature: 37.8 °C
<b>Results Remarks:</b>	
<b>Study/Method - Vapor Pressure</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	ASTM D5191
<b>GLP:</b>	Yes
<b>Study Reference:</b>	See: CONCAWE (1995) Physico-chemical characterization of gasoline samples, study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995
<b>Reliability/Data Quality - Vapor Pressure</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	(1) valid without restriction



<b>Vapor Pressure</b>	
<b>Test Substance - Vapor Pressure</b>	
<b>Category Chemical:</b>	(64741-54-4) Naphtha, petroleum, heavy catalytic cracked
<b>Test Substance:</b>	(64741-54-4) Naphtha, petroleum, heavy catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>The sample was identified by CONCAWE as MRD-95-046, gasoline sample W94/811, CAS No. 64741-54-4, a catalytically-cracked heavy naphtha.</p> <p>See: CONCAWE (1995) Physico-chemical characterization of gasoline samples, study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Vapor Pressure</b>	
<b>Vapor Pressure Value/Range (Pressure):</b>	= 5930 hPa @ Temperature: 37.8 °C
<b>Results Remarks:</b>	
<b>Study/Method - Vapor Pressure</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	ASTM D5191
<b>GLP:</b>	Yes
<b>Study Reference:</b>	See: CONCAWE (1995) Physico-chemical characterization of gasoline samples, study No. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995
<b>Reliability/Data Quality - Vapor Pressure</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	(1) valid without restriction



<b>Partition Coefficient</b>	
<b>Test Substance - Partition Coefficient</b>	
<b>Category Chemical:</b>	No CAS Number Provided
<b>Test Substance:</b>	No CAS Number Provided
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Read-Across
<b>Test Substance Result Type:</b>	Estimated
<b>Results - Partition Coefficient</b>	
<b>Partition Coefficient Value/Range (Log K<sub>ow</sub>):</b>	2.13 - 4.85 @ Temperature: 25 °C
<b>Results Remarks:</b>	<p>Partition coefficient (log Kow) values were reported for individual hydrocarbon constituents found in gasoline blending streams. Constituents were selected from detailed hydrocarbon analyses of selected category members so that a range in molecular weights and hydrocarbon types was represented. Thus, the range of partition coefficient values reported reflects the typical range for log Kow of individual hydrocarbon structures in these substances. When measured values were found, these were included in the reported ranges. In the absence of empirical measurements, the computer program, KOWWIN, a subroutine in EPI-Suite™ (US EPA, 2000), was used to provide calculated values for individual structures. Based on the cited data, the partition coefficient values of the hydrocarbons in these streams are expected to fall within the range 2.13 to 4.85.</p> <p>Note: the lower end of range not agree with Gasoline Blending Streams Category Analysis Document (CAD) Data Matrix - assume that there was a value transcription error from RSS to the CAD.</p>
<b>Study/Method - Partition Coefficient</b>	
<b>Key Study Sponsor Indicator:</b>	Weight of Evidence
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	Calculated by LOGKOWWIN ver. 1.65
<b>GLP:</b>	Not Applicable
<b>Study Reference:</b>	Meylan, M, SRC 1994-1999. LOGKOWWIN is contained in the computer program EPIWIN

(Estimate ver. 3.04), available from Syracuse Research Corp.

US EPA (2000). EPIWIN (Estimation Programs Interface) Suite 3.10. US Environmental Protection Agency, Office of Pollution Prevention and Toxics, Washington, DC.

### Reliability/Data Quality - Partition Coefficient

**Reliability:** Valid with Restrictions

**Reliability Remarks:** (2) Valid with restrictions  
RELIABILITY: Estimated partition coefficient values used to develop the category read across range were calculated using a validated computer model



<b>Partition Coefficient</b>											
<b>Test Substance - Partition Coefficient</b>											
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate										
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate										
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Substance type : Petroleum product            Physical status : Liquid            Remark: Paraffinic naphtha streams are obtained by alkylation (catalytic reaction), isomerisation (catalytic conversion) and solvent extraction. They contain mostly saturated hydrocarbons, generally in the range C5 to C10 and boil in the range of approximately 90 to 160°C. The paraffinic naphthas typically are composed of the following hydrocarbon classes:</p> <table border="0"> <thead> <tr> <th></th> <th>Content (volume %)</th> </tr> </thead> <tbody> <tr> <td>Paraffins</td> <td>99.4</td> </tr> <tr> <td>Olefins</td> <td>0</td> </tr> <tr> <td>Naphthenics</td> <td>0.6</td> </tr> <tr> <td>Aromatics</td> <td>0</td> </tr> </tbody> </table> <p>Light Alkylate Naphtha (CAS 64741-66-8) is a typical paraffinic naphtha stream.            The American Petroleum Institute have reported a thorough characterization of a specific sample (API 83-19) of Light Alkylate Naphtha (LAN).</p>		Content (volume %)	Paraffins	99.4	Olefins	0	Naphthenics	0.6	Aromatics	0
	Content (volume %)										
Paraffins	99.4										
Olefins	0										
Naphthenics	0.6										
Aromatics	0										
<b>Category Chemical Result Type:</b>	Estimated by Calculation										
<b>Test Substance Result Type:</b>	Estimated										
<b>Results - Partition Coefficient</b>											
<b>Partition Coefficient Value/Range (Log K<sub>ow</sub>):</b>	3.11 - 4.54 @ Temperature: 25 °C										
<b>Results Remarks:</b>	<p>Log P values represent the spread of calculated and/or measured values for the C5 to C9 hydrocarbon components found in LAN, CAS No. 64741-66-8. Detailed hydrocarbon analysis performed by Chevron Research was used to identify the components of this specific LAN sample. Calculated SAR result for surrogate structures contained in program database (smilecas.dat). Calculation based on an atom/fragment contribution method of W. Meylan and P. Howard.</p>										
<b>Study/Method - Partition Coefficient</b>											
<b>Key Study Sponsor Indicator:</b>	Key										
<b>Year Study Performed:</b>	2000										
<b>Method/Guideline Followed:</b>	Other										
<b>Method/Guideline and Test Condition Remarks:</b>	Calculated by LOGKOWWIN ver. 1.65										
<b>GLP:</b>	No										
<b>Study Reference:</b>	<p>Meylan, M, SRC 1994-1999.            LOGKOWWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.</p>										
<b>Reliability/Data Quality - Partition Coefficient</b>											

**Reliability:** Valid with Restrictions

**Reliability Remarks:** (2) Valid with restrictions  
RELIABILITY: Estimated partition coefficient values were calculated using a validated computer model



<b>Partition Coefficient</b>	
<b>Test Substance - Partition Coefficient</b>	
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Substance type : Petroleum product            Physical status : Liquid            Remark : The naphtha streams obtained from the catalytic cracking of heavy distillates into lighter fractions contain saturated, olefinic and aromatic hydrocarbons. However, their olefins content is higher than any of the naphtha streams derived by other processes. The catalytically cracked naphthas contain hydrocarbons in the range C4 to C10. The catalytically cracked naphthas typically are composed of the following hydrocarbon classes:            Approx. Content (volume %)            Paraffins 30            Olefins 46            Naphthenics 10            Aromatics 14            Light catalytically cracked naphtha (LCCN) (CAS 64741-55-5) is a typical olefinic naphtha stream. The American Petroleum Institute have reported (API, 1987) a thorough characterization of a specific sample of a light catalytically cracked naphtha (API 83-20), which has a high olefinic content and which was used in many of the mammalian toxicity studies.</p>
<b>Category Chemical Result Type:</b>	Estimated by Calculation
<b>Test Substance Result Type:</b>	Estimated
<b>Results - Partition Coefficient</b>	
<b>Partition Coefficient Value/Range (Log K<sub>ow</sub>):</b>	2.13 - 4 @ Temperature: 25 °C
<b>Results Remarks:</b>	<p>Log P values represent the spread of calculated and/or measured values for the C5 to C9 hydrocarbon components found in LCCN, CAS No. 64741-55-5. Detailed hydrocarbon analysis performed by Chevron Research was used to identify the components of this specific LCCN sample. Calculated SAR result for surrogate structures contained in program database (smilecas.dat). Calculation based on an atom/fragment contribution method of W. Meylan and P. Howard.</p> <p>Note: the lower end of range not agree with Gasoline Blending Streams Category Analysis Document (CAD) Data Matrix - assume that there was a value transcription error from RSS to the CAD.</p>
<b>Study/Method - Partition Coefficient</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	Calculated by LOGKOWWIN ver. 1.65.



<b>GLP:</b>	No
<b>Study Reference:</b>	Meylan, M, SRC 1994-1999. LOGKOWWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.
<b>Reliability/Data Quality - Partition Coefficient</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated partition coefficient values were calculated using a validated computer model



<b>Partition Coefficient</b>																																									
<b>Test Substance - Partition Coefficient</b>																																									
<b>Category Chemical:</b>	(64741-46-4) Naphtha, petroleum, light straight-run																																								
<b>Test Substance:</b>	(64741-46-4) Naphtha, petroleum, light straight-run																																								
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>LSRN (Low Naphthenic), CONCAWE sample CWE3            Substance type: Petroleum product            Physical status: Liquid            Remark: The naphtha streams that are rich in naphthenes are obtained from the atmospheric distillation of crude oil. The streams contain saturated and aromatic hydrocarbons, mainly in the range C4 to C10. The naphthenic naphthas typically are composed of the following hydrocarbon classes:            Approx. Content (volume %)            Paraffins 72            Olefins &lt;0.1            Naphthenics 21            Aromatics 7            Low naphthenic content            CONCAWE sample CWE3            CAS No. 64741-46-4            Density (g/ml @ 16°C) 0.6662            Sulfur (ppm) 83            Detailed hydrocarbon analysis (Method ASTM D 5134-92)</p> <table border="1"> <thead> <tr> <th></th> <th>Olefins</th> <th>Naphthenes</th> <th>Aromatics</th> <th>Paraffins</th> </tr> </thead> <tbody> <tr> <td>Total%</td> <td>1.04</td> <td>12.23</td> <td>3.27</td> <td>48.19</td> </tr> <tr> <td>C4</td> <td>0.00</td> <td>0.00</td> <td>0.00</td> <td>0.006</td> </tr> <tr> <td>C5</td> <td>0.085</td> <td>4.047</td> <td>0.00</td> <td>31.91</td> </tr> <tr> <td>C6</td> <td>0.830</td> <td>6.696</td> <td>2.252</td> <td>16.139</td> </tr> <tr> <td>C7</td> <td>0.119</td> <td>1.056</td> <td>0.382</td> <td>0.647</td> </tr> <tr> <td>C8</td> <td>0.00</td> <td>0.303</td> <td>0.334</td> <td>0.263</td> </tr> <tr> <td>C9</td> <td>0.00</td> <td>0.165</td> <td>0.243</td> <td>0.162</td> </tr> </tbody> </table> <p>Substance is in the Gasoline Blending Streams Category.            See Category Analysis Document(s) at  <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>		Olefins	Naphthenes	Aromatics	Paraffins	Total%	1.04	12.23	3.27	48.19	C4	0.00	0.00	0.00	0.006	C5	0.085	4.047	0.00	31.91	C6	0.830	6.696	2.252	16.139	C7	0.119	1.056	0.382	0.647	C8	0.00	0.303	0.334	0.263	C9	0.00	0.165	0.243	0.162
	Olefins	Naphthenes	Aromatics	Paraffins																																					
Total%	1.04	12.23	3.27	48.19																																					
C4	0.00	0.00	0.00	0.006																																					
C5	0.085	4.047	0.00	31.91																																					
C6	0.830	6.696	2.252	16.139																																					
C7	0.119	1.056	0.382	0.647																																					
C8	0.00	0.303	0.334	0.263																																					
C9	0.00	0.165	0.243	0.162																																					
<b>Category Chemical Result Type:</b>	Estimated by Calculation																																								
<b>Test Substance Result Type:</b>	Estimated																																								
<b>Results - Partition Coefficient</b>																																									
<b>Partition Coefficient Value/Range (Log K<sub>ow</sub>):</b>	2.13 - 4 @ Temperature: 25 °C																																								
<b>Results Remarks:</b>	<p>Log P values represent the spread of calculated and/or measured values for C5 to C7 hydrocarbon components found in LSRN, CAS No 64741-46-4. Detailed hydrocarbon analysis was used to identify the components of this specific low naphthenic LSRN sample. Calculation based on an atom/fragment contribution method of W. Meylan and P. Howard. Calculated SAR result for surrogate structure contained in program database (smilecas.dat).</p>																																								
<b>Study/Method - Partition Coefficient</b>																																									
<b>Key Study Sponsor Indicator:</b>	Key																																								
<b>Year Study Performed:</b>	2000																																								
<b>Method/Guideline Followed:</b>	Other																																								

<b>Method/Guideline and Test Condition Remarks:</b>	Calculated by LOGKOWWIN ver. 1.65
<b>GLP:</b>	Yes
<b>Study Reference:</b>	<p>CONCAWE (1995)  Physico-chemical characterization of gasoline samples, study no. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.</p> <p>Meylan, M, SRC 1994-1999.  LOGKOWWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.</p>
<b>Reliability/Data Quality - Partition Coefficient</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated partition coefficient values were calculated using a validated computer model



<b>Partition Coefficient</b>	
<b>Test Substance - Partition Coefficient</b>	
<b>Category Chemical:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Full -Range Catalytically Reformed Naphtha (FRCRN)- CAS No. 68955-35-1; API sample 83-05
<b>Category Chemical Result Type:</b>	Estimated by Calculation
<b>Test Substance Result Type:</b>	Estimated
<b>Results - Partition Coefficient</b>	
<b>Partition Coefficient Value/Range (Log K<sub>ow</sub>):</b>	2.13 - 4.76 @ Temperature: 25 °C
<b>Results Remarks:</b>	
<b>Study/Method - Partition Coefficient</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Calculated by LOGKOWWIN ver. 1.65. Log P values represent the spread of calculated and/or measured values for C5 to C9 hydrocarbon components found in FRCRN, CAS No 68955-35-1. Detailed hydrocarbon analysis was used to identify the components of this FRCRN (63% aromatics) sample. Calculated SAR result for surrogate structures contained in program database (smilecas.dat). Calculation based on an atom/fragment contribution method of W. Meylan and P. Howard.</p>
<b>GLP:</b>	No
<b>Study Reference:</b>	<p>American Petroleum Institute (1987) Comprehensive analytical analysis of API generic refinery streams.</p> <p>Meylan, M, SRC 1994-1999. LOGKOWWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.</p>
<b>Reliability/Data Quality - Partition Coefficient</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	<p>(2) Valid with restrictions</p> <p>RELIABILITY: Estimated partition coefficient values were calculated using a validated computer model</p>



<b>Partition Coefficient</b>	
<b>Test Substance - Partition Coefficient</b>	
<b>Category Chemical:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Light Catalytically Reformed Naphtha
<b>Category Chemical Result Type:</b>	Estimated by Calculation
<b>Test Substance Result Type:</b>	Estimated
<b>Results - Partition Coefficient</b>	
<b>Partition Coefficient Value/Range (Log K<sub>ow</sub>):</b>	2.13 - 4.54 @ Temperature: 25 °C
<b>Results Remarks:</b>	
<b>Study/Method - Partition Coefficient</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	Calculated by LOGKOWWIN ver. 1.65. Log P values represent the spread of calculated and/or measured values for C5 to C9 hydrocarbon components found in LCRN, CAS No 64741-63-5. Detailed hydrocarbon analysis performed by Chevron Research was used to identify the components of this specific LCRN sample. Calculation based on an atom/fragment contribution method of W. Meylan and P. Howard. Calculated SAR result for surrogate structure contained in program database (smilecas.dat).
<b>GLP:</b>	No
<b>Study Reference:</b>	Chevron Research (1995) Gasoline analysis Internal report  Meylan, M, SRC 1994-1999. LOGKOWWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.
<b>Reliability/Data Quality - Partition Coefficient</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated partition coefficient values were calculated using a validated computer model



<b>Partition Coefficient</b>	
<b>Test Substance - Partition Coefficient</b>	
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Gasoline CONCAWE sample, CWE5, Blend (match to API PS-6) Substance is in the Gasoline Blending Streams Category. See Category Analysis Document at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>  [Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]
<b>Category Chemical Result Type:</b>	Estimated by Calculation
<b>Test Substance Result Type:</b>	Estimated
<b>Results - Partition Coefficient</b>	
<b>Partition Coefficient Value/Range (Log K<sub>ow</sub>):</b>	2.13 - 4.5 @ Temperature: 25 °C
<b>Results Remarks:</b>	Log P values represent the spread of calculated and/or measured values for C5 to C8 hydrocarbon components found in gasoline, CAS No 86290-81-5. Detailed hydrocarbon analysis was used to identify the components of this specific gasoline sample. Calculation based on an atom/fragment contribution method of W. Meylan and P. Howard. Calculated SAR result for surrogate structure contained in program database (smilecas.dat).
<b>Study/Method - Partition Coefficient</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	Calculated by LOGKOWWIN ver. 1.65.
<b>GLP:</b>	No
<b>Study Reference:</b>	CONCAWE (1995) Physico-chemical characterization of gasoline samples, study no. 104990C. Study conducted by Exxon Biomedical Sciences Inc. Concawe, Brussels, 1995. Meylan, M, SRC 1994-1999. LOGKOWWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.  The most current version of EPIWIN (Estimation Programs Interface) Suite is available from the US EPA, Office of Pollution Prevention and Toxics, Washington, DC.
<b>Reliability/Data Quality - Partition Coefficient</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	

(2) Valid with restrictions  
RELIABILITY: Estimated partition coefficient values were  
calculated using a validated computer model.



<b>Water Solubility</b>	
<b>Test Substance - Water Solubility</b>	
<b>Category Chemical:</b>	No CAS Number Provided
<b>Test Substance:</b>	No CAS Number Provided
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Read-Across
<b>Test Substance Result Type:</b>	
<b>Results - Water Solubility</b>	
<b>Water Solubility Indicator:</b>	
<b>Water Solubility Value/Range (Solubility):</b>	1 - 2000 mg/L
<b>pH Value:</b>	
<b>pKa - Protein Kinase:</b>	
<b>pH Value at Saturation:</b>	
<b>Results Remarks:</b>	Individual components of complex petroleum substances have specific and differing water solubility values. For example, constituent hydrocarbons of gasoline blending streams have measured and calculated solubility values ranging from <1 to 2000 mg/L. However, for complex petroleum substances, the resulting aqueous concentration of each constituent hydrocarbon is a function of: 1) the loading rate (i.e., ratio of petroleum substance to water), 2) log Kow, 3) the amount of component present, and 4) the maximum water solubility of each component. Initially, as the complex petroleum substance is added to water in amounts below the solubility limit of the least soluble component, the aqueous concentration increases proportionally until the least soluble component reaches a saturation concentration. As more product is added to water, only the more soluble components continue to dissolve, resulting in a two phase system. Further addition of the complex petroleum substance results in an aqueous concentration that is a non-linear function of the amount added.
<b>Study/Method - Water Solubility</b>	
<b>Key Study Sponsor Indicator:</b>	Weight of Evidence
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	



<b>Method/Guideline and Test Condition Remarks:</b>	
<b>GLP:</b>	
<b>Study Reference:</b>	
<b>Reliability/Data Quality - Water Solubility</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: All studies used to derive the read across range were conducted under GLPs, but did not follow standard guidelines due to the hydrocarbon composition of the complex substance.



<b>Water Solubility</b>											
<b>Test Substance - Water Solubility</b>											
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate										
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate										
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Substance type : Petroleum product            Physical status : Liquid            Remark: Paraffinic naphtha streams are obtained by alkylation (catalytic reaction), isomerisation (catalytic conversion) and solvent extraction. They contain mostly saturated hydrocarbons, generally in the range C5 to C10 and boil in the range of approximately 90 to 160°C. The paraffinic naphthas typically are composed of the following hydrocarbon classes:</p> <table border="0"> <thead> <tr> <th colspan="2">Content (volume %)</th> </tr> </thead> <tbody> <tr> <td>Paraffins</td> <td>99.4</td> </tr> <tr> <td>Olefins</td> <td>0</td> </tr> <tr> <td>Naphthenics</td> <td>0.6</td> </tr> <tr> <td>Aromatics</td> <td>0</td> </tr> </tbody> </table> <p>Light Alkylate Naphtha (CAS 64741-66-8) is a typical paraffinic naphtha stream.            The American Petroleum Institute have reported a thorough characterization of a specific sample (API 83-19) of Light Alkylate Naphtha (LAN).</p> <p>Substance is in the Gasoline Blending Streams Category.            See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>	Content (volume %)		Paraffins	99.4	Olefins	0	Naphthenics	0.6	Aromatics	0
Content (volume %)											
Paraffins	99.4										
Olefins	0										
Naphthenics	0.6										
Aromatics	0										
<b>Category Chemical Result Type:</b>	Measured										
<b>Test Substance Result Type:</b>	Measured										
<b>Results - Water Solubility</b>											
<b>Water Solubility Indicator:</b>											
<b>Water Solubility Value/Range (Solubility):</b>	1 - 30 mg/L										
<b>pH Value:</b>											
<b>pKa - Protein Kinase:</b>											
<b>pH Value at Saturation:</b>											
<b>Results Remarks:</b>	<p>Gas chromatographic analysis of selected components indicated freshwater and saltwater solubilities of 1.6 and 0.9 ppm respectively. Measured test concentrations of the light alkylate naphtha were based on the total combined concentrations of 2,3 dimethyl butane; 2,4 dimethyl pentane; 2,2,4 trimethyl pentane; 2,5 dimethyl hexane; 2,3,4 trimethyl pentane, 2,3,3 trimethyl pentane and 1-methyl-1-ethyl cyclopentane, which represent 68% composition of the test substance. Concentrations for these components reached equilibrium in freshwater and saltwater by 24 and 12 hours respectively.</p> <p>Individual components of complex petroleum substances have specific and differing solubilities. Calculated and measured water solubilities for LAN components range from &lt;1 to approximately 30 mg/l. At any particular loading rate, the resulting aqueous concentration of each chemical constituent is a function of the relative volume of the two phases (aqueous and the petroleum mixture), the partition</p>										

coefficient between the phases, the amount of component present and the maximum water solubility of each component. Initially as the petroleum mixture is added in amounts below the solubility limit of the least soluble component the aqueous concentration increases proportionally until the least soluble component reaches a saturation concentration, and only the more soluble components continue to dissolve, resulting in a two phase system. Further addition of the petroleum mixture results in an aqueous concentration that is a non-linear function of the amount added.

### Study/Method - Water Solubility

<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	Preparation of Water Soluble Fraction Water Accommodated Fractions (WAFs) of LAN were prepared at 50 mg/l loading in freshwater and saltwater and equilibrated for 72 hours in tightly closed systems with minimal headspace.
<b>GLP:</b>	Yes
<b>Study Reference:</b>	CONCAWE (1992) Ecotoxicological Testing Of Petroleum Products : Test Methodology. Report 92/56, CONCAWE, Brussels. CONCAWE (1996) Environmental risk assessment of petroleum substances: the hydrocarbon block method. Report 96/52, CONCAWE, Brussels. ECETOC (1998) QSARS in the Assessment of the Environmental Fate and Effects of Chemicals. Technical Report No. 74 ECETOC, (1996) Aquatic Toxicity Testing of Sparingly Soluble, Volatile and Unstable Substances. Monograph 26. Stonybrook Laboratories, Inc. (1995) Method Validation for the Analysis of Whole Light Alkylate Naphtha (LAN) in Water Accommodated Fraction (WAF) using Purge-and-Trap and GC/FID, Study No. 65969. Stonybrook Laboratories Inc. Princeton, NJ.

### Reliability/Data Quality - Water Solubility

<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: GLP; not a guideline study



<b>Water Solubility</b>									
<b>Test Substance - Water Solubility</b>									
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked								
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked								
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Substance type : Petroleum product            Physical status : Liquid            Remark : The naphtha streams obtained from the catalytic cracking of heavy distillates into lighter fractions contain saturated, olefinic and aromatic hydrocarbons. However, their olefins content is higher than any of the naphtha streams derived by other processes. The catalytically cracked naphthas contain hydrocarbons in the range C4 to C10. The catalytically cracked naphthas typically are composed of the following hydrocarbon classes:</p> <p>Approx. Content (volume %)</p> <table border="0"> <tr><td>Paraffins</td><td>30</td></tr> <tr><td>Olefins</td><td>46</td></tr> <tr><td>Naphthenics</td><td>10</td></tr> <tr><td>Aromatics</td><td>14</td></tr> </table> <p>Light catalytically cracked naphtha (LCCN) (CAS 64741-55-5) is a typical olefinic naphtha stream. The American Petroleum Institute have reported (API, 1987) a thorough characterization of a specific sample of a light catalytically cracked naphtha (API 83-20), which has a high olefinic content and which was used in many of the mammalian toxicity studies.</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>	Paraffins	30	Olefins	46	Naphthenics	10	Aromatics	14
Paraffins	30								
Olefins	46								
Naphthenics	10								
Aromatics	14								
<b>Category Chemical Result Type:</b>	Measured								
<b>Test Substance Result Type:</b>	Measured								
<b>Results - Water Solubility</b>									
<b>Water Solubility Indicator:</b>									
<b>Water Solubility Value/Range (Solubility):</b>	3 - 2000 mg/L								
<b>pH Value:</b>									
<b>pKa - Protein Kinase:</b>									
<b>pH Value at Saturation:</b>									
<b>Results Remarks:</b>	<p>Gas chromatographic analysis of selected components indicated freshwater and saltwater solubilities of 4.6 and 4.3 ppm respectively. Measured test concentrations of the light alkylate naphtha were based on the total combined concentrations of benzene, toluene, ethylbenzene, o-xylene and p-xylene, which represent 13% composition of the test substance. Concentrations for these components reached equilibrium in freshwater and saltwater by 24 and 12 hours respectively.</p> <p>Conclusion : Individual components of complex petroleum substances have specific and differing solubilities. Calculated and measured water solubilities for LCCN components range from approximately 3 to 2000 mg/l. At any particular loading rate, the resulting aqueous concentration of each chemical constituent is a function of</p>								

the relative volume of the two phases (aqueous and the petroleum mixture), the partition coefficient between the phases, the amount of component present and the maximum water solubility of each component. Initially as the petroleum mixture is added in amounts below the solubility limit of the least soluble component the aqueous concentration increases proportionally until the least soluble component reaches a saturation concentration, and only the more soluble components continue to dissolve, resulting in a two phase system. Further addition of the petroleum mixture results in an aqueous concentration that is a non-linear function of the amount added.

### Study/Method - Water Solubility

**Key Study Sponsor Indicator:**

Key

**Year Study Performed:**

1995

**Method/Guideline Followed:**

Other

**Method/Guideline and Test Condition Remarks:**

Preparation of Water Soluble Fraction  
Water Accommodated Fractions (WAFs) of LCCN were prepared at 50 mg/l loading in freshwater and saltwater and equilibrated for 72 hours in tightly closed systems with minimal headspace.

**GLP:**

Yes

**Study Reference:**

CONCAWE (1992) Ecotoxicological Testing Of Petroleum Products : Test Methodology. Report 92/56, CONCAWE, Brussels.  
CONCAWE (1996) Environmental risk assessment of petroleum substances: the hydrocarbon block method. Report 96/52, CONCAWE, Brussels.ECETOC (1998) QSARS in the Assessment of the Environmental Fate and Effects of Chemicals. Technical Report No. 74.  
ECETOC, (1996) Aquatic Toxicity Testing of Sparingly Soluble, Volatile and Unstable Substances. Monograph 26.  
Stonybrook Laboratories, Inc.(1995) Method Validation for the Analysis of Whole Light Catalytically Cracked Naphtha (LCCN) in Water Accomodated Fraction (WAF) using Purge-and-Trap and GC/FID, Study No. 66232. Stonybrook Laboratories, Inc. Princeton, NJ.1995

### Reliability/Data Quality - Water Solubility

**Reliability:**

Valid with Restrictions

**Reliability Remarks:**

(2) Valid with restrictions  
RELIABILITY: GLP; not a guideline study



## Water Solubility

### Test Substance - Water Solubility

**Category Chemical:** (64741-46-4) Naphtha, petroleum, light straight-run

**Test Substance:** (64741-46-4) Naphtha, petroleum, light straight-run

**Test Substance Purity/Composition and Other Test Substance Comments:**

LSRN (Low Naphthenic), CONCAWE sample CWE39  
 Substance type: Petroleum product  
 Physical status: Liquid  
 Remark: The naphtha streams that are rich in naphthenes are obtained from the atmospheric distillation of crude oil. The streams contain saturated and aromatic hydrocarbons, mainly in the range C4 to C10  
 The naphthenic naphthas typically are composed of the following hydrocarbon classes:  
 Approx. Content (volume %)  
 Paraffins 72  
 Olefins <0.1  
 Naphthenics 21  
 Aromatics 7  
 Low naphthenic content  
 CONCAWE sample CWE3  
 CAS No. 64741-46-4  
 Density (g/ml @ 16°C) 0.6662  
 Sulfur (ppm) 83  
 Detailed hydrocarbon analysis (Method ASTM D 5134-92)

	Olefins		Naphthenes		Aromatics		Paraffins	
	n-	i-	n-	i-	n-	i-	n-	i-
Total%	1.04	12.23	3.27	48.19	34.02			
C4	0.00	0.00	0.00	0.006	0.000			
C5	0.085	4.047	0.00	31.91	8.228			
C6	0.830	6.696	2.252	16.139	23.917			
C7	0.119	1.056	0.382	0.647	1.241			
C8	0.00	0.303	0.334	0.263	0.324			
C9	0.00	0.165	0.243	0.162	0.178			

Substance is in the Gasoline Blending Streams Category.  
 See Category Analysis Document(s) at  
<http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

**Test Substance Result Type:** Measured

### Results - Water Solubility

**Water Solubility Indicator:**

**Water Solubility Value/Range (Solubility):** 3 - 2000 mg/L

**pH Value:**

**pKa - Protein Kinase:**

**pH Value at Saturation:**

**Results Remarks:** Gas chromatographic analysis of BTEX components indicated freshwater solubility at 24 hours of 4.9 ppm as benzene. Individual components of complex petroleum substances have specific and differing solubilities. Calculated and measured water solubilities for LSRN components range from approximately 3 to 2000 mg/l. At any particular loading rate, the resulting aqueous concentration of each chemical constituent is a function of the relative volume of the two phases (aqueous and the petroleum mixture), the partition

coefficient between the phases, the amount of component present and the maximum water solubility of each component. Initially as the petroleum mixture is added in amounts below the solubility limit of the least soluble component the aqueous concentration increases proportionally until the least soluble component reaches a saturation concentration, and only the more soluble components continue to dissolve, resulting in a two phase system. Further addition of the petroleum mixture results in an aqueous concentration that is a non-linear function of the amount added.

### Study/Method - Water Solubility

**Key Study Sponsor Indicator:**

Key

**Year Study Performed:**

1995

**Method/Guideline Followed:**

Other

**Method/Guideline and Test Condition Remarks:**

Preparation of Water Soluble Fractions  
Water Accommodated Fractions (WAFs) of LSRN were prepared at 1000 mg/L loading in freshwater and equilibrated for 48 hours in tightly closed systems with minimal headspace.

**GLP:**

Yes

**Study Reference:**

CONCAWE (1992) Ecotoxicological Testing Of Petroleum Products : Test Methodology. Report 92/56, CONCAWE, Brussels.  
CONCAWE (1996) Environmental risk assessment of petroleum substances: the hydrocarbon block method. Report 96/52, CONCAWE, Brussels.  
ECETOC (1998) QSARS in the Assessment of the Environmental Fate and Effects of Chemicals. Technical Report No. 74  
ECETOC, (1996) Aquatic Toxicity Testing of Sparingly Soluble, Volatile and Unstable Substances. Monograph 26.  
Springborn Laboratories, Inc. (1993) CWE3 (Straight Run Gasoline) Toxicity to freshwater Alga, Selenastrum capricornutum. SLI Report # 93-6-4805.  
Springborn Laboratories, Inc. Environmental Sciences division, 790 Main Street, Wareham, Massachusetts, USA.

### Reliability/Data Quality - Water Solubility

**Reliability:**

Valid with Restrictions

**Reliability Remarks:**

(2) Valid with restrictions  
RELIABILITY: GLP; not a guideline study



<b>Water Solubility</b>																																																																																																			
<b>Test Substance - Water Solubility</b>																																																																																																			
<b>Category Chemical:</b>	(64741-46-4) Naphtha, petroleum, light straight-run																																																																																																		
<b>Test Substance:</b>	(64741-46-4) Naphtha, petroleum, light straight-run																																																																																																		
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>LSRN-Hi Naphthenic, CONCAWE sample W94/809            Substance type: Petroleum product            Physical status: Liquid            Remark: The naphtha streams that are rich in naphthenes are obtained from the atmospheric distillation of crude oil. The streams contain saturated and aromatic hydrocarbons, mainly in the range C4 to C10            The naphthenic naphthas typically are composed of the following hydrocarbon classes:            Approx. Content (volume %)            Paraffins 72            Olefins &lt;0.1            Naphthenics 21            Aromatics 7            High naphthenic content            CONCAWE sample W94/809            CAS No. 64741-46-4            Density (g/ml @ 16°C) 0.7587            Sulfur (ppm) &lt;10            Detailed hydrocarbon analysis (Method ASTM D 5134-92)</p> <table border="1" style="margin-left: auto; margin-right: auto;"> <thead> <tr> <th rowspan="2"></th> <th colspan="2">Olefins</th> <th colspan="2">Naphthenes</th> <th colspan="2">Aromatics</th> <th colspan="2">Paraffins</th> </tr> <tr> <th>n-</th> <th>i-</th> <th>n-</th> <th>i-</th> <th>n-</th> <th>i-</th> <th>n-</th> <th>i-</th> </tr> </thead> <tbody> <tr> <td>Total%</td> <td>2.18</td> <td></td> <td>33.92</td> <td></td> <td>17.26</td> <td></td> <td>18.88</td> <td>26.83</td> </tr> <tr> <td>C4</td> <td>0.019</td> <td></td> <td>0.00</td> <td></td> <td>0.00</td> <td></td> <td>0.141</td> <td>0.059</td> </tr> <tr> <td>C5</td> <td>0.090</td> <td></td> <td>0.138</td> <td></td> <td>0.00</td> <td></td> <td>0.592</td> <td>0.468</td> </tr> <tr> <td>C6</td> <td>0.066</td> <td></td> <td>2.578</td> <td></td> <td>0.756</td> <td></td> <td>1.565</td> <td>1.341</td> </tr> <tr> <td>C7</td> <td>0.663</td> <td></td> <td>10.265</td> <td></td> <td>5.218</td> <td></td> <td>3.887</td> <td>3.811</td> </tr> <tr> <td>C8</td> <td>0.074</td> <td></td> <td>11.036</td> <td></td> <td>9.044</td> <td></td> <td>8.407</td> <td>9.409</td> </tr> <tr> <td>C9</td> <td>1.161</td> <td></td> <td>9.117</td> <td></td> <td>2.080</td> <td></td> <td>3.762</td> <td>8.834</td> </tr> <tr> <td>C10</td> <td>0.103</td> <td></td> <td>0.778</td> <td></td> <td>0.153</td> <td></td> <td>0.778</td> <td>0.103</td> </tr> <tr> <td>C11</td> <td>0.00</td> <td></td> <td>0.009</td> <td></td> <td>0.007</td> <td></td> <td>0.009</td> <td>0.145</td> </tr> </tbody> </table> <p>Substance is in the Gasoline Blending Streams Category.            See Category Analysis Document(s) at  <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>		Olefins		Naphthenes		Aromatics		Paraffins		n-	i-	n-	i-	n-	i-	n-	i-	Total%	2.18		33.92		17.26		18.88	26.83	C4	0.019		0.00		0.00		0.141	0.059	C5	0.090		0.138		0.00		0.592	0.468	C6	0.066		2.578		0.756		1.565	1.341	C7	0.663		10.265		5.218		3.887	3.811	C8	0.074		11.036		9.044		8.407	9.409	C9	1.161		9.117		2.080		3.762	8.834	C10	0.103		0.778		0.153		0.778	0.103	C11	0.00		0.009		0.007		0.009	0.145
	Olefins		Naphthenes		Aromatics		Paraffins																																																																																												
	n-	i-	n-	i-	n-	i-	n-	i-																																																																																											
Total%	2.18		33.92		17.26		18.88	26.83																																																																																											
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<b>Category Chemical Result Type:</b>	Measured																																																																																																		
<b>Test Substance Result Type:</b>	Measured																																																																																																		
<b>Results - Water Solubility</b>																																																																																																			
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<b>pKa - Protein Kinase:</b>																																																																																																			
<b>pH Value at Saturation:</b>																																																																																																			
<b>Results Remarks:</b>	Gas chromatographic analysis of TEX (toluene, ethyl benzene, and xylenes) components indicated freshwater solubility of 5.7-7.9 ppm (as TEX). Measured test concentrations of the LSRN were based on the total combined concentrations of TEXN which represent approximately 13% composition of the test substance. Concentrations for these																																																																																																		



components reached equilibrium by 19 hours. Individual components of complex petroleum substances have specific and differing solubilities. Calculated and measured water solubilities for LSRN components range from approximately 3 to 2000 mg/l. At any particular loading rate, the resulting aqueous concentration of each chemical constituent is a function of the relative volume of the two phases (aqueous and the petroleum mixture), the partition coefficient between the phases, the amount of component present and the maximum water solubility of each component. Initially as the petroleum mixture is added in amounts below the solubility limit of the least soluble component the aqueous concentration increases proportionally until the least soluble component reaches a saturation concentration, and only the more soluble components continue to dissolve, resulting in a two phase system. Further addition of the petroleum mixture results in an aqueous concentration that is a non-linear function of the amount added.

#### Study/Method - Water Solubility

**Key Study Sponsor Indicator:**

Key

**Year Study Performed:**

1995

**Method/Guideline Followed:**

Other

**Method/Guideline and Test Condition Remarks:**

Preparation of Water Soluble Fractions  
Water Accommodated Fractions (WAFs) of LSRN were prepared at 100 mg/L loading in freshwater and equilibrated for 48 hours in tightly closed systems with minimal headspace.

**GLP:**

Yes

**Study Reference:**

CONCAWE (1992) Ecotoxicological Testing Of Petroleum Products : Test Methodology. Report 92/56, CONCAWE, Brussels.  
CONCAWE (1995) Fish -acute toxicity test: study no. 104858, test substance MRD-95-048. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels.  
CONCAWE (1996) Environmental risk assessment of petroleum substances: the hydrocarbon block method. Report 96/52, CONCAWE, Brussels.  
ECETOC (1998) QSARS in the Assessment of the Environmental Fate and Effects of Chemicals. Technical Report No. 74  
ECETOC, (1996) Aquatic Toxicity Testing of Sparingly Soluble, Volatile and Unstable Substances. Monograph 26.

#### Reliability/Data Quality - Water Solubility

**Reliability:**

Valid with Restrictions

**Reliability Remarks:**

(2) Valid with restrictions  
RELIABILITY: GLP; not a guideline study



<b>Water Solubility</b>	
<b>Test Substance - Water Solubility</b>	
<b>Category Chemical:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Full -Range Catalytically Reformed Naphtha (FRCRN)-CAS No. 68955-35-1; API sample 83-05.</p> <p>AROMATIC NAPHTHAS            Substance type: Petroleum product            Physical status: Liquid            Remark: Aromatic naphtha streams are obtained from the catalytic reforming of mainly n-alkane and cycloparaffinic feedstocks into aromatic and branched chain hydrocarbons. The hydrocarbons are mainly in the range C5 to C12. A typical aromatic naphtha is composed of the following hydrocarbon classes in the approximate proportions shown:            Content (volume %)            Paraffins 32            Olefins 0.5            Naphthenics 4            Aromatics 63.5</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Water Solubility</b>	
<b>Water Solubility Indicator:</b>	
<b>Water Solubility Value/Range (Solubility):</b>	3 - 2000 mg/L
<b>pH Value:</b>	
<b>pKa - Protein Kinase:</b>	
<b>pH Value at Saturation:</b>	
<b>Results Remarks:</b>	<p>Gas chromatographic analysis of LCRN components benzene, toluene, ethylbenzene, ortho, meta and para-xylene in WAFs indicated freshwater solubility of 6.3 ppm. Concentrations for these components reached equilibrium by 48 hours. Individual components of complex petroleum substances have specific and differing solubilities. Calculated and measured water solubilities for FRCRN components range from approximately 3 to 2000 mg/l. At any particular loading rate, the resulting aqueous concentration of each chemical constituent is a function of the relative volume of the two phases (aqueous and the petroleum mixture), the partition coefficient between the phases, the amount of component present and the maximum water solubility of each component. Initially as the petroleum mixture is added in amounts below the solubility limit of the least soluble component the aqueous concentration increases proportionally until the least soluble component reaches a saturation concentration, and only the more soluble components continue to dissolve,</p>

resulting in a two phase system. Further addition of the petroleum mixture results in an aqueous concentration that is a non-linear function of the amount added.

### Study/Method - Water Solubility

**Key Study Sponsor Indicator:**

Key

**Year Study Performed:**

1995

**Method/Guideline Followed:**

Other

**Method/Guideline and Test Condition Remarks:**

Preparation of Water Soluble Fraction Water Accommodated Fractions (WAFs) of CONCAWE Reformate light naphtha (LCRN), CAS no. 64741-63-5 (CONCAWE sample ID W94/812) were prepared at 100 mg/l loading in freshwater and equilibrated for 48 hours in tightly closed systems with minimal headspace. Detailed hydrocarbon analysis was used to identify the components of this CONCAWE Light Cracked Naphtha (63% aromatics) sample. The analysis indicated that the composition of the CONCAWE LCRN sample was essentially identical to the composition of API 83-05 FRCRN sample. Therefore the water solubility information for the CONCAWE LCRN sample is applicable to the FRCRN sample.

**GLP:**

Yes

**Study Reference:**

CONCAWE (1992) Ecotoxicological Testing Of Petroleum Products : Test Methodology. Report 92/56, CONCAWE, Brussels.  
 CONCAWE (1995) Algal, Growth Inhibition Test: study no. 104767, test substance MRD-95-047. Study conducted by Exxon Biomedical Sciences Inc.  
 CONCAWE (1996) Environmental risk assessment of petroleum substances: the hydrocarbon block method. Report 96/52, CONCAWE, Brussels.  
 ECETOC (1998) QSARS in the Assessment of the Environmental Fate and Effects of Chemicals. Technical Report No. 74  
 ECETOC, (1996) Aquatic Toxicity Testing of Sparingly Soluble, Volatile and Unstable Substances. Monograph 26.

### Reliability/Data Quality - Water Solubility

**Reliability:**

Valid with Restrictions

**Reliability Remarks:**

(2) Valid with restrictions  
 RELIABILITY: GLP; not a guideline study



## Water Solubility

### Test Substance - Water Solubility

**Category Chemical:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:**

AROMATIC NAPHTHAS  
 Substance type: Petroleum product  
 Physical status: Liquid  
 Remark: Aromatic naphtha streams are obtained from the catalytic reforming of mainly n-alkane and cycloparaffinic feedstocks into aromatic and branched chain hydrocarbons. The hydrocarbons are mainly in the range C5 to C12. A typical aromatic naphtha is composed of the following hydrocarbon classes in the approximate proportions shown:  
 Content (volume %)  
 Paraffins 32  
 Olefins 0.5  
 Naphthenics 4  
 Aromatics 63.5  
 Light Catalytically Reformed Naphtha  
 Sample identified by Chevron Research as a light catalytically reformed naphtha CAS No. 64741-63-5  
 Detailed hydrocarbon analysis

	Olefins	Naphthenes	Aromatics	Paraffins	total n-
total%	0.90	2.36	39.40	57.34	17.51
C4	0.00	0.00	0.00	0.81	0.78
C5	0.34	0.26	0.00	19.45	8.05
C6	0.27	0.62	8.37	16.23	4.69
C7	0.28	1.18	29.77	17.70	3.59
C8	0.01	0.27	1.26	3.12	0.40

Substance is in the Gasoline Blending Streams Category.  
 See Category Analysis Document(s) at  
<http://www.petroleumhvp.org>

**Category Chemical Result Type:** Measured

**Test Substance Result Type:** Measured

### Results - Water Solubility

**Water Solubility Indicator:**

**Water Solubility Value/Range (Solubility):** 3 - 2000 mg/L

**pH Value:**

**pKa - Protein Kinase:**

**pH Value at Saturation:**

**Results Remarks:** Gas chromatographic analysis of selected components freshwater and saltwater solubilities of 13.7 and 14.0 ppm respectively. Measured test concentrations of the light catalytically reformed naphtha were based on the total combined concentrations of pentane, 2-methyl-pentane, benzene, toluene, ethylbenzene, ortho, meta and para-xylene, which represent more than 50% composition of the test substance. Concentrations for these components reached equilibrium by 24 hours.  
 Individual components of complex petroleum substances have specific and differing solubilities. Calculated and

measured water solubilities for LCRN components range from approximately 3 to 2000 mg/l. At any particular loading rate, the resulting aqueous concentration of each chemical constituent is a function of the relative volume of the two phases (aqueous and the petroleum mixture), the partition coefficient between the phases, the amount of component present and the maximum water solubility of each component. Initially as the petroleum mixture is added in amounts below the solubility limit of the least soluble component the aqueous concentration increases proportionally until the least soluble component reaches a saturation concentration, and only the more soluble components continue to dissolve, resulting in a two phase system. Further addition of the petroleum mixture results in an aqueous concentration that is a non-linear function of the amount added.

### Study/Method - Water Solubility

**Key Study Sponsor Indicator:**

Key

**Year Study Performed:**

1995

**Method/Guideline Followed:**

Other

**Method/Guideline and Test Condition Remarks:**

Preparation of Water Soluble Fraction Water Accommodated Fractions (WAFs) of LCRN were prepared at 50 mg/l loading in freshwater and saltwater and equilibrated for 48 hours in tightly closed systems with minimal headspace.

**GLP:**

Yes

**Study Reference:**

ABC Laboratories, Inc (1998) Method Validation for the Analysis of the Water Accomodated Fraction of Light Catalytically Cracked Naphtha using Purge-and-Trap and GC/FID, Study No. 43582. ABC Laboratories, Inc. Environmental Toxicology, 7200 E. ABC Lane, Columbia, Missouri  
 CONCAWE (1992) Ecotoxicological Testing Of Petroleum Products : Test Methodology. Report 92/56, CONCAWE, Brussels.  
 CONCAWE (1996) Environmental risk assessment of petroleum substances: the hydrocarbon block method. Report 96/52, CONCAWE, Brussels.  
 ECETOC (1998) QSARS in the Assessment of the Environmental Fate and Effects of Chemicals. Technical Report No. 74  
 ECETOC, (1996) Aquatic Toxicity Testing of Sparingly Soluble, Volatile and Unstable Substances. Monograph 26.

### Reliability/Data Quality - Water Solubility

**Reliability:**

Valid with Restrictions

**Reliability Remarks:**

(2) Valid with restrictions  
 GLP; not a guideline study



<b>Water Solubility</b>	
<b>Test Substance - Water Solubility</b>	
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Gasoline CONCAWE sample, CWE5, Blend (match to API PS-6), CAS No. 86290-81-5  [Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]  Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Water Solubility</b>	
<b>Water Solubility Indicator:</b>	
<b>Water Solubility Value/Range (Solubility):</b>	3 - 2000 mg/L
<b>pH Value:</b>	
<b>pKa - Protein Kinase:</b>	
<b>pH Value at Saturation:</b>	
<b>Results Remarks:</b>	Gas chromatographic analysis of BTEX components indicated freshwater solubility at 24 hours of 3.1, 3.1, <6.9E-3, and 0.92 ppm (as BTEX, respectively). Individual components of complex petroleum substances have specific and differing solubilities. Calculated and measured water solubilities for LSRN components range from approximately 3 to 2000 mg/l. At any particular loading rate, the resulting aqueous concentration of each chemical constituent is a function of the relative volume of the two phases (aqueous and the petroleum mixture), the partition coefficient between the phases, the amount of component present and the maximum water solubility of each component. Initially as the petroleum mixture is added in amounts below the solubility limit of the least soluble component the aqueous concentration increases proportionally until the least soluble component reaches a saturation concentration, and only the more soluble components continue to dissolve, resulting in a two phase system. Further addition of the petroleum mixture results in an aqueous concentration that is a non-linear function of the amount added.
<b>Study/Method - Water Solubility</b>	
<b>Key Study Sponsor Indicator:</b>	Key

<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	Preparation of Water Soluble Fraction Water Accommodated Fractions (WAFs) of LSRN were prepared at 100 mg/l loading in freshwater and equilibrated for 48 hours in tightly closed systems with minimal headspace.
<b>GLP:</b>	Yes
<b>Study Reference:</b>	CONCAWE (1992) Ecotoxicological Testing Of Petroleum Products : Test Methodology. Report 92/56, CONCAWE, Brussels. CONCAWE (1996) Environmental risk assessment of petroleum substances: the hydrocarbon block method. Report 96/52, CONCAWE, Brussels. ECETOC (1998) QSARS in the Assessment of the Environmental Fate and Effects of Chemicals. Technical Report No. 74 ECETOC, (1996) Aquatic Toxicity Testing of Sparingly Soluble, Volatile and Unstable Substances. Monograph 26.
<b>Reliability/Data Quality - Water Solubility</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: GLP; not a guideline study

# Physical-Chemical Other





<b>Density/Specific Gravity</b>	
<b>Test Substance - Density/Specific Gravity</b>	
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>API PS-6 gasoline</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p> <p>[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]</p>
<b>Category Chemical Result Type:</b>	
<b>Test Substance Result Type:</b>	
<b>Results - Density/Specific Gravity</b>	
<b>Density Type:</b>	Relative Density
<b>Density/Specific Gravity Value/Range:</b>	circa 50
<b>Results Remarks:</b>	
<b>Study/Method - Density/Specific Gravity</b>	
<b>Key Study Sponsor Indicator:</b>	
<b>Year Study Performed:</b>	1984
<b>Method/Guideline Followed:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	
<b>GLP:</b>	
<b>Study Reference:</b>	McFarland, H. N., Ulrich, C. E., Holdsworth, C. E., Kitchen, D. N., Halliwell, W. H. and Blum, S. C. (1984) A chronic Inhalation Study with unleaded gasoline vapor. J. Am. College of Toxicol. Vol. 3, No. 4, pp 231-248
<b>Reliability/Data Quality - Density/Specific Gravity</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	(1) Valid without restriction



<b>Density/Specific Gravity</b>	
<b>Test Substance - Density/Specific Gravity</b>	
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Paraffinic Naphtha Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Results - Density/Specific Gravity</b>	
<b>Density Type:</b>	Relative Density
<b>Density/Specific Gravity Value/Range:</b>	.697 @ Temperature: 15 °C
<b>Results Remarks:</b>	
<b>Study/Method - Density/Specific Gravity</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	Other
<b>Method/Guideline and Test Condition Remarks:</b>	ASTM D287
<b>GLP:</b>	No Data
<b>Study Reference:</b>	American Petroleum Institute (1987) Comprehensive analytical analysis of API generic refinery streams.  American Society for Testing and Materials (ASTM), 1991 Annual Book of ASTM Standards. Section 5, Petroleum Products, Lubricants and Fossil Fuels, ASTM, Philadelphia, Pa., 1991.
<b>Reliability/Data Quality - Density/Specific Gravity</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	Study conducted under standard oil industry method ASTM D287

# Fate SIDS



<b>Photodegradation</b>	
<b>Test Substance - Photodegradation</b>	
<b>Category Chemical:</b>	No CAS Number Provided
<b>Test Substance:</b>	No CAS Number Provided
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Read-Across
<b>Test Substance Result Type:</b>	Estimated
<b>Results - Photodegradation</b>	
<b>Photodegradation Result Description:</b>	Indirect Photolysis
<b>Photodegradation Value/Range:</b>	.0000000000006691 - .00000000008941 cm3/molecule*sec
<b>Half Life:</b>	
<b>Rate Constant:</b>	
<b>Photo Medium:</b>	
<b>Temperature:</b>	
<b>Sensitizer:</b>	Hydroxy Radicals
<b>Sensitizer Concentration and Units:</b>	1500000 OH radicals/cm3
<b>Light Source:</b>	Sunlight
<b>Light Source Spectrum:</b>	
<b>UV/VIS Absorption Spectrum:</b>	
<b>Quantum Yield:</b>	
<b>Breakdown Products Description:</b>	
<b>Results Remarks:</b>	Direct photodegradation is not expected to play an important role in the environmental fate of gasoline naphtha streams. Indirect photodegradation via reaction with hydroxyl radicals may be important in the gas-phase degradation of hydrocarbons that volatilize to the troposphere. An overall range of half-lives expected for individual components of these streams is 1.4 h to 16 days.
<b>Study/Method - Photodegradation</b>	
<b>Key Study Sponsor Indicator:</b>	Weight of Evidence
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	

<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	Estimated photodegradation values from studies used to develop the read across range were calculated using AOPWIN ver 1.89 (EPI Suite EPIWIN)
<b>GLP:</b>	
<b>Study Reference:</b>	
<b>Reliability/Data Quality - Photodegradation</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated photodegradation values from studies used to develop the read across range were calculated using a validated computer model



<b>Photodegradation</b>																																																																
<b>Test Substance - Photodegradation</b>																																																																
<b>Category Chemical:</b>	(64741-46-4) Naphtha, petroleum, light straight-run																																																															
<b>Test Substance:</b>	(64741-46-4) Naphtha, petroleum, light straight-run																																																															
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>LSRN-Moderate (19.7%) Naphthenic, LSRN (Moderate Naphthenic)            Substance type: Petroleum product            Physical status: Liquid            Remark: The naphtha streams that are rich in naphthenes are obtained from the atmospheric distillation of crude oil. The streams contain saturated and aromatic hydrocarbons, mainly in the range C4 to C10.</p> <p>The naphthenic naphthas typically are composed of the following hydrocarbon classes:            Approx. Content (volume %)            Paraffins 72            Olefins &lt;0.1            Naphthenics 21            Aromatics 7            Moderate naphthenic content            Chevron sample (Chevron, 1995)            CAS No. 64741-46-4            Detailed hydrocarbon analysis (Method ASTM D 5134-92)</p> <table border="1"> <thead> <tr> <th></th> <th>Olefins</th> <th>Naphthenes</th> <th>Aromatics</th> <th>Paraffins</th> <th>n-</th> <th>i-</th> </tr> </thead> <tbody> <tr> <td>Total%</td> <td>0.72</td> <td>22.41</td> <td>3.06</td> <td>73.31</td> <td>31.13</td> <td></td> </tr> <tr> <td>C4</td> <td>0.03</td> <td>0.00</td> <td>0.00</td> <td>5.85</td> <td>5.580</td> <td></td> </tr> <tr> <td>C5</td> <td>0.085</td> <td>1.73</td> <td>0.00</td> <td>38.80</td> <td>16.27</td> <td></td> </tr> <tr> <td>C6</td> <td>0.36</td> <td>6.24</td> <td>0.70</td> <td>18.18</td> <td>6.26</td> <td></td> </tr> <tr> <td>C7</td> <td>0.05</td> <td>7.11</td> <td>1.12</td> <td>5.58</td> <td>2.00</td> <td></td> </tr> <tr> <td>C8</td> <td>0.00</td> <td>5.31</td> <td>0.96</td> <td>3.22</td> <td>0.79</td> <td></td> </tr> <tr> <td>C9</td> <td>0.00</td> <td>1.95</td> <td>0.25</td> <td>1.20</td> <td>0.13</td> <td></td> </tr> <tr> <td>C10</td> <td>0.00</td> <td>0.07</td> <td>0.03</td> <td>0.46</td> <td>0.08</td> <td></td> </tr> </tbody> </table> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>		Olefins	Naphthenes	Aromatics	Paraffins	n-	i-	Total%	0.72	22.41	3.06	73.31	31.13		C4	0.03	0.00	0.00	5.85	5.580		C5	0.085	1.73	0.00	38.80	16.27		C6	0.36	6.24	0.70	18.18	6.26		C7	0.05	7.11	1.12	5.58	2.00		C8	0.00	5.31	0.96	3.22	0.79		C9	0.00	1.95	0.25	1.20	0.13		C10	0.00	0.07	0.03	0.46	0.08	
	Olefins	Naphthenes	Aromatics	Paraffins	n-	i-																																																										
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<b>Category Chemical Result Type:</b>	Estimated by Calculation																																																															
<b>Test Substance Result Type:</b>	Estimated																																																															
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<b>Sensitizer Concentration and Units:</b>	1500000 OH radicals/cm <sup>3</sup>																																																															
<b>Light Source:</b>	Sunlight																																																															

<b>Light Source Spectrum:</b>	
<b>UV/VIS Absorption Spectrum:</b>	
<b>Quantum Yield:</b>	
<b>Breakdown Products Description:</b>	
<b>Results Remarks:</b>	<p>Rate Constant: 0.6691E -12 (isopentane) to 13.5606E -12 (m-xylene) cm<sup>3</sup>/molecule-sec  Half-life: 0.789 to 15.985 days  AOPWIN ver. 1.89 calculates atmospheric oxidation half lives of hydrocarbons in contact with hydroxyl radicals in the troposphere, under the influence of sunlight and in contact with O<sub>3</sub>. Atmospheric oxidation rates were calculated for the C5 to C9 hydrocarbon components found in LSRN, CAS No. 64741-46-4. Detailed hydrocarbon analysis was used to identify the components of this specific moderate naphthenic LSRN sample. Based on a 12-hour day, the range for atmospheric half-lives for LSRN constituents is: 0.789 days (m-xylene) to 15.985 days (isopentane).</p>
<b>Study/Method - Photodegradation</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	AOPWIN (EPI Suite; EPIWIN)
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Calculated by AOPWIN ver. 1.89. Method based on the work of R. Atkinson
<b>Method/Guideline and Test Condition Remarks:</b>	Relative intensity : = 1 based on intensity of sunlight
<b>GLP:</b>	No
<b>Study Reference:</b>	Meylan, M, SRC 1994-1999. AOPWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.
<b>Reliability/Data Quality - Photodegradation</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated photodegradation values were calculated using a validated computer model



<b>Photodegradation</b>											
<b>Test Substance - Photodegradation</b>											
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate										
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate										
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Light alkylate naphtha            Paraffinic naphtha            Substance type : Petroleum product            Physical status : Liquid            Remark: Paraffinic naphtha streams are obtained by alkylation (catalytic reaction), isomerisation (catalytic conversion) and solvent extraction. They contain mostly saturated hydrocarbons, generally in the range C5 to C10 and boil in the range of approximately 90 to 160°C. The paraffinic naphthas typically are composed of the following hydrocarbon classes:</p> <table style="margin-left: 40px;"> <thead> <tr> <th colspan="2" style="text-align: center;">Content (volume %)</th> </tr> </thead> <tbody> <tr> <td>Paraffins</td> <td style="text-align: right;">99.4</td> </tr> <tr> <td>Olefins</td> <td style="text-align: right;">0</td> </tr> <tr> <td>Naphthenics</td> <td style="text-align: right;">0.6</td> </tr> <tr> <td>Aromatics</td> <td style="text-align: right;">0</td> </tr> </tbody> </table> <p>Light Alkylate Naphtha (CAS 64741-66-8) is a typical paraffinic naphtha stream.            The American Petroleum Institute has reported a thorough characterization of a specific sample (API 83-19) of Light Alkylate Naphtha (LAN). See the analytical data report at the website below.</p> <p>Substance is in the Gasoline Blending Streams Category.            See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>	Content (volume %)		Paraffins	99.4	Olefins	0	Naphthenics	0.6	Aromatics	0
Content (volume %)											
Paraffins	99.4										
Olefins	0										
Naphthenics	0.6										
Aromatics	0										
<b>Category Chemical Result Type:</b>	Estimated by Calculation										
<b>Test Substance Result Type:</b>	Estimated										
<b>Results - Photodegradation</b>											
<b>Photodegradation Result Description:</b>	Indirect Photolysis										
<b>Photodegradation Value/Range:</b>	.0000000000006691 - .000000000009956 cm3/molecule*sec										
<b>Half Life:</b>											
<b>Rate Constant:</b>											
<b>Photo Medium:</b>											
<b>Temperature:</b>											
<b>Sensitizer:</b>	Hydroxy Radicals										
<b>Sensitizer Concentration and Units:</b>	1500000 OH radicals/cm3										
<b>Light Source:</b>	Sunlight										
<b>Light Source Spectrum:</b>											
<b>UV/VIS Absorption Spectrum:</b>											
<b>Quantum Yield:</b>											



<b>Breakdown Products Description:</b>	
<b>Results Remarks:</b>	Rate Constant 0.6691E-12 (isopentane) cm <sup>3</sup> /mol-sec to 9.956E-12 (2,3,5 trimethyl hexane) Half-life 1.074 days to 15.985 days
<b>Study/Method - Photodegradation</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	AOPWIN (EPI Suite; EPIWIN)
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Calculated by AOPWIN ver. 1.89. Method based on the work of R. Atkinson
<b>Method/Guideline and Test Condition Remarks:</b>	AOPWIN ver. 1.89 calculates atmospheric oxidation half lives of hydrocarbons in contact with hydroxyl radicals in the troposphere, under the influence of sunlight and in contact with O <sub>3</sub> . Atmospheric oxidation rates were calculated for the C <sub>5</sub> to C <sub>8</sub> hydrocarbon components found in LAN, CAS No. 64741-66-8. Detailed hydrocarbon analysis performed by Chevron Research was used to identify the components of this specific LAN sample. Based on a 12-hour day, the range for atmospheric half-lives for LAN constituents is: 1.074 days (2,3,5 trimethyl hexane) to 15.985 days (isopentane).
<b>GLP:</b>	No
<b>Study Reference:</b>	Meylan, M, SRC 1994-1999. AOPWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.
<b>Reliability/Data Quality - Photodegradation</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated photodegradation values were calculated using a validated computer model



<b>Photodegradation</b>											
<b>Test Substance - Photodegradation</b>											
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked										
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked										
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Light catalytic cracked naphtha  Olefinic naphthas  Substance type : Petroleum product  Physical status : Liquid  Remark : The naphtha streams obtained from the catalytic cracking of heavy distillates into lighter fractions contain saturated, olefinic and aromatic hydrocarbons. However, their olefins content is higher than any of the naphtha streams derived by other processes. The catalytically cracked naphthas contain hydrocarbons in the range C4 to C10. The catalytically cracked naphthas typically are composed of the following hydrocarbon classes:</p> <table border="0"> <tr> <td colspan="2">Approx. Content (volume %)</td> </tr> <tr> <td>Paraffins</td> <td>30</td> </tr> <tr> <td>Olefins</td> <td>46</td> </tr> <tr> <td>Naphthenics</td> <td>10</td> </tr> <tr> <td>Aromatics</td> <td>14</td> </tr> </table> <p>Light catalytically cracked naphtha (LCCN) (CAS 64741-55-5) is a typical olefinic naphtha stream. The American Petroleum Institute have reported (API, 1987) a thorough characterization of a specific sample of a light catalytically cracked naphtha (API 83-20), which has a high olefinic content and which was used in many of the mammalian toxicity studies. The characterization of this sample can be found in the analytical data report at the website below.</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>	Approx. Content (volume %)		Paraffins	30	Olefins	46	Naphthenics	10	Aromatics	14
Approx. Content (volume %)											
Paraffins	30										
Olefins	46										
Naphthenics	10										
Aromatics	14										
<b>Category Chemical Result Type:</b>	Estimated by Calculation										
<b>Test Substance Result Type:</b>	Estimated										
<b>Results - Photodegradation</b>											
<b>Photodegradation Result Description:</b>	Indirect Photolysis										
<b>Photodegradation Value/Range:</b>	.0000000000006691 - .00000000008941 cm3/molecule*sec										
<b>Half Life:</b>											
<b>Rate Constant:</b>											
<b>Photo Medium:</b>											
<b>Temperature:</b>											
<b>Sensitizer:</b>	Hydroxy Radicals										
<b>Sensitizer Concentration and Units:</b>	1500000 OH radicals/cm3										
<b>Light Source:</b>	Sunlight										
<b>Light Source Spectrum:</b>											

<b>UV/VIS Absorption Spectrum:</b>	
<b>Quantum Yield:</b>	
<b>Breakdown Products Description:</b>	
<b>Results Remarks:</b>	<p>Rate constant: 0.6691E-12 cm<sup>3</sup>/mol-sec (isopentane) to 89.41 E-12 (1-methyl cyclopentene)  Half life: 1.44 hours to 15.985 days</p> <p>Sensitizer: O3 radical  Conc. of sensitizer: 7E1103/cm<sup>3</sup>  Rate constant: 1.2E-17 to 43-17 cm<sup>3</sup>/molecule-sec  Half life: 38.378 min to 22.920 Hrs.</p>
<b>Study/Method - Photodegradation</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	AOPWIN (EPI Suite; EPIWIN)
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Calculated by AOPWIN ver. 1.89. Method based on the work of R. Atkinson
<b>Method/Guideline and Test Condition Remarks:</b>	<p>AOPWIN ver. 1.89 calculates atmospheric oxidation half lives of hydrocarbons in contact with hydroxyl radicals in the troposphere, under the influence of sunlight and in contact with O3. Atmospheric oxidation rates were calculated for the C5 to C9 hydrocarbon components found in LCCN CAS No. 647415. Detailed hydrocarbon analysis performed by Chevron Research was used to identify the components of this specific LCCN sample.</p> <p>Based on a 12-hour day, the range for atmospheric half-lives for LCCN constituents due to OH reactions is: 1.44 hours (1-methyl cyclopentene) to 15.985 days (isopentane).</p> <p>The range for atmospheric half-lives due to O3 reactions for LCCN olefinic constituents (accounting for approximately 30% composition) is 38.378 min (1-methyl cyclopentene) to 22.920 Hrs (C5 olefins).</p>
<b>GLP:</b>	No
<b>Study Reference:</b>	Meylan, M, SRC 1994-1999. AOPWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.
<b>Reliability/Data Quality - Photodegradation</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated photodegradation values were calculated using a validated computer model



<b>Photodegradation</b>	
<b>Test Substance - Photodegradation</b>	
<b>Category Chemical:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Full -Range Catalytically Reformed Naphtha (FRCRN) -CAS No. 68955-35-1; API sample 83-05.            AROMATIC NAPHTHAS            Substance type: Petroleum product            Physical status: Liquid            Remark: Aromatic naphtha streams are obtained from the catalytic reforming of mainly n-alkane and cycloparaffinic feedstocks into aromatic and branched chain hydrocarbons. The hydrocarbons are mainly in the range C5 to C12. A typical aromatic naphtha is composed of the following hydrocarbon classes in the approximate proportions shown:            Content (volume %)            Paraffins 32            Olefins 0.5            Naphthenics 4            Aromatics 63.5</p> <p>Full range catalytically reformed naphtha (CAS 64741-66-8) is a typical aromatic naphtha stream and the American Petroleum Institute (API, 1987) have characterized a specific sample (API 83-05) of a Full range catalytic reformed naphtha. The results of this characterization can be found in the analytical report at the website below.</p> <p>Substance is in the Gasoline Blending Streams Category.            See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>
<b>Category Chemical Result Type:</b>	Estimated by Calculation
<b>Test Substance Result Type:</b>	Estimated
<b>Results - Photodegradation</b>	
<b>Photodegradation Result Description:</b>	Indirect Photolysis
<b>Photodegradation Value/Range:</b>	.0000000000006691 - .000000000016698 cm3/molecule*sec
<b>Half Life:</b>	
<b>Rate Constant:</b>	
<b>Photo Medium:</b>	
<b>Temperature:</b>	
<b>Sensitizer:</b>	Hydroxy Radicals
<b>Sensitizer Concentration and Units:</b>	1500000 OH radicals/cm3
<b>Light Source:</b>	Sunlight
<b>Light Source Spectrum:</b>	
<b>UV/VIS Absorption Spectrum:</b>	
<b>Quantum Yield:</b>	

<b>Breakdown Products Description:</b>	
<b>Results Remarks:</b>	Rate constant: 0.6691E -12 cm <sup>3</sup> /mol-sec (isopentane) to 16.698E -12 (1,2,4 trimethyl benzene) Half life: 0.641 to 15.985 days
<b>Study/Method - Photodegradation</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	AOPWIN (EPI Suite; EPIWIN)
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Calculated by AOPWIN ver. 1.89. Method based on the work of R. Atkinson
<b>Method/Guideline and Test Condition Remarks:</b>	Relative intensity: = 1 based on intensity of sunlight AOPWIN ver. 1.89 calculates atmospheric oxidation half lives of hydrocarbons in contact with hydroxyl radicals in the troposphere, under the influence of sunlight and in contact with O <sub>3</sub> . Atmospheric oxidation rates were calculated for the C5 to C9 hydrocarbon components found in FRCRN, CAS No. 68955-35-1. Detailed hydrocarbon analysis was used to identify the components of this specific FRCRN (63% aromatics) sample. Based on a 12-hour day, the range for atmospheric half-lives for FRCRN constituents is: 0.641 days (1, 2, 4 trimethylbenzene) to 15.985 days (isopentane).
<b>GLP:</b>	No
<b>Study Reference:</b>	Meylan, M, SRC 1994-1999. AOPWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.
<b>Reliability/Data Quality - Photodegradation</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated photodegradation values were calculated using a validated computer model



<b>Photodegradation</b>																																				
<b>Test Substance - Photodegradation</b>																																				
<b>Category Chemical:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed																																			
<b>Test Substance:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed																																			
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Light Catalytically Reformed Naphtha.            AROMATIC NAPHTHAS            Substance type: Petroleum product            Physical status: Liquid            Remark: Aromatic naphtha streams are obtained from the catalytic reforming of mainly n-alkane and cycloparaffinic feedstocks into aromatic and branched chain hydrocarbons. The hydrocarbons are mainly in the range C5 to C12. A typical aromatic naphtha is composed of the following hydrocarbon classes in the approximate proportions shown:            Content (volume %)            Paraffins 32            Olefins 0.5            Naphthenics 4            Aromatics 63.5            Sample identified by Chevron Research as a light catalytically reformed naphtha CAS No. 64741-63-5            Detailed hydrocarbon analysis</p> <table style="margin-left: auto; margin-right: auto;"> <thead> <tr> <th></th> <th>Olefins</th> <th>Naphthenes</th> <th>Aromatics</th> <th>Paraffins</th> </tr> </thead> <tbody> <tr> <td>total%</td> <td>0.90</td> <td>2.36</td> <td>39.40</td> <td>57.34</td> </tr> <tr> <td>C4</td> <td>0.00</td> <td>0.00</td> <td>0.00</td> <td>0.81</td> </tr> <tr> <td>C5</td> <td>0.34</td> <td>0.26</td> <td>0.00</td> <td>19.45</td> </tr> <tr> <td>C6</td> <td>0.27</td> <td>0.62</td> <td>8.37</td> <td>16.23</td> </tr> <tr> <td>C7</td> <td>0.28</td> <td>1.18</td> <td>29.77</td> <td>17.70</td> </tr> <tr> <td>C8</td> <td>0.01</td> <td>0.27</td> <td>1.26</td> <td>3.12</td> </tr> </tbody> </table> <p style="text-align: center;">total n-</p> <p>Substance is in the Gasoline Blending Streams Category.            See Category Analysis Document(s) at  <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>		Olefins	Naphthenes	Aromatics	Paraffins	total%	0.90	2.36	39.40	57.34	C4	0.00	0.00	0.00	0.81	C5	0.34	0.26	0.00	19.45	C6	0.27	0.62	8.37	16.23	C7	0.28	1.18	29.77	17.70	C8	0.01	0.27	1.26	3.12
	Olefins	Naphthenes	Aromatics	Paraffins																																
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<b>Category Chemical Result Type:</b>	Estimated by Calculation																																			
<b>Test Substance Result Type:</b>	Estimated																																			
<b>Results - Photodegradation</b>																																				
<b>Photodegradation Result Description:</b>	Indirect Photolysis																																			
<b>Photodegradation Value/Range:</b>	.0000000000006691 - .000000000071392 cm3/molecule*sec																																			
<b>Half Life:</b>																																				
<b>Rate Constant:</b>																																				
<b>Photo Medium:</b>																																				
<b>Temperature:</b>																																				
<b>Sensitizer:</b>	Hydroxy Radicals																																			
<b>Sensitizer Concentration and Units:</b>	1500000 OH radicals/cm3																																			
<b>Light Source:</b>	Sunlight																																			
<b>Light Source Spectrum:</b>																																				

<b>UV/VIS Absorption Spectrum:</b>	
<b>Quantum Yield:</b>	
<b>Breakdown Products Description:</b>	
<b>Results Remarks:</b>	Rate constant: 0.6691E-12 cm <sup>3</sup> /mol-sec (isopentane) to 7.1392E-12 (2,3 dimethyl pentane) Half life: 1.498 to 15.985 days
<b>Study/Method - Photodegradation</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	AOPWIN (EPI Suite; EPIWIN)
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Calculated by AOPWIN ver. 1.89. Method based on the work of R. Atkinson
<b>Method/Guideline and Test Condition Remarks:</b>	Relative intensity: = 1 based on intensity of sunlight AOPWIN ver. 1.89 calculates atmospheric oxidation half lives of hydrocarbons in contact with hydroxyl radicals in the troposphere, under the influence of sunlight and in contact with O <sub>3</sub> . Atmospheric oxidation rates were calculated for the C5 to C8 hydrocarbon components found in LCRN, CAS No. 64741-63-5. Detailed hydrocarbon analysis performed by Chevron Research was used to identify the components of this specific LCRN sample. Based on a 12-hour day, the range for atmospheric half-lives for LCRN constituents is: 1.498 days (2,3 dimethyl pentane) to 15.985 days (isopentane).
<b>GLP:</b>	No
<b>Study Reference:</b>	Meylan, M, SRC 1994-1999. AOPWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.
<b>Reliability/Data Quality - Photodegradation</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated photodegradation values were calculated using a validated computer model



## Photodegradation

### Test Substance - Photodegradation

**Category Chemical:** (86290-81-5) Antiknock Gasoline

**Test Substance:** (86290-81-5) Antiknock Gasoline

**Test Substance Purity/Composition and Other Test Substance Comments:** Gasoline CONCAWE sample, CWE5, Blend (match to API PS-6), CAS No. 86290-81-5

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]

**Category Chemical Result Type:** Estimated by Calculation

**Test Substance Result Type:** Estimated

### Results - Photodegradation

**Photodegradation Result Description:** Indirect Photolysis

**Photodegradation Value/Range:** .0000000000006991 - .0000000000135606 cm3/molecule\*sec

**Half Life:**

**Rate Constant:**

**Photo Medium:**

**Temperature:**

**Sensitizer:** Hydroxy Radicals

**Sensitizer Concentration and Units:** 1500000 OH radicals/cm3

**Light Source:** Sunlight

**Light Source Spectrum:**

**UV/VIS Absorption Spectrum:**

**Quantum Yield:**

**Breakdown Products Description:**

**Results Remarks:** Rate constant: 0.6991 E-12 (isopentane) to 13.5606 E-12 (m-xylene) cm3/molecule-sec  
Half-life: 0.789 to 15.985 days

### Study/Method - Photodegradation

**Key Study Sponsor Indicator:** Key



<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	AOPWIN (EPI Suite; EPIWIN)
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Calculated by AOPWIN ver. 1.89. Method based on the work of R. Atkinson
<b>Method/Guideline and Test Condition Remarks:</b>	AOPWIN ver. 1.89 calculates atmospheric oxidation half lives of hydrocarbons in contact with hydroxyl radicals in the troposphere, under the influence of sunlight and in contact with O <sub>3</sub> . Atmospheric oxidation rates were calculated for the C5 to C8 hydrocarbon components found in gasoline. Detailed hydrocarbon analysis was used to identify the components of this specific gasoline sample. Based on a 12-hour day, the range for atmospheric half-lives for gasoline constituents is: 0.789 days (m-xylene) to 15.985 days (isopentane).
<b>GLP:</b>	No
<b>Study Reference:</b>	<p>CONCAWE (1995) Physico-chemical characterization of gasoline samples, study no. 104990C. Study conducted by Exxon Biomedical Sciences Inc. Concawe, Brussels, 1995.</p> <p>Meylan, M, SRC 1994-1999. AOPWIN is contained in the computer program EPIWIN (Estimate ver. 3.04), available from Syracuse Research Corp.</p>
<b>Reliability/Data Quality - Photodegradation</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated photodegradation values were calculated using a validated computer model



<b>Stability in Water</b>			
<b>Test Substance - Stability In Water</b>			
<b>Category Chemical:</b>	No CAS Number Provided		
<b>Test Substance:</b>	No CAS Number Provided		
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>		
<b>Category Chemical Result Type:</b>	Read-Across		
<b>Test Substance Result Type:</b>			
<b>Results - Stability In Water</b>			
<b>Stability in Water Result Description:</b>			
<b>Stability in Water Value/Range:</b>			
<b>pH Value:</b>			
<b>Hydrolysis Indicator:</b>			
<b>Preliminary Test:</b>			
<b>Effect:</b>	<table border="1"> <tr> <td>Half-Life</td> <td>@ pH Value</td> </tr> </table>	Half-Life	@ pH Value
Half-Life	@ pH Value		
<b>Breakdown Products Description:</b>			
<b>Results Remarks:</b>	Hydrolysis unlikely  Hydrolysis is unlikely for gasoline and blending streams. Hydrolysis of an organic chemical is the transformation process in which a water molecule or hydroxide ion reacts to form a new carbon-oxygen bond. Chemicals that have a potential to hydrolyze include alkylhalides, amides, carbamates, carboxylic acid esters and lactones, epoxides, phosphate esters, and sulfonic acid esters (Harris, 1982b). The chemical components that comprise the gasoline blending streams category are hydrocarbons, which are not included in these chemical groups, and they are not subject to hydrolysis reactions with water.		
<b>Study/Method - Stability In Water</b>			
<b>Key Study Sponsor Indicator:</b>	Weight of Evidence		
<b>Year Study Performed:</b>			
<b>Method/Guideline Followed:</b>			
<b>Deviations from Method/Guideline:</b>			

<b>Method/Guideline Description:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	
<b>GLP:</b>	
<b>Study Reference:</b>	Harris, J.C. (1982) Rate of Hydrolysis. In Handbook of Chemical Property Estimation Methods. p. 7-6. W. J. Lyman, W.F. Reehl and D.H. Rosenblatt, eds. McGraw-Hill Book Company, New York, NY, USA.
<b>Reliability/Data Quality - Stability In Water</b>	
<b>Reliability:</b>	
<b>Reliability Remarks:</b>	



## Stability in Water

### Test Substance - Stability In Water

**Category Chemical:** (64741-46-4) Naphtha, petroleum, light straight-run

**Test Substance:** (64741-46-4) Naphtha, petroleum, light straight-run

**Test Substance Purity/Composition and Other Test Substance Comments:**

LSRN -Low Naphthenic, CONCAWE sample CWE39  
 Substance type: Petroleum product  
 Physical status: Liquid  
 Remark: The naphtha streams that are rich in naphthenes are obtained from the atmospheric distillation of crude oil. The streams contain saturated and aromatic hydrocarbons, mainly in the range C4 to C10.

The naphthenic naphthas typically are composed of the following hydrocarbon classes:

Approx. Content (volume %)

Paraffins 72

Olefins <0.1

Naphthenics 21

Aromatics 7

Low naphthenic content

CONCAWE sample CWE3

CAS No. 64741-46-4

Density (g/ml @ 16°C) 0.6662

Sulfur (ppm) 83

Detailed hydrocarbon analysis (Method ASTM D 5134-92)

	Olefins		Naphthenes		Aromatics		Paraffins	
	n-	i-	n-	i-	n-	i-	n-	i-
Total%	1.04	12.23	3.27	48.19	34.02	0.00	0.006	0.000
C4	0.00	0.00	0.00	31.91	8.228	0.00	0.263	0.324
C5	0.085	4.047	0.00	16.139	23.917	0.00	0.162	0.178
C6	0.830	6.696	2.252	0.647	1.241	0.382	0.334	0.263
C7	0.119	1.056	0.382	0.263	0.324	0.334	0.263	0.324
C8	0.00	0.303	0.334	0.263	0.324	0.334	0.263	0.324
C9	0.00	0.165	0.243	0.162	0.178	0.243	0.162	0.178

Substance is in the Gasoline Blending Streams Category.  
 See Category Analysis Document(s) at  
<http://www.petroleumhpv.org>

**Category Chemical Result Type:**

**Test Substance Result Type:**

### Results - Stability In Water

**Stability in Water Result Description:**

**Stability in Water Value/Range:**

**pH Value:**

**Hydrolysis Indicator:**

**Preliminary Test:**

**Effect:**

Half-Life	@ pH Value
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**Breakdown Products Description:**

**Results Remarks:** Hydrolysis unlikely

Hydrolysis is unlikely for gasoline and blending streams. Hydrolysis of an organic chemical is the transformation process in which a water molecule or hydroxide ion reacts to form a new carbon-oxygen bond. Chemicals that have a potential to hydrolyze include alkylhalides, amides, carbamates, carboxylic acid esters and lactones, epoxides, phosphate esters, and sulfonic acid esters (Harris, 1982b). The chemical components that comprise the gasoline blending streams category are hydrocarbons, which are not included in these chemical groups, and they are not subject to hydrolysis reactions with water.

### Study/Method - Stability In Water

**Key Study Sponsor Indicator:**

Key

**Year Study Performed:**

**Method/Guideline Followed:**

**Deviations from Method/Guideline:**

**Method/Guideline Description:**

**Method/Guideline and Test Condition Remarks:**

**GLP:**

**Study Reference:**

Harris, J.C. (1982) Rate of Hydrolysis. In Handbook of Chemical Property Estimation Methods. p. 7-6. W. J. Lyman, W.F. Reehl and D.H. Rosenblatt, eds. McGraw-Hill Book Company, New York, NY, USA.

### Reliability/Data Quality - Stability In Water

**Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

(1) Valid without restriction



## Stability in Water

### Test Substance - Stability In Water

**Category Chemical:** (68955-35-1) Naphtha, petroleum, catalytic reformed

**Test Substance:** (68955-35-1) Naphtha, petroleum, catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:**

Full -Range Catalytically Reformed Naphtha (FRCRN) -CAS No. 68955-35-1; API sample 83-05.

AROMATIC NAPHTHAS

Substance type: Petroleum product

Physical status: Liquid

Remark: Aromatic naphtha streams are obtained from the catalytic reforming of mainly n-alkane and cycloparaffinic feedstocks into aromatic and branched chain hydrocarbons. The hydrocarbons are mainly in the range C5 to C12. A typical aromatic naphtha is composed of the following hydrocarbon classes in the approximate proportions shown:

Content (volume %)

Paraffins 32

Olefins 0.5

Naphthenics 4

Aromatics 63.5

Full range catalytically reformed naphtha (CAS 64741-66-8) is a typical aromatic naphtha stream and the American Petroleum Institute (API, 1987) have characterized a specific sample (API 83-05) of a Full range catalytic reformed naphtha. The results of this characterization can be found in the analytical data report at the website below.

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:**

**Test Substance Result Type:**

### Results - Stability In Water

**Stability in Water Result Description:**

**Stability in Water Value/Range:**

**pH Value:**

**Hydrolysis Indicator:**

**Preliminary Test:**

**Effect:**

Half-Life	@ pH Value
-----------	------------

**Breakdown Products Description:**

**Results Remarks:** Hydrolysis unlikely

Hydrolysis is unlikely for gasoline and blending streams. Hydrolysis of an organic chemical is the transformation process in which a water molecule or hydroxide ion reacts to form a new carbon-oxygen bond. Chemicals that have a potential to hydrolyze include alkylhalides, amides, carbamates, carboxylic acid esters and lactones, epoxides, phosphate esters, and sulfonic

acid esters (Harris, 1982b). The chemical components that comprise the gasoline blending streams category are hydrocarbons, which are not included in these chemical groups, and they are not subject to hydrolysis reactions with water.

### Study/Method - Stability In Water

**Key Study Sponsor Indicator:**

Key

**Year Study Performed:**

**Method/Guideline Followed:**

**Deviations from Method/Guideline:**

**Method/Guideline Description:**

**Method/Guideline and Test Condition Remarks:**

**GLP:**

**Study Reference:**

Harris, J.C. (1982) Rate of Hydrolysis. In Handbook of Chemical Property Estimation Methods. p. 7-6. W. J. Lyman, W.F. Reehl and D.H. Rosenblatt, eds. McGraw-Hill Book Company, New York, NY, USA.

### Reliability/Data Quality - Stability In Water

**Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

(1) Valid without restriction



## Stability in Water

### Test Substance - Stability In Water

**Category Chemical:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance Purity/Composition and Other Test Substance Comments:**

Light alkylate naphtha  
 Paraffinnic naphtha  
 Substance type : Petroleum product  
 Physical status : Liquid  
 Remark: Paraffinnic naphtha streams are obtained by alkylation (catalytic reaction), isomerisation (catalytic conversion) and solvent extraction. They contain mostly saturated hydrocarbons, generally in the range C5 to C10 and boil in the range of approximately 90 to 160°C. The paraffinnic naphthas typically are composed of the following hydrocarbon classes:

Content (volume %)	
Paraffins	99.4
Olefins	0
Naphthenics	0.6
Aromatics	0

Light Alkylate Naphtha (CAS 64741-66-8) is a typical paraffinnic naphtha stream.  
 The American Petroleum Institute have reported a thorough characterization of a specific sample (API 83-19) of Light Alkylate Naphtha (LAN). The results of this characterization can be found in the analytical data report at the website listed below.

Substance is in the Gasoline Blending Streams Category.  
 See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:**

**Test Substance Result Type:**

### Results - Stability In Water

**Stability in Water Result Description:**

**Stability in Water Value/Range:**

**pH Value:**

**Hydrolysis Indicator:**

**Preliminary Test:**

**Effect:**

Half-Life	@ pH Value
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**Breakdown Products Description:**

**Results Remarks:** Hydrolysis unlikely

Hydrolysis is unlikely for gasoline and blending streams. Hydrolysis of an organic chemical is the transformation process in which a water molecule or hydroxide ion reacts to form a new carbon-oxygen bond. Chemicals that have a potential to hydrolyze include alkylhalides, amides, carbamates, carboxylic acid esters and lactones, epoxides, phosphate esters, and sulfonic



acid esters (Harris, 1982b). The chemical components that comprise the gasoline blending streams category are hydrocarbons, which are not included in these chemical groups, and they are not subject to hydrolysis reactions with water.

### Study/Method - Stability In Water

**Key Study Sponsor Indicator:**

Key

**Year Study Performed:**

**Method/Guideline Followed:**

**Deviations from Method/Guideline:**

**Method/Guideline Description:**

**Method/Guideline and Test Condition Remarks:**

**GLP:**

**Study Reference:**

Harris, J.C. (1982) Rate of Hydrolysis. In Handbook of Chemical Property Estimation Methods. p. 7-6. W. J. Lyman, W.F. Reehl and D.H. Rosenblatt, eds. McGraw-Hill Book Company, New York, NY, USA.

### Reliability/Data Quality - Stability In Water

**Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

(1) Valid without restriction



## Stability in Water

### Test Substance - Stability In Water

**Category Chemical:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance Purity/Composition and Other Test Substance Comments:**

Substance type : Petroleum product  
 Physical status : Liquid  
 Remark : The naphtha streams obtained from the catalytic cracking of heavy distillates into lighter fractions contain saturated, olefinic and aromatic hydrocarbons. However, their olefins content is higher than any of the naphtha streams derived by other processes. The catalytically cracked naphthas contain hydrocarbons in the range C4 to C10. The catalytically cracked naphthas typically are composed of the following hydrocarbon classes:  
 Approx. Content (volume %)  
 Paraffins 30  
 Olefins 46  
 Naphthenics 10  
 Aromatics 14  
 Light catalytically cracked naphtha (LCCN) (CAS 64741-55-5) is a typical olefinic naphtha stream. The American Petroleum Institute have reported (API, 1987) a thorough characterization of a specific sample of a light catalytically cracked naphtha (API 83-20), which has a high olefinic content and which was used in many of the mammalian toxicity studies. The characterization of this sample can be found in the analytical data report at the website below.

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:**

**Test Substance Result Type:**

### Results - Stability In Water

**Stability in Water Result Description:**

**Stability in Water Value/Range:**

**pH Value:**

**Hydrolysis Indicator:**

**Preliminary Test:**

**Effect:**

Half-Life	@ pH Value
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**Breakdown Products Description:**

**Results Remarks:** Hydrolysis unlikely

Hydrolysis is unlikely for gasoline and blending streams. Hydrolysis of an organic chemical is the transformation process in which a water molecule or hydroxide ion reacts to form a new carbon-oxygen bond. Chemicals that have a potential to hydrolyze include

alkylhalides, amides, carbamates, carboxylic acid esters and lactones, epoxides, phosphate esters, and sulfonic acid esters (Harris, 1982b). The chemical components that comprise the gasoline blending streams category are hydrocarbons, which are not included in these chemical groups, and they are not subject to hydrolysis reactions with water.

### Study/Method - Stability In Water

**Key Study Sponsor Indicator:**

Key

**Year Study Performed:**

**Method/Guideline Followed:**

**Deviations from Method/Guideline:**

**Method/Guideline Description:**

**Method/Guideline and Test Condition Remarks:**

**GLP:**

**Study Reference:**

Harris, J.C. (1982) Rate of Hydrolysis. In Handbook of Chemical Property Estimation Methods. p. 7-6. W. J. Lyman, W.F. Reehl and D.H. Rosenblatt, eds. McGraw-Hill Book Company, New York, NY, USA.

### Reliability/Data Quality - Stability In Water

**Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

(1) Valid without restriction



## Stability in Water

### Test Substance - Stability In Water

**Category Chemical:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:**

Light Catalytically Reformed Naphtha.  
 AROMATIC NAPHTHAS  
 Substance type: Petroleum product  
 Physical status: Liquid  
 Remark: Aromatic naphtha streams are obtained from the catalytic reforming of mainly n-alkane and cycloparaffinic feedstocks into aromatic and branched chain hydrocarbons. The hydrocarbons are mainly in the range C5 to C12. A typical aromatic naphtha is composed of the following hydrocarbon classes in the approximate proportions shown:  
 Content (volume %)  
 Paraffins 32  
 Olefins 0.5  
 Naphthenics 4  
 Aromatics 63.5  
 Full range catalytically reformed naphtha (CAS 64741-63-5) is a typical aromatic naphtha stream and the American Petroleum Institute (API, 1987) has characterized a specific sample (API 83-04) of a Full range catalytic reformed naphtha. (see website below for analytical report)

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:**

**Test Substance Result Type:**

### Results - Stability In Water

**Stability in Water Result Description:**

**Stability in Water Value/Range:**

**pH Value:**

**Hydrolysis Indicator:**

**Preliminary Test:**

**Effect:**

Half-Life	@ pH Value
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**Breakdown Products Description:**

**Results Remarks:** Hydrolysis unlikely

Hydrolysis is unlikely for gasoline and blending streams. Hydrolysis of an organic chemical is the transformation process in which a water molecule or hydroxide ion reacts to form a new carbon-oxygen bond. Chemicals that have a potential to hydrolyze include alkylhalides, amides, carbamates, carboxylic acid esters and lactones, epoxides, phosphate esters, and sulfonic

acid esters (Harris, 1982b). The chemical components that comprise the gasoline blending streams category are hydrocarbons, which are not included in these chemical groups, and they are not subject to hydrolysis reactions with water.

### Study/Method - Stability In Water

**Key Study Sponsor Indicator:**

Key

**Year Study Performed:**

**Method/Guideline Followed:**

**Deviations from Method/Guideline:**

**Method/Guideline Description:**

**Method/Guideline and Test Condition Remarks:**

**GLP:**

**Study Reference:**

Harris, J.C. (1982) Rate of Hydrolysis. In Handbook of Chemical Property Estimation Methods. p. 7-6. W. J. Lyman, W.F. Reehl and D.H. Rosenblatt, eds. McGraw-Hill Book Company, New York, NY, USA.

### Reliability/Data Quality - Stability In Water

**Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

(1) Valid without restriction



<b>Stability in Water</b>	
<b>Test Substance - Stability In Water</b>	
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Gasoline CONCAWE sample, CWE5, Blend (match to API PS-6), CAS No. 86290-81-5  Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>  [Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]
<b>Category Chemical Result Type:</b>	
<b>Test Substance Result Type:</b>	
<b>Results - Stability In Water</b>	
<b>Stability in Water Result Description:</b>	
<b>Stability in Water Value/Range:</b>	
<b>pH Value:</b>	
<b>Hydrolysis Indicator:</b>	
<b>Preliminary Test:</b>	
<b>Effect:</b>	Half-Life @ pH Value
<b>Breakdown Products Description:</b>	
<b>Results Remarks:</b>	Hydrolysis unlikely  Hydrolysis is unlikely for gasoline and blending streams. Hydrolysis of an organic chemical is the transformation process in which a water molecule or hydroxide ion reacts to form a new carbon-oxygen bond. Chemicals that have a potential to hydrolyze include alkylhalides, amides, carbamates, carboxylic acid esters and lactones, epoxides, phosphate esters, and sulfonic acid esters (Harris, 1982b). The chemical components that comprise the gasoline blending streams category are hydrocarbons, which are not included in these chemical groups, and they are not subject to hydrolysis reactions with water.
<b>Study/Method - Stability In Water</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	

<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	
<b>GLP:</b>	
<b>Study Reference:</b>	Harris, J.C. (1982) Rate of Hydrolysis. In Handbook of Chemical Property Estimation Methods. p. 7-6. W. J. Lyman, W.F. Reehl and D.H. Rosenblatt, eds. McGraw-Hill Book Company, New York, NY, USA.
<b>Reliability/Data Quality - Stability In Water</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	(1) Valid without restriction



## Transport Between Environmental Compartments Fugacity/Dist

### Test Substance - Transport Between Environmental Compartments Fugacity/Dist

**Category Chemical:** No CAS Number Provided

**Test Substance:** No CAS Number Provided

**Test Substance Purity/Composition and Other Test Substance Comments:** Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Read-Across

**Test Substance Result Type:** Estimated

### Results - Transport Between Environmental Compartments Fugacity/Dist

**Fugacity/Distribution Result Description:**

**Test Results:**

**Transport Table:**

	Emissions (kg/h)	Half-life (hr)	Mass Distribution (PERCENT)	Loss BY Reaction (PERCENT)	Loss BY Advection (PERCENT)
Air					
Water					
Soil					
Sediment					

**Temperature:**

**Level of Multi-media Model:** I

**Model Input (Water Solubility):**

**Model Input (Vapor Pressure):**

**Model Input (log K<sub>ow</sub>):**

**Model Input (Melting Point):**

**Henry's Law Constant:**

**Model Concentration -- Air:** 96.5 to 100

**Model Concentration -- Water:** 0.001 to 2.7

**Model Concentration -- Soil:** 0.00 to 1.83

**Model Concentration -- Sediment:** 0.00 to 0.03



<b>Results Remarks:</b>	The constituents of this complex petroleum mixture are expected to partition primarily to air, where these hydrocarbons will be rapidly oxidized by OH radicals.
<b>Study/Method - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Key Study Sponsor Indicator:</b>	Weight of Evidence
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	Read across ranges were calculated according to Mackay Level I  Model based on chemical fugacity. Physical properties input are those calculated by the EPIWIN Estimation 3.04 program.
<b>GLP:</b>	
<b>Study Reference:</b>	Mackay, D, A. DiGuardo, S. Paterson, & C. Cowan (1997) EQC Model, ver. 1.01, 1997, available from the Environmental Modelling Centre, Trent University, Canada.
<b>Reliability/Data Quality - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated values from studies used to determine read across range were calculated using a validated computer model



**Transport Between Environmental Compartments  
Fugacity/Dist**

**Test Substance - Transport Between Environmental Compartments  
Fugacity/Dist**

**Category Chemical:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance Purity/Composition and Other Test Substance Comments:**

Olefinic naphtha  
 Substance type : Petroleum product  
 Physical status : Liquid  
 Remark : The naphtha streams obtained from the catalytic cracking of heavy distillates into lighter fractions contain saturated, olefinic and aromatic hydrocarbons. However, their olefins content is higher than any of the naphtha streams derived by other processes. The catalytically cracked naphthas contain hydrocarbons in the range C4 to C10. The catalytically cracked naphthas typically are composed of the following hydrocarbon classes:  
 Approx. Content (volume %)  
 Paraffins 30  
 Olefins 46  
 Naphthenics 10  
 Aromatics 14  
 Light catalytically cracked naphtha (LCCN) (CAS 64741-55-5) is a typical olefinic naphtha stream. The American Petroleum Institute have reported (API, 1987) a thorough characterization of a specific sample of a light catalytically cracked naphtha (API 83-20), which has a high olefinic content and which was used in many of the mammalian toxicity studies. The characterization of this sample is provided in the Analytical Data Report at the website below.  
  
 Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Estimated by Calculation

**Test Substance Result Type:** Estimated

**Results - Transport Between Environmental Compartments  
Fugacity/Dist**

**Fugacity/Distribution Result Description:** Multimedia (Fugacity) Modeling

**Test Results:** Media: Soil, air, water, suspended sediment and sediment

Medium	% distribution
Air	97 to 100
Soil	0.00 to 1.2
Water	0.01 to 2.7
Sediment	<0.001 to 0.02
Suspended sediment	<0.001 to 0.02

**Transport Table:**

	Emissions (kg/h)	Half-life (hr)	Mass Distribution (PERCENT)	Loss BY Reaction (PERCENT)	Loss BY Advection (PERCENT)
<b>Air</b>					
<b>Water</b>					
<b>Soil</b>					
<b>Sediment</b>					

<b>Temperature:</b>	
<b>Level of Multi-media Model:</b>	I
<b>Model Input (Water Solubility):</b>	
<b>Model Input (Vapor Pressure):</b>	
<b>Model Input (log K<sub>ow</sub>):</b>	
<b>Model Input (Melting Point):</b>	
<b>Henry's Law Constant:</b>	
<b>Model Concentration -- Air:</b>	97 to 100
<b>Model Concentration -- Water:</b>	0.01 to 2.7
<b>Model Concentration -- Soil:</b>	0.00 to 1.2
<b>Model Concentration -- Sediment:</b>	<0.001 to 0.02
<b>Results Remarks:</b>	This complex petroleum mixture is expected to partition primarily to air.
<b>Study/Method - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	Other
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Type: Calculated according to Mackay Level 1
<b>Method/Guideline and Test Condition Remarks:</b>	Model based on chemical fugacity. Physical properties input are those calculated by the EPIWIN Estimation 3.04 program and presented in the corresponding physical endpoint Robust Study Summary. Values represent the spread of calculated values for C5 to C9 hydrocarbon components found in LCCN, CAS No 64741-55-5. Detailed hydrocarbon analysis performed by Chevron Research was used to identify the components of this specific LCCN sample. The majority of LCCN components will partition rapidly to air, where these hydrocarbons will be rapidly oxidized by OH radicals and ozone.
<b>GLP:</b>	
<b>Study Reference:</b>	Mackay, D, A. DiGuardo, S. Paterson, & C. Cowan (1997) EQC Model, ver. 1.01, 1997, available from the Environmental Modelling Centre, Trent University, Canada.
<b>Reliability/Data Quality - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Reliability:</b>	Valid with Restrictions

**Reliability Remarks:**

(2) Valid with restrictions

RELIABILITY: Estimated values were calculated using a validated computer model



**Transport Between Environmental Compartments  
Fugacity/Dist**

**Test Substance - Transport Between Environmental Compartments  
Fugacity/Dist**

**Category Chemical:** (68955-35-1) Naphtha, petroleum, catalytic reformed

**Test Substance:** (68955-35-1) Naphtha, petroleum, catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:** Full -Range Catalytically Reformed Naphtha (FRCRN) -CAS No. 68955-35-1; API sample 83-05.  
 AROMATIC NAPHTHAS  
 Substance type: Petroleum product  
 Physical status: Liquid  
 Remark: Aromatic naphtha streams are obtained from the catalytic reforming of mainly n-alkane and cycloparaffinic feedstocks into aromatic and branched chain hydrocarbons. The hydrocarbons are mainly in the range C5 to C12. A typical aromatic naphtha is composed of the following hydrocarbon classes in the approximate proportions shown:  
 Content (volume %)  
 Paraffins 32  
 Olefins 0.5  
 Naphthenics 4  
 Aromatics 63.5  
 Full range catalytically reformed naphtha (CAS 64741-66-8) is a typical aromatic naphtha stream and the American Petroleum Institute (API, 1987) have characterized a specific sample (API 83-05) of a Full range catalytic reformed naphtha. The results of this characterization are provided in the analytical data report at the website below.  
  
 Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Estimated by Calculation

**Test Substance Result Type:** Estimated

**Results - Transport Between Environmental Compartments  
Fugacity/Dist**

**Fugacity/Distribution Result Description:** Multimedia (Fugacity) Modeling

**Test Results:** Media: Soil, air, water, suspended sediment and sediment

Medium	% distribution
Air	96.5 to 99.98
Soil	0.01 to 1.83
Water	0.01 to 2.7
Sediment	<0.001 to 0.03
Suspended sediment	<0.001

**Transport Table:**

	Emissions (kg/h)	Half-life (hr)	Mass Distribution (PERCENT)	Loss BY Reaction (PERCENT)	Loss BY Advection (PERCENT)
<b>Air</b>					
<b>Water</b>					
<b>Soil</b>					
<b>Sediment</b>					

**Temperature:**

<b>Level of Multi-media Model:</b>	I
<b>Model Input (Water Solubility):</b>	
<b>Model Input (Vapor Pressure):</b>	
<b>Model Input (log K<sub>ow</sub>):</b>	
<b>Model Input (Melting Point):</b>	
<b>Henry's Law Constant:</b>	
<b>Model Concentration -- Air:</b>	96.5 to 99.98
<b>Model Concentration -- Water:</b>	0.01 to 2.7
<b>Model Concentration -- Soil:</b>	0.01 to 1.83
<b>Model Concentration -- Sediment:</b>	<0.001 to 0.03
<b>Results Remarks:</b>	The constituents of this complex petroleum mixture are expected to partition primarily to air.
<b>Study/Method - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	Other
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Calculated according to Mackay Level I Type: Calculated according to Mackay Level 1
<b>Method/Guideline and Test Condition Remarks:</b>	Model based on chemical fugacity. Physical properties input are those calculated by the EPIWIN Estimation 3.04 program and presented in the corresponding physical endpoint Robust Study Summary. Values represent the spread of calculated values for C5 to C8 hydrocarbon components found in LCRN, CAS No 64741-63-5. Detailed hydrocarbon analysis performed by Chevron Research was used to identify the components of this specific LCRN sample (see section 1.1.1.). The majority of LCRN components will partition rapidly to air, where these hydrocarbons will be rapidly oxidized by OH radicals.
<b>GLP:</b>	
<b>Study Reference:</b>	Mackay, D, A. DiGuardo, S. Paterson, & C. Cowan (1997) EQC Model, ver. 1.01, 1997, available from the Environmental Modelling Centre, Trent University, Canada.
<b>Reliability/Data Quality - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	

(2) Valid with restrictions  
RELIABILITY: Estimated values were calculated using a  
validated computer model



## Transport Between Environmental Compartments Fugacity/Dist

### Test Substance - Transport Between Environmental Compartments Fugacity/Dist

**Category Chemical:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:**

AROMATIC NAPHTHAS  
Substance type: Petroleum product  
Physical status: Liquid  
Remark: Aromatic naphtha streams are obtained from the catalytic reforming of mainly n-alkane and cycloparaffinic feedstocks into aromatic and branched chain hydrocarbons. The hydrocarbons are mainly in the range C5 to C12. A typical aromatic naphtha is composed of the following hydrocarbon classes in the approximate proportions shown:  
Content (volume %)  
Paraffins 32  
Olefins 0.5  
Naphthenics 4  
Aromatics 63.5  
Light Catalytically Reformed Naphtha  
Sample identified by Chevron Research as a light catalytically reformed naphtha CAS No. 64741-63-5  
Detailed hydrocarbon analysis

	Olefins	Naphthenes	Aromatics	Paraffins
				total n-
total%	0.90	2.36	39.40	57.34 17.51
C4	0.00	0.00	0.00	0.81 0.78
C5	0.34	0.26	0.00	19.45 8.05
C6	0.27	0.62	8.37	16.23 4.69
C7	0.28	1.18	29.77	17.70 3.59
C8	0.01	0.27	1.26	3.12 0.40

Substance is in the Gasoline Blending Streams Category.  
See Category Analysis Document(s) at  
<http://www.petroleumhpv.org>

**Category Chemical Result Type:** Estimated by Calculation

**Test Substance Result Type:** Estimated

### Results - Transport Between Environmental Compartments Fugacity/Dist

**Fugacity/Distribution Result Description:** Multimedia (Fugacity) Modeling

**Test Results:** Media: Soil, air, water, suspended sediment and sediment

Medium	% distribution
Air	97 to 99.98
Soil	0.01 to 0.8
Water	0.01 to 2.7
Sediment	0.00
Suspended sediment	0.00

**Transport Table:**

	Emissions (kg/h)	Half-life (hr)	Mass Distribution (PERCENT)	Loss BY Reaction (PERCENT)	Loss BY Advection (PERCENT)
<b>Air</b>					
<b>Water</b>					
<b>Soil</b>					



<b>Sediment</b>	
<b>Temperature:</b>	
<b>Level of Multi-media Model:</b>	I
<b>Model Input (Water Solubility):</b>	
<b>Model Input (Vapor Pressure):</b>	
<b>Model Input (log K<sub>ow</sub>):</b>	
<b>Model Input (Melting Point):</b>	
<b>Henry's Law Constant:</b>	
<b>Model Concentration -- Air:</b>	97 to 99.98
<b>Model Concentration -- Water:</b>	0.01 to 2.7
<b>Model Concentration -- Soil:</b>	0.01 to 0.8
<b>Model Concentration -- Sediment:</b>	0.00
<b>Results Remarks:</b>	The constituents of this complex petroleum mixture are expected to partition primarily to air.
<b>Study/Method - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	Other
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Calculated according to Mackay Level I Type: Calculated according to Mackay Level 1
<b>Method/Guideline and Test Condition Remarks:</b>	Model based on chemical fugacity. Physical properties input are those calculated by the EPIWIN Estimation 3.04 program and presented in the corresponding physical endpoint Robust Study Summary. Values represent the spread of calculated values for C5 to C8 hydrocarbon components found in LCRN, CAS No 64741-63-5. Detailed hydrocarbon analysis performed by Chevron Research was used to identify the components of this specific LCRN sample (see section 1.1.1.). The majority of LCRN components will partition rapidly to air, where these hydrocarbons will be rapidly oxidized by OH radicals.
<b>GLP:</b>	
<b>Study Reference:</b>	Mackay, D, A. DiGuardo, S. Paterson, & C. Cowan (1997) EQC Model, ver. 1.01, 1997, available from the Environmental Modelling Centre, Trent University, Canada.
<b>Reliability/Data Quality - Transport Between Environmental Compartments Fugacity/Dist</b>	

<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated values were calculated using a validated computer model



## Transport Between Environmental Compartments Fugacity/Dist

### Test Substance - Transport Between Environmental Compartments Fugacity/Dist

**Category Chemical:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance  
Purity/Composition  
and Other Test  
Substance  
Comments:**

Substance type : Petroleum product  
Physical status : Liquid  
Remark: Paraffinic naphtha streams are obtained by alkylation (catalytic reaction), isomerisation (catalytic conversion) and solvent extraction. They contain mostly saturated hydrocarbons, generally in the range C5 to C10 and boil in the range of approximately 90 to 160°C. The paraffinic naphthas typically are composed of the following hydrocarbon classes:

Content (volume %)	
Paraffins	99.4
Olefins	0
Naphthenics	0.6
Aromatics	0

Light Alkylate Naphtha (CAS 64741-66-8) is a typical paraffinic naphtha stream.

The American Petroleum Institute have reported a thorough characterization of a specific sample (API 83-19) of Light Alkylate Naphtha (LAN). The results of this characterization are found in the Analytical Data Report at the website below.

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical  
Result Type:** Estimated by Calculation

**Test Substance  
Result Type:** Estimated

### Results - Transport Between Environmental Compartments Fugacity/Dist

**Fugacity/Distribution  
Result Description:** Multimedia (Fugacity) Modeling

**Test Results:** Media: Soil, air, water, suspended sediment and sediment

Medium	% distribution
Air	99.4 to 100
Soil	0.01 to 0.27
Water	0.001 to 0.02
Sediment	<0.001
Suspended sediment	

**Transport Table:**

	Emissions (kg/h)	Half- life (hr)	Mass Distribution (PERCENT)	Loss BY Reaction (PERCENT)	Loss BY Advection (PERCENT)
<b>Air</b>					
<b>Water</b>					
<b>Soil</b>					
<b>Sediment</b>					

**Temperature:**

**Level of Multi-media  
Model:** I

<b>Model Input (Water Solubility):</b>	
<b>Model Input (Vapor Pressure):</b>	
<b>Model Input (log K<sub>ow</sub>):</b>	
<b>Model Input (Melting Point):</b>	
<b>Henry's Law Constant:</b>	
<b>Model Concentration -- Air:</b>	99.4 to 100
<b>Model Concentration -- Water:</b>	0.001 to 0.02
<b>Model Concentration -- Soil:</b>	0.01 to 0.27
<b>Model Concentration -- Sediment:</b>	<0.001
<b>Results Remarks:</b>	<p>This complex petroleum mixture is expected to partition primarily to air.</p> <p>Mobility in the aquatic and terrestrial environment is low due to low water solubility and high vapor pressure. The naphtha components will partition rapidly to air, where for the majority of these hydrocarbons will be rapidly oxidized by OH radicals.</p>
<b>Study/Method - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	Other
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Type: Calculated according to Mackay Level 1
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Model based on chemical fugacity. Multimedia distribution was calculated for the C5 to C9 hydrocarbon components found in LAN, CAS No. 64741-66-8. Detailed hydrocarbon analysis performed by Chevron Research was used to identify the components of this specific LAN sample.</p>
<b>GLP:</b>	
<b>Study Reference:</b>	Mackay, D, A. DiGuardo, S. Paterson, & C. Cowan (1997) EQC Model, ver. 1.01, 1997, available from the Environmental Modelling Centre, Trent University, Canada.
<b>Reliability/Data Quality - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	<p>(2) Valid with restrictions</p> <p>RELIABILITY: Estimated values were calculated using a validated computer model</p>



## Transport Between Environmental Compartments Fugacity/Dist

### Test Substance - Transport Between Environmental Compartments Fugacity/Dist

**Category Chemical:** (64741-46-4) Naphtha, petroleum, light straight-run

**Test Substance:** (64741-46-4) Naphtha, petroleum, light straight-run

**Test Substance Purity/Composition and Other Test Substance Comments:** Light straight-run naphtha (LSRN) - High (33.9%) naphthenic, CAS No. 64741-46-4

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Estimated by Calculation

**Test Substance Result Type:** Estimated

### Results - Transport Between Environmental Compartments Fugacity/Dist

**Fugacity/Distribution Result Description:** Multimedia (Fugacity) Modeling

**Test Results:** Media: Soil, air, water, suspended sediment, sediment

Medium % distribution  
Air: 97 to 99.97  
Soil: 0.03 to 1.2  
Water: 0.008 to 2.7  
Sediment 0.00 to 0.02  
Suspended sediment 0.00

**Transport Table:**

	Emissions (kg/h)	Half- life (hr)	Mass Distribution (PERCENT)	Loss BY Reaction (PERCENT)	Loss BY Advection (PERCENT)
<b>Air</b>					
<b>Water</b>					
<b>Soil</b>					
<b>Sediment</b>					

**Temperature:**

**Level of Multi-media Model:** I

**Model Input (Water Solubility):**

**Model Input (Vapor Pressure):**

**Model Input (log K<sub>ow</sub>):**

**Model Input (Melting Point):**

**Henry's Law Constant:**

97 to 99.97

<b>Model Concentration -- Air:</b>	
<b>Model Concentration -- Water:</b>	0.008 to 2.7
<b>Model Concentration -- Soil:</b>	0.03 to 1.2
<b>Model Concentration -- Sediment:</b>	0.00 to 0.02
<b>Results Remarks:</b>	The constituents of this complex petroleum mixture are expected to partition primarily to air.
<b>Study/Method - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	Other
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Calculated according to Mackay Level 1
<b>Method/Guideline and Test Condition Remarks:</b>	Model based on chemical fugacity. Physical properties input are those calculated by the EPIWIN Estimation 3.04 program and presented in the corresponding physical endpoint Robust Study Summary. Values represent the spread of calculated values for C5 to C9 hydrocarbon components found in LSRN, CAS No 64741-46-4. Detailed hydrocarbon analysis was used to identify the components of this specific high naphthenic LSRN sample. The majority of LSRN components will partition rapidly to air, where these hydrocarbons will be rapidly oxidized by OH radicals.
<b>GLP:</b>	
<b>Study Reference:</b>	CONCAWE (1995) Physico-chemical characterization of gasoline samples, study no. 104990C. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995. Mackay, D, A. DiGuardo, S. Paterson, & C. Cowan (1997) EQC Model, ver. 1.01, 1997, available from the Environmental Modelling Centre, Trent University, Canada.
<b>Reliability/Data Quality - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated values were calculated using a validated computer model



## Transport Between Environmental Compartments Fugacity/Dist

### Test Substance - Transport Between Environmental Compartments Fugacity/Dist

**Category Chemical:** (86290-81-5) Antiknock Gasoline

**Test Substance:** (86290-81-5) Antiknock Gasoline

**Test Substance Purity/Composition and Other Test Substance Comments:** Gasoline CONCAWE sample, CWE5, Blend (match to API PS-6)

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]

**Category Chemical Result Type:** Estimated by Calculation

**Test Substance Result Type:** Estimated

### Results - Transport Between Environmental Compartments Fugacity/Dist

**Fugacity/Distribution Result Description:** Multimedia (Fugacity) Modeling

**Test Results:** Media: Soil, air, water, suspended sediment, sediment

Medium	% distribution
Air:	97 to 99.99
Soil:	0.00 to 1.2
Water:	0.003 to 2.7
Sediment	<0.001 to 0.02
Suspended sediment	<0.001 to 0.02

**Transport Table:**

	Emissions (kg/h)	Half-life (hr)	Mass Distribution (PERCENT)	Loss BY Reaction (PERCENT)	Loss BY Advection (PERCENT)
<b>Air</b>					
<b>Water</b>					
<b>Soil</b>					
<b>Sediment</b>					

**Temperature:**

**Level of Multi-media Model:** I

**Model Input (Water Solubility):**

**Model Input (Vapor Pressure):**

**Model Input (log K<sub>ow</sub>):**

**Model Input (Melting Point):**

<b>Henry's Law Constant:</b>	
<b>Model Concentration -- Air:</b>	97 to 99.99 %
<b>Model Concentration -- Water:</b>	0.003 to 2.7
<b>Model Concentration -- Soil:</b>	0.00 to 1.2 %
<b>Model Concentration -- Sediment:</b>	<0.001 to 0.02
<b>Results Remarks:</b>	The constituents of this complex petroleum mixture are expected to partition primarily to air. Moderate partitioning to water and soil is predicted for the aromatic components of this mixture
<b>Study/Method - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	Other
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	Calculated according to Mackay Level I
<b>Method/Guideline and Test Condition Remarks:</b>	Model based on chemical fugacity. Physical properties input are those calculated by the EPIWIN Estimation 3.04 program and presented in the corresponding physical endpoint Robust Study Summary. Values represent the spread of calculated values for C5 to C8 hydrocarbon components found in gasoline. Detailed hydrocarbon analysis was used to identify the components of this specific gasoline sample. The majority of components in gasoline will partition rapidly to air, where these hydrocarbons will be rapidly oxidized by OH radicals. With the exception of toluene, partitioning to air is > 97% for all components.
<b>GLP:</b>	
<b>Study Reference:</b>	CONCAWE (1995) Physico-chemical characterization of gasoline samples, study no. 104990C. Study conducted by Exxon Biomedical Sciences Inc. Concauwe, Brussels, 1995. Mackay, D, A. DiGuardo, S. Paterson, & C. Cowan (1997) EQC Model, ver. 1.01, 1997, available from the Environmental Modelling Centre, Trent University, Canada.
<b>Reliability/Data Quality - Transport Between Environmental Compartments Fugacity/Dist</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	(2) Valid with restrictions RELIABILITY: Estimated values were calculated using a validated computer model





<b>Biodegradation</b>																	
<b>Test Substance - Biodegradation</b>																	
<b>Category Chemical:</b>	No CAS Number Provided																
<b>Test Substance:</b>	No CAS Number Provided																
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>																
<b>Category Chemical Result Type:</b>	Read-Across																
<b>Test Substance Result Type:</b>	Measured																
<b>Results - Biodegradation</b>																	
<b>Biodegradability Indicator:</b>	Inherently Biodegradable																
<b>Effect:</b>	<table border="1"> <thead> <tr> <th>Concentration Value</th> <th>Time in Days</th> <th>Biodegradation Value</th> <th>Biodegradation Value Range</th> </tr> </thead> <tbody> <tr> <td></td> <td>28</td> <td></td> <td>42 - 96</td> </tr> <tr> <td></td> <td>42</td> <td></td> <td>48 - 97</td> </tr> <tr> <td></td> <td>56</td> <td></td> <td>40 - 85</td> </tr> </tbody> </table>	Concentration Value	Time in Days	Biodegradation Value	Biodegradation Value Range		28		42 - 96		42		48 - 97		56		40 - 85
Concentration Value	Time in Days	Biodegradation Value	Biodegradation Value Range														
	28		42 - 96														
	42		48 - 97														
	56		40 - 85														
<b>Half Life:</b>																	
<b>Rate Constant:</b>																	
<b>Temperature:</b>																	
<b>Incubation Condition:</b>																	
<b>Inoculum Type:</b>																	
<b>Inoculum Concentration:</b>																	
<b>Inoculum Remarks:</b>																	
<b>Pre-Exposure Indicator:</b>																	
<b>Pre-Exposure Remarks:</b>																	
<b>Theoretical Carbon DiOxide:</b>																	
<b>Theoretical Oxygen Demand:</b>																	
<b>Chemical Oxygen Demand:</b>																	
<b>Control Substance Remarks:</b>																	
<b>Breakdown Products Description:</b>																	
<b>Results Remarks:</b>																	

<b>Study/Method - Biodegradation</b>	
<b>Key Study Sponsor Indicator:</b>	Weight of Evidence
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Method/Guideline Description:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	
<b>GLP:</b>	
<b>Study Reference:</b>	see CAS # 64741-66-8, 64741-55-5, 64741-63-5, 64741-41-9, and 86290-81-5
<b>Reliability/Data Quality - Biodegradation</b>	
<b>Reliability:</b>	
<b>Reliability Remarks:</b>	Studies used to develop read across ranges were either "reliable without restrictions" (reliability =1) or "reliable with restrictions" (reliability = 2)



## Biodegradation

### Test Substance - Biodegradation

**Category Chemical:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance Purity/Composition and Other Test Substance Comments:** Olefinic naphtha  
 Substance type : Petroleum product  
 Physical status : Liquid  
 Remark : The naphtha streams obtained from the catalytic cracking of heavy distillates into lighter fractions contain saturated, olefinic and aromatic hydrocarbons. However, their olefins content is higher than any of the naphtha streams derived by other processes. The catalytically cracked naphthas contain hydrocarbons in the range C4 to C10. The catalytically cracked naphthas typically are composed of the following hydrocarbon classes:

	Approx. Content (volume %)
Paraffins	30
Olefins	46
Naphthenics	10
Aromatics	14

Light catalytically cracked naphtha (LCCN) (CAS 64741-55-5) is a typical olefinic naphtha stream. The American Petroleum Institute have reported (API, 1987) a thorough characterization of a specific sample of a light catalytically cracked naphtha (API 83-20), which has a high olefinic content and which was used in many of the mammalian toxicity studies. The characterization of this sample can be found in the analytical data report at the website below.

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:** Measured

**Test Substance Result Type:** Measured

### Results - Biodegradation

**Biodegradability Indicator:** Inherently Biodegradable

**Effect:**

Concentration Value	Time in Days	Biodegradation Value	Biodegradation Value Range
	28	= 74 % Degradation	
	42	= 75 % Degradation	
	56	= 79 % Degradation	

**Half Life:**

**Rate Constant:**

**Temperature:**

**Incubation Condition:** Aerobic

<b>Inoculum Type:</b>	Other																																	
<b>Inoculum Concentration:</b>																																		
<b>Inoculum Remarks:</b>	Mixed, adapted inoculum of domestic activated sludge and soil Contact time: 56 day(s)																																	
<b>Pre-Exposure Indicator:</b>																																		
<b>Pre-Exposure Remarks:</b>																																		
<b>Theoretical Carbon DiOxide:</b>																																		
<b>Theoretical Oxygen Demand:</b>																																		
<b>Chemical Oxygen Demand:</b>																																		
<b>Control Substance Remarks:</b>																																		
<b>Breakdown Products Description:</b>																																		
<b>Results Remarks:</b>	<p>Test material was inherently biodegradable since it achieved &gt;20% biodegradability based on CO<sub>2</sub> production. By day 28 approximately 74% of the test material was degraded, then essentially reached a plateau in degradation rate until day 56. The test was considered valid according to CONCAWE criteria, as &gt;60% biodegradation of positive control (63% actual) was observed by day 14, and total blank CO<sub>2</sub> production at termination was less than 15% of the organic carbon added as test substance.</p> <p>Temperature ranged from 18 to 21 °C, which deviated from the protocol value of 22 ±2°C. This deviation was not expected to have affected the outcome of this study.</p> <table border="1"> <thead> <tr> <th colspan="3">% Degradation (sd)</th> </tr> <tr> <th>Day</th> <th>Test Hexadecane</th> <th>Test Material</th> </tr> </thead> <tbody> <tr> <td>3</td> <td>13.93 (1.85)</td> <td>16.83 (9.56)</td> </tr> <tr> <td>7</td> <td>34.40 (4.54)</td> <td>30.99 (0.56)</td> </tr> <tr> <td>14</td> <td>63.17 (0.94)</td> <td>51.66 (3.33)</td> </tr> <tr> <td>21</td> <td>77.26 (6.52)</td> <td>54.82 (6.24)</td> </tr> <tr> <td>28</td> <td>90.35 (7.14)</td> <td>74.30 (1.24)</td> </tr> <tr> <td>35</td> <td>85.13 (n=1)</td> <td>65.02 (1.37)</td> </tr> <tr> <td>42</td> <td>85.21 (n=1)</td> <td>74.82 (0.54)</td> </tr> <tr> <td>49</td> <td>96.93 (8.94)</td> <td>70.78 (6.48)</td> </tr> <tr> <td>56</td> <td>94.69 (4.10)</td> <td>79.22 (12.28)</td> </tr> </tbody> </table>	% Degradation (sd)			Day	Test Hexadecane	Test Material	3	13.93 (1.85)	16.83 (9.56)	7	34.40 (4.54)	30.99 (0.56)	14	63.17 (0.94)	51.66 (3.33)	21	77.26 (6.52)	54.82 (6.24)	28	90.35 (7.14)	74.30 (1.24)	35	85.13 (n=1)	65.02 (1.37)	42	85.21 (n=1)	74.82 (0.54)	49	96.93 (8.94)	70.78 (6.48)	56	94.69 (4.10)	79.22 (12.28)
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<b>Study/Method - Biodegradation</b>																																		
<b>Key Study Sponsor Indicator:</b>	Key																																	
<b>Year Study Performed:</b>	1999																																	
<b>Method/Guideline Followed:</b>	Other																																	
<b>Deviations from Method/Guideline:</b>																																		
<b>Method/Guideline Description:</b>	<p>CONCAWE test method for determining the inherent aerobic biodegradability of oil products. 1996/1997, and modification of ISO/DIS 14593</p> <p>Type (test type): Water quality-Evaluation of ultimate</p>																																	

aerobic biodegradability of organic compounds in aqueous medium-Method by analysis of inorganic carbon in sealed vessels (CO<sub>2</sub> headspace test)

**Method/Guideline  
and  
Test Condition  
Remarks:**

Mixed inoculum prepared from soil and activated sludge was incubated with test substance or hexadecane (positive control) during a two week adaptation period. Triplicate test systems were incubated for both the test substance and hexadecane fed inoculum. Two additional, similar test substances were concurrently incubated in separate 160 ml test systems using the same inoculum and acclimation procedure. Duplicate blank control test systems were prepared which consisted of the mixed inocula in mineral medium but no test or positive control substance. Test medium consisted of glass distilled water and mineral salts (phosphate buffer, ferric chloride, magnesium sulfate, calcium chloride) prepared as described in ISO method.

Acclimation procedure-Activated sludge from aeration basin of Wareham Wastewater Treatment Plant (Mass., U.S.A.) was sieved through 2 mm and centrifuged at 1000 rpm for 10 minutes. After removal of supernatant the concentrated solids were diluted to 5 mg/ml suspended solids with reagent grade water. Soil was collected from a site located in a mixed hardwood and pine forest (Mass., U.S.A.). Site of sampling was cleared of debris and approximately 500 g of soil was obtained at a depth between 5-10 cm from the soil surface. Soil was air-dried, sieved through a 2 mm sieve, and analyzed for moisture content (38%).

Test vessels (160 ml serum bottles) were filled with 103 ml of mineral medium containing 50 mg/l of yeast extract and 50 mg/l (dry weight) washed activated sludge, then approximately 0.16g of sieved soil (0.1 g dry wt) was added to each bottle. Test or reference substance were added directly to test systems using a 10 microliter Hamilton gas-tight syringe. The volume required to achieve the specified mg carbon/l concentrations were calculated based on % carbon and specific gravity of the respective substance. The test substance % carbon (0.8724) and specific gravity (0.7220 mg/ul) information was supplied by the Sponsor. Hexadecane % carbon (0.8496) was calculated from the empirical formula and specific gravity (0.7749 mg/ul) was obtained from Verschuere (1983). Addition of respective substance was performed on an incremental basis to the appropriate vessels as follows: 4, 8 and 8 mg C/l were added on days 0, 7 and 11, respectively. Test vessels were sealed with butyl rubber septa/aluminum crimp caps and incubated at 22 (±2°C) in the dark.

Biodegradation by CO<sub>2</sub> determination-test initiation and procedure. On day 14 of the acclimation phase, all test system inoculum from blanks, positive control, and each of the three test substances was combined and filtered through glass wool, and aerated prior to use. The aerated mixed inoculum was then added to mineral medium to achieve 10% concentration based on total volume (100 ml inoculum/l). Test vessels (160 ml serum bottles) were filled with 103 ml of inoculated mineral medium. Respective test systems were dosed with either test substance or hexadecane as described for the acclimation procedure to achieve 20 mg carbon/l concentration.

Duplicate test systems for each test substance, positive control and blank treatments were prepared for sacrifice at weekly sampling intervals for subsequent CO<sub>2</sub> analysis. After test system preparation, all vessels were placed in a walk-in chamber and incubated in the dark at 22°C (±2°).

On days 3, 7, 14, 21, 28, 35, 42, 49 and 56, 1ml of conc. H<sub>3</sub>PO<sub>4</sub> was injected through the septum of each sacrificed test vessel. The acidified samples were shaken for 1 hr at 200 ppm, then analyzed for CO<sub>2</sub> using gas chromatography-thermal conductivity detection. Quantitation of inorganic mg C/l evolved was determined by linear regression analysis based on response factors for sodium carbonate standards spanning 1-30 mg carbon/l concentrations.

**GLP:**

Yes

**Study Reference:** Springborn Laboratories, Inc. (1999) Light Catalytically Cracked Naphtha-Determination of Inherent Biodegradability. Study No. 13687.6109

**Reliability/Data Quality - Biodegradation**

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** (1) Valid without restriction  
RELIABILITY: GLP study with adequately detailed methods description



<b>Biodegradation</b>																	
<b>Test Substance - Biodegradation</b>																	
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate																
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate																
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Paraffinic naphtha            Substance type : Petroleum product            Physical status : Liquid            Remark: Paraffinic naphtha streams are obtained by alkylation (catalytic reaction), isomerisation (catalytic conversion) and solvent extraction. They contain mostly saturated hydrocarbons, generally in the range C5 to C10 and boil in the range of approximately 90 to 160°C. The paraffinic naphthas typically are composed of the following hydrocarbon classes:</p> <table border="1" style="margin-left: 40px;"> <thead> <tr> <th></th> <th style="text-align: center;">Content (volume %)</th> </tr> </thead> <tbody> <tr> <td>Paraffins</td> <td style="text-align: center;">99.4</td> </tr> <tr> <td>Olefins</td> <td style="text-align: center;">0</td> </tr> <tr> <td>Naphthenics</td> <td style="text-align: center;">0.6</td> </tr> <tr> <td>Aromatics</td> <td style="text-align: center;">0</td> </tr> </tbody> </table> <p>Light Alkylate Naphtha (CAS 64741-66-8) is a typical paraffinic naphtha stream. The American Petroleum Institute have reported a thorough characterization of a specific sample (API 83-19) of Light Alkylate Naphtha (LAN). The results of this characterization can be found in the analytical data report at the website below.</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>		Content (volume %)	Paraffins	99.4	Olefins	0	Naphthenics	0.6	Aromatics	0						
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<b>Test Substance Result Type:</b>	Measured																
<b>Results - Biodegradation</b>																	
<b>Biodegradability Indicator:</b>	Inherently Biodegradable																
<b>Effect:</b>	<table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th style="width: 25%;">Concentration Value</th> <th style="width: 15%;">Time in Days</th> <th style="width: 25%;">Biodegradation Value</th> <th style="width: 35%;">Biodegradation Value Range</th> </tr> </thead> <tbody> <tr> <td></td> <td style="text-align: center;">28</td> <td style="text-align: center;">= 42 % Degradation</td> <td></td> </tr> <tr> <td></td> <td style="text-align: center;">42</td> <td style="text-align: center;">= 48 % Degradation</td> <td></td> </tr> <tr> <td></td> <td style="text-align: center;">56</td> <td style="text-align: center;">= 40 % Degradation</td> <td></td> </tr> </tbody> </table>	Concentration Value	Time in Days	Biodegradation Value	Biodegradation Value Range		28	= 42 % Degradation			42	= 48 % Degradation			56	= 40 % Degradation	
Concentration Value	Time in Days	Biodegradation Value	Biodegradation Value Range														
	28	= 42 % Degradation															
	42	= 48 % Degradation															
	56	= 40 % Degradation															
<b>Half Life:</b>																	
<b>Rate Constant:</b>																	
<b>Temperature:</b>																	
<b>Incubation Condition:</b>	Aerobic																
<b>Inoculum Type:</b>	Other																

<b>Inoculum Concentration:</b>																																					
<b>Inoculum Remarks:</b>	Mixed, adapted inoculum of domestic activated sludge and soil Contact time: 56 day(s)																																				
<b>Pre-Exposure Indicator:</b>																																					
<b>Pre-Exposure Remarks:</b>																																					
<b>Theoretical Carbon DiOxide:</b>																																					
<b>Theoretical Oxygen Demand:</b>																																					
<b>Chemical Oxygen Demand:</b>																																					
<b>Control Substance Remarks:</b>																																					
<b>Breakdown Products Description:</b>																																					
<b>Results Remarks:</b>	<p>Test material was inherently biodegradable since it achieved &gt;20% biodegradability based on CO<sub>2</sub> production. By day 21 approximately 40% of the test material was degraded, a slight increase to 48% was observed by day 42, but by day 56 degradation had leveled back down to 40%. The test was considered valid according to CONCAWE criteria, as &gt;60% biodegradation of positive control (63% actual) was observed by day 14, and total blank CO<sub>2</sub> production at termination was less than 15% of the organic carbon added as test substance.</p> <p>Temperature ranged from 18 to 21 °C, which deviated from the protocol value of 22 ±2°C. This deviation was not expected to have affected the outcome of this study.</p> <table border="1"> <thead> <tr> <th colspan="3">% Degradation (sd)</th> </tr> <tr> <th>Test</th> <th>Hexadecane</th> <th>Test Material</th> </tr> </thead> <tbody> <tr> <td>Day</td> <td></td> <td></td> </tr> <tr> <td>3</td> <td>13.93 (1.85)</td> <td>0.12 (0.07)</td> </tr> <tr> <td>7</td> <td>34.40 (4.54)</td> <td>7.84 (7.80)</td> </tr> <tr> <td>14</td> <td>63.17 (0.94)</td> <td>26.59 (0.85)</td> </tr> <tr> <td>21</td> <td>77.26 (6.52)</td> <td>40.24 (5.00)</td> </tr> <tr> <td>28</td> <td>90.35 (7.14)</td> <td>42.41 (2.54)</td> </tr> <tr> <td>35</td> <td>85.13 (n=1)</td> <td>41.53 (9.90)</td> </tr> <tr> <td>42</td> <td>85.21 (n=1)</td> <td>48.12 (1.77)</td> </tr> <tr> <td>49</td> <td>96.93 (8.94)</td> <td>46.55 (1.04)</td> </tr> <tr> <td>56</td> <td>94.69 (4.10)</td> <td>40.44 (0.76)</td> </tr> </tbody> </table>	% Degradation (sd)			Test	Hexadecane	Test Material	Day			3	13.93 (1.85)	0.12 (0.07)	7	34.40 (4.54)	7.84 (7.80)	14	63.17 (0.94)	26.59 (0.85)	21	77.26 (6.52)	40.24 (5.00)	28	90.35 (7.14)	42.41 (2.54)	35	85.13 (n=1)	41.53 (9.90)	42	85.21 (n=1)	48.12 (1.77)	49	96.93 (8.94)	46.55 (1.04)	56	94.69 (4.10)	40.44 (0.76)
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Method by analysis of inorganic carbon in sealed vessels  
(CO<sub>2</sub> headspace test)

**Method/Guideline  
and  
Test Condition  
Remarks:**

Mixed inoculum prepared from soil and activated sludge was incubated with test substance or hexadecane (positive control) during a two week adaptation period. Triplicate test systems were incubated for both the test substance and hexadecane fed inoculum. Two additional, similar test substances were concurrently incubated in separate 160 ml test systems using the same inoculum and acclimation procedure. Duplicate blank control test systems were prepared which consisted of the mixed inocula in mineral medium but no test or positive control substance. Test medium consisted of glass distilled water and mineral salts (phosphate buffer, ferric chloride, magnesium sulfate, calcium chloride) prepared as described in ISO method.

Acclimation procedure-Activated sludge from aeration basin of Wareham Wastewater Treatment Plant (Mass., U.S.A.) was sieved through 2 mm and centrifuged at 1000 rpm for 10 minutes. After removal of supernatant the concentrated solids were diluted to 5 mg/ml suspended solids with reagent grade water. Soil was collected from a site located in a mixed hardwood and pine forest (Mass., U.S.A.). Site of sampling was cleared of debris and approximately 500 g of soil was obtained at a depth between 5-10 cm from the soil surface. Soil was air-dried, sieved through a 2 mm sieve, and analyzed for moisture content (38%).

Test vessels (160 ml serum bottles) were filled with 103 ml of mineral medium containing 50 mg/l of yeast extract and 50 mg/l (dry weight) washed activated sludge, then approximately 0.16g of sieved soil (0.1 g dry wt) was added to each bottle. Test or reference substance were added directly to test systems using a 10 microliter Hamilton gas-tight syringe. The volume required to achieve the specified mg carbon/l concentrations were calculated based on % carbon and specific gravity of the respective substance. The test substance % carbon (0.8505) and specific gravity (0.6690 mg/il) information was supplied by the Sponsor. Hexadecane % carbon (0.8496) was calculated from the empirical formula and specific gravity (0.7749 mg/il) was obtained from Verschuere (1983). Addition of respective substance was performed on an incremental basis to the appropriate vessels as follows: 4, 8 and 8 mg C/l were added on days 0, 7 and 11, respectively. Test vessels were sealed with butyl rubber septa/aluminum crimp caps and incubated at 22 (±2°C) in the dark.

Biodegradation by CO<sub>2</sub> determination-test initiation and procedure On day 14 of the acclimation phase, all test system inoculum from blanks, positive control, and each of the three test substances was combined and filtered through glass wool, and aerated prior to use. The aerated mixed inoculum was then added to mineral medium to achieve 10% concentration based on total volume (100 ml inoculum/l). Test vessels (160 ml serum bottles) were filled with 103 ml of inoculated mineral medium. Respective test systems were dosed with either test substance or hexadecane as described for the acclimation procedure to achieve 20 mg carbon/l concentration. Duplicate test systems for each test substance, positive control and blank treatments were prepared for sacrifice at weekly sampling intervals for subsequent CO<sub>2</sub> analysis. After test system preparation, all vessels were placed in a walk-in chamber and incubated in the dark at 22°C (±2°).

On days 3, 7, 14, 21, 28, 35, 42, 49 and 56, 1ml of conc. H<sub>3</sub>PO<sub>4</sub> was injected through the septum of each sacrificed test vessel. The acidified samples were shaken for 1 hr at 200 ppm, then analyzed for CO<sub>2</sub> using gas chromatography-thermal conductivity detection. Quantitation of inorganic mg C/l evolved was determined by linear regression analysis based on response factors for sodium carbonate standards spanning 1-30 mg carbon/l concentrations.

**GLP:** Yes

**Study Reference:**

Springborn Laboratories, Inc. (1999)  
Light Alkylate Naphtha-Determination of Inherent  
Biodegradability. Study No. 13687.6111

### Reliability/Data Quality - Biodegradation

<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	(1) Valid without restriction RELIABILITY: GLP study with adequately detailed methods description



## Biodegradation

### Test Substance - Biodegradation

**Category Chemical:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:** Aromatic naphtha  
Light Catalytically Reformed Naphtha.  
Sample identified by Chevron Research as a light catalytically reformed naphtha CAS No. 64741-63-5  
Detailed hydrocarbon analysis

	Olefins	Naphthenes	Aromatics	Paraffins total	n-
total%	0.90	2.36	39.40	57.34	17.51
C4	0.00	0.00	0.00	0.81	0.78
C5	0.34	0.26	0.00	19.45	8.05
C6	0.27	0.62	8.37	16.23	4.69
C7	0.28	1.18	29.77	17.70	3.59
C8	0.01	0.27	1.26	3.12	0.40

Substance is in the Gasoline Blending Streams Category.  
See Category Analysis Document(s) at  
<http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

**Test Substance Result Type:** Measured

### Results - Biodegradation

**Biodegradability Indicator:** Inherently Biodegradable

**Effect:**

Concentration Value	Time in Days	Biodegradation Value	Biodegradation Value Range
	28	= 96 % Degradation	
	42	= 97 % Degradation	
	56	= 85 % Degradation	

**Half Life:**

**Rate Constant:**

**Temperature:**

**Incubation Condition:** Aerobic

**Inoculum Type:** Other

**Inoculum Concentration:**

**Inoculum Remarks:** Mixed, adapted inoculum of domestic activated sludge and soil  
Contact time: 56 day(s)

**Pre-Exposure Indicator:**

<b>Pre-Exposure Remarks:</b>																															
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<b>Control Substance Remarks:</b>	Hexadecane was used as positive control and the blank control test systems consisted of the mixed inocula in mineral medium but no test or positive control substance.																														
<b>Breakdown Products Description:</b>																															
<b>Results Remarks:</b>	<p>Test material was inherently biodegradable since it achieved &gt;20% biodegradability based on CO<sub>2</sub> production. By day 28 approximately 96% of the test material was degraded, then essentially reached a plateau in degradation rate until day 56. The test was considered valid according to CONCAWE criteria, as &gt;60% biodegradation of positive control (63% actual) was observed by day 14, and total blank CO<sub>2</sub> production at termination was less than 15% of the organic carbon added as test substance.</p> <p>Temperature ranged from 18 to 21° C, which deviated from the protocol value of 22 ± 2°C. This deviation was not expected to have affected the outcome of this study.</p> <p>% Degradation (sd)</p> <table border="1"> <thead> <tr> <th>Test Day</th> <th>Hexadecane</th> <th>Test Material</th> </tr> </thead> <tbody> <tr> <td>3</td> <td>13.93 (1.85)</td> <td>30.85 (3.85)</td> </tr> <tr> <td>7</td> <td>34.40 (4.54)</td> <td>53.71 (3.52)</td> </tr> <tr> <td>14</td> <td>63.17 (0.94)</td> <td>77.25 (3.65)</td> </tr> <tr> <td>21</td> <td>77.26 (6.52)</td> <td>87.17 (8.87)</td> </tr> <tr> <td>28</td> <td>90.35 (7.14)</td> <td>96.17 (5.26)</td> </tr> <tr> <td>35</td> <td>85.13 (n=1)</td> <td>107.9 (n=1)</td> </tr> <tr> <td>42</td> <td>85.21 (n=1)</td> <td>96.95 (6.37)</td> </tr> <tr> <td>49</td> <td>96.93 (8.94)</td> <td>92.02 (n=1)</td> </tr> <tr> <td>56</td> <td>94.69 (4.10)</td> <td>84.92 (0.51)</td> </tr> </tbody> </table>	Test Day	Hexadecane	Test Material	3	13.93 (1.85)	30.85 (3.85)	7	34.40 (4.54)	53.71 (3.52)	14	63.17 (0.94)	77.25 (3.65)	21	77.26 (6.52)	87.17 (8.87)	28	90.35 (7.14)	96.17 (5.26)	35	85.13 (n=1)	107.9 (n=1)	42	85.21 (n=1)	96.95 (6.37)	49	96.93 (8.94)	92.02 (n=1)	56	94.69 (4.10)	84.92 (0.51)
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Biodegradation by CO<sub>2</sub> determination test initiation and procedure On day 14 of the acclimation phase, all test system inoculum from blanks, positive control, and each of the three test substances was combined and filtered through glass wool, and aerated prior to use. The aerated mixed inoculum was then added to mineral medium to achieve 10% concentration based on total volume (100 ml inoculum/l).

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**GLP:** Yes

**Study Reference:** Springborn Laboratories, Inc. (1999) Light Catalytically Reformed Naphtha-Determination of Inherent Biodegradability. Study No. 13687.6110

### Reliability/Data Quality - Biodegradation

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** (1) Valid without restriction  
RELIABILITY: GLP study with adequately detailed methods description



## Biodegradation

### Test Substance - Biodegradation

**Category Chemical:**

(86290-81-5) Antiknock Gasoline

**Test Substance:**

(86290-81-5) Antiknock Gasoline

**Test Substance Purity/Composition and Other Test Substance Comments:**

CAS No. 86290-81-5; a commercial unleaded gasoline topped at 76°C by distillation. It was free of hydrocarbons having less than six carbon atoms and contained no oxygenated compounds.

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]

**Category Chemical Result Type:**

Measured

**Test Substance Result Type:**

Measured

### Results - Biodegradation

**Biodegradability Indicator:**

Inherently Biodegradable

**Effect:**

Concentration Value	Time in Days	Biodegradation Value	Biodegradation Value Range
	25	94 % Degradation	

**Half Life:**
**Rate Constant:**
**Temperature:**
**Incubation Condition:**

Aerobic

**Inoculum Type:**

Other

**Inoculum Concentration:**
**Inoculum Remarks:**

Activated aerobic sludge obtained from an urban wastewater treatment plant.

**Pre-Exposure Indicator:**
**Pre-Exposure Remarks:**
**Theoretical Carbon DiOxide:**
**Theoretical Oxygen Demand:**
**Chemical Oxygen Demand:**

**Control Substance  
Remarks:****Breakdown  
Products  
Description:****Results Remarks:**

Biodegradation and Mineralization of Gasoline Gasoline was degraded up to 94% under non-limiting conditions after 25 d incubation (500 ml substrate/1 medium). The carbon balance of gasoline degradation showed that 61.7% of gasoline was mineralized to CO<sub>2</sub> and that microbial cell production accounted for the remaining carbon of gasoline degraded. Biomass formation and mineralization occurred mainly during the initial fast degradation phase whereas essentially mineralization occurred during the second slow degradation phase. Individual classes of hydrocarbons degraded and carbon balance were shown to be:

Hydrocarbon Class	Gasoline (mg/g)	After 2 days	After 25 days
Aromatics	789	88%	99%
Branched alkanes	165	14	74
Linear alkanes	23	17	92
Cyclic alkanes	17	10	99
Alkenes	6	71	99

Substrate or Products (mg C/l)	Initial amount (mg C/l)	Final amount (mg C/l)
Gasoline	357	18
Biomass	39	165
CO <sub>2</sub>	0	204
Total Carbon	396	387

Kinetic Experiments with Gasoline Two main degradation phases were found, one fast degradation phase (FDP), which started after an 18 h lag period and lasted until the 40th hour. The maximum rate of oxygen consumption during the FDP was 44 mg/l/h and the average rate was 24.5 mg/l/h. The FDP was followed by a slow degradation phase (SDP) where the rate of oxygen consumption slowed steadily from the 40th hour till the 25th day. The average rate was 15 mg/l/d, which was approximately 40 times slower than during the FDP.

Activated sludge microorganisms were found to biodegrade unleaded commercial gasoline up to 94% within 25 days. For each hydrocarbon class, degradation occurred at different rates. Aromatic compounds were found to be the most readily consumed, although compounds bearing neighboring substituents and those containing longer alkyl groups were consumed at a slower rate than those with no or only one alkyl chain. Likewise, linear alkanes (exception for undecane), alkenes with five to nine carbons, cyclohexane and substituted cyclopentanes were biodegraded. Residual components of gasoline most recalcitrant to biodegradation were found to be branched alkanes, particularly those containing a quaternary carbon and/or alkyl chains on consecutive carbon atoms.

**Study/Method - Biodegradation****Key Study Sponsor Indicator:**

Key

**Year Study Performed:**

1999

**Method/Guideline Followed:**

Other

**Deviations from Method/Guideline:****Method/Guideline Description:**

Non-guideline research method using a closed-system shake flask apparatus.  
Aerobic Biodegradability -Evaluation of biodegradability of

gasoline in aqueous medium. Method by analysis of disappearance of carbon compounds (gas chromatography with flame ionization detector), kinetics of O<sub>2</sub> consumption (respirometry), and CO<sub>2</sub> production (gas chromatography with thermal conductivity detector).

Exposure period was 16 or 25 days. See test conditions for more details.

**Method/Guideline and Test Condition Remarks:**

Activated sludge containing approximately 3 g/l dry weight was centrifuged at 15000 g for 20 min and re-suspending the biomass in the same volume of nutrient solution. The microbial suspension was used to inoculate nutrient solution at a final concentration of 100 mg dry weight/l. Gasoline (400 mg/l) or individual hydrocarbons (150 mg/l) were added to the medium as the sole carbon source. The nutrient solution was a vitamin-enriched mineral salt medium described by Bouchez et al. Appl. Microbiol. Biotechnol. 43:156-164 (1995).

**Biodegradation of Gasoline** The biodegradation tests were performed in 500-ml flasks with sidearms equipped with Mininert® valves. 25 ml of gasoline were added to 50 ml of inoculated nutrient medium (i.e., 500 ml substrate/1 medium) through the valve with a syringe. The flasks were incubated for 25 days at 30°C with alternate shaking (70 strokes per min). After the incubation period, 5 ml of CH<sub>2</sub>Cl<sub>2</sub> containing 600 mg/ml dodecane as internal standard was introduced to the flasks through the valve, and the remaining hydrocarbon compounds were extracted for 1 h under shaking. The flasks were refrigerated overnight at 4°C before opening. The suspensions were centrifuged at 35000 g for 30 min at 4°C. The CH<sub>2</sub>Cl<sub>2</sub> phase of each flask was then analyzed by gas chromatography for carbon compounds. Experiments were performed in duplicate and abiotic controls were prepared similarly to the other treatments with the exception that 1 g/l HgCl<sub>2</sub> were added to the flasks before incubation.

**Mineralization of Gasoline** Measurements of CO<sub>2</sub> evolved during the biodegradation of gasoline were conducted in 240-ml flasks closed by Viton® stoppers. 18 ml of inoculated culture medium were added to each flask along with 5 ml of gasoline (i.e., 500 ml substrate/1 medium). Flasks were incubated at 30°C for 25 days under alternate shaking. At the end of the incubation period the contents of each flask was acidified with 0.5 ml HNO<sub>3</sub> (68%) and CO<sub>2</sub> was measured by gas chromatography. Endogenous respiration of inoculated medium was measured in flasks without gasoline added.

**Kinetic Experiments with Gasoline** Kinetics of O<sub>2</sub> consumption during gasoline biodegradation were determined in duplicate at 30°C over 25 d by respirometry. 500 ml stirred culture flasks contained 250 ml of inoculated nutrient medium and 125 ml of gasoline (i.e., 500 ml substrate/1 medium). Control experiments without gasoline were also done. Kinetics of hydrocarbon degradation also was monitored by respirometry. Incubation was stopped at selected times and the remaining hydrocarbons were extracted as described above and analyzed by gas chromatography.

**Kinetic Experiments with Individual Hydrocarbons** Kinetics of CO<sub>2</sub> production during the degradation of individual hydrocarbons was carried out at 30°C over 16 days. Treatments were prepared in 125 ml shaken flasks with 25 ml of nutrient solution containing 70 mg/l of inoculum biomass and 5 ml of hydrocarbon (i.e., 200 ml substrate/1 medium). Flasks were closed with Teflon-coated stoppers and sealed. CO<sub>2</sub> was measured at various times by gas chromatography. Endogenous respiration was determined in flasks without hydrocarbon added.

**GLP:** No Data

**Study Reference:** Solano-Serena, F., R. Marchal, M. Ropars, J.-M. Lebeault, and J.-P. Vandecasteele. 1999. Biodegradation of Gasoline:



Kinetics, Mass Balance and Fate of Individual Hydrocarbons.  
J. Appl. Microbiol. 96:1006-1016.

### Reliability/Data Quality - Biodegradation

**Reliability:** Valid with Restrictions

**Reliability** (2) Valid with restrictions  
**Remarks:** RELIABILITY: GLP status of study unknown; non-guideline  
study with adequately detailed methods description



<b>Biodegradation</b>									
<b>Test Substance - Biodegradation</b>									
<b>Category Chemical:</b>	(64741-41-9) Naphtha, petroleum, heavy straight-run								
<b>Test Substance:</b>	(64741-41-9) Naphtha, petroleum, heavy straight-run								
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>High Naphthenic, Heavy Straight-Run Naphtha:            C6 - C11 aromatics: 13.7 % wt.            C6 - C12 iso-paraffins: 31.2 % wt.            C6 - C10 naphthenes: 29.5 % wt.            C7 - C10 olefins: 5.0 % wt.            C6 - C12 paraffins: 18.9 % wt.            Unidentified: 1.7 % wt.</p> <p>Substance is in the Gasoline Blending Streams Category.            See Category Analysis Document(s) at  <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>								
<b>Category Chemical Result Type:</b>	Measured								
<b>Test Substance Result Type:</b>	Measured								
<b>Results - Biodegradation</b>									
<b>Biodegradability Indicator:</b>	Readily Biodegradable								
<b>Effect:</b>	<table border="1"> <thead> <tr> <th>Concentration Value</th> <th>Time in Days</th> <th>Biodegradation Value</th> <th>Biodegradation Value Range</th> </tr> </thead> <tbody> <tr> <td>50 mg/L</td> <td>28</td> <td>= 77 % Degradation</td> <td>75 - 78</td> </tr> </tbody> </table>	Concentration Value	Time in Days	Biodegradation Value	Biodegradation Value Range	50 mg/L	28	= 77 % Degradation	75 - 78
Concentration Value	Time in Days	Biodegradation Value	Biodegradation Value Range						
50 mg/L	28	= 77 % Degradation	75 - 78						
<b>Half Life:</b>									
<b>Rate Constant:</b>									
<b>Temperature:</b>	22 °C								
<b>Incubation Condition:</b>	Aerobic								
<b>Inoculum Type:</b>	Activated Sludge								
<b>Inoculum Concentration:</b>	10 ml/L								
<b>Inoculum Remarks:</b>	The activated sludge was obtained from a domestic wastewater treatment plant receiving predominantly domestic sewage.								
<b>Pre-Exposure Indicator:</b>	No								
<b>Pre-Exposure Remarks:</b>									
<b>Theoretical Carbon DiOxide:</b>									
<b>Theoretical Oxygen Demand:</b>	3.41 mg/mg								
<b>Chemical Oxygen Demand:</b>									

<b>Control Substance Remarks:</b>	Sodium benzoate served as the positive control substance. The THOD = 1.67 mg/mg, and was used at a concentration of 50 mg/l.
<b>Breakdown Products Description:</b>	
<b>Results Remarks:</b>	<p>The average percent biodegradation of triplicate test systems of high naphthenic, heavy, straight-run naphtha was 77% of the THOD over a 28 day test period. 10% biodegradation was attained by Day 4 and had attained 60% by Day 12. Based on these results, the test substance passed the OECD criteria for ready biodegradability.</p> <p>Biodegradation of the positive reference substance, sodium benzoate, exceeded 60% of the THOD by Day 2 and 96% by Day 28.</p>
<b>Study/Method - Biodegradation</b>	
<b>Key Study Sponsor Indicator:</b>	Key
<b>Year Study Performed:</b>	2007
<b>Method/Guideline Followed:</b>	OECD 301F
<b>Deviations from Method/Guideline:</b>	No
<b>Method/Guideline Description:</b>	<p>Aerobic Ready Biodegradability: Manometric Respirometry Test</p> <p>Triplicate respirometer flasks were used to evaluate the biodegradability of the test and positive control substances at mean concentrations of 49 mg/L and 50 mg/L, respectively. A toxicity control (combination of positive control and test substance) was also evaluated at a mean concentration of 99 mg/L. Duplicate flasks containing test medium and inoculum but no test or positive control substances served as test blanks.</p> <p>Un-acclimated activated sludge was collected the day before test initiation from the Clinton Sanitary Wastewater Treatment Plant, Annandale, NJ, USA, which receives predominantly domestic sewage. The sample was aerated for approximately 24 hours with CO<sub>2</sub>-free air. The total suspended solids (TSS) of the activated sludge measured 3.5 g/L. After the aeration period, the sludge was homogenized in a blender for two minutes then allowed to settle for one hour and fifteen minutes. The supernatant was decanted and an aliquot of the supernatant was taken for measurement of the microbial activity. The colony-forming-units (CFU) of the supernatant measured 105 CFU/mL.</p> <p>Twenty liters of mineral medium was prepared according to OECD guidelines by adding mineral salt stock solutions to glass distilled water. After adding the mineral salt solutions, the activated sludge inoculum was added at a 1% loading volume of sludge supernatant to mineral medium. The medium was aerated for approximately 24 hours with CO<sub>2</sub> free air.</p> <p>One liter of test medium was added to each one liter respirometer flask. The test substance was weighed in an air tight syringe and injected into the test medium. The syringe was re-weighed after dosing and the weight difference equaled the amount of test substance added to the flask. Flasks were sealed immediately after addition of the test substance to minimize loss of volatile components. An aliquot of the positive control stock solution was added to the appropriate test flasks.</p> <p>All respirometer flasks were placed on a Coordinated Environmental Services (CES) automated respirometer which automatically recorded the oxygen uptake in general agreement with the OECD guideline. The 28-day study was</p>

conducted at a temperature range of approximately 21 to 24° C.

The composition of the test substance was characterized as follows:

C6 - C11 aromatics: 13.7 % wt.  
 C6 - C12 iso-paraffins: 31.2 % wt.  
 C6 - C10 naphthenes: 29.5 % wt.  
 C7 - C10 olefins: 5.0 % wt.  
 C6 - C12 paraffins: 18.9 % wt.  
 Unidentified: 1.7 % wt.

An elemental analysis of the test sample resulted in the following:

% Carbon 85.54  
 % Hydrogen 14.35  
 % Nitrogen 0.08  
 % Oxygen <0.1

The Theoretical Oxygen Demand (ThOD) was determined using the elemental analyses data and OECD 301F procedures.  
 ThOD = 3.41 mg O<sub>2</sub>/mg test substance.

**Method/Guideline  
and  
Test Condition  
Remarks:**

**GLP:** Yes

**Study Reference:** ExxonMobil Biomedical Sciences, Inc. 2006. Ready Biodegradability: Manometric Respirometry test on High Naphthenic, Heavy, Straight-Run Naphtha. Study # 0545979. ExxonMobil Biomedical Sciences, Annandale, NJ.

**Reliability/Data Quality - Biodegradation**

**Reliability:** Valid Without Restrictions

**Reliability  
Remarks:** RELIABILITY: Guideline study conducted under GLP



## Acute Toxicity to Aquatic Vertebrates

### Test Substance - Acute Toxicity To Aquatic Vertebrates

**Category Chemical:** *No CAS Number Provided*

**Test Substance:** *No CAS Number Provided*

**Test Substance Purity/Composition and Other Test Substance Comments:** Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:** Read-Across

**Test Substance Result Type:**

### Method - Acute Toxicity To Aquatic Vertebrates

**Year Study Performed:**

**Method/Guideline Followed:**

**Other Method/Guideline:**

**Deviations from Method/Guideline:**

**Species:**

**GLP:**

**Analytical Monitoring:**

**Test Type:**

**Test Vessel:**

**Water Media Type:**

**Test Concentrations:**

**Nominal and Measured Concentrations:**

**Exposure Period:**

**Vehicle Used:**

**Vehicle Name:**

**Vehicle Amount and Units:**

**Alkalinity:**

**Dissolved Oxygen:**

**pH Value:**

**Test Temperature and Units:**

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline Test Conditions Remarks:** Freshwater fish

**Limit Test:**

**Test Results - Acute Toxicity To Aquatic Vertebrates**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:** 96 Hours

**NOELR:** = 3.1 - 15 mg/L Nominal

**LOELR Exposure Duration:**

**LOELR:**

**LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration
96	Hours	LL	50%	=	2.09	46	mg/L	Mortality	

**Results Remarks:** The LL50 (lethal loading rate for 50% of the test population) range of acute toxicity values that may be used as read-across for members of the category is 2.09 - 46 mg/L (lethal loading rates) based on both calculated and measured data.  
The NOELR (no observable effect loading rate) range of values was 3.1 - 15 mg/L based on measured data.

**Reliability/Data Quality - Acute Toxicity To Aquatic Vertebrates**

**Reliability:**

**Reliability Remarks:** RELIABILITY: Studies used to determine the read across ranges were rated either as 1 or 2, 'valid without restrictions' or 'valid with restrictions', respectively.

**Key Study Sponsor Indicator:** Weight of Evidence

**Reference - Acute Toxicity To Aquatic Vertebrates**

**Reference:**



## Acute Toxicity to Aquatic Vertebrates

### Test Substance - Acute Toxicity To Aquatic Vertebrates

<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Gasoline CAS No. 86290-81-5</p> <p>Gasoline Sample W94/813, Blend Detailed hydrocarbon analysis:  N-paraffins: 20% total C3-C8,  Iso-paraffins: 28% total C4-C9  Olefins: 1%, C5-C7  Naphthenes: 5% C5-C10  Aromatics: 46% C6-C9</p> <p>Substance is in the Gasoline Blending Streams Category.  See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p> <p>[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]</p>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	OECD 203
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Oncorhynchus mykiss
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Static
<b>Test Vessel:</b>	
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 1, 5, 10, 25 and 50 mg/l
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	

<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	7.4 to 9.8 ppm
<b>pH Value:</b>	7.8 Upper Range: 8.1
<b>Test Temperature and Units:</b>	Value/Lower Range: 14.1 °C
<b>Photo (Light/Dark):</b>	16/8
<b>Salinity:</b>	
<b>TOC:</b>	
<b>Water Hardness:</b>	
<b>Method/Guideline</b>	LL50 at 96 hr calculated using Probit procedure (Finney, D.J., 1971. Probit Analysis, Third Edition, London: Cambridge University Press, and SAS computer statistics software.
<b>Test Conditions</b>	
<b>Remarks:</b>	<p>Test solutions were prepared as water accommodated fractions (WAF). The control and dilution water was a laboratory blend water prepared from carbon filtered well water and ion exchange softened, reverse osmosis dialyzed well water aged for &gt;24 hrs. To determine contact time required to achieve maximum solubility between test substance and aqueous solution, samples of WAF test solutions equilibrated for 20, 24 27 and 49 hours at 100 mg/l loading were analyzed by GC-FID for concentrations of the following: benzene, toluene, ethylbenzene, xylene isomers and naphthalene (BTEXN). Maximum WAF solubility for these components reached equilibrium within 24 hrs of stirring. Nominal loading rates of 0, 1, 5, 10, 25 and 50 mg/l were used to prepare test solutions for the fish toxicity tests. Test substance, added volumetrically, was mixed for each individual treatment in dilution water for 24 hours in 20liter stoppered containers with less than 10% headspace volume. The mixtures were allowed to settle for 1-2 hours prior to drawing off the aqueous solutions for testing. Fish were approximately four weeks old at test initiation and were obtained from Thomas Fish company, Anderson, CA, Lot 297. Loading of fish body mass to treatment was 0.2 g fish per liter of aqueous solution, mean length at termination was 2.7 cm (sd=0.2), and mean weight was 0.136 g (sd=0.034). Test vessels were 4 liter glass aspirator bottles with foil covered neoprene stoppers. Three replicates per treatment and 5 organisms per replicate were tested for each treatment and the control. Exposure containers were filled (no headspace volume) and tightly sealed to prevent volatilization. Test solution renewal was performed daily by removing 80% of the test solution and replacing it with fresh WAF solution prepared at least 24 hrs pri or to use. Freshly prepared and old WAF test solutions were analyzed by GC-FID for concentrations of BTEXN. Water temperature was 14.1 °C (0.03sd). Test photoperiod was 16 hrs. light and 8 hr dark, light intensity approx 619-622 Lux during full daylight periods. Dissolved oxygen measurements ranged from 7.4 to 9.8 ppm, pH values between 7.8 and 8.1.</p>
<b>Limit Test:</b>	No
<b>Test Results - Acute Toxicity To Aquatic Vertebrates</b>	
<b>NOEC Exposure Duration:</b>	
<b>NOEC:</b>	
<b>LOEC Exposure Duration:</b>	
<b>LOEC:</b>	
<b>NOELR Exposure Duration:</b>	96 Hours
<b>NOELR:</b>	= 5 mg/L
<b>LOELR Exposure Duration:</b>	
<b>LOELR:</b>	
<b>LC/EC Mean Value:</b>	



Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration
96	Hours	LL	50 %	=	11		mg/L	Mortality	Nominal

**Results Remarks:** Mortality (no. of deaths/treatment) at 96 hrs:

Treatment	No. of deaths
0	1
1.0	0
5	0
10	7
25	15
50	5

96-hr LL50 = 11 mg/l, 95% C.I.: 8.7-16 mg/l (as nominal loading rate) 96-hour No Observed Effect Loading (NOEL) was 5 mg/l, both calculated (Dunnett's Procedure) and observed. Results are quoted in terms of 50% Lethal Loading (LL50), the loading rate of test substance resulting in 50% mortality of the test species exposed to the WAF. At termination, abnormal behavior/appearance (lethargy, erratic swimming) was observed in all surviving fish at the 10 mg/l treatment. Losses of the soluble components from the WAF over each 24 hour period ranged from 5 to 25% for the 5, 10 and 25 mg/l loadings. Up to 57% loss was observed in the 1.0 mg/l treatment in 24 hrs samples. BETXN concentrations on 24hour samples of the 50 mg/l treatments due to complete mortality on day 0 were not determined.

Analytical results Measured BTEXN (mg/l)  
Nominal loading rate (mg/l)

Day	Control	1.0	5.0	10	25	50
0 (new)	ND	0.54	2.3	4.2	9.5	20
1 (old)	ND	0.50	2.3	4.0	10	NA
1 (new)	ND	0.47	1.7	4.2	NA	NA
2 (old)	ND	0.20	2.1	4.0	NA	NA
2 (new)	ND	0.52	2.0	4.1	NA	NA
3 (old)	ND	0.25	2.0	4.3	NA	NA
3 (new)	ND	0.57	1.6	4.0	NA	NA
4 (old)	ND	0.38	1.2	3.2	NA	NA

ND=not detected, NA=not analyzed due to 100% mortality

Guideline/protocol deviations: Body length (2.7cm av.) smaller than recommended range of 4-6 cm; smaller fish used to minimize DO depletion in closed vessel (no-headspace) systems.

**Reliability/Data Quality - Acute Toxicity To Aquatic Vertebrates**

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP; guideline study

**Key Study Sponsor Indicator:** Key

**Reference - Acute Toxicity To Aquatic Vertebrates**

**Reference:** CONCAWE (1995) Fish -acute toxicity test: study no. 104858, test substance MRD-95-048. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Acute Toxicity to Aquatic Vertebrates</b>	
<b>Test Substance - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Paraffinic naphtha Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Year Study Performed:</b>	1994
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Pimephales promelas
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Semi-Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 1.1, 5.2, 9.7, 19 and 74 mg/l
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	7.7 and 8.6
<b>pH Value:</b>	7.844 Upper Range: 8.23
<b>Test Temperature and Units:</b>	Value/Lower Range: 21.2 °C

**Photo (Light/Dark):** 16/8

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline Test Conditions Remarks:** No specific guideline was described as being used to conduct the test, however report references 1991 ASTM method E729-88a and 1982 EPA Support Documents for Environmental Testing: EPA 560/6-82-002. LL 50 calculated using binomial probability analysis. ASTM Special Technical Publication 634. 1977, pp 65-84. All NOEC values calculated using Fisher's exact test.

Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water was Mobil Technical Center well water. Nominal loading rates of 0, 1.1, 5.2, 9.7, 19 and 74 mg/l were used to prepare test solutions.

WAFs were prepared for each test concentration by mixing the appropriate mass of substance in 9.4 liters of water for 24 hr in 9 liter glass bottles. The bottles were filled to neck height with water to minimize volatility. A measured amount of test substance was added into each bottle, and the bottles were capped tightly with a positive pressure siphoning apparatus. The siphoning apparatus consisted of a teflon lined neoprene stopper housing two teflon tubes. One tube extended to the bottom of the bottle for removal of the WAF solution, the other tube ended above the WAF surface, and was used to control air pressure during siphoning. During WAF preparation, parafilm was used to seal the external joint between the neoprene stopper and glass bottle. After stirring for 24 hrs using 25% or less vortex, the contents of the WAF solution bottles were allowed to settle for approximately 45 minutes to two hours, then siphoned by the positive pressure apparatus port and used for testing. Samples were also analyzed by Purge & trap/GC-FID for concentrations of the following: 2,3 dimethyl butane; 2,4 dimethyl pentane; 2,2,4 trimethyl pentane; 2,5 dimethyl hexane; 2,3,4 trimethyl pentane, 2,3,3 trimethyl pentane and 1-methyl-1-ethyl cyclopentane, which represent 68% composition of the test substance. Measured test concentrations of the light alkylate naphtha were based on the total combined concentrations of all analytes.

Fish were hatched and raised in-house, and were acclimated prior to experimentation for a minimum of 14 days on a 16/8hr light/dark cycle. Test vessels were 3.8 liter glass containers with teflon lined caps. Two replicates per treatment and 10 organisms per replicate were tested for each treatment and the control. Exposure containers were filled with no headspace and tightly sealed to prevent volatilization. Test solution renewal was performed daily by removal of most of the test solution (leaving adequate volume to avoid stressing test organisms) and replacing it with fresh WAF solution prepared at least 24 hrs prior to use.

Water temperature was 21.2 °C (0.2 °C sd).  
 Test photoperiod was 16 hrs. light and 8 hr dark.  
 Dissolved oxygen measurements were between 7.7 and 8.6,  
 pH values between 7.844 and 8.23.

**Limit Test:** No

**Test Results - Acute Toxicity To Aquatic Vertebrates**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:** 96 Hours

**NOELR:** = 5.2 mg/L Nominal

**LOELR Exposure Duration:**

**LOELR:**

<b>LC/EC Mean Value:</b>	<b>Exposure Duration</b>	<b>Exposure Units</b>	<b>LC/EC</b>	<b>%</b>	<b>Value Description</b>	<b>Mean Value</b>	<b>Units</b>	<b>Effect Observed</b>	<b>Basis for Concentration</b>
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					or Lower Mean Value	Upper Mean Value			
96	Hours	LL	50 %	=	8.2		mg/L	Mortality	Nominal
			%						

**Results Remarks:** Mortality (no. of deaths/treatment) at 96 hrs: 0, 0, 0, 15, 20 and 20, respectively in 0, 1.1, 5.2, 9.7, 19 and 74 mg/l treatments. All surviving organisms exhibited normal behavior.

96-hr LL50 = 8.2 mg/l, (5.2-9.7 mg/l w/ 95% C.I.) as nominal loading rate 96-hr LC50 = 305 ppb, (164-384 ppb w/ 95% C.I.) measured concentrations  
 96-hr NOEL = 5.2 mg/l (as nominal loading rate)  
 96-hr NOEC = 166 ppb (measured concentrations)

Measured concentrations represented the sum of seven specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, there was insufficient information regarding the analytical measurements. It was not reported how many sample measurements were taken, nor whether the reported values were based on fresh or old solutions, initial measurements, or a mean of all measurements. Additionally, it was not reported to what degree measured concentrations declined between solution renewals. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

**Reliability/Data Quality - Acute Toxicity To Aquatic Vertebrates**

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:** Key

**Reference - Acute Toxicity To Aquatic Vertebrates**

**Reference:** Stonybrook Laboratories, Inc. (1995) Static Renewal 96-hour acute toxicity of the water accommodated fraction (WAF) of Whole Light Alkylate Naphtha (LAN) Product to Fathead Minnow. Study No. 65908. Stonybrook Laboratories, Inc. Princeton, NJ.1995.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Acute Toxicity to Aquatic Vertebrates</b>	
<b>Test Substance - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Category Chemical:</b>	(64741-46-4) Naphtha, petroleum, light straight-run
<b>Test Substance:</b>	(64741-46-4) Naphtha, petroleum, light straight-run
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	LSRN-Moderate naphthenic content Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Year Study Performed:</b>	1996
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Pimephales promelas
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Semi-Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 3.1, 6.3, 13, 25, 50 mg/L
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	144 to 154 mg/l
<b>Dissolved Oxygen:</b>	7.3 and 8.8
<b>pH Value:</b>	8.1 Upper Range: 8.3
<b>Test Temperature and Units:</b>	Value/Lower Range: 21 Upper Range: 22 °C

<b>Photo (Light/Dark):</b>	16/8
<b>Salinity:</b>	
<b>TOC:</b>	
<b>Water Hardness:</b>	134 - 144 mg/L
<b>Method/Guideline Test Conditions Remarks:</b>	<p>No specific guideline was described as being used to conduct the test, however report references 1991 ASTM method E729-88a and 1982 EPA Support Documents for Environmental Testing: EPA 560/6-82-002. LL50 and LC50 calculated using binomial probability analysis. ASTM Special Technical Publication 634. 1977, pp 65-84.</p> <p>Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water was prepared by blending naturally hard well water with water that had been demineralized by reverse osmosis. Nominal loading rates of 0, 3.1, 6.3, 13, 25 and 50 mg/l were used to prepare test solutions.</p> <p>WAFs were prepared for each test concentration by mixing the appropriate mass of substance in 9.4 liters of water for 24 hr in 9 liter glass bottles. The bottles were filled to neck height with water to minimize volatility. A measured amount of test substance was added into each bottle, and the bottles were capped tightly with a positive pressure siphoning apparatus. The siphoning apparatus consisted of a teflon lined neoprene stopper housing two teflon tubes. One tube extended to the bottom of the bottle for removal of the WAF solution, the other tube ended above the WAF surface, and was used to control air pressure during siphoning. During WAF preparation, parafilm was used to seal the external joint between the neoprene stopper and glass bottle, and the bottles were covered with aluminum foil. After stirring for 24 hrs using 25% or less vortex, the contents of the WAF solution bottles were allowed to settle for approximately 45 minutes to two hours, then siphoned by the positive pressure apparatus port and used for testing. Samples were also analyzed by Purge &amp; trap/GC-FID for concentrations of the following: 2-methyl-pentane, cyclohexane, benzene, toluene, ethylbenzene, ortho, meta and para-xylene. Measured test concentrations of the light straight run naphtha were based on the total combined concentrations of all analytes.</p> <p>Fish were hatched and raised from ABC Laboratories' in-house culture, and were acclimated prior to experimentation for a minimum of 14 days on a 16/8hr light/dark cycle. Test vessels were 3.8 liter glass containers with teflon lined caps. Fish were acclimated to the test water and temperature approximately 72 hr before the test, and were not fed during this 72 hr period. Two replicates per treatment and 10 organisms per replicate were tested for each treatment and the control, with the exception of the 50 mg/l treatment, where 11 organisms instead of 10 were placed in one replicate. Exposure containers were filled with no headspace and tightly sealed to prevent volatilization. Test solution renewal was performed daily by removal of most of the test solution (leaving approximately one liter of solution to avoid stressing test organisms) and replacing it with fresh WAF solution prepared at least 24 hrs prior to use.</p> <p>Water temperature was 21-22 °C. Test photoperiod was 16 hrs. light and 8 hr dark. Dissolved oxygen measurements were between 7.3 and 8.8, pH values between 8.1 and 8.3. Hardness values ranged from 134 to 144 mg/l; alkalinity values ranged from 144 to 154 mg/l and conductivity values ranged from 300 to 340 microsiemens.</p>
<b>Limit Test:</b>	No
<b>Test Results - Acute Toxicity To Aquatic Vertebrates</b>	
<b>NOEC Exposure Duration:</b>	
<b>NOEC:</b>	
<b>LOEC Exposure Duration:</b>	
<b>LOEC:</b>	
<b>NOELR Exposure Duration:</b>	96 Hours
<b>NOELR:</b>	= 6.3 mg/L Nominal
<b>LOELR Exposure Duration:</b>	

**LOELR:****LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration
96	Hours	LL	50 %	=	15		mg/L	Mortality	Nominal
			%						

**Results Remarks:**

Mortality (no. of deaths/treatment) at 96 hrs: 0, 0, 0, 6, 20 and 21 in 0, 3.1, 6.3, 13, 25 and 50 mg/l treatments. Abnormal behavior (surfacing, erratic swimming, quiescence) was observed at 96 hrs for 6 organisms in the 13 mg/l treatment.

96-hr LL50 = 15 mg/l, 6.3-25 mg/L w/ 95% C.I. (as nominal loading rate)

96-hr LC50 = 0.689 mg/l, 0.289-0.962 mg/l w/ 95% C.I. (measured concentrations) 96-hr NOEL = 6.3 mg/l (nominal)

96-hr NOEC = 0.287 mg/l (measured) based on lack of mortality and abnormal effects for these treatments.

Measured concentrations represented the sum of six specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, there was insufficient information regarding the analytical measurements. It was not reported how many sample measurements were taken, nor whether the reported values were based on fresh or old solutions, initial measurements, or a mean of all measurements. Additionally, it was not reported to what degree measured concentrations declined between solution renewals. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

A low boiling point naphtha sample w/ CAS no. 8030-30-6 (different from the sample used in toxicity testing, but similar in composition) was used to validate the analytical method being developed to identify water soluble hydrocarbons in aqueous 24 hour equilibrated samples. This does not appear to have affected the results of the study.

**Reliability/Data Quality - Acute Toxicity To Aquatic Vertebrates****Reliability:**

Valid with Restrictions

**Reliability Remarks:**

Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAF may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:**

Key

**Reference - Acute Toxicity To Aquatic Vertebrates****Reference:**

ABC Laboratories, Inc. (1998) Static Renewal 96 Hour-Acute Toxicity of the Water Accomodated Fraction (WAF) of Light Straight Run Naphtha, LSRN to Fathead Minnow (Pimephales promelas). Project ID. 43152. Environmental Toxicology, 7200 E. ABC Lane, Columbia, Missouri.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Acute Toxicity to Aquatic Vertebrates</b>	
<b>Test Substance - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Category Chemical:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Aromatic naphtha Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Year Study Performed:</b>	1998
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Pimephales promelas
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Semi-Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 3.1, 6.3, 13, 25 and 50 mg/l
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	150-158 mg/l
<b>Dissolved Oxygen:</b>	6.0-8.5
<b>pH Value:</b>	7.7 Upper Range: 8.5
<b>Test Temperature and Units:</b>	Value/Lower Range: 21 Upper Range: 22 °C



<b>Photo (Light/Dark):</b>	16/8
<b>Salinity:</b>	
<b>TOC:</b>	
<b>Water Hardness:</b>	138 - 148 mg/L
<b>Method/Guideline Test Conditions Remarks:</b>	<p>No specific guideline was described as being used to conduct the test, however report references 1991 ASTM method E729-88a and 1982 EPA Support Documents for Environmental Testing: EPA 560/6-82-002.</p> <p>Statistical Method: (FT -ME) LL 50 and LC50 calculated using binomial probability analysis. ASTM Special Technical Publication 634. 1977, pp 65-84.</p> <p>Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water was prepared by blending naturally hard well water with water that had been demineralized by reverse osmosis. Nominal loading rates of 0, 3.1, 6.3, 13, 25 and 50 mg/l were used to prepare test solutions.</p> <p>WAFs were prepared for each test concentration by mixing the appropriate mass of substance in 9.4 liters of water for 24 hr in 9 liter glass bottles. The bottles were filled to neck height with water to minimize volatility. A measured amount of test substance was added into each bottle, and the bottles were capped tightly with a positive pressure siphoning apparatus. The siphoning apparatus consisted of a teflon lined neoprene stopper housing two teflon tubes. One tube extended to the bottom of the bottle for removal of the WAF solution, the other tube ended above the WAF surface, and was used to control air pressure during siphoning. During WAF preparation, parafilm was used to seal the external joint between the neoprene stopper and glass bottle, and the bottles were covered with aluminum foil. After stirring for 24 hrs using 25% or less vortex, the contents of the WAF solution bottles were allowed to settle for approximately 45 minutes to two hours, then siphoned by the positive pressure apparatus port and used for testing. Samples were also analyzed by Purge &amp; trap/GC-FID for concentrations of the following: pentane, 2-methyl pentane, benzene, toluene, ethylbenzene, ortho, meta and para-xylene, which represent more than 50% composition of the test substance. Measured test concentrations of the light catalytically reformed naphtha were based on the total combined concentrations of all analytes.</p> <p>Fish were hatched and raised from ABC Laboratories' in-house culture, and were acclimated prior to experimentation for a minimum of 14 days on a 16/8hr light/dark cycle. Test vessels were 3.8 liter glass containers with teflon-lined caps. Fish were acclimated to the test water and temperature approximately 72 hr before the test, and were not fed during this 72 hr period. Two replicates per treatment and 10 organisms per replicate were tested for each treatment and the control. Exposure containers were filled with no headspace and tightly sealed to prevent volatilization. Test solution renewal was performed daily by removal of most of the test solution (leaving approximately one liter of solution to avoid stressing test organisms) and replacing it with fresh WAF solution prepared at least 24 hrs prior to use.</p> <p>Water temperature was 21-22 °C. Test photoperiod was 16 hrs. light and 8 hr dark. Dissolved oxygen measurements were between 6.0 and 8.5, pH values between 7.7 and 8.5. Hardness values ranged from 138 to 148 mg/l; alkalinity values ranged from 150 to 158 mg/l and conductivity values ranged from 299 to 313 microsiemens</p>
<b>Limit Test:</b>	No
<b>Test Results - Acute Toxicity To Aquatic Vertebrates</b>	
<b>NOEC Exposure Duration:</b>	
<b>NOEC:</b>	
<b>LOEC Exposure Duration:</b>	
<b>LOEC:</b>	
<b>NOELR Exposure Duration:</b>	96 Hours
<b>NOELR:</b>	= 3.1 mg/L Nominal
<b>LOELR Exposure Duration:</b>	

**LOELR:****LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration
96	Hours	LL	50 %	=	34		mg/L	Mortality	Nominal
			%						

**Results Remarks:**

Mortality (no. of deaths/treatment) at 96 hrs: 1, 0, 1, 0, 1 and 20 in 0, 3.1, 6.3, 13, 25 and 50 mg/l treatments. Abnormal behavior (surfacing, erratic swimming) was observed at 96 hrs for 3 organisms in 13 mg/l and 7 fish in 25 mg/l treatments.

96-hr LL50 = 34 mg/l, 25-50 mg/l w/ 95% C.I. (as nominal loading rate)  
 96-hr LC50 = 11 mg/l, 8.2-17.2 mg/l w/ 95% C.I. (measured concentrations)  
 96-hr NOEL = 3.1 mg/l (nominal)  
 96-hr NOEC = 1.03 mg/l (measured) based on lack of mortality and abnormal effects for these treatments.

Measured concentrations represented the sum of six specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, there was insufficient information regarding the analytical measurements. It was not reported how many sample measurements were taken, nor whether the reported values were based on fresh or old solutions, initial measurements, or a mean of all measurements. Additionally, it was not reported to what degree measured concentrations declined between solution renewals. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

### Reliability/Data Quality - Acute Toxicity To Aquatic Vertebrates

**Reliability:**

Valid with Restrictions

**Reliability Remarks:**

Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:**

Key

### Reference - Acute Toxicity To Aquatic Vertebrates

**Reference:**

ABC Laboratories, Inc. (1998) Static Renewal 96 Hour Acute Toxicity of the Water Accomodated Fraction (WAF) of Light Catalytically Reformed Naphtha, LCRN to Fathead Minnow (*Pimephales promelas*). Project ID. 43578. Environmental Toxicology, 7200 E. ABC Lane, Columbia, Missouri.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



## Acute Toxicity to Aquatic Vertebrates

### Test Substance - Acute Toxicity To Aquatic Vertebrates

**Category Chemical:** (68955-35-1) Naphtha, petroleum, catalytic reformed

**Test Substance:** (68955-35-1) Naphtha, petroleum, catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:** Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:** Estimated by Calculation

**Test Substance Result Type:** Estimated

### Method - Acute Toxicity To Aquatic Vertebrates

**Year Study Performed:**

**Method/Guideline Followed:**

**Other Method/Guideline:**

**Deviations from Method/Guideline:**

**Species:**

**GLP:**

**Analytical Monitoring:**

**Test Type:**

**Test Vessel:**

**Water Media Type:**

**Test Concentrations:**

**Nominal and Measured Concentrations:**

**Exposure Period:** 96 Hours

**Vehicle Used:**

**Vehicle Name:**

**Vehicle Amount and Units:**

**Alkalinity:**

**Dissolved Oxygen:**

**pH Value:**

**Test Temperature and Units:**

<b>Photo (Light/Dark):</b>																					
<b>Salinity:</b>																					
<b>TOC:</b>																					
<b>Water Hardness:</b>																					
<b>Method/Guideline Test Conditions Remarks:</b>	Calculated based on hydrocarbon block principle. In this procedure, the dissolved concentrations of individual hydrocarbons from a petroleum substance are estimated for a given loading rate and then normalised by their acute toxicity to yield Toxic Units (TU) which can be summed to predict the toxicity of the parent material (see below)																				
<b>Limit Test:</b>																					
<b>Test Results - Acute Toxicity To Aquatic Vertebrates</b>																					
<b>NOEC Exposure Duration:</b>																					
<b>NOEC:</b>																					
<b>LOEC Exposure Duration:</b>																					
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<b>LC/EC Mean Value:</b>	<table border="1"> <thead> <tr> <th>Exposure Duration</th> <th>Exposure Units</th> <th>LC/EC</th> <th>%</th> <th>Value Description</th> <th>Mean Value or Lower Mean Value</th> <th>Upper Mean Value</th> <th>Units</th> <th>Effect Observed</th> <th>Basis for Concentration</th> </tr> </thead> <tbody> <tr> <td>96</td> <td>Hours</td> <td>LL</td> <td>50 %</td> <td>=</td> <td>2.09</td> <td></td> <td>mg/L</td> <td></td> <td>Calculated</td> </tr> </tbody> </table>	Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration	96	Hours	LL	50 %	=	2.09		mg/L		Calculated
Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration												
96	Hours	LL	50 %	=	2.09		mg/L		Calculated												
<b>Results Remarks:</b>	Estimated 96 hour(s) fish acute toxicity LL50: 2.09 mg/l																				
<b>Reliability/Data Quality - Acute Toxicity To Aquatic Vertebrates</b>																					
<b>Reliability:</b>	Valid with Restrictions																				
<b>Reliability Remarks:</b>	RELIABILITY: Estimated values were calculated using a computer model																				
<b>Key Study Sponsor Indicator:</b>	Key																				
<b>Reference - Acute Toxicity To Aquatic Vertebrates</b>																					
<b>Reference:</b>	Peterson, D.R., (1994) Calculating the aquatic toxicity of hydrocarbon mixtures Chemosphere Vol.29, 12, pp. 2493-2506  Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003																				



## Acute Toxicity to Aquatic Vertebrates

### Test Substance - Acute Toxicity To Aquatic Vertebrates

<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Gasoline CAS No. 86290-81-5</p> <p>Gasoline Sample W94/814, Blend; Detailed hydrocarbon analysis:  N-paraffins: 16% total C4-C8  Iso-paraffins: 25% total C4-C11  Olefins: 12%, C4-C7  Naphthenes: 5% C6-C10  Aromatics: 42% C6-C11</p> <p>Substance is in the Gasoline Blending Streams Category.  See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p> <p>[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]</p>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	OECD 203
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Oncorhynchus mykiss
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Static
<b>Test Vessel:</b>	
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 0.1, 1, 5, 10, and 25 mg/l
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	

<b>Vehicle Amount and Units:</b>																			
<b>Alkalinity:</b>																			
<b>Dissolved Oxygen:</b>	5.4 to 9.7 ppm																		
<b>pH Value:</b>	6.8 Upper Range: 8.2																		
<b>Test Temperature and Units:</b>	Value/Lower Range: 15 °C																		
<b>Photo (Light/Dark):</b>	16/8																		
<b>Salinity:</b>																			
<b>TOC:</b>																			
<b>Water Hardness:</b>																			
<b>Method/Guideline Test Conditions Remarks:</b>	<p>LL50 at 96 hr calculated using binomial probability analysis. ASTM Special Technical Publication 634. 1977, pp 65-84</p> <p>Test solutions were prepared as water accommodated fractions (WAF). The control and dilution water was a laboratory blend water prepared from carbon filtered well water and ion exchange softened, reverse osmosis dialyzed well water aged for &gt;24 hrs. To determine contact time required to achieve maximum solubility between test substance and aqueous solution, samples of WAF test solutions equilibrated for 20, 24 27 and 49 hours at 100 mg/l loading were analyzed by GC-FID for concentrations of the following: benzene, toluene, ethylbenzene, xylene isomers and naphthalene (BTEXN). Maximum WAF solubility for these components reached equilibrium within 24 hrs of stirring. Nominal loading rates of 0, 0.1, 1, 5, 10, and 25 mg/l were used to prepare test solutions for the fish toxicity tests. Test substance, added volumetrically, was mixed for each liter stoppered containers with less than 10% headspace volume. The mixtures were allowed to settle for 1-2 hours prior to drawing off the aqueous solutions for testing. Fish were approximately five weeks old at test initiation and were obtained from Thomas Fish company, Anderson, CA, Lot 297. Loading of fish body mass to treatment was 0.3 g fish per liter of aqueous solution, mean length at termination was 3.3 cm (sd=0.2), and mean weight was 0.271 g (sd=0.064). Test vessels were 4 liter glass aspirator bottles with foil covered neoprene stoppers. Three replicates per treatment and 5 organisms per replicate were tested for each treatment and the control. Exposure containers were filled ( no headspace volume) and tightly sealed to prevent volatilization. Test solution renewal was performed daily by removing 80% of the test solution and replacing it with fresh WAF solution prepared at least 24 hrs prior to use. Freshly prepared and old WAF test solutions were analyzed by GC-FID for concentrations of BTEXN. Water temperature was 15 °C (0.1sd). Test photoperiod was 16 hrs. light and 8 hr dark, light intensity approx 609-614 Lux during full daylight periods. Dissolved oxygen measurements ranged from 5.4 to 9.7 ppm, pH values between 6.8 and 8.2.</p>																		
<b>Limit Test:</b>	No																		
<b>Test Results - Acute Toxicity To Aquatic Vertebrates</b>																			
<b>NOEC Exposure Duration:</b>																			
<b>NOEC:</b>																			
<b>LOEC Exposure Duration:</b>																			
<b>LOEC:</b>																			
<b>NOELR Exposure Duration:</b>	96 Hours																		
<b>NOELR:</b>	= 10 mg/L																		
<b>LOELR Exposure Duration:</b>																			
<b>LOELR:</b>																			
<b>LC/EC Mean Value:</b>	<table border="1"> <thead> <tr> <th>Exposure Duration</th> <th>Exposure Units</th> <th>LC/EC</th> <th>%</th> <th>Value Description</th> <th>Mean Value</th> <th>Units</th> <th>Effect Observed</th> <th>Basis for Concentration</th> </tr> </thead> <tbody> <tr> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> </tr> </tbody> </table>	Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value	Units	Effect Observed	Basis for Concentration									
Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value	Units	Effect Observed	Basis for Concentration											

					or Lower Mean Value	Upper Mean Value			
96	Hours	LL	50 %	=	16		mg/L	Mortality	Nominal

**Results Remarks:**

Mortality (no. of deaths/treatment) at 96 hrs:

Treatment	No. of deaths (mg/l)
0	0
0.1	0
1.0	0
5.0	0
10	0
25	15

96-hr LL50 = 16 mg/l, 99% C.I: 10-25 mg/l (as nominal loading rate) 96-hour No Observed Effect Loading (NOEL) was 10 mg/l, based on mortality, both calculated (Dunnett's Procedure) and observed. Results are quoted in terms of 50% Lethal Loading (LL50), the loading rate of test substance resulting in 50% mortality of the test species exposed to the WAF. At termination, loss of equilibrium was observed in all surviving fish at the 10 mg/l treatment.

Analytical results Losses of the soluble BTEXN components from the WAF over each 24 hour period ranged from 0 to 8% for the 1.0, 5, 10 and 25 mg/l loadings. Up to 100% loss was observed in the 0.1 mg/l treatment in 24 hrs samples.

Analytical results Measured BTEXN (mg/l)

rate (mg/l)		Nominal loading				
Day	Control	0.1	1.0	5.0	10	25
0 (new)	ND	0.12	0.31	1.7	3.1	7.7
1 (old)	ND	0.12	0.41	1.6	3.3	7.1
1 (new)	ND	0.16	0.44	1.7	1.9	6.5
2 (old)	ND	0.15	0.45	1.6	2.1	6.8
2 (new)	ND	0.07	0.43	1.6	3.2	NA
3 (old)	ND	0.12	0.43	1.6	3.1	NA
3 (new)	ND	0.16	0.57	1.8	3.3	NA
4 (old)	ND	ND	0.56	1.8	3.5	NA

ND=not detected, NA=not analyzed due to 100% mortality

Guideline/protocol deviations: Body length smaller than recommended range of 4-6 cm; smaller fish used to minimize DO depletion in closed vessel (no-headspace) systems.

### Reliability/Data Quality - Acute Toxicity To Aquatic Vertebrates

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP; guideline study

**Key Study Sponsor Indicator:** Key

### Reference - Acute Toxicity To Aquatic Vertebrates

**Reference:** Study conducted by Exxon Biomedical Sciences Inc. Fish acute toxicity test: study no. 104958, test substance MRD-95-049. CONCAWE, Brussels, 1995

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Acute Toxicity to Aquatic Vertebrates</b>	
<b>Test Substance - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Olefenic naphthas Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Pimephales promelas
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Semi-Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 3.0, 7.4, 15, 37 and 74 mg/l
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	5.2 and 8.6
<b>pH Value:</b>	7.61 Upper Range: 8.2
<b>Test Temperature and Units:</b>	Value/Lower Range: 21.4 Upper Range: 21.8 °C



**Photo (Light/Dark):** 16/8

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline Test Conditions Remarks:** No specific guideline was described as being used to conduct the test, however report references 1991 ASTM method E729-88a and 1982 EPA Support Documents for Environmental Testing: EPA 560/6-82-002.

Statistical Method: (FT -ME) LL50 and LC50 calculated using binomial probability analysis. ASTM Special Technical Publication 634. 1977, pp 65-84.

Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water was Mobil Technical Center well water. Nominal loading rates of 0, 3.0, 7.4, 15, 37 and 74 mg/l were used to prepare test solutions.

WAFs were prepared for each test concentration by mixing the appropriate mass of substance in 9.4l of water for 24 hr in 9l glass bottles. The bottles were filled to neck height with water to minimize volatility. A measured amount of test substance was added into each bottle, and the bottles were capped tightly with a positive pressure siphoning apparatus. The siphoning apparatus consisted of a teflon lined neoprene stopper housing two teflon tubes. One tube extended to the bottom of the bottle for removal of the WAF solution, the other tube ended above the WAF surface, and was used to control air pressure during siphoning. During WAF preparation, parafilm was used to seal the external joint between the neoprene stopper and glass bottle. After stirring for 24 hrs using 25% or less vortex, the contents of the WAF solution bottles were allowed to settle for approximately 45 minutes to two hours, then siphoned by the positive pressure apparatus port and used for testing. Samples were also analyzed by Purge & trap/GC-PID for concentrations of the following: benzene, toluene, ethylbenzene, and p-xylene, which represent 13% composition of the test substance. Measured test concentrations of the light catalytically cracked naphtha were based on the total combined concentrations of all analytes.

Fish were hatched and raised in-house, and were acclimated prior to experimentation for a minimum of 14 days on a 16/8hr light/dark cycle. Test vessels were 3.8l glass containers with teflon lined caps. Two replicates per treatment and 10 organisms per replicate were tested for each treatment and the control. Exposure containers were filled with no headspace and tightly sealed to prevent volatilization. Test solution renewal was performed daily by removal of most of the test solution (leaving adequate volume to avoid stressing test organisms) and replacing it with fresh WAF solution prepared at least 24 hrs prior to use.

Water temperature was 21.4-21.8 °C. Test photoperiod was 16 hrs. light and 8 hr dark. Dissolved oxygen measurements were between 5.2 and 8.6, pH values between 7.61 and 8.2.

**Limit Test:** No

**Test Results - Acute Toxicity To Aquatic Vertebrates**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:** 96 Hours

**NOELR:** = 15 mg/L Nominal

**LOELR Exposure Duration:**

**LOELR:**

**LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or	Upper Mean Value	Units	Effect Observed	Basis for Concentration
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					<b>Lower Mean Value</b>				
96	Hours	LL	50 %	=	46		mg/L	Mortality	Nominal
			%						

**Results Remarks:** Mortality (no. of deaths/treatment) at 96 hrs: 0, 1, 0, 0, 4 and 20, respectively 0, 3.0, 7.4, 15, 37 and 74 mg/l treatments. All surviving organisms exhibited normal behavior.  
96-hr LL50 = 46 mg/l, 37-74 mg/l w/ 95% C.I. (as nominal loading rate)  
96-hr LC50 = 4.1 mg/l, 3.2-7.0 mg/l mg/l w/ 95% C.I. (measured concentrations)

Measured concentrations represented the sum of four specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, there was insufficient information regarding the analytical measurements. It was not reported how many sample measurements were taken, nor whether the reported values were based on fresh or old solutions, initial measurements, or a mean of all measurements. Additionally, it was not reported to what degree measured concentrations declined between solution renewals. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

### Reliability/Data Quality - Acute Toxicity To Aquatic Vertebrates

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Measured concentrations may not represent 100% of components. Remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations.

NOEC values not reported as sublethal effects and moderate mortality (20%) were observed at the 37 ppm (nominal loading) treatment which is reported to be the NOEC.

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:** Key

### Reference - Acute Toxicity To Aquatic Vertebrates

**Reference:** Stonybrook Laboratories, Inc. (1995) Static Renewal 96-hour acute toxicity of the water accommodated fraction (WAF) of FR 15799 FCC Light to Fathead Minnow. Study No. 66234 Stonybrook Laboratories Inc. Princeton, NJ.1995.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Acute Toxicity to Aquatic Vertebrates</b>	
<b>Test Substance - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Category Chemical:</b>	(64741-70-4) Naphtha, petroleum, isomerization
<b>Test Substance:</b>	(64741-70-4) Naphtha, petroleum, isomerization
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	PARAFFINIC NAPHTHA CAS 64741-70-4, CONCAWE sample W94/810 Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Vertebrates</b>	
<b>Year Study Performed:</b>	1996
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Oncorhynchus mykiss
<b>GLP:</b>	
<b>Analytical Monitoring:</b>	
<b>Test Type:</b>	
<b>Test Vessel:</b>	
<b>Water Media Type:</b>	
<b>Test Concentrations:</b>	
<b>Nominal and Measured Concentrations:</b>	maximum loadings of 50 mg/l or less
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	
<b>Test Temperature and Units:</b>	

**Photo  
(Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline  
Test Conditions  
Remarks:**

Experimental studies with fish, invertebrates and algae tested in closed systems with minimal head-space were performed on WAFS of low boiling point naphthas prepared at maximum loadings of 50 mg/l or less. Results show that acute aquatic toxicity lethal loading (LL), effective loading (EL) or inhibition of growth rate values (IrL) affecting 50% of the organism population are greater than 1 mg/l and mostly in the range 1-100 mg/l. Summarized CONCAWE test data indicating the extent of aquatic toxicity are as follows in Results Remarks Section, and 95% confidence intervals are included in parentheses.

**Limit Test:**

### Test Results - Acute Toxicity To Aquatic Vertebrates

**NOEC Exposure  
Duration:**

**NOEC:**

**LOEC Exposure  
Duration:**

**LOEC:**

**NOELR Exposure  
Duration:**

**NOELR:**

**LOELR Exposure  
Duration:**

**LOELR:**

**LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration
96	Hours	LL	50 %	=	10		mg/L		Nominal

**Results Remarks:**

PARAFFINIC NAPHTHA CAS 64741-70-4, CONCAWE sample W94/810

Fish (*Oncorhynchus mykiss*) LL50, 96h 10 mg/l (5-23)

Invertebrate (*Daphnia magna*) EL50, 48h 10 mg/l (8.5-13)

Algae (*Selenastum capricornutum*) IrL50, 72h >50 mg/l (not calculable)

95% confidence intervals are included in parentheses

### Reliability/Data Quality - Acute Toxicity To Aquatic Vertebrates

**Reliability:**

**Reliability  
Remarks:**

**Key Study Sponsor  
Indicator:**

Key

### Reference - Acute Toxicity To Aquatic Vertebrates

**Reference:**

CONCAWE. 1996c. Acute Aquatic Toxicity of Gasolines - Report on CONCAWE Test Programme. Report No. 96/57. CONCAWE, Brussels, Belgium.



## Acute Toxicity to Aquatic Vertebrates

### Test Substance - Acute Toxicity To Aquatic Vertebrates

<b>Category Chemical:</b>	(64741-54-4) Naphtha, petroleum, heavy catalytic cracked
<b>Test Substance:</b>	(64741-54-4) Naphtha, petroleum, heavy catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	OLEFINIC NAPHTHA CAS . 64741-54-4, CONCAWE sample W94/811 Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured

### Method - Acute Toxicity To Aquatic Vertebrates

<b>Year Study Performed:</b>	1996
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Oncorhynchus mykiss
<b>GLP:</b>	
<b>Analytical Monitoring:</b>	
<b>Test Type:</b>	
<b>Test Vessel:</b>	
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	
<b>Nominal and Measured Concentrations:</b>	maximum loadings of 50 mg/l or less
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	
<b>Test Temperature and Units:</b>	

<b>Photo (Light/Dark):</b>									
<b>Salinity:</b>									
<b>TOC:</b>									
<b>Water Hardness:</b>									
<b>Method/Guideline Test Conditions Remarks:</b>									
Experimental studies with fish, invertebrates and algae tested in closed systems with minimal head-space were performed on WAFS of low boiling point naphthas prepared at maximum loadings of 50 mg/l or less. Results show that acute aquatic toxicity lethal loading (LL), effective loading (EL) or inhibition of growth rate values (IrL) affecting 50% of the organism population are greater than 1 mg/l and mostly in the range 1-100 mg/l. Summarized CONCAWE test data indicating the extent of aquatic toxicity are as follows in Results Remarks Section, and 95% confidence intervals are included in parentheses.									
<b>Limit Test:</b>									
<b>Test Results - Acute Toxicity To Aquatic Vertebrates</b>									
<b>NOEC Exposure Duration:</b>									
<b>NOEC:</b>									
<b>LOEC Exposure Duration:</b>									
<b>LOEC:</b>									
<b>NOELR Exposure Duration:</b>									
<b>NOELR:</b>									
<b>LOELR Exposure Duration:</b>									
<b>LOELR:</b>									
<b>LC/EC Mean Value:</b>									
<b>Exposure Duration</b>	<b>Exposure Units</b>	<b>LC/EC</b>	<b>%</b>	<b>Value Description</b>	<b>Mean Value or Lower Mean Value</b>	<b>Upper Mean Value</b>	<b>Units</b>	<b>Effect Observed</b>	<b>Basis for Concentration</b>
96	Hours	LL	50 %	=	15		mg/L		Nominal
<b>Results Remarks:</b>									
OLEFINIC NAPHTHA CAS . 64741-54-4, CONCAWE sample W94/811									
Fish (Oncorhynchus mykiss) LL50, 96h 15 mg/l (10-23)									
Invertebrate (Daphnia magna) EL50, 48h 13 mg/l (12-15)									
Algae (Selenastum capricornutum) IrL50, 72h 3.1 mg/l (3.6-14)									
95% confidence intervals are included in parentheses									
<b>Reliability/Data Quality - Acute Toxicity To Aquatic Vertebrates</b>									
<b>Reliability:</b>									
<b>Reliability Remarks:</b>									
<b>Key Study Sponsor Indicator:</b>									
Key									
<b>Reference - Acute Toxicity To Aquatic Vertebrates</b>									
<b>Reference:</b>									
CONCAWE. 1996c. Acute Aquatic Toxicity of Gasolines - Report on CONCAWE Test Programme. Report No. 96/57. CONCAWE, Brussels, Belgium.									



## Acute Toxicity to Aquatic Invertebrates

### Test Substance - Acute Toxicity To Aquatic Invertebrates

**Category Chemical:** No CAS Number Provided

**Test Substance:** No CAS Number Provided

**Test Substance Purity/Composition and Other Test Substance Comments:** Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Read-Across

**Test Substance Result Type:**

### Method - Acute Toxicity To Aquatic Invertebrates

**Year Study Performed:** 1996

**Method/Guideline Followed:**

**Other Method/Guideline:**

**Deviations from Method/Guideline:**

**Species/In Vitro System:**

**GLP:**

**Analytical Monitoring:**

**Test Type:**

**Test Vessel:**

**Water Media Type:**

**Test Concentrations:**

**Nominal and Measured Concentrations:**

**Exposure Period:**

**Vehicle Used:**

**Vehicle Name:**

**Vehicle Amount and Units:**

**Alkalinity:**

**Dissolved Oxygen:**

**pH Value:**

**Test Temperature and Units:**

<b>Photo (Light/Dark):</b>									
<b>Salinity:</b>									
<b>TOC:</b>									
<b>Water Hardness:</b>									
<b>Method/Guideline Test Conditions Remarks:</b> Aquatic invertebrate acute toxicity values for members of the Gasoline Blending Streams Category are expected to be similar based on the common mode of action for acute toxicity of petroleum hydrocarbons. These values may be expected to fall within the range of calculated and measured data cited in the robust summaries for tested or modeled members of the Gasoline Blending Streams Category.									
<b>Limit Test:</b>									
<b>Test Results - Acute Toxicity To Aquatic Invertebrates</b>									
<b>NOEC Exposure Duration:</b>									
<b>NOEC:</b>									
<b>LOEC Exposure Duration:</b>									
<b>LOEC:</b>									
<b>NOELR Exposure Duration:</b>									
<b>NOELR:</b>									
<b>LOELR Exposure Duration:</b>									
<b>LOELR:</b>									
<b>LC/EC Mean Value:</b>									
<b>Exposure Duration</b>	<b>Exposure Units</b>	<b>LC/EC</b>	<b>%</b>	<b>Value Description</b>	<b>Mean Value or Lower Mean Value</b>	<b>Upper Mean Value</b>	<b>Units</b>	<b>Effect Observed</b>	<b>Basis for Concentration</b>
48	Hours	EL	50 %	=	.9	32	mg/L		
<b>Results Remarks:</b> The range of invertebrate acute toxicity values that may be used as read-across for members of the category is 0.9 to 32 mg/L (lethal loading rates) based on both calculated and measured data.  The basis for effect was either immobility or mortality.									
<b>Reliability/Data Quality - Acute Toxicity To Aquatic Invertebrates</b>									
<b>Reliability:</b> Valid with Restrictions									
<b>Reliability Remarks:</b> All studies used to derive the read across range for acute invertebrate toxicity were considered to be "2 - valid with restrictions".									
<b>Key Study Sponsor Indicator:</b> Weight of Evidence									
<b>Reference - Acute Toxicity To Aquatic Invertebrates</b>									
<b>Reference:</b> See Acute Toxicity to Aquatic Invertebrates Robust Study Summaries for CAS #: 64741-46-4, 64741-54-4, 64741-55-5, 64741-63-5, 64741-66-8, 64741-70-4, 68955-35-1, and 86290-81-5									





<b>Acute Toxicity to Aquatic Invertebrates</b>	
<b>Test Substance - Acute Toxicity To Aquatic Invertebrates</b>	
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Gasoline CAS No. 86290-81-5 Gasoline Sample W94/813, Blend Detailed hydrocarbon analysis: N-paraffins: 20% total C3-C8 Iso-paraffins: 28% total C4-C9 Olefins: 1% C5-C7 Naphthenes: 5% C5-C10 Aromatics: 46% C6-C9  Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>  [Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Invertebrates</b>	
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	OECD 202
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species/In Vitro System:</b>	Daphnia magna
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	
<b>Test Vessel:</b>	
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 0.1, 1, 5, 10, and 25 mg/l
<b>Exposure Period:</b>	48 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	

<b>Vehicle Amount and Units:</b>										
<b>Alkalinity:</b>										
<b>Dissolved Oxygen:</b> 7.2-9.2										
<b>pH Value:</b> 7.5 Upper Range: 7.8										
<b>Test Temperature and Units:</b> Value/Lower Range: 19 °C										
<b>Photo (Light/Dark):</b>										
<b>Salinity:</b>										
<b>TOC:</b>										
<b>Water Hardness:</b>										
<b>Method/Guideline Test Conditions Remarks:</b> EL50 calculated using the probit procedure (Finney, D.J., 1971. Probit Analysis, 3rd Ed. London: Cambridge Univ. Press)										
<p>Test solutions were prepared as water accommodated fractions (WAF). The control and dilution water was a laboratory blend water prepared from carbon filtered well water and ion exchange softened, reverse osmosis dialyzed well water aged for &gt;24 hrs. To determine contact time required to achieve maximum solubility between test substance and aqueous solution, samples of WAF test solutions equilibrated for 20, 24, 27 and 49 hours at 100 mg/l loading were analyzed by GC-FID for concentrations of the following: benzene, toluene, ethylbenzene, xylene isomers and naphthalene (BTEXN). Maximum WAF solubility for these components reached equilibrium within 24 hrs of stirring. Nominal loading rates of 0, 0.1, 1, 5, 10, and 25 mg/l were used to prepare test solutions for the toxicity tests. Test substance, added volumetrically, was mixed for each individual treatment in dilution water for 24 hours in 4 liter stoppered containers with less than 10% headspace volume. The WAF mixtures were allowed to settle for 1-2 hours prior to drawing off the aqueous solutions for testing. WAF test solutions were analyzed by GC-FID for concentrations of BTEXN on day 0 and at termination. Test vessels for daphnid testing were 125 ml glass erlenmeyer flasks with foil covered neoprene stoppers. Four replicates per treatment and 5 organisms per replicate were tested for each treatment and the control. Exposure containers were filled ( no headspace volume) and tightly sealed to prevent volatilization. During the study test system solutions: dissolved oxygen concentration range: 7.2 to 9.2; pH ranged from 7.5 to 7.8; temperature was 19 °C (sd:0.2).Daphnia magna were supplied by testing laboratory; age &lt; 24 hours old; obtained from culture maintained in-house.</p>										
<b>Limit Test:</b> No										
<b>Test Results - Acute Toxicity To Aquatic Invertebrates</b>										
<b>NOEC Exposure Duration:</b>										
<b>NOEC:</b>										
<b>LOEC Exposure Duration:</b>										
<b>LOEC:</b>										
<b>NOELR Exposure Duration:</b> 48 Hours										
<b>NOELR:</b> = 1 mg/L Nominal										
<b>LOELR Exposure Duration:</b>										
<b>LOELR:</b>										
<b>LC/EC Mean Value:</b>										
	<b>Exposure Duration</b>	<b>Exposure Units</b>	<b>LC/EC</b>	<b>%</b>	<b>Value Description</b>	<b>Mean Value or Lower Mean Value</b>	<b>Upper Mean Value</b>	<b>Units</b>	<b>Effect Observed</b>	<b>Basis for Concentration</b>

48	Hours	EL	50%	=	7.6	mg/L	Immobilization	Nominal
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**Results Remarks:** 48 hr results-number of organisms affected and analytical results  
 Measured Measured

Treatment	Immobilization	BTEXN -day 0	BTEXN -day 2
Control	0	ND	ND
0.5 mg/l	0	0.29	0.10
1.0 mg/l	0	0.28	0.10
5.0 mg/l	3	2.3	1.7
10 mg/l	16	3.9	3.1
25 mg/l	20	8.8	10

based upon nominal loading rate 48-hr EL50 = 7.6 mg/l (95% C.I. 6.4 to 9.3 mg/l) 48-hr  
 NOEL = 1.0 mg/l

### Reliability/Data Quality - Acute Toxicity To Aquatic Invertebrates

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Three previous attempts to conduct study were invalidated due to excessive (>20%) control mortality. Two more studies (this study and one other) had acceptably low mortality rates and were considered valid.

Guideline study conducted under GLP

**Key Study Sponsor Indicator:** Key

### Reference - Acute Toxicity To Aquatic Invertebrates

**Reference:** CONCAWE (1995) Daphnia -acute toxicity test: study no. 104842, test substance MRD-95-048. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Acute Toxicity to Aquatic Invertebrates</b>	
<b>Test Substance - Acute Toxicity To Aquatic Invertebrates</b>	
<b>Category Chemical:</b>	(64741-46-4) Naphtha, petroleum, light straight-run
<b>Test Substance:</b>	(64741-46-4) Naphtha, petroleum, light straight-run
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	LSRN-Moderate naphthenic content Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Invertebrates</b>	
<b>Year Study Performed:</b>	1996
<b>Method/Guideline Followed:</b>	ASTM E 729
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species/In Vitro System:</b>	Daphnia magna
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Semi-Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	3.0, 6.0, 12, 24 and 48 mg/l
<b>Exposure Period:</b>	48 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	142-150 mg/l
<b>Dissolved Oxygen:</b>	8.0-8.5
<b>pH Value:</b>	8.3 Upper Range: 8.4
	Value/Lower Range: 20 Upper Range: 21 °C

**Test Temperature and Units:**

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:** 132 - 140 mg/L

**Method/Guideline Test Conditions Remarks:** Procedure patterned after:1991 ASTM method E729-88a and 1985 USEPA TSCA Test Guidelines: Daphnid Acute Toxicity Test. Fed. Reg., vol. 50 (No. 188) Sept 27, 1985, 797.1300.

Statistical Method: (FT -ME) EL50 and EC50 calculated using binomial probability analysis. ASTM Special Technical Publication 634. 1977, pp 65-84.

Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water were a blend of aged well water and reverse osmosis well water.

WAFS were prepared for each test concentration by mixing the appropriate mass of substance in 2.4l of water for 24 hr in aluminum foil covered 2.5 liter aspirator bottles fitted w/ a port near the bottle bottom. The bottles were filled to neck height with water to minimize volatility. A measured amount of test substance was pipetted below the water surface. After stirring for 24 hrs using 25% or less vortex, the contents of the aspirator bottles were allowed to settle for approximately one hour, then drained from the port and used for testing. Samples were also analyzed by purge & trap/GC-FID for concentrations of the following: 2-methyl-pentane, cyclohexane, benzene, toluene, ethylbenzene, ortho, meta and para-xylene. Measured test concentrations of the light straight run naphtha were based on the total combined concentrations of all analytes.

Range finding toxicity studies were conducted at 0.5, 1.0, 10 and 100 mg/l loading, using WAFS which were divided into duplicate aliquots and tested. Definitive toxicity studies were conducted at 3.0, 6.0, 12, 24 and 48 mg/l loading, using WAFS which were divided into duplicate aliquots and tested. Test vessels were teflon cap-sealed 8 oz. glass jars with 10 daphnids per jar and were completely filled to overflowing with approximately 273 ml test solution.

During the study test system solutions: dissolved oxygen concentration range: 8.0 to 8.5; pH ranged from 8.3 to 8.4; temperature was 20 to 21 °C; hardness (mg/l) ranged from 132 -140; alkalinity (mg/l) was 142-150 and conductivity (imhos) values were 280 -300.

Daphnia magna, were supplied by testing laboratory; age < 24 hours old; obtained from 11 day culture maintained in-house since October 1996.

**Limit Test:** No

**Test Results - Acute Toxicity To Aquatic Invertebrates**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:** 48 Hours

**NOELR:** = 6 mg/L Nominal

**LOELR Exposure Duration:**

**LOELR:**

LC/EC Mean Value:	Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower	Upper Mean Value	Units	Effect Observed	Basis for Concentration

					Mean Value				
48	Hours	EL	50 %	=	18	mg/L	Immobilization	Nominal	
			%						

**Results Remarks:**

Immobility (no. of organisms) at 48 hrs: 1, 3, 0, 0, 19 and 20 for 0, 3.0, 6.0, 12, 24 and 48 mg/l treatments. At the 3 and 12mg/l nominal treatments, 1 and 20 organisms were observed at the bottom of the test chambers, respectively.

48-hr EL50 = 18 mg/l based upon nominal loading rate (95% C.I. 12 to 24 mg/l)

48 hr EC50 was 0.65 mg/l (95% C.I. 0.47 to 0.83 mg/l); based on total measured concentrations.

48-hr NOEL = 6.0 mg/l based upon nominal loading rate.

48 hr NOEC was 0.24 ppm based on total measured concentrations.

Measured concentrations represented the sum of six specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, there was insufficient information regarding the analytical measurements. It was not reported how many sample measurements were taken, nor whether the reported values were based on fresh or old solutions, initial measurements, or a mean of all measurements. Additionally, it was not reported to what degree measured concentrations declined between solution renewals. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

### Reliability/Data Quality - Acute Toxicity To Aquatic Invertebrates

**Reliability:**

Valid with Restrictions

**Reliability Remarks:**

Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAF may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP; guideline study

**Key Study Sponsor Indicator:**

Key

### Reference - Acute Toxicity To Aquatic Invertebrates

**Reference:**

ABC Laboratories, Inc. (1998) Static Renewal 48-hour Acute Toxicity of the Water Accomodated Fraction (WAF) of Light Straight Run Naphtha, LSRN to Daphnia Magna. Study No. 43150. Environmental Toxicology, 7200 E. ABC Lane, Columbia, Missouri.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



## Acute Toxicity to Aquatic Invertebrates

### Test Substance - Acute Toxicity To Aquatic Invertebrates

**Category Chemical:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:** Aromatic naphthas

Substance is in the Gasoline Blending Streams Category.  
See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:** Measured

**Test Substance Result Type:** Measured

### Method - Acute Toxicity To Aquatic Invertebrates

**Year Study Performed:** 1998

**Method/Guideline Followed:** ASTM E 729

**Other Method/Guideline:**

**Deviations from Method/Guideline:**

**Species/In Vitro System:** Daphnia magna

**GLP:** Yes

**Analytical Monitoring:** Yes

**Test Type:** Semi-Static

**Test Vessel:** Closed

**Water Media Type:** Freshwater

**Test Concentrations:** Nominal

**Nominal and Measured Concentrations:** 0, 3.0, 6.0, 12, 24 and 48 mg/l

**Exposure Period:** 48 Hours

**Vehicle Used:**

**Vehicle Name:**

**Vehicle Amount and Units:**

**Alkalinity:** 158-168

**Dissolved Oxygen:** 8.0-8.5

**pH Value:** 8.2 Upper Range: 8.4

Value/Lower Range: 20 Upper Range: 21 °C

**Test Temperature and Units:****Photo (Light/Dark):****Salinity:****TOC:****Water Hardness:** 146 - 152 mg/L**Method/Guideline Test Conditions Remarks:**

Procedure patterned after:1991 ASTM method E729-88a and 1985 USEPA TSCA Test Guidelines: Daphnid Acute Toxicity Test. Fed. Reg., vol. 50 (No. 188) Sept 27, 1985, 797.1300.

Statistical Method: (FT -ME) EL50 and EC50 calculated using binomial probability analysis. ASTM Special Technical Publication 634. 1977, pp 65-84.

Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water were a blend of aged well water and reverse osmosis well water.

WAFS were prepared for each test concentration by mixing the appropriate mass of substance in 2.4 liters of water for 24 hr in aluminum foil covered 2.5 liter aspirator bottles fitted w/ a port near the bottle bottom. The bottles were filled to neck height with water to minimize volatility. A measured amount of test substance was pipetted below the water surface. After stirring for 24 hrs using 25% or less vortex, the contents of the aspirator bottles were allowed to settle for approximately one hour, then drained from the port and used for testing. Samples were also analyzed by purge & trap/GC-FID for concentrations of the following: pentane, 2-methyl-pentane, benzene, toluene, ethylbenzene, ortho, meta and para-xylene, which represent more than 50% composition of the test substance. Measured test concentrations of the light catalytically reformed naphtha were based on the total combined concentrations of all analytes.

Range finding toxicity studies were conducted at 0.5, 1.0, 10 and 100 mg/l loading, using WAFS which were divided into duplicate aliquots and tested. Definitive toxicity studies were conducted at 3.0, 6.0, 12, 24 and 48 mg/l loading, using WAFS which were divided into duplicate aliquots and tested. Test vessels were teflon cap-sealed 273 ml glass jars with 10 daphnids per jar and were completely filled with test solution.

During the study test system solutions: dissolved oxygen concentration range: 8.0 to 8.5; pH ranged from 8.2 to 8.4; temperature was 20 to 21 °C; hardness (mg/l) ranged from 146 -152; alkalinity (mg/l) was 158-168 and conductivity (imhos) values were 312 -317.

Daphnia magna, were supplied by testing laboratory; age < 24 hours old; obtained from culture maintained in-house since January 1998.

**Limit Test:** No**Test Results - Acute Toxicity To Aquatic Invertebrates****NOEC Exposure Duration:****NOEC:****LOEC Exposure Duration:****LOEC:****NOELR Exposure Duration:** 48 Hours**NOELR:** = 3 mg/L Nominal**LOELR Exposure Duration:****LOELR:****LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower	Upper Mean Value	Units	Effect Observed	Basis for Concentration



					Mean Value				
48	Hours	EL	50 %	=	10	mg/L	Immobilization	Nominal	
			%						

**Results Remarks:**

Immobility (no. of organisms) at 48 hrs: 0, 0, 0, 15, 20 and 20 for 0, 3.0, 6.0, 12, 24 and 48 mg/l treatments. At the 6 and 12mg/l nominal treatments, 20 and 5 organisms were observed at the bottom of the test chambers, respectively.

48-hr EL50 = 10 mg/l based upon nominal loading rate (95% C.I. 6 to 12 mg/l);  
48 hr EC50 was 2.6 mg/l (95% C.I. 1.06 to 3.6 mg/l); based on total measured concentrations.

48-hr NOEL = 3 mg/l based upon nominal loading rate.

48 hr NOEC was 0.465 ppm based on total measured concentrations.

Measured concentrations represented the sum of six specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, there was insufficient information regarding the analytical measurements. It was not reported how many sample measurements were taken, nor whether the reported values were based on fresh or old solutions, initial measurements, or a mean of all measurements. Additionally, it was not reported to what degree measured concentrations declined between solution renewals. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

**Reliability/Data Quality - Acute Toxicity To Aquatic Invertebrates****Reliability:**

Valid with Restrictions

**Reliability Remarks:**

Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:**

Key

**Reference - Acute Toxicity To Aquatic Invertebrates****Reference:**

ABC Laboratories, Inc. (1998) Static Renewal 48-hour Acute Toxicity of the Water Accommodated Fraction (WAF) of Light Catalytically Reformed Naphtha, LCRN) to Daphnia Magna. Study No. 43577. Environmental Toxicology, 7200 E. ABC Lane, Columbia, Missouri. 1998.

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<b>Acute Toxicity to Aquatic Invertebrates</b>	
<b>Test Substance - Acute Toxicity To Aquatic Invertebrates</b>	
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Paraffinic naphthas Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Invertebrates</b>	
<b>Year Study Performed:</b>	1994
<b>Method/Guideline Followed:</b>	Other <span style="float: right;">x</span>
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species/In Vitro System:</b>	Daphnia magna
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Semi-Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	9, 18, 35, 70, & 140 mg/l
<b>Exposure Period:</b>	48 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	140-156
<b>Dissolved Oxygen:</b>	8.0 to 8.5
<b>pH Value:</b>	8 Upper Range: 8.2
<b>Test Temperature and Units:</b>	Value/Lower Range: 19.1 Upper Range: 21 °C

**Photo  
(Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:** 180 - 204 mg/L

**Method/Guideline  
Test Conditions  
Remarks:**

No specific guideline was described as being used to conduct the test, however report references 1991 ASTM method E729-88a and 1982 EPA Support Documents for Environmental Testing: EPA 560/6-82-002.

Statistical Method: (FT -ME) EL50 and EC calculated using binomial probability a analysis. ASTM Special Technical Publication 634. 1977, pp 65-84.

Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water was aged well water.

WAFS were prepared for each test concentration by mixing the appropriate mass of substance in 1.2 liters of water for 24 hr in aluminum foil covered 1-liter aspirator bottles fitted w/ a port near the bottle bottom. The bottles were filled to neck height with water to minimize volatility. A measured amount of test substance was pipetted below the water surface. After stirring for 24 hrs using 25% or less vortex, the contents of the aspirator bottles were allowed to settle for approximately 45 minutes, then drained from the port and used for testing. Samples were also analyzed by Purge & trap/GC-FID for concentrations of the following: 2,3 dimethyl butane; 2,4 dimethyl pentane; 2,2,4 trimethyl pentane; 2,5 dimethyl hexane; 2,3,4 trimethyl pentane, 2,3,3 trimethyl pentane and 1-methyl-1-ethyl cyclopentane, which represent 68% composition of the test substance. Measured test concentrations of the light alkylate naphtha were based on the total combined concentrations of all analytes.

Range finding toxicity studies were conducted at 1.2, 9.9 and 99 mg/l loading, using WAFS which were divided into duplicate aliquots and tested. Definitive toxicity studies were conducted at 9, 18, 35, 70, & 140 mg/l loading, using WAFS which were divided into duplicate aliquots and tested.

Test vessels were teflon cap-sealed 237 ml glass jars with 10 daphnids per jar and were completely filled with test solution.

During the study test system solutions:  
dissolved oxygen concentration range: 8.0 to 8.5  
pH ranged from 8.00 to 8.2  
temperature was 19.1 to 21.0 °C  
hardness (mg/l) ranged from 180 - 204  
alkalinity (mg/l) was 140-156  
conductivity (imhos) values were 385 -390.

Daphnia magna, were supplied by testing laboratory; age < 24 hours old; obtained from culture maintained in-house since January 1994. The primary culture was obtained from Aquatic Research organisms, Hampton, NH, which was derived from EPA laboratory culture, in Cincinnati, Ohio.

**Limit Test:** No

### Test Results - Acute Toxicity To Aquatic Invertebrates

**NOEC Exposure  
Duration:**

**NOEC:**

**LOEC Exposure  
Duration:**

**LOEC:**

**NOELR Exposure  
Duration:** 48 Hours

**NOELR:** = 18 mg/L Nominal

**LOELR Exposure  
Duration:**

**LOELR:**

LC/EC Mean Value:	Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration
	48	Hours	EL	50	=	32		mg/L	Immobilization	Nominal
				%						

**Results Remarks:** Immobility (no. of organisms) at 48 hrs: 0, 0, 0, 12, 13 and 20 for 0, 9, 18, 35, 70 and 140 mg/l treatments. At the 35 and 70 mg/l nominal treatments, 8 and 7 organisms were observed to show lethargic movement, respectively.

48-hr EL50 = 32 mg/l (95% C.I. 18 to 140 mg/l) based upon nominal loading rate. 48 hr EC50 was 556 ig/l (95% C.I. 339 to 1140 ig/l) based on total measured alkyl concentrations.

48-hr NOEL = 18 mg/l based upon nominal loading rate.

48 hr NOEC was 339 ppb based on total measured alkyl concentrations.

Measured concentrations represented the sum of seven specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, there was insufficient information regarding the analytical measurements. It was not reported how many sample measurements were taken, nor whether the reported values were based on fresh or old solutions, initial measurements, or a mean of all measurements. Additionally, it was not reported to what degree measured concentrations declined between solution renewals. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

#### Reliability/Data Quality - Acute Toxicity To Aquatic Invertebrates

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAF may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:** Key

#### Reference - Acute Toxicity To Aquatic Invertebrates

**Reference:** Stonybrook Laboratories, Inc. (1995) Static Renewal 48-hour acute toxicity of the water accomodated fraction (WAF) of Whole Light Alkylate Naphtha (LAN) Product to Daphnia Magna. Study No. 65907. Stonybrook Laboratories Inc. Princeton, NJ.

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## Acute Toxicity to Aquatic Invertebrates

### Test Substance - Acute Toxicity To Aquatic Invertebrates

**Category Chemical:** (68955-35-1) Naphtha, petroleum, catalytic reformed

**Test Substance:** (68955-35-1) Naphtha, petroleum, catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:** Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Estimated by Calculation

**Test Substance Result Type:** Estimated

### Method - Acute Toxicity To Aquatic Invertebrates

**Year Study Performed:**

**Method/Guideline Followed:**

**Other Method/Guideline:**

**Deviations from Method/Guideline:**

**Species/In Vitro System:**

**GLP:**

**Analytical Monitoring:**

**Test Type:**

**Test Vessel:**

**Water Media Type:**

**Test Concentrations:**

**Nominal and Measured Concentrations:**

**Exposure Period:** 48 Hours

**Vehicle Used:**

**Vehicle Name:**

**Vehicle Amount and Units:**

**Alkalinity:**

**Dissolved Oxygen:**

**pH Value:**

**Test Temperature and Units:**

**Photo  
(Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline  
Test Conditions  
Remarks:** Calculated based on hydrocarbon block principle. In this procedure, the dissolved concentrations of individual hydrocarbons from a petroleum substance are estimated for a given loading rate and then normalized by their acute toxicity to yield Toxic Units (TU) which can be summed to predict the toxicity of the parent material (see below).

**Limit Test:** Not Applicable

### Test Results - Acute Toxicity To Aquatic Invertebrates

**NOEC Exposure  
Duration:**

**NOEC:**

**LOEC Exposure  
Duration:**

**LOEC:**

**NOELR Exposure  
Duration:**

**NOELR:**

**LOELR Exposure  
Duration:**

**LOELR:**

**LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration
48	Hours	EL	50 %	=	.9		mg/L		Calculated

**Results Remarks:** Estimated 48 hour(s) Daphnid acute toxicity EL50: 0.9 mg/l.

### Reliability/Data Quality - Acute Toxicity To Aquatic Invertebrates

**Reliability:** Valid with Restrictions

**Reliability  
Remarks:** RELIABILITY: Estimated values were calculated using a petroleum substance specific model.

**Key Study Sponsor  
Indicator:** Key

### Reference - Acute Toxicity To Aquatic Invertebrates

**Reference:** Peterson, D.R., (1994) Calculating the aquatic toxicity of hydrocarbon mixtures Chemosphere Vol.29, 12, pp. 2493-2506

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## Acute Toxicity to Aquatic Invertebrates

### Test Substance - Acute Toxicity To Aquatic Invertebrates

<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Olefenic naphthas Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured

### Method - Acute Toxicity To Aquatic Invertebrates

<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species/In Vitro System:</b>	Daphnia magna
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Semi-Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	6.4, 13, 25, 51 and 102 mg/l
<b>Exposure Period:</b>	48 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	124-132 mg/l
<b>Dissolved Oxygen:</b>	8.06
<b>pH Value:</b>	7.94 Upper Range: 8.4
<b>Test Temperature and Units:</b>	Value/Lower Range: 19.1 Upper Range: 20.2 °C

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:** 172 - 180 mg/L

**Method/Guideline Test Conditions Remarks:** No specific guideline was described as being used to conduct the test, however report references 1991 ASTM method E729-88a and 1982 EPA Support Documents for Environmental Testing: EPA 560/6-82-002.

Statistical Method: (FT -ME) EL50 and EC50 calculated using binomial probability analysis. ASTM Special Technical Publication 634. 1977, pp 65-84.

Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water was aged well water.

WAFS were prepared for each test concentration by mixing the appropriate mass of substance in 1.2l of water for 24 hr in aluminum foil covered 1l aspirator bottles fitted with a port near the bottle bottom. The bottles were filled to neck height with water to minimize volatility. A measured amount of test substance was pipetted below the water surface. After stirring for 24 hrs using 25% or less vortex, the contents of the aspirator bottles were allowed to settle for approximately 45 minutes, then drained from the port and used for testing. Samples were also analyzed by Purge & trap/GC-FID for concentrations of the following: benzene, toluene, ethylbenzene, and p-xylene, which represent 13% composition of the test substance. Measured test concentrations of the light catalytically cracked naphtha were based on the total combined concentrations of all analytes.

Range finding toxicity studies were conducted at 1.3, 10 and 102 mg/l loading, using WAFS which were divided into duplicate aliquots and tested. Definitive toxicity studies were conducted at 6.4, 13, 25, 51 and 102 mg/l loading, using WAFS which were divided into duplicate aliquots and tested. Test vessels were teflon cap-sealed 265 ml glass jars with 10 daphnids per jar and were completely filled with test solution. During the study test system solutions: dissolved oxygen concentration range: 8.0 to 8.6; pH ranged from 7.94 to 8.40; temperature was 19.1 to 20.2 °C; hardness (mg/l) ranged from 172 -180; alkalinity (mg/l) was 124-132 and conductivity (µmhos) values were 360 -405.

Daphnia magna, were supplied by testing laboratory; age < 24 hours old; obtained from culture maintained in-house since January 1994. The primary culture was obtained from Aquatic Research organisms, Hampton, NH, which was derived from EPA laboratory culture, in Cincinnati, Ohio.

**Limit Test:** No

**Test Results - Acute Toxicity To Aquatic Invertebrates**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:** 48 Hours

**NOELR:** = 13 mg/L Nominal

**LOELR Exposure Duration:**

**LOELR:**

**LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration



48	Hours	EL	50 %	=	18	mg/L	Mortality	Nominal
			%	=				
			%					

**Results Remarks:**

Mortality (no. of deaths/treatment) at 48 hrs: 0, 0, 0, 20, 20 and 20 for 0, 6.4, 13, 25, 51 and 102 mg/l treatments.

48-hr EL50 = 18 mg/l (95% C.I. 13 to 25 mg/l) based upon nominal loading rate. 48 hr EC50 was 1.4 ppm (95% C.I. 0.99 to 1.95 mg/l); based on total measured concentrations.

48-hr NOEC = 13 mg/l based upon nominal loading rate.

48 hr EC50 was 0.99 ppm based on total measured concentrations.

Measured concentrations represented the sum of four specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, there was insufficient information regarding the analytical measurements. It was not reported how many sample measurements were taken, nor whether the reported values were based on fresh or old solutions, initial measurements, or a mean of all measurements. Additionally, it was not reported to what degree measured concentrations declined between solution renewals. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

### Reliability/Data Quality - Acute Toxicity To Aquatic Invertebrates

**Reliability:**

Valid with Restrictions

**Reliability Remarks:**

Measured concentrations represent only 13-20% of components, remaining hydrocarbon components in WAF may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:**

Key

### Reference - Acute Toxicity To Aquatic Invertebrates

**Reference:**

Stonybrook Laboratories, Inc. (1995) Static Renewal 48-hour Acute Toxicity of the Water Accomodated Fraction (WAF) of FR 15799 FCC Light (Light Catalytically Cracked Naphtha, LCCN) to Daphnia Magna. Study No. 66233. Stonybrook Laboratories Inc. Princeton, NJ.1995.

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## Acute Toxicity to Aquatic Invertebrates

### Test Substance - Acute Toxicity To Aquatic Invertebrates

**Category Chemical:** (86290-81-5) Antiknock Gasoline

**Test Substance:** (86290-81-5) Antiknock Gasoline

**Test Substance Purity/Composition and Other Test Substance Comments:** Gasoline CAS No. 86290-81-5

Gasoline Sample W94/814, Blend Detailed hydrocarbon analysis:

N-paraffins: 16% total C4-C8

Iso-paraffins: 25% total C4-C11

Olefins: 12% C4-C7

Naphthenes: 5% C6-C10

Aromatics: 42% C6-C11

Substance is in the Gasoline Blending Streams Category.

See Category Analysis Document(s) at <http://www.petroleumhvp.org>

[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]

**Category Chemical Result Type:** Measured

**Test Substance Result Type:** Measured

### Method - Acute Toxicity To Aquatic Invertebrates

**Year Study Performed:** 1995

**Method/Guideline Followed:** OECD 202

**Other Method/Guideline:**

**Deviations from Method/Guideline:**

**Species/In Vitro System:** Daphnia magna

**GLP:** Yes

**Analytical Monitoring:** Yes

**Test Type:** Static

**Test Vessel:** Closed

**Water Media Type:** Freshwater

**Test Concentrations:** Nominal

**Nominal and Measured Concentrations:** 0, 0.1, 1, 5, 10, and 25 mg/l

**Exposure Period:** 48 Hours

**Vehicle Used:**

**Vehicle Name:**

**Vehicle Amount and Units:**

**Alkalinity:**

**Dissolved Oxygen:** 7.2-9.2

**pH Value:** 7.5 Upper Range: 7.8

**Test Temperature and Units:** Value/Lower Range: 19 °C

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline Test Conditions Remarks:**

EL50 calculated using the probit procedure (Finney, D.J., 1971. Probit Analysis, 3rd Ed. London: Cambridge Univ. Press)

Test solutions were prepared as water accommodated fractions (WAF). The control and dilution water was a laboratory blend water prepared from carbon filtered well water and ion exchange softened, reverse osmosis dialyzed well water aged for >24 hrs. To determine contact time required to achieve maximum solubility between test substance and aqueous solution, samples of WAF test solutions equilibrated for 20, 24, 27 and 49 hours at 100 mg/l loading were analyzed by GC-FID for concentrations of the following: benzene, toluene, ethylbenzene, xylene isomers and naphthalene (BTEXN). Maximum WAF solubility for these components reached equilibrium within 24 hrs of stirring. Nominal loading rates of 0, 0.1, 1, 5, 10, and 25 mg/l were used to prepare test solutions for the toxicity tests. Test substance, added volumetrically, was mixed for each individual treatment in dilution water for 24 hours in 4 liter stoppered containers with less than 10% headspace volume. The WAF mixtures were allowed to settle for 1-2 hours prior to drawing off the aqueous solutions for testing. WAF test solutions were analyzed by GC-FID for concentrations of BTEXN on day 0 and at termination. Test vessels for daphnid testing were 125 ml glass erlenmeyer flasks with foil covered neoprene stoppers. Four replicates per treatment and 5 organisms per replicate were tested for each treatment and the control. Exposure containers were filled ( no headspace volume) and tightly sealed to prevent volatilization. During the study test system solutions: dissolved oxygen concentration range: 7.2 to 9.2; pH ranged from 7.5 to 7.8; temperature was 19 °C (sd:0.2).Daphnia magna were supplied by testing laboratory; age < 24 hours old; obtained from culture maintained in-house.

**Limit Test:** No

**Test Results - Acute Toxicity To Aquatic Invertebrates**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:** 48 Hours

**NOELR:** = 5 mg/L Nominal

**LOELR Exposure Duration:**

**LOELR:**

**LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration

48	Hours	EL	50%	=	12	mg/L	Immobilization	Nominal
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**Results Remarks:** 48 hr results-number of organisms affected and analytical results

Treatment	Measured		Measured	
	Immobilization	BTEXN -day 0	BTEXN -day 2	
Control	2	ND	ND	
0.1 mg/l	1	0.12	0.20	
1.0 mg/l	1	0.31	0.42	
5.0 mg/l	1	1.7	1.4	
10 mg/l	5	3.1	3.2	
25 mg/l	20	7.7	7.1	

based upon nominal loading rate 48-hr EL50 = 12 mg/l (95% C.I. 7.3 to 22 mg/l)

### Reliability/Data Quality - Acute Toxicity To Aquatic Invertebrates

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Three previous attempts to conduct study were invalidated due to excessive (>20%) control mortality. Two more studies (this study and one other) had acceptably low mortality rates and were considered valid.

Guideline study conducted under GLPs

**Key Study Sponsor Indicator:** Key

### Reference - Acute Toxicity To Aquatic Invertebrates

**Reference:** CONCAWE (1995) Daphnia -acute toxicity test: study no. 104942A, test substance MRD-95-049. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.

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## Acute Toxicity to Aquatic Invertebrates

### Test Substance - Acute Toxicity To Aquatic Invertebrates

<b>Category Chemical:</b>	(64741-70-4) Naphtha, petroleum, isomerization
<b>Test Substance:</b>	(64741-70-4) Naphtha, petroleum, isomerization
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	PARAFFINIC NAPHTHA CAS 64741-70-4, CONCAWE sample W94/810 Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Invertebrates</b>	
<b>Year Study Performed:</b>	1996
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species/In Vitro System:</b>	Daphnia magna
<b>GLP:</b>	
<b>Analytical Monitoring:</b>	
<b>Test Type:</b>	
<b>Test Vessel:</b>	
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	maximum loadings of 50 mg/l or less
<b>Exposure Period:</b>	48 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	
<b>Test Temperature and Units:</b>	

<b>Photo (Light/Dark):</b>									
<b>Salinity:</b>									
<b>TOC:</b>									
<b>Water Hardness:</b>									
<b>Method/Guideline Test Conditions Remarks:</b>									
Experimental studies with fish, invertebrates and algae tested in closed systems with minimal head-space were performed on WAFS of low boiling point naphthas prepared at maximum loadings of 50 mg/l or less. Results show that acute aquatic toxicity lethal loading (LL), effective loading (EL) or inhibition of growth rate values (IrL) affecting 50% of the organism population are greater than 1 mg/l and mostly in the range 1-100 mg/l. Summarized CONCAWE test data indicating the extent of aquatic toxicity are as follows in Results Remarks Section, and 95% confidence intervals are included in parentheses.									
<b>Limit Test:</b>									
<b>Test Results - Acute Toxicity To Aquatic Invertebrates</b>									
<b>NOEC Exposure Duration:</b>									
<b>NOEC:</b>									
<b>LOEC Exposure Duration:</b>									
<b>LOEC:</b>									
<b>NOELR Exposure Duration:</b>									
<b>NOELR:</b>									
<b>LOELR Exposure Duration:</b>									
<b>LOELR:</b>									
<b>LC/EC Mean Value:</b>									
<b>Exposure Duration</b>	<b>Exposure Units</b>	<b>LC/EC</b>	<b>%</b>	<b>Value Description</b>	<b>Mean Value or Lower Mean Value</b>	<b>Upper Mean Value</b>	<b>Units</b>	<b>Effect Observed</b>	<b>Basis for Concentration</b>
48	Hours	EL	50 %	=	10		mg/L		Nominal
<b>Results Remarks:</b>									
PARAFFINIC NAPHTHA CAS 64741-70-4, CONCAWE sample W94/810									
Invertebrate (Daphnia magna) EL50, 48h 10 mg/l (8.5-13)									
Fish (Oncorhynchus mykiss) LL50, 96h 10 mg/l (5-23)									
Algae (Selenastum capricornutum) IrL50, 72h >50 mg/l (not calculable)									
95% confidence intervals are included in parentheses									
<b>Reliability/Data Quality - Acute Toxicity To Aquatic Invertebrates</b>									
<b>Reliability:</b>									
<b>Reliability Remarks:</b>									
<b>Key Study Sponsor Indicator:</b>									
Key									
<b>Reference - Acute Toxicity To Aquatic Invertebrates</b>									
<b>Reference:</b>									
CONCAWE. 1996. Acute Aquatic Toxicity of Gasolines - Report on CONCAWE Test Programme. Report No. 96/57. CONCAWE, Brussels, Belgium.									



<b>Acute Toxicity to Aquatic Invertebrates</b>	
<b>Test Substance - Acute Toxicity To Aquatic Invertebrates</b>	
<b>Category Chemical:</b>	(64741-54-4) Naphtha, petroleum, heavy catalytic cracked
<b>Test Substance:</b>	(64741-54-4) Naphtha, petroleum, heavy catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	OLEFINIC NAPHTHA CAS . 64741-54-4, CONCAWE sample W94/811 Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Invertebrates</b>	
<b>Year Study Performed:</b>	1996
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species/In Vitro System:</b>	Daphnia magna
<b>GLP:</b>	
<b>Analytical Monitoring:</b>	
<b>Test Type:</b>	
<b>Test Vessel:</b>	
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	maximum loadings of 50 mg/l or less
<b>Exposure Period:</b>	48 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	
<b>Test Temperature and Units:</b>	

**Photo  
(Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline  
Test Conditions  
Remarks:**

Experimental studies with fish, invertebrates and algae tested in closed systems with minimal head-space were performed on WAFS of low boiling point naphthas prepared at maximum loadings of 50 mg/l or less. Results show that acute aquatic toxicity lethal loading (LL), effective loading (EL) or inhibition of growth rate values (IrL) affecting 50% of the organism population are greater than 1 mg/l and mostly in the range 1-100 mg/l. Summarized CONCAWE test data indicating the extent of aquatic toxicity are as follows in Results Remarks Section, and 95% confidence intervals are included in parentheses.

**Limit Test:**

### Test Results - Acute Toxicity To Aquatic Invertebrates

**NOEC Exposure  
Duration:**

**NOEC:**

**LOEC Exposure  
Duration:**

**LOEC:**

**NOELR Exposure  
Duration:**

**NOELR:**

**LOELR Exposure  
Duration:**

**LOELR:**

**LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration
48	Hours	EL	50 %	=	13		mg/L		Nominal

**Results Remarks:**

OLEFINIC NAPHTHA CAS . 64741-54-4, CONCAWE sample W94/811  
 Invertebrate (Daphnia magna) EL50, 48h 13 mg/l (12-15)  
 Fish (Oncorhynchus mykiss) LL50, 96h 15 mg/l (10-23)  
 Algae (Selenastum capricornutum) IrL50, 72h 3.1 mg/l (3.6-14)  
 95% confidence intervals are included in parentheses

### Reliability/Data Quality - Acute Toxicity To Aquatic Invertebrates

**Reliability:**

**Reliability  
Remarks:**

**Key Study Sponsor  
Indicator:** Key

### Reference - Acute Toxicity To Aquatic Invertebrates

**Reference:**

CONCAWE. 1996. Acute Aquatic Toxicity of Gasolines - Report on CONCAWE Test Programme. Report No. 96/57. CONCAWE, Brussels, Belgium.





<b>Acute Toxicity to Aquatic Plants</b>	
<b>Test Substance - Acute Toxicity To Aquatic Plants</b>	
<b>Category Chemical:</b>	No CAS Number Provided
<b>Test Substance:</b>	No CAS Number Provided
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Read-Across
<b>Test Substance Result Type:</b>	
<b>Method - Acute Toxicity To Aquatic Plants</b>	
<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	
<b>GLP:</b>	
<b>Analytical Monitoring:</b>	
<b>Test Type:</b>	
<b>Test Vessel:</b>	
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	
<b>Nominal and Measured Concentrations:</b>	
<b>Exposure Period:</b>	
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	
<b>Test Temperature and Units:</b>	

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline**

**Test Conditions**

**Remarks:**

**Limit Test:**

**Test Results - Acute Toxicity To Aquatic Plants**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:**

**NOELR:**

**LOELR Exposure Duration:**

**LOELR:**

**Effect:**

Exposure Duration	Exposure Units	Type	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Basis for Effect	Basis for Concentration
72	Hours	EL	50 %	=	1.1	64	mg/L		Nominal

**Results Remarks:** The acute toxicity of gasoline blending streams to freshwater algae is expected to fall within the approximate range 1.1 - 64 mg/L (lethal loading rates). This range of acute toxicity values can be used as "read-across" to untested members of the gasoline blending streams category.

**Reliability/Data Quality - Acute Toxicity To Aquatic Plants**

**Reliability:**

**Reliability Remarks:** RELIABILITY: Studies used to develop the read across range had reliability ratings of either "1 = reliable without restriction" or "2- reliable with restrictions".

**Key Study Sponsor Indicator:** Weight of Evidence

**Reference - Acute Toxicity To Aquatic Plants**

**Reference:** See Robust Study Summaries for CAS #: 64741-46-4, 64741-54-4, 64741-55-5, 64741-63-5, 64741-66-8, 64741-70-4, 68955-35-1, and 86290-81-5



<b>Acute Toxicity to Aquatic Plants</b>	
<b>Test Substance - Acute Toxicity To Aquatic Plants</b>	
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Gasoline CAS No. 86290-81-5 Gasoline Sample W94/814, Blend Detailed hydrocarbon analysis: N-paraffins: 16% total C4-C8 Iso-paraffins: 25% total C4-C11 Olefins: 12% C4-C7 Naphthenes: 5% C6-C10 Aromatics: 42% C6-C11  Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>  [Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Plants</b>	
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	OECD 201
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Selenastrum capricornutum
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	No
<b>Test Type:</b>	Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 0.5, 1, 5, 10, and 25 mg/l
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	

<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	7.6 Upper Range: 9.5
<b>Test Temperature and Units:</b>	Value/Lower Range: 23 °C
<b>Photo (Light/Dark):</b>	
<b>Salinity:</b>	
<b>TOC:</b>	
<b>Water Hardness:</b>	
<b>Method/Guideline Test Conditions Remarks:</b>	<p>EL50 values were calculated using the inverse extrapolation method of Snedecor and Cochran, Statistical Methods, 8th Ed., 1989, Iowa State University Press/Ames. NOEL values calculated using ANOVA (Duncan D.B., 1975, Biometrics, 31, 339-359)</p> <p>Individual test treatment solutions were prepared as Water Accommodated Fractions (WAFs). To determine contact time required to achieve maximum solubility between test substance and aqueous solution, samples of WAF test solutions equilibrated for 20, 24, 27 and 49 hours at 100 mg/l loading were analyzed by GC-FID for concentrations of the following: benzene, toluene, ethylbenzene, xylene isomers and naphthalene (BTEXN). Maximum WAF solubility for these components reached equilibrium within 24 hrs of stirring. Nominal loading rates of 0, 0.5, 1, 5, 10, and 25 mg/l were used to prepare test solutions for the algal toxicity tests. Test material was added volumetrically to 2.0 liters of sterilized algal nutrient media (enriched with 100 mg/l of sodium bicarbonate) in 2.0 liter aspirator bottles covered with aluminum foil. The mixing vessels were sealed with foil covered stoppers and mixed on magnetic stir plates with teflon coated stir bars for approximately 24 hours at room temperature. After mixing the solutions were allowed to settle for one hour and the WAF was removed and used for testing. Test vessels were 125ml glass Erlenmeyer flasks containing ten 4mm glass balls that were completely filled (140 ml) with treatment solution, inoculated with algae and sealed with glass stoppers. Algal cells were obtained from 6 day old laboratory stock cultures maintained in nutrient enriched media, at 24 °C (±2°) C under continuous illumination of 4300(±10%) lux. Original algal cultures (Strain 1648) were provided by the Department of Botany, University of Texas. Cell density of the algal stock culture inoculum was determined prior to study initiation with a Turner filter-fluorometer. Fluorometer readings were converted to cell number s using a regression formula developed through cell counts. Three replicates were prepared for each treatment level and six replicates were prepared as control systems. The initial algal concentration was approximately 1.0 x 10<sup>3</sup> cells/ml in each replicate chamber. All test replicates were placed on a shaker table at 150 oscillations per minute during the study and exposed to continuous fluorescent light, illumination at 4300 to 4400 Lux as measured daily using a Licor photometric sensor. A sample volume of 3.5 ml was taken daily for density determinations, and an equivalent volume of reserve 24 hour WAF was used to replenish the displaced sample volume. WAF test solutions were analyzed by GC FID for concentrations of BTEXN at day 0 and 96 hr termination. BTEXN total concentration at termination was at least 80% of the initial concentration for all treatments, with the exception of the 5.0 mg/l exposure, which showed a loss of 73%. This excessive loss compared to the other treatments was determined to be due to sampling technique. Test temperature was 23 °C (sd=0.08 C. The average pH was 7.5 at initiation; and ranged from 9.2 (control) to 7.8 (25 mg/l loading) at termination.</p>
<b>Limit Test:</b>	No
<b>Test Results - Acute Toxicity To Aquatic Plants</b>	
<b>NOEC Exposure Duration:</b>	
<b>NOEC:</b>	
<b>LOEC Exposure Duration:</b>	

**LOEC:**

**NOELR Exposure Duration:**

**NOELR:**

**LOELR Exposure Duration:**

**LOELR:**

**Effect:**

Exposure Duration	Exposure Units	Type	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Basis for Effect	Basis for Concentration
72	Hours	EL	50 %	=	3.3		mg/L	Growth Rate	Nominal
72	Hours	EL	50 %	=	4.2		mg/L	Biomass	Nominal
96	Hours	EL	50 %	=	2.5		mg/L	Growth Rate	Nominal
			%						

**Results Remarks:**

Percent inhibition:  
 72 hour EL50 for average growth rate=3.3 mg/l(0.24 to >25 mg/l CI @95% )  
 72 hour EL50 for area under the growth curve=4.2 mg/l(0 to 24 mg/l CI @95%)  
 96 hour EL50 for average growth rate=2.5 mg/l(0.62 to 14 mg/l CI @95%)  
  
 72 hour NOEL for average growth rate and area under the growth curve =0.5 mg/l  
 96 hour NOEL for average growth rate =0.5 mg/l  
 96 hour NOEL for area under the growth curve =0.5 mg/l

Nominal (mg/l)	Average		% Inhibition		Area	
	cell density (cells/ml)		growth rate		under growth curve	
	72hr	96hr	72hr	96 hr	72hr	96hr
Control	9.9E4	3.8E5	0	0	0	0
0.5	7.7E4	2.6E5	7.7	7.8	21	27
1.0	5.5E4	1.7E5	15	17	36	50
5.0	2.5E4	2.2E4	33	51	54	81
10	3.7E3	2.0E3	76	95	90	97
25	BMDL	BMDL	99	100	98	99

BMDL=below method detection limit

Analytical results

Nominal (mg/l)	Measured Concentration (mg/l as BTEXN)	
	Day 0	Day 4
Control	none detected	none detected
0.5	0.22	0.23
1.0	0.47	0.51
5.0	1.5	1.3
10	3.5	3.3
25	9.5	7.7

**Reliability/Data Quality - Acute Toxicity To Aquatic Plants**

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP; guideline study

**Key Study Sponsor Indicator:** Key

**Reference - Acute Toxicity To Aquatic Plants**

**Reference:** CONCAWE (1995) Algal, Growth Inhibition Test: study no. 104967, test substance MRD-95-049. Study conducted by Exxon Biomedical Sciences Inc. CONCAWE, Brussels, 1995.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Acute Toxicity to Aquatic Plants</b>	
<b>Test Substance - Acute Toxicity To Aquatic Plants</b>	
<b>Category Chemical:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Aromatic naphtha Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Plants</b>	
<b>Year Study Performed:</b>	1998
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Selenastrum capricornutum
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 1.3, 2.5, 5.0, 10, 20 and 40 mg/l
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	7.5 Upper Range: 8.9
<b>Test Temperature and Units:</b>	Value/Lower Range: 24 Upper Range: 25 °C

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline Test Conditions Remarks:**

No specific guideline was described as being used to conduct the test, however report references 1991 ASTM method E729-88a and 1982 EPA Support Documents for Environmental Testing: EPA 560/6-82-002.

Statistical Method: EL 50 and EC50 calculated using nonlinear logistics sigmoid model (SAS). All NOEL/NOEC values based on visual review and Dunnett's test for significance.

Individual test treatment solutions were prepared as Water Accommodated Fractions (WAFs). Test material was added to 2.4 liters of sterilized AAP test media (enriched with 515 mg/l of sodium bicarbonate, 300 ug/l EDTA chelator, pH adjusted to 7.5 ±0.1 with 0.1NHCl and sterilized by 0.45 micron filtration) in 2.5 liter aspirator bottles. The mixing vessels were sealed with foil covered stoppers, covered with aluminum foil and the contents mixed on magnetic stir plates with teflon coated stir bars for approximately 24 hours at room temperature. After mixing the solutions were allowed to settle for one hour and the WAF was removed from the spout at the base of each bottle and used for testing. Test vessels were 125ml glass Erlenmeyer flasks that were completely filled (148 ml) with treatment solution and inoculated with 6 day old algae. Algal cells obtained from testing laboratory cultures grown in sterile, nutrient enriched AAP media. Original algal cultures (stock UTEX-1648) obtained from Dept of Botany, Culture Collection of Algae, University of Texas at Austin, 1997. Cell density of the algal stock culture inoculate was determined prior to study initiation with a hemacytometer cell and compound microscope. Twelve replicates were prepared for each treatment level. Nominal treatment levels were 0, 1.3, 2.5, 5.0, 10, 20 and 40 mg/l. The initial algal concentration was 1.0 x 10<sup>3</sup> cells/ml. All test replicates were placed on a shaker table at 100 oscillations per minute during the study and exposed to continuous fluorescent light, illumination range 371 to 442 ft candles. Triplicate samples were taken daily for cell counts and analytical testing. Cell densities were determined by direct microscopic examination. Samples at 0, 24, 48, 72 and 96 hrs were also analyzed by Purge & trap/GC-FID for concentrations of the following: pentane, 2 methyl-pentane, benzene, toluene, ethylbenzene, ortho, meta and para-xylene, which represent more than 50% composition of the test substance. Measured test concentrations of the light catalytically reformed naphtha were based on the total combined concentrations of all analytes.

Test temperature was 24-25 °C. Test solution pH ranged from 7.5 to 8.9.

**Limit Test:** No

**Test Results - Acute Toxicity To Aquatic Plants**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:** 96 Hours

**NOELR:** = 5 mg/L Nominal

**LOELR Exposure Duration:**

**LOELR:**

**Effect:**

Exposure Duration	Exposure Units	Type	%	Value Description	Mean Value or Lower	Upper Mean Value	Units	Basis for Effect	Basis for Concentration



				Mean Value				
96	Hours	EL	50 %	8.5		mg/L	Cell Number	Calculated
			%					
96	Hours	EL	10 %	6	=	mg/L	Cell Number	Calculated
96	Hours	EL	90 %	12		mg/L	Cell Number	Calculated

**Results Remarks:**

Percent inhibition on growth determined by cell density (cells/mL):

96 hour EL10=6.0 mg/l (3.1-8.8 mg/l CI @95% )

96 hour EL50=8.5mg/l (7.3-9.8 mg/l CI @95% )

96 hour EL90=12 mg/l (9.9-14 mg/l CI @95% )

96 hour NOEL=5.0 mg/l

96 hour EC10=1.1 mg/l (0.41-1.8 mg/l CI @95% )

96 hour EC50= 1.7mg/l (1.4-2.1 mg/l CI @95% )

96 hour EC90=2.7 mg/l (2.1-3.4 mg/l CI @95% )

96 hour NOEC=0.866 mg/l

Subcultures of the 10, 20 and 40 mg/l treatment cultures were placed in fresh media (no test substance) after acute testing for ten days indicated that growth inhibition was algistatic in all treatments. No excursions from the protocol were noted which would have affected the integrity of the study.

## Concentration

nominal (mg/l)	measured (mg/l)	96hr cell density (cells/ml)
Control	0.0147	40.5 x10 <sup>4</sup>
1.3	0.126	40.92 x10 <sup>4</sup>
2.5	0.211	42.33 x10 <sup>4</sup>
5.0	0.866	41.17 x10 <sup>4</sup>
10	2.12	11.11 x10 <sup>4</sup>
20	5.26	0.70 x10 <sup>4</sup>
40	13.3	0.04 x10 <sup>4</sup>

Measured concentrations represented the sum of six specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, it was not reported to what degree measured concentrations declined between the beginning and end of the test. Because of this uncertainty, test endpoints based on the measured components should be viewed and interpreted with an understanding of this limitation.

**Reliability/Data Quality - Acute Toxicity To Aquatic Plants****Reliability:**

Valid with Restrictions

**Reliability Remarks:**

Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:**

Key

**Reference - Acute Toxicity To Aquatic Plants****Reference:**

ABC Laboratories, Inc. (1998). Static Renewal 96 Hour Acute Toxicity of the Water Accomodated Fraction (WAF) of Light Catalytically Reformed Naphtha, LCRN to to a Freshwater Alga, Selenastrum capricornutum. Project ID. 43579. ABC Laboratories, Inc. Environmental Toxicology, 7200 E. ABC Lane, Columbia, Missouri.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Acute Toxicity to Aquatic Plants</b>	
<b>Test Substance - Acute Toxicity To Aquatic Plants</b>	
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Paraffinic naphthas Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Plants</b>	
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	Other
<b>Other Method/Guideline:</b>	EPA. 1982. Guidelines and Support Documents for Environmental Effects Testing. EPA 560/6-82-002. Sections EG-8, ES-5.
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Selenastrum capricornutum
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 18, 70, 146, 292 and 1157 mg/l
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	7.5
<b>Test Temperature and Units:</b>	Value/Lower Range: 22 Upper Range: 26 °C

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline Test Conditions Remarks:**

Statistical Method: EL50 and EC50 calculated using binomial probability analysis. ASTM Special Technical Publication 634. 1977, pp 65-84. All NOEL/NOEC values calculated using Fisher's exact test.

Individual test treatment solutions were prepared as Water Accommodated Fractions (WAFs). Test material was added to 2.3 liters of sterilized AAP test media (enriched with 515 mg/l of sodium bicarbonate, pH adjusted to 7.5 ± 0.1 with 0.1 NHCl and sterilized by 0.22 micron filtration) in 2.0 liter aspirator bottles, covered with aluminum foil. The mixing vessels were sealed with foil covered stoppers and mixed on magnetic stir plates with teflon coated stir bars for approximately 24 hours at room temperature. After mixing the solutions were allowed to settle for one hour and the WAF was removed and used for testing. Test vessels were 125ml glass Erlenmeyer flasks that were completely filled (135 ml) with treatment solution and inoculated with algae. Algal cells obtained from testing laboratory cultures grown in sterile, nutrient enriched AAP media, and transferred every 5-9 days to fresh media. Original algal cultures obtained from American Type Culture Collection (ATCC Strain 22662), Rockville, MD, June 1994. Cell density of the algal stock culture inoculate was determined prior to study initiation with a hemacytometer cell and compound microscope. Twelve replicates were prepared for each treatment level. Nominal treatment levels were 0, 18, 70, 146, 292 and 1157 mg/l The initial algal concentration was 1.0 x 10<sup>3</sup> cells/ml. All test replicates were placed on a shaker table at 100 oscillations per minute during the study and exposed to continuous fluorescent light, illumination at 400 ± 50-ft candles. Triplicate samples were taken daily for cell counts and analytical testing. Cell densities were determined by direct microscopic examination Samples at 0,24, 48, 72 and 96 hrs were also analyzed by Purge & trap/GC-FID for concentrations of the following: 2,3 dimethyl but ane; 2,4 dimethyl pentane; 2,2,4 trimethyl pentane; 2,5 dimethyl hexane; 2,3,4 trimethyl pentane, 2,3,3 trimethyl pentane and 1-methyl-1-ethyl cyclopentane, which represent 68% composition of the test substance. Measured test concentrations of the light alkylate naphtha were based on the total combined concentrations of all analytes.

Test temperature was 24 ± 2°C. The pH was 7.5 at test initiation, pH value at test termination not included in report.

**Limit Test:** No

**Test Results - Acute Toxicity To Aquatic Plants**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:** 96 Hours

**NOELR:** = 18 mg/L Nominal

**LOELR Exposure Duration:**

**LOELR:**

**Effect:**

Exposure Duration	Exposure Units	Type	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Basis for Effect	Basis for Concentration

96	Hours	EL	50	=	45	mg/L	Cell	Calculated
			%				Number	
			%					

**Results Remarks:**

Percent inhibition on growth determined by cell density (cells/ml):  
 96 hour EL50=45mg/l (18-70 mg/l CI @95% )  
 96 hour NOEL=18 mg/l

Subcultures placed in fresh media (no test substance) after acute testing for nine days indicated that growth inhibition was algistatic in all treatments. No excursions from the protocol were noted. However, range finding and two previous definitive tests were performed and considered inconclusive due to inconsistencies in control and treatment cell densities, which presumably were resolved by modification of the AAP media. Additionally, control growth showed a lag during the first 48 hours of the study.

Concentration (mg/l)		96hr cell density (cells/ml)	(% Inhibition)
Nominal mg/L	Measured ug/L		
Control		5.7x104	na
18	0.112	5.53x104	3.1
70	0.305	1.27x104	77.7
146	0.498	3.46x103	93.9
292	0.610	1.36x103	97.6
1157	0.612	1.60x103	97.2

Measured concentrations represented the sum of seven specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, it was not reported to what degree measured concentrations declined between the beginning and end of the test. Because of this uncertainty, test endpoints based on the measured components should be viewed and interpreted with an understanding of this limitation.

**Reliability/Data Quality - Acute Toxicity To Aquatic Plants**

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP study with adequately detailed methods description (guideline = "other")

**Key Study Sponsor Indicator:** Key

**Reference - Acute Toxicity To Aquatic Plants**

**Reference:** Stonybrook Laboratories, Inc. (1995) Static Renewal 96-hour acute toxicity of the water accommodated fraction (WAF) of Whole Light Alkylate Naphtha (LAN) Product to a Freshwater Alga, *Selenastrum capricornutum*, Study No. 65909. Stonybrook Laboratories Inc. Princeton, NJ.1995.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Acute Toxicity to Aquatic Plants</b>	
<b>Test Substance - Acute Toxicity To Aquatic Plants</b>	
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Olefenic naphthas Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Plants</b>	
<b>Year Study Performed:</b>	1995
<b>Method/Guideline Followed:</b>	Other
<b>Other Method/Guideline:</b>	EPA. 1982. Guidelines and Support Documents for Environmental Effects Testing. EPA 560/6-82-002. Sections EG-8, ES-5.
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Selenastrum capricornutum
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 6.4, 13, 25, 51 and 102 mg/l
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	7.5
<b>Test Temperature and Units:</b>	Value/Lower Range: 22 Upper Range: 26 °C

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline Test Conditions Remarks:** Statistical Method: LL50 and LC50 calculated using probit analysis. ASTM Special Technical Publication 634. 1977, pp 65-84. All NOEL/NOEC values calculated using Fisher's exact test.

Individual test treatment solutions were prepared as Water Accommodated Fractions (WAFs). Test material was added to 4.4 liters of sterilized AAP test media (enriched with 515 mg/l of sodium bicarbonate, pH adjusted to 7.5 ± 0.1 with 0.1NHCl and sterilized by 0.22 micron filtration) in 4.0 liter aspirator bottles. The mixing vessels were sealed with foil covered stoppers and mixed on magnetic stir plates with teflon coated stir bars for approximately 24 hours at room temperature in a hood darkened with aluminum foil. After mixing the solutions were allowed to settle for one hour and the WAF was removed and used for testing. Test vessels were 125ml glass Erlenmeyer flasks that were completely filled (140 ml) with treatment solution and inoculated with algae. Algal cells obtained from testing laboratory cultures grown in sterile, nutrient enriched AAP media, and transferred every 4-8 days to fresh media. Original algal cultures obtained from American Type Culture Collection (ATCC Strain 22662), Rockville, MD, September 1995. Cell density of the algal stock culture inoculate was determined prior to study initiation with a hemacytometer cell and compound microscope. Twelve replicates were prepared for each treatment level. Nominal treatment levels were 0, 6.4, 13, 25, 51 and 102 mg/l. The initial algal concentration was 1.0 x 10<sup>3</sup> cells/ml. All test replicates were placed on a shaker table at 100 oscillations per minute during the study and exposed to continuous fluorescent light, illumination at 400 + 50-ft candles. Triplicate samples were taken daily for cell counts and analytical testing. Cell densities were determined by direct microscopic examination. Samples at 0, 24, 48, 72 and 96 hrs were also analyzed by Purge & trap/GC-FID for concentrations of the following: benzene, toluene, ethylbenzene, o-xylene and p-xylene, which represent 13% composition of the test substance. Measured test concentrations of the light catalytically cracked naphtha were based on the total combined concentrations of all analytes.

Test temperature was 24 ± 2 °C. The pH was 7.5 at test initiation, pH value at test termination not included in report.

**Limit Test:** No

**Test Results - Acute Toxicity To Aquatic Plants**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:** 96 Hours

**NOELR:** = 51 mg/L Nominal

**LOELR Exposure Duration:**

**LOELR:**

**Effect:**

Exposure Duration	Exposure Units	Type	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Basis for Effect	Basis for Concentration
96	Hours	EL	50 %	=	64		mg/L	Cell Number	Calculated
			%						

**Results Remarks:**

Percent inhibition on growth determined by cell density (cells/ml):

96 hour EL50=64 mg/l (44-111 mg/l CI @95% )

96 hour EC50= 4.6mg/l (2.9-8.8 mg/l CI @95% )

96 hour NOEL=51 mg/l

96 hour NOEC=3.5 mg/l

Subcultures placed in fresh media (no test substance) after acute testing for six days indicated that growth inhibition was algistatic in all treatments, with the exception of the 102 ppm, which was determined to be algicidal. No excursions from the protocol were noted. However, range finding and two previous definitive tests were performed and considered inconclusive due to inconsistencies in control and treatment cell densities, which presumably were resolved by modification of the AAP media. Additionally, control growth showed a lag during the first 72 hours of the study.

Concentration (mg/l)		96hr cell density	
Nominal	Measured	(cells/ml)	(% Inhibition)
Control		8.4 x103	na
6.4	0.093	3.2 x104	-281.1
13	0.130	9.73x103	-16.00
25	0.429	1.99x104	-136.9
51	1.87	1.36x103	53.0
102	4.85	2.59x103	69.2

Measured concentrations represented the sum of four specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, it was not reported to what degree measured concentrations declined between the beginning and end of the test. Because of this uncertainty, test endpoints based on the measured components should be viewed and interpreted with an understanding of this limitation.

**Reliability/Data Quality - Acute Toxicity To Aquatic Plants****Reliability:**

Valid with Restrictions

**Reliability Remarks:**

Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:**

Key

**Reference - Acute Toxicity To Aquatic Plants****Reference:**

Stonybrook Laboratories, Inc. (1995) Static Renewal 96-hour acute toxicity of the water accommodated fraction (WAF) FR 15799 FCC Light (Light Catalytically Cracked Naphtha, LCCN) to a Freshwater Alga, *Selenastrum capricornutum*, Study No. 66235 Stonybrook Laboratories, Inc Princeton, NJ.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Acute Toxicity to Aquatic Plants</b>	
<b>Test Substance - Acute Toxicity To Aquatic Plants</b>	
<b>Category Chemical:</b>	(64741-46-4) Naphtha, petroleum, light straight-run
<b>Test Substance:</b>	(64741-46-4) Naphtha, petroleum, light straight-run
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	LSRN-Moderate naphthenic content Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Plants</b>	
<b>Year Study Performed:</b>	1997
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Selenastrum capricornutum
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 1.9, 4.0, 7.8, 16 and 31 mg/l
<b>Exposure Period:</b>	96 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	8 Upper Range: 8.5
<b>Test Temperature and Units:</b>	Value/Lower Range: 24 Upper Range: 26



<b>Photo (Light/Dark):</b>	
<b>Salinity:</b>	
<b>TOC:</b>	
<b>Water Hardness:</b>	
<b>Method/Guideline</b>	No specific guideline was described as being used to conduct the test, however report references 1991 ASTM method E729-88a and 1982 EPA Support Documents for Environmental Testing: EPA 560/6-82-002
<b>Test Conditions</b>	
<b>Remarks:</b>	<p>Statistical Method: EL50 and EC50 calculated using nonlinear logistics sigmoid model (SAS). All NOEL/NOEC values based on visual review and Dunnett's test for significance.</p> <p>Individual test treatment solutions were prepared as Water Accommodated Fractions (WAFs). Test material was added to 9.4-9.6l of sterilized AAP test media (enriched with 515 mg/l of sodium bicarbonate, 300 ig/l EDTA chelator, pH adjusted to <math>7.5 \pm 0.1</math> with 0.1 NHCl and sterilized by 0.45 micron filtration) in 9.5 liter aspirator bottles. A measured amount of test substance was added into each bottle, and the bottles were capped tightly with a positive pressure siphoning apparatus. The siphoning apparatus consisted of a teflon lined neoprene stopper housing two teflon tubes. One tube extended to the bottom of the bottle for removal of the WAF solution, the other tube ended above the WAF surface, and was used to control air pressure during siphoning. During WAF preparation, parafilm was used to seal The external joint between the neoprene stopper and glass bottle, and the bottles were covered with aluminum foil. The contents were stirred with teflon coated stir bars in the mixing vessels which were placed on magnetic stir plates at room temperature. After stirring for 24 hrs using 25% or less vortex, the contents of the WAF solution bottles were allowed to settle for approximately 45 minutes to two hours, then siphoned by the positive pressure apparatus port and used for testing. Test vessels were 125ml glass Erlenmeyer flasks that were completely filled (148 ml) with treatment solution and inoculated with 3 day old algae. Algal cells obtained from testing laboratory cultures grown in sterile, nutrient enriched AAP media. Original algal cultures (stock UTEX-1648) obtained from Dept of Botany, Culture Collection of Algae, University of Texas at Austin, 1996. Cell density of the algal stock culture inoculate was determined prior to study initiation with a hemacytometer cell and compound microscope. Twelve replicates were prepared for each treatment level. Nominal treatment levels were 0, 1.9, 4.0, 7.8, 16 and 31 mg/l The initial algal concentration was <math>1.0 \times 10^3</math> cells/ml.</p> <p>All test replicates were placed on a shaker table at 100 oscillations per minute during the study and exposed to continuous fluorescent light, illumination 400 +50 ft candles. Triplicate samples were taken daily for cell counts and analytical testing. Cell densities were determined by direct microscopic examination. Samples at 0,24, 48, 72 and 96 hrs were also analyzed by Purge &amp; trap/GC-FID for concentrations of the following: 2-methyl-pentane, cyclohexane, benzene, toluene, ethylbenzene, ortho, meta and para-xylene. Measured test concentrations of the light straight run naphtha were based on the total combined concentrations of all analytes.</p> <p>Test temperature was 24-26 °C. Test solution pH ranged from 8.0 to 8.5.</p>
<b>Limit Test:</b>	No
<b>Test Results - Acute Toxicity To Aquatic Plants</b>	
<b>NOEC Exposure Duration:</b>	
<b>NOEC:</b>	
<b>LOEC Exposure Duration:</b>	
<b>LOEC:</b>	
<b>NOELR Exposure Duration:</b>	96 Hours
<b>NOELR:</b>	$\approx$ 1.9 mg/L Nominal
<b>LOELR Exposure Duration:</b>	

**LOELR:****Effect:**

Exposure Duration	Exposure Units	Type	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Basis for Effect	Basis for Concentration
96	Hours	EL	10 %	=	2.7		mg/L	Cell Number	Calculated
96	Hours	EL	50 %	=	6.4		mg/L	Cell Number	Calculated
96	Hours	EL	90 %	=	15		mg/L	Cell Number	Calculated
			%						

**Results Remarks:**

Percent inhibition on growth determined by cell density (cells/ml):  
 96 hour EL10=2.7 mg/l (1.9-3.5 mg/l CI @95% )  
 96 hour EL50=6.4mg/l (5.7-7.1 mg/l CI @95% )  
 96 hour EL90=15 mg/l (12-18 mg/l CI @95% )  
 96 hour NOEL=1.9 mg/L

96 hour EC10=0.1 mg/l (0.061-0.15 mg/l CI @95% )  
 96 hour EC50= 0.26 mg/l (0.22-0.30 mg/l CI @95% )  
 96 hour EC90=0.66 mg/l (0.50-0.83 mg/l CI @95% )  
 96 hour NOEC=0.0326 mg/l

Subcultures of the 31 mg/l treatment cultures were placed in fresh media (no test substance) after acute testing for ten days and indicated that growth inhibition was algistic in this treatment. Conduct of the range-finder and definitive tests were acceptable (no repeats). No excursions from the protocol were noted which would have affected the integrity of the study.

**Concentration**

Nominal (mg/L)	Measured (mg/L)	96hr cell density (cells/ml)	
Control	1.9	0.0322	42.33 x104
4.0	0.130	29.25	x104
7.8	0.329	18.42	x104
16	0.704	1.74	x104
31	1.29	0.04	x104

Measured concentrations represented the sum of six specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, it was not reported to what degree measured concentrations declined between the beginning and end of the test. Because of this uncertainty, test endpoints based on the measured components should be viewed and interpreted with an understanding of this limitation.

**Reliability/Data Quality - Acute Toxicity To Aquatic Plants****Reliability:**

Valid with Restrictions

**Reliability Remarks:**

Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:**

Key

**Reference - Acute Toxicity To Aquatic Plants****Reference:**

ABC Laboratories, Inc. (1998) Static Renewal 96 Hour Acute Toxicity of the Water Accomodated Fraction (WAF) of Light Straight Run Naphtha, LSRN to a Freshwater Alga, Selenastrum capricornutum. Project ID. 43151. Environmental Toxicology, 7200 E. ABC Lane, Columbia, Missouri.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



## Acute Toxicity to Aquatic Plants

### Test Substance - Acute Toxicity To Aquatic Plants

<b>Category Chemical:</b>	(64741-70-4) Naphtha, petroleum, isomerization
<b>Test Substance:</b>	(64741-70-4) Naphtha, petroleum, isomerization
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	PARAFFINIC NAPHTHA CAS 64741-70-4, CONCAWE sample W94/810 Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured

### Method - Acute Toxicity To Aquatic Plants

<b>Year Study Performed:</b>	1996
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Selenastrum sp.
<b>GLP:</b>	
<b>Analytical Monitoring:</b>	
<b>Test Type:</b>	
<b>Test Vessel:</b>	
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	maximum loadings of 50 mg/l or less
<b>Exposure Period:</b>	72 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	
<b>Test Temperature and Units:</b>	

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline Test Conditions Remarks:**

Experimental studies with fish, invertebrates and algae tested in closed systems with minimal head-space were performed on WAFS of low boiling point naphthas prepared at maximum loadings of 50 mg/l or less. Results show that acute aquatic toxicity lethal loading (LL), effective loading (EL) or inhibition of growth rate values (IrL) affecting 50% of the organism population are greater than 1 mg/l and mostly in the range 1-100 mg/l. Summarized CONCAWE test data indicating the extent of aquatic toxicity are as follows in Results Remarks Section, and 95% confidence intervals are included in parentheses.

**Limit Test:**

**Test Results - Acute Toxicity To Aquatic Plants**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:**

**NOELR:**

**LOELR Exposure Duration:**

**LOELR:**

**Effect:**

Exposure Duration	Exposure Units	Type	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Basis for Effect	Basis for Concentration
72	Hours	EL	50 %	>	50		mg/L	Growth Rate	Nominal
72	Hours	EL	50 %	=	25		mg/L	Other	Nominal

**Results Remarks:**

PARAFFINIC NAPHTHA CAS 64741-70-4, CONCAWE sample W94/810  
 Algae (Selenastum capricornutum) IrL50, 72h >50 mg/l (not calculable)  
 Fish (Oncorhynchus mykiss) LL50, 96h 10 mg/l (5-23)  
 Invertebrate (Daphnia magna) EL50, 48h 10 mg/l (8.5-13)  
 Algae endpoints for toxicity were Growth Rate and Area Under Growth Curve  
 95% confidence intervals are included in parentheses

**Reliability/Data Quality - Acute Toxicity To Aquatic Plants**

**Reliability:**

**Reliability Remarks:**

**Key Study Sponsor Indicator:** Key

**Reference - Acute Toxicity To Aquatic Plants**

**Reference:**

CONCAWE. 1996. Acute Aquatic Toxicity of Gasolines - Report on CONCAWE Test Programme. Report No. 96/57. CONCAWE, Brussels, Belgium.



<b>Acute Toxicity to Aquatic Plants</b>	
<b>Test Substance - Acute Toxicity To Aquatic Plants</b>	
<b>Category Chemical:</b>	(64741-54-4) Naphtha, petroleum, heavy catalytic cracked
<b>Test Substance:</b>	(64741-54-4) Naphtha, petroleum, heavy catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	OLEFINIC NAPHTHA CAS . 64741-54-4, CONCAWE sample W94/811 Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Acute Toxicity To Aquatic Plants</b>	
<b>Year Study Performed:</b>	1996
<b>Method/Guideline Followed:</b>	
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species:</b>	Selenastrum sp.
<b>GLP:</b>	
<b>Analytical Monitoring:</b>	
<b>Test Type:</b>	
<b>Test Vessel:</b>	
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	maximum loadings of 50 mg/l or less
<b>Exposure Period:</b>	72 Hours
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	
<b>pH Value:</b>	
<b>Test Temperature and Units:</b>	

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline Test Conditions Remarks:**

Experimental studies with fish, invertebrates and algae tested in closed systems with minimal head-space were performed on WAFS of low boiling point naphthas prepared at maximum loadings of 50 mg/l or less. Results show that acute aquatic toxicity lethal loading (LL), effective loading (EL) or inhibition of growth rate values (IrL) affecting 50% of the organism population are greater than 1 mg/l and mostly in the range 1-100 mg/l. Summarized CONCAWE test data indicating the extent of aquatic toxicity are as follows in Results Remarks Section, and 95% confidence intervals are included in parentheses.

**Limit Test:**

**Test Results - Acute Toxicity To Aquatic Plants**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:**

**NOELR:**

**LOELR Exposure Duration:**

**LOELR:**

**Effect:**

Exposure Duration	Exposure Units	Type	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Basis for Effect	Basis for Concentration
72	Hours	EL	50 %	=	3.1		mg/L	Growth Rate	Nominal
			%						

**Results Remarks:**

OLEFINIC NAPHTHA CAS . 64741-54-4, CONCAWE sample W94/811  
 Algae (Selenastum capricornutum) IrL50, 72h 3.1 mg/l (3.6-14)  
 Fish (Oncorhynchus mykiss) LL50, 96h 15 mg/l (10-23)  
 Invertebrate (Daphnia magna) EL50, 48h 13 mg/l (12-15)  
 95% confidence intervals are included in parentheses

**Reliability/Data Quality - Acute Toxicity To Aquatic Plants**

**Reliability:**

**Reliability Remarks:**

**Key Study Sponsor Indicator:**

Key

**Reference - Acute Toxicity To Aquatic Plants**

**Reference:**

CONCAWE. 1996. Acute Aquatic Toxicity of Gasolines - Report on CONCAWE Test Programme. Report No. 96/57. CONCAWE, Brussels, Belgium.





# **EcoToxicity Other**



## Chronic Aquatic Vertebrate Toxicity

### Test Substance - Chronic Aquatic Vertebrate Toxicity

**Category Chemical:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:** Aromatic naphtha  
Substance is in the Gasoline Blending Streams Category.  
See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

**Test Substance Result Type:** Measured

### Method - Chronic Aquatic Vertebrate Toxicity

**Year Study Performed:** 1999

**Method/Guideline Followed:** OECD 204

**Other Method/Guideline:**

**Deviations from Method/Guideline:**

**Species/In Vitro System:** Pimephales promelas

**GLP:** Yes

**Analytical Monitoring:** Yes

**Test Type:** Semi-Static

**Test Vessel:** Closed

**Test Concentrations:** Nominal

**Nominal and Measured Concentrations:** 0, 0.39, 1.0, 2.6, 6.3, 16, and 40mg/l

**Exposure Period:** 14 Days

**Vehicle Used:**

**Vehicle Name:**

**Vehicle Amount and Units:**

**Alkalinity:**

**Dissolved Oxygen:** 8.2 to 8.9 in the new solutions and 3.6 to 5.8 in the old solutions

**pH Value:** 7.2 Upper Range: 8.2

**Test Temperature and Units:** Value/Lower Range: 24 Upper Range: 26 °C

16/8

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline Test Conditions Remarks:**

LL50/LC50 and EL50/EC50 calculated using linear interpolation. NOEL/NOEC for survival determined by Steel's Many-One Rank Test. NOEL/NOEC for growth determined by Williams Test. TOXSTAT program was used to determine endpoints.

Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water was prepared by fortifying well water according to the formula for hard water (USEPA, 1975, EPA-660/3-75-009) and filtering through Amberlite XAD-7 resin to remove potential organic contaminants. The water used in this study had a total hardness range of 160-170 mg/l as CaCO<sub>3</sub>, total alkalinity of 120 mg/l as CaCO<sub>3</sub>, pH range of 7.9 to 8.1, and a specific conductivity range of 480 to 500 mmhos/cm. Nominal loading rates of 0, 0.39, 1.0, 2.6, 6.3, 16, and 40mg/l were used to prepare test solutions. WAFs were prepared for each test concentration by mixing the appropriate volume of substance in 9.4l of fortified well water for 24 hr in 9.5l screw-capped glass jars. The volume of test substance added was based on the experimentally determined density of 0.742 g/ml. After stirring for 24 hrs with a vortex of no more than 25% of the solution depth, the contents of the WAF solution bottles were allowed to settle for 1 to 1.5 hrs prior to use. The WAF was removed from an outlet port located 2 cm from the bottom of the jar directly into each exposure vessel. A control solution was prepared similarly except without test substance addition. Test solutions were renewed daily with fresh WAFs in which 80% of the old solutions were siphoned and excess debris removed from the exposure vessel prior to refilling with fresh WAF. Renewed solutions were then siphoned again and refilled a second time to achieve an exposure solution of ~96% fresh WAF. Duplicate samples of freshly prepared WAFs and composited replicate old test solutions were collected each day and analyzed by Purge & trap/GC-FID for concentrations of the following: pentane, methylpentane, benz ene, toluene, ethylbenzene, ortho, meta and para-xylene. Measured test concentrations of the light catalytically reformed naphtha were based on the concentrations of all analytes. Fish were hatched and raised from laboratory in-house culture. Fish were 8 days old at the start of the test. Test vessels were 1l screw-capped glass jars containing 980 ml of WAF with minimal headspace. Four replicates per treatment and 10 organisms per replicate were tested for each treatment and the control. Fish were fed 0.15 ml of live brine shrimp nauplii (<48 hr old) twice daily during the test. Water temperature was 24 to 26° C. Test photoperiod was 16 hrs light and 8 hrs dark. Dissolved oxygen concentrations were 8.2 to 8.9 in the new solutions and 3.6 to 5.8 in the old solutions. pH values were 7.2 to 8.2.

**Limit Test:** No

**Test Results - Chronic Aquatic Vertebrate Toxicity**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:** 14 Days

**NOELR:** = 2.6 mg/L Nominal

**LOELR:**

**LOELR Exposure Duration:**

**LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration

14	Days	LL	50 %	=	5.2		mg/L	Other	Calculated
			%						

**Results Remarks:**

The mean measured concentrations for nominal loading rates of 0.39, 1.0, 2.6, 6.3, 16, and 40 mg/l were 0.079, 0.15, 0.38, 0.80, 5.2, and 15 mg/l representing the average of total analytes measured in the new and old WAFs. The average total analyte concentration in the controls was 0.030 mg/l.  
 14-d LL50 for survival = 5.2 mg/l (95% C.I. 4.4 -7.0)  
 14-d LC50 for survival = 0.67 mg/l (95% C.I. 0.58 -0.93)  
 14-d NOEL for survival = 2.6 mg/l  
 14-d NOEC for survival = 0.38 mg/l.

Mortality (no. of deaths/treatment) at 14 days: 1, 0, 0, 2, 28, 40, and 40 in the 0, 0.39, 1.0, 2.6, 6.3, 16, and 40 mg/l treatments.

14-d NOEL for growth = 2.6 mg/l

14-d NOEC for growth = 0.38 mg/l.

14-d EL50 and EC50 for growth could not be calculated because none of the treatment group means were <50% of control.

Since there were significant mortality at the three highest treatments, these treatments were excluded in the analysis.

Measured concentrations represented the sum of six specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, the stability of the dissolved components between renewals was not reported. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

**Reliability/Data Quality - Chronic Aquatic Vertebrate Toxicity****Reliability:**

Valid with Restrictions

**Reliability Remarks:**

Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations. Low dissolved oxygen could have contributed to fish mortality.

RELIABILITY: GLP; guideline study with limitations for chemical analysis of relatively insoluble complex polymolecular substance

**Key Study Sponsor Indicator:**

Key

**Reference - Chronic Aquatic Vertebrate Toxicity****Reference:**

Springborn Laboratories, Inc. (1999) Light Catalytically Reformed Naphtha -Prolonged (14-Day) Toxicity Test with Fathead Minnow, *Pimephales promelas*, Under Static-Renewal Conditions Following OECD Guideline 204. Project ID. No. 13687.0598.6107.124.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



## Chronic Aquatic Vertebrate Toxicity

### Test Substance - Chronic Aquatic Vertebrate Toxicity

<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Olefenic naphtha Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured

### Method - Chronic Aquatic Vertebrate Toxicity

<b>Year Study Performed:</b>	1999
<b>Method/Guideline Followed:</b>	OECD 204
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species/In Vitro System:</b>	Pimephales promelas
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Semi-Static
<b>Test Vessel:</b>	Closed
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 0.38, 0.99, 2.6, 6.4, 16, and 40 mg/l
<b>Exposure Period:</b>	14 Days
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	8.1 to 8.5 in the new solutions and 3.7 to 5.9 in the old solutions.
<b>pH Value:</b>	7.2 Upper Range: 8.3
<b>Test Temperature and Units:</b>	Value/Lower Range: 24 Upper Range: 26 °C
	16/8

**Photo (Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:**

**Method/Guideline Test Conditions Remarks:**

Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water was prepared by fortifying well water according to the formula for hard water (USEPA, 1975, EPA-660/3-75-009) and filtering through Amberlite XAD-7 resin to remove potential organic contaminants. The water used in this study had a total hardness range of 170-180 mg/l as CaCO<sub>3</sub>, total alkalinity of 120-130 mg/l as CaCO<sub>3</sub>, pH range of 8.0 to 8.2, and a specific conductivity of 500 mmhos/cm. Nominal loading rates of 0, 0.38, 0.99, 2.6, 6.4, 16, and 40mg/l were used to prepare test solutions. WAFs were prepared for each test concentration by mixing the appropriate volume of substance in 9.4l of fortified well water for 24 hr in 9.5l screw-capped glass jars. The volume of test substance added was based on the experimentally determined density of 0.718 g/ml. After stirring for 24 hrs with a vortex of no more than 25% of the solution depth, the contents of the WAF solution bottles were allowed to settle for 0.75 to 1.25 hrs prior to use. The WAF was removed from an outlet port located 2 cm from the bottom of the jar directly into each exposure vessel. A control solution was prepared similarly except without test substance addition. Test solutions were renewed daily with fresh WAFs in which 80% of the old solutions were siphoned and excess debris removed from the exposure vessel prior to refilling with fresh WAF. Renewed solutions were then siphoned again and refilled a second time to achieve an exposure solution of ~96% fresh WAF. Duplicate samples of freshly prepared WAFs and composited replicate old test solutions were collected each day and analyzed by Purge & trap/GC-FID for concentrations of the following: benzene, toluene, ethylbenzene, ortho, meta and para-xylene. Measured test concentrations of the light catalytically cracked naphtha were based on the concentrations of all analytes. Fish were hatched and raised from laboratory in-house culture. Fish were 10 days old at the start of the test. Test vessels were 1l screw-capped glass jars containing 980 ml of WAF with minimal headspace. Four replicates per treatment and 10 organisms per replicate were tested for each treatment and the control. Fish were fed 0.15 ml of live brine shrimp nauplii (<48 hr old) twice daily during the test. Water temperature was 24 to 26° C. Test photoperiod was 16 hrs light and 8 hrs dark. Dissolved oxygen concentrations were 8.1 to 8.5 in the new solutions and 3.7 to 5.9 in the old solutions. pH values were 7.2 to 8.3.

**Limit Test:** No

**Test Results - Chronic Aquatic Vertebrate Toxicity**

**NOEC Exposure Duration:**

**NOEC:**

**LOEC Exposure Duration:**

**LOEC:**

**NOELR Exposure Duration:** 14 Days

**NOELR:** = 6.4 mg/L Nominal

**LOELR:**

**LOELR Exposure Duration:**

**LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration
14	Days	LL	50 %	=	23		mg/L	Mortality	Nominal
			%						

**Results Remarks:** The mean measured concentrations for nominal loading rates of 0.38, 0.99, 2.6, 6.4, 16, and 40 mg/l were 0.009, 0.024, 0.12, 0.28, 0.64, and 3.4 mg/l representing the average of total analytes measured in the new and old WAFs. The average total analyte concentration in the controls was 0.004 mg/l.

14-d LL50 for survival = 23 mg/l (95% C.I. 19 -26)  
 14-d LC50 for survival = 1.5 mg/l (95% C.I. 1.1 -1.8)  
 14-d NOEL for survival = 6.4 mg/l  
 14-d NOEC for survival = 0.28 mg/l.

Mortality (no. of deaths/treatment) at 14 days: 0, 1, 0, 0, 3, 11, and 40 in the 0, 0.38, 0.99, 2.6, 6.4, 16, and 40 mg/l treatments.

14-d NOEL for growth = 6.4 mg/l  
 14-d NOEC for growth = 0.28 mg/l

14-d EL50 and EC50 for growth could not be calculated because none of the treatment group means were <50% of control. Since there were significant mortality at the two highest treatments, these treatments were excluded in the analysis of growth data. The mean (standard deviation) for dry weights were 2.49 (0.08), 2.58 (0.21), 2.76 (0.07), 2.67 (0.19), and 2.79 (0.42) in the 0, 0.38, 0.99, 2.6, and 6.4 mg/l treatments. Light intensity was not measured during the study due to an oversight and had no impact on the results of the study.

Measured concentrations represented the sum of four specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, the stability of the dissolved components between renewals was not reported. Because of the uncertainty in what the measured values represented, test endpoints based on the measured data should be viewed and interpreted with an understanding of this limitation.

#### Reliability/Data Quality - Chronic Aquatic Vertebrate Toxicity

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP; non-guideline study with limitations in analytical monitoring of complex polymolecular substance

**Key Study Sponsor Indicator:** Key

#### Reference - Chronic Aquatic Vertebrate Toxicity

**Reference:** Springborn Laboratories, Inc. (1999) Light Catalytically Cracked Naphtha -Prolonged (14-Day) Toxicity Test with Fathead Minnow, Pimephales promelas, Under Static-Renewal Conditions Following OECD Guideline 204. Project ID. No. 13687.0598.6106.124.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



## Chronic Aquatic Vertebrate Toxicity

### Test Substance - Chronic Aquatic Vertebrate Toxicity

**Category Chemical:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance Purity/Composition** Paraffinic naphtha

**and Other Test Substance Comments:** Substance is in the Gasoline Blending Streams Category.  
See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

**Test Substance Result Type:** Measured

### Method - Chronic Aquatic Vertebrate Toxicity

**Year Study Performed:** 1999

**Method/Guideline Followed:** OECD 204

**Other Method/Guideline:**

**Deviations from Method/Guideline:**

**Species/In Vitro System:** Pimephales promelas

**GLP:** Yes

**Analytical Monitoring:**

**Test Type:**

**Test Vessel:**

**Test Concentrations:** Nominal

**Nominal and Measured Concentrations:** 0, 0.44, 1.0, 2.6, 6.4, 16, and 40 mg/L

**Exposure Period:** 14 Days

**Vehicle Used:**

**Vehicle Name:**

**Vehicle Amount and Units:**

**Alkalinity:** 120mg/l as CaCO<sub>3</sub>

**Dissolved Oxygen:** 8.7 to 8.9 in the new solutions and 5.7 to 7.8 in the old solutions

**pH Value:** 7.3

**Test Temperature and Units:** Value/Lower Range: 24 °C

16/8



**Photo  
(Light/Dark):**

**Salinity:**

**TOC:**

**Water Hardness:** 170

**Method/Guideline  
Test Conditions  
Remarks:**

**Limit Test:** No

### Test Results - Chronic Aquatic Vertebrate Toxicity

**NOEC Exposure  
Duration:**

**NOEC:**

**LOEC Exposure  
Duration:**

**LOEC:**

**NOELR Exposure  
Duration:** 14 Days

**NOELR:** = 2.6 mg/L Nominal

**LOELR:**

**LOELR Exposure  
Duration:**

**LC/EC Mean Value:**

Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower Mean Value	Upper Mean Value	Units	Effect Observed	Basis for Concentration
			%						Other
14	Days	LL	50 %	=	8		mg/L	Mortality	Nominal

**Results Remarks:**

The mean measured concentrations for nominal loading rates of 0.44, 1.0, 2.6, 6.4, 16, and 40 mg/l were 0.011, 0.021, 0.041, 0.10, 0.38, and 0.62 mg/l representing the average of total analytes measured in the new and old WAFs. The average total analyte concentration in the controls was 0.005 mg/l.

14-d LL50 for survival = 8.0 mg/l (95% C.I. 5.4 -9.8),

14-d LC50 for survival = 0.15 mg/l (95% C.I. 0.073 -0.20)

14-d NOEL for survival = 2.6 mg/l,

14-d NOEC for survival = 0.041 mg/l.

Mortality (no. of deaths/treatment) at 14 days: 0, 0, 3, 2, 16, 40, and 40 in the 0, 0.44, 1.0, 2.6, 6.4, 16, and 40 mg/l treatments. All surviving fish in the 6.4 mg/l treatment were lethargic.

14-d NOEL for growth = 2.6 mg/l,

14-d NOEC for growth = 0.041 mg/l.

14-d EL50 and EC50 for growth could not be calculated because none of the treatment group means were <50% of control.

Since there were significant mortality at the three highest treatments, these treatments were excluded in the analysis of growth data. The mean (standard deviation) for dry weights were 4.08 (0.26), 4.28 (0.20), 4.69 (0.43), and 4.85 (0.38) in the 0, 0.44, 1.0, and 2.6 mg/l treatments. Dissolved oxygen concentrations in the aged exposure solutions at all loading rates were occasionally below 60% of saturation between day 10 and day 14 due to oxygen consumption by fish and bacteria in the closed test systems and could not be avoided. Light intensity was not measured during the study due to an oversight and had no impact on the results of the study.

Measured concentrations represented the sum of seven specific hydrocarbon compounds

measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, the stability of the dissolved components between renew als was not reported. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

### Reliability/Data Quality - Chronic Aquatic Vertebrate Toxicity

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations. Dissolved oxygen in the test solutions were occasionally below 60% of saturation.

RELIABILITY: GLP; non-guideline study with limitations in analytical monitoring of complex polymolecular substance

**Key Study Sponsor Indicator:** Key

### Reference - Chronic Aquatic Vertebrate Toxicity

**Reference:** Springborn Laboratories, Inc.(1999) Light Alkylate Naphtha -Prolonged (14-Day) Toxicity Test with Fathead Minnow, Pimephales promelas, Under Static-Renewal Conditions Following OECD Guideline 204. Project ID. No. 13687.0598.6108.124.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Chronic Aquatic Invertebrate Toxicity</b>	
<b>Test Substance - Chronic Aquatic Invertebrate Toxicity</b>	
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Olefinic naphtha Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Test Substance Result Type:</b>	Measured
<b>Method - Chronic Aquatic Invertebrate Toxicity</b>	
<b>Year Study Performed:</b>	1999
<b>Method/Guideline Followed:</b>	OECD 211
<b>Other Method/Guideline:</b>	
<b>Deviations from Method/Guideline:</b>	
<b>Species/In Vitro System:</b>	Daphnia magna
<b>GLP:</b>	Yes
<b>Analytical Monitoring:</b>	Yes
<b>Test Type:</b>	Semi-Static
<b>Test Vessel:</b>	Closed
<b>Water Media Type:</b>	Freshwater
<b>Test Concentrations:</b>	Nominal
<b>Nominal and Measured Concentrations:</b>	0, 0.38, 0.99, 2.6, 6.4, 16, and 40 mg/l
<b>Exposure Period:</b>	21 Days
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Alkalinity:</b>	
<b>Dissolved Oxygen:</b>	8.7 to 8.8 in the new solutions and 8.4 to 9.1 in the old solutions.
<b>pH Value:</b>	7.2 Upper Range: 8.2
<b>Test Temperature and Units:</b>	Value/Lower Range: 19 Upper Range: 21 °C

<b>Photo (Light/Dark):</b>	16/8																				
<b>Salinity:</b>																					
<b>TOC:</b>																					
<b>Water Hardness:</b>																					
<b>Method/Guideline Test Conditions Remarks:</b>	<p>For NOEL/NOEC, Fisher's Exact Test was used for survival of adult daphnids and Kruskal-Wallis Test with Dunn's Multiple Comparison was used for reproduction. For EL50/EC50, survival data were analyzed using the Spearman-Kärber method and reproduction data were analyzed by linear interpolation. TOXSTAT program was used to determine the endpoints.</p> <p>Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water was prepared by fortifying well water according to the formula for hard water (USEPA, 1975, EPA-660/3-75-009) and filtering through Amberlite XAD-7 resin to remove potential organic contaminants. The water used in this study had a total hardness range of 170-180 mg/l as CaCO<sub>3</sub>, total alkalinity of 120-130 mg/l as CaCO<sub>3</sub>, pH range of 8.0 to 8.2, and a specific conductivity of 500 µmhos/cm. Nominal loading rates of 0, 0.38, 0.99, 2.6, 6.4, 16, and 40mg/l were used to prepare test solutions. WAFs were prepared for each test concentration by mixing the appropriate volume of substance in 9.4l of fortified well water for 24 hr in 9.5l screw-capped glass jars. The volume of test substance added was based on the experimentally determined density of 0.718 g/ml. After stirring for 24 hrs with a vortex of no more than 25% of the solution depth, the contents of the WAF solution bottles were allowed to settle for 45 min to 1.25 hrs prior to use. The WAF was removed from an outlet port located 2 cm from the bottom of the jar directly into each exposure vessel. A control solution was prepared similarly except without test substance addition. Test solutions were renewed daily with 70 ml of fresh WAFs added to a second set of beakers. Food was added to the fresh WAFs and daphnids were then transferred from the old test solutions to the fresh WAFs. Duplicate samples of freshly prepared WAFs and composited replicate old test solutions were collected each day and analyzed by Purge &amp; trap/GC-FID for concentrations of the following: benzene, toluene, ethylbenzene, ortho, meta and para-xylene. Measured test concentrations of the light catalytically cracked naphtha were based on the concentrations of all analytes. Daphnids used in the test were from laboratory in-house culture. Daphnids were =24 hrs old at the start of the test. Test vessels were 70 ml screw-capped glass jars containing 70 ml of WAF with minimal headspace. Ten replicates per treatment and 1 daphnid per replicate were tested for each treatment and the control. Daphnids were fed 0.2 ml of algal suspension (<i>Ankistrodesmus falcatus</i>, 4 x 10<sup>7</sup> cells/ml) and 0.05 ml of a yeast, cereal leaves and digested flaked fish food (YCT) suspension daily during the test. Water temperature was 19 to 21° C. Test photoperiod was 16 hrs light and 8 hrs dark. Dissolved oxygen concentrations were 8.7 to 8.8 in the new solutions and 8.4 to 9.1 in the old solutions. pH values were 7.2 to 8.2.</p>																				
<b>Limit Test:</b>	No																				
<b>Test Results - Chronic Aquatic Invertebrate Toxicity</b>																					
<b>NOEC Exposure Duration:</b>																					
<b>NOEC:</b>																					
<b>LOEC Exposure Duration:</b>																					
<b>LOEC:</b>																					
<b>NOELR Exposure Duration:</b>	21 Days																				
<b>NOELR:</b>	= 2.6 - 16 mg/L Nominal																				
<b>LOELR Exposure Duration:</b>																					
<b>LOELR:</b>																					
<b>LC/EC Mean Value:</b>																					
	<table border="1"> <thead> <tr> <th>Exposure Duration</th> <th>Exposure Units</th> <th>LC/EC</th> <th>%</th> <th>Value Description</th> <th>Mean Value or Lower</th> <th>Upper Mean Value</th> <th>Units</th> <th>Effect Observed</th> <th>Basis for Concentration</th> </tr> </thead> <tbody> <tr> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td> </tr> </tbody> </table>	Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower	Upper Mean Value	Units	Effect Observed	Basis for Concentration										
Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower	Upper Mean Value	Units	Effect Observed	Basis for Concentration												

					Mean Value				
21	Days	EL	50 %	=	27		mg/L	Mortality	Nominal
21	Days	EL	50 %	=	13		mg/L	Reproduction	Nominal
			%						
			%						

**Results Remarks:**

The mean measured concentrations for nominal loading rates of 0.38, 0.99, 2.6, 6.4, 16, and 40 mg/l were 0.007, 0.022, 0.11, 0.27, 0.68, and 3.1 mg/l representing the average of total analytes measured in the new and old WAFs. The average total analyte concentration in the controls was 0.004 mg/l.

21-d EL50 for survival = 27 mg/l (95% C.I. 26 -29)

21-d EC50 for survival = 1.9 mg/l (95% C.I. 1.8 -2.0)

21-d NOEL for survival = 16 mg/l

21-d NOEC for survival = 0.68 mg/l

Daphnid immobilization at 21 days: 0, 1, 0, 0, 0, 0, and 10 in the 0, 0.38, 0.99, 2.6, 6.4, 16, and 40 mg/l treatments.

21-d EL50 for reproduction = 13 mg/l (95% C.I. 12-15)

21-d EC50 for reproduction = 0.55 mg/l (95% C.I. 0.49-0.64)

21-d NOEL for reproduction = 2.6 mg/l

21-d NOEC for reproduction = 0.11 mg/l

Since there was significant immobilization in the highest treatment, it was excluded in the analysis of reproduction data. The mean numbers (standard deviation) of offspring released per female daphnid were 150 (9), 139 (12), 141 (7), 139 (10), 123 (10), and 55 (28) in the 0, 0.38, 0.99, 2.6, 6.4, and 16 mg/l treatments. The numbers of offspring released in the 6.4 and 16 mg/l treatments were significantly less than the controls. First brood release for organisms exposed to =6.4 mg/l and the controls occurred by day 8. First brood release for organisms exposed to 16 mg/l occurred on day 10. From day 17 to day 21, immobilized offspring were released in the 16 mg/l treatment.

Measured concentrations represented the sum of four specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, the stability of the dissolved components between renewals was not reported. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

**Reliability/Data Quality - Chronic Aquatic Invertebrate Toxicity**

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP; guideline study with limitations in analytical monitoring of complex polymolecular substance

**Key Study Sponsor Indicator:** Key

**Reference - Chronic Aquatic Invertebrate Toxicity**

**Reference:** Springborn Laboratories, Inc. (1999) Light Catalytically Cracked Naphtha -Full Life Cycle Toxicity Test with Water Fleas, Daphnia magna, Under Static-Renewal Conditions Following OECD Guideline 211. Project ID. No. 13687.0598.6103.130.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



## Chronic Aquatic Invertebrate Toxicity

### Test Substance - Chronic Aquatic Invertebrate Toxicity

**Category Chemical:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance Purity/Composition and Other Test Substance Comments:** Paraffinic naphtha  
Substance is in the Gasoline Blending Streams Category.  
See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

**Test Substance Result Type:** Measured

### Method - Chronic Aquatic Invertebrate Toxicity

**Year Study Performed:** 1999

**Method/Guideline Followed:** OECD 211

**Other Method/Guideline:**

**Deviations from Method/Guideline:**

**Species/In Vitro System:** Daphnia magna

**GLP:** Yes

**Analytical Monitoring:** Yes

**Test Type:** Semi-Static

**Test Vessel:** Closed

**Water Media Type:** Freshwater

**Test Concentrations:** Nominal

**Nominal and Measured Concentrations:** 0, 0.44, 1.0, 2.6, 6.4, 16, and 40 mg/L

**Exposure Period:** 21 Days

**Vehicle Used:**

**Vehicle Name:**

**Vehicle Amount and Units:**

**Alkalinity:**

**Dissolved Oxygen:** 9.1 to 9.2 in the new solutions and 8.7 to 9.4 in the old

**pH Value:** 7.5 Upper Range: 8.5

**Test Temperature and Units:** Value/Lower Range: 19 Upper Range: 21 °C

<b>Photo (Light/Dark):</b>	16/8																				
<b>Salinity:</b>																					
<b>TOC:</b>																					
<b>Water Hardness:</b>																					
<b>Method/Guideline Test Conditions Remarks:</b>	<p>For NOEL/NOEC, Fisher's Exact Test was used for survival of adult daphnids and Kruskal-Wallis Test with Dunn's Multiple Comparison was used for reproduction. For EL50/EC50, reproduction data were analyzed by linear interpolation. Survival data were not analyzed because survival was &gt;50% at all loading rates. TOXSTAT program was used to determine the endpoints.</p> <p>Test solutions were prepared as water accommodated fractions (WAF). Control and dilution water was prepared by fortifying well water according to the formula for hard water (USEPA, 1975, EPA-660/3-75-009) and filtering through Amberlite XAD-7 resin to remove potential organic contaminants. The water used in this study had a total hardness range of 170-180 mg/l as CaCO<sub>3</sub>, total alkalinity of 120-130 mg/l as CaCO<sub>3</sub>, pH range of 8.0 to 8.2, and a specific conductivity range of 500-550 mmhos/cm. Nominal loading rates of 0, 0.44, 1.0, 2.6, 6.4, 16, and 40 mg/l were used to prepare test solutions. WAFs were prepared for each test concentration by mixing the appropriate volume of substance in 9.4l of fortified well water for 24 hr in 9.5l screw-capped glass jars. The volume of test substance added was based on the experimentally determined density of 0.69 g/ml. After stirring for 24 hrs with a vortex of no more than 25% of the solution depth, the contents of the WAF solution bottles were allowed to settle for 1 to 1.5 hrs prior to use. The WAF was removed from an outlet port located 2 cm from the bottom of the jar directly into each exposure vessel. A control solution was prepared similarly except without test substance addition. Test solutions were renewed daily with 70 ml of fresh WAFs added to a second set of beakers. Food was added to the fresh WAFs and daphnids were then transferred from the old test solutions to the fresh WAFs. Duplicate samples of freshly prepared WAFs and composited replicate old test solutions were collected each day and analyzed by Purge &amp; trap/GC-FID for concentrations of the following : 2,3-dimethylbutane, 2,4-dimethylpentane, 2,2,4-trimethylpentane, 2,5-dimethylhexane, 2,3,3-trimethylpentane, and 2,3,4-trimethylpentane. Measured test concentrations of the light alkylate naphtha were based on the concentrations of all analytes. Daphnids used in the test were from laboratory in-house culture. Daphnids were =24 hrs old at the start of the test. Test vessels were 70 ml screw-capped glass jars containing 70 ml of WAF with minimal headspace. Ten replicates per treatment and 1 daphnid per replicate were tested for each treatment and the control. Daphnids were fed 0.2 ml of algal suspension (<i>Ankistrodesmus falcatus</i>, 4 x 10<sup>7</sup> cells/ml) and 0.05 ml of a yeast, cereal leaves and digested flaked fishfood (YCT) suspension daily during the test. Water temperature was 19 to 21° C. Test photoperiod was 16 hrs light and 8 hrs dark. Dissolved oxygen concentrations were 9.1 to 9.2 in the new solutions and 8.7 to 9.4 in the old solutions. pH values were 7.5 to 8.5.</p>																				
<b>Limit Test:</b>	No																				
<b>Test Results - Chronic Aquatic Invertebrate Toxicity</b>																					
<b>NOEC Exposure Duration:</b>																					
<b>NOEC:</b>																					
<b>LOEC Exposure Duration:</b>																					
<b>LOEC:</b>																					
<b>NOELR Exposure Duration:</b>	21 Days																				
<b>NOELR:</b>	= 2.6 - 16 mg/L Nominal																				
<b>LOELR Exposure Duration:</b>																					
<b>LOELR:</b>																					
<b>LC/EC Mean Value:</b>	<table border="1" style="width:100%; border-collapse: collapse; text-align: center;"> <thead> <tr> <th>Exposure Duration</th> <th>Exposure Units</th> <th>LC/EC</th> <th>%</th> <th>Value Description</th> <th>Mean Value or Lower</th> <th>Upper Mean Value</th> <th>Units</th> <th>Effect Observed</th> <th>Basis for Concentration</th> </tr> </thead> <tbody> <tr> <td> </td> <td> </td> <td> </td> <td> </td> <td> </td> <td> </td> <td> </td> <td> </td> <td> </td> <td> </td> </tr> </tbody> </table>	Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower	Upper Mean Value	Units	Effect Observed	Basis for Concentration										
Exposure Duration	Exposure Units	LC/EC	%	Value Description	Mean Value or Lower	Upper Mean Value	Units	Effect Observed	Basis for Concentration												

					Mean Value				
21	Days	EL	50 %	>=	40		mg/L	Mortality	Nominal
21	Days	EL	50 %	=	10		mg/L	Reproduction	Nominal
			%						
			%						

**Results Remarks:**

The mean measured concentrations for nominal loading rates of 0.44, 1.0, 2.6, 6.4, 16, and 40 mg/l were 0.010, 0.016, 0.032, 0.084, 0.23, and 0.46 mg/l representing the average of total analytes measured in the new and old WAFs. The average total analyte concentration in the controls was 0.005 mg/l.

21-d EL50 for survival = >40 mg/l  
 21-d EC50 for survival = >0.46 mg/l  
 21-d NOEL for survival = 16 mg/l  
 21-d NOEC for survival = 0.23 mg/l.

Daphnid immobilization at 21 days: 0, 2, 0, 0, 0, 1, and 4 in the 0, 0.44, 1.0, 2.6, 6.4, 16, and 40 mg/l treatments.

21-d EL50 for reproduction = 10 mg/l (95% C.I. 8.7-11)  
 21-d EC50 for reproduction = 0.14 mg/l (95% C.I. 0.12-0.16)  
 21-d NOEL for reproduction = 2.6 mg/l  
 21-d NOEC for reproduction = 0.032 mg/l.

Since there was significant immobilization in the highest treatment, it was excluded in the analysis of reproduction data. The mean numbers (standard deviation) of offspring released per female daphnid were 137 (11), 125 (7), 125 (6), 117 (20), 96 (21), and 28 (10) in the 0, 0.44, 1.0, 2.6, 6.4, and 16 mg/l treatments. The numbers of offspring released in the 6.4 and 16 mg/l treatments were significantly less than the controls. First brood release for organisms exposed to =16 mg/l and the controls occurred by day 8.

Measured concentrations represented the sum of seven specific hydrocarbon compounds measured in the WAF solutions. However, these compounds do not represent 100% of the hydrocarbons in the dissolved fraction. Therefore, test endpoints calculated from measured values would be expected to be lower than if all dissolved components were included in the measurements. Additionally, the stability of the dissolved components between renewals was not reported. Because of the uncertainty in what the measured values represented, test endpoints based on measured data should be viewed and interpreted with an understanding of this limitation.

**Reliability/Data Quality - Chronic Aquatic Invertebrate Toxicity**

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Measured concentrations may not represent 100% of components, remaining hydrocarbon components in WAFs may be equally toxic and should have been quantitated to determine total measured concentrations.

RELIABILITY: GLP; guideline study with limitations in analytical monitoring of complex polymolecular substance

**Key Study Sponsor Indicator:** Key

**Reference - Chronic Aquatic Invertebrate Toxicity**

**Reference:** Springborn Laboratories, Inc., (1999) Light Alkylate Naphtha -Full Life Cycle Toxicity Test with Water Fleas, *Daphnia magna*, Under Static-Renewal Conditions Following OECD Guideline 211. Project ID. No. 13687.0598.6105.130.

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## Other

**Category Chemical:** *No CAS Number Provided*

**Test Substance:** *No CAS Number Provided*

**Test Substance Purity/Composition and Other Test Substance Comments:**

**Other End Point Name:** Ecotoxicity

**Other End Point Description:**

**Reference:** CONCAWE (1996) Acute aquatic toxicity of gasolines: report on CONCAWE test programme. CONCAWE Report No. 96/57.<br><br>Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003

**Description:** Experimental studies with fish, invertebrates and algae tested in closed systems with minimal head-space were performed on WAFS of low boiling point naphthas prepared at maximum loadings of 50 mg/l or less. Results show that acute aquatic toxicity lethal loading (LL), effective loading (EL) or inhibition of growth rate values (IrL) affecting 50% of the organism population are greater than 1 mg/l and mostly in the range 1-100 mg/l. Summarized CONCAWE test data indicating the extent of aquatic toxicity are as follows, and 95% confidence intervals are included in parentheses:

**Results:** PARAFFINIC NAPHTHA CAS 64741-70-4, CONCAWE sample W94/810<br>Fish (Oncorhynchus mykiss) LL50, 96h 10 mg/l (5-23)<br>Invertebrate (Daphnia magna) EL50, 48h 10 mg/l (8.5-13)<br>Algae (Selenastum capricornutum) IrL50, 72h >50 mg/l (not calculable)<br><br>OLEFINIC NAPHTHA CAS 64741-54-4, CONCAWE sample W94/811<br>Fish (Oncorhynchus mykiss) LL50, 96h 15 mg/l (10-23)<br>Invertebrate (Daphnia magna) EL50, 48h 13 mg/l (12-15)<br>Algae (Selenastum capricornutum) IrL50, 72h 3.1 mg/l (3.6-14)<br><br>NAPHTHENIC NAPHTHA, CAS 64741-46-4, CONCAWE sample W94/809<br>Fish (Oncorhynchus mykiss) LL50, 96h 18 mg/l (15-20)<br>Invertebrate (Daphnia magna) EL50, 48h 4.5 mg/l (not calculable)<br>Algae (Selenastum capricornutum) IrL50, 72h 4.1 mg/l (not calculable)<br><br>AROMATIC NAPHTHA, CAS 64741-63-5, CONCAWE sample W94/812<br>Fish (Oncorhynchus mykiss) LL50, 96h 12 mg/l (9-16)<br>Invertebrate (Daphnia magna) EL50, 48h 8.4 mg/l (6.7-11)<br>Algae (Selenastum capricornutum) IrL50, 72h 6.4 mg/l (1-280)

# **Mammalian Health Effects**

## **SIDS**



## Acute Toxicity

### Test Substance - Acute Toxicity

**Category Chemical:** No CAS Number Provided

**Test Substance:** No CAS Number Provided

**Test Substance Purity/Composition and Other Test Substance Comments:** Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhqv.org>

**Category Chemical Result Type:** Read-Across

### Method - Acute Toxicity

**Route of Administration:** Inhalation

**Type of Exposure:**

**Species:**

**Mammalian Strain:**

**Gender:**

**Number of Animals per Dose:**

**Dose:**

**Year Study Performed:**

**Method/Guideline Followed:**

**GLP:**

**Method/Guideline and Test Condition Remarks:**

### Test Results - Acute Toxicity

**Concentration (LC/LD):**

LC/LD	%	Value Description	Value/Lower Concentration	Upper Concentration	Units
LC	50 %	>	5000		mg/m3

**Number of Deaths ( Male ):**

**Number of Deaths ( Female ):**

**Number of Deaths ( Total ):**

**Results Remarks:** Results of testing naphtha blending streams for acute toxicity indicate that these materials demonstrate consistently low acute toxicity by the oral [Rat LD50 >5g/kg], dermal [Rabbit LD50 >2g/kg] and inhalation [Rat LC50 >5g/m3] exposure routes, are mild to moderate eye and skin irritants and are not skin

sensitizers. Acute data for gasoline gave comparable results.

**Conclusion:** The inhalation acute toxicity read-across value for untested category members is LC50 > 5,000 mg/m3

### Reliability/Data Quality - Acute Toxicity

**Reliability:**

**Reliability Remarks:** All studies used to develop the read across value were rated as

Reliability = 1, Valid without restrictions

**Key Study Sponsor Indicator:**

### Reference - Acute Toxicity

**Reference:** See records for CAS 64741-55-5, 64741-66-8, 64741-87-3, 68955-35-1, and 86290-81-5.



## Acute Toxicity

### Test Substance - Acute Toxicity

**Category Chemical:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance Purity/Composition and Other Test Substance Comments:** Test sample API #83-20 Light Catalytic Cracked Naphtha (LCCN). See compositional data file attached to category at website below.

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

### Method - Acute Toxicity

**Route of Administration:** Inhalation

**Type of Exposure:** Vapor

**Species:** Rat

**Mammalian Strain:** Sprague-Dawley

**Gender:** Both M/F

**Number of Animals per Dose:** 5

**Dose:** 5 mg/l

**Year Study Performed:** 1987

**Method/Guideline Followed:** Unknown

**GLP:** Yes

**Method/Guideline and Test Condition Remarks:** A group of 5 male and 5 female rats were exposed by whole body inhalation to API 83-20 at a nominal concentration of 5mg/l for 4 hours. After the 4 hour exposure the rats were observed twice daily for mortality. The animals were weighed prior to exposure and again on days 7 and 14 post exposure. On day 14 all surviving animals were killed and subjected to a gross post-mortem examination. For all animals, including those found dead during the study, the lungs were removed, fixed and examined histologically.

### Test Results - Acute Toxicity

**Concentration (LC/LD):**

LC/LD	%	Value Description	Value/Lower Concentration	Upper Concentration	Units
LC	50 %	>	5300		mg/m3

**Number of Deaths ( Male ):** 0 of 5

**Number of Deaths ( Female ):** 0 of 5

**Number of Deaths ( Total ):** 0 of 10

**Results Remarks:** The mean analytical exposure concentration was measured and found to be 5.28 ±0.55 mg/L. Gravimetric samples, collected on glass fiber filters suggested little or no aerosol in the chamber. Most animals exhibited languid behavior and squinted eyes during the second hour of the exposure. Polypnea was observed in all animals when removed from the chamber at the one hour post exposure observation period. Rhinorrhea was exhibited by two animals on day two of the test. All animals appeared normal subsequently and there were no mortalities during the study. With the exception of one animal (female) all animals had body weights that were considered unremarkable. There were no remarkable gross or microscopic findings.

**Conclusion:** Inhalation LC50 > 5.3 mg/L (5.3 g/m<sup>3</sup>; 5300 mg/m<sup>3</sup>) in male and female rats

#### Reliability/Data Quality - Acute Toxicity

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:** Key

#### Reference - Acute Toxicity

**Reference:** American Petroleum Institute (1987) Acute inhalation toxicity evaluation of a petroleum derived hydrocarbon in rats API 83-20 Light catalytic cracked naphtha (CAS 64741-55-5). Study conducted by Hazleton Laboratories America Inc. Health and Environmental Sciences Dept. Publ. No. 34-32777

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



## Acute Toxicity

### Test Substance - Acute Toxicity

**Category Chemical:** (64741-87-3) Naphtha, petroleum, sweetened

**Test Substance:** (64741-87-3) Naphtha, petroleum, sweetened

**Test Substance Purity/Composition and Other Test Substance Comments:** Test material API #81-08 Sweetened Naphtha. See compositional data file attached to category at website below.

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

### Method - Acute Toxicity

**Route of Administration:** Inhalation

**Type of Exposure:** Vapor

**Species:** Rat

**Mammalian Strain:** Sprague-Dawley

**Gender:** Both M/F

**Number of Animals per Dose:** 5

**Dose:** 5 mg/l (5,000 mg/m3)

**Year Study Performed:** 1986

**Method/Guideline Followed:** Unknown

**GLP:** Yes

**Method/Guideline and Test Condition Remarks:** A group of 5 male and 5 female rats were exposed by whole body inhalation to API 81-08 at a nominal concentration of 5mg/l for 4 hours. After the 4 hour exposure the rats were observed twice daily for mortality. The animals were weighed prior to exposure and again on days 7 and 14 post exposure. On day 14 all surviving animals were killed by exsanguination following sodium pentobarbital anesthesia and were subjected to a full necropsy. For all animals, including those found dead during the study the lungs were removed, fixed and examined histologically.

### Test Results - Acute Toxicity

**Concentration (LC/LD):**

LC/LD	%	Value Description	Value/Lower Concentration	Upper Concentration	Units
LC	50 %	>	5200		mg/m3

**Number of Deaths ( Male ):** 0 of 5

**Number of Deaths ( Female ):** 0 of 5

**Number of Deaths ( Total ):** 0 of 10

**Results Remarks:** The actual chamber concentrations were found to be 5.2 mg/l.  
No deaths occurred during the study. There were no unusual pharmacotoxic signs or behavior observed in the control animals. There was however, a slight incidence of nasal discharge (2/5 males and 1/5 females) during the exposure period but none during the following 14 day observation period.  
The body weight gains for the males exposed to API 81-08 were considered normal but the female body weight gains were marginally less than that of the controls on day 14 post exposure (8.2% compared to 13.8% increase over pre-exposure body weight).  
No significant macro or microscopic changes were observed that were considered to be treatment related.

Number of deaths are reported for treated animals, only.

**Conclusion:** The inhalation LC50 > 5.2 mg/L (5,200 mg/m<sup>3</sup>) in male and female Sprague-Dawley rats

### Reliability/Data Quality - Acute Toxicity

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:** Key

### Reference - Acute Toxicity

**Reference:** American Petroleum Institute (1986) LC50 Acute inhalation toxicity evaluation of a petroleum derived hydrocarbon in rats API 81-08 Sweetened Naphtha CAS 64741-87-3 API Health and Environmental Sciences Dept. Publication No. 33-31827. June 1986

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003





<b>Acute Toxicity</b>													
<b>Test Substance - Acute Toxicity</b>													
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate												
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate												
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Sample API 83-19 is a Light Alkylate Naphtha (LAN). See compositional data file attached to category at website below. Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>												
<b>Category Chemical Result Type:</b>	Measured												
<b>Method - Acute Toxicity</b>													
<b>Route of Administration:</b>	Inhalation												
<b>Type of Exposure:</b>	Vapor												
<b>Species:</b>	Rat												
<b>Mammalian Strain:</b>	Sprague-Dawley												
<b>Gender:</b>	Both M/F												
<b>Number of Animals per Dose:</b>	5												
<b>Dose:</b>	5 mg/L (5,000 mg/m3)												
<b>Year Study Performed:</b>	1987												
<b>Method/Guideline Followed:</b>	Unknown												
<b>GLP:</b>	Yes												
<b>Method/Guideline and Test Condition Remarks:</b>	A group of 5 male and 5 female rats were exposed by whole body inhalation to API 83-19 at a nominal concentration of 5mg/l for 4 hours. This was achieved by total volatilization of the test material and appropriate dilution with air. After the 4 hour exposure the rats were observed twice daily for mortality. The animals were weighed prior to exposure and again on days 7 and 14 post exposure. On day 14 all surviving animals were killed by exsanguination following sodium pentobarbital anesthesia. For all animals, including those found dead during the study the lungs were removed, fixed and examined histologically.												
<b>Test Results - Acute Toxicity</b>													
<b>Concentration (LC/LD):</b>	<table border="1"> <thead> <tr> <th>LC/LD</th> <th>%</th> <th>Value Description</th> <th>Value/Lower Concentration</th> <th>Upper Concentration</th> <th>Units</th> </tr> </thead> <tbody> <tr> <td>LC</td> <td>50 %</td> <td>&gt;</td> <td>5000</td> <td></td> <td>mg/m3</td> </tr> </tbody> </table>	LC/LD	%	Value Description	Value/Lower Concentration	Upper Concentration	Units	LC	50 %	>	5000		mg/m3
LC/LD	%	Value Description	Value/Lower Concentration	Upper Concentration	Units								
LC	50 %	>	5000		mg/m3								
<b>Number of Deaths ( Male ):</b>	0 of 5												
<b>Number of Deaths ( Female ):</b>	0 of 5												

<b>Number of Deaths ( Total ):</b>	0 of 10
<b>Results Remarks:</b>	The mean analytical and nominal exposure concentrations were 5.04 ± 0.74 and 6.31 mg/l respectively. All animals survived the study but exhibited languid behavior and a hunched appearance during the exposure. Female body weights were decreased at day 15 but this was attributed to pre-necropsy fasting. At necropsy there were no remarkable findings and histopathology of the lungs was normal.
<b>Conclusion:</b>	Inhalation LD50 > 5 mg/L (5,000 mg/m3) in male and female Sprague-Dawley rats
<b>Reliability/Data Quality - Acute Toxicity</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	RELIABILITY: GLP study with adequately detailed methods description
<b>Key Study Sponsor Indicator:</b>	Key
<b>Reference - Acute Toxicity</b>	
<b>Reference:</b>	American Petroleum Institute (1987) Acute inhalation toxicity evaluation of a petroleum derived hydrocarbon in rats API 83-19 Light Alkylate Naphtha (CAS# 64741-66-8). Study conducted by Hazleton laboratories. API Health and Environmental Sciences Dept. Report 34-30636  Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



## Acute Toxicity

### Test Substance - Acute Toxicity

**Category Chemical:** (68955-35-1) Naphtha, petroleum, catalytic reformed

**Test Substance:** (68955-35-1) Naphtha, petroleum, catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:** API Test Material: API#83-05 Catalytically reformed naphtha

Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (see website below)

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

### Method - Acute Toxicity

**Route of Administration:** Inhalation

**Type of Exposure:** Vapor

**Species:** Rat

**Mammalian Strain:** Sprague-Dawley

**Gender:** Both M/F

**Number of Animals per Dose:** 5

**Dose:** 5000 mg/m3

**Year Study Performed:** 1984

**Method/Guideline Followed:** Unknown

**GLP:** Yes

**Method/Guideline and Test Condition Remarks:** A group of 5 male and 5 female rats were exposed by whole body inhalation to API 83-05 at a nominal concentration of 5mg/l for 4 hours. After the 4 hour exposure the rats were observed twice daily for mortality. The animals were weighed prior to exposure and again on days 7 and 14 post exposure. On day 14 all surviving animals were killed by exsanguination following methoxyflurane anesthesia and were subjected to a full necropsy. For all animals, including those found dead during the study the lungs were removed, fixed and examined histologically.

### Test Results - Acute Toxicity

**Concentration (LC/LD):**

LC/LD	%	Value Description	Value/Lower Concentration	Upper Concentration	Units
LC	50 %	>	5220		mg/m3

**Number of Deaths ( Male ):** 0/5

0/5

<b>Number of Deaths ( Female ):</b>	
<b>Number of Deaths ( Total ):</b>	0/10
<b>Results Remarks:</b>	<p>The exposure chamber TWA concentration was determined to be <math>5.22 \pm 0.14</math> mg/l. No animal died during the study and no clinical signs of systemic toxicity were observed. There were no significant gross observations at necropsy. Histological examination of lung tissues yielded minimal to moderate pulmonary findings. The possibility that these could be due to the exposure could not be ruled out.</p> <p>The number of deaths/sex is provided for treated animals only.</p> <p>The 4 hour LC50 was therefore greater than 5220 mg/m3.</p>
<b>Conclusion:</b>	The 4 hour LC50 was therefore greater than 5220 mg/m3.
<b>Reliability/Data Quality - Acute Toxicity</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	RELIABILITY: GLP study with adequately detailed methods description
<b>Key Study Sponsor Indicator:</b>	Key
<b>Reference - Acute Toxicity</b>	
<b>Reference:</b>	<p>American Petroleum Institute (1984)          Acute inhalation toxicity evaluation of a petroleum derived hydrocarbon in rats, full range catalytically reformed naphtha, API sample 83-05.          Study conducted by Litton Bionetics, Inc.          API Medical Research Publication No. 31-30681, February 1984.</p>



<b>Repeated-Dose Toxicity</b>																			
<b>Test Substance - Repeated-Dose Toxicity</b>																			
<b>Category Chemical:</b>	No CAS Number Provided																		
<b>Test Substance:</b>	No CAS Number Provided																		
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>																		
<b>Category Chemical Result Type:</b>	Read-Across																		
<b>Method - Repeated-Dose Toxicity</b>																			
<b>Route of Administration:</b>	Inhalation																		
<b>Type of Exposure:</b>	Vapor																		
<b>Species:</b>	Rat																		
<b>Mammalian Strain:</b>																			
<b>Gender:</b>																			
<b>Number of Animals per Dose:</b>																			
<b>Dose:</b>																			
<b>Year Study Performed:</b>																			
<b>Method/Guideline Followed:</b>																			
<b>GLP:</b>																			
<b>Exposure Period:</b>																			
<b>Frequency of Treatment:</b>																			
<b>Post-Exposure Period:</b>																			
<b>Method/Guideline and Test Condition Remarks:</b>																			
<b>Test Results - Repeated-Dose Toxicity</b>																			
<b>Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):</b>	<table border="1"> <thead> <tr> <th>LOAEL/ LOAEC/ NOAEL/ NOAEC</th> <th>Population</th> <th>Value Description</th> <th>Value/Lower Concentration</th> <th>Upper Concentration</th> <th>Units</th> </tr> </thead> <tbody> <tr> <td>LOAEC</td> <td></td> <td>=</td> <td>6572</td> <td>27800</td> <td>mg/m3</td> </tr> <tr> <td>NOAEC</td> <td></td> <td>=</td> <td>1507</td> <td>10153</td> <td>mg/m3</td> </tr> </tbody> </table>	LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units	LOAEC		=	6572	27800	mg/m3	NOAEC		=	1507	10153	mg/m3
LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units														
LOAEC		=	6572	27800	mg/m3														
NOAEC		=	1507	10153	mg/m3														
<b>Results Remarks:</b>	Results are for male and female rats combined																		
<b>Conclusion:</b>																			

Gasoline blending streams have a low inhalation repeat dose hazard potential. The inhalation NOAELs and LOAELs were similar between the different hydrocarbon classes of streams (PONA) and the formulated product, gasoline in rats. Since there were no appreciable differences between paraffinic, olefinic, naphthenic, and aromatic (PONA) streams, a range of values derived from all of the repeated dose inhalation studies will be used to read across to all untested category members. These read-across values are:  
 LOAEL: 6572 mg/m<sup>3</sup> - 27,800mg/m<sup>3</sup> (1864 - 7885ppm)  
 NOAEL: 1507mg/m<sup>3</sup> - 10,153mg/m<sup>3</sup> (427 - 2880ppm)

### Reliability/Data Quality - Repeated-Dose Toxicity

**Reliability:**

**Reliability  
Remarks:**

Studies used to derive the read across range were rated as either as "1 - valid without restrictions", or "2 - valid with restrictions"

**Key Study Sponsor  
Indicator:**

Weight of Evidence

### Reference - Repeated-Dose Toxicity

**Reference:**

See Repeated-Dose Robust Study Summaries for CAS #, 64741-41-9, 64741-55-5, 64741-63-5, 64741-66-8, and 86290-81-5



<b>Repeated-Dose Toxicity</b>	
<b>Test Substance - Repeated-Dose Toxicity</b>	
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	LCCN-D (Distillate of LCCN; Petroleum Product Stewardship Council test material)  Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (see website below)  Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Repeated-Dose Toxicity</b>	
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Number of Animals per Dose:</b>	16
<b>Dose:</b>	Target: 750, 2500 & 7500 ppm. (0, 2300, 7700, 23400 mg/m3) Actual: 756, 2507 & 7533 ppm.
<b>Year Study Performed:</b>	2001
<b>Method/Guideline Followed:</b>	EPA OTS 798.2450
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	15 Weeks
<b>Frequency of Treatment:</b>	6 hours/day, 5 days/week
<b>Post-Exposure Period:</b>	4 Weeks
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Groups of 16 male and 16 female rats underwent whole body exposures to 750, 2500 and 7500 ppm LCCN-D. Exposures were for 6 hours each day, 5 days per week, for at least 65 exposures, over a period of 15 weeks.</p> <p>Extra groups of 16 rats of each sex were exposed to the high dose level and also for a recovery control group. These animals were maintained untreated for 28 days following cessation of the 15 weeks exposure.</p> <p>Neurobehavioral evaluations of motor activity and functional activity were performed pretest and during weeks 5, 9, 14/15 and after the 4 week recovery period for the recovery animals. Animals were not exposed to LCCN-D during these tests.</p> <p>Following 15 weeks of exposure, 16 animals/sex/group were necropsied and microscopic examination was performed on selected</p>

tissues. Nervous tissue from 6 rats/sex/group was also examined microscopically.

At the end of the 4 week recovery period, 16 animals of each sex from the high and control groups were necropsied and selected tissues were examined microscopically.

During the study clinical observations were made twice daily. Ophthalmoscopic evaluations were performed pretest and just prior to the scheduled sacrifices at 15 weeks and 20 weeks (recovery groups). Body weights and food consumption were measured throughout the study. Blood samples were taken from 10 fasted rats/sex/group at 14 and 18 weeks for hematological and clinical chemical measurements.

At termination (after 15 weeks exposure for the main study and after 19 weeks for the recovery animals) all animals were killed and subjected to a complete macroscopic examination. 10 animals of each sex were designated for non-neuropathological examination and 6 of each sex for neuropathological examination. For the non neuropathology animals, the following organs were weighed: adrenals, brain, heart, kidneys, liver, lung, ovaries, prostate, spleen, testes (with epididymes), thymus and uterus. Brain lengths and widths were measured for each rat.

A wide range of tissues (39) were removed from the control and high dose animals and were fixed and examined histopathologically. Additionally, kidneys from selected animals were stained with Mallory-Heidenhain and examined. Tissues were also removed from the nervous system (central and peripheral) of all animals for subsequent special staining and histopathological examination.

Animals designated for neuropathological examination were subjected to a detailed examination of central and peripheral nervous tissues.

Neurobehavioral studies were undertaken as follows:

Motor activity Locomotor activity was monitored as the number of beam breaks in an activity box. Monitoring sessions were for 60 minutes, divided into twelve 5-minute intervals. Evaluation was made pretest and during weeks 5, 9, 15 and at the end of the 4 week recovery period. [A detailed description of the evaluation and analysis is provided in the publication but is not included here.]

Functional Operational Battery An assessment of the following was made:

Home cage evaluations for Posture, vocalization, palpebral closure.

Handling evaluations for reactivity to general stimuli, signs of autonomic function.

Open field behavior: arousal level, gait, urination and defecation frequency, convulsions, tremor, abnormal behavior, piloerection and exophthalmos.

Reflex assessments for: response to visual and auditory stimuli, tail pinch, pupillary function.

Animals were also evaluated for fore limb and hind limb grip strength, landing foot splay and air righting ability.

The test atmospheres were generated by wholly vaporizing the test material (LCCN-D) and diluting with air to achieve the required concentrations. The highest concentration was approximately 75% of the lower explosive limit.

Actual exposure concentrations were determined six times per exposure session for treated groups and once for controls.

Particle size determinations were carried out once during each exposure using an aerodynamic particle sizer. Mean mass aerodynamic diameter (MMAD), geometric standard deviation (GSD) and total mass concentration (TMC) were calculated. The actual concentrations for each of the target dose levels were:

Dose group	Actual TMC*
(ppm)	(ppm) (mg/m3)
0 (Control)	0 0.005820
750	756 0.005506
2500	2507 0.005085
7500	7533 0.004348

\* TMC = Total Mass Aerosol Concentration



### Test Results - Repeated-Dose Toxicity

Concentration  
(LOAEL/ LOAEC/  
NOAEL/ NOAEC):

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
LOAEC	Male	=	23400		mg/m3
NOAEC	Male	=	7700		mg/m3
LOAEC	Female	=	23400		mg/m3
NOAEL	Female	=	7700		mg/m3

#### Results Remarks:

No exposure-related clinical observations were noted either during exposure or during non-exposure periods and no ocular abnormalities were observed. Although the males in the high dose group were slightly lighter than the controls (total weight gain 344g compared to 323g), the difference was not significant. In the females however, the difference (total weight gain 165g compared to 154g) was statistically significant. At the end of the 4 week recovery period body weights of the high dose males and females were comparable to the corresponding controls. During the 4 week recovery period, the high dose males and females had food consumption that were greater (statistically significant) than controls. [Note: actual data not included in the draft publication]. At 15 weeks the following hematological changes were recorded.

7500 ppm males: Decreased hemoglobin concentration (8%) Decreased hematocrit (7%)

2500 ppm males: Decreased MCHC (3%)

7500 ppm females: Decreased MCHC (4%)

After the 4 week recovery period, all hematological values were considered to be normal.

At 15 weeks there were no abnormal clinical chemistry values. After the 4 week recovery period however, glucose and albumin was raised in the 7500 ppm females by 21 and 15% respectively. Since the values were within the normal range they were not considered to be toxicologically significant.

Neurobehavioral studies: There was no evidence of any effect on motor activity either after 15 weeks exposure or after the 4 week recovery period. There was no evidence of a treatment-related effect in the functional operational battery that was carried out.

Pathology: With the exception of those listed below, absolute and relative organ weights were not affected by treatment.

#### Group

Parameter 2500 ppm 7500 ppm Recovery

#### MALES

Abs Kidney 21% up

Rel Kidney 15% up 32% up

Rel Liver 23% up

#### FEMALES

Rel Kidney 18% up

Abs Liver 24% up

Rel Liver 12% up

Rel Brain 9% down

There were no microscopic findings in either the liver or brain of the groups in which organ weight changes had been recorded. The only treatment-related microscopic changes were found in the nasal turbinates and kidneys as follows.

Nasal turbinates: The following table summarizes the incidence of selected microscopic findings in the nasoturbinal tissues.

Numbers in table are male incidence/female incidence

Dose group (ppm)

0 750 2500 7500

Incidence at 15 weeks

No. evaluated 10/10 10/10 10/10 10/10

Goblet cell hypertrophy/hyperplasia

Score 1 3/2 1/4 1/4 1/1

2 7/6 5/5 7/3 5/5  
 3 0/1 2/1 2/3 3/3  
 Nasal mucosa hyperplasia  
 Score 1 0/1 0/4 1/2 1/0  
 2 2/3 3/3 6/5 5/5  
 3 0/0 0/0 1/1 1/2

Incidence in post-exposure animals  
 No. evaluated 10/10 0/0 0/0 10/10  
 Goblet cell hypertrophy/hyperplasia  
 Score 1 2/4 2/2

2 5/2 5/3  
 3 3/1 3/0  
 Nasal mucosa hyperplasia  
 Score 1 2/4 2/3  
 2 6/4 5/5  
 3 0/0 1/0

These findings are considered indicative of exposure to a mild irritant.

Kidney: At the end of 15 weeks exposure several changes were observed and at the end of the 4 week recovery period there was an indication of some reversibility of the kidney effects. The findings are summarized in the following table.

Finding Terminal Post-exposure  
 0 750 2500 7500 0 7500  
 No of animals evaluated  
 10 10 10 10 10 10  
 Bilateral cortex: eosinophilic hyaline droplets in proximal convoluted tubular epithelium with severity greater than or equal to 2  
 0 3 8 10 0 2  
 Postive Mallory/Heidenhain staining hyaline droplets in proximal convoluted tubular epithelium with severity greater than or equal to 2  
 1 4 9 10 1 2  
 Bilateral interstitium subacute/chronic inflammation  
 0 0 3 5 1 3  
 Bilateral cortex/cortico-medullary junction tubules dilated with granular casts  
 0 0 1 4 0 0  
 Bilateral cortex convoluted tubular basophilic epithelium  
 0 0 0 3 0 0

Similar effects were not observed in the females. In the post exposure animals, brain length and width measurements showed no test-material-related effects.

**Conclusion:**

In both males and females, the subchronic LOAEC = 7,500 ppm (23,400 mg/m3) and the NOAEC = 2500 ppm (7,700 mg/m3). These finding were based on nasal mucosa cell hyperplasia and goblet cell hypertrophy/hyperplasia.

Light hydrocarbon nephropathy was observed in males at all exposure levels. Since this finding is specific to male rats and not relevant for human risk assessment (U.S. Environmental Protection Agency. Alpha 2 microglobulin: association with chemically induced renal toxicity and neoplasia in the male rat. 1991. In Risk Assessment Forum. US Government Printing Office, Washington, DC: EPA: 85) it was excluded when deriving the subchronic LOAEL/NOAEL.

In both males and females the neurotoxicity NOAEL > 7500 ppm (23,400 mg/m3), the highest dose tested.

**Reliability/Data Quality - Repeated-Dose Toxicity**

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP; guideline study

**Key Study Sponsor Indicator:** Key

**Reference - Repeated-Dose Toxicity**

**Reference:** Lapin, C., Bui, Q., Breglia, R., Burnett, D., Koschier, F., Roth, R., Schreiner, C., White, R., Mandella, R. and Hoffman, G.

(2001) Toxicity evaluation of petroleum blending streams:  
Inhalation subchronic toxicity/neurotoxicity study of a light  
catalytic cracked naphtha distillate in rats. Int. J. Toxicol.  
Vol 20, pp 307-319

Posting dates of documents from HPV Challenge web site from which  
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<b>Repeated-Dose Toxicity</b>																																																								
<b>Test Substance - Repeated-Dose Toxicity</b>																																																								
<b>Category Chemical:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed																																																							
<b>Test Substance:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed																																																							
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Partially vaporized (30-40%) full range catalytic reformed naphtha (FR-CRN). See compositional data file attached to category at the website cited below.</p> <p>Test atmospheres were generated by partially vaporizing FRCRN. The concentrations in the chamber were adjusted by dilution with air. Concentrations were monitored throughout the study. The actual concentrations for each of the dose levels are shown below.</p> <table border="1"> <thead> <tr> <th rowspan="2">Parameter</th> <th colspan="3">Exposure group</th> </tr> <tr> <th>Low</th> <th>Medium</th> <th>High</th> </tr> </thead> <tbody> <tr> <td>Target conc. (mg/m3)</td> <td>500</td> <td>2000</td> <td>8000</td> </tr> <tr> <td>Actual conc. (mg/m3)</td> <td>410</td> <td>1970</td> <td>8050</td> </tr> <tr> <td>Butane</td> <td>4.33</td> <td>3.91</td> <td>4.05</td> </tr> <tr> <td>Methylbutane</td> <td>20.56</td> <td>17.26</td> <td>17.55</td> </tr> <tr> <td>Pentane</td> <td>13.24</td> <td>11.44</td> <td>11.86</td> </tr> <tr> <td>Hexane</td> <td>6.53</td> <td>5.71</td> <td>6.36</td> </tr> <tr> <td>Heptane</td> <td>2.32</td> <td>2.35</td> <td>2.33</td> </tr> <tr> <td>Benzene</td> <td>2.19</td> <td>4.93</td> <td>5.79</td> </tr> <tr> <td>Toluene</td> <td>10.02</td> <td>12.23</td> <td>10.93</td> </tr> <tr> <td>m-and p-Xylenes</td> <td>3.57</td> <td>4.05</td> <td>3.4</td> </tr> <tr> <td>2-Ethyltoluene</td> <td>0.43</td> <td>0.35</td> <td>0.17</td> </tr> <tr> <td>Trimethylbenzene</td> <td>0.01</td> <td>0.01</td> <td>0.04</td> </tr> </tbody> </table> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>	Parameter	Exposure group			Low	Medium	High	Target conc. (mg/m3)	500	2000	8000	Actual conc. (mg/m3)	410	1970	8050	Butane	4.33	3.91	4.05	Methylbutane	20.56	17.26	17.55	Pentane	13.24	11.44	11.86	Hexane	6.53	5.71	6.36	Heptane	2.32	2.35	2.33	Benzene	2.19	4.93	5.79	Toluene	10.02	12.23	10.93	m-and p-Xylenes	3.57	4.05	3.4	2-Ethyltoluene	0.43	0.35	0.17	Trimethylbenzene	0.01	0.01	0.04
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<b>Type of Exposure:</b>	Vapor																																																							
<b>Species:</b>	Rat																																																							
<b>Mammalian Strain:</b>	Sprague-Dawley																																																							
<b>Gender:</b>	Both M/F																																																							
<b>Number of Animals per Dose:</b>	15																																																							
<b>Dose:</b>	410, 1970 and 8050 mg/m3																																																							
<b>Year Study Performed:</b>	1996																																																							
<b>Method/Guideline Followed:</b>	Other																																																							
<b>GLP:</b>	No Data																																																							
<b>Exposure Period:</b>	13 Weeks																																																							
<b>Frequency of Treatment:</b>	6 hours/day, 5 days/week																																																							
<b>Post-Exposure Period:</b>	0																																																							

**Method/Guideline and Test Condition Remarks:**

Groups of 15 rats of each sex were individually housed in 1m<sup>3</sup> inhalation chambers. The rats underwent whole body exposures to partially vaporized full range catalytic reformed naphtha (FRCRN). Exposures were [6 hours/day, 5 days/week] for 13 weeks at nominal concentrations of 500, 2000 and 8000 mg/m<sup>3</sup>.

Two extra groups of 15 rats of each sex served as sham and untreated controls. (NB. This is not stated in the publication but from other comments in the paper, it is clear that exposure was not continual during the study).

Water was available ad lib, but food was withheld during the exposure periods.

Clinical observations were made regularly and body weights were recorded weekly.

At the end of the 13 weeks exposure, blood samples were taken for hematological and clinical chemical measurements. The rats were then sacrificed and necropsied. Organs were weighed and a wide range of tissues fixed for subsequent histology and microscopic examination. The wet weight of the right middle lung lobe was also weighed. The lobes were then dried and their dry weights determined. The cauda epididymis of the control and high dose male rats was used to determine the morphology and number of sperm and the left testis was used to determine the number of testicular spermatids.

The following tissues from the high dose animals were examined histologically: adrenals, bone and marrow (sternum), pancreas (head), brain (three sections), submaxillary salivary gland, eye, optic nerve, spleen, heart, stomach (squamous and glandular), colon, testes or ovaries, duodenum, kidneys, thymus, thyroid, liver, tracheobronchial lymph nodes, lung (left lobe), nasal turbinates (four sections), thigh muscle, urinary bladder, sciatic nerve, and any gross lesions. In addition, tracheobronchial lymph nodes and any gross lesions from untreated control animals were also evaluated.

**Test Results - Repeated-Dose Toxicity**

**Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):**

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
LOAEC	Male	=	8050		mg/m <sup>3</sup>
NOAEC	Male	=	1970		mg/m <sup>3</sup>
LOAEC	Female	=	8050		mg/m <sup>3</sup>
NOAEC	Female	=	1970		mg/m <sup>3</sup>

**Results Remarks:**

There were no treatment-related clinical signs during the study, no effects on serum chemistry values or parameters of the male reproductive system a terminal sacrifice. Body weights of males were exposed to the mid and high dose groups were higher than the controls throughout the study and the differences were statistically significant in the high dose group from week 10 onwards.

WBC count was significantly lower in sham treated controls and all three treated groups in both sexes compared to untreated controls. Additionally the WBC count was decreased by approximately 24% in the high dose females when compared to the sham controls. No other parameters were affected.

The only organ weights affected were the liver and kidney. In the male high dose group, mean kidney weight was approximately 13% greater than the sham treated animals (but not the untreated controls), and the liver weight was approximately 14% greater.

No treatment-related gross lesions were observed at necropsy and no treatment-related abnormalities were noted during microscopic examination. No hydrocarbon-induced nephropathy was observed in male rats in this study. Because of the lack of effects in the histology, no tissues were examined from the lower dose groups.

<b>Conclusion:</b>	The LOAEC = 8050mg/m3 based on increased liver and kidney weights in males, decreased WBC in females. NOAEC = 1970mg/m3 in both male and female rats.
<b>Reliability/Data Quality - Repeated-Dose Toxicity</b>	
<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	The publication is not clear in its description of the frequency and duration of exposures. However, it is assumed that the exposures are 6 hours/day, 5 days/week since this would be consistent with other studies reported from the same laboratory.
<b>Key Study Sponsor Indicator:</b>	Key
<b>Reference - Repeated-Dose Toxicity</b>	
<b>Reference:</b>	<p>Dalbey, W. and Feuston, M. (1996) Partially vaporized full range catalytic reformed naphtha: Subchronic and developmental toxicity studies in rats. Inhalation Toxicology. Vol 8., pp 271-284</p> <p>Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003</p>



<b>Repeated-Dose Toxicity</b>	
<b>Test Substance - Repeated-Dose Toxicity</b>	
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Repeated-Dose Toxicity</b>	
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Number of Animals per Dose:</b>	20
<b>Dose:</b>	<p>Target Concentrations:            Unleaded gasoline: 0, 400 &amp; 1500 ppm (0, 1570, and 6,350 mg/m<sup>3</sup>)            Leaded gasoline: 0, 100 &amp; 400 ppm (0, 420, and 1,530 mg/m<sup>3</sup>).</p> <p>Actual Concentrations:            Unleaded gasoline: 0, 384, and 1552 ppm (0, 1507, and 6571 mg/m<sup>3</sup>)            Leaded gasoline: 0, 103, 374 ppm (0, 420, and 1530 g/m<sup>3</sup>)</p>
<b>Year Study Performed:</b>	1984
<b>Method/Guideline Followed:</b>	
<b>GLP:</b>	No Data
<b>Exposure Period:</b>	13 Weeks
<b>Frequency of Treatment:</b>	6 hr/day, 5 days/week
<b>Post-Exposure Period:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	<p>This study was conducted as a preliminary range finding study prior to conducting a two year study on the same test materials.</p> <p>20 rats and 4 monkeys of each sex were housed in 1m<sup>3</sup> glass and stainless steel exposure chambers 24 hours a day and were only removed for cleaning purposes.</p> <p>Target exposure vapor concentrations of the test materials were:            Unleaded gasoline: 400 and 1500 ppm            Leaded gasoline: 100 and 400 ppm</p> <p>A control group of 20 rats and 4 monkeys of each sex were exposed to air only.            Exposures were for 6 hours each day, 5 days each week for 13</p>

weeks.

Blood was taken from 10 rats of each sex at the end of the study from the highest dose groups only for hematological evaluation. Blood was taken from all monkeys in the highest dose group at 1.5, and 3 months. Urine samples were analyzed for all animals at 1.5 and 3 months for levels of protein, glucose, ketones, bilirubin, blood and lead.

CNS evaluations were conducted on the monkeys in the control and high level dose groups at before exposure and at 3 months. The CNS evaluations consisted of recording simultaneous and evoked responses and this was accomplished using electrodes that had been implanted permanently in the visual cortex. Pulmonary function tests similar to those reported by Alarie were conducted on all monkeys prior to exposure and at 1.5 and 3 months on the control and high level unleaded groups. All animals that died or were sacrificed at termination of the study were subjected to a gross necropsy. Organ weights were recorded and lungs, kidneys, spleen, heart, brain and bone marrow from the control and high dose groups were evaluated for histopathology. All male and female animal from the control and high exposure groups were also evaluated for the presence of IgG in the renal glomerulus and lungs. A lead analysis was also made on rat brain, kidney, liver, urine and blood from both the leaded dose groups and controls.

The gasoline samples were piped to an atomizer to which nitrogen heated to 105 °C was also fed at a pressure of 10 psig. The atomized gasoline was then carried to the exposure chamber with air. Exposure chamber atmospheres were analyzed for gasoline vapor concentration twice daily. The mean exposure concentrations for the two gasoline samples were as follows:  
 Target concentration Gasoline vapor exposure concentration  
 mg/liter ppm Alkyl lead  
 ±SD µg Pb/l ±SD  
 0 ppm (control)  
 Unleaded gasoline  
 1500 ppm 6.35 ± 0.44 1552 -  
 400 ppm 1.57 ± 0.15 384 -  
 Leaded gasoline  
 400 ppm 1.53 ± 0.23 374 0.72 ± 0.10  
 100 ppm 0.42 ± 0.04 103 0.19 ± 0.04

#### Test Results - Repeated-Dose Toxicity

Concentration  
(LOAEL/ LOAEC/  
NOAEL/ NOAEC):

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
LOAEC	Male	=	6350		mg/m3
NOAEC	Male	=	1570		mg/m3
LOAEC	Female	=	6350		mg/m3
NOAEC	Female	=	1570		mg/m3

#### Results Remarks:

Test results presented in the Test Results table are for UNLEADED GASOLINE



Gasoline vapor  
 exposure concentration Alkyl lead  
 Group Mg/l ppm µg Pb/l  
 ±SD ±SD  
 Control - - -  
 Unleaded 1500 ppm 6.35±0.44 1552 -  
 Unleaded 400 ppm 1.57±0.15 384 -  
 Leaded 400 ppm 1.53±0.23 374 0.72±0.1  
 Leaded 100 ppm 0.42±0.04 103 0.19±0.04

Three rats at different dose levels and three monkeys also at different dose levels died during the study. These deaths were not considered to be treatment-related.

Two female monkeys in each of the high dose groups exhibited emesis, 13 and 17 days after commencing exposure for the 1500 ppm unleaded and 400 ppm leaded groups respectively. Although there was a reduction in body weights in males in the lowest dose group of each of the test materials but by the end of the study they were demonstrating increased weights. No differences were observed in any of the other treated groups.

The hematological values for the monkeys exposed to either test material at either dose level were similar to those for the control animals. In the rats the only changes observed were:  
 unleaded (1500 ppm males) 64% increase in thrombocytes  
 unleaded (1500 ppm females) 150% increase in reticulocytes  
 leaded (400 ppm males) 4% decrease in MCHC  
 leaded (400 ppm females) 10% increase in hematocrit  
 leaded (400 ppm females) 11% increase in MCV  
 leaded (400 ppm females) 25% decrease in WBC

Mean flash-evoked response time for the monkeys was measured prior to exposure and was unaffected by exposure.

The results of the mean pulmonary function data are summarised in the following table. Only increases (+%) or decreases (-%) compared to controls are shown in the table. All other parameters were similar for treated and control animals.

Pre-exposure 42 days 90 days  
 Respiratory rate  
 Unleaded 1500 ppm F - - -  
 Unleaded 1500 ppm M -30% -21% -  
 Leaded 400 ppm F - - -  
 Leaded 400 ppm M - - -  
 Tidal volume  
 Unleaded 1500 ppm F - - -22%  
 Unleaded 1500 ppm M - - -  
 Leaded 400 ppm F - - -  
 Leaded 400 ppm M - - -  
 Minute volume  
 Unleaded 1500 ppm F - - -  
 Unleaded 1500 ppm M - - +36%  
 Leaded 400 ppm F - - -  
 Leaded 400 ppm M - - +53%

There were no effects on airway resistance, dynamic compliance or breaths to 1% nitrogen.

Urinalysis showed no differences between treated and control animals in either species. There was no evidence of IgG deposition in the kidneys of rats or monkeys of either sex following exposure to the test materials for 90 days.  
 Group mean lead levels in the rat tissues were as follows:

Leaded Unleaded  
 Control 400 ppm 100 ppm  
 Brain M 1.26 9.49 7.23  
 F 1.44 5.39 2.32  
 Kidney M 1.71 12.4 7.06  
 F 2.97 9.57 13  
 Liver M 0.71 17.9 6.51  
 F 1.21 19.7 8.41  
 Blood M 0.61 6.1 0.77  
 F 0.24 1.32 0.46  
 Urine M 0.17 0.21 0.19  
 F 0.31 0.18 0.25

No actual values are given on organ weights or organ/body weight ratios but the following effects are reported:

Rats  
 Liver wt Kidney wt  
 Unleaded 400 ppm M increased  
 Unleaded 400 ppm F  
 Unleaded 1500 ppm M  
 Unleaded 1500 ppm F  
 Leaded 400 ppm M  
 Leaded 400 ppm F decreased  
 Leaded 100 ppm M increased  
 Leaded 100 ppm F increased

Monkeys  
 Thyroid Kidney  
 Unleaded 400 ppm M increased  
 Unleaded 400 ppm F  
 Unleaded 1500 ppm M increased  
 Unleaded 1500 ppm F  
 Leaded 400 ppm M decreased  
 Leaded 400 ppm F  
 Leaded 100 ppm M  
 Leaded 100 ppm F

Organ weights were also expressed as % of body weight and the following effects were recorded:

Rats:  
 Decreased heart weight in both male leaded groups  
 Decreased brain weight in both male unleaded groups  
 Decreased liver weight in 400 ppm female leaded group  
 Decreased adrenal weight in 1500 ppm female unleaded group.  
 Monkeys:  
 Decreased kidney weight in 400 ppm male unleaded group.

No evidence of treatment-related histopathology was observed in either rats or monkeys, with the exception of lesions noted in the kidneys of all male rats.

The lesions were characterized by subtle but discernible increases in the incidence and severity of regenerative epithelium and dilated tubules. The latter were seen to contain protein in their lumens.

The kidney lesions in males are now attributed to light hydrocarbon nephropathy (LHN) that is specific to male rats. Since male rat LHN is not relevant for human risk, it was not taken into account to determine the study LOAEL and NOAEL.

**Conclusion:**

LOAEC and NOAEC values for rats are as follows: (see separate RSS for Monkey results)

UNLEADED GASOLINE: [NOTE unleaded gasoline results ONLY in results table]  
 Male LOAEC = 1552 ppm (6.35 g/m<sup>3</sup>), based upon increased thrombocyte count  
 Male NOAEC = 384 ppm (1.57 g/m<sup>3</sup>)  
 Female LOAEC = 1552 ppm (6.35 g/m<sup>3</sup>), based upon increased reticulocyte count  
 Female NOAEC = 384 ppm (1.57 g/m<sup>3</sup>)

LEADED GASOLINE: [results NOT presented in table above]  
 Male LOAEC = 374 ppm (1.53 g/m<sup>3</sup>), based upon increased thrombocyte count  
 Male NOAEC = 103 ppm (0.42 g/m<sup>3</sup>)  
 Female LOAEC = 374 ppm (1.53 g/m<sup>3</sup>), based upon decreased WBC count  
 Female NOAEC = 103 ppm (0.42 g/m<sup>3</sup>)

Male LOAEL/NOAEL values excluded male rat specific nephropathy findings

### Reliability/Data Quality - Repeated-Dose Toxicity

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Although the GLP status of this study is unknown, the study is generally well described in the peer reviewed publication.

**Key Study Sponsor Indicator:** Key

### Reference - Repeated-Dose Toxicity

**Reference:** Kuna, R. A. and Ulrich, C. E. (1984) Subchronic inhalation toxicity of two motor fuels. J. American College of Toxicology. Vol 3. No. 4. 217-229.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Repeated-Dose Toxicity</b>																																															
<b>Test Substance - Repeated-Dose Toxicity</b>																																															
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate																																														
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<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>The test material (LAN-D) was a distillate of a Light Alkylate Naphtha (LAN). The composition and uniformity chamber gas chromatographic results (% weight) were:</p> <table border="1"> <thead> <tr> <th rowspan="2">Component</th> <th rowspan="2">Liquid</th> <th colspan="2">Vapor</th> </tr> <tr> <th>At Start</th> <th>Termination</th> </tr> </thead> <tbody> <tr> <td>n-butane</td> <td>2.442</td> <td>3.217</td> <td>3.210</td> </tr> <tr> <td>iso-pentane</td> <td>29.854</td> <td>33.517</td> <td>34.343</td> </tr> <tr> <td>2,3-dimethylbutane</td> <td>12.437</td> <td>11.963</td> <td>12.977</td> </tr> <tr> <td>2-methylpentane</td> <td>4.064</td> <td>4.775</td> <td>4.096</td> </tr> <tr> <td>2,4-dimethylpentane</td> <td>5.923</td> <td>5.663</td> <td>5.663</td> </tr> <tr> <td>2,3-dimethylpentane</td> <td>2.904</td> <td>2.794</td> <td>2.680</td> </tr> <tr> <td>2,2,4-trimethylpentane</td> <td>18.35</td> <td>16.897</td> <td>16.885</td> </tr> <tr> <td>2,3,4-trimethylpentane</td> <td>4.343</td> <td>3.772</td> <td>3.578</td> </tr> <tr> <td>2,3,3-trimethylpentane</td> <td>5.258</td> <td>4.614</td> <td>4.505</td> </tr> <tr> <td>2,2,5-trimethylhexane</td> <td>3.096</td> <td>2.641</td> <td>2.499</td> </tr> </tbody> </table> <p>Chamber concentrations were monitored throughout the study. Actual chamber concentrations were close to target concentrations. Particle mass distribution measurements confirmed that no measurable test material was present as aerosol.</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>	Component	Liquid	Vapor		At Start	Termination	n-butane	2.442	3.217	3.210	iso-pentane	29.854	33.517	34.343	2,3-dimethylbutane	12.437	11.963	12.977	2-methylpentane	4.064	4.775	4.096	2,4-dimethylpentane	5.923	5.663	5.663	2,3-dimethylpentane	2.904	2.794	2.680	2,2,4-trimethylpentane	18.35	16.897	16.885	2,3,4-trimethylpentane	4.343	3.772	3.578	2,3,3-trimethylpentane	5.258	4.614	4.505	2,2,5-trimethylhexane	3.096	2.641	2.499
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	<p>The nominal and actual concentrations for each of the target dose levels were:</p> <table border="1"> <thead> <tr> <th>Dose group (ppm)</th> <th>Nominal (ppm)</th> <th>Actual (ppm)</th> <th>TMC* (mg/m3)</th> </tr> </thead> <tbody> <tr> <td>0 (Control)</td> <td>0</td> <td>0</td> <td>3.8</td> </tr> <tr> <td>675</td> <td>719</td> <td>668</td> <td>3.7</td> </tr> <tr> <td>2250</td> <td>2073</td> <td>2220</td> <td>3.9</td> </tr> <tr> <td>6750</td> <td>7127</td> <td>6669</td> <td>4.2</td> </tr> </tbody> </table>	Dose group (ppm)	Nominal (ppm)	Actual (ppm)	TMC* (mg/m3)	0 (Control)	0	0	3.8	675	719	668	3.7	2250	2073	2220	3.9	6750	7127	6669	4.2																										
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	6750 (recovery) 6768      6623      3.2 * TMC = Total Mass Aerosol Concentration
<b>Year Study Performed:</b>	1998
<b>Method/Guideline Followed:</b>	OECD 413
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	13 Weeks
<b>Frequency of Treatment:</b>	6 hours/day, 5 days/week
<b>Post-Exposure Period:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Groups of 12 male and 12 female rats underwent whole body exposures to 668, 2220 and 6646 ppm LAN-D. Exposures were for 6 hours each day, 5 days per week for 13 weeks. Extra groups of 12 rats of each sex were exposed to the high dose level and also for a recovery control group. These animals were maintained untreated for 28 days following cessation of the 13 weeks exposure. Neurobehavioural evaluations of motor activity and functional activity were performed pretest and during weeks 5, 9, 14 and 18 recovery groups. Animals were not exposed to LAN-D during these tests. Following 13 weeks of exposure, 12 animals/sex/group were necropsied and microscopic examination was performed on selected tissues. Nervous tissue from 6 rats/sex/group was also examined microscopically. At the end of the 4 week recovery period, 12 animals of each sex from the high and control groups were necropsied and selected tissues were examined microscopically. During the study clinical observations were made twice daily. Ophthalmoscopic evaluations were performed pretest and just prior to the scheduled sacrifices at 14 weeks and 18 weeks (recovery groups). Body weights and food consumption was measured throughout the study. Blood samples were taken from 12 fasted rats/sex/group at 14 and 18 weeks for hematological and clinical chemical measurements. At termination (after 13 weeks exposure for the main study and after 18 weeks for the recovery animals) all animals were killed and subjected to a complete macroscopic examination. The following organs were weighed: adrenals, brain, heart, kidneys, liver, lung, ovaries, prostate, spleen, testes (with epididymes), thymus and uterus. Brain lengths and widths were measured for each rat. A wide range of tissues (39) were removed from the control and high dose animals, were fixed and examined histopathologically. Additionally, kidneys from selected animals were stained with Mallory-Heidenhain and examined. Tissues were also removed from the nervous system (central and peripheral) of all animals for subsequent special staining and histopathological examination. Nervous system tissues were selected randomly from 6 rats/sex/group in the high dose and controls at the end of 13 weeks for microscopic examination. Specific brain regions examined were forebrain, cerebral cortex, hippocampus, basal ganglia, midbrain cerebellum and pons and medulla.</p> <p>Neurobehavioural studies were undertaken as follows: Motor activity: Locomotor activity was monitored as the number of beam breaks in an activity box. Monitoring sessions were for 60 minutes, divided into twelve 5-minute intervals. Evaluation was made pretest and during weeks 5, 9, 14 and at the end of the 4 week recovery period. [A detailed description of the evaluation and analysis is provided in the publication but is not included here.]</p> <p>Functional Operational Battery: An assessment of the following was made: Home cage evaluations for Posture, vocalization, palpebral closure. Handling evaluations for reactivity to general stimuli, signs of autonomic function. open field behavior: arousal level, gait, urination and defecation frequency, convulsions, tremor, abnormal behavior, piloerection and exophthalmos. Reflex assessments for: response to visual and auditory stimuli, tail pinch, pupillary function.</p> <p>Animals were also evaluated for fore limb and hind limb grip strength, landing foot splay and air righting ability.</p>

The test atmospheres were generated by wholly vaporizing the test material (LAN-D) and diluting with air to achieve the required concentrations. The highest concentration was approximately 75% of the lowest explosive limit. Nominal concentrations were calculated from the loss of weight from the generation apparatus divided by the total airflow through the chamber during exposure. Actual exposure concentrations were determined three times daily by gas chromatography. Particle size determinations were also carried out once during each exposure using an aerodynamic particle sizer. Mean mass aerodynamic diameter (MMAD), geometric standard deviation (GSD) and total mass concentration (TMC) were calculated. The nominal and actual concentrations for each of the target dose levels were:

### Test Results - Repeated-Dose Toxicity

Concentration  
(LOAEL/ LOAEC/  
NOAEL/ NOAEC):

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
LOAEC	Male	=	24300		mg/m3
NOAEC	Male	=	8102		mg/m3
LOAEC	Female	=	24300		mg/m3
NOAEC	Female	=	8102		mg/m3

### Results Remarks:

There were no mortalities during the study and there were no treatment related signs of toxicity. A possible treatment related sign was an increased incidence of red facial staining in rats of both sexes in the high dose group. Mean body weights, body weight gains and food consumption was unaffected by treatment.

Hematological and clinical chemical measurements were unaffected except for a 5% decrease in hemoglobin, a 5% decrease in hematocrit and a 7% decrease in erythrocytes. The hemoglobin was still decreased (4%) after the 4 week recovery period. However, it was considered that these differences were toxicologically unimportant because they were small and within the historical range for the test laboratory.

Although there were some changes in AST and ALT in high dose females they were not considered to be toxicologically significant because several control animals also had elevated levels for these enzymes in the control groups and also relative to historical controls. The organ weight changes were few. Statistically significant increases in kidney weights in high dose males correlated with microscopically observed hyaline droplet formation and degeneration of proximal renal tubules were observed, indicative of alpha 2-microglobulin mediated nephropathy, also identified as light hydrocarbon nephropathy, a species and sex specific syndrome not relevant to humans (U.S. Environmental Protection Agency. Alpha 2 microglobulin: association with chemically induced renal toxicity and neoplasia in the male rat. 1991. In Risk Assessment Forum. US Government Printing Office, Washington, DC: EPA: 85.).

Absolute and relative liver weights were observed in the high dose males and females at 13 weeks but the differences had disappeared after the recovery period. There were no pathological findings associated with this increase. The magnitude of the organ weight increases is shown below.

Dose level (ppm)  
6 68 2220 6646 Recovery  
Males  
Abs. Kidney wt. 13.2 19.8 27 23  
Rel. Kidney wt. 18 30 11  
Abs. Liver wt. 21  
Rel. Liver wt. 25

Females  
Abs. Liver wt. 17 12  
Rel. Liver wt. 12

In the neurobehavioral studies no treatment-related effects were observed in the functional operational battery. In the study of motor activity there were some statistically significant differences, but overall they did not occur in a dose related manner and furthermore were smaller than some of the differences seen during the pre dosing period.

**Conclusion:**

LAN-D was not a neurotoxicant in the neurobehavioral studies that were conducted. LAN-D did induce a light hydrocarbon nephropathy in the male rats at all exposure levels, but this is regarded as species and sex specific and not relevant for human health risk assessment (U.S. Environmental Protection Agency. Alpha 2 microglobulin: association with chemically induced renal toxicity and neoplasia in the male rat. 1991. In Risk Assessment Forum. US Government Printing Office, Washington, DC: EPA: 85.). The light hydrocarbon nephropathy was not taken into account for determination of subchronic LOEC and NOAEC values.

Excluding the nephropathy, the NOAEC for subchronic toxicity in male and female Sprague-Dawley rats was 2220 ppm (8102 mg/m<sup>3</sup>; based on reversible increased liver weights); the LOAEC for subchronic toxicity in male and female Sprague-Dawley rats was 6646 ppm (24300 mg/m<sup>3</sup>; based on reversible increased liver weights).

The neurotoxicity NOAEC in male and female Sprague-Dawley rats was 6646 ppm (24300 mg/m<sup>3</sup>; no neurotoxicity effects at highest dose tested).

### Reliability/Data Quality - Repeated-Dose Toxicity

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP; guideline study

**Key Study Sponsor Indicator:** Key

### Reference - Repeated-Dose Toxicity

**Reference:** Schreiner, C., Lapadula, E., Breglia, R., Bui, Q., Burnett, D., Koschier, F., Podhasky, P. and White, R. (1998) Toxicity evaluation of petroleum blending streams: inhalation subchronic toxicity/neurotoxicity study of a light alkylate naphtha distillate in rats. J. Toxicol. and Env. Health, Part A, Vol 55, pp 277-296

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



## Repeated-Dose Toxicity

### Test Substance - Repeated-Dose Toxicity

**Category Chemical:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance:** (64741-63-5) Naphtha, petroleum, light catalytic reformed

**Test Substance Purity/Composition and Other Test Substance Comments:** Distillate of a Light catalytically reformed naphtha (LCRN-D). See compositional data file attached to category.

The LCRN-D was pumped onto the central glass helix of a volatilization chamber. Nitrogen was passed upwards through the chamber over the heated coil and the volatilized material was suitably diluted with air to achieve the desired concentration. A separate volatilization chamber was used for each dose concentration. During the study the exposure chamber concentrations were monitored hourly. The composition of the vapor (vol. %) is shown in the following table:

```
Parameter LCRN-D
vapor
Olefins 1.37
Paraffins 88.3
Naphthenes ) 1.24
Aromatics 9.09
Benzene 4.65
Carbon No.
4 3.6
5 59.11
6 25.18
7 11.65
8 0.46
9 0
```

There was a gas chromatographic analysis of the LCRN-D at the beginning and at the end of the study. The results (expressed in wt %) were as follows:

```
Component LCRN-D vapor
(wt %)
Study Study
Beginning Termination
n-Butane 3.34 3.16
n-Pentane 20.38 20.43
Isopentane 35.31 34.70
1-Pentene 0.05 0.05
2-Methyl-2-butene 0.35 0.36
2-Methyl-1-butene 0.20 0.20
2,2-Dimethylbutane 2.22 2.22
n-Hexane 4.27 4.34
Methylcyclopentane 0.48 0.49
2,3-Dimethylbutane 1.54 1.59
2-Methylpentane 6.62 6.73
3-Methylpentane 4.54 4.62
Benzene 6.42 6.61
2-Methylhexane 1.65 1.67
3-Methylhexane 1.83 1.86
n-Heptane 0.90 0.91
Toluene 5.65 5.76
```

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

### Method - Repeated-Dose Toxicity

Inhalation



<b>Route of Administration:</b>	
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Number of Animals per Dose:</b>	16
<b>Dose:</b>	0, 750, 2500 and 7500 ppm (0, 2775, 9250, and 27750 mg/m3)
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	OECD 413
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	13 Weeks
<b>Frequency of Treatment:</b>	6 hours/day, 5 days/week
<b>Post-Exposure Period:</b>	4 Weeks
<b>Method/Guideline and Test Condition Remarks:</b>	<p>The method used was described in OECD guideline 413. Groups of 16 male and 16 female rats underwent whole body exposures to 750, 2500 and 7500 ppm LCRN-D(0, 2775, 9250, and 27750 mg/m3). Exposures were for 6 hours each day, 5 days per week for 13 weeks. Extra groups of 16 rats of each sex were exposed to the high dose level and also for a recovery control group. These animals were maintained untreated for 28 days following cessation of the 13 weeks exposure.</p> <p>Neurobehavioural evaluations of motor activity and functional activity were performed pretest and during weeks 5, 9, 14 and 19 (recovery groups). Animals were not exposed to LCRN-D during these tests.</p> <p>Following 13 weeks of exposure, 16 animals/sex/group were necropsied and microscopic examination was performed on selected tissues. Nervous tissue from 6 rats/sex/group was also examined microscopically.</p> <p>At the end of the 4 week recovery period, 16 animals of each sex from the high and control groups were necropsied and selected tissues were examined microscopically.</p> <p>During the study, clinical observations were made twice daily. Ophthalmoscopic evaluations were performed pretest and just prior to the scheduled sacrifices at 14 weeks and 19 weeks (recovery groups). Body weights and food consumption was measured throughout the study. Blood samples were taken from 10 fasted rats/sex/group at 14 and 18 weeks for hematological and clinical chemical measurements.</p> <p>At termination (after 13 weeks exposure for the main study and after 19 weeks for the recovery animals) all animals were killed and subjected to a complete macroscopic examination. 10 animals of each sex were designated for non-neuropathological examination and 6 of each sex for neuropathological examination.</p> <p>For the non-neuropathology animals, the following organs were weighed: adrenals, brain, heart, kidneys, liver, lung, ovaries, prostate, spleen, testes (with epididymes), thymus and uterus. Brain lengths and widths were measured for each rat.</p> <p>A wide range of tissues (39) was removed from the control and high dose animals, were fixed and examined histopathologically. Additionally, kidneys from selected animals were stained with Mallory-Heidenhain and examined. Tissues were also removed from</p>

the nervous system (central and peripheral) of all animals for subsequent special staining and histopathological examination. Animals designated for neuropathological examination were subjected to a detailed examination of central and peripheral nervous tissues.

Neurobehavioural studies were undertaken as follows:

#### Motor activity

Locomotor activity was monitored as the number of beam breaks in an activity box. Monitoring sessions were for 60 minutes, divided into twelve 5-minute intervals. Evaluation was made pretest and during weeks 5, 9, 14 and at the end of the 4 week recovery period. [A detailed description of the evaluation and analysis is provided in the publication but is not included here.]

#### Functional Operational Battery

An assessment of the following was made:

Home cage evaluations for Posture, vocalization, palpebral closure.

Handling evaluations for reactivity to general stimuli, signs of autonomic function.

open field behavior: arousal level, gait, urination and defecation frequency, convulsions, tremor, abnormal behavior, piloerection and exophthalmos.

Reflex assessments for: response to visual and auditory stimuli, tail pinch, pupillary function.

Animals were also evaluated for fore limb and hind limb grip strength, landing foot splay and air righting ability.

The LCRN-D was pumped onto the central glass helix of a volatilization chamber. Nitrogen was passed upwards through the chamber over the heated coil and the volatilized material was suitably diluted with air to achieve the desired concentration. A separate volatilization chamber was used for each dose concentration.

During the study the exposure chamber concentrations were monitored hourly. The composition of the vapor (vol. %) is shown in the following table:

Parameter LCRN-D vapor

Olefins	1.37
Paraffins	88.3
Naphthenes )	1.24
Aromatics	9.09
Benzene	4.65
Carbon No.	
4	3.6
5	59.11
6	25.18
7	11.65
8	0.46
9	0

There was a gas chromatographic analysis of the LCRN-D at the beginning and at the end of the study. The results (expressed in wt %) were as follows:

Component LCRN-D vapor (wt %)

Study	Study
Beginning	Termination
n-Butane	3.34 3.16
n-Pentane	20.38 20.43
Isopentane	35.31 34.70
1-Pentene	0.05 0.05
2-Methyl-2-butene	0.35 0.36
2-Methyl-1-butene	0.20 0.20
2,2-Dimethylbutane	2.22 2.22
n-Hexane	4.27 4.34
Methylcyclopentane	0.48 0.49
2,3-Dimethylbutane	1.54 1.59
2-Methylpentane	6.62 6.73
3-Methylpentane	4.54 4.62
Benzene	6.42 6.61
2-Methylhexane	1.65 1.67
3-Methylhexane	1.83 1.86

n-Heptane 0.90 0.91  
Toluene 5.65 5.76

### Test Results - Repeated-Dose Toxicity

Concentration  
(LOAEL/ LOAEC/  
NOAEL/ NOAEC):

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
LOAEC	Male	=	27750		mg/m3
NOAEC	Male	=	9250		mg/m3
NOAEC	Female	>	27750		mg/m3

#### Results Remarks:

There were no mortalities during the study and there were no treatment-related signs of toxicity. The ophthalmic examinations did not reveal any treatment-related effects. Mean body weights, body weight gains and food consumption were unaffected by treatment.

No treatment-related effects were recorded in the Functional Operational Battery. In the examinations of motor activity, there were no treatment-related effects recorded during the 13 week exposure period but a slight increased activity was found in the highest treatment group after the 4 week recovery period.

After 13 weeks exposure there was a significant decrease in total WBC count (36%) and lymphocyte counts in the high dose males and a slight decrease in neutrophil counts for the mid dose males. A trend towards decreased WBC (2.1%) and lymphocyte counts was also seen in the mid dose males and high dose females. After the 4 week recovery period, leukocyte values were comparable to control values. However, MCV was slightly decreased (2.8%) in the high dose males. It was concluded that these changes were suggestive of a reversible slight effect of the LCRN-D.

Clinical chemistry parameters were unaffected by treatment.

After 13 weeks exposure relative kidney weights in the high dose males were increased (15.9%) and this correlated with the occurrence of hyaline droplets in the proximal convoluted tubules. This finding has been described as a "light hydrocarbon nephropathy" and is sex and species specific and is not relevant for human health risk assessment.

In the high dose males decreased absolute (25.7%) and relative (22%) spleen weights were also recorded. It was concluded that this was associated with the minor hematological changes that had been observed. These differences were not apparent after the recovery period and no abnormal microscopic findings were found in either the spleen or bone marrow.

Brain length and width measurements were unaffected by treatment and there were no abnormal microscopic findings in the brain, spinal cord or peripheral nerves.

#### Conclusion:

The male systemic LOAEC exclusive of kidney effects was 27,750 mg/m3 based upon the decreased WBC and lymphocyte counts. The male neurotoxicity LOAEL was also 27,750 mg/m3, based on the increased motor activity in the high dose recovery group. The system and neurotoxicity NOAEC for male rats was 9250 mg/m3.

There were no systemic or neurotoxic effects observed in female rats; the NOAEC for both endpoints was greater than 27,750 mg/m3.

### Reliability/Data Quality - Repeated-Dose Toxicity

#### Reliability:

Valid Without Restrictions

#### Reliability Remarks:

RELIABILITY: GLP; guideline study

#### Key Study Sponsor Indicator:

Key

**Reference - Repeated-Dose Toxicity****Reference:**

Schreiner, C., Bui, Q., Breglia, R., Burnett, D., Koschier, F., Lapadula, E., Podhasky, P., White, R., Hoffman, G. and Mandella, R. (2000) Toxicity evaluation of petroleum blending streams: Inhalation subchronic toxicity/neurotoxicity study of a light catalytic reformed naphtha distillate in rats. *J. Tox. and Env. Health, Part A*. Vol. 60, pp 489-512

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Repeated-Dose Toxicity</b>																											
<b>Test Substance - Repeated-Dose Toxicity</b>																											
<b>Category Chemical:</b>	(64741-41-9) Naphtha, petroleum, heavy straight-run																										
<b>Test Substance:</b>	(64741-41-9) Naphtha, petroleum, heavy straight-run																										
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Naphtha, petroleum, heavy straight-run, Colorless liquid. MW 111.25. The test substance is a mixture that contains approximately 225 volatile hydrocarbons. The purity of the mixture is 100%            Stable based on analyses of chamber atmosphere.            12 Representative Components monitored in Study</p> <table border="1"> <thead> <tr> <th>Component</th> <th>Volume %</th> </tr> </thead> <tbody> <tr><td>2-Methyl C6 + C7-olefin</td><td>4.50</td></tr> <tr><td>3-Methylhexane</td><td>3.52</td></tr> <tr><td>t-1,3-Dimethylcyclopentane</td><td>1.45</td></tr> <tr><td>t-1,2-Dimethylcyclopentane</td><td>1.61</td></tr> <tr><td>n-Heptane</td><td>7.23</td></tr> <tr><td>Methylcyclohexane</td><td>6.76</td></tr> <tr><td>Toluene</td><td>3.44</td></tr> <tr><td>2-Methylheptane</td><td>3.25</td></tr> <tr><td>n-Octane</td><td>5.81</td></tr> <tr><td>Ethylcyclohexane</td><td>1.95</td></tr> <tr><td>m-Xylene</td><td>1.71</td></tr> <tr><td>n-Nonane</td><td>4.47</td></tr> </tbody> </table> <p>Substance is in the Gasoline Blending Streams Category.            See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>	Component	Volume %	2-Methyl C6 + C7-olefin	4.50	3-Methylhexane	3.52	t-1,3-Dimethylcyclopentane	1.45	t-1,2-Dimethylcyclopentane	1.61	n-Heptane	7.23	Methylcyclohexane	6.76	Toluene	3.44	2-Methylheptane	3.25	n-Octane	5.81	Ethylcyclohexane	1.95	m-Xylene	1.71	n-Nonane	4.47
Component	Volume %																										
2-Methyl C6 + C7-olefin	4.50																										
3-Methylhexane	3.52																										
t-1,3-Dimethylcyclopentane	1.45																										
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Ethylcyclohexane	1.95																										
m-Xylene	1.71																										
n-Nonane	4.47																										
<b>Category Chemical Result Type:</b>	Measured																										
<b>Method - Repeated-Dose Toxicity</b>																											
<b>Route of Administration:</b>	Inhalation																										
<b>Type of Exposure:</b>	Vapor																										
<b>Species:</b>	Rat																										
<b>Mammalian Strain:</b>	Sprague-Dawley																										
<b>Gender:</b>	Both M/F																										
<b>Number of Animals per Dose:</b>	12																										
<b>Dose:</b>	<p>Target: 0, 100, 500, 3000ppm (0, 455, 2275, 13650mg/m3)            Actual: 0, 100, 520, 2950ppm (0, 455, 2366, 13423mg/m3)            The mean concentrations (SE) representing the total area for the approximately 225 components contained in the test substance over the test period were 100(0.8), 500(2.0), and 3000(8.3)ppm in chambers targeted at 100, 500, and 3000ppm, respectively. Results from the cryogenic GC analysis indicated that the components were present in the chamber atmosphere within expected concentrations.</p>																										
<b>Year Study Performed:</b>	2008																										
<b>Method/Guideline Followed:</b>	OECD 422																										
<b>GLP:</b>	Yes																										
<b>Exposure Period:</b>	30 - 31 Days																										
<b>Frequency of Treatment:</b>	6 hours/day, 7 days/week																										

**Post-Exposure Period:**

0

**Method/Guideline and Test Condition Remarks:**

Note - details of the reproductive/developmental portion of the OECD 422 screen are presented in a separate Robust Study Summary

Concentrations of Naphtha vapor were generated by flash evaporation of the test material. An air control group was also evaluated using a similar generation apparatus; however, no test material was supplied to this vapor generator. Vapor concentrations of Naphtha were measured by gas chromatography (GC) using the area sum function and integrating all of the eluted peaks. Additional air samples were collected weekly and analyzed for 12 of the larger, most representative components of the test substance using a cryogenic GC. Temperature, humidity, and airflow were also recorded periodically during each exposure day. Exposures were conducted for 6 hours per day, 7 days per week.

Groups of 12 young, adult, male or nulliparous, non-pregnant female Crl:CD(SD) rats were exposed to atmospheres containing 0, 100, 500, or 3000ppm of Naphtha for 30- 31 days. Satellite females without evidence of mating during the 2-week period continued to be exposed for 26 days after the end of the cohabitation period.

Body weights, clinical signs, and food consumption were recorded throughout the study. Body weight data were collected weekly for males, subchronic females and satellite females without evidence of copulation. After approximately 30 days of exposure, blood samples were collected from all male and all subchronic female rats for measurement of haematology and clinical chemistry parameters. An abbreviated neurobehavioral evaluation was conducted on all males, subchronic females, and satellite females prior to test substance administration in order to obtain baseline measurements, and again during week 4 [test days 26-29] in the morning prior to daily exposure for males and subchronic females. Neurobehavioral evaluation consisted of motor activity and a modified Functional Observational Battery [FOB] of open field (approach and touch response, auditory response and tail pinch), papillary response, and fore and hind limb grip strength. Males and subchronic females were sacrificed after approximately 30 days of exposure, organs (liver, kidneys, lungs, adrenal glands, thymus, brain, spleen, heart, testes with epididymides, prostate, ovaries with oviducts, and uterus with cervix) were weighed, and 36 selected tissues were evaluated microscopically. Statistical analysis: Preliminary statistical analyses included Levene's test for homogeneity and Shapiro-Wilk test for normality, followed by one-way analysis of variance [ANOVA] and Dunnett/Tamhane-Dunnett's test or Kruskal-Wallis and Dunn's test as appropriate. Repeated measure ANOVA with Linear contrasts or Jonckheere-Terpstra trend test was used for motor activity and grip strength.

**Test Results - Repeated-Dose Toxicity**

**Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):**

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
LOAEC	Female	=	13423		mg/m3
NOAEC	Male	=	2366		mg/m3
NOAEC	Female	=	2366		mg/m3
LOAEC	Male	=	13423		mg/m3

**Results Remarks:**

Mortality did not occur at any exposure concentration. Test substance-related increases in the incidence of stained and wet fur in males and subchronic females were observed in the 3000ppm group; however, this did not adversely impact the health of the animals. In 3000ppm subchronic females, adverse, test substance-related, decreases in body weight, and weight gain (35% lower than controls) occurred. Slightly decreased body weight and/or weight gain occurred in 3000ppm males however, the magnitude of the effects was not statistically significant. No adverse effect

on body weight or weight gain were seen in any animals in the 500 or 100ppm groups

Summary of Body Weight, Weight Gain in Subchronic Rats exposed to Naphtha

Dose (ppm) Male Female

Day 29 BW (g) BW gain (g) Day 29 BW (g) BW gain(g)

Days 1-29 Days 1-29

0	433.7	132.7	269.5	51.1
100	422.5	127.0	263.7	46.9
500	413.6	120.0	266.7	48.7
3000	413.7	112.6	248.1	33.3*

Note: BW = body weight; BWG = body weight gain,

\*Statistically significant at  $p < 0.05$  by Dunnett/Tamhane-Dunnett

Decreases in food consumption correlated with decreased body weight and weight gains for animals in the 3000ppm group. Food efficiency was significantly reduced in subchronic 3000ppm females (0.057 vs 0.085 control). No test-substance related effects were seen on food consumption or efficiency in 100 or 500ppm groups or on food efficiency in 3000ppm males.

There were no adverse or test substance related effects on haematology or clinical chemistry parameters. Liver weight parameters were increased in 3000ppm males and females, which correlated with hepatocellular hypertrophy and were consistent with pharmacological induction of hepatic enzymes. Kidney weight parameters were increased in 500ppm and above males and in 3000ppm females. In males, the increased absolute/relative kidney weights correlated with hyaline droplet accumulation, which was observed in 100ppm and above males, indicative of light hydrocarbon nephropathy, a species and sex specific syndrome not relevant to humans (EPA, 1991). In 3000ppm subchronic females, the increased kidney weight parameters were not associated with any functional or microscopic change, and therefore were considered secondary to non-adverse enzyme induction. Minimal hypertrophy of thyroid follicular epithelium occurred in 3000ppm males and females, possibly secondary to the liver enzyme induction. The systemic toxicity LOAEL exclusive of kidney effects = 3000ppm (13650mg/m<sup>3</sup>) based on decreased body weight, weight gain and decreased food efficiency in females and on hypertrophy of thyroid follicular epithelium in 3000ppm males and females. Systemic NOAEL = 500ppm (2275mg/m<sup>3</sup>)

Neurobehavioral Toxicology: There were no test substance-attributed or statistically significant differences in forelimb or hindlimb grip strength in subchronic males and females at any concentration of the test substance. Pupillary constriction response and open field parameters consisting of approach and touch response, auditory response and tail pinch response were comparable for all treated groups and controls. Motor activity [duration of movement and number of movements] did not demonstrate any test substance related adverse effects. Males assigned to the 3000ppm group had statistically significantly lower motor activity compared to the control group mean at both the baseline pre-exposure evaluation and the week 4 evaluation, indicating that this effect was a function of the group composition and was not induced by high naphthenic naphtha vapor exposure. The NOAEL for neurobehavioral toxicity was 3000ppm (13650mg/m<sup>3</sup>), the highest concentration tested.

#### Conclusion:

Exposure to this heavy straight run naphtha at 3000ppm induced some systemic toxicity in male and female rats expressed as reduced body weight and weight gain, decreased food efficiency in females and hypertrophy of thyroid follicular epithelium in both sexes. Hydrocarbon nephropathy was seen in male rats at all doses but is a species and sex-specific syndrome not relevant to human risk assessment [U.S. Environmental Protection Agency. Alpha 2 microglobulin: association with chemically induced renal toxicity and neoplasia in the male rat. 1991. In Risk Assessment Forum. US Government Printing Office, Washington, DC: EPA: 85.] and thus was not included in determining the systemic NOAEL of 500ppm. This naphtha did not induce neurotoxic adverse effects and is not considered a neurobehavioral toxicant.

Male systemic (exclusive of kidney effects) NOAEC = 2366 mg/m<sup>3</sup>

Female systemic NOAEC = 2366 mg/m<sup>3</sup>

Male systemic (exclusive of kidney effects) LOAEC = 13423 mg/m<sup>3</sup>

Female systemic LOAEC = 13423 mg/m<sup>3</sup>

Male & female neurotoxicity NOAEL = 13423 mg/m3 (highest dose tested)

### Reliability/Data Quality - Repeated-Dose Toxicity

<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	RELIABILITY: GLP; guideline study  Key study  The reproductive/developmental toxicity segment of this study is described in a separate Robust Summary.
<b>Key Study Sponsor Indicator:</b>	Key

### Reference - Repeated-Dose Toxicity

<b>Reference:</b>	<p>Naphtha, Petroleum, Heavy Straight-run: Combined Repeated Dose Toxicity Study With the Reproduction/Developmental Toxicity Screening Test in Rats (OECD 422). 2008. DuPont Haskell Global Centers for Health and Environmental Sciences Project ID DuPont-18331. Newark, DE. Sponsored by Petroleum HPV Testing Group, API, Washington, DC.</p> <p>US EPA 1991. Alpha 2 microglobulin: Association of chemically induced renal toxicity and neoplasia in male rats. In Risk Assessment Forum, p.85. US Govt Printing Office, Washington DC</p>
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## Genetic Toxicity in vivo

### Test Substance - Genetic Toxicity in vivo

**Category Chemical:** No CAS Number Provided

**Test Substance:** No CAS Number Provided

**Test Substance Purity/Composition and Other Test Substance Comments:** Substance is in the Gasoline Blending Streams Category.  
See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:** Read-Across

### Method - Genetic Toxicity in vivo

**Type of Study:**

**Type of Test:**

**Route of Administration:**

**Type of Exposure:**

**Species:**

**Strain:**

**Gender:**

**Dose:**

**Year Study Performed:**

**Method/Guideline Followed:**

**GLP:**

**Duration of Treatment/ Exposure Period and Units:**

**Frequency of Treatment:**

**Positive, Negative, and Solvent Control Substance(s):**

**Number of Animals per Sex and Dose Group:**

**Method/Guideline and Test Condition Remarks:**

### Test Results - Genetic Toxicity in vivo

**Systemic Toxicity:**

**Genotoxic Effect:** Negative

**Results Remarks:** All PONA (Paraffinic, Olefinic, Naphthenic, and Aromatic) streams are negative for induction of chromosome aberrations in rats. One high olefinic sample induced sister chromatid exchanges in mice. Although the SCE assay demonstrated interaction of the LCCN (light catalytic cracked naphtha) sample and DNA, it was not considered definitive for clastogenic activity since no genetic material was unbalanced or lost, but rather a biomarker of

exposure. Negative results in two assays in rats, which monitor actual cytogenetic damage demonstrated that LCCN was not a clastogenic material. Gasoline did not induce cytogenetic damage in rats or adverse effects on spermatogenic cycle in mice. Although SCEs were induced in cultured peripheral blood from rats exposed to baseline gasoline vapor concentrate, the parallel micronucleus study was negative. Overall gasoline refinery blending streams are not clastogenic.

**Conclusion:** The read-across conclusion for untested streams in this category is negative for in vivo genetic toxicity.

### Reliability/Data Quality - Genetic Toxicity in vivo

**Reliability:**

**Reliability Remarks:**

**Key Study Sponsor Indicator:** Weight of Evidence

### Reference - Genetic Toxicity in vivo

**Reference:** See records for CAS No. 64741-66-8, 64741-55-5, 64741-63-5, 64741-87-3, 68955-35-1, and gasoline (86290-81-5).



<b>Genetic Toxicity in vivo</b>	
<b>Test Substance - Genetic Toxicity in vivo</b>	
<b>Category Chemical:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	API 83-05, Catalytic Reformed Naphtha. Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at website below)  Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Genetic Toxicity in vivo</b>	
<b>Type of Study:</b>	Bone Marrow Chromosomal Aberration
<b>Type of Test:</b>	Chromosome aberration assay
<b>Route of Administration:</b>	Intraperitoneal
<b>Type of Exposure:</b>	
<b>Species:</b>	Rat
<b>Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Dose:</b>	0, 0.26, 0.82 & 2.42 g/kg
<b>Year Study Performed:</b>	1985
<b>Method/Guideline Followed:</b>	
<b>GLP:</b>	Yes
<b>Duration of Treatment/Exposure Period and Units:</b>	
<b>Frequency of Treatment:</b>	once
<b>Positive, Negative, and Solvent Control Substance (s):</b>	Positive: Triethylenemelamine at a dose of 0.8 mg/kg in corn oil. Corn oil was used as the solvent control.
<b>Number of Animals per Sex and Dose Group:</b>	5/sex/dose/sacrifice times
<b>Method/Guideline and Test Condition Remarks:</b>	Two studies were carried out. In the first study, the test material did not induce a significant increase in the percentage of aberrant cells above the controls in either sex. Furthermore, the positive control (TEM at a dose of 0.8 mg/kg) did not induce a significant elevation in the percentage of cells with structural aberrations. Consequently, the first study was considered 'invalid' because the positive control did not induce chromosomal

damage. The assay was, therefore, repeated using a higher dose of TEM. In this robust summary, only the results of the repeat study are described. The study design was as follows:

Treatment Animals/sex/sacrifice time  
 6 hrs. 24 hrs. 48 hrs  
 Corn oil (vehicle) 5 5 5  
 API 83-05, 2.42 g/kg 5 5 5  
 API 83-05, 0.82 g/kg 5 5 5  
 API 83-05, 0.26 g/kg 5 5 5  
 Triethylenemelamine 5  
 (Positive control)

Test material in vehicle was given intraperitoneally at a dose of 5 ml/kg to groups of rats as shown above. Corn oil was used as vehicle control and TEM (1.5 mg/kg) as the positive control. Four hours prior to sacrifice the rats were given a single intraperitoneal dose of colchicine (4 mg/kg). One male in the 2.42 g/kg group and one male in the 0.82 g/kg dose group died immediately after dosing, these were replaced by substitute animals.

Immediately after sacrifice, bone marrow was obtained from the tibiae of the animals. The marrow was washed and the cells were fixed before being spread on slides (at least 3 from each animal) for examination. Slides were scored for chromosomal aberrations. Where possible, a minimum of 50 metaphase cells from each animal were examined and scored for chromatid and chromosome gaps and breaks, fragments, structural rearrangements and ploidy (1-3). A mitotic index (= No. of cells in mitosis/500 counted x 100) was calculated and recorded.

The type of aberration, its frequency, the statistical significance of any increases and its correlation to dose in a given time period will all be considered in evaluating a test article as being mutagenically positive or negative.

Criteria for a positive response are generally a statistically significant dose-related increase in the number of structural aberrations at three dose levels. The final decision is based on scientific judgment.

Similar cytogenetics assays have been reported for two other aromatic naphtha samples (API 83-04 and API 83-06, approx. 42 and 90% aromatics respectively) and both were negative.

### Test Results - Genetic Toxicity in vivo

**Systemic Toxicity:** No Effects

**Genotoxic Effect:** Negative

**Results Remarks:** The dose levels used in the assay were selected on the basis of a preliminary screen. In the cytogenetics assay, one male died at each of the dose levels 2.42 and 0.82 g/kg, the mortality occurred immediately after dosing. Toxic signs included lethargy and a moribund appearance at the high dose and slow uncoordinated movement in the mid dose group. The results of the cytogenetics evaluations are summarized in the following table.

MALES

Dose level (g/kg)	Positive	Vehicle		
0.26	0.82	2.42	control	control
% Cells with 1 or more aberrations				
6 hrs	0.5	0.4	1.0	0.5
24 hrs	0.4	0.8	1.0	32.4
48 hrs	0	1.6	0.5	0.8
% Cells with 2 or more aberrations				
6 hrs	0	0	0	0.5
24 hrs	0	0	0	10.8
48 hrs	0	0	0	0
Frequency of structural aberrations				
6 hrs	.005	.004	.01	.05
24 hrs	.004	.008	.01	.708
48 hrs	0	.016	.005	.008
Frequency of numerical aberrations				
6 hrs	.005	0	.016	.015
24 hrs	.008	.008	.01	.008
			.005	

48 hrs .01 .008 0 .004

FEMALES

Cells with 1 or more aberrations

6 hrs 0 0.5 1.6 0.8

24 hrs 0 0.4 1.5 33.2 0

48 hrs 0 1.2 0.8 1.2

Cells with 2 or more aberrations

6 hrs 0 0 0 0

24 hrs 0 0 0.5 13.20 0

48 hrs 0 0 0 0.5

Frequency of structural aberrations\*

6 hrs 0 .005 .016 0.008

24 hrs 0 .004 .02 0.804 0

48 hrs 0 .012 .008 0.012

Frequency of numerical aberrations\*

6 hrs .005 .005 .020 0

24 hrs .01 .016 .005 0.020 0.020

48 hrs .008 0 .012 0.005

Mitotic Index

6 hrs 5.4 6.3 3.1 6.1

24 hrs 4.9 5.4 4.1 4.7 4.8

48 hrs 5.5 4.9 7.0 5.2

\* Frequency based upon the aberration frequency per cell per animal

Note that for simplicity only, mean values without standard errors are shown in the above table although they are given in the laboratory report.

On the basis of the criteria defined for assessing the results, the authors concluded that API 83-05 was not mutagenic in this assay.

**Conclusion:**

Catalytic reformed naphtha did not cause bone marrow chromosomal aberrations at i.p. dosed up to 2.42 g/kg, which was the highest dose tested.

**Reliability/Data Quality - Genetic Toxicity in vivo**

**Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:**

Key

**Reference - Genetic Toxicity in vivo**

**Reference:**

American Petroleum Institute (1985) Mutagenicity evaluation of 83-05 in the rat bone marrow cytogenetic assay. Study conducted by Litton Bionetics, Inc. API Mes. Res. Publ. 32-32289, June 1985.

American Petroleum Institute (1985) Activity of API 83-06 (heavy catalytic reformed naphtha) in the acute in-vivo cytogenetics assay in male and female rats API HESD Report No. 33-30494

American Petroleum Institute (1986) Mutagenicity evaluation in the rat bone marrow cytogenetic assay. API 83-04, light catalytically cracked reformed naphtha (CAS 64741-63-5) API HESD Report No. 33-31092

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



## Genetic Toxicity in vivo

### Test Substance - Genetic Toxicity in vivo

**Category Chemical:** (86290-81-5) Antiknock Gasoline

**Test Substance:** (86290-81-5) Antiknock Gasoline

**Test Substance Purity/Composition and Other Test Substance Comments:** Substance is in the Gasoline Blending Streams Category.  
See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

### Method - Genetic Toxicity in vivo

**Type of Study:** Bone Marrow Chromosomal Aberration

**Type of Test:** Dominant lethal assay

**Route of Administration:** Inhalation

**Type of Exposure:** Vapor

**Species:** Mouse

**Strain:** CD-1

**Gender:** Both M/F

**Dose:** 0, 400 & 1600 ppm; approximately 1, 1500, 6000 mg/m<sup>3</sup>

**Year Study Performed:** 1980

**Method/Guideline Followed:** Unknown

**GLP:** Yes

**Duration of Treatment/Exposure Period and Units:** 8 Weeks

**Frequency of Treatment:** 6 hours/day, 5 days/week

**Positive, Negative, and Solvent Control Substance(s):** Positive control (10 male rats) was a single i.p. injection of triethylenemelamine at 0.3 mg/kg in saline. Negative control (10 male rats) was filtered air.

**Number of Animals per Sex and Dose Group:** 10 males per group; females were untreated.

**Method/Guideline and Test Condition Remarks:** Groups of 10 male mice were exposed to either filtered air (negative controls) or test material at concentrations of 400 or 1600 ppm. Generation of test atmospheres was accomplished by bubbling air through the test material. Exposures were for 6 hours a day, 5 days each week for 8 weeks. On the final day of exposure a positive control group of 10 male mice were given Triethylenemelamine (TEM) intraperitoneally as a single i.p. dose, at a dose level of 0.3 mg/kg. The dose volume was 0.1 ml/mouse and the TEM was dissolved in 0.9% saline. Chamber concentrations were monitored at least hourly during the exposure

periods.  
 After 2 days rest following termination of exposures, each male was caged with 2 unexposed virgin female mice. At the end of 5 days, the females were removed. This weekly mating sequence was continued for 2 weeks. Each pair of mated females were transferred to a fresh cage and after 14 days after the midweek of being caged with the male were sacrificed. The uterine contents of the females were examined and scored for the numbers of dead and living implants and total implants.

**Evaluation Criteria:**

Dominant lethality was determined from a) a mutation index derived from the ratio of total to dead implants; or b) the number of dead implants per pregnant female.

If true dominant lethality is observed then a significant increase in the number of dead implants per pregnant female should be accompanied by a significant decrease in the number of living implants per pregnant female.

The two ratios are compared with both concurrent and comparable historical control values. Dose-related trends are also looked for. Any statistically significant differences must also be strongly evaluated for their biological significance.

In this study the following parameters were determined:

Fertility index ie. Proportion of pregnant females.  
 Average No of implants/pregnant female.  
 Average No. of dead implants/pregnant female.  
 Proportion of females with one or more dead implants.  
 Proportion of females with two or more dead implants.

### Test Results - Genetic Toxicity in vivo

**Systemic Toxicity:** No Effects

**Genotoxic Effect:** Negative

**Results Remarks:**

During the exposure phase actual chamber concentrations were found to be 0, 396.4 and 1524.6 ppm. One male died in the 1600 ppm group and another animal in the same group exhibited excessive lacrimation in the seventh week but this cleared in the final week.

The data for each of the parameters determined are as follows for untreated control, historical control, positive control and the two groups exposed to test material.

Week	Hist.-ve	-ve	+ve	400ppm	1600ppm
Fertility index					
1	22/24	21/23	19/24	17/20	21/22
2	16/24	19/24	13/24	18/19	16/22
Av. No. of implants/pregnant female					
1	267/22	240/21	140/19	203/17	214/21
2	193/16	220/19	91/13	219/18	183/16
Av. No. of dead implants/pregnant female					
1	12/22	14/21	83/19	9/17	9/21
2	13/16	5/19	66/13	9/18	12/16
Proportion of females with one or more dead implants					
1	11/22	9/21	19/19	6/17	8/21
2	9/16	4/19	13/13	8/18	7/16

Proportion of females with two or more dead implants					
1	1/22	3/21	17/19	3/17	1/21
2	2/16	1/19	13/13	1/18	3/16
No of dead implants/total implants					
1	12/267	14/240	83/140	9/203	9/214
2	13/193	5/220	66/91	9/219	12/183

Interpretation of the results: The test material did not cause any significant reduction in the fertility index. The test material had no effect on the average number of implants per pregnant female. With respect to the number of dead implants per pregnant female, the test material showed no significant differences from the values of the concurrent as well as the negative controls.

The results support the conclusion that the test material did not cause increases in post-implantation deaths.

**Conclusion:** PS-6 unleaded gasoline was not genotoxic in an in vivo dominant lethal assay in CD-1 mice.

#### Reliability/Data Quality - Genetic Toxicity in vivo

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:** Key

#### Reference - Genetic Toxicity in vivo

**Reference:** American Petroleum Institute (1980) Mutagenicity evaluation of Gasoline, API PS-6 fuel in the mouse dominant lethal assay. Study conducted by Litton Bionetics Inc. API Publication No. 28-31344. April 1980

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003





## Genetic Toxicity in vivo

### Test Substance - Genetic Toxicity in vivo

**Category Chemical:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance Purity/Composition and Other Test Substance Comments:** Sample API 83-19 is a Light Alkylate Naphtha (LAN). Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at website below)

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:** Measured

### Method - Genetic Toxicity in vivo

**Type of Study:** Bone Marrow Chromosomal Aberration

**Type of Test:** Chromosome aberration assay

**Route of Administration:** Intraperitoneal

**Type of Exposure:**

**Species:** Rat

**Strain:** Sprague-Dawley

**Gender:** Both M/F

**Dose:** 0, 0.3, 1.0 & 3.0 g/kg

**Year Study Performed:** 1985

**Method/Guideline Followed:** Other

**GLP:** Yes

**Duration of Treatment/Exposure Period and Units:** 48 Hours

**Frequency of Treatment:** single treatment

**Positive, Negative, and Solvent Control Substance (s):** Corn oil was used as vehicle control and 0.5 mg/kg TEM (Triethylenemelamine) as the positive control

**Number of Animals per Sex and Dose Group:** 6 rats/sex/dose and sacrifice time

**Method/Guideline and Test Condition Remarks:** Type: Cytogenetic assay  
The study design was as follows:  
Treatment Animals/sex/sacrifice time  
6 hrs. 24 hrs. 48 hrs  
Corn oil (vehicle) 5 5 5  
API 83-19, 3 g/kg 5 5 5  
API 83-19, 1 g/kg 5 5 5

API 83-19, 0.3 g/kg 5 5 5  
 Triethylenemelamine 5  
 (Positive control)

Test material in vehicle was given intraperitoneally at a dose of 5 ml/kg to groups of rats as shown above. Corn oil was used as vehicle control and TEM (0.5 mg/kg) as the positive control. Two to four hours prior to sacrifice the rats were given a single intraperitoneal dose of colchicine (1 mg/kg). 2 Males and one female in the high dose group died, these were replaced by substitute animals that were killed approximately 50 hours after administration of the test

material. Immediately after sacrifice bone marrow was obtained from the femurs of the animals. The marrow was washed and the cells were fixed before being spread on slides (at least 3 from each animal) for examination. Slides were scored for chromosomal aberrations. Where possible, a minimum of 50 metaphase cells from each animal were examined and scored for chromatid and chromosome gaps and breaks, fragments, structural rearrangements and ploidy (1-3). A mitotic index (= No. of cells in mitosis/500 counted X 100) was calculated and recorded.

The data were evaluated according to the following criteria:

For the test to be considered to be valid, the % of cells in the negative control group demonstrating aberrations of any type, other than gaps, must not exceed 4%. The % of cells with aberrations in the positive control group must be statistically increased ( $p=0.05$ ) relative to the vehicle control using Chi-square statistics.

The test material is considered positive when the % of cells with aberrations in any treatment group is significantly increased ( $p = 0.05$ ) relative to the vehicle control using Chi-square analysis and the number of aberrations per cell is also significantly increased ( $p = 0.05$ ) relative to the vehicle control using t-test statistics.

### Test Results - Genetic Toxicity in vivo

**Systemic Toxicity:** No Effects

**Genotoxic Effect:** Negative

**Results Remarks:**

The dose levels used in the assay were selected on the basis of a preliminary screen in which only one male rat died within 24 hours following the administration of API 83-19 as a single i.p. dose to 4 rats of each sex. In the cytogenetics assay, 5 of 18 males and 4 of 18 females receiving 3 g/kg API 83-19 died within 3 days. At this dose level, there was a weight loss of 10% and 9% in males and females respectively within 48 hours of administration. Other signs of toxicity included piloerection, crusty eyes and noses and excess lacrimation. No sex-related differences were noted in the study and therefore the data for males and females were combined for the cytogenetics evaluation. The results are summarized in the following table.

	0.3 g/kg	1 g/kg	3 g/kg	Positive	Vehicle
Cells with aberrations					
6 hrs	0	2	0	0	
24 hrs	1	0	1	171	0
48 hrs	1	0	1	0	
Incidence of aberrations (%)					
6 hrs	0	0.4	0	0	
24 hrs	0.2	0	0.2	34.2	0
48 hrs	0.2	0	0.3	0	
No. Gaps					
6 hrs	0	2	0	0	
24 hrs	0	0	0	15	1
48 hrs	0	0	4	1	
No. Breaks					
6 hrs	0	2	0	0	
24 hrs	1	0	1	197	0
48 hrs	2	0	1	0	
Aberrations per cell					

6 hrs 0 0.004 0 0  
 24 hrs 0.002 0 0.002 2.336 0  
 48 hrs 0.004 0 0.003 0

NB.1. 500 cells were evaluated for each time point at each dose level.

NB.2. In the API 83-19 and vehicle control groups no rearrangements were observed and no aberrations from severely damaged cells were seen. In contrast 51 rearrangements and 920 aberrations from severely damaged cells were seen in the positive control group.

**Conclusion:** Light Alkylate Naphtha did not induce bone marrow chromosomal aberrations in male or female Sprague-Dawley rats.

#### Reliability/Data Quality - Genetic Toxicity in vivo

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:** Key

#### Reference - Genetic Toxicity in vivo

**Reference:** American Petroleum Institute (1985) Acute In Vivo cytogenetics assay in male and female rats of API sample 83-19. Study conducted by Microbiological Associate Inc. API Health & Environmental Sciences Department, Publication No. 32-32409

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<b>Genetic Toxicity in vivo</b>	
<b>Test Substance - Genetic Toxicity in vivo</b>	
<b>Category Chemical:</b>	(64741-87-3) Naphtha, petroleum, sweetened
<b>Test Substance:</b>	(64741-87-3) Naphtha, petroleum, sweetened
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	API 81-08, Sweetened Naphtha. Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at website below)  Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Genetic Toxicity in vivo</b>	
<b>Type of Study:</b>	Bone Marrow Chromosomal Aberration
<b>Type of Test:</b>	Chromosome aberration assay
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Dose:</b>	0, 65, 300 & 2050 ppm, nominal concentrations; measured average concentrations were 0, 69, 293, and 2012 ppm [approximately 0, 150, 725, 5000 mg/m <sup>3</sup> ]
<b>Year Study Performed:</b>	1986
<b>Method/Guideline Followed:</b>	
<b>GLP:</b>	Yes
<b>Duration of Treatment/Exposure Period and Units:</b>	5 Days
<b>Frequency of Treatment:</b>	6 hr/day
<b>Positive, Negative, and Solvent Control Substance (s):</b>	A positive control group of 10 rats of each sex were given a single dose (0.8 mg/kg) of TEM intraperitoneally 24 hours before sacrifice. A negative control group of 10 rats of each sex were exposed to air only.
<b>Number of Animals per Sex and Dose Group:</b>	10
<b>Method/Guideline and Test Condition Remarks:</b>	Groups of 10 male and 10 female Sprague Dawley rats were exposed (whole body) to nominal concentrations of 65, 300 and 2050 ppm of test material. Animals were exposed to vapor of the test material 6 hours each day for 5 consecutive days. A positive control group of 10 rats of each sex were given a single dose (0.8 mg/kg) of TEM

intraperitoneally 24 hours before sacrifice. A negative control group of 10 rats of each sex were exposed to air only. For the treated and negative control groups bone marrow was harvested 6 hours after the final exposure. For the positive control group the bone marrow was harvested 24 hours after administration of the TEM.

Three hours prior to sacrifice by carbon monoxide the rats were given a single intraperitoneal dose of colchicine (4 mg/kg). Immediately after sacrifice bone marrow was obtained from the tibiae of the animals. The marrow was washed and the cells were fixed before being spread on slides for examination. Routinely 50 spreads were prepared for each animal. The location of cells bearing aberrations were identified. A mitotic index based on at least 500 cells counted was also recorded. It was calculated by scoring the number of cells in mitosis per 500 cells on each slide read. Slides were scored for chromosomal aberrations.

The authors give the following as the criteria for a positive response and data interpretation. Gaps were not counted as significant aberrations. Indicators of genetic damage were considered to be: Open breaks, configurations resulting from the repair of breaks. The latter included translocations, multiradials, rings, multicentrics etc. Reunion figures such as these were weighted slightly higher than breaks since they usually resulted from more than one break. The number of cells with aberrations per animal was also considered to indicate more genetic damage than those containing evidence of single events. Consistent variations from the euploid number were also considered in the evaluation of mutagenic potential. Often it is not possible to locate 50 suitable metaphase spreads for each animal, even after preparing additional spreads. Possible causes for this appear to be related to cytotoxic effects which alter the duration of the cell cycle, kill the cell or cause clumping of the chromosomes.

Additional information can be gained from the mitotic index which also appears to reflect cytotoxic effects. The type of aberration, its frequency and its correlation to dose in a given time period was considered in evaluating a test article as being mutagenically positive or negative. Statistical analysis employed a Kruskal-Wallis test of aberrations per cell on a per animal basis.

Vapor of the test material was generated by bubbling nitrogen through heated distillation columns packed with glass beads. The test material was delivered to the top of the glass beads using syringe pumps, a different delivery rate being used for each target dose level. Chamber concentrations were monitored hourly each exposure period. Results of chamber monitoring are:

Target (ppm)	Actual (ppm)
0	0
65	69 ± 18
300	293 ± 42
2050	2012 ± 16

### Test Results - Genetic Toxicity in vivo

**Systemic Toxicity:** No Effects

**Genotoxic Effect:** Negative

**Results Remarks:** The mean exposure chamber concentrations were found to be: 0, 69±18, 293±42 and 2012±16 ppm.

No signs of toxicity were observed in the rats during the exposure phase of the study. The results of the cytogenetic evaluation are summarized in the following table. NB. Mean values without standard errors are given in the table, although these data are available in the report.

Exposure concentration (ppm)	Control
69	293
2012	Positive
	Negative
Total No. of cells	

Male 470 500 410 400 500  
 Female 500 500 500 474 500  
 M+F 970 1000 910 874 1000

Frequency of structural aberrations  
 Male .009 .006 .029 >.708 .016  
 Female 0 >.014 .030 >.970 .008  
 M+F .005 >0.01 .029 >.853 .012

Frequency of numerical aberrations  
 Male .012 0 .013 .023 .01  
 Female .012 .016 .006 .015 .008  
 M+F .01 .008 .01 .019 .009

% Cells with structural aberrations/animal  
 1 or more  
 Male .9 .4 2.2 20 .6  
 Female 0 1.4 2.6 19 .8  
 M+F .5 .9 2.4 19.5 .7

2 or more  
 Male 0 .2 .4 11.3 .4  
 Female 0 .2 .4 14.4 0  
 M+F 0 .2 .4 12.9 .2

%MI  
 Male 6.5 6.6 3.8 1 5.7  
 Female 4.1 4.8 4.5 1 4.2  
 M+F 5.3 5.7 4.2 12.9 .2

On the basis of the above data, the authors concluded that there was no evidence of a clastogenic effect of the test material and that there was no significant increase in chromosomal aberration in the dosed animals when compared to the negative controls.

**Conclusion:** Sweetened naphtha at inhalation exposure concentrations up to 2012 ppm was not clastogenic in adult male or female Sprague Dawley rats under these test conditions.

#### Reliability/Data Quality - Genetic Toxicity in vivo

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:** Key

#### Reference - Genetic Toxicity in vivo

**Reference:** American Petroleum Institute (1986) Mutagenicity evaluation in the rat bone marrow cytogenetic assay, API 81-08, Sweetened naphtha (CAS 64741-87-3) Study conducted by Litton Bionetics Inc. API, HESD Research Publication 33-31093, April 1986.

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<b>Genetic Toxicity in vivo</b>	
<b>Test Substance - Genetic Toxicity in vivo</b>	
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	API 81-03, Sweetened Naphtha. Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at website below)  Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Genetic Toxicity in vivo</b>	
<b>Type of Study:</b>	Bone Marrow Chromosomal Aberration
<b>Type of Test:</b>	Chromosome aberration assay
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Dose:</b>	0, 65, 300 & 2050 ppm, nominal concentrations; measured average concentrations were 0, 63, 297, and 2046 ppm [approximately 0, 150, 725, 5000 mg/m <sup>3</sup> ]
<b>Year Study Performed:</b>	1983
<b>Method/Guideline Followed:</b>	
<b>GLP:</b>	Yes
<b>Duration of Treatment/Exposure Period and Units:</b>	5 Days
<b>Frequency of Treatment:</b>	6 hr/day
<b>Positive, Negative, and Solvent Control Substance (s):</b>	A positive control group of 10 male and 8 female rats of each sex were given a single dose (0.8 mg/kg) of TEM intraperitoneally 24 hours before sacrifice. A negative control group of 10 rats of each sex were exposed to air only.
<b>Number of Animals per Sex and Dose Group:</b>	10
<b>Method/Guideline and Test Condition Remarks:</b>	Groups of 10 male and 10 female Sprague Dawley rats were exposed (whole body) to nominal concentrations of 65, 300 and 2050 ppm of test material. Animals were exposed to vapor of the test material 6 hours each day for 5 consecutive days. A positive control group of 10 rats of each sex were given a single dose (0.8 mg/kg) of TEM

intraperitoneally 24 hours before sacrifice. A negative control group of 10 rats of each sex were exposed to air only. For the treated and negative control groups bone marrow was harvested 6 hours after the final exposure. For the positive control group the bone marrow was harvested 24 hours after administration of the TEM.

Four hours prior to sacrifice by carbon monoxide the rats were given a single intraperitoneal dose of colchicine (4 mg/kg). Immediately after sacrifice bone marrow was obtained from the tibiae of the animals. The marrow was washed and the cells were fixed before being spread on slides for examination. Routinely 50 spreads were prepared for each animal. The location of cells bearing aberrations were identified. A mitotic index based on at least 500 cells counted was also recorded. It was calculated by scoring the number of cells in mitosis per 500 cells on each slide read. Slides were scored for chromosomal aberrations.

The authors give the following as the criteria for a positive response and data interpretation. Gaps were not counted as significant aberrations. Indicators of genetic damage were considered to be: Open breaks, configurations resulting from the repair of breaks. The latter included translocations, multiradials, rings, multicentrics etc. Reunion figures such as these were weighted slightly higher than breaks since they usually resulted from more than one break. The number of cells with aberrations per animal was also considered to indicate more genetic damage than those containing evidence of single events. Consistent variations from the euploid number were also considered in the evaluation of mutagenic potential. Often it is not possible to locate 50 suitable metaphase spreads for each animal, even after preparing additional spreads. Possible causes for this appear to be related to cytotoxic effects which alter the duration of the cell cycle, kill the cell or cause clumping of the chromosomes.

Additional information can be gained from the mitotic index which also appears to reflect cytotoxic effects. The type of aberration, its frequency and its correlation to dose in a given time period was considered in evaluating a test article as being mutagenically positive or negative. Statistical analysis employed Student t-test, Wilcoxin rank sum test, and Kruskal-Wallis test.

Vapor of the test material was generated by bubbling nitrogen through heated distillation columns packed with glass beads. The test material was delivered to the top of the glass beads using syringe pumps, a different delivery rate being used for each target dose level. Chamber concentrations were monitored hourly each exposure period. Results of chamber monitoring are:

Target	Actual
(ppm)	(ppm)
0	0
65	63 ± 14
300	297 ± 12
2050	2046 ± 29

### Test Results - Genetic Toxicity in vivo

**Systemic Toxicity:** No Effects

**Genotoxic Effect:** Negative

**Results Remarks:** The mean exposure chamber concentrations were found to be: 0, 63, 297, and 2046 ppm [approximately 0, 150, 725, 5000 mg/m<sup>3</sup>].

No signs of toxicity were observed in the rats during the exposure phase of the study. The results of the cytogenetic evaluation are summarized in the following table. NB. Mean values without standard errors are given in the table, although these data are available in the report.

Exposure concentration (ppm)	Control
63 297 2046	Positive Negative



Total No. of cells  
 Male 421 463 500 442 500  
 Female 413 500 450 338 500  
 M+F 834 963 950 780 1000

Frequency of structural aberrations  
 Male .000 .009 .002 >.434 .006  
 Female .000 .006 .002 >.268 .000  
 M+F .000 .007 .002 >.357 .003

Frequency of numerical aberrations  
 Male .033 .017 .008 .018 .014  
 Female .027 .020 .020 .044 .024  
 M+F .030 .019 .014 .030 .019

% Cells with structural aberrations/animal  
 1 or more  
 Male .0 .9 .2 13.6 .6  
 Female .0 .4 .2 11.3 .0  
 M+F .0 .6 .2 12.2 .3

2 or more  
 Male .0 .0 .0 6.6 .4  
 Female .0 .2 .0 4.1 .0  
 M+F .0 .1 .0 5.4 .2

%MI  
 Male 5.7 3.9 5.2 1.4 5.0  
 Female 4.9 3.3 5.8 1.6 5.0  
 M+F 5.3 3.6 5.4 1.5 5.0

On the basis of the above data, the authors concluded that there was no evidence of a clastogenic effect of the test material and that there was no significant increase in chromosomal aberration in the dosed animals when compared to the negative controls.

**Conclusion:** Sweetened naphtha at inhalation exposure concentrations up to 2046 ppm (~ 5000 mg/m3) was not clastogenic in adult male or female Sprague Dawley rats under these test conditions.

### Reliability/Data Quality - Genetic Toxicity in vivo

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:** Key

### Reference - Genetic Toxicity in vivo

**Reference:** American Petroleum Institute (1985) Mutagenicity evaluation studies in the rat bone marrow cytogenetic assay in the mouse lymphoma forward mutation assay Light catalytic cracked naphtha API sample 81-03 API Med. Res. Pub. 32-31300

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## Genetic Toxicity in vitro

### Test Substance - Genetic Toxicity in vitro

**Category Chemical:** No CAS Number Provided

**Test Substance:** No CAS Number Provided

**Test Substance Purity/Composition and Other Test Substance Comments:** Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:** Read-Across

### Method - Genetic Toxicity in vitro

**Type of Study:**

**Concentrations:**

**Year Study Performed:**

**Method/Guideline Followed:**

**GLP:**

**Positive, Negative, and Solvent Control Substance (s):**

**Method/Guideline and Test Condition Remarks:**

### Test Results - Genetic Toxicity in vitro

**Details on Cytogenetic Assay:**

**Statistics:**

**Effect:**

Species	Other Species	Strain	Other Strain	Metabolic Activation	Genotoxic Effect	Conclusion
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**Results Remarks:** Results from representative samples from each of the PONA (Paraffinic, Olefinic, Naphthenic, or Aromatic Hydrocarbons) categories indicate that most gasoline blending streams are not mutagenic in mammalian cells except for those substances with fairly high aromatic content where equivocal or in one case positive activity was seen with metabolic activation. Gasoline tested in both bacterial and mammalian cell assays did not induce mutation in either test system.

**Conclusion:** The read-across conclusion is that all streams in this category are negative with and without metabolic activation with the exception of streams with aromatic content greater than 60% that can be classified as negative/equivocal without metabolic activation and equivocal/positive with metabolic activation.

<b>Reliability/Data Quality - Genetic Toxicity in vitro</b>	
<b>Reliability:</b>	
<b>Reliability Remarks:</b>	
<b>Key Study Sponsor Indicator:</b>	Weight of Evidence
<b>Reference - Genetic Toxicity in vitro</b>	
<b>Reference:</b>	See Robust Study Summaries for CAS # 64741-55-5, 64741-63-5, 64741-66-8, 6474168-0, 64741-87-3, 68955-35-1, and 86290-81-5



<b>Genetic Toxicity in vitro</b>	
<b>Test Substance - Genetic Toxicity in vitro</b>	
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Genetic Toxicity in vitro</b>	
<b>Type of Study:</b>	Mammalian cell gene mutation assay
<b>Concentrations:</b>	0.065 to 1.04 ul/ml
<b>Year Study Performed:</b>	1977
<b>Method/Guideline Followed:</b>	
<b>GLP:</b>	Yes
<b>Positive, Negative, and Solvent Control Substance (s):</b>	The test material was dissolved in acetone for this assay. The positive control substances were Ethyl methane sulphonate (EMS) and Dimethylnitrosamine (DMN).
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Type: Mouse lymphoma assay System of testing: Forward mutation assay using cell line L5178Y TK+/-</p> <p>A cytotoxicity study was carried out prior to the mutagenicity assay. For the mutation assay the lymphoma cells were exposed for 5 hours to test material at concentrations ranging from 0.065 to 1.04 ul/ml for both the activation and non-activation assays. Metabolic activation was accomplished using Araclor-induced rat liver S-9 suspension. After exposure to the test material, the cells were allowed to recover for 3 days and then cultures were selected for cloning and mutant selection. Surviving cell populations were determined by plating diluted aliquots in non-selective growth medium.</p> <p>A mutation index was derived by dividing the number of clones formed in the BUDR-containing selection medium by the number found in the same medium without BUDR. The ratio was then compared to that obtained from other dose levels and from positive and negative controls.</p> <p>A compound is considered mutagenic if: A dose response relationship is observed over 3 of the 4 dose levels employed. The minimum increase at the high level of the dose response curve is at least 2.5 times greater than the solvent control value. The solvent control data are within the normal range of the spontaneous</p>

background for the TK locus.

### Test Results - Genetic Toxicity in vitro

#### Details on Cytogenetic Assay:

#### Statistics:

#### Effect:

Species	Other Species	Strain	Other Strain	Metabolic Activation	Genotoxic Effect	Conclusion
Mammalian Cell Line		Mouse Lymphoma L5178Y Cells	-	With and Without	Negative	Negative

#### Results Remarks:

Little toxicity was observed with the test material. Positive control values exhibited significant responses over the negative controls, and the negative controls were within the normal range.

All results for the test material from the non-activation assay were negative. The results from the activation assay were also considered to be negative. There was an increase in the number of mutants at the 0.52 ul/ml concentration but this appeared to result from a slight increase in the number of viable clones. There was no trend indicating a dose-related response and, therefore, the increases were not believed to be compound related. The results are summarized below.

Dose (ul/ml)	Rel. susp. growth	Mutant clones	Viable clones	%Rel. growth	Mutant frequency
Non-activation					
0.065	121.8	76	159	139.3	0.478
0.13	103.7	29	215	160.4	0.1349
0.26	114.6	44	211	174	0.2085
0.52	141.8	66	161	164.3	0.4099
1.04	107.5	58	270	208.9	0.2148
Solvent	100	14	139	100	0.1007
Negative	129.9	41	140	130.8	0.2929
EMS	58.7	227	67	28.3	3.3881
Activation					
0.065	120.6	66	87	79.5	0.7586
0.13	108.6	46	126	103.7	0.3651
0.26	106	70	130	104.4	0.5385
0.52	112.4	92	108	92	0.8519
1.04	68.9	21	193	100.8	0.1088
Solvent	100	30	132	100	0.2273
Negative	92.1	41	150	104.7	0.2733
DMN	16.7	91	7	0.9	13

#### Conclusion:

Unleaded gasoline was negative with or without metabolic activation in the mouse lymphoma forward mutation assay.

### Reliability/Data Quality - Genetic Toxicity in vitro

#### Reliability:

Valid Without Restrictions

#### Reliability Remarks:

RELIABILITY: GLP study with adequately detailed methods description

#### Key Study Sponsor Indicator:

Key

### Reference - Genetic Toxicity in vitro

#### Reference:

American Petroleum Institute (1977) Mutagenicity evaluation of unleaded gasoline  
Study conducted by Litton Bionetics, Inc. API HESD Publication No. 28-30173,

March 1977

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data have been  
entered into the HPVIS: 10/28/2003



<b>Genetic Toxicity in vitro</b>	
<b>Test Substance - Genetic Toxicity in vitro</b>	
<b>Category Chemical:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Test material was a catalytically reformed naphtha, CAS # 68955-35-1</p> <p>Test material designation by study sponsor was API # 83-05. Compositional information on this test material can be found in the analytical data report attached to the Gasoline Blending Stream Category - see website.</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Genetic Toxicity in vitro</b>	
<b>Type of Study:</b>	Mammalian cell gene mutation assay
<b>Concentrations:</b>	The non-activated cultures that were cloned were treated with 6.25, 25, 37.5 50, 75 and 100 nl/ml of test material. The activated cultures that were cloned were treated with 18.8, 37.5, 75, 100, 150 and 200 nl/ml of test material.
<b>Year Study Performed:</b>	1985
<b>Method/Guideline Followed:</b>	
<b>GLP:</b>	Yes
<b>Positive, Negative, and Solvent Control Substance (s):</b>	<p>Three positive control substances were used viz Ethyl methane sulphonate (EMS) at concentrations of 0.25 &amp; 0.4 µl/ml for the non activation assay, Dimethylnitrosamine (DMN) at a concentration of 0.3 and Methylcholanthrene (MCA) at a concentration of 2.5 µg/ml for the activation assay.</p> <p>Three negative controls were used: 2 solvent (ethanol) controls (not to exceed 1% of of growth medium) and one untreated control. For test substances assayed with activation, the solvent controls included the activation mixture.</p>
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Type: Mouse lymphoma assay System of testing: Forward mutation assay using cell line L5178Y TK+/-</p> <p>The test material was dissolved in ethanol for this assay. Three positive control substances were used viz Ethyl methane sulphonate (EMS) at concentrations of 0.25 &amp; 0.4 µl/ml for the non activation assay, Dimethylnitrosamine (DMN) at a concentration of 0.3 and Methylcholanthrene (MCA) at a concentration of 2.5 µg/ml for the activation assay. Three negative controls were used: 2 solvent (ethanol) controls (not to exceed 1% of of growth medium) and one untreated control. For test substances assayed with activation, the solvent controls included the activation mixture. Araclor-induced rat liver was the source of the S-9 homogenate for the activation assay. A cytotoxicity study was carried out prior to the mutagenicity assay. The test material was lethal at a concentration of 500 nl/ml and highly toxic at 250 nl/ml without S-9. These results were used to select a dose range of 6.25 to 500 nl/ml for the non-activation assay and 3.13 to 400 µl/ml for the activation assay.</p> <p>For the mutation assay, the lymphoma cells were exposed for 4</p>

hours to test material. After exposure to the test material, the cells were allowed to recover for 2 days and then cultures were selected for cloning and mutant selection; 5-Trifluorothymidine (TFT) was used as the restrictive agent. six non-activated and six activated cultures were selected for cloning based on their degree of toxicity. The non-activated cultures that were cloned were treated with 6.25, 25, 37.5 50, 75 and 100 nl/ml of test material and resulted in a range of growth of 30 to 97% compared to the solvent control. The activated cultures that were cloned were treated with 18.8, 37.5, 75, 100, 150 and 200 nl/ml of test material. This resulted in growth ranging from 4.6 to 67.9% compared to solvent control. Plates were prepared from TFT and from the viable cultures (VC) and after 10 to 12 days incubation these plates were scored for total number of colonies per plate. A mutation frequency was then determined.

The following criteria were used in judging the significance of the activity of the test article.

Positive -if there is a positive dose response and one or more of the 3 highest doses exhibit a mutant frequency which is two-fold greater than background level. The minimum criterion for mutagenesis in this assay was a mutant frequency exceeding  $47.0 \times 10^{-6}$ .

Equivocal -if there is no dose response but any one or more doses exhibit a two-fold increase in mutant frequency over background.

Negative -if there is no dose response and none of the test cultures exhibit mutant frequencies which are two-fold greater than background.

### Test Results - Genetic Toxicity in vitro

#### Details on Cytogenetic Assay:

#### Statistics:

#### Effect:

Species	Other Species	Strain	Other Strain	Metabolic Activation	Genotoxic Effect	Conclusion
Mammalian Cell Line		Mouse Lymphoma L5178Y Cells	-	Without	Negative	Negative Without Metabolic Activation
Mammalian Cell Line		Mouse Lymphoma L5178Y Cells	-	With	Positive	Positive With Metabolic Activation

#### Results Remarks:

The mutant frequencies and the percentage total growth at each of the test concentrations is summarized in the following table.

Concentration Mutant % Relative  
(nl/ml) frequency growth

Non-Activated

6.25 24.2 97.3

25 22.5 64.3

37.5 18.2 32.6

50 23 47.8

75 39.6 59.4

100 22.3 29.6

Solvent 1 22.7 100

Solvent 2 30.6 100

Untreated control 20.7 110.6

EMS 0.25 µl/ml 364.5 53.8

EMS 0.4 µl/ml 504.5 23.2

S-9 Activated

18.8 54.2 67.9

37.5 57.3 56.1

75 72.1 60.3

100 85.2 32.8

150 73 27.4

200 146.2 4.6

Solvent 1 31.3 100



Solvent 2 30.8 100  
 Untreated control 42.1 123.9  
 DMN 0.3 µl/ml 258.8 12.7  
 MCA 2.5 µl/ml 243.6 78.5

The authors concluded that the test material was not mutagenic in the non-activated assay because there was no dose response relationship and furthermore the mutant frequency was not significantly different from the solvent and untreated controls. The minimum criteria for indicating mutagenesis would have been  $47 \times 10^{-6}$  and  $62.1 \times 10^{-6}$  for non-activated and S-9 activated groups, respectively. Since the 100 µl/ml treatment represented a close approach to the excessively toxic treatment at 150 nl/ml, this assay was considered sufficient to evaluate the test material as non-mutagenic under non-activation conditions.

In the presence of the S-9 mix, the test material was converted into one or more mutagenic products. The minimum criterion for a significant response was a mutant frequency exceeding  $62.1 \times 10^{-6}$ . This value was exceeded for 4 of the 6 analyzed cultures. The response was dose related. The results were judged sufficient to evaluate the test material as mutagenic in the presence the metabolic activation system.

Mouse lymphoma forward mutation assays have been carried out on two other aromatic naphtha samples. The results, including this study, were:

Sample No.	Aromatic	Response
	content (vol. %)	with S-9
		without S-9
83-04	42.1	negative negative
83-05	62.5	(this study) positive negative
83-06	89.8	
Laboratory 1		positive negative
Laboratory 2		equivocal equivocal

These additional studies are summarized in separate robust study summary records.

**Conclusion:**

The test material API#83-05, CAS # 68955-35-1 was tested in a mouse lymphoma assay with L5178Y TK+/- cell with and without metabolic activation.

The test material was negative for causing forward mutations without metabolic activation.

The test material was positive for causing forward mutations with metabolic activation.

**Reliability/Data Quality - Genetic Toxicity in vitro****Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:**

Key

**Reference - Genetic Toxicity in vitro****Reference:**

American Petroleum Institute (1985) Mutagenicity evaluation of catalytically reformed naphtha API #83-05 in the mouse lymphoma forward mutation assay. Study conducted by Litton Bionetics, Inc. API Med. Res. Publ. 32-32459, July 1985.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Genetic Toxicity in vitro</b>											
<b>Test Substance - Genetic Toxicity in vitro</b>											
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked										
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked										
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>API 83-20. PONA composition is</p> <table border="0"> <tr> <td>Paraffins</td> <td>44.5%</td> </tr> <tr> <td>Olefins</td> <td>46.5%</td> </tr> <tr> <td>Naphthenics</td> <td>0.0%</td> </tr> <tr> <td>Aromatics</td> <td>9.0%</td> </tr> <tr> <td>(Benzene</td> <td>1.2%)</td> </tr> </table> <p>Additional compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at website below)</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>	Paraffins	44.5%	Olefins	46.5%	Naphthenics	0.0%	Aromatics	9.0%	(Benzene	1.2%)
Paraffins	44.5%										
Olefins	46.5%										
Naphthenics	0.0%										
Aromatics	9.0%										
(Benzene	1.2%)										
<b>Category Chemical Result Type:</b>	Measured										
<b>Method - Genetic Toxicity in vitro</b>											
<b>Type of Study:</b>	Mammalian cell gene mutation assay										
<b>Concentrations:</b>	Treatments from 50 to 800 nl/ml without activation and with treatments from 25 to 500 nl/ml with Araclor-induced rat liver S-9 activation.										
<b>Year Study Performed:</b>	1987										
<b>Method/Guideline Followed:</b>											
<b>GLP:</b>	Yes										
<b>Positive, Negative, and Solvent Control Substance (s):</b>	<p>Ethanol was the solvent control</p> <p>Ethyl methane sulfonate (EMS) at concentrations of 0.25 &amp; 0.5 ul/ml was the positive control for non activation assays</p> <p>3-methylcholanthrene (MCA) at concentrations of 2.5 &amp; 4.0 ug/ml was the positive control for activation assays</p>										
<b>Method/Guideline and Test Condition Remarks:</b>	<p>The test material was dissolved in Ethanol for this assay. Two positive control substances were used viz Ethyl methane sulfonate (EMS) at concentrations of 0.25 &amp; 0.5 ul/ml for non activation assays and 3-methylcholanthrene (MCA) at concentrations of 2.5 &amp; 4.0 ug/ml for activation assays. A cytotoxicity study carried out prior to the mutagenicity assay established that the sample was highly toxic at 500 nl/ml without activation and lethal at the same concentration in the presence of metabolic activation. Therefore, for the mutation assay the lymphoma cells were exposed for 4 hours to test material at treatments from 50 to 800 nl/ml without activation and with treatments from 25 to 500 nl/ml with Araclor-induced rat liver S-9 activation. After exposure to the test material, the cells were allowed to recover for 2 days and then cultures were selected for cloning and mutant selection. Plates containing colonies of selected cells were incubated for 10 to 14 days after which they were scored for total number of colonies per plate. A mutation frequency was then determined.</p> <p>Assay evaluation criteria were: The minimum criterion considered necessary to demonstrate mutagenesis for any given treatment is a mutant frequency that is at least 150% of the concurrent background frequency plus <math>10 \times 10^{-6}</math>. The background frequency is defined as the average mutant frequency of the solvent negative</p>										

controls. The minimum increase is based on extensive experience which indicates that assay variability increases with higher backgrounds and the calculated minimum increase as defined above is often a repeatable result; statistical analysis for the confidence limits is not yet available.

The observation of a mutant frequency that meets the minimum criterion for a single treated culture within a range of assayed concentrations is not sufficient evidence to evaluate a test material as a mutagen. The following test results must be obtained to reach this conclusion for either activation or non-activation conditions. A dose-related or toxicity-related increase in mutant frequency should be observed. It is desirable to obtain this relation for at least three doses, but this depends on the concentration steps chosen for the assay and the toxicity at which mutagenic activity appears. If an increase of about two times the minimum criterion or greater is observed for a single dose near the highest testable toxicity, as defined in the Assay acceptance criteria, the test material will be considered mutagenic. Smaller increases at a single dose near the highest testable toxicity will require confirmation by a repeat assay. Treatments that induce less than 10% relative growth are included in the assay, but are not used as primary evidence for mutagenicity as it relates to risk assessment.

In the assay reported in this particular study, under non-activation conditions, the test material was excessively toxic at 300 nl/ml. Five treatments from 50 to 250 nl/ml were therefore chosen for the analysis of mutant induction and non-detectable to moderate toxicities were induced (relative growths 205.3% to 25.5%). None of the assayed treatments induced a mutant frequency that exceeded the minimum criterion of  $89.7 \times 10^{-6}$ . However, since it is desirable to include highly toxic treatments (10 to 20% relative growth) in an analysis, another non activation assay was performed in an attempt to obtain a wider range of toxicities. In the second assay the test material was analyzed for mutant induction from 50 to 150 nl/ml.

In the presence of metabolic activation six treatments from 75 to 400 nl/ml were analyzed for mutant induction and a wide range of toxicities was induced (86.3 to 6.9% relative growths). The minimum criterion for mutagenesis in this assay was a mutant frequency exceeding  $81.4 \times 10^{-6}$ . None of the acceptable treatments induced a mutant frequency that exceeded the minimum criterion. One treatment with less than 10% relative growth (400 nl/ml) induced a mutant frequency that exceeded the minimum criterion, but the treatment was not acceptable for analysis because it did not fulfill the requirements of the assay evaluation criteria. A second assay was therefore performed at treatments ranging from 200 to 300 nl/ml.

Two other olefinic naphtha streams have been tested in a mouse lymphoma assay. The results are summarized below.  
 Sample Result API Report  
 API 81-03 Negative 32-31300  
 With or without S9  
 (American Petroleum Institute (1985) Mutagenicity evaluation studies in the rat bone marrow cytogenetic assay in the mouse lymphoma forward mutation assay Light catalytic cracked naphtha API sample 81-03 API Med. Res. Pub. 32-31300)

API 81-04 Negative without S9 32-31710  
 Equivocal with S9  
 S9 source: Araclor-induced rat liver  
 (American Petroleum Institute (1985) L5178Y TK +/- Mouse lymphoma mutagenesis assay of API 81-04 API Med. Res. Pub. 32-31710)

**Test Results - Genetic Toxicity in vitro**

**Details on Cytogenetic Assay:**

**Statistics:**

**Effect:**

Species	Other Species	Strain	Other Strain	Metabolic Activation	Genotoxic Effect	Conclusion
		Mouse Lymphoma	-	Without	Negative	Negative Without

Mammalian Cell Line	L5178Y Cells				Metabolic Activation
Mammalian Cell Line	Mouse Lymphoma L5178Y Cells	-	With	Negative	Negative With Metabolic Activation

**Results Remarks:**

Only the results of the second assays are summarized since the first assay was not considered acceptable (for the reasons given in the method section above).

## Cloning Relative Mutant

Test condition efficiency growth frequency (%) (10E units)

## Non activation

Solvent control 100.5 100 49.1  
 Solvent control 109.7 100 42.5  
 Solvent control 107.5 100 45.6  
 EMS 0.25 ul/ml 86.8 82.5 286.7  
 EMS 0.4 ul/ml 72.7 56.2 469.7  
 Sample 83-20  
 50 nl/ml 128.4\* 144.7 38.5  
 100 nl/ml 100.1\* 88.8 58.2  
 150 nl/ml 78.5\* 81.6 60.5

## S9 activation

Solvent control 114.7 100 46.5  
 Solvent control 121.0 100 44.3  
 Solvent control 100.2 100 57.2  
 MCA 2.5 ug/ml 87.3 55.5 235.1  
 MCA 4 ug/ml 73.2 53.5 210.0  
 Sample 83-20  
 200 nl/ml 88.7\* 62.4 48.3  
 250 nl/ml 88.1\* 68.8 65.5  
 250 nl/ml 64.3\* 8.6 66.2  
 300 nl/ml 63.7\* 11.1 74.3  
 300 nl/ml 59.7\* 7.3 96.7

\* Cloning efficiency relative to solvent control

In the non activation assay, at most, low toxicities were induced without inducing significant increases above the background mutant frequency (average of solvent controls). Higher toxicities could not be assayed because of a very sharp toxicity curve; a small increase in concentration from 150 to 175 nl/ml was excessively toxic. The test material was, therefore, considered non mutagenic without activation at concentrations that approached excessive toxicity.

In the activation assay, the 250 and 300 nl/ml treatments were duplicated to determine reproducibility. Low and high toxicities were induced by the assayed treatments (68.8 to 7.3% relative growths). For a treatment to be considered mutagenic in this trial, a mutant frequency exceeding  $84.0 \times 10^{-6}$  was required. One treatment at 300 nl/ml induced a mutant frequency that exceeded this criterion but the increase was observed at less than 10% relative growth and a duplicate treatment at the same concentration was inactive. The test material was, therefore, considered non-mutagenic with activation in this assay.

In the assays used in this evaluation, the average cloning efficiencies for the solvent controls varied from 70.5% and 105.9% without activation to 96.2% and 112.0% with activation, which demonstrated acceptable cloning conditions for the assays. The negative control mutant frequencies were all within the expected range and the positive control compounds yielded mutant frequencies that were greatly in excess of the background. Sample 83-20 is considered inactive in the mouse lymphoma assay, with and without metabolic activation.

**Conclusion:**

Sample 83-20 is considered inactive in the mouse lymphoma assay, with and without metabolic activation.

**Reliability/Data Quality - Genetic Toxicity in vitro****Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor  
Indicator:**

Key

**Reference - Genetic Toxicity in vitro**

**Reference:**

American Petroleum Institute (1987) Mutagenicity of API 83-20,  
Light catalytic cracked naphtha (CAS 64741-55-5) in a mouse  
lymphoma mutation assay HESD Pub. No. 34-30633

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data have been entered into the HPVIS: 10/28/2003



<b>Genetic Toxicity in vitro</b>	
<b>Test Substance - Genetic Toxicity in vitro</b>	
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Sample API 83-19 is a Light Alkylate Naphtha (LAN)</p> <p>Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at website below)</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Genetic Toxicity in vitro</b>	
<b>Type of Study:</b>	Other
<b>Concentrations:</b>	0.005 to 0.08 µl/ml without activation and 0.00004 to 0.8 µl/ml with Araclor-induced rat liver S-9 activation
<b>Year Study Performed:</b>	1985
<b>Method/Guideline Followed:</b>	Other
<b>GLP:</b>	Yes
<b>Positive, Negative, and Solvent Control Substance (s):</b>	<p>The test material was dissolved in acetone for this assay.</p> <p>Two positive control substances were used viz Ethyl methane sulphonate (EMS) at concentrations of 1.0 &amp; 0.5 il/ml and 7, 12-DMBA at concentrations of 7.5 &amp; 5.0 µg/ml.</p>
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Type: Mouse lymphoma assay System of testing: Forward mutation assay using cell line L5178Y TK+/-</p> <p>The test material was dissolved in acetone for this assay. Two positive control substances were used viz Ethyl methane sulphonate (EMS) at concentrations of 1.0 &amp; 0.5 il/ml and 7, 12-DMBA at concentrations of 7.5 &amp; 5.0 µg/ml.</p> <p>A cytotoxicity study was carried out prior to the mutagenicity assay. The results were difficult to interpret and as a consequence a second study was carried out and the results from this were used to determine the concentrations to be used in the subsequent lymphoma assay. It was established that complete toxicity occurred at 0.05 µl/ml for the non-activated cultures and at 0.5 ul/ml for S-9 activated cultures.</p> <p>For the mutation assay the lymphoma cells were exposed for 4 hours to test material at concentrations ranging from 0.005 to 0.08 µl/ml without activation and 0.00004 to 0.8 ul/ml with Araclor-induced rat liver S-9 activation. After exposure to the test material, the cells were allowed to recover for 2 days and then cultures were selected for cloning and mutant selection; trifluorothymidine (TFT) was used as the restrictive agent. Eight non-activated and nine activated cultures were selected for cloning based on their degree of toxicity. The non-activated cultures that were cloned were treated with 0.005, 0.01, 0.015, 0.02, 0.025, 0.03, 0.035 or 0.04 ul/ml and resulted in a range of growth of 6 to 97%. The activated cultures that were cloned were treated with 0.0002, 0.0009, 0.0028, 0.008, 0.02, 0.045, 0.09, 0.7 or 0.75</p>

µl/ml and produced a range of growth from 24 to 109%. Plates were prepared from TFT-restricted and from the Viable cultures (VC) and after 10 to 12 days incubation these plates were scored for total number of colonies per plate. A mutation frequency was then determined. The following criteria were used in judging the significance of the activity of the test article.

Positive -if there is a positive dose response and one or more of the 3 highest doses exhibit a mutant frequency which is two-fold greater than background level.

Equivocal -if there is no dose response but any one or more doses exhibit a two-fold increase in mutant frequency over background.

Negative -if there is no dose response and none of the test cultures exhibit mutant frequencies which are two-fold greater than background.

Six mouse lymphoma assays were conducted but for technical reasons four of the assays were invalid. In the fifth assay none of the cultures that were cloned, whether in the presence or absence of S-9 activation exhibited mutant frequencies that were greater than those for the solvent control. However, the toxic response in the S-9 activation portion of the assay was erratic and this portion of the assay was repeated. This summary includes information from the fifth and sixth assays only, since they are the only ones considered to be valid.

### Test Results - Genetic Toxicity in vitro

#### Details on Cytogenetic Assay:

#### Statistics:

#### Effect:

Species	Other Species	Strain	Other Strain	Metabolic Activation	Genotoxic Effect	Conclusion
Mammalian Cell Line		Mouse Lymphoma L5178Y Cells	-	With and Without	Negative	Negative

#### Results Remarks:

The results of the fifth assay are as follows: After the 2 day recovery period, eight non-activated cultures and nine S-9 activated cultures were cloned based on their degree of toxicity. The mutant frequencies and the percentage total growth at each of the test concentrations is summarized in the following table.

Concentration Mutant %Total  
(µl/ml) frequency growth

Non-Activated

0.04 0 34

0.035 0.5 3

0.03 0.2 30

0.025 0 46

0.02 0 93

0.015 -0.2 102

0.01 0 79

0.005 0 93

Solvent 1 0.5

Solvent 2 0.6

DMBA 7.5 µl/ml 3.6 27

DMBA 5 µl/ml 1.9 57

S-9 Activated

0.75 0.2 101

0.7 0.2 16

0.09 0 88

0.045 -0.1 107

0.02 0 107

0.008 0.1 104

0.0028 0 100

0.0009 0 113

0.0002 -0.1 111

Solvent 1 0.6

Solvent 2 0.6

EMS 1µl/ml 8.7 3

EMS 0.5 µl/ml 6.8 29

The sixth assay was with S-9 activation only and the results were as follows:

S-9 Activated  
 0.8 0.2 50  
 0.75 0 84  
 0.7 -0.1 90  
 0.65 -0.4 143  
 0.6 -0.1 99  
 0.5 -0.1 18  
 0.45 0.1 89  
 0.4 -0.1 72  
 0.35 0.1 76  
 0.25 -0.3 31  
 Solvent 1 0.8  
 Solvent 2 0.8  
 DMBA 7.5 µl/ml 1.4 62  
 DMBA 5 µl/ml 1.1 86

The authors concluded that according to the criteria used to judge the activity of the test material, the sample produced a negative response in the presence and absence of S-9 activation.

**Conclusion:** The authors concluded that according to the criteria used to judge the activity of the test material, the sample produced a negative response in the presence and absence of S-9 activation.

#### Reliability/Data Quality - Genetic Toxicity in vitro

**Reliability:** Valid with Restrictions

**Reliability Remarks:** GLP study with adequate description of the methods. Many technical difficulties were encountered before finally producing technically valid results in the 5th and 6th repeats of the assay.

**Key Study Sponsor Indicator:** Key

#### Reference - Genetic Toxicity in vitro

**Reference:** American Petroleum Institute (1985) L5178Y +/-Mouse lymphoma assay, API 83-19 Light Alkylate Naphtha. Study conducted by Microbiological Associates Inc. API Health and Environmental Sciences Dept. Report 32-32746

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<b>Genetic Toxicity in vitro</b>																		
<b>Test Substance - Genetic Toxicity in vitro</b>																		
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline																	
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline																	
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>																	
<b>Category Chemical Result Type:</b>	Measured																	
<b>Method - Genetic Toxicity in vitro</b>																		
<b>Type of Study:</b>	Bacterial reverse mutation assay																	
<b>Concentrations:</b>	<table border="1"> <thead> <tr> <th rowspan="2">Test doses</th> <th colspan="2">% Concentration</th> </tr> <tr> <th>Bacteria</th> <th>Yeast</th> </tr> </thead> <tbody> <tr> <td>1/8 50% survival</td> <td>0.375</td> <td>0.625</td> </tr> <tr> <td>1/4 50% survival</td> <td>0.75</td> <td>1.25</td> </tr> <tr> <td>1/2 50% survival</td> <td>1.5</td> <td>2.5</td> </tr> <tr> <td>50% survival</td> <td>3</td> <td>5</td> </tr> </tbody> </table>	Test doses	% Concentration		Bacteria	Yeast	1/8 50% survival	0.375	0.625	1/4 50% survival	0.75	1.25	1/2 50% survival	1.5	2.5	50% survival	3	5
Test doses	% Concentration																	
	Bacteria	Yeast																
1/8 50% survival	0.375	0.625																
1/4 50% survival	0.75	1.25																
1/2 50% survival	1.5	2.5																
50% survival	3	5																
<b>Year Study Performed:</b>	1977																	
<b>Method/Guideline Followed:</b>																		
<b>GLP:</b>	Yes																	
<b>Positive, Negative, and Solvent Control Substance (s):</b>	DMSO was used as solvent.																	
<b>Method/Guideline and Test Condition Remarks:</b>	<p>The solubility, toxicity and dose levels for the test material were determined prior to the mutagenicity screening.</p> <p>Plate tests: For non-activation assays cells in broth were exposed to the test material at the concentrations shown above. The contents of the tubes of broth plus test material were poured over selective agar plates which were then incubated. The test was conducted with and without Araclor-induced rat liver S-9 metabolic activation. Positive control substances (see results section) were also run in the same assay.</p> <p>The following evaluation criteria were used in this plate test.</p> <p>Strains TA1535, 1537 and 1538: If the solvent control value is within the normal range a chemical which produces a positive response over three concentrations with the lowest increase equal to twice the solvent control value is considered to be mutagenic.</p> <p>Strains TA98, 100 and D4 (yeast): If the solvent control value is within the normal range, a chemical which produces a positive response over three concentrations with the highest increase equal to twice the solvent control value for TA100 and two to three times the solvent control value for</p>																	

strains  
TA98 and D4 is considered to be mutagenic. For these strains, the dose response increase should start at approximately the solvent control value.

Pattern: Because TA1535 and TA100 were both derived from the same parental strain (G-46) and because TA1538 and TA98 were both derived from the same parental strain (D3052), there is a built-in redundancy in the microbial assay. In general the two strains of a set respond to the same mutagen and such a pattern is sought. It is also anticipated that if a given strain responds to a mutagen in non-activation tests it will generally do so in activation tests, but the converse of this is not anticipated. While similar response patterns are not required for all mutagens, they can be used to enhance the reliability of an evaluation decision.

Reproducibility: If a chemical produces a response in a single test which cannot be repeated in one or more additional runs, the initial positive test data loses significance.

The above criteria are not absolute and other extenuating factors may enter into a final evaluation decision.

Suspension tests: Bacteria and yeast cultures were grown in complete broth. The cells were removed, washed and exposed to the test material at the concentrations shown in the results section. For the yeast cells exposure to the test material was for 4 hours whereas for the bacterial cells exposure was for 1 hour. Aliquots of the cells were plated onto the appropriate complete media. After suitable incubation periods, the number of revertant colonies were counted. This assay was also conducted with and without metabolic activation and positive control substances were also included.

The following criteria were used in the suspension assay.

Surviving population counts: A certain level of chemically-induced toxicity is anticipated, but occasionally isolated tests show very low (<25%) survival compared to the tissue controls. Data of this type are generally unacceptable and these experiments are repeated at a lower dose level.

Total mutant counts: For non mutagens, the ratio of mutant to surviving population should be roughly equivalent for each test point in a given experiment. A mutagenic chemical will produce an altered mutant/surviving population ratio. An attempt is made to keep the surviving population of cells high and to look for positive responses that show increases in both numbers of mutants and mutation frequencies.

Dose-response: Dose-related increases in mutants and mutation frequencies are the most convincing data when assessing mutagenic activity. To ensure a proper dose response, dose levels are kept within a relatively low range.

### Test Results - Genetic Toxicity in vitro

**Details on  
Cytogenetic Assay:**

**Statistics:****Effect:**

Species	Other Species	Strain	Other Strain	Metabolic Activation	Genotoxic Effect	Conclusion
Bacteria		S. typhimurium TA 98	-	With and Without	Negative	Negative
Bacteria		S. typhimurium TA 100	-	With and Without	Negative	Negative
Bacteria		S. typhimurium TA 1535	-	With and Without	Negative	Negative
Bacteria		S. typhimurium TA 1537	-	With and Without	Negative	Negative
Bacteria		S. typhimurium TA 1538	-	With and Without	Negative	Negative
Yeast		Saccharomyces cerevisiae	-	With and Without	Negative	Negative

**Results Remarks:**

Plate test: There was no increase in revertants caused by exposure to the test material at any concentration. The results in this assay were negative both with and without metabolic activation.

Suspension test: The mutation frequencies are summarized in the following table for assays with and without metabolic activation.

## Non activation assay

Salmonella strains Yeast

Dose TA100 TA1535 TA1537 TA1538 TA98\* D4\*\* level

-ve control 5.48 3.59 6.15 7.1 41.99 23.69

+ve control 125.51 185.65 161.54 84.75 100 66.29

1 (low) 18.18 2.26 12.54 27.78 233.33 9.52

2 2.9 2.15 8.97 11.76 63.04 36.99

3 3.1 2.98 7.19 10 9.56 30.02

4 (high) 4.13 2.66 9.68 3.21 35.74 32.38

Assay repeated for negative control and lowest 2 doses. Results were 54.59 for -

ve control, 10.84 for lowest dose, 14.11 for next highest dose

\*\* Assay repeated at all dose levels

Results were: -ve control 4.66, +ve control 97.73, dose level 1 1.3, dose level

2 8.33, dose level 4 12.65.

Slight increases are observed at the high dose levels with TA100, TA1537 and TA1538. However the responses are not adequate enough to be considered positive.

The increases with TA98 could not be reproduced.

## With activation

Salmonella strains Yeast

Dose TA100 TA1535 TA1537 TA1538 TA98\* D4\*\* level

-ve controls\*

A+C 17.08 5.25 6.01 4.8 21.01 52.66

A-C 17.29 8.77 9.29 8.25 62.02 7.96

AL1 17.34 7.32 3.99 6.48 45.03 30.06

+ve control 25.51 89.92 0.22 1253.4 555.35 115.3

1 (low) 22.97 41.67 100 71.43 100

2 15.64 7.21 0 300 30.66 27.22

3 17.26 9.57 20 15.38 83.33 27.03

4 22 .31 7.21 5.43 6.93 60.13 29.04

\* Controls were

A+C No activation system but including positive control

A-C Solvent control, no test chemical or activation system

AL1 Liver homogenate control plus solvent

Scattered increases were found at one or more dose levels (see table above). All apparent positive effects were repeated and were not reproducible indicating problems associated with the initial runs. When the raw data were inspected it was observed that the increases were due to anomalous reductions in viable cell counts. The results of this assay were therefore considered to be negative.

**Conclusion:** Unleaded gasoline was not mutagenic with and without metabolic activation in 5 strains of bacteria or one strain of yeast.

#### **Reliability/Data Quality - Genetic Toxicity in vitro**

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Valid with restrictions due to poor quality of initial assay.

**Key Study Sponsor Indicator:** Key

#### **Reference - Genetic Toxicity in vitro**

**Reference:** American Petroleum Institute (1977) Mutagenicity evaluation of unleaded gasoline Study conducted by Litton Bionetics, Inc. API HESD Publication No. 28-30173, March 1977

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Genetic Toxicity in vitro</b>	
<b>Test Substance - Genetic Toxicity in vitro</b>	
<b>Category Chemical:</b>	(64741-87-3) Naphtha, petroleum, sweetened
<b>Test Substance:</b>	(64741-87-3) Naphtha, petroleum, sweetened
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>API 81-08</p> <p>Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at website below)</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Genetic Toxicity in vitro</b>	
<b>Type of Study:</b>	Mammalian cell gene mutation assay
<b>Concentrations:</b>	12.5 -300 µl/ml
<b>Year Study Performed:</b>	1985
<b>Method/Guideline Followed:</b>	
<b>GLP:</b>	Yes
<b>Postive, Negative, and Solvent Control Substance (s):</b>	<p>Two positive control substances were used viz Ethyl methane sulphonate (EMS) at a concentration of 0.5 µl/ml in the assay without activation and Dimethylnitrosamine (DMN) at a concentrations of 0.3 µl/ml.</p> <p>Ethanol was the solvent; 2 solvent controls were included in the assays.</p>
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Type: Mouse lymphoma assay System of testing: Forward mutation assay using cell line L5178Y TK+/-</p> <p>Based on a preliminary test, ethanol was selected as solvent for this assay. Concentrations of 0.061 to 1000 µl/ml appeared soluble in the assay medium and no change in color was noted. Two positive control substances were used viz Ethyl methane sulphonate (EMS) at a concentration of 0.5 µl/ml in the assay without activation and Dimethylnitrosamine (DMN) at a concentrations of 0.3 µl/ml.</p> <p>A cytotoxicity study was carried out prior to the mutagenicity assay. The results were difficult to interpret and as a consequence a second study was carried out and the results from this were used to determine the concentrations to be used in the subsequent lymphoma assay. It was established that complete toxicity occurred at 0.05 µl/ml for the non-activated cultures and at 0.5 µl/ml for Araclor-induced rat liver S-9 activated cultures.</p> <p>For the mutation assay the lymphoma cells were exposed for 4 hours to test material at concentrations ranging from 0.005 to 0.08 µl/ml without activation and 0.00004 to 0.8 µl/ml with S-9 activation. After exposure to the test material, the cells were allowed to recover for 2 days and then cultures were selected for cloning and mutant selection; 5-trifluorothymidine (TFT) was used as the restrictive agent.</p> <p>Eight non-activated and nine activated cultures were selected for</p>

cloning based on their degree of toxicity. The non-activated cultures that were cloned were treated with 0.005, 0.01, 0.015, 0.02, 0.025, 0.03, 0.035 or 0.04 µl/ml and resulted in a range of growth of 6 to 97%. The activated cultures that were cloned were treated with 0.0002, 0.0009, 0.0028, 0.008, 0.02, 0.045, 0.09, 0.7 or 0.75 µl/ml and produced a range of growth from 24 to 109%. Plates were prepared from TTF and from the viable culture (VC) and after 10 to 12 days incubation these plates were scored for total number of colonies per plate. A mutation frequency was then determined.

The following criteria were used in judging the significance of the activity of the test article.

Positive -if there is a positive dose response and one or more of the 3 highest doses exhibit a mutant frequency which is two-fold greater than background level.

Equivocal -if there is no dose response but any one or more doses exhibit a two-fold increase in mutant frequency over background.

Negative -if there is no dose response and none of the test cultures exhibit mutant frequencies which are two-fold greater than background.

### Test Results - Genetic Toxicity in vitro

#### Details on

#### Cytogenetic Assay:

Cycotoxic concentr.: 0.05 µl/ml without activation; 0.5 µl/ml with activation

#### Statistics:

#### Effect:

Species	Other Species	Strain	Other Strain	Metabolic Activation	Genotoxic Effect	Conclusion
Mammalian Cell Line		Mouse Lymphoma L5178Y Cells	-	With and Without	Negative	Negative

#### Results Remarks:

The data from each of the 3 trials that were considered valid are tabulated below.

Conc. Relative growth Mutant frequency  
(µl.ml) (%) (10<sup>-6</sup> units)

TRIAL 1 No activation

15.6 118.6 18.1

31.3 64.4 27

62.5 97.8 15.7

125 78.2 24.2

250 20.1 48.8

Solvent control 1 100 13.9

Solvent control 2 100 20.3

Untreated control 191.7 21.5

EMS 0.5 µl/ml 17.4 258.2

TRIAL 1 with S-9 activation

15.6 78.1 59.1

31.3 53.8 49.3

62.5 63.3 49

125 46.5 79.7

250 46.3 41.6

Solvent control 1 100 34

Solvent control 2 100 24

Untreated control 100.7 30.5

DMN 0.3 µl/ml 5 327.5

TRIAL 4 No activation

12.5 47.8 19.5

25 49.7 19.2

50 37.7 13.5

100 113.3 8.5

200 86.2 9.3

300 19.8 36.4

Solvent control 1 100 18.3

Solvent control 2 100 18.5

Untreated control 163.9 16.2

EMS 0.5 µl/ml 13.5 700

TRIAL 4 with S-9 activation

12.5 81.6 52.3  
 25 60.2 85.7  
 50 57.3 59.1  
 100 44.7 63.8  
 200 71.8 21  
 300 3.1 19.3  
 Solvent control 1 100 23.2  
 Solvent control 2 100 22.9  
 Untreated control 78.2 22.2  
 DMN 0.3 µl/ml 8.8 469.4

TRIAL 5 with S-9 activation  
 150 76.9 13.6  
 150 28.4 25.2  
 200 42.5 24  
 200 41.9 15.3  
 250 59.6 24.2  
 250 15.6 31.1  
 300 4.9 30.2  
 300 7.3 32  
 Solvent control 1 100 27.1  
 Solvent control 2 100 19.2  
 Solvent control 3 100 22.4  
 Solvent control 4 100 24.5  
 Untreated control 1 63.8 31  
 Untreated control 2 49.9 29.2  
 DMN 0.3 µl/ml 16.6 352.9  
 DMN 0.3 µl/ml 2.2 333.3

#### TRIAL 1:

Non activation conditions.: The percent relative growths of the assayed treatments ranged from 118.6% to 20.1% which demonstrated non-detectable to moderate toxicities. The minimum criterion for mutagenesis in this assay was a mutant frequency that exceeding  $37.8 \times 10^{-6}$ . The highest, most toxic treatment (250 µl/ml) induced a mutant frequency that exceeded the minimum criterion, but the increase in the mutant frequency was not accompanied by an increase in the total mutant clones. In order to determine if the increase was repeatable, another nonactivation assay was performed.

Activated assay: Test material was assayed at concentrations ranging from 15.6 to 250 µl.ml. The minimum criterion for mutagenesis in this assay was a mutant frequency exceeding  $54.2 \times 10^{-6}$ . Two treatments induced mutant frequencies that exceeded the minimum criterion, but the increases were sporadic and unrelated to dose or toxicity. Another assay was therefore performed.

#### TRIAL 4

Non activated assay.: The test material was assayed at concentrations ranging from 12.5 to 300 µl/ml. In order for a treatment to be considered mutagenic in this assay, a mutant frequency of  $36.5 \times 10^{-6}$  was required. None of the assayed treatments induced mutant frequencies that exceeded the minimum criterion. The observed toxicities ranged from non toxic to moderate toxicity. Although it is preferable to consider results from treatments that induce high toxicity, it was not possible in this assay because of a sharp toxicity curve. The test material was therefore considered non mutagenic without activation in this assay at treatments that approached lethality.

Activated assay.: Concentrations ranging from 12.5 to 300 µl/ml were used in this assay and low to very high toxicity was induced. Sporadic increases in the mutant frequency were induced. The minimum criterion for mutagenesis in this assay was a mutant frequency exceeding  $44.2 \times 10^{-6}$  and three treatments did exceed the minimum criterion (25, 50 & 100 nl/ml). However, the highest concentrations assayed were non-mutagenic. A further assay was therefore performed.

#### TRIAL 5:

Activated assay: The test material was assayed in duplicate at concentrations ranging from 150 to 300 nl/ml. A wide range of toxicities were induced. The sporadic increases in mutant frequency observed in Trials 1 and 4 were not repeatable. None of the treatments induced mutant frequencies that exceeded the minimum criterion of  $48.4 \times 10^{-6}$ . The test material was therefore considered non-mutagenic with activation in this assay.

#### Conclusion:

The investigators concluded that the test material, API 81-08 was non-mutagenic in the absence and presence of metabolic activation in the mouse lymphoma forward mutation assay.

However, due to wide ranges of toxicity and sporadic increases in mutant frequencies, five trials were performed in order to verify the absence of genetic toxicity in this assay system.

#### Reliability/Data Quality - Genetic Toxicity in vitro

<b>Reliability:</b>	Valid with Restrictions
<b>Reliability Remarks:</b>	multiple assays needed to get usable studies
<b>Key Study Sponsor Indicator:</b>	Key

#### Reference - Genetic Toxicity in vitro

<b>Reference:</b>	American Petroleum Institute (1985) Mutagenicity evaluation studies in the mouse lymphoma forward mutation assay, sweetened naphtha, sample 81-08. Study carried out by Litton Bionetics Inc. API Medical Research Publication No. 32-31233
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<b>Genetic Toxicity in vitro</b>	
<b>Test Substance - Genetic Toxicity in vitro</b>	
<b>Category Chemical:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Test material was a light catalytically reformed naphtha, CAS # 64741-63-5</p> <p>Test material designation by study sponsor was API # 83-04. Compositional information on this test material can be found in the analytical data report attached to the Gasoline Blending Stream Category at website below.</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Genetic Toxicity in vitro</b>	
<b>Type of Study:</b>	Other
<b>Concentrations:</b>	The non-activated cultures that were cloned were treated with 25, 50, 75, 100 and 125 nl/ml of test material. The activated cultures that were cloned were treated with 25, 50, 75, 100, and 150 nl/ml of test material.
<b>Year Study Performed:</b>	1985
<b>Method/Guideline Followed:</b>	No Data
<b>GLP:</b>	Yes
<b>Positive, Negative, and Solvent Control Substance (s):</b>	<p>Three positive control substances were used: Ethyl methane sulphonate (EMS) at concentrations of 0.25 &amp; 0.4 µl/ml for the non activation assay, Dimethylnitrosamine (DMN) at a concentration of 0.3 and Methylcholanthrene (MCA) at a concentration of 2.5 µg/ml for the activation assay.</p> <p>Three negative controls were used: 2 solvent (ethanol) controls (not to exceed 1% of of growth medium) and one untreated control. For test substances assayed with activation, the solvent controls included the activation mixture.</p>
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Type: Mouse lymphoma assay System of testing: Forward mutation assay using cell line L5178Y TK+/-</p> <p>The test material was dissolved in ethanol for this assay. Three positive control substances were used viz Ethyl methane sulphonate (EMS) at concentrations of 0.25 &amp; 0.4 µl/ml for the non activation assay, Dimethylnitrosamine (DMN) at a concentration of 0.3 and Methylcholanthrene (MCA) at a concentration of 2.5 µg/ml for the activation assay. Three negative controls were used: 2 solvent (ethanol) controls (not to exceed 1% of of growth medium) and one untreated control. For test substances assayed with activation, the solvent controls included the activation mixture. Araclor-induced rat liver was the source of the S-9 homogenate for the activation assay. A cytotoxicity study was carried out prior to the mutagenicity assay.</p> <p>For the mutation assay, the lymphoma cells were exposed for 4 hours to test material. After exposure to the test material, the cells were allowed to recover for 2 days and then cultures were selected for cloning and mutant selection; 5-Trifluorothymidine</p>

(TFT) was used as the restrictive agent. Six non-activated and six activated cultures were selected for cloning based on their degree of toxicity. The non-activated cultures that were cloned were treated with 25, 50, 75, 100, and 125 nl/ml of test material and resulted in a range of growth of 1.6 to 53.1% compared to the solvent control. The highest concentration of test material was not used to determine non-activated mutagenicity due to the low (1.6%) growth rate. The activated cultures that were cloned were treated with 25, 50, 75, 100, 125, and 150 nl/ml of test material. This resulted in growth ranging from 14.1 to 94.4% compared to solvent control. Plates were prepared from TFT and from the viable cultures (VC) and after 10 to 12 days incubation these plates were scored for total number of colonies per plate. A mutation frequency was then determined.

The following criteria were used in judging the significance of the activity of the test article.

**Positive** -if there is a positive dose response and one or more of the 3 highest doses exhibit a mutant frequency which is two-fold greater than background level. The minimum criterion for mutagenesis in this assay was a mutant frequency exceeding  $52.0 \times 10^{-6}$  and  $72.9 \times 10^{-6}$ , for non-activated and S-9 activated groups, respectively.

**Equivocal** -if there is no dose response but any one or more doses exhibit a two-fold increase in mutant frequency over background.

**Negative** -if there is no dose response and none of the test cultures exhibit mutant frequencies which are two-fold greater than background.

### Test Results - Genetic Toxicity in vitro

#### Details on Cytogenetic Assay:

#### Statistics:

#### Effect:

Species	Other Species	Strain	Other Strain	Metabolic Activation	Genotoxic Effect	Conclusion
Mammalian Cell Line		Mouse Lymphoma L5178Y Cells	-	Without	Negative	Negative Without Metabolic Activation
Mammalian Cell Line		Mouse Lymphoma L5178Y Cells	-	With	Negative	Negative With Metabolic Activation

#### Results Remarks:

The mutant frequencies and the percentage total growth at each of the test concentrations is summarized in the following table.

Concentration Mutant % Relative  
(nl/ml) frequency growth

Non-Activated

25 32.1 53.1

50 34.6 43.7

75 31.7 26.4

100 48.7 14.2

125\*\* 63.3\*\* 1.6\*\*

Solvent 1 33.0 100

Solvent 2 27.1 100

Untreated control 23.9 110.6

EMS 0.25 µl/ml 350.2 58.9

EMS 0.4 µl/ml 585.3 22.2

\*\* not used to determine mutagenicity due to low relative growth

S-9 Activated

25 49.0 94.4

50 80.1 49.4

75 77.5 55.8

100 98.8 36.7

125 72.5 28.7

150 66.6 14.1

Solvent 1 45.2 100

Solvent 2 37.5 100  
 Untreated control 43.1 123.9  
 DMN 0.3 µl/ml 237.8 12.7  
 MCA 2.5 µl/ml 340.9 78.5

The authors concluded that the test material was not mutagenic in the non-activated assay because there was no dose response relationship and furthermore the mutant frequency was not significantly different from the solvent and untreated controls. The highest concentration was not used in the mutagenicity determination due to excessive toxicity (1.6% of solvent control growth). The minimum criteria for indicating mutagenesis would have been  $52.0 \times 10^{-6}$ . This assay was considered sufficient to evaluate the test material as non-mutagenic under non-activation conditions.

In the presence of metabolic activation, three treatments (50, 75, and 100 nl/ml) induced mutation frequencies that exceeded the  $72.9 \times 10^{-6}$  mutation frequency criterion determined to designate a positive response. The increases were small (80.1, 77.5 and 98.8, respectively), and only one of the three doses (100 nl/ml) was more than 2-fold above background. The response was not dose related, and no increases with respect to control were observed at the two highest concentrations, i.e. higher, more toxic concentrations were not mutagenic. The investigators concluded that the observed increases were spurious and the test material was considered nonmutagenic with activation in this assay.

Mouse lymphoma forward mutation assays have been carried out on two other aromatic naphtha samples. The results, including this study, were:

Sample No.	Aromatic	Response
	content (vol. %)	with S-9 without S-9
83-04	42.1	(this study) negative negative
83-05	62.5	positive negative
83-06	89.8	
Laboratory 1	positive	negative
Laboratory 2	equivocal	equivocal

These additional studies are summarized in separate robust study summary records.

**Conclusion:**

The test material API#83-04, CAS # 64741-63-5 was tested in a mouse lymphoma assay with L5178Y TK+/- cell with and without metabolic activation.

The test material was negative for causing forward mutations without metabolic activation

The test material was negative for causing forward mutations in the presence of metabolic activation.

**Reliability/Data Quality - Genetic Toxicity in vitro**

**Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:**

Key

**Reference - Genetic Toxicity in vitro**

**Reference:**

American Petroleum Institute (1985) Mutagenicity evaluation of API 83-04 in the mouse lymphoma forward mutation assay API Report No. 32-32168

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<b>Genetic Toxicity in vitro</b>	
<b>Test Substance - Genetic Toxicity in vitro</b>	
<b>Category Chemical:</b>	(64741-68-0) Naphtha, petroleum, heavy catalytic reformed
<b>Test Substance:</b>	(64741-68-0) Naphtha, petroleum, heavy catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Test material was a heavy catalytically reformed naphtha, CAS # 64741-68-0</p> <p>Test material designation by study sponsor was API # 83-06. Compositional information on this test material can be found in the analytical data report attached to the Gasoline Blending Stream Category at website below.</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Genetic Toxicity in vitro</b>	
<b>Type of Study:</b>	Mammalian cell gene mutation assay
<b>Concentrations:</b>	<p>The non-activated cultures that were cloned were treated with 6.25, 18.8, 37.5, 50, 62.5 75 and 100 nl/ml of test material.</p> <p>Trial # 1: The activated cultures that were cloned were treated with 6.25, 18.8, 37.5, 50, 62.5 75 and 100 nl/ml of test material.</p> <p>Trial #2 : The activated cultures that were cloned were treated with 10.0, 30.0, 60.0, 80.0, 100.0, and 120.0 nl/ml of test material.</p>
<b>Year Study Performed:</b>	1985
<b>Method/Guideline Followed:</b>	
<b>GLP:</b>	Yes
<b>Positive, Negative, and Solvent Control Substance (s):</b>	<p>Two positive control substances were used viz Ethyl methane sulphonate (EMS) at concentrations of 0.25 &amp; 0.4 µl/ml for the non activation assay, Methylcholanthrene (MCA) was used at a concentration of 2.5 &amp; 4.0 µg/ml for the activation assay.</p> <p>Three negative solvent (DMSO) controls were used (not to exceed 1% of of growth medium). For test substances assayed with activation, the solvent controls included the activation mixture.</p>
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Type: Mouse lymphoma assay</p> <p>System of testing: Forward mutation assay using cell line L5178Y TK+/-</p> <p>The test material was dissolved in dimethyl sulfoxide (DMSO) for this assay. Two positive control substances were used viz Ethyl methane sulphonate (EMS) at concentrations of 0.25 &amp; 0.4 µl/ml for the non activation assay, Methylcholanthrene (MCA) was used at a concentration of 2.5 &amp; 4.0 µg/ml for the activation assay. Three negative solvent (DMSO) controls were used (not to exceed 1% of of growth medium). For test substances assayed with activation, the solvent controls included the activation mixture. Araclor-induced rat liver was the source of the S-9 homogenate for the activation assay. A cytotoxicity study was carried out prior to the mutagenicity assay.</p> <p>For the mutation assay, the lymphoma cells were exposed for 4 hours to test material. After exposure to the test material, the</p>

cells were allowed to recover for 2 days and then cultures were selected for cloning and mutant selection; 5-Trifluorothymidine (TFT) was used as the restrictive agent. six non-activated and six activated cultures were selected for cloning based on their degree of toxicity. The non-activated cultures that were cloned were treated with 6.25, 18.8, 37.5, 50, 62.5 75 and 100 nl/ml of test material and resulted in a range of growth of 7.3 - 39.5% compared to the solvent control. The activated cultures that were cloned were treated with 16.25, 18.8, 37.5, 50, 62.5 75 and 100 nl/ml of test material in Trial #1 and 10.0, 30.0, 60.0, 80.0, 100.0, and 120.0 nl/ml in Trial #2. This resulted in growth ranging from 24.9% to 79.6% (compared to solvent control) in Trial #1 and 15.0% to 100.0% (as compared to controls) in Trial #2. Plates were prepared from TFT and from the viable cultures (VC) and after 10 to 12 days incubation these plates were scored for total number of colonies per plate. A mutation frequency was then determined.

The following criteria were used in judging the significance of the activity of the test article.

Positive -if there is a positive dose response and one or more of the 3 highest doses exhibit a mutant frequency which is two-fold greater than background level. The minimum criterion for mutagenesis in this assay was a mutant frequency exceeding  $68.7 \times 10^{-6}$  and  $85.3 \times 10^{-6}$  for Trials one (1) and two (2), respectively.

Equivocal -if there is no dose response but any one or more doses exhibit a two-fold increase in mutant frequency over background.

Negative -if there is no dose response and none of the test cultures exhibit mutant frequencies which are two-fold greater than background.

### Test Results - Genetic Toxicity in vitro

#### Details on Cytogenetic Assay:

#### Statistics:

#### Effect:

Species	Other Species	Strain	Other Strain	Metabolic Activation	Genotoxic Effect	Conclusion
Mammalian Cell Line		Mouse Lymphoma L5178Y Cells	-	Without	Negative	Negative Without Metabolic Activation
Mammalian Cell Line		Mouse Lymphoma L5178Y Cells	-	With	Positive	Positive With Metabolic Activation

#### Results Remarks:

TRIAL # 1  
The mutant frequencies and the percentage total growth at each of the test concentrations are summarized in the following tables. The minimum criterion for mutagenesis in this assay was a mutant frequency exceeding  $68.7 \times 10^{-6}$  for activated preparations.

Test Group % Relative Mutant growth frequency  
Non-Activated  
Solvent Control 100.0 19.0  
Solvent Control 100.0 14.7  
Solvent Control 100.0 20.3  
EMS 0.25 µl/ml 44.4 334.4  
EMS 0.4 µl/ml 21.3 622.8  
Test Compound (nl/ml)  
6.25 39.5 25.0  
18.8 40.9 13.7  
37.5 35.9 21.1  
50.0 24.0 16.1  
62.5 10.9 24.0  
75.0 7.3 27.4  
100.0 excessive toxicity; treatment not cloned

Test Group % Relative Mutant  
 growth frequency  
 S-9 Activated  
 Solvent Control 100.0 39.7  
 Solvent Control 100.0 41.7  
 Solvent Control 100.0 35.9  
 MCA 2.5 µl/ml 59.3 196.0  
 MCA 4.0 µl/ml 24.8 349.6  
 Test Compound (nl/ml) < br>6.25 79.6 44.4  
 18.8 62.4 69.4  
 37.5 42.2 64.3  
 50.0 24.7 87.4  
 62.5 23.1 76.1  
 75.0 24.9 85.54  
 100.0 excessive toxicity; treatment not cloned

TRIAL # 2 (S-9 Activated groups only)  
 The mutant frequencies and the percentage total growth at each of the test concentrations are summarized in the following tables. The minimum criterion for mutagenesis in this assay was a mutant frequency exceeding  $85.3 \times 10^{-6}$  for activated preparations.

Test Group % Relative Mutant  
 growth frequency  
 S-9 Activated  
 Solvent Control 100.0 39.7  
 Solvent Control 100.0 41.7  
 Solvent Control 100.0 35.9  
 MCA 2.5 µl/ml 85.0 196.0  
 MCA 4.0 µl/ml 32.3 349.6  
 Test Compound (nl/ml)  
 10.0 100.0 63.8  
 30.0 93.0 73.2  
 60.0 67.8 84.1  
 80.0 60.7 78.4  
 100.0 31.7 91.0  
 120.0 15.0 90.6

The authors concluded that the test material was not mutagenic in the non-activated assay because there was no dose response relationship and furthermore the mutant frequency was not significantly different from the solvent and untreated controls. Since the 75 nl/ml treatment represented a close approach to the excessively toxic treatment, this assay was considered sufficient to evaluate the test material as non-mutagenic under non-activation conditions.

In the presence of the S-9 mix, the test material was converted into one or more mutagenic products. The minimum criterion for a significant response was a mutant frequency exceeding  $68.7$  and  $85.3 \times 10^{-6}$  for Trials 1 and 2. This value was exceeded for the three highest concentrations of test material; the increases were from 1.9- to 2.2-fold above background mutant frequency (average of solvent controls). While these increases were considered significant, they were small and required confirmation in a second trial.

The two highest concentrations in the second trial (100 and 120 ng/ml) induced small but significant increases above the background mutant frequency. The increases observed in Trial #1 were therefore repeatable and the test material was considered weakly mutagenic in the presence of the metabolic activation system.

Mouse lymphoma forward mutation assays have been carried out on two other aromatic naphtha samples, as well as a repeat study (API report # 33-31641) for 83-06 in a separate testing facility. The results were:

Sample No. Aromatic Response  
 content (vol. %) with S-9 without S-9  
 83-04 42.1 negative negative  
 83-05 62.5 positive negative  
 83-06 89.8  
 Laboratory 1 (this study) positive negative  
 Laboratory 2 equivocal equivocal

These additional studies are summarized in separate robust study summary records.

**Conclusion:** The test material API#83-06, CAS # 64741-68-0 was tested in a mouse lymphoma assay with L5178Y TK+/- cell with and without metabolic activation.

The test material was negative for causing forward mutations without metabolic activation.

The test material was positive for causing forward mutations with metabolic activation.

#### Reliability/Data Quality - Genetic Toxicity in vitro

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:** Key

#### Reference - Genetic Toxicity in vitro

**Reference:** American Petroleum Institute (1985) Mutagenicity evaluation in the mouse lymphoma forward mutation assay. Study conducted by Litton Bionetics, Inc. Litton Project No. 20989. API 83-06 Heavy catalytically reformed naphtha API Report No. 32-32460

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Genetic Toxicity in vitro</b>	
<b>Test Substance - Genetic Toxicity in vitro</b>	
<b>Category Chemical:</b>	(64741-68-0) Naphtha, petroleum, heavy catalytic reformed
<b>Test Substance:</b>	(64741-68-0) Naphtha, petroleum, heavy catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Test material was a heavy catalytically reformed naphtha, CAS # 64741-68-0</p> <p>Test material designation by study sponsor was API # 83-06. Compositional information on this test material can be found in the analytical data report attached to the Gasoline Blending Stream Category at the website below.</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Genetic Toxicity in vitro</b>	
<b>Type of Study:</b>	Mammalian cell gene mutation assay
<b>Concentrations:</b>	<p>The non-activated cultures that were cloned were treated with 18, 24, 32, 42, 56, and 75 nl/ml of test material.</p> <p>Trial # 1: The activated cultures that were cloned were treated with 67, 89, and 120 nl/ml of test material.</p> <p>Trial #2 : The activated cultures that were cloned were treated with 70, 110, 150, 180, and 220 µl/ml of test material.</p>
<b>Year Study Performed:</b>	1986
<b>Method/Guideline Followed:</b>	No Data
<b>GLP:</b>	Yes
<b>Positive, Negative, and Solvent Control Substance (s):</b>	<p>Two positive control substances were used: Ethyl methane sulphonate (EMS) at concentrations of 0.5 &amp; 1.0 µl/ml for the non-activation assay, Dimethybenzanthrene (DMBA) was used at a concentration of 5.0 &amp; 7.5 µg/ml for the activation assay.</p> <p>Two negative solvent (ethanol) controls were used. For test substances assayed with activation, the solvent controls included the activation mixture.</p>
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Type: Mouse lymphoma assay</p> <p>System of testing: Forward mutation assay using cell line L5178Y TK+/-</p> <p>The test material was dissolved in ethanol for this assay. Two positive control substances were used: Ethyl methane sulphonate (EMS) at concentrations of 0.5 &amp; 1.0 µl/ml for the non activation assay, Dimethybenzanthrene (DMBA) was used at a concentration of 5.0 &amp; 7.5 µg/ml for the activation assay.</p> <p>Two negative solvent (ethanol) controls were used. For test substances assayed with activation, the solvent controls included the activation mixture. Araclor-induced rat liver was the source of the S-9 homogenate for the activation assay. A cytotoxicity study was carried out prior to the mutagenicity assay.</p> <p>For the mutation assay, the lymphoma cells were exposed for 4 hours to test material. After exposure to the test material, the cells were allowed to recover for 2 days and then cultures were selected for cloning and mutant selection; 3 g/ml</p>



5-Trifluorothymidine (TFT) was used as the restrictive agent. The non-activated cultures (A & B) that were cloned were treated with 18, 24, 32, 42, 56, and 75 nl/ml of test material and resulted in a range of growth of 1 - 115% compared to the solvent control. Two activated cultures (A & B) that were cloned were treated with 67, 89, and 120 nl/ml of test material in Trial #1 and 70, 110, 150, 180, and 220 µl/ml in Trial #2. This resulted in growth ranging from 4% to 28% (compared to solvent control) in Trial #1 and 10% to 91% (as compared to controls) in Trial #2. Plates were prepared from TFT and from the viable cultures (VC) and after 10 to 12 days incubation these plates were scored for total number of colonies per plate. A mutation frequency was then determined.

Trials 1 and 2 were conducted more than one year apart, so the cytotoxicity study was repeated for trial 2.

The following criteria were used in judging the significance of the activity of the test article.

Positive -if there is a positive dose response and one or more of the doses in the 10% or greater total growth range exhibit a mutant frequency which is two-fold greater than background level.

Equivocal -if there is no dose response but any one or more of the three highest doses with 10% or greater total growth exhibit a two-fold increase in mutant frequency over background; or if there is a dose response but no culture exhibits a two-fold increase in mutant frequency over background.

Negative -if there is no dose response in cultures with 10% or greater growth and none of the test cultures exhibit mutant frequencies which are two-fold greater than background.

### Test Results - Genetic Toxicity in vitro

#### Details on Cytogenetic Assay:

#### Statistics:

#### Effect:

Species	Other Species	Strain	Other Strain	Metabolic Activation	Genotoxic Effect	Conclusion
Mammalian Cell Line		Mouse Lymphoma L5178Y Cells	-	Without	Equivocal	Equivocal Without Metabolic Activation
Mammalian Cell Line		Mouse Lymphoma L5178Y Cells	-	With	Equivocal	Equivocal With Metabolic Activation

#### Results Remarks:

TRIAL # 1  
The mutant frequencies and the percentage total growth at each of the test concentrations are summarized in the following tables.

Test Group % Total Mutant  
Growth frequency  
Non-Activated

Solvent Control 1 100 0.6

Solvent Control 2 100 0.7

EMS 0.5 µl/ml 31 10.2

EMS 1.4 µl/ml 2 27.3

Test Compound (nl/ml)

18 A 115 0.5

18 B 109 0.8

24 A 110 0.6

24 B 83 0.7

32 A 85 0.7

32 B 89 0.7

42 A 31 1.0

42 B 47 0.9

56 A 85 0.6

56 B 10 1.4

75 A 1 21.7

75 B 2 2.5

Test Group % Total Mutant  
 Growth frequency  
 S-9 Activated  
 Solvent Control 100 0.9  
 Solvent Control 100 1.0  
 DMBA 5 µl/ml 22 6.8  
 DMBA 7.5 µl/ml too toxic to clone  
 Test Compound (nl/ml)  
 67 A 11 1.8  
 67 B 27 1.3  
 89 A 7 2.5  
 89 B 16 1.7  
 120 A 13 1.7  
 120 B 4 2.1

TRIAL # 2 (S-9 Activated groups only)

The mutant frequencies and the percentage total growth at each of the test concentrations are summarized in the following tables.

Test Group % Relative Mutant  
 growth frequency  
 S-9 Activated  
 Solvent Control 100 0.9  
 Solvent Control 100 1.0  
 DMBA 5 µl/ml 55 3.9  
 DMBA 7.5 µl/ml 34 4.8  
 Test Compound (nl/ml)  
 70 A 83 1.4  
 70 B 91 1.5  
 110 A 61 1.6  
 110 B 53 1.4  
 150 A 44 1.6  
 150 B 28 1.5  
 180 A 28 1.6  
 180 B 23 1.6  
 220 A 20 1.7  
 220 B 10 2.1

Three non-activated cultures (75 A, 75 B and 56 B nl/ml) exhibited mutant frequencies which were 31.0, 3.6 or 2.0 times, respectively, the mean mutant frequency of the solvent controls. The Total Growth of these cultures was 1%, 2%, and 10%, respectively. The remaining non-activated culture mutant frequencies did not differ significantly from control. It is customary to consider that significant increases observed only at highly toxic concentrations (<10% Total Growth) may be due to epigenetic events. Because this test article has a very steep toxic response curve in this system, very minute differences in dose result in large differences in Total Growth. The Total Growth exhibited by each culture may be more representative of dose delivered than the test article concentration indicated. A comparison of induced Mutant Frequency with Total Growth indicated a dose dependent response and the data are judged to be equivocal.

Two trials were conducted in the presence of the S-9 mix, with a time interval exceeding one year between each trial. In Trial #1, two cloned cultures, 89 nl/ml A and 120 nl/ml B, had mutant frequencies that were 2.5 and 2.1 times greater than solvent controls, respectively. Percent Total Growth was 4% for the 120 nl/ml culture, and 7% for the 89 nl/ml culture. Since the Total Growth of these cultures was below 10%, the increase in mutant frequency was not considered significant because TFT resistance observed at these highly toxic levels may be due to epigenetic events.

In Trial #2, one culture, 220 nl/ml B, exhibited a mutant frequency that was 2.1 times greater than solvent controls, and had a Total Growth of 10%. A dose dependent response was noted. The mutagenic response of the test article in this trial was judged to be equivocal since a reproducible positive response was not observed at any dose.

Overall, the results from these tests indicated that, under the conditions of these tests, test article API 83-06 produced an equivocal response in the presence and absence of exogenous

metabolic activation.

Mouse lymphoma forward mutation assays have been carried out on two other aromatic naphtha samples, as well as another study (API report # 32-32460) for 83-06 in a separate testing facility. The results were:

Sample No. Aromatic Response  
 content (vol. %) with S-9 without S-9  
 83-04 42.1 negative negative  
 83-05 62.5 positive negative  
 83-06 89.8  
 Laboratory 1 positive negative  
 Laboratory 2 (this study) equivocal equivocal

These additional studies are summarized in separate robust study summary records.

**Conclusion:**

The test material API#83-06, CAS # 64741-68-0 was tested in a mouse lymphoma assay with L5178Y TK+/- cell with and without metabolic activation.

The results indicated that the test material was equivocal for causing forward mutations both with and without metabolic activation.

**Reliability/Data Quality - Genetic Toxicity in vitro**

**Reliability:**

Valid Without Restrictions

**Reliability  
Remarks:**

RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor  
Indicator:**

Key

**Reference - Genetic Toxicity in vitro**

**Reference:**

American Petroleum Institute (1986) L5178Y TK +/-Mouse lymphoma assay API 83-06 Heavy catalytically cracked reformed naphtha (CAS 64741-68-0). Study conducted by Microbiological Associates, Inc. Testing facility No. MAT 2420.701 and T2420.701012. API HESD Publ. 33-31641, May 1986.

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Reproductive Toxicity</b>													
<b>Test Substance - Reproductive Toxicity</b>													
<b>Category Chemical:</b>	No CAS Number Provided												
<b>Test Substance:</b>	No CAS Number Provided												
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>												
<b>Category Chemical Result Type:</b>	Read-Across												
<b>Method - Reproductive Toxicity</b>													
<b>Route of Administration:</b>													
<b>Type of Exposure:</b>													
<b>Species:</b>													
<b>Mammalian Strain:</b>													
<b>Gender:</b>													
<b>Number of Animals per Dose:</b>													
<b>Dose:</b>													
<b>Year Study Performed:</b>													
<b>Method/Guideline Followed:</b>													
<b>GLP:</b>													
<b>Exposure Period:</b>													
<b>Frequency of Treatment:</b>													
<b>Post-Exposure Period:</b>													
<b>Method/Guideline and Test Condition Remarks:</b>													
<b>Pre-Mating Exposure / Males:</b>													
<b>Pre-Mating Exposure / Females:</b>													
<b>Test Results - Reproductive Toxicity</b>													
<b>Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):</b>	<table border="1"> <thead> <tr> <th>LOAEL/ LOAEC/ NOAEL/ NOAEC</th> <th>Population</th> <th>Value Description</th> <th>Value/Lower Concentration</th> <th>Upper Concentration</th> <th>Units</th> </tr> </thead> <tbody> <tr> <td>NOAEC</td> <td></td> <td>=</td> <td>13650</td> <td>27750</td> <td>mg/m3</td> </tr> </tbody> </table>	LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units	NOAEC		=	13650	27750	mg/m3
LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units								
NOAEC		=	13650	27750	mg/m3								

<b>Results:</b>	
<b>Results Remarks:</b>	Parental systemic LOAEL and NOAEL values over all studies reflect primarily decreases in body weights at maximum doses.
<b>Conclusion:</b>	<p>Reproductive NOAEC = 13650 mg/m3 to 27750 mg/m3</p> <p>Parental systemic toxicity LOAEC = 13650 - 27750 mg/m3 NOAEC = 2275 - 25000 mg/m3</p> <p>[Parental toxicity values were determined exclusive of male kidney effects indicative of alpha 2-microglobulin mediated nephropathy, also identified as light hydrocarbon induced nephropathy, a species and sex specific syndrome that does not occur in female rats or other species, including humans and is not relevant to humans ((U.S. Environmental Protection Agency. Alpha 2 microglobulin: association with chemically induced renal toxicity and neoplasia in the male rat. 1991. In Risk Assessment Forum. US Government Printing Office, Washington, DC: EPA: 85)]</p>
<b>Reliability/Data Quality - Reproductive Toxicity</b>	
<b>Reliability:</b>	
<b>Reliability Remarks:</b>	
<b>Key Study Sponsor Indicator:</b>	
<b>Reference - Reproductive Toxicity</b>	
<b>Reference:</b>	See Reproductive Toxicity Robust Study Summaries for CAS #, 64741-41-9, 64741-55-5, 64741-63-5, 64741-66-8, 68955-35-1, and 86290-81-5



<b>Reproductive Toxicity</b>	
<b>Test Substance - Reproductive Toxicity</b>	
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Reproductive Toxicity</b>	
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Number of Animals per Dose:</b>	30
<b>Dose:</b>	target chamber concentrations: 5000, 10000 & 20000 mg/m <sup>3</sup> actual chamber concentrations: 5076, 10274, & 20241 mg/m <sup>3</sup>
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	OECD 416
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	91 - 132 Days
<b>Frequency of Treatment:</b>	6 hours/day, seven days/week
<b>Post-Exposure Period:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	Type: Two generation study Premating exposure period: 10 weeks (male and female)  Groups of 30 male and 30 female Sprague Dawley rats were exposed 6 hours/day, seven days/week to volatilized test material at target concentrations of 5000, 10000 and 20000 mg/m <sup>3</sup> . Singly housed animals were exposed for 10 weeks prior to mating. There was then a 3 week mating period and mating was confirmed by either presence of sperm in a vaginal rinse or by the presence of a vaginal plug. Exposure of females was continued until gestation day 20. Exposure was then suspended until post partum day 5 to avoid unduly stressing the dams during birth and was then re-commenced and continued until sacrifice of parental females after weaning. The pups were culled on a random basis to approximately

5/sex/litter. At weaning on postnatal day 28, the F1 pups were selected for the second generation. Among the pups not selected, 3/sex/litter were sacrificed and examined for internal abnormalities. The remainder were examined for external abnormalities, sacrificed and discarded.

The pups selected for F1 were exposed for a 13 week pre-mating period and the for a 3 week mating period as described above. The males were sacrificed at this time and the females continued to be exposed until gestation day 20. As described above exposures were resumed on post partum day 5 and was continued until weaning, when all remaining animals were sacrificed. Other than during the period from gestation day 20 until post partum day 5, all F1 offspring were exposed from conception to sacrifice.

All animals were examined regularly for viability and clinical observations. Body weights and food intakes were also recorded regularly throughout the study. All pups were counted and examined externally on a daily basis and weighed at regular intervals until post natal day 21. F1 pups were examined regularly between post natal days 21 to 28 and were weighed on days 28 and 35. All surviving F1 and F2 pups were examined for developmental landmarks, including pinna detachment, hair growth, incisor eruption, eye opening and the development of the surface righting reflex. Surviving F1 female offspring were monitored for vaginal opening and males were examined for preputial separation. Reproductive parameters evaluated included: male and female fertility indices, male mating index, female fecundity and gestational indices, mean litter size, mean days of gestation, female estrous cycle length and number of females cycling normally. Live birth index, survival index, survival indices (post partum days 1, 4, 7, 14 and 21), viability index at weaning, mean live and dead offspring on day 0, sex ratio at day 0, offspring in-life observations, offspring body weight and offspring gross postmortem findings were also assessed. All animals dying or sacrificed in a moribund condition were necropsied. Culled pups were examined externally but were only necropsied if external evidence warranted it. Randomly selected pups were necropsied and the weight of the following organs was determined: ovaries, liver, adrenals, testes, kidneys, spleen and brain. Additionally a wide range of tissues were taken for histology. Similar evaluations were also carried out on all adults surviving to scheduled sacrifice. Tissues taken from the high dose group and controls were evaluated histologically and since there were no untoward findings, tissues from the lower dose groups were not examined. Samples of sperm from the left distal cauda epididymis were collected from all males at terminal sacrifice for evaluation of sperm parameters. These included assessments of total caudal epididymal sperm numbers, % progressively motile sperm and homogenization resistant spermatid count, %

morphologically normal sperm and % sperm with an identified abnormality. An ovarian examination was carried out in the females that included confirmation of growing follicles and corporea lutea and quantification of primordial oocytes. This was done in the high dose and control groups and since there were no abnormal findings other groups were no evaluated.

**Pre-Mating Exposure / Males:** 10

**Pre-Mating Exposure / Females:** 10

### Test Results - Reproductive Toxicity

**Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):**

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
NOAEC	Parental (F0)	=	20000		mg/m3
NOAEC	Offspring (F1)	=	20000		mg/m3

#### Results:

There were no treatment-related clinical signs, or effects on mortality, body weight or food intake in either parents or pups. Furthermore, there were no treatment-related post mortem findings.

There were no significant differences in absolute organ weights in either males or females from the first parental generation. In the second parental generation, however, there were some statistically significant increases in absolute organ weights, including liver, kidneys and testis in the males and lungs in the females, but none of the differences between controls and the high dose group were statistically significant. In the absence of a clear dose-response relationship the significance of the result was unclear. When expressed as organ/body weight ratios, the only significant difference was seen in male kidney weights in the lowest dose group of the first parental generation and an increase in the highest dose group of the second parental generation. Although this latter may have been treatment related it was not considered to be of clinical importance.

There were no compound-related microscopic changes in any of the reproductive tissues or in the upper or lower respiratory tract from any of the P1 or P2 rats exposed to 20 000 mg/m<sup>3</sup>. The only microscopic changes seen were in the kidneys of males of both generations. There was an exposure related increase in the amount and size of hyaline droplets. In three male rats of the high exposure group from both P1 and P2 animals granular casts were observed in the medullary tubules of the kidneys. These kidney changes and the accompanying weight increases are regarded as a sex and species specific effect and of no relevance for man.

In the first generation there were no differences in mating



index, fecundity, pregnancy or length of gestation. Among the offspring there were no differences in litter size, fraction of live births or sex ratio. Results in the second generation were similar. There were no differences in survival of offspring through weaning in the first generation and in the second generation early survival was slightly higher among the offspring from the exposed dams. There were no differences in the weight of the offspring through weaning in either generation. There were no unusual post mortem observations.

The sperm analysis carried out on both P1 and P2 (F1) males revealed no effects on sperm count, progressive motility or gross appearance.

No effects were found on the estrous cycle length, quantification of primordial oocytes or % females with abnormal cycles in the P1 or P2 generations.

There were no significant differences in incisor eruption, pinna detachment, or surface righting reflex in the F1 or F2 offspring. Hair growth was delayed by just less than one day in males only of the F1 pups and in both sexes of the lowest dose group (approx half day) for the F2 pups. Eye opening was advanced by approximately one-half day for the high dose males of the F2 offspring.

#### Results Remarks:

#### Conclusion:

NOAEC for parental and reproductive toxicity = 7400ppm (20000mg/m<sup>3</sup>), which was the highest dose tested.

There were no treatment related systemic effects in parental females and only the species and sex specific increased hyaline droplet formation consistent with alpha 2-microglobulin mediated nephropathy was observed in kidneys of male rats of both generations. These kidney lesions have been determined not relevant to humans (U.S. Environmental Protection Agency. Alpha 2 microglobulin: association with chemically induced renal toxicity and neoplasia in the male rat. 1991. In Risk Assessment Forum. US Government Printing Office, Washington, DC: EPA: 85) and were excluded in parental NOAEL determination. No reproductive parameters were affected and there were no deleterious effects on offspring survival and growth in either generation. Sperm count and quality in both P1 and P2 (F1) males were comparable in all dose groups.

#### Reliability/Data Quality - Reproductive Toxicity

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP; guideline study

**Key Study Sponsor Indicator:** Key

#### Reference - Reproductive Toxicity

**Reference:** McKee, R. H., Trimmer, G. W., Whitman, F. T., Nessel, C. S., Mackerer, C. R., Hagemann, R., Priston, R. A. J., Riley, A. J., Simpson, B. J. and

Urbanus, J. H.  
(2000) Assessment in rats of the reproductive toxicity of  
gasoline from a  
gasoline vapor recovery unit. Reproductive Toxicology Vol 14, No.  
4, pp 337-353

Posting dates of documents from HPV Challenge web site from which  
data have been  
entered into the HPVIS: 10/28/2003



<b>Reproductive Toxicity</b>	
<b>Test Substance - Reproductive Toxicity</b>	
<b>Category Chemical:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Light Catalytic Reformed Naphtha (LCRN-D) Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a> Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a> )
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Reproductive Toxicity</b>	
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Number of Animals per Dose:</b>	10
<b>Dose:</b>	Target conc.: 750, 2500 & 7500 ppm. (2775, 9250, & 27750 mg/m3) Actual conc.: 750, 2490 & 7480 ppm
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	OECD 421
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	46 - 51 Days
<b>Frequency of Treatment:</b>	6 hours/day, 7 days/week
<b>Post-Exposure Period:</b>	0 Days
<b>Method/Guideline and Test Condition Remarks:</b>	NOTE - this same study is also described in the Developmental Toxicity/Teratogenicity section of HPVIS for this test material. Groups of 10 rats of each sex were exposed to 750, 2500 or 7500 ppm. LCRN-D for 6 hours /day, seven days/week. A group of 10 rats of each sex served as sham treated controls. Parental females were exposed for 14 consecutive days prior to mating, throughout mating and days 0-10 of gestation. Dams and their litters were sacrificed on post partum day 4. Unmated females and parental males were exposed to the test material for 14 days prior to mating, throughout mating and 18 additional days following completion of the mating period. These animals were sacrificed shortly after the last litters were delivered reached post partum day 4.  Mating: Within each group one male was co-housed with the same female until evidence of mating was observed (presence of sperm

in vaginal smear or copulatory plug). The day of mating was designated day 0 of gestation. Following mating, the females were housed individually and continued their exposures to test material until day 19 of gestation. Females not showing evidence of mating following a 14 day mating period continued their exposures. If such a female showed signs of being pregnant, it was removed from the exposure regimen and observed for parturition.

Observations: All parental animals were regularly observed for mortality and gross pharmacologic signs. A physical examination, including palpation for tissue masses was carried out daily 30 mins. after removal from the exposure chambers. Body weights and food consumption were measured throughout the study. From day 20 of gestation, females (pregnant and non-pregnant) were observed for signs of parturition. As soon as possible after delivery, litters were observed for the number of live and dead pups and for any abnormalities. Litters were also observed twice daily for unusual findings and dead pups. On days 0 and 4 of lactation, the pups were counted, weighed and sex was determined by external observation.

Pathology: Males were killed as a group shortly after the last litters delivered had reached day 4 of lactation. Females with litters that reached day 4 of lactation were killed the next day or shortly thereafter. Unmated females and those that did not deliver were killed 23 days after completion of the mating period. At post mortem, a complete macroscopic examination was carried out on all adult animals. The following organs were weighed and organ/body weight ratios were calculated: adrenals, brain, heart, kidneys, liver, lung, spleen, epididymes, testes and thymus. Post mortem examination of females included a count of uterine implantation scars when present.

Pups were sacrificed on day 4 of lactation and underwent a complete macroscopic examination and a determination of sex by internal examination. All pups were preserved with viscera intact. Pups found dead at birth and that died prior to day 4 of lactation also underwent a gross external and internal examination. Dead pups were not eviscerated, the intact pups were preserved.

27 tissues were preserved from all adult animals in all dose groups. Ovaries, testes, epididymes, nose with nasal turbinates, and any grossly observed abnormalities were processed and sections examined histologically for all males and female parental animals in the control and highest dose group. Four sections were prepared and examined microscopically of the skull containing the nasal turbinates. These were area between upper incisor and incisive papilla area between incisive papilla and first palatal ridge area between second palatal ridge and first upper molar area between first upper molar and nasopharynx.

Premating exposure period  
Male: 2 weeks  
Female: 2 weeks

**Pre-Mating  
Exposure / Males:** 14

**Pre-Mating  
Exposure /  
Females:** 14

#### Test Results - Reproductive Toxicity

**Concentration  
(LOAEL/ LOAEC/  
NOAEL/ NOAEC):**

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
LOAEC	Parental (F0)	=	27750		mg/m3
NOAEC	Parental (F0)	=	9250		mg/m3
NOAEC	Offspring (F1)	>=	27750		mg/m3

<b>Results:</b>	<p>All parental animals survived to scheduled sacrifice and no treatment related clinical signs were observed. Except for a slight reduction in body weights in the high dose males there were no other effects on either body weight or food consumption. When compared to the controls, at week 3 the decrease in weight of the high dose males was 3.8% and at week 7 was 7.8. The only treatment related organ weight changes was an increase in relative kidney (15%) and relative liver (5%) weights in the high dose males. No other organ weight changes were recorded. There were no treatment-related microscopic changes in the testes, epididymes, ovaries or nasal turbinates in the animals in the high dose group.</p> <p>Reproductive/fertility effects: All groups had a mating index and a fertility index of 100% and all animals in all groups had mated within 4 days of cohabitation. Delivery and litter data did not demonstrate any effects of treatment see data summarized below.</p> <p>Parameter Dose group (ppm)  0 750 2500 7500</p> <p>Females on study 10 10 10 10  Litters with liveborn 10 10 10 10  Implantation sites 147 154 155 154  Mean 14.7 15.1 15.5 15.4  Pups delivered (total) 145 151 146 145  Liveborn 142 151 143 144  Live birth index (%) 98 100 98 99  Pups dying  Day 0 0 1 1 1  Days 1-4 2 4 0 0  Pups surviving 4 days 140 146 142 143  Viability index (%) 99 97 99 99  pup sex distribution  Day 0 M/F (ratio) 63/79 67/84 9/74 68/76  Day 4 M/F (ratio) 63/77 64/82 68/74 68/75  Pup weight/litter (g)  Day 0 6.0 6.6 6.2 6.1  Day 4 9.3 8.9 9.2 9.6</p> <p>External and internal examination of pups sacrificed on day 4 of lactation resulted in only one pup in a single litter of the control group with abnormalities.</p>
<b>Results Remarks:</b>	NOTE - this same study is also described in the Developmental Toxicity/Teratogenicity section of HPVIS for this test material.
<b>Conclusion:</b>	<p>Parental toxicity LOAEL = 7500ppm (27750 mg/m3) based on slightly decreased body weight and increased relative liver weight; NOAEL parental toxicity = 2500ppm (9250 mg/m3).</p> <p>NOAEL for reproductive performance/ developmental toxicity &gt;= 7500ppm (27750mg/m3), the highest concentration tested.</p>
<b>Reliability/Data Quality - Reproductive Toxicity</b>	
<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	RELIABILITY: GLP; guideline study
<b>Key Study Sponsor Indicator:</b>	Key
<b>Reference - Reproductive Toxicity</b>	
<b>Reference:</b>	<p>Schreiner, C., Bui, Q., Brelia, R., Burnett, D., Koschier, F., Podhasky, P., White, R., Hoffman, G. and Schroder, R. (2000) Toxicity evaluation of petroleum blending streams: reproductive and developmental effects of light catalytic reformed naphtha distillate in rats. J. Tox. and Env. Health, part A., Vol 60, pp 101-116</p>



<b>Reproductive Toxicity</b>																																				
<b>Test Substance - Reproductive Toxicity</b>																																				
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate																																			
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate																																			
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Distillate of light alkylate naphtha (LAN-D)</p> <p>The test material (LAN-D) was prepared to be representative of the fraction of light alkylate naphtha to which man would normally be exposed during normal handling and use. It was obtained by the distillation of light alkylate naphtha (LAN) and collecting that fraction that boiled over the temperature range 78 to 145°F. The sample was analyzed and its composition compared to the light alkylate naphtha from which it was derived (See section 1.1.1. above).</p> <p>The compositions of the distillate and starting material were as follows:</p> <table border="1"> <thead> <tr> <th rowspan="2">Compound</th> <th colspan="2">Weight %</th> </tr> <tr> <th>LAN-D</th> <th>LAN</th> </tr> </thead> <tbody> <tr> <td>n-butane</td> <td>3.42</td> <td>0.84</td> </tr> <tr> <td>isopentane</td> <td>63.59</td> <td>12.61</td> </tr> <tr> <td>n-pentane</td> <td>1.33</td> <td>0.23</td> </tr> <tr> <td>2,3-dimethylbutane</td> <td>22.51</td> <td>4.74</td> </tr> <tr> <td>2-methylpentane</td> <td>6.44</td> <td>1.57</td> </tr> <tr> <td>3-methylpentane</td> <td>2.26</td> <td>0.74</td> </tr> <tr> <td>2,4-dimethylpentane</td> <td>0.29</td> <td>4.09</td> </tr> <tr> <td>2,2,4-trimethylpentane</td> <td>0.06</td> <td>23.92</td> </tr> <tr> <td>2,3,3-trimethylpentane</td> <td>0</td> <td>8.99</td> </tr> <tr> <td>2,3,4-trimethylpentane</td> <td>0</td> <td>11.56</td> </tr> </tbody> </table> <p>Additional compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>)</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>	Compound	Weight %		LAN-D	LAN	n-butane	3.42	0.84	isopentane	63.59	12.61	n-pentane	1.33	0.23	2,3-dimethylbutane	22.51	4.74	2-methylpentane	6.44	1.57	3-methylpentane	2.26	0.74	2,4-dimethylpentane	0.29	4.09	2,2,4-trimethylpentane	0.06	23.92	2,3,3-trimethylpentane	0	8.99	2,3,4-trimethylpentane	0	11.56
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<b>Category Chemical Result Type:</b>	Measured																																			
<b>Method - Reproductive Toxicity</b>																																				
<b>Route of Administration:</b>	Inhalation																																			
<b>Type of Exposure:</b>	Vapor																																			
<b>Species:</b>	Rat																																			
<b>Mammalian Strain:</b>	Sprague-Dawley																																			
<b>Gender:</b>	Both M/F																																			
<b>Number of Animals per Dose:</b>	10																																			
<b>Dose:</b>	Actual 5.09, 12.5 and 24.7 g/m <sup>3</sup> (5090, 12500, & 24700 mg/m <sup>3</sup> ) Target: 5, 12.5, and 25 g/m <sup>3</sup> (1650, 4040, & 8000 ppm)																																			
<b>Year Study Performed:</b>	1995																																			
<b>Method/Guideline Followed:</b>	OECD 421																																			
	Yes																																			

<b>GLP:</b>																			
<b>Exposure Period:</b>	49 - 56 Days																		
<b>Frequency of Treatment:</b>	6 hr/day																		
<b>Post-Exposure Period:</b>	0 Days																		
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Type: One generation study  Premating exposure period: Male and Female: 14 days  Duration of test: Females 7 weeks, males 8 weeks  Method: Adaptation of OECD No. 421</p> <p>The test material was totally vaporized and diluted with air to achieve the desired concentrations for the study. Exposures were conducted in one cubic meter whole-body chambers. Chamber concentrations were monitored three times daily by GC/FID. All animals were housed individually in suspended mesh cages. 10 animals of each sex were exposed 6 hours each day to test material at target concentrations of 5, 12.5 and 25 g/m<sup>3</sup>. The animals were exposed for 6 hours each day. Parental females were exposed for 14 days prior to mating, throughout mating and gestation days 0-19 (7 consecutive weeks). Dams and their litters were sacrificed on postpartum day 4. Parental males were also exposed for 14 days prior to mating, during mating, throughout the female gestation and post partum period and throughout the female necropsy period (8 consecutive weeks). Rats were mated in a 1:1 ratio and females were monitored for evidence of mating by the examination of a vaginal lavage sample for sperm or vaginal plug. If sperm or a vaginal plug were observed the female was considered to be at day 0 of gestation and the male was removed from the female at this stage. If there was no evidence that mating had occurred the pairs were allowed to remain together up to a period of 2 weeks after which time the female was assumed to be pregnant. All animals were observed for clinical signs at least twice daily throughout the study. Body weights and food consumption were recorded throughout the study. Each litter was examined as soon as possible after delivery to establish number and sex of pups, stillbirths, live births and presence of gross abnormalities. Neonatal survival was monitored and all pups were killed postpartum days 4 or 5. Parental females were killed on gestation day 25 if they had not delivered, otherwise they were killed on postpartum days 4 or 5. At necropsy each parental animal was examined macroscopically for structural abnormalities and pathological changes with emphasis on reproductive organs. Additionally the number of implantation sites and corpora lutea of each female were recorded. Lungs, trachea and larynx were removed in their entirety. The right middle lobe of the lung was weighed, the remaining lobes were fixed for subsequent histopathological examination. The testes and epididymes of the males were weighed and then fixed for histological examination as were the ovaries of the females.</p> <p>This study has also been reported in the open literature (Bui et al, 1998) but the open literature publication does not contain as much information as the original laboratory report summarized here.</p>																		
<b>Pre-Mating Exposure / Males:</b>	14																		
<b>Pre-Mating Exposure / Females:</b>	14																		
<b>Test Results - Reproductive Toxicity</b>																			
<b>Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):</b>	<table border="1"> <thead> <tr> <th>LOAEL/ LOAEC/ NOAEL/ NOAEC</th> <th>Population</th> <th>Value Description</th> <th>Value/Lower Concentration</th> <th>Upper Concentration</th> <th>Units</th> </tr> </thead> <tbody> <tr> <td>NOAEC</td> <td>Parental (F0)</td> <td>&gt;</td> <td>24700</td> <td></td> <td>mg/m3</td> </tr> <tr> <td>NOAEC</td> <td>Offspring (F1)</td> <td>&gt;</td> <td>24700</td> <td></td> <td>mg/m3</td> </tr> </tbody> </table>	LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units	NOAEC	Parental (F0)	>	24700		mg/m3	NOAEC	Offspring (F1)	>	24700		mg/m3
	LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units													
	NOAEC	Parental (F0)	>	24700		mg/m3													
NOAEC	Offspring (F1)	>	24700		mg/m3														

**Results:** The chamber concentrations of test material were found to be between 96 and 104% of nominal, the mean highest dose concentration being 24.7 mg/m<sup>3</sup>. The vapor compositions were also found to be similar to that of the parent test material. No parent animals died or were killed during the study and there were no clinical signs. Body weights and food consumption were unaffected by exposure to test material. Results on reproductive capacity and fertility are summarized in the following table.

Parameter	Treatment group (g/m <sup>3</sup> )			
	0	5	12.5	25
Pregnancy (%)	80	80	100	80
Litters with live pups	8	8	9	8
Implantation sites	14.9	16.8	13.9	17.3
Pups delivered	14.4	15.6	14.3	15.6
Live pups/litter	14.4	14.8	13.8	15.5
No. liveborn	115	118	124	124
Live birth index (%)	100	94	96	99
Pups surviving 4 days	113	114	122	123
Viability index (%)	98	97	98	99
Pup wt./Litter day 1	7.2	7.3	7.1	7.1
Pup wt./Litter day 4	10.8	11.1	11.2	10.5

There were no treatment-related findings observed at necropsy. Organ weights were unaffected by treatment and there were no treatment-related histological findings.

**Results Remarks:**

**Conclusion:** No adverse reproductive or systemic effects were induced in treated male and female rats. All pregnant females had comparable delivery data and pups in all groups showed comparable birth weights, weight gain, and viability at postnatal day 4. No histopathological changes were seen at necropsy for adults or offspring, and reproductive organs of adult animals were normal histologically. NOAECs for Reproductive, Developmental, and Parental Systemic toxicities > 25 g/m<sup>3</sup> [24700 mg/m<sup>3</sup>], the highest dose tested.

**Reliability/Data Quality - Reproductive Toxicity**

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP; "adaptation" of guideline study with adequate methods & results description

**Key Study Sponsor Indicator:** Key

**Reference - Reproductive Toxicity**

**Reference:** Bui, Q., Burnett, D. M., Breglia, R. J., Koschier, F. J., Lapadula, E. S., Podhasky, P. I., Schreiner, C. A. and White, R. D. (1998) Toxicity evaluation of petroleum blending streams: reproductive and developmental effects of a distillate from light alkylate naphtha. J. Tox. Env. Health, Part A, Vol 53, pp 121-133

Stonybrook Laboratories Inc (1995) Reproductive/developmental toxicity screening test of light alkylate naphtha distillate in rats Study No. 65874 Stonybrook Laboratories Inc. Princeton, NJ





<b>Reproductive Toxicity</b>	
<b>Test Substance - Reproductive Toxicity</b>	
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a> Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhpv.org">www.petroleumhpv.org</a> )
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Reproductive Toxicity</b>	
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Other
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Number of Animals per Dose:</b>	10
<b>Dose:</b>	Target: 750, 2500 & 7500 ppm. (2700, 9000, & 27000 mg/m3) Actual: 752, 2512 & 7518 ppm
<b>Year Study Performed:</b>	1999
<b>Method/Guideline Followed:</b>	OECD 421
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	47 - 51 Days
<b>Frequency of Treatment:</b>	6 hours/day, 7 days/week
<b>Post-Exposure Period:</b>	0 Days
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Groups of 10 rats of each sex were exposed to 750, 2500 or 7500 ppm. ICRN-D for 6 hours /day, seven days/week. A group of 10 rats of each sex served as sham treated controls. Parental females were exposed for 14 consecutive days prior to mating, throughout mating and days 0-19 of gestation. Dams and their litters were sacrificed on post partum day 4. Unmated females and parental males were exposed to the test material for 14 days prior to mating, throughout mating and for 23 additional days following completion of the mating period. These animals were sacrificed shortly after the last litters were delivered reached post partum day 4.</p> <p>Mating: Within each group one male was co-housed with the same female until evidence of mating was observed (presence of sperm in vaginal smear or copulatory plug). The day of mating was designated day 0 of gestation. Following mating, the females were housed individually and continued their exposures to test material until day 19 of gestation. Females not showing evidence of mating following a 14 day mating period continued their</p>

exposures. If such a female showed signs of being pregnant it was removed from the exposure regimen and observed for parturition.

Observations: All parental animals were regularly observed for mortality and gross pharmacologic signs. A physical examination, including palpation for tissue masses was carried out daily 30 mins. after removal from the exposure chambers. Body weights and food consumption were measured throughout the study. From day 20 of gestation, females (pregnant and non-pregnant) were observed for signs of parturition. As soon as possible after delivery, litters were observed for the number of live and dead pups and for any abnormalities. Litters were also observed twice daily for unusual findings and dead pups. On days 0 and 4 of lactation, the pups were counted, weighed and their sex was determined by external observation.

Pathology: Males were killed as a group shortly after the last litters delivered had reached day 4 of lactation. Females with litters that reached day 4 of lactation were killed the next day or shortly thereafter. Unmated females and those that did not deliver were killed 23 days after completion of the mating period. At post mortem, a complete macroscopic examination was carried out on all adult animals. The following organs were weighed and organ/body weight ratios were calculated: adrenals, brain, heart, kidneys, liver, lung, spleen, epididymes, testes and thymus. Post mortem examination of females included a count of uterine implantation scars when present.

Pups were sacrificed on day 4 of lactation and underwent a complete macroscopic examination and a determination of sex by internal examination. All pups were preserved with viscera intact. Pups found dead at birth and that died prior to day 4 of lactation also underwent a gross external and internal examination. Dead pups were not eviscerated, but were preserved intact. 27 tissues were preserved from all adult animals in all dose groups. Ovaries, testes, epididymes, nose with nasal turbinates, and any grossly observed abnormalities were processed and sections examined histologically for all males and female parental animals in the control and highest dose group. Four sections were prepared and examined microscopically of the skull containing the nasal turbinates. These were:  
 area between upper incisor and incisive papilla  
 area between incisive papilla and first palatal ridge  
 area between second palatal ridge and first upper molar  
 area between first upper molar and nasopharynx.

The LCCN-D was wholly vaporized using a countercurrent volatilization chamber. The volatilized LCCN-D was diluted with air to achieve the desired atmospheric concentrations. The target and actual chamber concentrations are as follows:

Target concentration (ppm)	Actual concentration (ppm)	Total concentration (µg/m <sup>3</sup> )
0	0	4.8 ± 4.5
750	752 ± 35	4.1 ± 3.5
2500	2512 ± 66	3.7 ± 3.3
7500	7518 ± 146	4.1 ± 3.9

The LCCN-D was characterized pre-and post study. The results (given in weight %) of the characterization are as follows:

Component	LCCN-D Study	Study	Study
liquid start			
n-Butane	0.43	0.40	0.43
n-Pentane	3.28	3.23	3.24
iso-Pentane	15.22	15.87	15.74
1-Pentane	2.82	2.64	2.70
2-Pentene (trans)	7.30	6.96	7.02
2-Pentene (cis)	4.12	3.96	4.02
2-Methyl-2-butene	10.81	10.37	10.43
2-Methyl-1-butene	5.60	5.23	5.33
Cyclopentane	1.34	1.28	1.31
n-Hexane	1.56	1.58	1.56
Methylcyclopentane	1.95	2.15	2.12
2,3-Dimethylbutane	1.36	2.30	2.27
2-Methylpentane	5.57	6.28	6.15
3-Methylpentane	3.08	3.18	3.12
1-Methylcyclopentane	1.23	1.24	1.25
Benzene	1.15	1.30	1.20
2-Methylhexane	1.09	1.20	1.17

**Pre-Mating  
Exposure / Males:** 14

**Pre-Mating  
Exposure /  
Females:** 14

### Test Results - Reproductive Toxicity

**Concentration  
(LOAEL/ LOAEC/  
NOAEL/ NOAEC):**

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
LOAEC	Parental (F0)	=	27000		mg/m3
NOAEC	Parental (F0)	=	9000		mg/m3
NOAEC	Offspring (F1)	=	27000		mg/m3

**Results:**

All animals survived to scheduled sacrifice. Red staining on the snout was seen with increasing frequency in the mid and high dose animals of both sexes throughout the study. Microscopic examination of the nasal turbinates of the sham-exposed and high dose animals did not reveal any significant changes.

Although all treated groups gained slightly less weight than the sham treated controls the differences were not statistically significant. Food consumption was comparable in all groups. Apart from those listed below, absolute and relative organ weights were unaffected by treatment.

**High dose Males:**

Absolute kidney weight increased (18%)  
Relative kidney weights increased (24%)  
Relative liver weights increased (15%)

**High dose females:**

Absolute spleen weights increased by (19%)  
Relative spleen weights increased by (19%)

At necropsy, no organs appeared abnormal. Microscopic examination of kidneys from one high dose male with a dilated renal pelvis at necropsy revealed hyaline droplet formation and tubular dilatation of tubules in the cortico-medullary junction. This finding is consistent with male-rat-specific light hydrocarbon nephropathy. No test-related microscopic changes were observed in the testes or epididymes of adult male rats or ovaries of adult female rats in the high dose group.

Reproductive/fertility effects: All groups had a fertility index of >90% and all groups had a live birth index greater than or equal to 98%. Data are summarized below.

Parameter Dose group (ppm)  
0 750 2500 7500  
Females on study 10 10 10 10  
Litters with liveborn 9 8 9 10  
Implantation sites 155 126 139 160  
Mean 17.2 15.8 15.4 16  
Pups delivered (total) 149 110 132 152  
Liveborn 149 108 131 151  
Live birth index (%) 100 98 99 99  
Pups dying  
Day 0 0 2 1 1  
Days 1-4 4 2 2 1  
Pups surviving 4 days 145 106 129 150  
Viability index (%) 97 98 99 99  
pup sex distribution  
Day 0 M/F (ratio) 72/77 50/58 65/66 87/64  
Day 4 M/F (ratio) 72/73 49/57 65/64 87/63  
Pup weight/litter (g)  
Day 0 6.3 6.6 6.4 6.4  
Day 4 9.9 10.8 10.1 10.3

External and internal examination of pups sacrificed on day 4 of

lactation were unremarkable.

**Results Remarks:**

All groups had a fertility index of >90% and a live birth index greater than or equal to 98%. Offspring showed comparable body weights, weight gain, and viability index at postnatal day 4. Parental male rats had increased kidney weights and relative liver weights at the highest dose, and high dose females had increased spleen weights. Reproductive organs and nasal turbinates from high dose and control animals were examined by a pathologist and no histological changes were observed in tissue from treated rats.

Male rat kidney effects were consistent with alpha 2-microglobulin mediated nephropathy, also identified as light hydrocarbon induced nephropathy, a species and sex specific syndrome that does not occur in female rats or other species, including humans and is not relevant to humans (U.S. Environmental Protection Agency. Alpha 2 microglobulin: association with chemically induced renal toxicity and neoplasia in the male rat. 1991. In Risk Assessment Forum. US Government Printing Office, Washington, DC: EPA: 85).

**Conclusion:**

LOAEL parental toxicity = 7500ppm [27000mg/m3]  
 NOAEL parental toxicity = 2500ppm [9000mg/m3];  
 NOAEL reproductive performance/ developmental toxicity = 7500ppm [27000mg/m3]

**Reliability/Data Quality - Reproductive Toxicity****Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

RELIABILITY: GLP; guideline study

**Key Study Sponsor Indicator:**

Key

**Reference - Reproductive Toxicity****Reference:**

Schreiner, C. Bui, Q., Burnett, D., Koschier, F., Podhasky, P., Lapadula, E., White, R. and Schroeder, R. E. (1999) Toxicity evaluation of petroleum blending streams: reproductive and developmental effects of light catalytic cracked naphtha distillate in rats. J. Toxicol. and Env. Health, Part A, Vol 58, pp 365-382

Posting dates of documents from HPV Challenge web site from which data have been entered into the HPVIS: 10/28/2003



<b>Reproductive Toxicity</b>																																							
<b>Test Substance - Reproductive Toxicity</b>																																							
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline																																						
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline																																						
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p> <p>[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]</p> <p>Unleaded baseline gasoline API 99-01 Vapor Condensate Test material is a complex mixture of volatile hydrocarbons. The purity of mixture is 100% and stable based on analysis of chamber atmospheres.</p> <p>Representative Components [98.8%] monitored in Study:</p> <table border="1"> <thead> <tr> <th>COMPONENT</th> <th>AREA %</th> </tr> </thead> <tbody> <tr><td>Isobutane</td><td>2.70</td></tr> <tr><td>n-butane</td><td>12.78</td></tr> <tr><td>3-methyl-1-butene</td><td>0.41</td></tr> <tr><td>Isopentane</td><td>36.50</td></tr> <tr><td>n-pentane</td><td>9.36</td></tr> <tr><td>Trans-2-pentene</td><td>3.60</td></tr> <tr><td>2,3-dimethylbutane</td><td>1.75</td></tr> <tr><td>2-methylpentane</td><td>7.25</td></tr> <tr><td>3-methylpentane</td><td>4.27</td></tr> <tr><td>n-hexane</td><td>3.62</td></tr> <tr><td>Methylcyclopentane</td><td>1.87</td></tr> <tr><td>2,4-dimethylpentane</td><td>1.36</td></tr> <tr><td>Benzene</td><td>2.75</td></tr> <tr><td>2-methylhexane</td><td>1.73</td></tr> <tr><td>2,3-dimethylpentane</td><td>1.52</td></tr> <tr><td>3-methylhexane</td><td>1.73</td></tr> <tr><td>Isooctane</td><td>1.92</td></tr> <tr><td>Toluene</td><td>3.91</td></tr> </tbody> </table> <p>Additional compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>)</p>	COMPONENT	AREA %	Isobutane	2.70	n-butane	12.78	3-methyl-1-butene	0.41	Isopentane	36.50	n-pentane	9.36	Trans-2-pentene	3.60	2,3-dimethylbutane	1.75	2-methylpentane	7.25	3-methylpentane	4.27	n-hexane	3.62	Methylcyclopentane	1.87	2,4-dimethylpentane	1.36	Benzene	2.75	2-methylhexane	1.73	2,3-dimethylpentane	1.52	3-methylhexane	1.73	Isooctane	1.92	Toluene	3.91
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<b>Method - Reproductive Toxicity</b>																																							
<b>Route of Administration:</b>	Inhalation																																						
<b>Type of Exposure:</b>	Vapor																																						
<b>Species:</b>	Rat																																						
<b>Mammalian Strain:</b>	Sprague-Dawley																																						
<b>Gender:</b>	Both M/F																																						
<b>Number of Animals per Dose:</b>	26																																						
<b>Dose:</b>	26 males, 26 females/group																																						
	Target: 0, 2000, 10,000, and 20,000mg/m3 Actual: 0, 2014, 10,139, and 20,004 mg/m3																																						
<b>Year Study Performed:</b>	2006																																						

<b>Method/Guideline Followed:</b>	EPA 870.3800
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	112 - 133 Other
<b>Frequency of Treatment:</b>	6 hours/day, 7 days/week
<b>Post-Exposure Period:</b>	0
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Baseline Gasoline Vapor Condensate was administered via whole-body exposures to Sprague Dawley rats over 2 generations at target concentrations of 2000, 10000 and 20000 mg/m3 for 6 hours/day, 7 days/week. In addition, an Air Control group received nitrogen-enriched air only while in chamber. Exposure levels were determined using an infra-red spectrophotometer 4 times per chamber per day. The test substance's major components were assayed once per chamber per week. Particle size distribution measurements were also made once per chamber per week using a TSI Aerodynamic Particle Sizer. Viability checks were performed twice daily to check for mortality and signs of severe toxic or pharmacologic effects. Physical observations and body weights were collected twice pretest (P0 generation) and at least weekly during the study (P0 and F1). Feed consumption was measured beginning the week prior to treatment initiation (P0 generation) and at least weekly during the study (P0 and F1). For P0 and F1 dams, body weight and food consumption were measured on Gestation Days [GD] 0, 7, 14, 20 and on Lactation Days [LD] 1,4,7,14,21 and 28. After approximately 16 weeks of exposure, all parental male animals (P0 and F1) were sacrificed and all parental females (P0 and F1) were sacrificed on their respective LD28. Females that failed to mate were sacrificed 25 days after the end of the mating period and females with confirmed mating but without delivery were sacrificed on presumed GD25. Selected organs [adrenals, brain, heart, liver, lungs, kidneys, spleen, thymus, ovaries, uterus testes, seminal vesicles, prostate, epididymides] were weighed and organ/body weight and organ/brain weight ratios calculated. Macroscopic examinations were performed on all parental rats and histological evaluations of the tissue samples from the weighed organs of 10 randomly selected rats in the Control and 20000mg/m3 groups were performed. Reproductive organs from all male and bred female rats in control and high dose groups were evaluated. Sperm evaluations included motility, testicular homogenization-resistant sperm and cauda epididymal sperm count, sperm morphology in the cauda epididymis. Ovary histopathology included evaluation of the primordial follicle population, number of growing follicles and corpora lutea. Mating: Vaginal smears were taken daily for each female beginning three weeks prior to cohabitation for P0 and F1 rats and continuing until there was evidence of mating or until the 14-day mating period was ended. Following 10 weeks</p>

pre-mating exposure, one male and one female from the same group were mated overnight until evidence of mating was observed or 14 days had elapsed. Animals were not paired during the daily exposure period. During mating of F1 generation, male and female littermates were never paired together. At weaning of each F1 litter on Lactation day 28, one pup/sex/litter was chosen at random to continue with exposure to BGVC as the F1 parental generation. When less than 26 litters were available in a group, additional pups from other litters within the group were selected at random to make up 26 mating pairs/group. Parturition and Lactation: On Day 18 of gestation exposure was ended and each female was transferred to a plastic shoebox with bedding material and observed for evidence of parturition. The day on which parturition was observed was Day 0 of Lactation. These females were not exposed from GD19 [P0 and F1 dams] until exposure was resumed on LD5 to weaning at LD28. Pups (F1 and F2 generations) were observed as soon as possible after delivery for sex, number of live and dead pups and pup abnormalities. Pup dead at delivery were identified as stillborn or liveborn found dead based on lung floatation evaluation. Thereafter litters were observed twice daily. On LD 4, F1 litters with more than 10 pups were randomly culled to 10 pups with sex distribution equalized if possible. Pups were examined and weighed on LD1 (delivery day), 4 (pre-culled), 7, 14, 21 and 28. At weaning one pup/sex/group was selected for mating to produce the F2 generation. F1 pups [5/sex/group/assessment] not selected for F1 mating were evaluated for standard Tier 2 neuropathology [40 CFR79.66] or for GFAP assessments [40 CFR79.67] on postpartum day 28 [Results of GFAP study are reported in separate Neurotoxicity Robust Summary]. The remaining pups were sacrificed. Three pups/sex/litter in each group (F1 and F2) were selected from macroscopic examination and selected organs [brain, spleen, thymus] were weighed from one pup /sex/litter.

Statistical methods: For continuous data [Body weights, Body weight change, Feed consumption, Organ weight data, Gestation length, Pup body weights, Number of pups (live, dead, total), Mean age-to-criteria for vaginal opening and preputial separation], mean values of all exposure groups were compared to the mean value for the control group at each time interval. Evaluation of equality of group means was made with standard one-way analysis of variance (ANOVA) using the F ratio followed by Dunnett's if needed.

Sperm and ovary analysis: The following parameters were analysed statistically: Mean sperm count (testicular sperm count and caudal epididymal sperm count) and motility data and numbers of primordial and growing follicles by ovary and total. If a significant difference occurred ( $p < 0.05$ ) between groups using the nonparametric Kruskal-Wallis test, the Wilcoxon (Mann-Whitney U) test was used for pair-wise comparisons of each treated group to the vehicle control group. Incidence data [Mortality, Mating Indices, Pregnancy rates, Male fertility Indices, Live birth indices, and P up viability indices (Days

0-4) and lactation indices (Days 4-28)] were analyzed using the Chi-square test (2 x n). If Chi-square analysis was not significant, no additional analyses were performed. If Chi-square is significant, a Fisher Exact Test with Bonferroni correction was performed to identify differences between the groups.

**Pre-Mating Exposure / Males:** 10

**Pre-Mating Exposure / Females:** 10

### Test Results - Reproductive Toxicity

**Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):**

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
NOAEC		>=	20000		mg/m3
NOAEC	Female (Maternal)	=	10000		mg/m3
NOAEC	Offspring (F1)	=	10000		mg/m3
LOAEC	Female (Maternal)	=	20000		mg/m3
LOAEC	Offspring (F1)	=	20000		mg/m3

**Results:** NOAEC Reproductive Toxicity greater than or equal to 20000 mg/m3  
 LOAEC Systemic Toxicity for P0 females and F1 males = 20000 mg/m3  
 NOAEC Systemic Toxicity for P0 females and F1 males = 10000 mg/m3

**Results Remarks:** Exposure conditions: The analytically measured exposure levels of the airborne test substance were reasonably close to the targeted exposure levels. Chamber environmental conditions averaged 24°C and 43% relative humidity. Particle sizing results indicated that the atmospheres were essentially vapor only. Analysis of the major components in the neat test substance and the test atmospheres showed a reasonably close comparison between the neat test substance and the vaporized test substance. This data demonstrated that the test animals were exposed, as expected, to all of the major components of the test substance in their reasonably proper proportion. The data was consistent from week-to-week during the study indicating stability of the test substance and the atmosphere generation techniques. Parental data (P0 and F1 generations): There was no effect of treatment on survival. The test animals were generally unremarkable in-chamber during the exposure periods and during the non-exposure periods (afternoon evaluations) during the pre-mating period in both sexes, the mating/postmating period in the male rats, and the gestation and lactation periods in the female rats. There were exposure-related differences in body weights or weight changes in the test substance exposed animals compared to the Air Control animals. These



differences were decreases in weight gain in the P0 female rats in the 20000mg/m<sup>3</sup> group during the latter 3 weeks of the pre-mating period and in the F1 male rats in the 20000 mg/m<sup>3</sup> group during the initial 8 weeks of the pre-mating period. There were no exposure-related differences in feed consumption in the test substance exposed animals compared to the Air Control animals. There were no exposure-related differences in estrous cycle data (as measured by cycle length and number of estrous cycles) in the test substance exposed animals compared to the Air Control animals. Mating indices for the male rats treated with the test substance were comparable to the Air Control group. Mating, fertility and gestation indices for the female rats treated with the test substance were comparable to the Air Control group. The pregnancy rates for the Air Control, 2000, 10000 and 20000 mg/m<sup>3</sup> groups were 96.0%, 96.2%, 92.3% and 100%, respectively, for the P0 animals and 100%, 100%, 91.7% and 100%, respectively, for the F1 animals. Treatment with the test substance also resulted in no statistically significant differences in most other reproductive parameters including the percent of females completing delivery and the duration of gestation, when compared to the Air Control group. There were no exposure-related differences in body weights or weight changes in the test substance exposed animals compared to the Air Control animals during the gestation and lactation periods. There were no exposure-related differences in feed consumption during the gestation and lactation periods in the test substance exposed animals compared to the Air Control animals. Treatment with the test substance resulted in no statistically significant differences in all parturition parameters including the total number of pups delivered, the number of pups dying, the viability (4 day survival) and lactation (28 day survival) indices, the number of implantation sites per litter, the sex ratio and the number of live pups/litter, when compared to the Air Control group. There were no exposure-related temporal differences in males showing preputial separation and females showing vaginal opening in the F1 pups weaned from test substance exposed animals compared to the F1 pups weaned from Air Control animals. There were no exposure-related differences in macroscopic postmortem evaluations in the test substance exposed animals compared to the Air Control animals. Exposure-related effects on organ weights included statistically significant increases in kidney weights (absolute and relative to body and brain weight) at the 2 higher exposure levels in the P0 and F1 males and at the highest exposure level in the P0 females. These differences for the males (but not the females) were consistent with the microscopic findings discussed below. The percent sperm motility, caudal epididymal and homogenization-resistant testicular sperm counts, sperm morphology, and primordial and growing follicle counts, as individual ovaries and total per animal, were not affected by treatment with

test substance at an exposure level of 20,000 mg/m<sup>3</sup>. Microscopic findings that were considered exposure-related were found only in the kidneys of male animals exposed to 20,000 mg/m<sup>3</sup> of test substance and are consistent with hyaline droplet nephropathy, attributable to accumulation of alpha-2 microglobulin within renal tubular epithelial cells. This species- and gender-specific change has been well documented in male rats exposed to a variety of hydrocarbon compounds and is not considered relevant to humans. No test substance related microscopic changes were noted in male and female reproductive organs or other protocol-specified tissues in this study. Pup data (F1 & F2 generations): There were no exposure-related differences in body weights and weight changes in the pups from test substance exposed animals compared to the pups from Air Control animals. The pups were unremarkable during the lactation period. There were no exposure-related differences in macroscopic postmortem evaluations and organ weights in the pups from test substance exposed animals compared to the pups from Air Control animals. No adverse neuropathological findings were observed.

**Conclusion:** Exposure of rats to 2000, 10000 and 20000mg/m<sup>3</sup> of vapor of test substance resulted in decreased body weight gains in the P0 females and F1 males prior to mating in the 20000 mg/m<sup>3</sup> exposed group. Increases in kidney weights in parental male animals exposed to the 2 higher exposure levels of vapor were consistent with hydrocarbon nephropathy seen in these animals, a finding has been generally accepted not to be relevant to human risk assessment (US EPA, 1991). There was no effect at any of the exposure levels on reproductive performance in the study, including mating, fertility, parturition, lactation, offspring survival and development or maturation, in either the P0 or F1 generations. There was no evidence of any neuropathology in F1 pups as a result of the exposures [GFAP results reported in separate Robust summary]. The NOAEL for systemic toxicity [excluding kidney effects in male rats] is 10000mg/m<sup>3</sup>. The NOAEL for neuropathology in F1 animals is >20,000mg/m<sup>3</sup>. The Reproductive NOAEL is greater than or equal to 20,000mg/m<sup>3</sup>.

#### Reliability/Data Quality - Reproductive Toxicity

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** HPV Supporting study from Section 211(b) Testing Consortium, Fuels and Fuel Additives Health Effects Testing Regulation, administered by API, Washington DC

RELIABILITY: GLP; guideline study

**Key Study Sponsor Indicator:** Key

#### Reference - Reproductive Toxicity

**Reference:** Baseline Gasoline Vapor Condensate: A Two-Generation Whole Body Inhalation Reproductive Study in Rats. 2006. HLS Study No. 00-4207. Huntingdon Life

Sciences Laboratories, East Millstone, NJ  
US EPA 1991. Alpha 2 microglobulin: Association of chemically  
induced renal  
toxicity and neoplasia in male rats. In Risk Assessment Forum,  
p.85. US Govt  
Printing Office, Washington DC



<b>Reproductive Toxicity</b>	
<b>Test Substance - Reproductive Toxicity</b>	
<b>Category Chemical:</b>	(64741-41-9) Naphtha, petroleum, heavy straight-run
<b>Test Substance:</b>	(64741-41-9) Naphtha, petroleum, heavy straight-run
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Naphtha, petroleum, heavy straight-run, Colorless liquid. MW 111.25. The test substance is a mixture that contains approximately 225 volatile hydrocarbons. The purity of the mixture is 100%</p> <p>Stable based on analyses of chamber atmosphere.</p> <p>12 Representative Components monitored in Study</p> <p>Component Volume %</p> <p>2-Methyl C6 + C7-olefin 4.50</p> <p>3-Methylhexane 3.52</p> <p>t-1,3-Dimethylcyclopentane 1.45</p> <p>t-1,2-Dimethylcyclopentane 1.61</p> <p>n-Heptane 7.23</p> <p>Methylcyclohexane 6.76</p> <p>Toluene 3.44</p> <p>2-Methylheptane 3.25</p> <p>n-Octane 5.81</p> <p>Ethylcyclohexane 1.95</p> <p>m-Xylene 1.71</p> <p>n-Nonane 4.47</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Reproductive Toxicity</b>	
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Number of Animals per Dose:</b>	12
<b>Dose:</b>	<p>Target: 0, 100, 500, 3000ppm (0, 455, 2275, 13650mg/m3)</p> <p>Actual: 0, 100, 520, 2950ppm (0, 455, 2366, 13423mg/m3)</p> <p>The mean concentrations (SE) representing the total area for the approximately 225 components contained in the test substance over the test period were 100(0.8), 500(2.0), and 3000(8.3)ppm in chambers targeted at 100, 500, and 3000ppm, respectively. Results from the cryogenic GC analysis indicated that the components were present in the chamber atmosphere within expected concentrations</p>
<b>Year Study Performed:</b>	2008
<b>Method/Guideline Followed:</b>	OECD 422
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	6hr/d, 7d/wk

<b>Frequency of Treatment:</b>	
<b>Post-Exposure Period:</b>	4 Days
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Concentrations of Naphtha vapor were generated by flash evaporation of the test material. An air control group was also evaluated using a similar generation apparatus; however, no test material was supplied to this vapor generator. Vapor concentrations of Naphtha were measured by gas chromatography (GC) using the area sum function and integrating all of the eluted peaks. Additional air samples were collected weekly and analyzed for 12 of the larger, most representative components of the test substance using a cryogenic GC. Temperature, humidity, and airflow were also recorded periodically during each exposure day. Exposures were conducted for 6 hours per day, 7 days per week.</p> <p>Groups of 12 young, adult, male Crl:CD(SD) rats were exposed to atmospheres containing 0, 100, 500, or 3000ppm of Naphtha for 30 days. Satellite groups of 12 young, nulliparous, non-pregnant female rats were exposed to 0, 100, 500, or 3000ppm during a pre-mating period of approximately 2 weeks, a cohabitation period of up to 2 weeks, and a gestation period of approximately 3 weeks. Following the 2 week pre-mating period, each satellite female was paired with a male of the same respective dosage group during an approximately 2 week cohabitation period. Presumed pregnant females were exposed from gestation day [GD] 0-19 but were not exposed after gestation day 19, or during the approximately 4-day lactation period [LD]. Females without evidence of mating continued to be exposed for 26 days after the end of the cohabitation period.</p> <p>Body weights, clinical signs, and food consumption were recorded throughout the study. Body weight data were collected weekly for males, and satellite females without evidence of copulation. Satellite females were weighed weekly during pre-mating and cohabitation, on GD0, 7, 14, 21 and on LD0 and 4. Food consumption data were collected at the same intervals except for non-bred satellite females post cohabitation. After approximately 30 days of exposure, blood samples were collected from all males for measurement of haematology and clinical chemistry parameters. An abbreviated neurobehavioral evaluation was conducted on all males, and satellite females prior to test substance administration in order to obtain baseline measurements, and again during week 4 in the morning prior to daily exposure for males and on lactation day 4 for satellite females with litters. Neurobehavioral evaluation consisted of motor activity and a modified Functional Observational Battery [FOB] of open field (approach and touch response, auditory response and tail pinch), papillary response, and fore and hind limb grip strength. Males were sacrificed after 30 days of exposure, organs (liver, kidneys, lungs, adrenal glands, thymus, brain, spleen, heart, testes with epididymides, prostate, were weighed, and 36 selected tissues were evaluated microscopically. On postpartum day 4, lactating females and offspring were sacrificed, organs (liver, kidneys, lungs, ovaries with oviducts and uterus with cervix) were weighed, and reproductive organs were evaluated microscopically. Offspring were evaluated for external abnormalities.</p> <p>Statistical analysis: Preliminary statistical analyses included Levene's test for homogeneity and Shapiro-Wilk test for normality, followed by one-way analysis of variance [ANOVA] and Dunnett/Tamhane-Dunnett's test or Kruskal-Wallis and Dunn's test as appropriate. Analysis of covariance [ANCOVA] and Dunnett-Hsu, or non-parametric ANCOVA was used for pup sex ratio and pup weights. Repeated measure ANOVA with Linear contrasts or Jonckheere-Terpstra trend test was used for motor activity and grip strength.</p>
<b>Pre-Mating Exposure / Males:</b>	30
<b>Pre-Mating Exposure / Females:</b>	14
<b>Test Results - Reproductive Toxicity</b>	

Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):	LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
	NOAEC		>=	13423		mg/m3
	NOAEC	Female (Maternal)	=	2366		mg/m3
	NOAEC	Male (Paternal)	=	2366		mg/m3

**Results:**

A NOAEL for reproductive performance was 3000ppm (13650mg/m3).

The systemic NOAEL of 500ppm (2275 mg/m3) reported in the Subchronic Toxicity robust summary for (parental) males were based on hypertrophy of thyroid follicular epithelium observed in 3000ppm males.

The systemic NOAEC for dams was 500 ppm (2275 mg/m3) due to test substance related effects on body weight and weight gain observed in 3000ppm females during the three-week gestation period.

**Results Remarks:**

Details of systemic effects in males are presented in detail in the Repeated Dose Subchronic Toxicity Robust Summary for this OECD 422 study. Slightly decreased body weight and/or weight gain occurred in 3000ppm males; however the magnitude of the effect was not considered adverse. Successful reproductive performance was observed for all males although of one high dose female failed to mate but the other female bred to that male became pregnant. No adverse effects were seen in histo- pathological evaluation of reproductive organs. A NOAEL for reproductive performance in males was 3000ppm (13650mg/m3). The systemic NOAEL of 500ppm (2275 mg/m3) reported in the Subchronic Toxicity robust summary for males were based on hypertrophy of thyroid follicular epithelium observed in 3000ppm males.

Mortality did not occur at any exposure concentration. Test substance-related increases in the incidence of stained and wet fur in males and satellite females were observed in the 3000ppm group; however, this did not adversely impact the health of the animals. Satellite females showed no significant test substance related effects on body weight or weight gain during pre-mating or mating. Test substance related effects on body weight and weight gain were observed in 3000ppm females during the three-week gestation period. Body weight at GD 21 was 7% lower than controls, and weight gain from GD0-21 was 14% lower than controls. The lower body weight on GD21 correlates with the statistically significant lower weight on LD0 and was considered an adverse effect. The statistically significant lower maternal body weight at LD0 (7%) correlated with the lower weight trend in high dose females during gestation. The overall weight gain from LD0-4 was comparable to controls although the absolute weight of high dose females did not fully recover to control levels but was not significantly lower. No adverse effect on body weight or weight gain were seen in any animals in the 500 or 100 ppm groups. Decreases in food consumption correlated with decreased body weight and weight gains for animals in the 3000ppm group. Food efficiency was slightly decreased in 3000ppm satellite females during pre-mating and gestation but not during LD0-4. No test-substance related effects were seen on food consumption or efficiency in 100 or 500ppm groups.

No adverse clinical chemistry, haematologic, or histopathology effects compared to controls were seen in females with litters at Day 4 of lactation.

Reproductive Toxicology: There were no significant test substance related differences in mean number of pregnant animals, number of animals delivering, mating index, fertility index, pre-coital interval, gestation length, number of corpora lutea, number of implantation sites or percent of post implantation loss for any exposure group. No test substance-related differences were observed in number of fetuses born, live born index, viability index, sex ratio incidence, or clinical observations or mean pup body weight on postnatal days 0 or 4. One female in the 3000ppm group failed to mate. The Mating Indices were 100% for controls and groups 100 and 500 and 91.7% in the 3000ppm group. The duration of gestation was 22 days for controls and 21.9 days for treated groups. There were 12 viable litters in controls, 100 and 500ppm groups and 11 litters in the 3000ppm group. One dam [#434]

in the 3000ppm group was not identified as pregnant and delivered her litter during exposure in the chamber. Her mating date could not be determined, pups were small and 5/12 pups died between lactation days 0-4. Liveborn index was 100% in all groups. There were no statistically significant differences in average number of pups born alive: 15.3, 14.3, 15.1 and 13.8 pups in control, 100, 500 and 3000ppm groups respectively. By LD4, one pup each died in control and 100ppm groups, none died in 500ppm group and the only deaths in the 3000ppm group were the 5/12 pups indicated above, all other litters at 3000ppm had 100% survival. Viability Indices at LD4 were 99.5%, 99.5%, 100%, and 100% in control, 100, 500 and 3000ppm groups respectively. Combined average pup weights at birth were 6.5g, 6.6g, 6.4g, and 6.2g and 10.3g, 10.6g, 10.0g and 9.7g in control, 100, 500 and 3000ppm groups respectively. When LD4 pup weights from dam #434 were omitted from the mean and offspring body weights re-analyzed, the 3000ppm weights were comparable to controls. Pup weight gains from LD0 to LD4 for all treated groups were comparable to controls. The NOAEL for reproductive toxicity was 3000ppm (13650mg/m3), the highest concentration tested.

Neurobehavioral Toxicology: There were no test substance-attributed or statistically significant differences in forelimb or hindlimb grip strength in satellite lactating females at any concentration of the test substance. Pupillary constriction response and open field parameters consisting of approach and touch response, auditory response and tail pinch response were comparable for all treated groups and controls. Motor activity [duration of movement and number of movements] did not demonstrate any test substance related adverse effects. The NOAEL for neurobehavioral toxicity in lactating females was 3000ppm (13650mg/m3), the highest concentration tested.

**Conclusion:**

Exposure to this heavy straight run naphtha at 3000ppm induced some systemic toxicity in breeding female rats expressed as reduced body weight and weight gain, and slight decreased food consumption at 3000ppm (13650mg/m3). No significant adverse reproductive effects were seen for breeding males. This naphtha did not induce reproductive, or neurotoxic adverse effects in maternal animals and is not considered a reproductive/developmental or a maternal neurobehavioral toxicant.

**Reliability/Data Quality - Reproductive Toxicity****Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

RELIABILITY: GLP; guideline study

**Key Study Sponsor Indicator:**

Key

**Reference - Reproductive Toxicity****Reference:**

API (American Petroleum Institute) 2008a. OECD 422 inhalation combined repeated dose toxicity study with the reproductive/developmental toxicity screening test of Heavy straight run naphtha [CAS # 64741-41-9]. Haskell Laboratories, Project ID: DuPont -18331. Wilmington, DE



Developmental Toxicity/Teratogenicity																															
Test Substance - Developmental Toxicity/Teratogenicity																															
<b>Category Chemical:</b>	No CAS Number Provided																														
<b>Test Substance:</b>	No CAS Number Provided																														
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>																														
<b>Category Chemical Result Type:</b>	Read-Across																														
Method - Developmental Toxicity/Teratogenicity																															
<b>Route of Administration:</b>	Inhalation																														
<b>Type of Exposure:</b>																															
<b>Species:</b>																															
<b>Mammalian Strain:</b>																															
<b>Gender:</b>																															
<b>Number of Animals per Dose:</b>																															
<b>Dose:</b>																															
<b>Year Study Performed:</b>																															
<b>Method/Guideline Followed:</b>																															
<b>GLP:</b>																															
<b>Exposure Period:</b>																															
<b>Frequency of Treatment:</b>																															
<b>Post-Exposure Period:</b>																															
<b>Method/Guideline and Test Condition Remarks:</b>																															
Test Results - Developmental Toxicity/Teratogenicity																															
<b>Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):</b>	<table border="1"> <thead> <tr> <th>LOAEL/ LOAEC/ NOAEL/ NOAEC</th> <th>Population</th> <th>Value Description</th> <th>Value/Lower Concentration</th> <th>Upper Concentration</th> <th>Units</th> </tr> </thead> <tbody> <tr> <td>NOAEC</td> <td>Fetal</td> <td>=</td> <td>5970</td> <td>27750</td> <td>mg/m3</td> </tr> <tr> <td>NOAEC</td> <td>Female (Maternal)</td> <td>=</td> <td>5970</td> <td>27750</td> <td>mg/m3</td> </tr> <tr> <td>LOAEC</td> <td>Parental (F0)</td> <td>=</td> <td>13650</td> <td>27750</td> <td>mg/m3</td> </tr> <tr> <td>NOAEC</td> <td>Parental (F0)</td> <td>=</td> <td>2275</td> <td>25000</td> <td>mg/m3</td> </tr> </tbody> </table>	LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units	NOAEC	Fetal	=	5970	27750	mg/m3	NOAEC	Female (Maternal)	=	5970	27750	mg/m3	LOAEC	Parental (F0)	=	13650	27750	mg/m3	NOAEC	Parental (F0)	=	2275	25000	mg/m3
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**Results Remarks:**

**Conclusion:**

**Reliability/Data Quality - Developmental Toxicity/Teratogenicity**

**Reliability:**

**Reliability  
Remarks:**

**Key Study Sponsor  
Indicator:** Weight of Evidence

**Reference - Developmental Toxicity/Teratogenicity**

**Reference:** For a complete discussion of the potential developmental toxicity hazards associated with gasoline blending streams please see Gasoline Blending Streams Category Analysis Document(s) at <http://www.petroleumhvp.org>.

In HPVIS, see Developmental Toxicity/Teratogenicity Robust Study Summaries for  
CAS #, 64741-41-9, 64741-55-5, 64741-63-5, 64741-66-8, 68955-35-1, and 86290-81-5 (inhalation; most relevant for human hazard)

Also in HPVIS via other routes of administration:  
CAS# 64741-55-5 and 68513-02-0 (dermal)  
CAS# 64741-55-5 (oral)



<b>Developmental Toxicity/Teratogenicity</b>																																							
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<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline																																						
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline																																						
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p> <p>[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]</p> <p>Unleaded baseline gasoline API 99-01 Vapor Condensate Test material is a complex mixture of volatile hydrocarbons. The purity of mixture is 100% and stable based on analysis of chamber atmospheres.</p> <p>Representative Components [98.8%] monitored in Study:</p> <table border="1"> <thead> <tr> <th>COMPONENT</th> <th>AREA %</th> </tr> </thead> <tbody> <tr><td>Isobutane</td><td>2.70</td></tr> <tr><td>n-butane</td><td>12.78</td></tr> <tr><td>3-methyl-1-butene</td><td>0.41</td></tr> <tr><td>Isopentane</td><td>36.50</td></tr> <tr><td>n-pentane</td><td>9.36</td></tr> <tr><td>Trans-2-pentene</td><td>3.60</td></tr> <tr><td>2,3-dimethylbutane</td><td>1.75</td></tr> <tr><td>2-methylpentane</td><td>7.25</td></tr> <tr><td>3-methylpentane</td><td>4.27</td></tr> <tr><td>n-hexane</td><td>3.62</td></tr> <tr><td>Methylcyclopentane</td><td>1.87</td></tr> <tr><td>2,4-dimethylpentane</td><td>1.36</td></tr> <tr><td>Benzene</td><td>2.75</td></tr> <tr><td>2-methylhexane</td><td>1.73</td></tr> <tr><td>2,3-dimethylpentane</td><td>1.52</td></tr> <tr><td>3-methylhexane</td><td>1.73</td></tr> <tr><td>Isooctane</td><td>1.92</td></tr> <tr><td>Toluene</td><td>3.91</td></tr> </tbody> </table> <p>Additional compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>)</p>	COMPONENT	AREA %	Isobutane	2.70	n-butane	12.78	3-methyl-1-butene	0.41	Isopentane	36.50	n-pentane	9.36	Trans-2-pentene	3.60	2,3-dimethylbutane	1.75	2-methylpentane	7.25	3-methylpentane	4.27	n-hexane	3.62	Methylcyclopentane	1.87	2,4-dimethylpentane	1.36	Benzene	2.75	2-methylhexane	1.73	2,3-dimethylpentane	1.52	3-methylhexane	1.73	Isooctane	1.92	Toluene	3.91
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<b>Method - Developmental Toxicity/Teratogenicity</b>																																							
<b>Route of Administration:</b>	Inhalation																																						
<b>Type of Exposure:</b>	Vapor																																						
<b>Species:</b>	Mouse																																						
<b>Mammalian Strain:</b>	CD-1																																						
<b>Gender:</b>	Female																																						
<b>Number of Animals per Dose:</b>	25																																						
<b>Dose:</b>	Target: 0, 2000, 10000, 20000 mg/m3 Analytical: 0, 2086, 10625, 20903 mg/m3																																						
<b>Year Study Performed:</b>	2008																																						
<b>Method/Guideline Followed:</b>	EPA 870.3700																																						

<b>GLP:</b>	Yes
<b>Exposure Period:</b>	5 - 17 Days
<b>Frequency of Treatment:</b>	6 hours/day, 5 days/week
<b>Post-Exposure Period:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	<p>EPA OPPTS 870.3600 (likely a mistake; dev tox guideline is 870.3700)</p> <p>A developmental toxicity study in rats of Baseline Gasoline Vapor Condensate (BGVC), a 20% light fraction of whole unleaded gasoline was performed according to OPPTS 870.3600, 870.3700 and OECD 414 guidelines. This test material was a representative evaporative emission tested under the USEPA 211(b) Fuels and Fuel Additives Health Effects Testing Program (1994b). BGVC was administered to 25 confirmed-mated female Crl:CD-1@ICR)BR mice/exposure group at target concentrations of 0, 2000, 10,000, and 20,000 mg/m<sup>3</sup> (mean analytical concentrations 0, 2086, 10625 and 20,903 mg/m<sup>3</sup>; 0, 680, 3463, and 6814 ppm) in air. The animals were exposed daily for six hours from Gestation Day 5 through Gestation Day 17. The Sponsor selected the exposure levels based upon safety considerations and previously conducted mammalian toxicity studies. The highest exposure level was one-half the lower explosive limit. The concentration of the test atmosphere in each chamber and the chamber room was determined approximately hourly during each exposure by on-line gas chromatography. The chamber concentrations were measured in the breathing zone of the rats. Additionally, a sorbent tube sample of the test atmosphere was collected once during each week of the study. These samples were analyzed by the detailed capillary/GC method used for the initial characterization analysis of the liquid test substance. This analysis was done to determine component proportions of the test material atmosphere compared to the liquid test material. Chamber Homogeneity was evaluated during the validation of the exposure system for this study. Distribution samples were drawn from twelve different points within the chamber at each exposure level. A particle size determination of the aerosol portion of the test atmosphere was conducted three times during the chamber trials from the 20,000 mg/m<sup>3</sup> concentration. The samples were taken using a multistage cascade impactor. Preweighed glass fiber filters were used to collect aerosol on each stage, which are associated with specific cutoff diameters for aerodynamic particle size in microns. Since minimal aerosol was present, no further calculations were performed. Clinical observations were made daily during gestation. Body weight and food consumption measurements were made on GD 0, 5, 8, 11, 14, 17, and 18. On GD 18, animals were sacrificed by CO<sub>2</sub> asphyxiation followed by exsanguination. and cesarean sections (C-sections) were performed. . The reproductive organs and the abdominal and thoracic cavities were examined grossly.</p>

Evaluations of dams during cesarean section were conducted without knowledge of treatment group in order to minimize bias. Uterine weights with ovaries attached were recorded. Uterine contents were examined, and the numbers of live, dead and resorbed fetuses were recorded. Corpora lutea were also counted. All fetuses were weighed, sexed externally, and examined externally for gross malformations. Apparent non-gravid uteri were placed in 10% ammonium sulfide solution for confirmation of non-pregnancy status. The fetuses were placed in a refrigerator to slow down and eventually terminate vital signs after the external examination and weighing. The viscera of approximately one-half of the fetuses of each litter were examined by fresh dissection. After these fetuses were examined, they were decapitated. The heads were preserved in Bouin's solution for at least two weeks, rinsed, and subsequently stored in 70% ethanol. The fetal heads were sectioned and examined with a dissecting microscope for the presence of abnormalities. The remaining fetuses judged to be alive at the C-section were eviscerated, processed for skeletal staining, stained for bone and cartilage, and examined for the presence of skeletal malformations and variations.

Statistical Analysis: Statistical evaluation of equality of means was done by an appropriate one way analysis of variance and a test for ordered response in the dose groups. First, Bartlett's Test was performed to determine if the dose groups had equal variance (Snedecor and Cochran, 1989). If the variances were equivalent, the hypothesis that there was no difference in response between the groups was tested using a standard one-way analysis of variance (Snedecor and Cochran, 1989). If the variances were equal, the testing was done using parametric methods, otherwise nonparametric techniques were used. Continuous data will be tested for statistical significance as follows: Where applicable, percentages were calculated and transformed by Cochran's transformation, followed by the arc sine transformation (Snedecor and Cochran, 1989). The raw percentages and the transformed percentages both were tested for statistical significance. For the parametric procedures, a standard one way ANOVA using the F distribution to assess significance was used (Snedecor and Cochran, 1989). If significant differences among the means were indicated, Dunnett's Test was used to determine which treatment groups differed significantly from control (Dunnett, 1964). In addition to the ANOVA, a standard regression analysis for linear response in the dose groups was performed. The regression also tested for linear lack of fit in the model. For the nonparametric procedures, the test of equality of means was performed using the Kruskal-Wallis Test (Hollander and Wolfe, 1973). If significant differences among the means were indicated, Dunn's Summed Rank Test was used to determine which treatment groups differed significantly from the control (Hollander and Wolfe, 1973). In addition to the Kruskal-Wallis Test,

Jonckheere's Test for monotonic trend in the dose response was performed.  
 Bartlett's Test for equal variance was conducted at the 1% level of significance. All other tests were conducted at the 5% and 1% level of significance.  
 The following data was not included in the statistical analyses:  
 ? Gestation body weight and body weight change data for females that were not pregnant  
 ? Gestation food consumption for females that were not pregnant

Means and standard deviations were calculated for animal, exposure and chamber environmental data. The coefficient of variation also was calculated when considered relevant for the exposure data.  
 Fetal body weight was analyzed by a mixed model analysis of variance that provided an accurate statistical model of the biology. The analysis used the litter as the basis for analysis and effectively used the litter size as a covariate. The model considered dose group, litter size, and fetal sex as explanatory variables. If the overall effect of dose, or the dose by sex effect, was statistically significant the dose groups means were tested pairwise vs. the control group using least squares means. The least squares means allowed comparisons that accounted for differences in litter size and sex. The mathematical model was based on a paper by Chen, et al (1996). The analysis was run using SAS with code suggested in Little, et al (1997). The analysis of anomalies (malformations or variations) was based on a Generalized Estimating Equation (GEE) application of the linearized model, Ryan (1992). The model used the litter as the basis for analysis and considered correlation among littermates by incorporating an estimated constant correlation and the litter size as a covariate. If the overall effect of dose, or the dose by sex effect, was statistically significant the dose groups were tested pairwise vs. the control group using least squares means. The least squares means allowed comparisons that accounted for differences in litter size. Three categories of anomalies were tested, and within each category specific anomalies also were tested. In addition to the category specific anomalies a series of combined analyses were performed within each category as applicable:  
 Combined Malformations and Variations for All Fetuses  
 Combined Malformations and Variations for Alive Fetuses  
 Malformations for All Fetuses  
 Malformations for Alive Fetuses  
 Variations for All Fetuses  
 Variations for Alive Fetuses

**Test Results - Developmental Toxicity/Teratogenicity**

**Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):**

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
LOAEL	Female (Maternal)	=	20000		mg/m3
NOAEL	Female (Maternal)	=	10000		mg/m3
LOAEL	Fetal	>=	10000		mg/m3

NOAEL	Fetal	>=	2000	mg/m3
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**Results Remarks:**

All dams were free of clinical or postmortem findings attributable to treatment with BGVC. One control, one 10,000 mg/m3, and two 20,000 mg/m3 dams were determined at the scheduled terminal sacrifice to be not pregnant. Additionally, one control and one 20,000 mg/m3 dam delivered their litters on Day 18, prior to their scheduled sacrifice.

Maternal toxicity was evident as statistically significant decreases in mean gestation body weight and mean gestation body weight change in the 20,000 mg/m3 target concentration group. The only clinical sign observed was emaciation, noted in a single dam at 20,000 mg/m3 on GD 11; since this finding was not seen in other dams at this target concentration, this is unlikely to be related to exposure to the test substance.

Statistically significant reduced fetal body weights, compared with the control fetal weights, were noted in the 10,000 and 20,000 mg/m3 target concentration groups. The reduction of these fetal weights occurred in the absence of statistically significant reductions in maternal body weight and body weight change in the 10,000 mg/m3 target concentration group. There were no statistically significant differences detected in the incidence of other fetal observations.

The uterine implantation data revealed a statistically significant decrease in the number of live fetuses in the 20,000 mg/m3 target concentration group and also a statistically significant increase in the transformed resorptions to implantation ratio in this group. This difference is not considered to be exposure-related for two reasons. First, the mean number of corpora lutea (CL) per litter at 20,000 mg/m3 was nearly two CL less than the control group. The number of corpora lutea was determined prior to initiation of exposure to the test material, and hence cannot be due to exposure. The difference in the mean number of corpora lutea per litter alone is insufficient to explain the cascading differences in mean litter implantation number and live fetuses per litter. The reduced number of live fetuses primarily was a function of a reduction in the number of implantations prior to commencement of exposure. Additionally the litter of one dam in this group was completely resorbed. The dam lost 26% of her body weight on GD 8-11. Although there is no apparent explanation for this animal's weight loss, weight loss during gestation in mice due to food restriction is associated with increased resorptions (Chapin et al., 1993). When the uterine implantation data for this litter was removed from the statistical analyses as an outlier, there was no statistical significance in the transformed resorptions to implantation ratio. This dam also was noted as emaciated on GD 11 and its body weight data indicates

that resorption of the litter probably occurred between GD 5 and GD 8.

The NOAELs for developmental and maternal toxicity were considered to be 2000 (680 ppm) and 10,000 mg/m<sup>3</sup> (3,463 ppm) target concentrations, respectively.

**Conclusion:** Based upon reduced fetal body weights in the absence of reduced maternal body weights, BGVC was determined to be a developmental toxicant in CD-1 mice. The NOAEL for developmental toxicity was 2,086 mg/m<sup>3</sup> (680 ppm); the LOAEL for developmental toxicity was 10,625 mg/m<sup>3</sup> (3,463 ppm). Based upon reduced gestation body weight and mean gestation body weight change, the Maternal NOAEL was 10,625 mg/m<sup>3</sup> (3,463 ppm); the maternal LOAEL was 20,903 mg/m<sup>3</sup> (6,814 ppm).

#### Reliability/Data Quality - Developmental Toxicity/Teratogenicity

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** Guideline study

**Key Study Sponsor Indicator:** Key

#### Reference - Developmental Toxicity/Teratogenicity

**Reference:** Whole-Body Inhalation Developmental Toxicity Study in Mice with Baseline Gasoline Vapor Condensate (MRD-00-695). Laboratory (EMBSI) study number 169534. ExxonMobil Biomedical Sciences, Inc., Annadale, NJ. Study conducted for the American Petroleum Institute 211(b) Research Group in compliance of the Clean Air Act 211(b) testing requirements.

Other references cited in study summary:  
Dunnett, C., New Tables for Multiple Comparisons with a Control, Biometrics 20, 1964, pp. 482-491.

Hollander, M. and Wolfe, D.A. Nonparametric Statistical Methods, John Wiley and Sons, New York, 1973.

Little, Milliken, Stroup, and Wolfinger, ?SAS System for Mixed Models?, SAS Institute, Cary, NC, 1997, section 5.6.2, pg 203.

Ryan, L., ?The use of generalized estimating equations for risk assessment in developmental toxicity?, Risk Analysis, 12(3), pg 439-447, 1992.

Snedecor, G.W., and Cochran, W.G., Statistical Methods, 8th ed., Iowa State University Press, Ames, Iowa, 1989.



<b>Developmental Toxicity/Teratogenicity</b>	
<b>Test Substance - Developmental Toxicity/Teratogenicity</b>	
<b>Category Chemical:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance:</b>	(64741-55-5) Naphtha, petroleum, light catalytic cracked
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Light Catalytically Cracked Naphtha (LCCN), CAS# 64741-55-5. The test material contained approximately 41% olefins. Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Developmental Toxicity/Teratogenicity</b>	
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Female
<b>Number of Animals per Dose:</b>	15
<b>Dose:</b>	Target: 2000 & 8000 mg/m <sup>3</sup> . Actual: 2150 ± 260 & 7660 ± 570 mg/m <sup>3</sup>
<b>Year Study Performed:</b>	1996
<b>Method/Guideline Followed:</b>	Other
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	20 Days
<b>Frequency of Treatment:</b>	6 hr/day
<b>Post-Exposure Period:</b>	0
<b>Method/Guideline and Test Condition Remarks:</b>	Control group: Yes  Four groups of 15 presumed-pregnant female rats were assigned to the following groups: Untreated controls, sham-treated controls, 2000 and 8000 mg/m <sup>3</sup> test material. Exposures were for 6 hours each day on days 0 to 19 of gestation. All animals were observed daily and body weights were recorded on days 0, 6, 13 and 20 of gestation. On day 20 each female was sacrificed and all organs were examined grossly. Serum samples were analyzed for a variety of parameters, including serum iron and lactic dehydrogenase.  The number of corpora lutea per ovary and the gravid uterine weights were recorded. Uterine contents were examined and the numbers of implantation sites, early resorptions and live and dead fetuses recorded. Each fetus was identified for its sex, weighed and the crown-rump distance was measured. Each fetus was examined for external anomalies. Half the fetuses were fixed in Bouin's solution and examined for visceral anomalies and the



remaining fetuses were prepared for examination for skeletal anomalies.

**CHAMBER TEST MATERIAL CONCENTRATION GENERATION:**

Vapors of LCCN were generated in a glass countercurrent generator (one for each concentration). As liquid LCCN flowed down the coil, nitrogen passed upwards and carried off vapors of the more volatile components. Main stream air was used to dilute the vapor to the required concentration.

Vapor concentration was monitored at approximately hourly intervals during each exposure period.

Concentrations (Target and actual) are shown below.

Target	Actual
(mg/m <sup>3</sup> )	(mg/m <sup>3</sup> )
2000	2150 ± 260
8000	7660 ± 570

The test material contained approximately 41% olefins.

**Test Results - Developmental Toxicity/Teratogenicity**

**Concentration  
(LOAEL/ LOAEC/  
NOAEL/ NOAEC):**

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
LOAEC	Fetal	=	7660		mg/m <sup>3</sup>
NOAEC	Fetal	=	2150		mg/m <sup>3</sup>
NOAEC	Female (Maternal)	>=	7660		mg/m <sup>3</sup>

**Results Remarks:**

There were no treatment-related clinical abnormalities or differences in body weight among dams. Results of the reproductive parameters are listed below.

LCCN LCCN

Control 2150 7660

Parameter No Sham mg/m<sup>3</sup> mg/m<sup>3</sup>

treat treat

Females mated 15 15 15 15

Females pregnant 14 13 14 15

Corpora lutea 18 18 16 18

Implantation sites 16 16 14 16

Primplantation loss (&) 10 12 14 8

Viable fetuses/litter 15 14 14 15

Resorptions 0.7 0.6 0.8 1.7 \*a

Resorptions (%) 4.6 3.9 4.7 10.4 \*a

Dams with resorptions 9 5 8 13 \*b

\*a Significant difference from untreated and sham treated controls

\*b Significant difference from sham treated controls

It is clear that with the exception of resorptions, no other parameter was affected by exposure.

During the external examination of fetuses, a sham treated animal had gastroschisis and one fetus from the 2150 mg/m<sup>3</sup> group had a tail that was short and filamentous.

Fetal body weights and crown-rump lengths were unaffected by treatment.

No visceral abnormalities were observed.

There was an increased number of skeletal variations in animals housed in the exposure chambers (exposed and sham treated controls) when compared to the untreated controls. The authors concluded that these alterations were not related to LCCN since they occurred at the same incidence in the sham treated controls as well. The findings are tabulated below.

The numbers of fetuses with the specific anomaly are shown. The numbers in parenthesis are the % of fetuses.

LCCN LCCN

Control 2150 7660

Parameter No Sham mg/m<sup>3</sup> mg/m<sup>3</sup>

treat treat

Caudal vertebrae:  
 transverse process 18(16) 42(40) 41(40) 45(39)  
 incompletely ossified

Sacral vertebrae  
 transverse process 7(6) 23(22) 17(17) 28(24)  
 incompletely ossified

Incompletely ossified 83(75) 80(76) 91(89) 101(88)  
 sternebrae

[Note: IF DATA TABLE(S) FORMATTING IS LOST (i.e. tables are unclear/unreadable), please see Gasoline Blending Streams Category Robust Study Summaries at <http://www.petroleumhqv.org> for easier to read tables.]

**Conclusion:** Based upon increased resorptions, the developmental LOAEC = 7660 mg/m<sup>3</sup> (2128ppm), which was the highest dose tested. The developmental NOAEC = 2150mg/m<sup>3</sup> (597ppm).

The maternal systemic NOAEC = 7660 mg/m<sup>3</sup> (2128ppm), the highest dose tested.

#### Reliability/Data Quality - Developmental Toxicity/Teratogenicity

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** RELIABILITY: GLP study with adequately detailed methods description

**Key Study Sponsor Indicator:** Key

#### Reference - Developmental Toxicity/Teratogenicity

**Reference:** Dalbey, W. E., Feuston, M. H., Yang, J. J., Kommineni, C. V and Roy, T. A. (1996) Light Catalytically cracked naphtha: subchronic toxicity of vapors in rats and mice and developmental toxicity screen in rats. J. Toxicol. and Env. Health Vol 47, pp 77-91



<b>Developmental Toxicity/Teratogenicity</b>	
<b>Test Substance - Developmental Toxicity/Teratogenicity</b>	
<b>Category Chemical:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Partially vaporized full range catalytic reformed naphtha (FR-CRN) with > 60% aromatic compounds. The test material was tested as a 40% vapor. Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Developmental Toxicity/Teratogenicity</b>	
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Female
<b>Number of Animals per Dose:</b>	11
<b>Dose:</b>	0, 508, and 1835 ppm. (0, 2160 and 7800 mg/m3) There were two control groups; untreated and sham treated controls
<b>Year Study Performed:</b>	1996
<b>Method/Guideline Followed:</b>	Other
<b>GLP:</b>	No Data
<b>Exposure Period:</b>	14 Days
<b>Frequency of Treatment:</b>	6 hours/ day
<b>Post-Exposure Period:</b>	0
<b>Method/Guideline and Test Condition Remarks:</b>	Groups of 11 or 12 presumed pregnant female rats were exposed 6 hours each day from days 6-19 of gestation to whole body exposures of 508 or 1835 ppm (2160 and 7800 mg/m3) partially vaporized FR-CRN (40%). Two extra groups served as untreated and sham treated controls. All animals were observed daily and body weights were recorded on days 0, 6, 13 and 20 of gestation. On day 20 each female was sacrificed and blood samples removed for serum chemistry evaluations. Parameters measured were the same as those in the subchronic study by the same authors, and in addition included iron and lactic dehydrogenase. All organs were examined grossly and liver and thymus weights were recorded. In addition, the number of corporea lutea per ovary and the gravid uterine weights were recorded. Uterine contents were examined and the numbers of implantation sites, early and late resorptions and live and dead fetuses were recorded. Each fetus was gendered, weighed and grossly examined for external abnormalities. Half the fetuses were fixed in Bouin's fluid and examined subsequently for

soft tissue abnormalities. Remaining fetuses were stained with Alizarin red and examined for skeletal anomalies.

### Test Results - Developmental Toxicity/Teratogenicity

**Concentration  
(LOAEL/ LOAEC/  
NOAEL/ NOAEC):**

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
NOAEL	Female (Maternal)	>=	7800		mg/m3
NOAEL	Fetal	>=	7800		mg/m3

**Results Remarks:**

There were no adverse effects on maternal body weight gain, liver weight or thymus weight. In the high dose group, maternal serum glucose levels were significantly decreased (1.5%) and potassium levels increased (1%) relative to the untreated controls. Reproductive performance during gestation and in-utero survival and development of concepti were unaffected by treatment. Furthermore, there were no treatment-related increases in gross abnormalities or anomalies of soft or skeletal tissues.

**Conclusion:**

Partially vaporized full range catalytic reformed naphtha (FR-CRN) with > 60% aromatic compounds did not produce maternal or developmental toxicity under the test conditions described. The changes in maternal serum chemistry were not considered to be relevant in setting the NOAEL.

The NOAEL for maternal and developmental endpoints >= 7800 mg/m3 (1835 ppm), which was the highest dose tested.

### Reliability/Data Quality - Developmental Toxicity/Teratogenicity

**Reliability:**

Valid with Restrictions

**Reliability  
Remarks:**

GLP status for test was unknown; study design and endpoints evaluated were similar to standard guideline developmental/teratogenicity protocols.

**Key Study Sponsor  
Indicator:**

Key

### Reference - Developmental Toxicity/Teratogenicity

**Reference:**

Dalbey, W. and Feuston, M. (1996) Partially vaporized full range catalytic reformed naphtha: Subchronic and developmental toxicity studies in rats. Inhalation Toxicology. Vol 8., pp 271-284



<b>Developmental Toxicity/Teratogenicity</b>																																							
<b>Test Substance - Developmental Toxicity/Teratogenicity</b>																																							
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline																																						
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline																																						
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p> <p>[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]</p> <p>Unleaded baseline gasoline API 99-01 Vapor Condensate Test material is a complex mixture of volatile hydrocarbons. The purity of mixture is 100% and stable based on analysis of chamber atmospheres.</p> <p>Representative Components [98.8%] monitored in Study:</p> <table border="1"> <thead> <tr> <th>COMPONENT</th> <th>AREA %</th> </tr> </thead> <tbody> <tr><td>Isobutane</td><td>2.70</td></tr> <tr><td>n-butane</td><td>12.78</td></tr> <tr><td>3-methyl-1-butene</td><td>0.41</td></tr> <tr><td>Isopentane</td><td>36.50</td></tr> <tr><td>n-pentane</td><td>9.36</td></tr> <tr><td>Trans-2-pentene</td><td>3.60</td></tr> <tr><td>2,3-dimethylbutane</td><td>1.75</td></tr> <tr><td>2-methylpentane</td><td>7.25</td></tr> <tr><td>3-methylpentane</td><td>4.27</td></tr> <tr><td>n-hexane</td><td>3.62</td></tr> <tr><td>Methylcyclopentane</td><td>1.87</td></tr> <tr><td>2,4-dimethylpentane</td><td>1.36</td></tr> <tr><td>Benzene</td><td>2.75</td></tr> <tr><td>2-methylhexane</td><td>1.73</td></tr> <tr><td>2,3-dimethylpentane</td><td>1.52</td></tr> <tr><td>3-methylhexane</td><td>1.73</td></tr> <tr><td>Isooctane</td><td>1.92</td></tr> <tr><td>Toluene</td><td>3.91</td></tr> </tbody> </table> <p>Additional compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>)</p>	COMPONENT	AREA %	Isobutane	2.70	n-butane	12.78	3-methyl-1-butene	0.41	Isopentane	36.50	n-pentane	9.36	Trans-2-pentene	3.60	2,3-dimethylbutane	1.75	2-methylpentane	7.25	3-methylpentane	4.27	n-hexane	3.62	Methylcyclopentane	1.87	2,4-dimethylpentane	1.36	Benzene	2.75	2-methylhexane	1.73	2,3-dimethylpentane	1.52	3-methylhexane	1.73	Isooctane	1.92	Toluene	3.91
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<b>Dose:</b>	Target: 0, 2000, 10000, 20000 mg/m3 Analytical: 0, 1979, 10676, 20638 mg/m3																																						
<b>Year Study Performed:</b>	2008																																						
<b>Method/Guideline Followed:</b>	EPA 870.3700																																						

<b>GLP:</b>	Yes
<b>Exposure Period:</b>	5 - 20 Days
<b>Frequency of Treatment:</b>	6 hours/day, 5 days/week
<b>Post-Exposure Period:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	<p>EPA OPPTS 870.3600 (likely a mistake - as of 1/10/2014, there is no 870-3600; guideline number for teratogenicity is 870.3700)</p> <p>Baseline Gasoline Vapor Condensate (BGVC) was administered by whole-body inhalation exposure to 25 confirmed-mated Crl:CD®(SD)IGSBR female rats at target doses of 0 (air control) 2000, 10,000, and 20,000 mg/m<sup>3</sup> for six hours (plus the theoretical equilibration time) daily from Gestation Day (GD) 5 through GD 20.</p> <p>The Sponsor selected the exposure levels based upon safety considerations and previously conducted mammalian toxicity studies. The highest exposure level was one-half the lower explosive limit.</p> <p>The concentration of the test atmosphere in each chamber and the chamber room was determined approximately hourly during each exposure by on-line gas chromatography. The chamber concentrations were measured in the breathing zone of the rats.</p> <p>Additionally, a sorbent tube sample of the test atmosphere was collected once during each week of the study. These samples were analyzed by the detailed capillary/GC method used for the initial characterization analysis of the liquid test substance. This analysis was done to determine component proportions of the test material atmosphere compared to the liquid test material. Chamber Homogeneity was evaluated during the validation of the exposure system for this study. Distribution samples were drawn from twelve different points within the chamber at each exposure level.</p> <p>A particle size determination of the aerosol portion of the test atmosphere was conducted three times during the chamber trials from the 20,000 mg/m<sup>3</sup> concentration. The samples were taken using a multistage cascade impactor.</p> <p>Preweighed glass fiber filters were used to collect aerosol on each stage, which are associated with specific cutoff diameters for aerodynamic particle size in microns. Since minimal aerosol was present, no further calculations were performed.</p> <p>Clinical observations were made daily during gestation. Body weight and food consumption measurements were made on GD 0, 5, 8, 11, 14, 17, 20, and 21. On GD 21, animals were sacrificed by CO<sub>2</sub> asphyxiation followed by exsanguination.</p> <p>Cesarean sections were then conducted. The reproductive organs and the abdominal and thoracic cavities were examined grossly. Evaluations of dams during cesarean section were conducted without knowledge of treatment group in order to minimize bias. Uterine weights with ovaries attached were recorded.</p> <p>Uterine contents were examined, and the numbers of live, dead and resorbed fetuses were recorded. Corpora lutea were also counted. All fetuses were weighed, sexed externally, and examined externally for gross</p>

malformations.

Apparent non-gravid uteri were placed in 10% ammonium sulfide solution for confirmation of non-pregnancy status. The fetuses were placed in a refrigerator to slow down and eventually terminate vital signs after the external examination and weighing. The viscera of approximately one-half of the fetuses of each litter were examined by fresh dissection. After these fetuses were examined, they were decapitated. The heads were preserved in Bouin's solution for at least two weeks, rinsed, and subsequently stored in 70% ethanol. The fetal heads were sectioned and examined with a dissecting microscope for the presence of abnormalities. The remaining fetuses judged to be alive at the C-section were eviscerated, processed for skeletal staining, stained for bone and cartilage, and examined for the presence of skeletal malformations and variations.

Statistical Analysis: Statistical evaluation of equality of means was done by an appropriate one way analysis of variance and a test for ordered response in the dose groups. First, Bartlett's Test was performed to determine if the dose groups had equal variance (Snedecor and Cochran, 1989). If the variances were equivalent, the hypothesis that there was no difference in response between the groups was tested using a standard one-way analysis of variance (Snedecor and Cochran, 1989). If the variances were equal, the testing was done using parametric methods, otherwise nonparametric techniques were used. Continuous data will be tested for statistical significance as follows: Where applicable, percentages were calculated and transformed by Cochran's transformation, followed by the arc sine transformation (Snedecor and Cochran, 1989). The raw percentages and the transformed percentages both were tested for statistical significance.

For the parametric procedures, a standard one way ANOVA using the F distribution to assess significance was used (Snedecor and Cochran, 1989). If significant differences among the means were indicated, Dunnett's Test was used to determine which treatment groups differed significantly from control (Dunnett, 1964). In addition to the ANOVA, a standard regression analysis for linear response in the dose groups was performed. The regression also tested for linear lack of fit in the model.

For the nonparametric procedures, the test of equality of means was performed using the Kruskal-Wallis Test (Hollander and Wolfe, 1973). If significant differences among the means were indicated, Dunn's Summed Rank Test was used to determine which treatment groups differed significantly from the control (Hollander and Wolfe, 1973). In addition to the Kruskal-Wallis Test, Jonckheere's Test for monotonic trend in the dose response was performed.

Bartlett's Test for equal variance was conducted at the 1% level of significance. All other tests were conducted at the 5% and 1% level of significance.

The following data was not included in the statistical analyses:  
? Gestation body weight and body weight change data for females

that were  
not pregnant  
? Gestation food consumption for females that were not pregnant

Means and standard deviations were calculated for animal, exposure and chamber environmental data. The coefficient of variation also was calculated when considered relevant for the exposure data. Fetal body weight was analyzed by a mixed model analysis of variance that provided an accurate statistical model of the biology. The analysis used the litter as the basis for analysis and effectively used the litter size as a covariate. The model considered dose group, litter size, and fetal sex as explanatory variables. If the overall effect of dose, or the dose by sex effect, was statistically significant the dose groups means were tested pairwise vs. the control group using least squares means. The least squares means allowed comparisons that accounted for differences in litter size and sex. The mathematical model was based on a paper by Chen, et al (1996). The analysis was run using SAS with code suggested in Little, et al (1997). The analysis of anomalies (malformations or variations) was based on a Generalized Estimating Equation (GEE) application of the linearized model, Ryan (1992). The model used the litter as the basis for analysis and considered correlation among littermates by incorporating an estimated constant correlation and the litter size as a covariate. If the overall effect of dose, or the dose by sex effect, was statistically significant the dose groups were tested pairwise vs. the control group using least squares means. The least squares means allowed comparisons that accounted for differences in litter size. Three categories of anomalies were tested, and within each category specific anomalies also were tested. In addition to the category specific anomalies a series of combined analyses were performed within each category as applicable:  
Combined Malformations and Variations for All Fetuses  
Combined Malformations and Variations for Alive Fetuses  
Malformations for All Fetuses  
Malformations for Alive Fetuses  
Variations for All Fetuses  
Variations for Alive Fetuses

### Test Results - Developmental Toxicity/Teratogenicity

Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):	LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
	NOAEC	Female (Maternal)	>=	20000		mg/m3
	NOAEC	Fetal	>=	20000		mg/m3

#### Results Remarks:

The mean analytical exposure concentrations [ $\pm$  standard deviation (S.D.)] were 1979  $\pm$  98.0, 10676  $\pm$  309.8, and 20638  $\pm$  452.1 for the target concentrations of 2000, 10000, and 20000 mg/m<sup>3</sup>, respectively. Chamber uniformity was also within acceptable limits with 12 point sampling means ( $\pm$  S.D.) of 1997  $\pm$  56.4, 10495  $\pm$



195.0, and 19996 ± 275.8 mg/m<sup>3</sup> for the respective target concentrations.

There was no evidence of maternal toxicity in this study at any concentration tested. All dams survived to scheduled terminal sacrifice on GD 21 and were free of clinical or postmortem effects attributable to treatment with BGVC. However there was a statistically significant linear trend (decrease) in dose response in the GD 5-8 body weight change and a statistically significant linear trend (increase) in dose response in the GD 14-17 body weight change. However, the pairwise analyses of the control data versus each treated group was not statistically significant; mean maternal body weight for the 20,000 mg/m<sup>3</sup> target concentration group on GD 8 was 98.9% of the control mean value. The linear trend for the GD 14-17 body weight change was also not considered biologically significant due to the absence of statistically significant differences between the treated and control groups.

There were no statistically significant differences between the control and the BGVC treated groups for uterine implantation data, and external, visceral, and skeletal observations. The most frequently noted observation during fetal examinations was rudimentary lumbar ribs. The incidence of this observation was similar across all groups and was within the historical control range of this laboratory.

A statistically significant decrease in mean fetal body weight was evident in all exposed groups. This could be interpreted as an indication of developmental toxicity. However, these decreases are probably neither treatment related nor biologically significant for the following reasons:

\* The mean fetal weights of the treatment groups were within the historical control range of the laboratory. The mean fetal body weights determined in the control group were greater than this laboratory's historical control mean fetal body weight range and likewise the MARTA historical control data base (mean fetal body weights) for Charles River (Raleigh facility) rat fetuses obtained from dams on GD 21.

\* A comparison of mean litter weights (mean of the sum of all fetus weights/group) revealed that the litter weights of all groups were comparable and the control litter weights were the most variable.

\* The mean litter size in the control group was smaller than any treated group. Consequently, it must be remembered, however, that among animals which deliver multiple offspring, individual fetal body weights tend to be heavier in smaller litters, as was seen in this study (Romero, 1992).

\* There was no dose response in the mean fetal weights of the treated groups. The fetal weights of the treated groups were not statistically significantly different from each other. If the lower fetal weights in the treated groups were related to treatment, one would expect that the mean fetal weight of the group exposed to a target concentration of 20,000 mg/m<sup>3</sup> would be at least substantially lower than the mean fetal weight of the group exposed to a target concentration of 2000 mg/m<sup>3</sup>.

No other observations were evident in the treated groups that were statistically or biologically significantly different from the observations in the control group.

In conclusion, administration of the test substance to rats by whole-body inhalation exposure during the period of organogenesis and fetal growth did not result in maternal or developmental toxicity.

Therefore, the No Observable Adverse Effect Concentrations (NOAECs) for maternal and developmental toxicity in this study was established at 20,000 mg/m<sup>3</sup> target concentration.

**Conclusion:** BGVC was not a developmental toxicant in Sprague Dawley rats at exposure concentrations up to 20000 mg/m<sup>3</sup>. The NOAEC for both maternal and developmental toxicity was  $\geq$  20000 mg/m<sup>3</sup>. This was the highest concentration tested.

### Reliability/Data Quality - Developmental Toxicity/Teratogenicity

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** Guideline study conducted according to GLPs

**Key Study Sponsor Indicator:** Key

### Reference - Developmental Toxicity/Teratogenicity

**Reference:** Whole-Body Inhalation Developmental Toxicity Study in Rats with Baseline Gasoline Vapor Condensate (MRD-00-695). Laboratory (EMBSI) study number 169534. ExxonMobil Biomedical Sciences, Inc., Annadale, NJ. Study conducted for the American Petroleum Institute 211(b) Research Group in compliance of the Clean Air Act 211(b) testing requirements.

Other references cited in study summary:  
Dunnnett, C., New Tables for Multiple Comparisons with a Control, Biometrics 20, 1964, pp. 482-491.

Hollander, M. and Wolfe, D.A. Nonparametric Statistical Methods, John Wiley and Sons, New York, 1973.

Little, Milliken, Stroup, and Wolfinger, ?SAS System for Mixed Models?, SAS Institute, Cary, NC, 1997, section 5.6.2, pg 203.

Romero, A., Villamayor, F., Grau, M. T., Sacristan, A., and Ortiz, J. A. ?Relationship between Fetal Weight and Litter Size in Rats: Application to Reproductive Toxicology Studies?, Reproductive Toxicology 6: 453-456, 1992.

Ryan, L., ?The use of generalized estimating equations for risk assessment in developmental toxicity?, Risk Analysis, 12(3), pg 439-447, 1992.

Snedecor, G.W., and Cochran, W.G., Statistical Methods, 8th ed., Iowa State University Press, Ames, Iowa, 1989.



<b>Developmental Toxicity/Teratogenicity</b>																																				
<b>Test Substance - Developmental Toxicity/Teratogenicity</b>																																				
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate																																			
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate																																			
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Distillate of light alkylate naphtha (LAN-D)</p> <p>The test material (LAN-D) was prepared to be representative of the fraction of light alkylate naphtha to which man would normally be exposed during normal handling and use. It was obtained by the distillation of light alkylate naphtha (LAN) and collecting that fraction that boiled over the temperature range 78 to 145°F. The sample was analyzed and its composition compared to the light alkylate naphtha from which it was derived (See section 1.1.1. above).</p> <p>The compositions of the distillate and starting material were as follows:</p> <table border="1"> <thead> <tr> <th rowspan="2">Compound</th> <th colspan="2">Weight %</th> </tr> <tr> <th>LAN-D</th> <th>LAN</th> </tr> </thead> <tbody> <tr> <td>n-butane</td> <td>3.42</td> <td>0.84</td> </tr> <tr> <td>isopentane</td> <td>63.59</td> <td>12.61</td> </tr> <tr> <td>n-pentane</td> <td>1.33</td> <td>0.23</td> </tr> <tr> <td>2,3-dimethylbutane</td> <td>22.51</td> <td>4.74</td> </tr> <tr> <td>2-methylpentane</td> <td>6.44</td> <td>1.57</td> </tr> <tr> <td>3-methylpentane</td> <td>2.26</td> <td>0.74</td> </tr> <tr> <td>2,4-dimethylpentane</td> <td>0.29</td> <td>4.09</td> </tr> <tr> <td>2,2,4-trimethylpentane</td> <td>0.06</td> <td>23.92</td> </tr> <tr> <td>2,3,3-trimethylpentane</td> <td>0</td> <td>8.99</td> </tr> <tr> <td>2,3,4-trimethylpentane</td> <td>0</td> <td>11.56</td> </tr> </tbody> </table> <p>Additional compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>)</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>	Compound	Weight %		LAN-D	LAN	n-butane	3.42	0.84	isopentane	63.59	12.61	n-pentane	1.33	0.23	2,3-dimethylbutane	22.51	4.74	2-methylpentane	6.44	1.57	3-methylpentane	2.26	0.74	2,4-dimethylpentane	0.29	4.09	2,2,4-trimethylpentane	0.06	23.92	2,3,3-trimethylpentane	0	8.99	2,3,4-trimethylpentane	0	11.56
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<b>Mammalian Strain:</b>	Sprague-Dawley																																			
<b>Gender:</b>	Both M/F																																			
<b>Number of Animals per Dose:</b>	10																																			
<b>Dose:</b>	<p>Actual 5.09, 12.5 and 24.7 g/m<sup>3</sup> (5090, 12500, &amp; 24700 mg/m<sup>3</sup>)</p> <p>Target: 5, 12.5, and 25 g/m<sup>3</sup> (1650, 4040, &amp; 8000 ppm)</p>																																			
<b>Year Study Performed:</b>	1995																																			
<b>Method/Guideline Followed:</b>	OECD 421																																			

<b>GLP:</b>	Yes
<b>Exposure Period:</b>	7 - 8 Weeks
<b>Frequency of Treatment:</b>	6 hr/day
<b>Post-Exposure Period:</b>	0
<b>Method/Guideline and Test Condition Remarks:</b>	<p>This study forms part of the fertility study described in the Reproductive Toxicity Section for this CAS# 64741-66-8, where the method is also described. For the examination for developmental effects, the pups were sacrificed on day 4 or 5 post partum and were necropsied and examined grossly for any abnormalities.</p> <p>As a courtesy for readers, the Methods have been copied here from Reproductive Toxicity summary for the same study:</p> <p>"Type: One generation study Premating exposure period: Male and Female: 14 days Duration of test: Females 7 weeks, males 8 weeks Method: Adaptation of OECD No. 421</p> <p>The test material was totally vaporized and diluted with air to achieve the desired concentrations for the study. Exposures were conducted in one cubic meter whole-body chambers. Chamber concentrations were monitored three times daily by GC/FID. All animals were housed individually in suspended mesh cages. 10 animals of each sex were exposed 6 hours each day to test material at target concentrations of 5, 12.5 and 25 g/m<sup>3</sup>. The animals were exposed for 6 hours each day. Parental females were exposed for 14 days prior to mating, throughout mating and gestation days 0-19 (7 consecutive weeks). Dams and their litters were sacrificed on postpartum day 4. Parental males were also exposed for 14 days prior to mating, during mating, throughout the female gestation and post partum period and throughout the female necropsy period (8 consecutive weeks). Rats were mated in a 1:1 ratio and females were monitored for evidence of mating by the examination of a vaginal lavage sample for sperm or vaginal plug. If sperm or a vaginal plug were observed the female was considered to be at day 0 of gestation and the male was removed from the female at this stage. If there was no evidence that mating had occurred the pairs were allowed to remain together up to a period of 2 weeks after which time the female was assumed to be pregnant. All animals were observed for clinical signs at least twice daily throughout the study. Body weights and food consumption were recorded throughout the study. Each litter was examined as soon as possible after delivery to establish number and sex of pups, stillbirths, live births and presence of gross abnormalities. Neonatal survival was monitored and all pups were killed postpartum days 4 or 5. Parental females were killed on gestation day 25 if they had not delivered, otherwise they were killed on postpartum days 4 or 5. At necropsy each parental animal was examined macroscopically for structural abnormalities and pathological changes with emphasis on reproductive organs. Additionally the number of implantation sites and corpora lutea of each female were recorded. Lungs, trachea and larynx were removed in their entirety. The right middle lobe of the lung was weighed, the remaining lobes were fixed for subsequent histopathological examination. The testes and epididymes of the males were weighed and then fixed for histological examination as were the ovaries of the females."</p> <p>This study has also been reported in the open literature (Bui et al, 1998) but the open literature publication does not contain as much information as the original laboratory report summarized here.</p>

### Test Results - Developmental Toxicity/Teratogenicity

Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):	LOAEL/	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
	LOAEC/ NOAEL/ NOAEC					
NOAEC	Offspring (F1)	>=	24700			mg/m3

NOAEC	Parental (F0)	>=	24700	mg/m <sup>3</sup>
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**Results Remarks:**

The chamber concentrations of test material were found to be between 96 and 104% of nominal, the mean highest dose concentration being 24.7 mg/m<sup>3</sup>. The vapor compositions were also found to be similar to that of the parent test material. No parent animals died or were killed during the study and there were no clinical signs. Body weights and food consumption were unaffected by exposure to test material. Results on developmental toxicity endpoints are summarized below.

At necropsy, the following incidence of developmental endpoint observations (which were not dose related) was recorded:

Dose group  
0 5 12.5 25  
N(%) N(%) N(%) N(%)  
Litters examined 8 8 9 8  
Pups examined 113 114 122 123  
Observations (Litter incidence)

**LIVER**  
Pale left lateral lobe  
0(0) 1(0.9) 0(0) 0(0)  
Patchy tan area both surfaces, all liver lobes  
1(0.9) 0(0) 0(0) 0(0)

**LIMBS**  
Broken rt hind limb  
0(0) 0(0) 1(0.8) 0(0)

**THORACIC CAVITY**  
Adhesion between apex of heart and diaphragm  
0(0) 0(0) 0(0) 1(13)

**HEAD**  
Red focus on rt. side of brain  
0(0) 0(0) 0(0) 1(13)  
Red focus on meninges  
0(0) 1(13) 0(0) 1(13)  
Depression on right ventricle  
0(0) 1(0.9) 0(0) 0(0)  
Red focus (1mmx1mm) on top of brain  
1(13) 1(13) 0(0) 0(0)

**TAIL**  
Fleshy tab at tip of tail  
1(13) 0(0) 0(0) 0(0)  
V ring (constriction)  
0(0) 0(0) 0(0) 1(0.8)  
Necrotic tail tip  
1(13) 1(13) 0(0) 0(0)

**TOTAL PUP NECROPSY OBSERVATIONS**  
Pup 4(3.5) 6(5.3) 1(0.8) 3(2.4)  
Litter 4(50) 3(38) 1(11) 2(25)

Again, as a courtesy for readers, the results for the reproductive toxicity and parental systemic toxicity have been copied here from Reproductive Toxicity summary for the same study:

"Results on reproductive capacity and fertility are summarized in the following table.

Treatment group (g/m<sup>3</sup>)  
Parameter 0 5 12.5 25  
Pregnancy (%) 80 80 100 80  
Litters with live pups 8 8 9 8  
Implantation sites 14.9 16.8 13.9 17.3  
Pups delivered 14.4 15.6 14.3 15.6  
Live pups/litter 14.4 14.8 13.8 15.5  
No. liveborn 115 118 124 124  
Live birth index (%) 100 94 96 99  
Pups surviving 4 days 113 114 122 123  
Viability index (%) 98 97 98 99  
Pup wt./Litter day 1 7.2 7.3 7.1 7.1  
Pup wt./Litter day 4 10.8 11.1 11.2 10.5

There were no treatment-related findings observed at necropsy for parental treated rats. Organ weights were unaffected by treatment and there were no treatment-related histological findings."

**Conclusion:**

NOAECs in Sprague-Dawley rats for Developmental, Reproductive, and Parental Systemic toxicities > 25 g/m<sup>3</sup> [24700 mg/m<sup>3</sup>], which

was the highest dose tested.

No treatment-related adverse developmental, reproductive, or parental systemic effects were observed. All pregnant females had comparable delivery data and pups in all groups showed comparable birth weights, weight gain, and viability at postnatal day 4. No histopathological changes were seen at necropsy for adults or offspring, and reproductive organs of adult animals were normal histologically.

### Reliability/Data Quality - Developmental Toxicity/Teratogenicity

**Reliability:** Valid with Restrictions

**Reliability Remarks:** RELIABILITY: GLP; "adaptation" of guideline study with adequate methods & results description

This developmental study did not include skeletal staining for an examination of structural abnormalities. Nevertheless the study did not demonstrate skeletal abnormalities by gross observation at necropsy.

**Key Study Sponsor Indicator:** Key

### Reference - Developmental Toxicity/Teratogenicity

**Reference:** Bui, Q., Burnett, D. M., Breglia, R. J., Koschier, F. J., Iapadula, E. S., Podhasky, P. I., Schreiner, C. A. and White, R. D. (1998) Toxicity evaluation of petroleum blending streams: reproductive and developmental effects of a distillate from light alkylate naphtha. J. Tox. Env. Health, Part A, Vol 53, pp 121-133

Stonybrook Laboratories Inc (1995) Reproductive/developmental toxicity screening test of light alkylate naphtha distillate in rats Study No. 65874 Stonybrook Laboratories Inc. Princeton, NJ



<b>Developmental Toxicity/Teratogenicity</b>	
<b>Test Substance - Developmental Toxicity/Teratogenicity</b>	
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Developmental Toxicity/Teratogenicity</b>	
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Female
<b>Number of Animals per Dose:</b>	25
<b>Dose:</b>	0, 400, and 1600 ppm [equivalent to 0, 1493, and 5970 mg/m3]
<b>Year Study Performed:</b>	1978
<b>Method/Guideline Followed:</b>	Other
<b>GLP:</b>	No Data
<b>Exposure Period:</b>	10 Days
<b>Frequency of Treatment:</b>	6 hours each day
<b>Post-Exposure Period:</b>	0
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Female rats were mated with sexually mature males of the same strain. The females were examined daily for evidence of a copulatory plug and when this was observed it was designated day 0 of gestation.</p> <p>The mated female rats were assigned sequentially into three groups of 25 animals for the 0, 400 ppm (1493 mg/m3), and 1600 ppm (5970 mg/m3) dose groups and were caged individually. The animals were subjected to whole body exposure to gasoline vapors at the concentrations shown above for 6 ours each day from day 6 through day 15 of gestation. Mated females were weighed on days 0, 6, 15 and 20 of gestation. Food consumption was recorded daily during the periods 0-6, 6-15 and 15-20 days of gestation. Observations were made daily for clinical signs.</p>

On day 20 of gestation the female rats were anesthetized and their visceral and thoracic organs were examined. The uterus was removed and opened and the number of implantation sites, their placement in the uterine horns, live and dead fetuses and resorption sites recorded.

The fetuses were removed, examined externally for abnormalities and weighed. One third of the fetuses from each litter were fixed in Bouin's and examined later for changes in the soft tissues of the head, thoracic and visceral organs. The remaining fetuses in each litter were stained with Alizarin Red S and examined for skeletal abnormalities.

The uterus and ovaries from the adult females were preserved for possible future examination.

### Test Results - Developmental Toxicity/Teratogenicity

Concentration  
(LOAEL/ LOAEC/  
NOAEL/ NOAEC):

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
NOAEC	Female (Maternal)	=	5970		mg/m3
NOAEC	Fetal	=	5970		mg/m3

#### Results Remarks:

Chamber concentrations were found to be:

Nominal ppm Actual ppm Calculated equivalent  
in mg/m3  
0 0 0  
400 442 ± 42 1493  
1600 1573 ± 80 5970

There were no deaths during the study and all animals appeared normal throughout. There were no treatment-related effects on body weight or food consumption in the dams.

There were no treatment related effects on any of the reproductive parameters recorded. These data are summarized as follows:

Historical 0 400 1600  
control ppm ppm ppm  
Pregnancy ratio - 20/22 22/22 20/21  
(pregnant/bred)  
Live litters 99% 20 22 20  
Implantation sites 46/54% 123/145 149/158 143/152  
(left /right horn)  
Resorptions 252 16 22 15  
Litters with resorptions 50% 65% 41% 55%  
Dead fetuses 1 0 0 0  
Litters with 1 0 0 0  
dead fetuses  
Live fetuses/ 92% 95% 93% 95%  
Implantation site  
Mean live litter size 12.2 13 13 14  
Average fetal wt. (g) 3.5 3.8 3.7 3.6

No treatment related effects were observed during the examination for soft tissue changes in the fetuses. Results of the skeletal examination of the stained fetuses are summarized below:

Dose Fetuses Fetuses Fetuses with  
(ppm) examined normal commonly Unusual  
encountered skeletal



changes variations only  
 0 177 (20)\* 112 60 (18) 5 (5)  
 400 197\*\* (22) 128 55 (16) 14 (4)  
 1600 196 (20) 131 47\*\*\* (14) 18\*\*\* (7)  
 \* Average No. of litters in parenthesis  
 \*\* Two specimens of one litter lost on processing  
 \*\*\*  $p < 0.05$

The unusual changes were mainly related to retarded ossification and were not considered as malformations.

Statistical analysis of data on a pup basis revealed a significant difference between the 1600 and 0 ppm groups. However when analyzed on a litter basis no statistically significant differences were found.

**Conclusion:**

NO(A)EL for maternal and developmental toxicity = 1600ppm (5970mg/m<sup>3</sup>), which was the highest concentration tested.

The effects of exposure of pregnant rats to vapors of unleaded gasoline at concentrations of 400 (1493 g/m<sup>3</sup>) or 1600 ppm (5970mg/m<sup>3</sup>) were comparable to concurrent control rats. There were no treatment-related effects on body weight or food consumption. There were no treatment related effects on any reproductive parameter (pregnancy ratio, live litters, implantation sites, litters with resorptions, dead fetuses, litter size, fetal weights), or fetal soft tissue or skeletal examination.

**Reliability/Data Quality - Developmental Toxicity/Teratogenicity****Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

Although the GLP status of this study is unknown, the study methods are well described and follow established teratogenicity study design, with presentation of relevant results.

**Key Study Sponsor Indicator:**

Key

**Reference - Developmental Toxicity/Teratogenicity****Reference:**

American Petroleum Institute (1978) Teratology study in rats unleaded gasoline  
 Study conducted by Litton Bionetics Inc. API HESD Res. Publ. 26-60014



<b>Developmental Toxicity/Teratogenicity</b>	
<b>Test Substance - Developmental Toxicity/Teratogenicity</b>	
<b>Category Chemical:</b>	(64741-41-9) Naphtha, petroleum, heavy straight-run
<b>Test Substance:</b>	(64741-41-9) Naphtha, petroleum, heavy straight-run
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Naphtha, petroleum, heavy straight-run, Colorless liquid. MW 111.25. The test substance is a mixture that contains approximately 225 volatile hydrocarbons. The purity of the mixture is 100%            Stable based on analyses of chamber atmosphere.            12 Representative Components monitored in Study            Component Volume %            2-Methyl C6 + C7-olefin 4.50            3-Methylhexane 3.52            t-1,3-Dimethylcyclopentane 1.45            t-1,2-Dimethylcyclopentane 1.61            n-Heptane 7.23            Methylcyclohexane 6.76            Toluene 3.44            2-Methylheptane 3.25            n-Octane 5.81            Ethylcyclohexane 1.95            m-Xylene 1.71            n-Nonane 4.47</p> <p>Additional compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>). Test material is identified as:            "HPV High Naphthenic Naphtha Test Material            CAS# 64741-41-9 HPU [error, should be HPV] Test Material"</p> <p>Substance is in the Gasoline Blending Streams Category.            See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Developmental Toxicity/Teratogenicity</b>	
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Number of Animals per Dose:</b>	12
<b>Dose:</b>	<p>Target: 0, 100, 500, 3000ppm (0, 455, 2275, 13650mg/m3)            Actual: 0, 100, 520, 2950ppm (0, 455, 2366, 13423mg/m3)            The mean concentrations (<math>\pm</math> SE) representing the total area for the approximately 225 components contained in the test substance over the test period were <math>100 \pm 0.8</math>, <math>500 \pm 2.0</math>, and <math>3000 \pm 8.3</math>ppm in chambers targeted at 100, 500, and 3000ppm, respectively. Results from the cryogenic GC analysis indicated that the components were present in the chamber atmosphere within expected concentrations</p>
<b>Year Study Performed:</b>	2008
	OECD 422

<b>Method/Guideline Followed:</b>	
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	30 - 54 Days
<b>Frequency of Treatment:</b>	6 hours/day, 7 days/week
<b>Post-Exposure Period:</b>	0 Days
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Exposure Period: 30 days for subchronic males used for breeding; approximately 34-47 days for pregnant satellite females included 14 days pre-mating, up to 14 days mating and Gestation days 0-19; and 54 days for females with no evidence of copulation.</p> <p>Concentrations of Naphtha vapor were generated by flash evaporation of the test material. An air control group was also evaluated using a similar generation apparatus; however, no test material was supplied to this vapor generator. Vapor concentrations of Naphtha were measured by gas chromatography (GC) using the area sum function and integrating all of the eluted peaks. Additional air samples were collected weekly and analyzed for 12 of the larger, most representative components of the test substance using a cryogenic GC. Temperature, humidity, and airflow were also recorded periodically during each exposure day. Exposures were conducted for 6 hours per day, 7 days per week.</p> <p>Groups of 12 young, adult, male Crl:CD(SD) rats were exposed to atmospheres containing 0, 100, 500, or 3000ppm of Naphtha for 30 days. Satellite groups of 12 young, nulliparous, non-pregnant female rats were exposed to 0, 100, 500, or 3000ppm during a pre-mating period of approximately 2 weeks, a cohabitation period of up to 2 weeks, and a gestation period of approximately 3 weeks. Following the 2 week pre-mating period, each satellite female was paired with a male of the same respective dosage group during an approximately 2 week cohabitation period. Presumed pregnant females were exposed from gestation day [GD] 0-19 but were not exposed after gestation day 19, or during the approximately 4-day lactation period [LD]. Females without evidence of mating continued to be exposed for 26 days after the end of the cohabitation period.</p> <p>Body weights, clinical signs, and food consumption were recorded throughout the study. Body weight data were collected weekly for males, and satellite females without evidence of copulation. Satellite females were weighed weekly during pre-mating and cohabitation, on GD0, 7, 14, 21. Food consumption data were collected at the same intervals except for non-bred satellite females post cohabitation. After approximately 30 days of exposure, blood samples were collected from all males for measurement of haematology and clinical chemistry parameters. An abbreviated neurobehavioral evaluation was conducted on all males, and satellite females prior to test substance administration in order to obtain baseline measurements, and again during week 4 in the morning prior to daily exposure for males and on lactation day 4 for satellite females with litters [details available in Repeated Dose Toxicity robust summary section for males and Reproductive Toxicity robust summary section for LD4 females]. On postpartum day 4, lactating females and offspring were sacrificed, organs (liver, kidneys, lungs, ovaries with oviducts and uterus with cervix) were weighed, and reproductive organs were evaluated microscopically. Offspring were evaluated for external abnormalities [see Reproductive Toxicity robust summary].</p> <p>Statistical analysis: Preliminary statistical analyses included Levene's test for homogeneity and Shapiro-Wilk test for normality, followed by one-way analysis of variance [ANOVA] and Dunnett/Tamhane-Dunnett's test or Kruskal-Wallis and Dunn's test as appropriate. Analysis of covariance [ANCOVA] and Dunnett-Hsu, or non-parametric ANCOVA was used for pup sex ratio and pup weights. Repeated measure ANOVA with Linear contrasts or Jonckheere-Terpstra trend test was used for motor activity and grip strength.</p>
<b>Test Results - Developmental Toxicity/Teratogenicity</b>	

Concentration (LOAEL/ LOAEC/ NOAEL/ NOAEC):	LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
	NOAEC	Male (Paternal)	>=	13650		mg/m3
	LOAEC	Female (Maternal)	=	13650		mg/m3
	NOAEC	Female (Maternal)	=	2275		mg/m3
	NOAEC	Offspring (F1)	>=	13650		mg/m3

**Results Remarks:**

Systemic effects in males bred to satellite females are presented in detail in the Repeated Dose Subchronic Toxicity Robust Summary for this OECD 422 study.

Mortality did not occur at any exposure concentration. Test substance-related increases in the incidence of stained and wet fur in satellite females were observed in the 3000ppm group; however, this did not adversely impact the health of the animals. Satellite females showed no significant test substance related effects on body weight or weight gain during pre-mating or mating. Test substance related effects on body weight and weight gain were observed in 3000ppm females during the three-week gestation period. Body weight gain at GD 21 was 7% lower than controls, and weight gain from GD0-21 was 14% lower than controls. The lower body weight on GD21 correlates with the statistically significant lower weight on LD0 [See Reproductive toxicity section robust summary] and was considered an adverse effect. No adverse effects on body weight or weight gain were seen in any dams in the 500 or 100ppm groups

Decreases in food consumption correlated with decreased body weight and weight gains for animals in the 3000ppm group. Food efficiency was slightly decreased in 3000ppm satellite females during pre-mating and gestation. No test-substance related effects were seen on food consumption or efficiency in 100 or 500ppm groups

Reproductive/Developmental Toxicology: There were no test substance related significant differences in mean number of pregnant animals, number of animals delivering, mating index, fertility index, pre-coital interval, gestation length, number of corpora lutea, number of implantation sites or percent of post implantation loss for any exposure group. No test substance-related differences were observed in number of fetuses born, live born index, viability index, sex ratio incidence, or clinical observations or mean pup body weight at delivery. One female in the 3000ppm group failed to mate. The duration of gestation was 22 days for controls and 21.9 days for treated groups. There were 12 viable litters in controls, 100 and 500ppm groups and 11 litters in the 3000ppm group. One dam [#434] in the 3000ppm group was not identified as pregnant and delivered her litter during exposure in the chamber. Her mating date could not be determined, pups were small and 5/12 pups died between lactation days 0-4. There were no statistically significant differences in average number of pups born alive: 15.3, 14.3, 15.1 and 13.8 pups in control, 100, 500 and 3000ppm groups respectively. Liveborn index was 100% in all groups. Combined average pup weights at birth were 6.5g, 6.6g, 6.4g, and 6.2g in control, 100, 500 and 3000ppm groups respectively. No overt teratogenic abnormalities were seen in any pup. Pups were allowed to nurse to day 4 of lactation. No soft tissue or skeletal evaluations were performed. The NOAEL for developmental toxicity was ? 3000ppm (13650mg/m3), the highest concentration tested.

**Conclusion:**

Exposure to this heavy straight run naphtha at 3000ppm induced some systemic toxicity in breeding female rats expressed as reduced body weight and weight gain, and slight decreased food consumption at 3000ppm (13650mg/m3), which was the highest dose tested. This naphtha did not induce reproductive or developmental adverse effects and is not considered a developmental toxicant under conditions of this screening procedure.

The NOAEL for developmental toxicity was >= 3000ppm (13,650mg/m3), the highest concentration tested.

Based upon reduced body weight and weight gain, the LOEL for maternal toxicity was = 500ppm (2,275mg/m3); the NOAEL for maternal toxicity was = 3000ppm (13650mg/m3).

The NOAEL for paternal toxicity was  $\geq$  3000ppm (13,650mg/m3), the highest concentration tested.

### Reliability/Data Quality - Developmental Toxicity/Teratogenicity

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** OECD 422 is a Developmental screening protocol, not a complete developmental study. According to OECD 422 protocol, this study did not include skeletal staining for examination of structural abnormalities.

(OECD 422: Combined Repeated Dose Toxicity with the Reproductive/Developmental Toxicity Screening Test)

**Key Study Sponsor Indicator:** Key

### Reference - Developmental Toxicity/Teratogenicity

**Reference:** API (American Petroleum Institute) 2008. OECD 422 inhalation combined repeated dose toxicity study with the reproductive/developmental toxicity screening test of Heavy straight run naphtha [CAS # 64741-41-9]. Haskell Laboratories, Project ID: DuPont -18331. Wilmington, DE.



<b>Developmental Toxicity/Teratogenicity</b>	
<b>Test Substance - Developmental Toxicity/Teratogenicity</b>	
<b>Category Chemical:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Light Catalytic Reformed Naphtha (LCRN-D) Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhqv.org">http://www.petroleumhqv.org</a>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Developmental Toxicity/Teratogenicity</b>	
<b>Route of Administration:</b>	Inhalation
<b>Type of Exposure:</b>	Vapor
<b>Species:</b>	Rat
<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Both M/F
<b>Number of Animals per Dose:</b>	10
<b>Dose:</b>	Target conc.: 750, 2500 & 7500 ppm. (2775, 9250, & 27750 mg/m3) Actual conc.: 750, 2490 & 7480 ppm
<b>Year Study Performed:</b>	2000
<b>Method/Guideline Followed:</b>	OECD 421
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	46 - 51
<b>Frequency of Treatment:</b>	6 hours/day, 7 days/week
<b>Post-Exposure Period:</b>	0
<b>Method/Guideline and Test Condition Remarks:</b>	<p>NOTE - this same study is also described in the Reproductive Toxicity section of HPVIS for this test material.</p> <p>Groups of 10 rats of each sex were exposed to 750, 2500 or 7500 ppm. LCRN-D for 6 hours /day, seven days/week. A group of 10 rats of each sex served as sham treated controls.</p> <p>Parental females were exposed for 14 consecutive days prior to mating, throughout mating and days 0-10 of gestation. Dams and their litters were sacrificed on post partum day 4. Unmated females and parental males were exposed to the test material for 14 days prior to mating, throughout mating and 18 additional days following completion of the mating period. These animals were sacrificed shortly after the last litters were delivered reached post partum day 4.</p> <p>Mating: Within each group one male was co-housed with the same female until evidence of mating was observed (presence of sperm in vaginal smear or copulatory plug). The day of mating was designated day 0 of gestation. Following mating, the females were</p>

housed individually and continued their exposures to test material until day 19 of gestation. Females not showing evidence of mating following a 14 day mating period continued their exposures. If such a female showed signs of being pregnant, it was removed from the exposure regimen and observed for parturition.

Observations: All parental animals were regularly observed for mortality and gross pharmacologic signs. A physical examination, including palpation for tissue masses was carried out daily 30 mins. after removal from the exposure chambers. Body weights and food consumption were measured throughout the study. From day 20 of gestation, females (pregnant and non-pregnant) were observed for signs of parturition. As soon as possible after delivery, litters were observed for the number of live and dead pups and for any abnormalities. Litters were also observed twice daily for unusual findings and dead pups. On days 0 and 4 of lactation, the pups were counted, weighed and sex was determined by external observation.

Pathology: Males were killed as a group shortly after the last litters delivered had reached day 4 of lactation. Females with litters that reached day 4 of lactation were killed the next day or shortly thereafter. Unmated females and those that did not deliver were killed 23 days after completion of the mating period. At post mortem, a complete macroscopic examination was carried out on all adult animals. The following organs were weighed and organ/body weight ratios were calculated: adrenals, brain, heart, kidneys, liver, lung, spleen, epididymes, testes and thymus. Post mortem examination of females included a count of uterine implantation scars when present.

Pups were sacrificed on day 4 of lactation and underwent a complete macroscopic examination and a determination of sex by internal examination. All pups were preserved with viscera intact. Pups found dead at birth and that died prior to day 4 of lactation also underwent a gross external and internal examination. Dead pups were not eviscerated, the intact pups were preserved.

27 tissues were preserved from all adult animals in all dose groups. Ovaries, testes, epididymes, nose with nasal turbinates, and any grossly observed abnormalities were processed and sections examined histologically for all males and female parental animals in the control and highest dose group. Four sections were prepared and examined microscopically of the skull containing the nasal turbinates. These were area between upper incisor and incisive papilla area between incisive papilla and first palatal ridge area between second palatal ridge and first upper molar area between first upper molar and nasopharynx.

Premating exposure period

Male: 2 weeks

Female: 2 weeks

### Test Results - Developmental Toxicity/Teratogenicity

Concentration  
(LOAEL/ LOAEC/  
NOAEL/ NOAEC):

LOAEL/ LOAEC/ NOAEL/ NOAEC	Population	Value Description	Value/Lower Concentration	Upper Concentration	Units
LOAEC	Parental (F0)	=	27750		mg/m3
NOAEC	Parental (F0)	=	9250		mg/m3
NOAEC	Offspring (F1)	>=	27750		mg/m3

#### Results Remarks:

NOTE - this same study is also described in the Reproductive Toxicity section of HPVIS for this test material.

Reproductive/fertility effects: All groups had a mating index and a fertility index of 100% and all animals in all groups had mated within 4 days of cohabitation. Delivery and litter data did not demonstrate any effects of treatment see data summarized below.

Parameter    Dose group (ppm)

	0	750	2500	7500
Females on study	10	10	10	10
Litters with liveborn	10	10	10	10
Implantation sites	147	154	155	154
Mean	14.7	15.1	15.5	15.4
Pups delivered (total)	145	151	146	145
Liveborn	142	151	143	144
Live birth index (%)	98	100	98	99
Pups dying				
Day 0	0	1	1	1
Days 1-4	2	4	0	0
Pups surviving 4 days	140	146	142	143
Viability index (%)	99	97	99	99
pup sex distribution				
Day 0 M/F (ratio)	63/79	67/84	9/74	68/76
Day 4 M/F (ratio)	63/77	64/82	68/74	68/75
Pup weight/litter (g)				
Day 0	6.0	6.6	6.2	6.1
Day 4	9.3	8.9	9.2	9.6

External and internal examination of pups sacrificed on day 4 of lactation resulted in only one pup in a single litter of the control group with abnormalities.

**Conclusion:**

Parental toxicity LOAEC = 7500ppm (27750 mg/m<sup>3</sup>) based on slightly decreased body weight and increased relative liver weight; NOAEC parental toxicity = 2500ppm (9250 mg/m<sup>3</sup>).

NOAEC for reproductive performance/ developmental toxicity >= 7500ppm (27750mg/m<sup>3</sup>), the highest concentration tested.

**Reliability/Data Quality - Developmental Toxicity/Teratogenicity****Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

RELIABILITY: GLP; guideline study

This developmental study did not include skeletal staining for an examination of structural abnormalities, an endpoint which is not included in the OECD 421 repro/dev tox screening study.

**Key Study Sponsor Indicator:**

Key

**Reference - Developmental Toxicity/Teratogenicity****Reference:**

Schreiner, C., Bui, Q., Brelia, R., Burnett, D., Koschier, F., Podhasky, P., White, R., Hoffman, G. and Schroder, R. (2000) Toxicity evaluation of petroleum blending streams: reproductive and developmental effects of light catalytic reformed naphtha distillate in rats. J. Tox. and Env. Health, part A., Vol 60, pp 101-116



# **Mammalian Health Effects**

## **Other**



<b>Skin Irritation</b>	
<b>Test Substance - Skin Irritation</b>	
<b>Category Chemical:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>API Test Material 83-05, Catalytically reformed naphtha (CAS# 68955-35-1)</p> <p>Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>)</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Skin Irritation</b>	
<b>Species:</b>	Rabbit
<b>Mammalian Strain:</b>	Unknown
<b>Type of Coverage:</b>	Occlusive
<b>Preparation of Test Site:</b>	Other
<b>Gender:</b>	Unknown
<b>Number of Animals per Dose:</b>	6
<b>Amount/Concentration Applied:</b>	0.5 ml
<b>Year Study Performed:</b>	1985
<b>Method/Guideline Followed:</b>	Other
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	24 Hours
<b>Total Volume applied and Units:</b>	.5 ml
<b>Control Group Type:</b>	None
<b>Vehicle Used:</b>	No
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Post-Exposure Period:</b>	14 Days
<b>Grading Scale:</b>	Draize scale
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Method: Draize Test</p> <p>0.5 ml of undiluted test material was applied to two areas on each rabbit. One area was intact and the other abraded skin. The treated area was then covered with an</p>

occlusive dressing.

After 24 hours the dressing was removed and the treated skin was wiped to remove any residue of test material. The degree of erythema and edema was recorded according to the Draize scale. A second reading of skin responses was made at 72 hours again at 5, 7 and 14 days. Results of the 24 and 72 hour readings were used to determine the Primary Irritation Index.

### Test Results - Skin Irritation

**Grade:**

**Primary Irritation Index:** 3.1

**Lesions:** none

**Erythema:**

**Edema:**

**Results Remarks:**

Erythema and edema was observed at all evaluation times except the last one at day 14. The scores for erythema and edema at each of the observation times were as follows:

	Erythema		Edema	
	Intact	Abraded	Intact	Abraded
24 h	1.2	1.5	1.5	1.8
72 h	1.5	1.5	1.7	1.8
5 days	1.0	1.3	1.5	1.7
7 days	0.8	1.0	1.0	1.0
14 days	0	0	0	0

The primary dermal irritation index was 3.1, which is considered to be a moderate skin irritant.

Growth rates were normal throughout the study and there were no clinical signs of systemic toxicity.

**Interpretation of Results:** Moderately Irritating

**Conclusion:** The test material was a moderate primary skin irritant under the test conditions; the Primary Dermal Irritation Index was 3.1.

### Reliability/Data Quality - Skin Irritation

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** GLP study with adequately described methods

**Key Study Sponsor Indicator:** Key

### Reference - Skin Irritation

**Reference:** American Petroleum Institute (1985)  
Acute oral toxicity study in rats, acute dermal toxicity study in rabbits, primary dermal irritation study in rabbits, primary eye irritation study in rabbits in API 83-05 full range catalytically reformed naphtha.

Study conducted by Hazleton Laboratories America, Inc.  
API Med research publication No. 32-31474, April 1985.



<b>Skin Sensitization</b>	
<b>Test Substance - Skin Sensitization</b>	
<b>Category Chemical:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance:</b>	(64741-66-8) Naphtha, petroleum, light alkylate
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Sample API 83-19 is a Light Alkylate Naphtha (LAN)</p> <p>Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>)</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>
<b>Category Chemical Result Type:</b>	Measured
<b>Method - Skin Sensitization</b>	
<b>Test Type:</b>	In Vivo
<b>Study Type:</b>	Buehler Test
<b>Species:</b>	Guinea pig
<b>Mammalian Strain:</b>	
<b>Route of Induction:</b>	Epicutaneous, Occlusive
<b>Route of Challenge Exposure:</b>	Epicutaneous, Occlusive
<b>Gender:</b>	Both M/F
<b>Number of Animals per Dose:</b>	10
<b>Concentration:</b>	1st: Induction 50 % occlusive epicutaneous 2nd: Challenge 25 % occlusive epicutaneous
<b>Year Study Performed:</b>	1986
<b>Method/Guideline Followed:</b>	Other
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	6 Hours
<b>Induction Frequency of Treatment:</b>	weekly for 3 weeks
<b>Challenge Exposure Period:</b>	6 Hours
<b>Challenge Frequency of Treatment:</b>	once at 2 weeks post-induction
<b>Total Volume applied and Units:</b>	.4 ml
	Positive

<b>Control Group Type:</b>	
<b>Vehicle Used:</b>	Yes
<b>Vehicle Name:</b>	Other
<b>Other Vehicle Name:</b>	paraffin oil
<b>Vehicle Amount and Units:</b>	
<b>Positive Control Substance:</b>	2,4-dinitrochlorobenzene
<b>Negative Control Substance:</b>	Paraffin oil
<b>Post-Exposure Period:</b>	48 Hours
<b>Method/Guideline and Test Condition Remarks:</b>	<p>0.4 ml of a 50% mixture of test material and paraffin oil was applied under an occlusive dressing to the shorn skin of 10 male and 10 female animals. 6 hours after application the dressings were removed and the skin wiped to remove residues of test material. The animals received one application each week for 3 weeks. The same application site was used each time. 2 weeks following the third application a challenge dose (0.4 ml of a 25% mixture in paraffin oil) was applied in the same manner as the sensitizing doses. A previously untreated site was used for the challenge application. The application sites for sensitizing and challenge doses were read for erythema and edema 24 and 48 hours after patch removal. To assist in the reading of the response to the final challenge dose the test site was depilated 3 hours prior to reading by using a commercially available depilatory cream.</p> <p>Positive control (2,4-dinitrochlorobenzene), vehicle control and naive control groups were included in this study and the procedure for these was the same as for the test groups.</p>
<b>Test Results - Skin Sensitization</b>	
<b>Measurement Period and Units:</b>	48 Hours
<b>Percent Sensitized Test Substance:</b>	0
<b>Percent Sensitized Positive Control:</b>	100
<b>Percent Sensitized Negative Control:</b>	0
<b>Sensitization Score:</b>	
<b>Results Remarks:</b>	At challenge, a very slight erythema was exhibited by one animal. The other 9 animals had no response. In contrast all 20 of the positive controls responded with reactions ranging from slight to severe irritation. Only one naive control exhibited a very slight erythema upon challenge.
<b>Interpretation of Results:</b>	Not Sensitizing
<b>Conclusion:</b>	Test material was not sensitizing in Guinea Pigs tested by the Buehler Test
<b>Reliability/Data Quality - Skin Sensitization</b>	
<b>Reliability:</b>	Valid Without Restrictions

**Reliability Remarks:** GLP study with adequate methods description

**Key Study Sponsor Indicator:** Key

#### Reference - Skin Sensitization

**Reference:** American Petroleum Institute (1986) Acute oral toxicity study in rats, acute dermal toxicity study in rabbits, primary dermal irritation study in rabbits, primary eye irritation study in rabbits, dermal sensitization study in guinea pigs on API 83-19, Light Alkylate Naphtha (CAS 64741-66-8). Study conducted by Hazleton Laboratories. Health and Environmental Sciences Dept. Report 33-30594



## Skin Sensitization

### Test Substance - Skin Sensitization

<b>Category Chemical:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance:</b>	(68955-35-1) Naphtha, petroleum, catalytic reformed
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Test Material: API 83-05, full range catalytically reformed naphtha</p> <p>Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a>)</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhvp.org">http://www.petroleumhvp.org</a></p>

<b>Category Chemical Result Type:</b>	Measured
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### Method - Skin Sensitization

<b>Test Type:</b>	In Vivo
<b>Study Type:</b>	Buehler Test
<b>Species:</b>	Guinea pig
<b>Mammalian Strain:</b>	Other
<b>Other Strain:</b>	Albino
<b>Route of Induction:</b>	Epicutaneous, Occlusive
<b>Route of Challenge Exposure:</b>	Epicutaneous, Occlusive
<b>Gender:</b>	Both M/F
<b>Number of Animals per Dose:</b>	10
<b>Concentration:</b>	<p>Induction: 0.4 ml of 50% occlusive epicutaneous</p> <p>Challenge: 0.4 ml of 25% occlusive epicutaneous</p>
<b>Year Study Performed:</b>	1986
<b>Method/Guideline Followed:</b>	Other
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	6 Hours
<b>Induction Frequency of Treatment:</b>	weekly for 3 weeks
<b>Challenge Exposure Period:</b>	6 Hours
<b>Challenge Frequency of Treatment:</b>	once at 2 weeks post-induction
<b>Total Volume applied and Units:</b>	.4 ml
<b>Control Group Type:</b>	Positive

<b>Vehicle Used:</b>	Yes
<b>Vehicle Name:</b>	Other
<b>Other Vehicle Name:</b>	Paraffin oil
<b>Vehicle Amount and Units:</b>	.4 Other
<b>Positive Control Substance:</b>	2,4-dinitrochlorobenzene, as a 0.3% w/v solution in 80% aqueous ethanol
<b>Negative Control Substance:</b>	vehicle control (paraffin oil) and naive control groups
<b>Post-Exposure Period:</b>	48 Hours
<b>Method/Guideline and Test Condition Remarks:</b>	<p>0.4 ml of a 50% mixture of test material and paraffin oil was applied under an occlusive dressing to the shorn skin of 10 male and 10 female animals. 6 hours after application, the dressings were removed and the skin wiped to remove residues of test material. The animals received one application each week for 3 weeks. The same application site was used each time.</p> <p>2 weeks following the third application a challenge dose (0.4 ml of a 25% mixture in paraffin oil) was applied in the same manner as the sensitizing doses. A previously untreated site was used for the challenge application.</p> <p>The application sites for sensitizing and challenge doses were read for erythema and edema 24 and 48 hours after patch removal. To assist in the reading of the response to the final challenge dose, the test site was depilated 3 hours prior to reading by using a commercially available depilatory cream.</p> <p>Positive control (2,4-dinitrochlorobenzene), vehicle control, and naive control groups were included in this study and the procedure for these was the same as for the test groups. The positive control was used at a concentration of 0.3% in 80% aqueous ethanol for the induction doses and at 0.1% w/v in ~acetone for the challenge dose.</p>
<b>Test Results - Skin Sensitization</b>	
<b>Measurement Period and Units:</b>	48 Hours
<b>Percent Sensitized Test Substance:</b>	0
<b>Percent Sensitized Positive Control:</b>	100
<b>Percent Sensitized Negative Control:</b>	5
<b>Sensitization Score:</b>	
<b>Results Remarks:</b>	<p>There was no abnormal appearance in any of the animals exposed to the test material during the study. The skin reactions to the challenge dose are summarized as follows:</p> <p>Test material</p>



No dermal reactions by any animal  
 Naive control  
 Very slight erythema in 2/20 animals  
 Vehicle control  
 No dermal reactions by any animal  
 Positive control  
 Very slight to moderate irritation by all 20 animals.  
 The reactions of 16 of the animals exceeded the highest  
 reaction observed in the naive positive control animals.  
 Naive positive control  
 10/20 animals exhibited very slight erythema.

**Interpretation of Results:**

Not Sensitizing

**Conclusion:**

Test material was not a sensitizer in guinea pigs  
 assessed in the Buelher Test

**Reliability/Data Quality - Skin Sensitization****Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

GLP study with adequately described methods

**Key Study Sponsor Indicator:**

Key

**Reference - Skin Sensitization****Reference:**

American Petroleum Institute (1986)  
 Dermal sensitization study in guinea pigs, API 83-05, full  
 range catalytically reformed naphtha (CAS 68955-35-1).  
 Study conducted by Hazleton Laboratories America Inc.  
 API HESD Research Publication 33-30497, January 1986.



## Skin Sensitization

### Test Substance - Skin Sensitization

**Category Chemical:** (86290-81-5) Antiknock Gasoline

**Test Substance:** (86290-81-5) Antiknock Gasoline

**Test Substance Purity/Composition and Other Test Substance Comments:** API PS-6 unleaded gasoline sample

[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]

Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <http://www.petroleumhpv.org>)

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

### Method - Skin Sensitization

**Test Type:** In Vivo

**Study Type:** Buehler Test

**Species:** Guinea pig

**Mammalian Strain:**

**Route of Induction:** Epicutaneous, Occlusive

**Route of Challenge Exposure:** Epicutaneous, Occlusive

**Gender:** Both M/F

**Number of Animals per Dose:** 10

**Concentration:** 0.5 ml

**Year Study Performed:** 1979

**Method/Guideline Followed:** Other

**GLP:** Yes

**Exposure Period:** 3 Weeks

**Induction Frequency of Treatment:** 6 hr/day, 3 day/week

**Challenge Exposure Period:** 3 Weeks

**Challenge Frequency of Treatment:** 6 hr/day, 3 day/week

**Total Volume applied and Units:** .5 ml

<b>Control Group Type:</b>	Positive																			
<b>Vehicle Used:</b>	Yes																			
<b>Vehicle Name:</b>	Other																			
<b>Other Vehicle Name:</b>	Mineral oil																			
<b>Vehicle Amount and Units:</b>																				
<b>Positive Control Substance:</b>	0.05% 2,4-dinitrochlorobenzene in ethanol																			
<b>Negative Control Substance:</b>	vehicle and naive control groups																			
<b>Post-Exposure Period:</b>	0																			
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Concentration: 1st: Induction 50 % occlusive epicutaneous 2nd: Challenge 50 % occlusive epicutaneous</p> <p>0.5 ml of undiluted test material was applied under an occlusive dressing to the shorn skin of 10 male and 10 female animals. 6 hours after application the dressings were removed and the skin wiped to remove residues of test material. After the first application, irritation was sufficiently severe that for further dosing a 50% dilution in mineral oil was used. The animals received one application 3 times each week for 3 weeks. The same application site was used each time. 2 weeks following the third application a challenge dose (0.5 ml of a 50% dilution in mineral oil) was applied in the same manner as the sensitizing doses. A previously untreated site was used for the challenge application. The application sites for sensitizing and challenge doses were read for erythema and edema 24 and 48 hours after patch removal. To assist in the reading of the response to the final challenge dose the test site was depilated 3 hours prior to reading by using a commercially available depilatory cream.</p> <p>Positive control (0.05% 2,4-dinitrochlorobenzene in ethanol), vehicle control and naive control groups were included in this study and the procedure for these was the same as for the test groups.</p>																			
<b>Test Results - Skin Sensitization</b>																				
<b>Measurement Period and Units:</b>																				
<b>Percent Sensitized Test Substance:</b>																				
<b>Percent Sensitized Positive Control:</b>																				
<b>Percent Sensitized Negative Control:</b>																				
<b>Sensitization Score:</b>																				
<b>Results Remarks:</b>	<p>On a subjective basis, the challenge treatment did not appear to be more reactive than the sensitizing treatments. The average scores for erythema and edema following induction and challenge are summarized below.</p> <table border="1"> <thead> <tr> <th rowspan="2">Average scores</th> <th colspan="2">PS-6 gasoline</th> <th colspan="2">Positive control</th> </tr> <tr> <th>Erythema</th> <th>Edema</th> <th>Erythema</th> <th>Edema</th> </tr> </thead> <tbody> <tr> <td>Induction</td> <td>0.9</td> <td>0.3</td> <td>1.3</td> <td>0.3</td> </tr> <tr> <td>Challenge</td> <td>0.1</td> <td>0</td> <td>1.9</td> <td>1.7</td> </tr> </tbody> </table>	Average scores	PS-6 gasoline		Positive control		Erythema	Edema	Erythema	Edema	Induction	0.9	0.3	1.3	0.3	Challenge	0.1	0	1.9	1.7
Average scores	PS-6 gasoline		Positive control																	
	Erythema	Edema	Erythema	Edema																
Induction	0.9	0.3	1.3	0.3																
Challenge	0.1	0	1.9	1.7																

The authors concluded that the test material was not sensitizing.

**Interpretation of Results:** Not Sensitizing

**Conclusion:** Test material was not a sensitizer in guinea pigs assessed in the Buehler Test.

#### Reliability/Data Quality - Skin Sensitization

**Reliability:** Valid with Restrictions

**Reliability Remarks:** Although the study was conducted to GLP, the results from the positive controls were not convincing, suggesting that the study may be invalid due to this lack of response of the positive controls. The positive control concentration used in this study was 0.05% 2,4-dinitrochlorobenzene. In similar studies conducted by the sponsor on different test materials, the concentration of the same positive control substance was 0.3% w/v, almost an order of magnitude higher than the concentration used in this study.

**Key Study Sponsor Indicator:** Key

#### Reference - Skin Sensitization

**Reference:** American Petroleum Institute (1980) Acute toxicity tests, API #PS-6 unleaded motor gasoline. Study conducted by Elars Bioresearch Laboratories Inc. API Report No. 27-32130.



## Skin Sensitization

### Test Substance - Skin Sensitization

**Category Chemical:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance Purity/Composition and Other Test Substance Comments:** Test material: API 83-20

Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <http://www.petroleumhvp.org>)

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:** Measured

### Method - Skin Sensitization

**Test Type:** In Vivo

**Study Type:** Buehler Test

**Species:** Guinea pig

**Mammalian Strain:**

**Route of Induction:** Epicutaneous, Occlusive

**Route of Challenge Exposure:** Epicutaneous, Occlusive

**Gender:** Both M/F

**Number of Animals per Dose:** 10

**Concentration:** 0.4 ml

**Year Study Performed:** 1986

**Method/Guideline Followed:** Other

**GLP:** Yes

**Exposure Period:** 6 Hours

**Induction Frequency of Treatment:** weekly for 3 weeks

**Challenge Exposure Period:** 6 Hours

**Challenge Frequency of Treatment:** once at 2 weeks post-induction

**Total Volume applied and Units:** .4 ml

Positive

<b>Control Group Type:</b>	
<b>Vehicle Used:</b>	Yes
<b>Vehicle Name:</b>	Other
<b>Other Vehicle Name:</b>	Paraffin oil
<b>Vehicle Amount and Units:</b>	.4 Other
<b>Positive Control Substance:</b>	2,4-dinitrochlorobenzene, as a 0.3% w/v solution in 80% aqueous ethanol
<b>Negative Control Substance:</b>	vehicle control (paraffin oil) and naive control groups
<b>Post-Exposure Period:</b>	48 Hours
<b>Method/Guideline and Test Condition Remarks:</b>	0.4 ml of undiluted test material was applied under an occlusive dressing to the shorn skin of 10 male and 10 female animals. 6 hours after application the dressings were removed and the skin wiped to remove residue of test material. The animals received one application each week for 3 weeks. The same application site was used each time. 2 weeks following the third application a challenge dose (0.4 ml of a 25% mixture in paraffin oil) was applied in the same manner as the sensitizing doses. A previously untreated site was used for the challenge application. The application sites for sensitizing and challenge doses were read for erythema and edema 24 and 48 hours after patch removal. To assist in the reading of the response to the final challenge dose the test site was depilated 3 hours prior to reading by using a commercially available depilatory cream. Positive control (2,4-dinitrochlorobenzene, as a 0.3% w/v solution in 80% aqueous ethanol), vehicle control (paraffin oil) and naive control groups were included in this study and the procedure for these was the same as for the test groups.
<b>Test Results - Skin Sensitization</b>	
<b>Measurement Period and Units:</b>	
<b>Percent Sensitized Test Substance:</b>	0
<b>Percent Sensitized Positive Control:</b>	100
<b>Percent Sensitized Negative Control:</b>	
<b>Sensitization Score:</b>	
<b>Results Remarks:</b>	No skin reactions were observed following the application of the challenge dose in either the naive controls or the group that had been exposed to test material. Scores of 0.2, 0.3 and 0.5 for erythema were recorded for the paraffin oil controls. In contrast all positive control animals developed a skin response following the challenge procedure.
<b>Interpretation of Results:</b>	Not Sensitizing
<b>Conclusion:</b>	Test material was not a sensitizer as assessed by the Buehler Test
<b>Reliability/Data Quality - Skin Sensitization</b>	
<b>Reliability:</b>	Valid Without Restrictions

<b>Reliability Remarks:</b>	GLP study with adequately described methods
<b>Key Study Sponsor Indicator:</b>	Key
<b>Reference - Skin Sensitization</b>	
<b>Reference:</b>	American Petroleum Institute (1986) Acute oral toxicity study in rats, acute dermal toxicity study in rabbits, primary dermal irritation study in rabbits, primary eye irritation study in rabbits, dermal sensitization study in guinea pigs. Final Report. API 83-20, light catalytic cracked naphtha (petroleum). (CAS#64741-55-5). Study conducted by Hazleton Laboratories Inc. Health and Environmental Sciences Dept. Publ. No. 33-32722



## Carcinogenicity

### Test Substance - Carcinogenicity

**Category Chemical:** (86290-81-5) Antiknock Gasoline

**Test Substance:** (86290-81-5) Antiknock Gasoline

**Test Substance Purity/Composition and Other Test Substance Comments:** Test Material API 99-01 and API 02-08 unleaded gasoline vapor condensate (two batches of the same test material)

[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]

Unleaded baseline gasoline API 99-01 (and API 02-08) Vapor Condensate (BGVC) Test material is a complex mixture of volatile hydrocarbons. The purity of mixture is 100% and stable based on analysis of chamber atmospheres.

Representative Components [98.8%] monitored in Study:

COMPONENT	AREA %
Isobutane	2.70
n-butane	12.78
3-methyl-1-butene	0.41
Isopentane	36.50
n-pentane	9.36
Trans-2-pentene	3.60
2,3-dimethylbutane	1.75
2-methylpentane	7.25
3-methylpentane	4.27
n-hexane	3.62
Methylcyclopentane	1.87
2,4-dimethylpentane	1.36
Benzene	2.75
2-methylhexane	1.73
2,3-dimethylpentane	1.52
3-methylhexane	1.73
Isooctane	1.92
Toluene	3.91

Additional compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <http://www.petroleumhvp.org>). The test materials are referred to as "211(b) Unleaded Gasoline Vapor Condensate Batch A" or "211(b) Unleaded Gasoline Vapor Condensate Batch B"

Substance is in the Gasoline Blending Streams Category.  
See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:** Measured

### Method - Carcinogenicity

**Route of Administration:** Inhalation

**Type of Exposure:** Vapor

**Species:** Rat

**Mammalian Strain:** Fischer 344

**Gender:** Both M/F

**Number of Animals per Dose:** 50

**Dose:** Nominal whole body exposure concentrations:  
0, 2000, 10000, and 20000 mg/m3

Analytical concentrations:  
0, 2020 ± 70, 10100 ± 340, and 20300 ± 740 mg/m3



<b>Year Study Performed:</b>	
<b>Method/Guideline Followed:</b>	Other
<b>GLP:</b>	Yes
<b>Exposure Period:</b>	104 Weeks
<b>Frequency of Treatment:</b>	6 hours/day, 5 days/week
<b>Animals at Interim Sacrifice:</b>	
<b>Interim Sacrifice Time:</b>	
<b>Animals at Final Sacrifice:</b>	
<b>Final Sacrifice Time:</b>	
<b>Control Group Type:</b>	Negative
<b>Method/Guideline and Test Condition Remarks:</b>	<p>In compliance with the Clean Air Act Section 211(b) for fuel and fuel additive registration, the petroleum industry and oxygenate manufacturers have conducted comparative chronic toxicology testing of evaporative emissions from unleaded gasoline (baseline gasoline vapor condensate [BGVC]). Groups of 50 male/50 female CDF(F344)CrlBR rats were exposed in H2000 whole-body inhalation chambers at BGVC vapor nominal concentrations of 0 mg/m<sup>3</sup> (negative control), 2000 g/m<sup>3</sup> (low level), 10000 g/m<sup>3</sup> (mid level), and 20000 g/m<sup>3</sup> (high level) 6 hours/day, 5 days/week for 104 weeks (518 exposure days).</p> <p><b>Test Materials</b>  Baseline gasoline vapor condensate (BGVC, lots API 99-01 and API 02-08) was prepared and supplied in 420-pound and 20-pound gas cylinders by Chevron Research and Technology Center (CRTC, Richmond, CA).  The test materials were fabricated to mimic vapors people would be exposed to during refueling of their vehicle. The starting gasoline for the generation of both vapor condensate samples was certified to meet the 1990 industry average gasoline properties in 40 CFR 79.55. Briefly, vapor condensate, was generated by a single-step distillation from a 1000-gallon Pfaudler glass-lined kettle wherein approximately 15 to 23% of the starting material was slowly vaporized, separated, condensed by chilling, and recovered as test sample. The liquid temperature during collection was approximately 150°F, which resulted in a vapor temperature of approximately 130°F.  Original characterization of the test substances was performed by ExxonMobil Biomedical Sciences, Inc. (EMBSI, 2009). Twenty-pound cylinders and some 420-pound cylinders were stored at ambient temperature in a dedicated storage building. The remaining 420-pound cylinders were stored in an outside, controlled area at ambient temperature. The test substance was transferred, as needed, from the 420-pound to the 20 pound cylinders. Before dispensing test article from each 420-pound tank, a sample was removed from the tank and analyzed by gas chromatography (GC) with flame ionization detection (FID) using a Shimadzu Model GC-17A/FID (Shimadzu Scientific Instruments, Columbia, MD). The gas chromatographic profile of the 18 major peaks (retention time and relative peak area) was compared with that originally determined for the BGVC by ExxonMobil Biomedical Sciences, Inc. (EMBSI, 2009) to ensure the stability of the test mixtures throughout the study.</p> <p><b>Animals</b>  Four hundred-forty CDF(F344)CrlBR rats (5-6 wk old when received) were purchased from Charles River Laboratories (Raleigh, NC) for the study. All animals were quarantined and acclimated to whole-body inhalation chambers for at least 14 d. Healthy animals were randomly assigned by weight to the core exposure groups (400 total; 50 rats/sex/exposure level/test material). Following randomization, the rats assigned to study were identified by tail tattoo. Five unassigned male and female rats were sacrificed before exposures began to evaluate their health status as an indicator of the health status of the population on study. Five male and five female rats were assigned as sentinels and housed in the respective control chamber. Sentinels were screened for health status prior to the beginning of the study and at 6 mo intervals. The animals were anesthetized and blood collected via retro-orbital puncture. Serum was prepared for serology testing and submitted to BioReliance, Rockville, MD, for analysis of antibodies</p>

against common rodent pathogens.

#### Environmental Conditions

The rats were housed 24 h per day during quarantine and exposure in Hazleton 2000 whole-body inhalation chambers. Initially, all rats were housed separately in 3.8-inch wide by 11-inch long by 8-inch high compartments within stainless steel baskets. When the male rats reached 400 g, they were transferred to baskets with 5.7- by 11- by 8-inch compartments. Each chamber contained six baskets. The chambers were held at approximately 1 inch of water negative pressure with respect to the exposure room, and the chamber flow rates were maintained at 12 to 15 air changes per hour (400-500 liters per minute [LPM]). Chamber temperatures were maintained at 20° to 24°C. Temperature, relative humidity, and chamber air flow rates were continuously monitored, 24 h per day. Values for the three parameters were recorded at 30-min intervals. Oxygen concentration in the chambers was maintained at 19%. A 12-h light/dark cycle was maintained with lights on at 0600. Light levels in the exposure room and noise levels in the chambers were determined periodically.

#### Diet and Drinking Water

Unlimited municipal tap water was available at all times. Rats were fed Teklad Certified Rodent Diet (8728C; Harlan Teklad, Madison, WI). Food was available at all times except during the daily exposure period.

#### Experimental Design

Fifty rats/sex/exposure level/test article were exposed 6 h/d (plus T90, the time for the vapor concentration to reach 90% of equilibrium), 5 d/wk for 104 wk (518 exposure days) in Hazleton 2000 whole body chambers. Target total hydrocarbon concentrations were 0 g/m<sup>3</sup>, 2 g/m<sup>3</sup>, 10 g/m<sup>3</sup> or 20 g/m<sup>3</sup> (0, 2000, 10000, or 20000 mg/m<sup>3</sup>) for the control, low-, mid-, and high-level exposures respectively.

#### Inhalation Exposure System

The daily supply of test article for each exposure chamber was contained in 20-pound gas storage cylinders. Exposure atmospheres were generated by controlling the flow of pressurized test article through a rotameter, into a heated stainless steel transfer line where the test article was completely vaporized. Chamber concentrations were controlled by adjusting the flow rates of the test article and dilution air rate. Chamber exhaust was carried to an oxidizer on the roof of the exposure facility where it was burned.

#### Aerosol Generation and Characterization

Vapor concentrations in the exposure chambers were continuously monitored using Miran 1A infrared analyzers (Foxboro Wilks, Foxboro, CT). The high-, mid-, and low-level exposure chambers were each monitored with their own analyzer. A fourth analyzer was devoted to monitoring the control chamber, the room air, and the hood enclosing the 20 pound tank of test substance. The Miran analyzers were operated at a wavelength of 10.4 μm. The path length for the instrument monitoring the low chamber was 15.7 meters, while the path length for the instruments monitoring the mid and high level chambers was 6.75 meters. The Miran 1A analyzers dedicated to the mid- and high-level chambers were calibrated using test article over a range of 6-35 g/m<sup>3</sup> (6000 - 35000 mg/m<sup>3</sup>). The Miran 1A analyzers for the control chamber and the low-level chamber were calibrated over a concentration range of 1-7 g/m<sup>3</sup> (1000 - 7000 mg/m<sup>3</sup>).

#### Qualitative Assessment of Exposure Atmospheres.

The qualitative composition of the exposure atmosphere in each chamber was determined weekly by gas chromatography using a Shimadzu Model GC-17A/FID. The percent peak area of each of 18 major components was determined and recorded weekly.

#### Determination of Nominal Concentration.

Daily nominal or "anticipated" usage was calculated by multiplying the average test article concentration in each chamber (low, mid, high; g/m<sup>3</sup>) by the total flow through each respective chamber ([L/min × min]/1000 m<sup>3</sup>) and then summing the values for all three chambers. This value was compared to the actual test article usage determined by taking the difference between the weight of the 20-pound cylinder before and after each exposure.

#### In-Life Observations:

All animals were individually weighed using the Path-Tox® data acquisition system (Version 4.2.2., Xybion, Cedar Knolls, NJ) on study Day -7 (to randomly assign rats to groups by weight), Day -1, weekly for 13 wk, and then every 4 wk thereafter. Thorough clinical examinations were made at randomization, on Day -1, and weekly thereafter.

#### Post-Exposure Endpoints:

##### Gross Necropsy.

A complete gross examination was performed on all animals at final sacrifice and on those animals that died naturally or were sacrificed in a moribund condition. Sacrifices of rats surviving 518 d of exposure occurred during the week

following the last exposure day for each sex. Animals were randomly assigned to a sacrifice day.

#### Necropsy, Lung Harvest, and Tissue Processing.

All study animals received a complete necropsy. Animals were euthanized with an overdose of intraperitoneally injected barbiturate anesthetic (Euthasol®, Virbac AH Inc., Fort Worth, TX). Body weights and fresh organ weights were collected on lungs, liver, kidneys, adrenals, testes, epididymides, ovaries, uterus, spleen, brain and heart of final sacrifice and moribund sacrifice animals. Animals found dead received a complete necropsy with tissue collection.

Lungs were gently instilled via the trachea with 10% neutral buffered formalin (NBF) to approximate normal volume. Organs and tissues were immersion fixed in 10% NBF for subsequent histopathologic examination. Tissues were trimmed, processed routinely, paraffin embedded, sectioned at 5 µm and stained with hematoxylin and eosin for histopathologic examination.

#### Histopathology:

All collected tissues and lesions were examined histologically in control (0 g/m<sup>3</sup>) animals, high-level (20 g/m<sup>3</sup>; 20000 mg/m<sup>3</sup>) animals, and dead or moribund animals of all groups. Respiratory tissues (lungs, larynx, trachea and nasal turbinate sections at 4 levels), potential target tissues (testes, kidneys of males) and gross lesions from non-target tissues were examined histologically from final sacrifice low-level (2 g/m<sup>3</sup>; 2000 mg/m<sup>3</sup>) and mid-level (10 g/m<sup>3</sup>; 10000 mg/m<sup>3</sup>) animals. Nomenclature of proliferative lesions was based on the international harmonized nomenclature recommended by the Rat Nomenclature Reconciliation Subcommittee of the Society of Toxicologic Pathologists (see <http://www.toxpath.org>; Standardized Rat Nomenclature). Nomenclature for other lesions was routine, widely understood usage (see Boorman et al., 1990).

#### STATISTICAL ANALYSIS

##### Body and Organ Weights:

Group mean body weight, organ weight, percent organ-to-body weight, and percent organ-to-brain weight data were tested for statistical significance using Path-Tox® software. After testing for an overall trend among test groups by an analysis of variance, Bartlett's test was used to establish the homogeneity of the data. If the data were homogeneous, group differences were evaluated using a modified Dunnett's test. If data were non-homogeneous, group differences were assessed using a modified t-test. Significance levels were set at p less than or equal to 0.05.

##### Survival Analysis:

The probability of survival was estimated by the Kaplan-Meier product-limit method using PROC LIFETEST in SAS Version 8.2 (SAS Institute, Cary, NC). Mean numbers of survival days and time to 25% mortality were estimated for each exposure group by the PROC LIFETEST program. Log-rank tests were used to test the hypothesis that there are differences among the four groups for each sex. The significance level was set at p = 0.05. All reported p-values for the survival analysis are two-sided.

##### Histopathology:

The incidences of all neoplastic and non-neoplastic lesions are given as the ratio of the number of affected animals to the number of animals with the site examined microscopically. Three statistical evaluations were performed on the histopathology lesion incidence data: 1) Cochran-Armitage test, which tests whether the incidence of lesions shows a trend across exposure groups; 2) logistic regression test that takes death date into account when assessing the presence of an exposure-dependent trend; and 3) the Fisher's exact test, which compares incidences among the four exposure groups. The two-sided significance level was set at p = 0.05. If a significant difference was detected by the Fisher's exact test, six possible pair-wise comparisons were calculated. Using the Bonferroni correction for pair-wise comparisons, each pair-wise comparison would be considered significant if p < 0.008.

Fisher's exact test and the Cochran-Armitage test do not use survival information and are appropriate in situations where survival is similar among exposure groups as is the case for this study. The Fisher's exact test tests the null hypothesis of equality of prevalences across exposure groups against the alternate hypothesis that the prevalences are not equal while the Cochran-Armitage analysis tests the null hypothesis of equality across exposures against the alternate hypothesis of a monotonic increasing or decreasing trend. Additionally, differences between groups with regard to both the severity and incidence of non-proliferative lesions were analyzed by the Kolmogorov-Smirnov two-sample, one-tailed test as performed by the Path-Tox system. The significance level was set at p = 0.05.

### Test Results - Carcinogenicity

**MTD Indicator:**

**Neoplastic Effect:** MALES: Significant increases in kidney adenoma and carcinomas; increased trends in testicular mesothelioma, nasal squamous cell and thyroid follicular cell carcinomas.

FEMALES: No significant increases as compared to untreated controls

**Male Survival Rate:****Female Survival Rate:****Total Survival Rate:**

**Clinical Observations:** There were no clinical signs of toxicity attributable to BGVC inhalation. As this was a chronic toxicity evaluation, many of the observations during the second year were related to aging (e.g. mammary masses, jaundice).

**Carcinogenic Effect:** Yes

**Results Remarks:** Survival of the BGVC-exposed rats was not significantly different from that of control rats.

**SUMMARY OF MALE FINDINGS:**

\*Reduced body weight (sporadic organ relative weight increases)  
 \*Chronic progressive nephropathy, treatment-related increase in severity, but not incidence  
 \*Increased nasal respiratory epithelial cell degeneration  
 Also decreased olfactory epithelial cell degeneration; however the meaning of the olfactory finding is unclear  
 \*Kidney adenomas and carcinomas  
 \*Nasal squamous cell carcinoma (possible association)  
 \*Testicular mesothelioma (possible association)  
 \*Thyroid follicular cell carcinoma (possible association)  
 \*CONCLUSION - test material was carcinogenic in MALE rats

**SUMMARY OF FEMALE FINDINGS:**

\*Reduced body weight (sporadic organ relative weight increases)  
 \*Increased nasal respiratory epithelial cell degeneration  
 Also decreased olfactory epithelial cell degeneration; however, the meaning of the olfactory finding is unclear  
 \*No enhancement of proliferative lesions (hyperplastic lesions, neoplasms)  
 [NOTE: there was increased incidence in mononuclear cell leukemia in low- and mid-level exposed females; however study investigators concluded that this finding was not treatment related]  
 \*CONCLUSION - test material was NOT CARCINOGENIC in FEMALE rats

**BGVC: Incidence of Proliferation Lesions**

Tissue Diagnosis	DOSE (mg/m3)		
Control	Low	Mid	High
(0)	(2000)	(10,000)	(20,000)

**MALES****Kidney**

No. examined	50	50	50	50
Adenoma, renal tubule	1 (2%)	1 (2%)	4 (8%)	0 (0%)
Carcinoma, renal tubule	0 (0%)	0 (0%)	3 (6%)	0 (0%)
Renal tubule adenoma and carcinoma, combined(a)	1 (2%)	1 (2%)	7 (14%)	0 (0%)

**Nasal Passages:**

Turbinate Level	2			
No. examined	50	50	50	50
Carcinoma, squamous cell	0 (0%)	0 (0%)	0 (0%)	1 (2%)
Turbinate Level 3				
No. examined	50	50	49	50
Carcinoma, squamous cell(b)	0 (0%)	0 (0%)	0 (0%)	3 (6%)
Turbinate Level 4				
No. examined	50	50	49	50
Carcinoma, squamous cell	0 (0%)	1 (2%)	0 (0%)	3 (6%)

## Testes

No. examined 50 49 50 50  
 Mesothelioma, malignant(c) 0 (0%) 0 (0%) 4 (8%) 0 (0%)  
 Adenoma, interstitial cell 48 (96%) 46 (94%) 50 (100%) 49 (98%)

## Thyroid

No. examined 50 29 27 50  
 Hyperplasia, follicular cell(d,e) 1 (2%) 0 (0%) 2 (7%) 6 (12%)  
 Avg. severity 0.0 0.0 0.1 0.2  
 Adenoma, follicular cell 0 (0%) 2 (7%) 0 (0%) 2 (4%)  
 Carcinoma, follicular cell(c) 0 (0%) 0 (0%) 2 (7%) 0 (0%)  
 Follicular cell adenoma and carcinoma, combined(g) 0 (0%) 2 (7%) 2 (7%) 2 (4%)

## Spleen

No. examined 50 34 38 50  
 Leukemia, mononuclear cell 32 (64%) 23(68%) 25 (66%) 32 (64%)

## FEMALES

## Spleen

No. examined 50 25 32 50  
 Leukemia, mononuclear cell(c) 13(26%) 14(56%) 18 (56%) 15 (30%)

- (a) Mid-dose turbinate levels 3 & 4 for one animal were autolytic, resulting in an n of 49 (Note: same tumor may occur at more than one turbinate level)  
 (b) Significant trend for increased incidence with increasing exposure concentration, Cochran-Armitage test.  
 (c) Significant trend for increased incidence with increasing exposure concentration, Fisher's exact test.  
 (d) Average of the severity score for all animals examined (both affected and unaffected). Unaffected animals were assigned a severity score of zero.  
 (e) Significant increasing trend with increasing exposure concentration, Cochran-Armitage and logistic tests.

It should be noted that numerous studies have determined that male rat kidney adenomas and carcinomas have little relevance to human risk. Renal tumors develop secondary to alpha-2-microglobulin (alpha 2u) accumulation in the male rat kidney. Alpha-2u accumulation does not occur in humans, hence the lack of risk correlation (EPA, 1991).

The relevance to human risk of thyroid gland proliferative lesions in male rats might also be questioned, given that mechanisms of chemical thyroid carcinogenesis are believed to be different between humans and rodents (USEPA, 1998). Mutation of thyroid follicular cell DNA may lead directly to cancer and is the only mechanism verified to be carcinogenic in humans, but rodents are believed to be more susceptible to carcinogenic processes involving stimulation of thyroid follicular cell growth through disruption of pituitary-thyroid hormonal physiology. Mutagenesis of thyroid follicular cells and hormone disruption were not evaluated in this study.

**Conclusion:**

In summary, chronic BGVC inhalation suppressed body weight in males, and to a greater extent in females, increased the severity of chronic progressive nephropathy in males and caused epithelial degeneration in the nasal passages of both sexes. The degenerative nasal effects were most likely caused by the test material.

Consistent with previous studies and a concurrent Gasoline + MTBE Vapor Condensate chronic study, chronic exposure to BGVC did enhance the development of renal adenomas and carcinomas in male rats. BGVC may also have enhanced squamous cell carcinoma in the nasal passages, testicular mesothelioma, and thyroid follicular tumors in male rats. Consequently, due primarily to treatment-related effects in the male kidney, and to possible treatment-related effects on testes, and nose, and thyroid, chronic inhalation of Baseline Gasoline Vapor Condensate was determined to be carcinogenic in male rats in this study.

Chronic exposure to BGVC did not enhance the development of proliferative lesions (hyperplastic lesions, neoplasms) in female rats. There was an increased incidence in mononuclear cell leukemia in low- and mid-level exposed females, however, study investigators concluded that this finding was not treatment-related. Chronic inhalation of Baseline Gasoline Vapor Condensate was determined not to be carcinogenic in female rats in this study.

**Reliability/Data Quality - Carcinogenicity**

<b>Reliability:</b>	Valid Without Restrictions
<b>Reliability Remarks:</b>	USEPA Clean Air Act mandated chronic inhalation study conducted according to GLPs
<b>Key Study Sponsor Indicator:</b>	Key
<b>Reference - Carcinogenicity</b>	
<b>Reference:</b>	<p>211(b) Chronic Carcinogenicity Study: Baseline Gasoline Vapor Condensate (BGVC). Laboratory (LRRI) study number FY01-027. Lovelace Respiratory Research Institute, Albuquerque, NM. Study conducted for the American Petroleum Institute 211(b) Research Group in compliance of the EPA Clean Air Act 211(b) testing requirements.</p> <p>Benson, JM, Gigliotti AP, March TH, Barr, EB, Tibbetts, BM, Skipper, BJ, Clark, CR, and Twerdok, L. (2011) Chronic carcinogenicity study of gasoline vapor condensate (GVC) and GVC containing methyl tertiary-butyl ether in F344 rats. <i>Journal of Toxicology and Environmental Health, Part A</i>, 74:638-657.</p> <p>ExxonMobil Biomedical Sciences, Inc. (2009) Gasoline vapor condensate characterization. Study Number 167490, ExxonMobil Biomedical Sciences, Inc, Annandale, NJ.</p> <p>Boorman, GA, Eustis, SL, Elwell, JR, and Leininger, JR., eds. (1990). <i>Pathology of the Fischer rat</i>. San Diego, CA: Academic Press.</p> <p>U.S. Environmental Protection Agency. Alpha 2 microglobulin: association with chemically induced renal toxicity and neoplasia in the male rat. 1991. In <i>Risk Assessment Forum</i>. US Government Printing Office, Washington, DC: EPA: 85</p> <p>U.S. Environmental Protection Agency (USEPA). 1998. Risk assessment forum. Assessment of thyroid follicular cell tumors. EPA/630/R-97/002, Washington, DC.</p>



Public Tabs? & soname= Immunotoxicity Reptises - Mammalian Health Effects of Other Substances and Categories HPVIS

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# Endpoint Details

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Immunotoxicity																																							
Test Substance - Immunotoxicity																																							
<b>Category Chemical:</b>	(86290-81-5) Antiknock Gasoline																																						
<b>Test Substance:</b>	(86290-81-5) Antiknock Gasoline																																						
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	<p>Test Material API 91-01 unleaded gasoline vapor condensate</p> <p>[Note - there is no CAS Number for Gasoline in the US TSCA Inventory. CAS Number 68290-81-5 is on the European Inventory and added to the Gasoline Category as a "Supplemental Chemical"]</p> <p>Unleaded baseline gasoline API 99-01 Vapor Condensate Test material is a complex mixture of volatile hydrocarbons. The purity of mixture is 100% and stable based on analysis of chamber atmospheres.</p> <p>Representative Components [98.8%] monitored in Study:</p> <table border="1"> <thead> <tr> <th>COMPONENT</th> <th>AREA %</th> </tr> </thead> <tbody> <tr><td>Isobutane</td><td>2.70</td></tr> <tr><td>n-butane</td><td>12.78</td></tr> <tr><td>3-methyl-1-butene</td><td>0.41</td></tr> <tr><td>Isopentane</td><td>36.50</td></tr> <tr><td>n-pentane</td><td>9.36</td></tr> <tr><td>Trans-2-pentene</td><td>3.60</td></tr> <tr><td>2,3-dimethylbutane</td><td>1.75</td></tr> <tr><td>2-methylpentane</td><td>7.25</td></tr> <tr><td>3-methylpentane</td><td>4.27</td></tr> <tr><td>n-hexane</td><td>3.62</td></tr> <tr><td>Methylcyclopentane</td><td>1.87</td></tr> <tr><td>2,4-dimethylpentane</td><td>1.36</td></tr> <tr><td>Benzene</td><td>2.75</td></tr> <tr><td>2-methylhexane</td><td>1.73</td></tr> <tr><td>2,3-dimethylpentane</td><td>1.52</td></tr> <tr><td>3-methylhexane</td><td>1.73</td></tr> <tr><td>Isooctane</td><td>1.92</td></tr> <tr><td>Toluene</td><td>3.91</td></tr> </tbody> </table> <p>Additional compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>)</p> <p>Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a></p>	COMPONENT	AREA %	Isobutane	2.70	n-butane	12.78	3-methyl-1-butene	0.41	Isopentane	36.50	n-pentane	9.36	Trans-2-pentene	3.60	2,3-dimethylbutane	1.75	2-methylpentane	7.25	3-methylpentane	4.27	n-hexane	3.62	Methylcyclopentane	1.87	2,4-dimethylpentane	1.36	Benzene	2.75	2-methylhexane	1.73	2,3-dimethylpentane	1.52	3-methylhexane	1.73	Isooctane	1.92	Toluene	3.91
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Method - Immunotoxicity																																							
<b>Species:</b>	Rat																																						

<b>Mammalian Strain:</b>	Sprague-Dawley
<b>Gender:</b>	Female
<b>Number of Animals per Dose:</b>	10
<b>Dose:</b>	Target: 0, 2000, 10,000, and 20,000mg/m <sup>3</sup> Actual: 0, 2050, 10,153, and 20,324 mg/m <sup>3</sup>
<b>Year Study Performed:</b>	2005
<b>Method/Guideline Followed:</b>	Other
<b>GLP:</b>	Yes
<b>Vehicle Used:</b>	
<b>Vehicle Name:</b>	
<b>Vehicle Amount and Units:</b>	
<b>Method/Guideline and Test Condition Remarks:</b>	<p>Method/Guideline Followed: Modified hemolytic plaque assay to comply with EPA Clean Air Act 211(b) immunotoxicity testing guidelines Exposure Period: 4 weeks, [20 exposures] Frequency of Whole-Body Inhalation Treatment: 6 hours/day, 5 days/week Post-Exposure Period: None</p> <p>This study was conducted as a satellite study of the 13 week inhalation toxicity study reported in the Repeated Dose section. Baseline Gasoline Vapor Condensate was administered via whole-body exposures to female Sprague Dawley rats at target concentrations of 2000, 10000 and 20000 mg/m<sup>3</sup> for 6 hours/day, 5 days/week for 4 weeks. An Air Control group received nitrogen-enriched air only while in chamber. A separate positive control group was treated by intraperitoneal injection with 50mg/kg cyclophosphamide daily for 4 days prior to sacrifice, the last 4 days of exposure for inhalation females.</p> <p>Four days prior to sacrifice, rats were sensitized by intravenous [tail vein] administration of sheep erythrocytes. Day 4 after antigen sensitization is the peak day for the sRBC [sheep red blood cell] IgM antibody-forming cell [AFC] cell response in rats.</p> <p>On the day after the last exposure blood was collected from the orbital sinus, serum was frozen (-700C) for possible future use [serum was discarded at end of the study], and animals were sacrificed. The thymus of each animal was removed, weighed and preserved in 10% neutral buffered formalin for possible histopathology [No subsequent histopathology was performed]. The spleen of each rat was aseptically removed, weighed and shipped intact in HEPES/EBSS/Gentamicin solution on wet ice for overnight delivery to ImmunoTox Inc, Richmond, VA.</p> <p>Single-cell suspensions were prepared from each spleen, and viability of splenocytes was determined. A 0.1 ml aliquot of spleen cells from each suspension was added to separate test tubes containing 25µl guinea pig complement, 25µl sRBC and 0.5ml warm agar. Each mixture was plated onto a separate petri disk, covered with a microscope cover slip and incubated at 36-380C for 3 hours. The spleen weight, cells/spleen, AFC/10<sup>6</sup> spleen cells and AFC/spleen were determined. The developed plaques were counted using a Bellco Plaque viewer.</p> <p>Each plaque is generated from a single IgM antibody-producing B cell, permitting the number of AFC present in the whole spleen to be calculated. A significant modulation of the IgM AFC response to the T-dependant</p>



antigen (sRBC) compared to vehicle controls indicates that a test agent is capable of modifying the humoral immune response in the whole animal.

Statistical analysis: Data were first tested for homogeneity of variances using the Bartlett's Chi Square Test. Homogeneous data were evaluated by a parametric one-way analysis of variance. When significant differences occur, exposed groups were compared to the vehicle control group using Dunnett's t test. Non-homogeneous data were evaluated using a non-parametric analysis of variance (Kruskall & Wallis). When significant differences occur, exposed groups were compared to vehicle control group using the Gehan-Wilcoxon Test when appropriate. The Jonckheere's Test was used to test for exposure level-related trends across the vehicle and exposed groups. The positive control was compared to the vehicle control using the Student t Test.

**Test Results - Immunotoxicity**

**Test Results:**

The humoral response NOAEC in female Sprague-Dawley rats was  $\geq 20,000$  mg/m<sup>3</sup>, which was the highest dose of Baseline Gasoline Vapor Condensate tested

**Results Remarks:**

No adverse effect was observed on absolute or relative spleen or thymus weight of treated animals weighed at sacrifice. The viabilities of all splenocyte cultures were  $>95\%$ . Spleen weights of treated animals were not significantly different to the vehicle control group, and there was no significant difference in the spleen cell number. For test substance treated animals, there was no statistical difference in the IgM antibody-forming cell response as compared to the vehicle control group when evaluated as either specific activity (AFC/10<sup>6</sup> spleen cells) or as total spleen activity (AFC/spleen). Cyclophosphamide (the positive control) as expected produced a significant decrease on both relative and absolute weights of both spleen and thymus compared to vehicle control. Spleen cell numbers were decreased when compared to the vehicle control. The positive control produced a significant decrease in specific activity (100%) and total spleen cell activity (100%) when compared to the vehicle control. Baseline gasoline vapor condensate did not adversely affect the humoral immune response of female SD rats in this assay system.

The NOAEC  $\geq 20,000$  mg/m<sup>3</sup>.

**Conclusion:**

Baseline Gasoline Vapor Condensate administered by inhalation to female rats for 4 weeks did not result in alterations of the humoral immune response as evaluated in the IgM anti-body forming cell response to the T-dependent antigen sheep erythrocytes. There was no statistically significant effect on spleen weight, spleen cell number or IgM antibody production evaluated as specific activity or as total spleen activity.

The humoral response NOAEC in female Sprague-Dawley rats was  $\geq 20,000$  mg/m<sup>3</sup>, which was the highest dose of Baseline (Unleaded) Gasoline Vapor Condensate tested.

**Reliability/Data Quality - Immunotoxicity**

**Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

GLP study conducted by methods stipulated by EPA in Clean Air Act section 211(b).

HPV Supporting study from Section 211(b) Testing Consortium, Fuels and Fuel Additives Health Effects Testing Regulation, administered by API, Washington DC

**Key Study Sponsor Indicator:**

Not Key

**Reference - Immunotoxicity****Reference:**

Baseline Gasoline Vapor Condensate: A 13-Week Whole Body Inhalation Toxicity Study in Rats with Neurotoxicity Assessments and 4-Week In Vivo Genotoxicity and Immunotoxicity Assessments. HLS Study No. 00-6125: Vol IV, Appendix Z. Immunological Evaluation of Baseline Gasoline Vapor Condensate Using the Plaque Forming Assay. Kimber White, Principal Investigator, ImmunoTox Inc Study Designation ITI-900. 2005. Huntingdon Life Sciences Laboratories, East Millstone, NJ and ImmunoTox Inc., Richmond, VA



## Neurotoxicity

### Test Substance - Neurotoxicity

**Category Chemical:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance:** (64741-55-5) Naphtha, petroleum, light catalytic cracked

**Test Substance Purity/Composition and Other Test Substance Comments:** Test material PPSC-LLCN (light catalytically cracked naphtha), CAS# 64741-55-5  
Test was conducted by the Petroleum Product Stewardship Council

Compositional information on this substance can be found in the Analytical Data attachment for the Gasoline Blending Streams Category (at <http://www.petroleumhpv.org>)

Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhpv.org>

**Category Chemical Result Type:** Measured

### Method - Neurotoxicity

**Species:** Rat

**Mammalian Strain:** Sprague-Dawley

**Gender:** Both M/F

**Number of Animals per Dose:** 16

**Dose:** The actual concentrations for each of the target dose levels were:

Dose group nominal (ppm)	Actual (ppm)	Actual in (mg/m3)
0 (Control)	0	-0-
750	756	2300
2500	2507	7700
7500	7533	23400

\* TMC = Total Mass Aerosol Concentration

**Year Study Performed:** 2001

**Method/Guideline Followed:** Other

**GLP:** Yes

**Vehicle Used:**

**Vehicle Name:**

**Vehicle Amount and Units:**

**Method/Guideline and Test Condition Remarks:** Neurotoxicity evaluations (neuropathology, motor activity, and functional observational battery) were included in a standard inhalation subchronic study, conducted according to EPA OTS 798.2450 guideline. See repeated-dose RSS for complete description of study; CAS # 64741-55-5 with the study reference given below

Briefly:  
Groups of 16 male and 16 female rats underwent whole body exposures to 750, 2500 and 7500 ppm LCCN. Exposures were

for 6 hours each day, 5 days per week, for at least 65 exposures, over a period of 15 weeks. Extra groups of 16 rats of each sex were exposed to the high dose level and also for a recovery control group. These animals were maintained untreated for 28 days following cessation of the 15 weeks exposure.

Neurobehavioral evaluations of motor activity and functional activity were performed pretest and during weeks 5, 9, 14/15 and after the 4 week recovery period for the recovery animals. Animals were not exposed to LCCN during these tests.

Following 15 weeks of exposure, 16 animals/sex/group were necropsied and microscopic examination was performed on selected tissues. Nervous tissue from 6 rats/sex/group was also examined microscopically.

A wide range of tissues (39) were removed from the control and high dose animals and were fixed and examined histopathologically. Additionally, kidneys from selected animals were stained with Mallory-Heidenhain and examined. Tissues were also removed from the nervous system (central and peripheral) of all animals for subsequent special staining and histopathological examination. Animals designated for neuropathological examination were subjected to a detailed examination of central and peripheral nervous tissues.

Neurobehavioral studies were undertaken as follows: Motor activity Locomotor activity was monitored as the number of beam breaks in an activity box. Monitoring sessions were for 60 minutes, divided into twelve 5-minute intervals. Evaluation was made pretest and during weeks 5, 9, 15 and at the end of the 4 week recovery period. [A detailed description of the evaluation and analysis is provided in the publication but is not included here.]

Functional Operational Battery An assessment of the following was made:  
Home cage evaluations for Posture, vocalization, palpebral closure.  
Handling evaluations for reactivity to general stimuli, signs of autonomic function.  
Open field behavior: arousal level, gait, urination and defecation frequency, convulsions, tremor, abnormal behavior, piloerection and exophthalmos.  
Reflex assessments for: response to visual and auditory stimuli, tail pinch, pupillary function.

Animals were also evaluated for fore limb and hind limb grip strength, landing foot splay and air righting ability.

The test atmospheres were generated by wholly vaporizing the test material (LCCN) and diluting with air to achieve the required concentrations. The highest concentration was approximately 75% of the lower explosive limit.

Actual exposure concentrations were determined six times per exposure session for treated groups and once for controls.

Particle size determinations were carried out once during each exposure using an aerodynamic particle sizer. Mean mass aerodynamic diameter (MMAD), geometric standard deviation (GSD) and total mass concentration (TMC) were calculated. The actual concentrations for each of the target dose levels were:

Dose group	Actual (ppm)	TMC* (ppm)	(mg/m <sup>3</sup> )
0 (Control)	0	0	0.005820
750	756	0.005506	
2500	2507	0.005085	
7500	7533	0.004348	

\* TMC = Total Mass Aerosol Concentration

### Test Results - Neurotoxicity

<b>Effect Level:</b>	<b>Effect Type</b>	<b>Population</b>	<b>Value Description</b>	<b>Effect Level</b>	<b>Effect Level Upper Value</b>	<b>Units</b>
	NOAEL	Male	>	23400		mg/m3 air (analytical)
	NOAEL	Female	>	23400		mg/m3 air (analytical)
<b>Results Remarks:</b>	<p>See repeated-dose RSS for complete description of study results; CAS # 64741-55-5</p> <p>Neurobehavioral studies: There was no evidence of any effect on motor activity either after 15 weeks exposure or after the 4 week recovery period. There was no evidence of a treatment-related effect in the functional operational battery that was carried out.</p>					
<b>Conclusion:</b>	<p>In both males and females the neurotoxicity NOAEC &gt; 7500 ppm (23,400 mg/m3), the highest dose tested.</p>					
<b>Reliability/Data Quality - Neurotoxicity</b>						
<b>Reliability:</b>	Valid Without Restrictions					
<b>Reliability Remarks:</b>	RELIABILITY: GLP; guideline study					
<b>Key Study Sponsor Indicator:</b>	Key					
<b>Reference - Neurotoxicity</b>						
<b>Reference:</b>	<p>Lapin, C., Bui, Q., Breglia, R., Burnett, D., Koschier, F., Roth, R., Schreiner, C., White, R., Mandella, R. and Hoffman, G. (2001) Toxicity evaluation of petroleum blending streams: Inhalation subchronic toxicity/neurotoxicity study of a light catalytic cracked naphtha distillate in rats. Int. J. Toxicol. Vol 20, pp 307-319</p>					



<b>Neurotoxicity</b>																									
<b>Test Substance - Neurotoxicity</b>																									
<b>Category Chemical:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed																								
<b>Test Substance:</b>	(64741-63-5) Naphtha, petroleum, light catalytic reformed																								
<b>Test Substance Purity/Composition and Other Test Substance Comments:</b>	Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <a href="http://www.petroleumhpv.org">http://www.petroleumhpv.org</a>																								
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<b>Vehicle Name:</b>																									
<b>Vehicle Amount and Units:</b>																									
<b>Method/Guideline and Test Condition Remarks:</b>	See rat inhalation subchronic study (OECD 413) with the same reference as this summary for methods & test material information.																								
<b>Test Results - Neurotoxicity</b>																									
<b>Effect Level:</b>	<table border="1"> <thead> <tr> <th>Effect Type</th> <th>Population</th> <th>Value Description</th> <th>Effect Level</th> <th>Effect Level Upper Value</th> <th>Units</th> </tr> </thead> <tbody> <tr> <td>LOAL</td> <td>Male</td> <td>=</td> <td>27775</td> <td></td> <td>mg/m3 air</td> </tr> <tr> <td>NOEL</td> <td>Male</td> <td>=</td> <td>9250</td> <td></td> <td>mg/m3 air</td> </tr> <tr> <td>NOEL</td> <td>Female</td> <td>&gt;</td> <td>27775</td> <td></td> <td>mg/m3 air</td> </tr> </tbody> </table>	Effect Type	Population	Value Description	Effect Level	Effect Level Upper Value	Units	LOAL	Male	=	27775		mg/m3 air	NOEL	Male	=	9250		mg/m3 air	NOEL	Female	>	27775		mg/m3 air
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NOEL	Male	=	9250		mg/m3 air																				
NOEL	Female	>	27775		mg/m3 air																				
<b>Results Remarks:</b>	A 13 week with 4 week recovery inhalation repeated dose study for CAS#64741-63-5 included evaluation of neurotoxicity (motor activity, functional activity, and nervous tissue histopathology). Please see the Repeated-dose toxicity summary for details.																								
<b>Conclusion:</b>	The male neurotoxicity LOEC was 27,775 mg/m3 based on the increased motor activity in the high dose recovery group.																								

The systemic and neurotoxicity NOEC for male rats was 9,250 mg/m3.

There were no neurotoxic effects observed in female rats; the female NOEC > 27,775 mg/m3.

### Reliability/Data Quality - Neurotoxicity

**Reliability:** Valid Without Restrictions

**Reliability Remarks:** guideline study conducted according to GLPs

**Key Study Sponsor Indicator:** Key

### Reference - Neurotoxicity

**Reference:** Schreiner, C., Bui, Q., Breglia, R., Burnett, D., Koschier, F., Lapadula, E., Podhasky, P., White, R., Hoffman, G. and Mandella, R. (2000) Toxicity evaluation of petroleum blending streams: Inhalation subchronic toxicity/neurotoxicity study of a light catalytic reformed naphtha distillate in rats. J. Tox. and Env. Health, Part A. Vol. 60, pp 489-512



## Neurotoxicity

### Test Substance - Neurotoxicity

**Category Chemical:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance:** (64741-66-8) Naphtha, petroleum, light alkylate

**Test Substance Purity/Composition and Other Test Substance Comments:** Substance is in the Gasoline Blending Streams Category. See Category Analysis Document(s) at <http://www.petroleumhvp.org>

**Category Chemical Result Type:** Derived from Other Endpoint Data

### Method - Neurotoxicity

**Species:** Rat

**Mammalian Strain:**

**Gender:** Both M/F

**Number of Animals per Dose:**

**Dose:**

**Year Study Performed:**

**Method/Guideline Followed:**

**GLP:**

**Vehicle Used:**

**Vehicle Name:**

**Vehicle Amount and Units:**

**Method/Guideline and Test Condition Remarks:** Neurotoxicity was evaluated as part of a rat inhalation subchronic study with 30 days recovery; OECD 413 guideline

#### NEUROTOXICITY ENDPOINTS:

Tissues were also removed from the nervous system (central and peripheral) of all animals for subsequent special staining and histopathological examination. Nervous system tissues were selected randomly from 6 rats/sex/group in the high dose and controls at the end of 13 weeks for microscopic examination. Specific brain regions examined were forebrain, cerebral cortex, hippocampus, basal ganglia, midbrain cerebellum and pons and medulla.

Neurobehavioural studies were undertaken as follows: Motor activity: Locomotor activity was monitored as the number of beam breaks in an activity box. Monitoring sessions were for 60 minutes, divided into twelve 5-minute intervals. Evaluation was made pretest and during weeks 5, 9, 14 and at the end of the 4 week recovery period. [A detailed description of the evaluation and analysis is provided in the publication but is not included here.] Functional Operational Battery: An assessment of the following was made: Home cage evaluations for Posture, vocalization, palpebral closure. Handling evaluations for reactivity to general stimuli, signs of autonomic function. open field behavior: arousal level, gait, urination and defecation frequency, convulsions, tremor, abnormal



behavior, piloerection and exophthalmos. Reflex assessments for: response to visual and auditory stimuli, tail pinch, pupillary function.

Animals were also evaluated for fore limb and hind limb grip strength, landing foot splay and air righting ability.

### Test Results - Neurotoxicity

**Effect Level:**

Effect Type	Population	Value Description	Effect Level	Effect Level Upper Value	Units
NOEL	Male	>	24300		mg/m3 air (analytical)
NOEL	Female	>	24300		mg/m3 air (analytical)

**Results Remarks:**

See repeated-dose RSS for CAS # 64741-66-8 (with the same Schreiner et al, 1998 reference) for additional methods and test material information.

**Conclusion:**

No neurotoxicity was observed in either sex. The neurotoxicity NOEL for both males and females was the highest dose tested, 24,300 mg/m3.

### Reliability/Data Quality - Neurotoxicity

**Reliability:**

Valid Without Restrictions

**Reliability Remarks:**

OECD subchronic guideline study conducted according to GLPs

**Key Study Sponsor Indicator:**

Key

### Reference - Neurotoxicity

**Reference:**

Schreiner, C., Lapadula, E., Breglia, R., Bui, Q., Burnett, D., Koschier, F., Podhasky, P. and White, R. (1998) Toxicity evaluation of petroleum blending streams: inhalation subchronic toxicity/neurotoxicity study of a light alkylate naphtha distillate in rats. J. Toxicol. and Env. Health, Part A, Vol 55, pp 277-296