201-14858

Petroleum HPV

November 24, 2003

The Honorable Michael O. Leavitt, Administrator
U.S. Environmental Protection Agency
P.O. Box 1473
Merrifield, VA 22116

Attention: **Chemical Right-to-Know**
HPV CONSORTIUM
Crude Oil Test Plan and Robust Summary

Dear Administrator Leavitt:

The American Petroleum Institute, on behalf of the Petroleum HPV Testing Group, is pleased to submit the Crude Oil Test Plan and Robust Summary. Our consortium has chosen not to use the HPV Tracker system for submission of our test plans due to the complexity of petroleum substances categories and the associated test plans. We are therefore submitting this test plan, as well as the robust summary, directly to EPA to make available for public comment.

Electronic copies of the test plan (in .pdf format) and robust summary (in .pdf format and as an IUCLID export file) are accompanying this letter via email to the EPA HPV robust summary email address (http://www.epa.gov/chemrtk/srbstsum.htm). This submission is also being sent, via email, to the individuals listed below, including Mr. Charles Auer.

Please feel free to contact me (202-682-8344; twerdokl@api.org) or Tom Gray (202-682-8480; grayt@api.org) with any comments or questions you may have regarding this submission.

Sincerely,

Lorraine Twerdok, Ph.D., DABT
Administrator, Petroleum HPV Testing Program

Cc: C. Auer, USEPA
R. Hefter, USEPA
O. Hernandez, USEPA
Petroleum HPV Testing Group Oversight Committee and Technical Workgroup

HIGH PRODUCTION VOLUME (HPV) CHEMICAL CHALLENGE PROGRAM

TEST PLAN
CRUDE OIL CATEGORY

Submitted to the US EPA

by

The American Petroleum Institute
Petroleum HPV Testing Group

Consortium Registration

November 21, 2003

TEST PLAN

CRUDE OIL CATEGORY

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PLAIN LANGUAGE SUMMARY

This test plan addresses the potential for mammalian and environmental toxicity resulting from exposure to crude petroleum. Crude oils are naturally occurring substances derived from decomposition of plant and animal matter under high temperature and pressure for thousands of years. Crude oil is a complex combination of hydrocarbons consisting predominantly of aliphatic, alicyclic and aromatic hydrocarbons covering the carbon number range from C1 to C60+. It also contains sulfur, oxygen and nitrogen compounds, organometallic complexes notably of sulfur and vanadium, and dissolved gases such as hydrogen sulfide (CONCAWE, 2001). In appearance, crude oils range from thin, light colored oils consisting mainly of gasoline-quality stock to heavy, thick tar-like materials. The chemical composition of crude oils from different producing regions, and even from within a particular formation, can vary tremendously. Crude oils are identified by the predominant proportion of similar hydrocarbon molecules. Paraffinic crude oils are rich in straight chain and branched paraffins, have a high API gravity, low density and viscosity, and contain a higher concentration of gasoline grade naphtha. Naphthenic crude oils contain mainly cycloparaffins and aromatic hydrocarbons, have low API gravity, higher density and viscosity and contain residual materials and heteroatoms (e.g. sulfur, nitrogen, oxygen-containing hydrocarbon analogs).

Mammalian Toxicity Summary

The petroleum crude category contains only CAS Number 8002059 to identify all conventional crude oils and those derived from tar sands, regardless of source or hydrocarbon distribution. Crude oils tested demonstrated similar and relatively low acute toxicity. Crude oils have induced gene mutation in bacteria only after solvent extraction to concentrate biologically active aromatic hydrocarbons in the test fraction. Crude oils tested in rat and mouse micronucleus assays yielded negative results, although one crude did induce sister chromatid exchange in mice. Limited repeat dose studies performed by the oral or dermal routes demonstrated similar effects on hematologic endpoints, enlargement of the liver and thymic atrophy, with some skin irritation from thirteen weeks of dermal exposure. Three developmental toxicity studies suggest that in general, crude oils cause fetal death, decreased fetal weight and delayed skeletal ossification at doses that are maternally toxic. An occurrence of decreased pup viability at Day 4 of lactation was observed from maternal exposure to a heavy crude oil.

Although studies on crude oils are relatively few by comparison to some other petroleum categories, there is a substantial body of data on products derived from crude oils, such as gasoline, diesel fuels, kerosene and jet fuels, lubricating oils and white oils, which are subjects of other HPV test plans. Extrapolation from these studies provides insight into biologically active components of crude oils. Occurrence and severity of toxic effects appear correlated with concentration of polynuclear aromatic hydrocarbons (PAH) and PAH-containing nitrogen or sulfur heteroatoms (PAC). In addition there are significant data developed from monitoring effects of unintentional oil spills, providing “real world” environmental information.

Environmental Summary

Due to their complex composition, crude oils vary widely in their physical/chemical properties. The melting point, boiling point, vapor pressure, partition coefficient and water solubility characteristics of crude oils can differ between oil producing regions as well as within a specific production field. Despite these wide-ranging physical and chemical characteristics, some generalizations can be made regarding the environmental behavior of crude oil. When a release to the environment occurs, components of crude oil will partition into various environmental compartments. The lower molecular weight components may dissolve in water or volatilize to the atmosphere, intermediate fractions may float and spread out on water where they may form emulsions and/or adsorb to soil and sediment, and the viscous, heavy components may agglomerate and float or sink in water or adhere to soil and sediment. The rate at which partitioning occurs depends not only on the nature of the crude but also on the severity of the weathering processes it encounters.

When components of crude oil disperse, they may undergo further chemical and physical transformations. Constituents that partition to the air interact with hydroxyl radicals in the atmosphere and thus are subject to indirect photodegradation. Atmospheric half-lives range from 0.4 days (e.g., n-dodecane) to 6.5 days (e.g., benzene). Crude oils are subject to biodegradation, but biodegradation rates vary considerably, and no crude oils would be considered to be readily biodegradable in standard tests. Low molecular weight components may readily biodegrade, but as molecular weight increases, hydrocarbons become increasingly insoluble in water, so that their bioavailability is limited. In general, hydrocarbons are regarded as being inherently biodegradable, although the degradation rates of the more complex high molecular weight fractions may be very slow.

Crude oil is, in general, harmful to aquatic organisms. In both marine and freshwater environments, a spill event may cause extensive mortality to non-motile susceptible species such as phytoplankton, crustaceans and larvae or eggs of fish and invertebrates. In contrast, spills of crude oil may not acutely affect highly mobile species such as adult fish, and mollusks and polychaete worms have an apparent tolerance to oil contamination. Acute aquatic toxicity of crude oil ranged from 10 to >100 mg/l in studies of whole oil dispersions in water or as water-accommodated fractions (WAFs) (CONCAWE, 2001). Acute toxicity is attributed to those water-soluble hydrocarbon components that are either saturates (aliphatic and alicyclic) or mono- and di-aromatics. Polyaromatic hydrocarbons (PAHs) in crude oil are not expected to contribute significantly to acute aquatic toxicity due to limited bioavailability. However, their partition coefficients (log Kow 3 to >6) indicate they have the potential to bio-accumulate, thus chronic toxicity of PAHs may be a concern.

Other risks to aquatic species, semi-aquatic birds, and sea mammals include physical fouling of plumage, fur, gills etc, by floating oil product. This results in loss of buoyancy, insulation and smothering of inter-tidal animals. Ingestion of oil resulting from attempts by animals to clean contaminated body parts may result in severe enteritis and toxicity.

Spills in freshwater environments have been shown to adversely affect the aquatic macro-invertebrate community, with the observed effects associated with oil sorption and substrate coating. Recovery of such communities in some habitats may be rapid (e.g., riffle areas of streams/rivers), while impacts to backwater areas may persist for months.

Ultimately, the type of crude oil and the local conditions and habitats will dictate the potential and extent to which crude oil persists and cause effects in the environment.

As a result of crude oil spills and continuous long-term release of crude oil components, a plethora of real-world data is available on the acute and chronic effects of crude oil. In summary, there is adequate literature on most of the physical/chemical properties, transformation (fate) processes and aquatic and terrestrial effects endpoints. Where there is not specific 'data', a technical discussion has been prepared to characterize crude oil behavior/impact with respect to the required endpoint based upon information of those products and their components derived from refining of crude oil (e.g. gasoline, lubricating oils, diesel fuels, kerosene, jet fuels, white oils and waxes). Therefore, it is not necessary to propose any further testing for crude oil to satisfy the basic data set requirements for physical/chemical properties, transformation processes or environmental toxicity.

The strategy of this test plan is to provide more definitive data on hazards from crude oil by selecting samples that compositionally reflect the extremes of hydrocarbon distribution. Proposed samples are a light crude oil (high API gravity, high paraffinic, lower aromatic content) and a heavy crude oil (lower API gravity, higher naphthenic and aromatic content) to be tested by the dermal route of exposure in a Combined Repeat Dose Toxicity Study with the Reproductive/Developmental Toxicity Screening Test (OECD protocol 422).

The currently available data on crude oils and refinery streams and the proposed testing outlined in Table 4, combined with chemical characterization, will provide sufficient information to profile mammalian and environmental toxicity for a wide range of naturally occurring crude oils.

Description of the Category

Crude oils are naturally occurring substances formed millions of years ago from the remains of tiny aquatic plants and animals that settled with mud and silt into the bottoms of ancient seas, and the remains of fish and terrestrial animals trapped in swamps and bogs. Successive layers built up, subjecting the remains to high pressures and temperature and causing chemical transformation to hydrocarbons and other crude oil constituents. In many areas crude oil migrated and accumulated in porous rock formations overlaid by impervious rock formations that prevented further travel, geologically speaking, the petroleum trap. Usually a layer of salt water underlies the oil pool.

Among the earliest known uses of natural crude oil and tar, collected from swamps and seepage, was for waterproofing and medicinal purposes. The "fiery furnace of Nebuchadnezzar" is believed to have been an oil seepage that caught fire. In the 1850s, it became known that crude petroleum could be distilled to produce kerosene for illumination, a cheaper and better source of light than whale oil. The industry began with the successful drilling of the first commercial well near Titusville, Pennsylvania in 1859 and the opening of the first refinery in 1862 to process crude into kerosene by atmospheric distillation. Development of the internal combustion engine led to production of gasoline and diesel fuel, and the advent of the airplane created the need for high-octane aviation fuel. A brief chronology of the development of refining processing is found in Appendix 2, Table A2-1 (OSHA, 1993; Mobil, 1997).

Although petroleum in the United States was originally found in Pennsylvania, Texas, California and Louisiana with their offshore fields are now the largest producers of crude oil. Oil reserves also exist in Alaska. The large oil fields of the Middle East provide substantial crude and supplies also come from Canada and South America. Access to oil bearing strata may require drilling as deep as 5 miles underground. Crude oil frequently comes to the surface under great pressure and in combination with large volumes of gas. Gas is separated from oil and processed to remove additional liquids of high volatility to form "natural gasoline" for blending into motor gasoline. The remaining "dry gas" is sold for fuel or recycled back to the underground formations to maintain pressure in the oil pool and thus increase recovery of crude oil. Steam is sometimes injected under pressure into wells to force out remaining oil from a depleted pool.

Types and composition:
Crude oils range from thin, light colored oils consisting mainly of gasoline to thick, black oil similar to melted tar, varying in appearance and composition from one oil field to another. An "average" crude contains 84% carbon, 14% hydrogen, 1-3% sulfur, and approximately 1.0% nitrogen, 1.0% oxygen and 0.1% minerals and salts. Crude oils are composed of paraffinic, naphthenic (cycloparaffinic) and aromatic compounds, and are identified based on the predominant proportion of similar hydrocarbon molecules. Paraffinic crude oils are rich in straight chain and branched paraffins; naphthenic crude oils contain mainly naphthenic and aromatic hydrocarbons. Mixed base crude oils have varying amounts of each type of hydrocarbon. Table 1 provides examples of various crude oil compositions.

Most of these oils would be considered light crude (=33^0 API). The heavy crude oils = 28^0 API) are Prudhoe Bay, Saudi Heavy, Venezuela Heavy, Belridge Heavy. Mid-range crude oils are Kuwait, Venezuela Light, USA West Texas sour.

Table 1: Properties of Whole Crude Oils

Crude Source	Paraffins % vol	Naphthenes % vol	Aromatics % vol	Sulfur % wt.	API gravity (^{0}API)
		Light Crude oils			
Saudi Light	63	18	19	2.0	34
South Louisiana	79	45	19	0.0	35
Beryl	47	34	19	0.4	37
North Sea Brent	50	34	16	0.4	37
Nigerian Light					
Lost Hills Light	Non-aromatics 50%		50	0.9	-
USA Mid Contint. sweet	-	-	-	0.4	40
		Mid range Crude oils			
Venezuela Light	52	34	14	1.5	30
Kuwait	63	20	24	2.4	31
USA West Texas sour	46	32	22	1.9	32
		Heavy Crude oils			
Prudhoe Bay	27	36	28	0.9	28
Saudi Heavy	60	20	15	2.1	28
Venezuela Heavy	35	53	12	2.3	24
Belridge Heavy	Non-aromatics 37%		63	1.1	-

IARC, 1989, Mobil, 1997; OSHA, 1993

Appendices 1 and 2 describe hydrocarbon chemistry and the distribution of products from processing of crude oil. From 1927-1967, the American Petroleum Institute sponsored research in petroleum composition that resulted in the isolation of 266 hydrocarbons from the gaseous, gasoline, kerosene, light gas oil, heavy gas oil, and light lubricant portions of a Ponca Oklahoma crude oil (King, 1988).

Crude oils are further classified by viscosity, specific gravity (density) and by API gravity. API gravity is an indication of the gasoline potential of crude oil; $^0\text{API} = \frac{141.5}{\text{Sp.Gr.}} - 131.5.$

The higher the API gravity (the lower the specific gravity), the more valuable is the crude. A crude with a high API gravity (=33^{0}API, Platts, 2003), and high % hydrogen usually contains a higher concentration of naphtha, which can be processed readily to make gasoline and is considered a light crude. Crude oils with high % carbon and low %

hydrogen are usually rich in aromatics and tend to contain more residual material (e.g. asphalts) and heteroatoms (e.g. sulfur, nitrogen, oxygen-containing hydrocarbon analogs). The “heavy crude oils” require more steps in processing, and are more costly to refine (Appendix 2, Fig. A2-1).

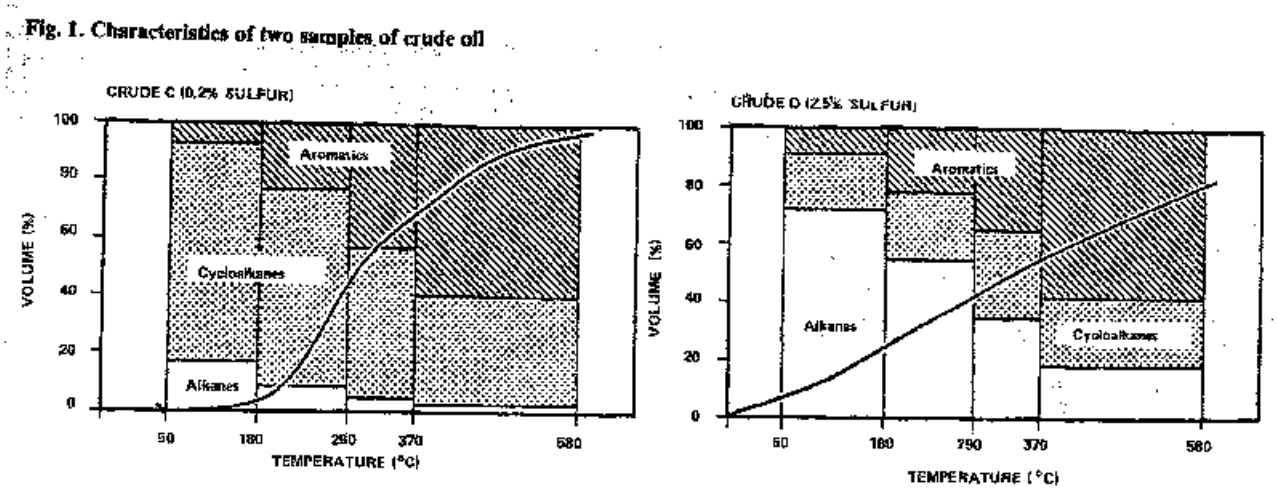
Crude oils also contain varying amounts of non-hydrocarbon sulfur, nitrogen, oxygen and trace metals. Sulfur is present as hydrogen sulfide (H_2S), as thiols, mercaptans, sulfides, benzothiophenes, polysufides, or as elemental sulfur. As a rule, the proportion, stability and complexity of sulfur compounds are greater in heavier crude oil fractions. H_2S is a primary contributor to corrosion in refinery process units and combustion of sulfur-containing petroleum products can produce undesirable byproducts such as sulfuric acid and sulfur dioxide. Total sulfur contents of crude oils spans a range of <0.1% - 5.0% by elemental analysis. In general, as API gravity decreases, sulfur content increases. For example, a light US crude (Rodessa, Louisiana has an API gravity of 42.8 and a sulfur content of 0.28%, while an extremely heavy crude from Venezuela has an API gravity of 9.5 and contains 5.25% sulfur (Dickey, 1981; IARC, 1989). Crude oils high in sulfur are designated “sour crude oils”, and those low in sulfur are “sweet crude oils”. Sulfur is removed during refining by catalytic hydrotreating or by caustic wash (sweetening) processes. Nitrogen types include anilines, pyridines, quinolines, pyrols, carbazoles, benzonitrils and amides. Nitrogen is found in lighter fractions as basic compounds and in heavier fractions as non-basic compounds. Total nitrogen varies from <0.01% -1.0% by elemental analysis. Oxygen compounds are generally phenols, ketones and carboxylic acids. Metals found in crude oil include nickel, iron, vanadium, and arsenic in small quantities. These are removed during refining to avoid poisoning of catalysts, and when burning heavy fuel oils, to minimize deposits of vanadium oxide, and nickel oxide in furnace boxes, ducts and tubes. Inorganic salts such as magnesium chloride or calcium chloride are suspended as minute crystals in crude oil or dissolved in entrained water (brine). These salts are removed or neutralized prior to processing to prevent catalyst poisoning, equipment corrosion and fouling.

CATEGORY RATIONALE

The petroleum crude category contains only one CAS number to identify all crude oil regardless of source. The definition for Crude Petroleum, CAS #8002-05-9 on the TSCA Inventory (1985) is “a complex combination of hydrocarbons consisting predominantly of aliphatic, alicyclic and aromatic hydrocarbons. It may also contain small amounts of nitrogen, oxygen, and sulfur compounds. This category encompasses light, medium and heavy petroleum, as well as the oils extracted from tar sands.” Oil extracted from tar sands undergoes upgrading at or near the production site to produce a higher quality, lighter material referred to as synthetic crude oil.

All petroleum crude oils reach the refinery in a natural state, except for the removal of some contaminants (e.g. sediment, water, mineral salts) at the wellhead. Crude oils with very similar boiling ranges may differ considerably in other physical properties, hydrocarbon composition and distribution of paraffinics, naphthenics and aromatics and

sulfur content. These variations are illustrated in Figure 1 for two typical crude oils from the American Petroleum Institute (API): Crude C, a naphthenic crude with low sulfur content, and Crude D, a paraffinic crude with higher sulfur content.



IARC, 1989

Synthetic crude oil, from upgraded tar sands, is compositionally similar to high quality conventional crude oil (>33^{0}API). The conventional technologies such as delayed and fluid coking, hydrotreating, and hydrocracking, used to upgrade heavy crude oils and bitumens, are used to convert tar sands into an essentially "bottomless" crude, consisting of blends of hydrotreated naphthas, diesel and gas oil without residual heavier oils. Table 2 presents side by side comparative data for a Canadian Synthetic crude oil and a high quality Light Louisiana Sweet Crude, showing the similarity in paraffin, naphthene and aromatic content over the gasoline boiling range and a lower sulfur and metal [nickel, vanadium] content in the synthetic crude oil. This information was supplied to EPA under the requirements of the 211(b) Fuel and Fuel Additives Test Rule [40CFR Part 79, 6/27/94) to support the position that tar sands-derived synthetic crude oil is comparable to conventional crude oils for health effects and environmental testing, a position with which EPA concurred.

TABLE 2. Specification Comparisons: Petroleum and Synthetic Crude Oils

	Canadian Synthetic Crude	Lt Louisiana Sweet Crude
^{0}API	32.2	35.3
Sulfur, (wt %)	0.16	0.49
Viscosity SUS@100^{0}F	38.6	41.8
Nickel ppm	4.1	7.7
Vanadium ppm	5.4	11.0
Hydrocarbons: Avg Vol % over gasoline boiling range [IBP-400^{0}F]		
Paraffinic (vol%)	69	72
Naphthenic	27	24
Aromatics	14	12

Courtesy of Marathon Ashland Petroleum LLC, 1994

Human exposure to petroleum crude oils is primarily through skin contact, however some airborne exposure to crude oil components, such as hydrogen sulfide, mercaptans and gaseous and volatile hydrocarbons can occur via explosive events at wellheads, during transport and in the refinery. Known carcinogens such as benzene, certain polycyclic aromatic hydrocarbons, nickel and arsenic compounds are found in crude oil. Environmental exposure to marine organisms by dermal and oral routes occurs from accidental spills or spillage during loading for transport. Exposure to terrestrial species also occurs due to spillage and tank leakage (IARC, 1989).

Review of existing health effects data indicates that acute toxicity is relatively low and similar over a range of crude oils: oral LD50 rats> 5.0g/kg, dermal LD50 rabbits> 2.0g/kg. Skin irritation was minimal and skin sensitization unlikely; some eye irritation has been observed with a heavy oil sample. Crude oils induced gene mutation in *Salmonella typhimurium* when extractions of polycyclic aromatic compounds (PAC) rich in 3-7 ring polynuclear aromatic hydrocarbons (PAH) and N- or S- containing heteroatoms, were tested with metabolic activation. Limited repeat dose studies performed by the oral and dermal routes demonstrated similar effects on hematological endpoints, enlarged liver and thymic atrophy. These effects were also reported in sea otters exposed to Prudhoe Bay crude from the Valdez oil spill (Alaskan Crude Oil Conference, 1989).

Although few large-scale studies have been performed with crude oils, significant resources have been committed to performing mammalian and environmental toxicity studies on products derived from refining of crude oil (e.g., gasoline, lubricating oils, diesel fuels, kerosene, jet fuels, white oils and waxes). Extrapolation from these refinery stream studies and from available data from mammalian studies with crude oil either in the laboratory or from mammals exposed to environmental spills indicates that crude oil-induced mammalian toxicity is correlated with increasing levels of aromatic components rich in 3-7-ring PAC. The lighter the crude oil, the lower the PAC levels, and the heavier the crude oil, the higher the PAC levels.

For environmental endpoints, the toxicity of crude oils does not correlate with levels of 3-7 ring PAC, but is defined by the range of components of crude oil in general. The majority of constituent chemicals are neutral organic hydrocarbons whose toxic mode of action to aquatic organisms is non-polar narcosis. Acute toxicity is attributed to those water-soluble components that are either saturates (aliphatic or alicyclic) or mono- or di-aromatics. Polyaromatic hydrocarbons (PAH) in crude oil are not expected to contribute significantly to acute aquatic toxicity due to limited bioavailability, but by bioaccumulating, may contribute to chronic effects on aquatic organisms, the benthic community and fish species. Bioaccumulation is species dependent, and many aquatic organisms have shown the capacity to metabolize PAHs and thus do not bioaccumulate to levels predicted by partitioning characteristics. The extent of bioaccumulation depends on the species as well as the type of PAH compound (Eisler 1987).

TEST MATERIALS

The Petroleum HPV testing program proposes to test a light crude oil (high ^{0}API, high paraffinic content), and a heavy crude oil (lower ^{0}API, higher naphthenic, aromatic content) by the most appropriate route of mammalian exposure (dermal administration) to cover the extremes of component composition of materials in this category. Results from these studies, combined with published data and environmental monitoring following accidental releases of crude, will be used to delineate the range of toxicity from petroleum crude oils.

EVALUATION OF EXISTING HEALTH EFFECTS DATA and PROPOSED TESTING

Results of studies from various crude oils are summarized in this section. Studies are comprised of both laboratory and environmental monitoring data collected from monitoring species exposed by unintentional oil spills. Detailed study information is available in the Robust Summaries organized in the IUCLID data set format employed by the European Union (Appendix 3). The currently available data submitted to the HPV program and any additional testing will be developed with the goal of facilitating international harmonization of hazard and risk characterization worldwide.

Mammalian Toxicity

Acute Toxicity

Acute oral and dermal toxicity and eye and skin irritation data from 5 crude oils [4 light crude oils and 1 heavy crude] are summarized in Table 3. Crude oils were not significantly toxic orally in rats or dermally in rabbits, and demonstrated minimal skin irritation. Only Lost Hills light crude induced substantial conjunctival irritation at 24 hours. In a separate study of 3 samples of crude oils, Smith (1981) reported oral toxicity in the mouse as LD_{50} ranging from >10.0g/kg (mixed petroleum crude oils) to >16.0g/kg (Wilmington crude 18^{0}API; Recluse crude).

Table 3. Acute Toxicity of Crude Oils

Sample	Oral LD_{50} (Rat) g/kg	Dermal LD_{50} (Rabbit) g/kg	Skin Irritation (Rabbit)[a]		Eye Irritation (Rabbit 24hr) Conjunctiva
			Erythema	Edema	
Beryl [36.5^{0}API]	>5.0	>2.0	ND	ND	1.7
Arab Lt [34.5 API]	>5.0	>2.0	0.9	0.1	1.3
Mid-Continent [40^{0}API]	>5.0	>2.0	ND	ND	0.3
Lost Hills Light	>5.0	>2.0	1.6	1.3	3.7
Belridge Heavy	>5.0	>2.0	0.6	0.8	0.8

a- Mean scores of reactions at 24, 48, and 72 hrs. Mobil (1984a, 1985, 1990a)

Evaluation of Lost Hills Light and Belridge Heavy for sensitization in the guinea pig (Bueler method) indicated that crude oils were unlikely to cause skin sensitization (Mobil, 1991a,b).

Summary: Crude oils show little toxicity from short duration exposure. **There are sufficient data to characterize acute toxicity endpoints for a range of light to heavy crude oils and no additional testing is proposed.**

Repeat Dose Toxicity

Three crude oils (Arab Light, 34.5^{0}API Prudhoe Bay, 28^{0}API, heavy; and South Louisiana. 35^{0}API, light) were administered orally to CD-1 mice once daily for 5 days (Leighton, 1990). Prudhoe Bay was administered at concentrations of 0, 2, 4, 8, 10, 12, or 16ml/kg/day; Arab Light and South Louisiana were given at 10ml/kg/day only. All three crude oils induced minor hematologic changes, liver enlargement and thymic and splenic atrophy without concurrent pathological effects on tissue structure. Liver enlargement was considered an adaptive, physiological response and thymic atrophy a non-specific, stress-related secondary effect.

Lost Hills Light (LHL- low viscosity, 50.0 wt% PAC, 0.86 wt% S) and Belridge Heavy (BH - high viscosity, 63 wt% PAC, 1.05 wt% S) crude oils were administered dermally to the clipped backs of male and female Sprague Dawley rats at dose levels of 0, 30, 125, and 500mg/kg/day for 13 weeks, 5 days/wk; sites were not occluded (Feuston et al., 1997b).
Effects of dermal exposure at 500mg/kg of heavy crude oil [Belridge Heavy] included reduced mean body wt gain in male rats only, depressed hemoglobin and hematocrit counts in both sexes and decreased red blood cell counts in females. Liver weight was increased and thymus weight decreased in both sexes at 500mg/kg and relative liver weight was increased in males of the 125mg/kg group. Microscopically, thymic atrophy was observed in most 500mg/kg rats (both sexes) but hypertrophy and hyperplasia of the thyroid was only observed in some 500mg/kg males and no females. Hyperplasia of treated skin was slightly less severe in BH treated rats than in those exposed to the light crude oil. Dermal exposure to the light crude oil [Lost Hills Light} at 500mg/kg did not affect body weight or body weight gain but did depress hemoglobin and hematocrit counts in male rats only. Increased liver weight was seen in both sexes at 500mg/kg but no significant weight changes were seen in the thymus. Microscopically, hyperplasia of treated skin was slightly more severe than with exposure to heavy crude oil. Thymic atrophy occurred in only a few rats compared to most rats treated with the heavy crude oil at 500mg/kg. Hypertrophy and hyperplasia of the follicular epithelium of the thyroid was seen in some male rats at all dose levels but not in females. LOAEL = 30mg/kg based on irritation and marginal thyroid effects, for both crude oils. However, Belridge Heavy, richer in 3-5 and 4-5 ring polynuclear aromatic compounds (PAC), demonstrated more severe toxicity in target systems than Lost Hills Light. These results correlated with data from similar studies performed on refinery streams which revealed a relationship between endpoints of general toxicity and increasing levels of 3-7 and 4-7 ring PAC (Feuston et al., 1994; Mobil, 1992a,b).

Summary: The available data from oral and dermal repeat dose toxicity studies on crude oils demonstrate similar toxic effects for both light and heavy crude oils (e.g. hematologic changes, liver enlargement and thymic atrophy) by either route. **These studies are adequate to define the repeated dose toxicity of crude oils.** However, additional systemic toxicity data on a light and a heavy crude will be collected as part of the OECD protocol 422 proposed to evaluate reproductive/developmental toxicity of crude oils.

***In Vitro* Genetic Toxicity**

Gene mutation assays performed with *Salmonella typhimurium* with and without metabolic activation from rodent liver homogenate were negative when neat (unextracted) crude oils (Arab Light, 34.5^0API,light crude, Petrilli et al., 1980); Wilmington, 18^0API, heavy crude, Lockard et al., 1982) were tested. Solvent extraction of crude oils with dimethyl sulfoxide produced a polycyclic aromatic compound fraction enriched in 3-5 and 3-7 ring PAH and heteroatoms, which when tested in the modified Ames test with *S. typhimurium* strain TA98 with metabolic activation resulted in increased incidence of revertant colonies for several oils – light crude oils - Arab Light, Beryl, Mid-Continent, and Belridge Heavy. Lost Hills Light which contains less aromatic constituents was negative for bacterial mutagenesis (Mobil, 1984b, 1990b). *In vitro* studies employing mammalian cells did not demonstrate genetic damage from exposure of neat Belridge Heavy or Lost Hills Light to Chinese Hamster ovary cells (Mobil, 1991c,d), or transformation from exposure of Syrian Hamster ovary cells to Wilmington crude (Lockard et al., 1982). Wilmington crude also did not induce sister chromatid exchange *in vitro* (Lockard et al., 1982).

Summary: *In vitro* gene mutation has been demonstrated in bacterial assays for a variety of crude oil extracts. Where activity is present, it occurs with metabolic activation only and severity is correlated with higher ratios of PAC. *In vitro* tests with neat (unextracted) crude oils in bacterial or mammalian cells are negative, due to limited solubility of the whole oil in aqueous medium and possible competition of non-biologically active components for available metabolic sites. **There are sufficient data to characterize the *in vitro* genetic toxicity endpoint for crude oils and no additional testing is proposed.**

***In Vivo* Genetic Toxicology**

Results of micronucleus assays in Sprague Dawley rats treated dermally with Lost Hills Light or Belridge Heavy crude oils at concentrations of 0, 30, 125 or 500 mg/kg for 13 wks demonstrated that these crude oils did not induce cytogenetic damage in bone marrow of treated rats (Mobil, 1990c;1991e). Administration of Wilmington [heavy] crude oil at the same doses and regime in ICR mice also produced negative results, but a single intra-peritoneal injection at doses of 1.8, 3.6 or 7.2g/kg did induce a slight increase in sister chromatid exchanges in mouse bone marrow, indicative of some DNA perturbation (Lockard et al., 1982).

Summary: The data available are sufficient to characterize the *in vivo* cytogenetic toxicity endpoint for crude oils. **No additional testing is proposed.**

Carcinogenicity:

A number of crude oil samples have been investigated for their potential to cause skin cancer in mouse skin painting studies of 104-110 wk duration. These samples include Naphthenic, Gulf Coast [API Crude C], Paraffinic, high sulfur [API Crude D] (Lewis et al., 1984), San Joaquin Valley [21^0API] (Clark et al., 1988), and Wilmington crude (Renne et al., 1981). All crude oils produced skin tumors in 33-100% of mice with latency periods of 40-76 wks, and are considered dermal carcinogens. Tumor incidence and latency depended on crude oil composition and dose. In a limited initiation study, mice were treated with a single dermal dose (50µl) of South Louisiana [light] crude, followed by treatment with a phorbol ester promoter for 180 days. Calkins et al (1981) concluded that South Louisiana crude oil was not a tumor initiator. Interestingly, this conclusion is not inconsistent with the skin painting results since compounds which induce tumors after fairly long latency periods to first tumor, are frequently assumed to act by promoting the expression of genetic events induced by initiating compounds rather than causing these events directly. In the International Agency for Research on Cancer (IARC, 1989) evaluation of carcinogenic risk to humans, crude oils were assigned to Group 3 – not classifiable as to carcinogenicity to humans, based on "inadequate evidence" for carcinogenicity in humans, and 'limited evidence" for carcinogenicity in experimental animals.

Summary: These studies are summarized for information only and are beyond the scope of HPV testing. **No testing is proposed**

Reproductive and Developmental Toxicity

Prudhoe Bay crude oil (28^0API, heavy crude) was administered orally to Sprague Dawley pregnant rats in a single dose at levels up to 10ml/kg on specified days of gestation or as repeated doses up to 2ml/kg/day on gestation days (GD) 6-17 (Khan et al., 1987). Increased incidence of resorptions, increased fetal death and decreased fetal weight were observed with both treatment regimes at maternally toxic doses. NOEL maternal and developmental toxicity = 893mg/kg.

Lost Hills Light (LHL- low viscosity, 50 weight% PAC, 0.86 weight% S) and Belridge Heavy (BH- high viscosity, 63 wt%, 1.05 wt% S) were evaluated for pre- and post-natal developmental toxicity by the dermal route (Feuston et al., 1997a; Mobil, 1991f,g). LHL was applied to clipped backs of presumed pregnant rats at concentrations of 0, 125, 500, 1000 (postnatal group only) and 2000 (prenatal group only) mg/kg/day on GD 0-19. BH was applied under the same regimen at concentrations of 0, 30, 125, and 500mg/kg/day. Application sites were not occluded. Prenatal rats were killed on GD20; postnatal rats delivered naturally and remain, untreated, with their litters until sacrifice at 3-4wk postpartum. Both crude oils produced maternal and developmental toxicity. Maternal effects included slight (LHL) to moderate (BH) skin irritation, lower body weight gain and increased relative liver weights at concentrations = 500mg/kg for both crude oils. Parturition was delayed in the BH 500mg/kg group. A significant increase in resorptions with concomitant decrease in litter size was observed in animals exposed to LHL at 2000mg/kg and the BH at 500mg/kg. The 4-day pup viability index was decreased in all

BH exposed groups. Reductions in mean fetal body weights were observed at these doses and reduced ossification of skeletal elements were also reported. For the light crude oil, Lost Hills Light, developmental effects were observed at doses that were maternally toxic; NOEL maternal toxicity = 125mg/kg, NOEL developmental = 500mg/kg. The heavy crude oil, Belridge Heavy, induced reduction in the day 4 pup viability index at all dose levels; NOEL maternal = 125mg/kg, NOEL developmental could not be established. Generally, the greater severity of effects was seen in animals from groups exposed to Belridge Heavy, compositionally higher in aromatic compounds with 3-5 and 4-5 hydrocarbon rings. These results are consistent with results of studies performed with refinery products derived from these and other petroleum crude oils.

Sperm morphology was examined after 5 days intra-peritoneal injections of Wilmington crude [18^0API, heavy] to mice at levels up to 2.1g/kg/day. Evaluation of samples 35 days after exposure did not reveal any significant increase in the incidence of abnormal sperm (Lockard et al., 1982).

Summary: These studies demonstrate, in general, that petroleum crude oils induce developmental toxicity at doses which are also toxic to the dam and do not appear to be uniquely hazardous to the fetus. Sperm do not appear to be susceptible to crude oil induced toxicity. Decreases in viability at postnatal day 4 observed in pups from dams exposed to Belridge Heavy at both maternally toxic and non-toxic doses, may suggest the possibility of weakened systems and failure to thrive not expressed as overt toxicity at earlier stages of development. More severe effects correlated with higher levels of PAC in the crude sample. **To provide additional reproductive toxicity information, the Reproductive/Developmental Toxicity screen as part of a Repeat Dose systemic toxicity study (OECD protocol 422) is proposed.**

EVALUATION OF EXISTING PHYSICOCHEMICAL AND ENVIRONMENTAL FATE DATA

The physicochemical endpoints for the EPA HPV chemical program include melting point, boiling point, vapor pressure, water solubility, and octanol/water partition coefficient (Kow). Environmental fate endpoints include biodegradation, photodegradation, hydrolysis, and fugacity. Because the HPV substances covered under the testing plan are mixtures of differing compositions, it is not possible to measure or calculate a single numerical value for some of the physicochemical properties. For example, a product that is a mixture of chemicals does not have a melting point, but rather a melting point range. Melting point and boiling point ranges will be reported because these substances are complex mixtures. Where appropriate, values for PC properties will be represented as a range of values according to the product's component composition. Although some data for materials in this category exist, not all of these endpoints are defined and a consensus database for chemicals that represent materials in this category does not exist. Therefore, calculated and measured representative data were identified and a technical discussion provided where appropriate. The EPIWIN© computer model, as discussed in the US EPA document entitled "The Use of Structure-Activity Relationships (SAR) in the High

Production Volume Chemicals Challenge Program. " is used to calculate physical/chemical properties of representative PONA constituents of crude oils. Log P_{ow}, atmospheric oxidation half-lives and environmental media partitioning were calculated for individual hydrocarbon constituents identified previously in crude oils, and the range of these properties are summarized.

Summary: Where measured data does not exist and is impractical to develop, calculated physicochemical and environmental data for selected constituents of crude oils have been developed using the EPIWIN© computer model

Melting point: For complex mixtures like petroleum products, melting point may be characterized by a range of temperatures reflecting the melting points of the individual components. To better describe phase or flow characteristics of petroleum products, the pour point is routinely used. The range of figures quoted in the robust summary, -30 - 30 ° C, are a typical range for the pour point as measured by a standard oil industry procedure (ASTM Method D97; ASTM 1991). Some low wax crude oils have pour points below -30 °C.

Summary: The pour point of various crude oils has been adequately characterized. **No additional testing is necessary**

Boiling point: Distillation temperatures for crude oil range from approximately -1 - 565 ° C at 1013 hPa. The ASTM D86 method measures the distillation range of petroleum products. The figures quoted represent the approximate range for crude oils and are based on (a) the boiling point of n-butane for the lower value and (b) an upper limit quoted by Speight (1990). In practice, atmospheric distillation of crude oil is not practiced above 275-300 °C, to avoid thermal decomposition of the residue. The residue is normally vacuum-distilled in a subsequent operation.

Summary: The distillation range for crude oil has been adequately described. **No additional testing is necessary**

Vapor pressure: The range of vapor pressures has been reported from 6 - 45 kPa (Jokuty, et al. 2002). Crude oil vapor pressure is a function of oil temperature and composition. Reported values represent vapor pressures of different crude oil types as reported in Environment Canada database.

Summary: The vapor pressure of various crude oils has been adequately measured. **No additional testing is necessary**

Partition Coefficient: The range of partition coefficients for constituent hydrocarbons in crude oil extend from 2 to > 6, based on the calculated log P_{ow} at 25^0C (European Chemicals Bureau, 2000). The calculation was done by the CLOGP Version 3.5 program (Calculation of LOG Partition coefficient octanol/water). The figures represent the spread of calculated and/or measured values for typical hydrocarbon components of crude oil. Calculated values for higher molecular weight hydrocarbons are above 6, but such values

are notional, since no correlation has been established between calculated and experimental values.

Summary: The reported range of partition coefficients represents varied constituent hydrocarbons in different crude oils. **No additional testing is necessary**

Water Solubility: The aqueous solubilities of the main classes of hydrocarbons present in crude oil differ and increase in the order n-alkanes<isoalkanes< cycloalkanes< aromatics (McAuliffe, 1966). Solubilities of crude oil components may extend up to one or two percent individually, however, total solubility of all components will be dictated by component composition and loading rates of oil to water.

Summary: Calculated and measured water solubilities differ for individual components of complex petroleum substances. At any particular loading rate, aqueous concentrations of each component is a function of relative volume of aqueous and petroleum phases, partition coefficient between phases, amount of component present and the maximum water solubility of each component. Aromatics represent the most abundant class of water-soluble hydrocarbons in crude oil. The water solubility of aromatics decreases with increasing number and size of alkyl substituents (McAuliffe, 1966). Although none of the components are appreciably water soluble, crude oils higher in aromatics and naphthenics demonstrate greater solubility than other oils. Available literature describing solubility determinations of components in crude oil are presented in robust summary formats. **No additional testing is necessary.**

Environmental Fate Data

Environmental fate endpoints include biodegradation, photodegradation, hydrolysis, and fugacity. Biodegradation data, available for several representative crude oils in this category, show that these materials can exhibit a moderate to rapid rate of biodegradation. For the photodegradation endpoint, data will be calculated. Most of the hydrocarbon components of crude oils are not subject to hydrolysis at measurable rates, therefore, hydrolysis is not a relevant endpoint for these materials.

A summary of fate processes indicates that most crude oil releases occur in seawater, where wind and wave action contribute significantly to the natural removal processes. Crude oil spreads as a film on the surface of water, facilitating the loss by volatilization of its lighter components. In air, the volatile hydrocarbons are photo-degraded by reaction with hydroxyl radicals, their half-lives varying from 0.5 day (e.g. for n-dodecane) to 6.5 days (e.g. for benzene). The water solubility of crude oil is low, with only the lower molecular weight aromatic hydrocarbons and some polar compounds showing low, but significant solubilities. The dissolved constituents gradually biodegrade in water. Some of the higher molecular weight compounds are removed by emulsification and these also slowly biodegrade; others adsorb to sediment and sink. A further removal process from the water column involving the heavier fraction is agglomeration to form tars, some of which are heavier than water and hence, sink (CONCAWE, 2001).

Equilibrium models are used to calculate chemical fugacity that can provide information on where a chemical is likely to partition in the environment. These data are useful in identifying environmental compartments that could potentially receive a released chemical. Fugacity data can only be calculated. In its guidance document for HPV data development, the US EPA states that it accepts Level I fugacity data as an estimate of chemical distribution values. The input data required to run a Level I model include basic physicochemical parameters; distribution is calculated as percent of chemical partitioned to the 6 environmental compartments within a unit world. Level I data are basic partitioning data that allow for comparisons between chemicals and indicate the compartment(s) to which a chemical is likely to partition in the environment. Calculated environmental partitioning behavior for selected constituents of the crude oils indicate that these chemicals will partition largely to the air, and therefore their fate in air is of environmental interest. The lower molecular weight components will mainly partition to air, with a maximum of about 1% of mono-aromatic hydrocarbons partitioning to water. As the molecular weights increase, there is less tendency for the hydrocarbons to partition to air, the environmental distribution being preferentially to soil and sediment. Collectively, the wide molecular weight range of the hydrocarbons in crude oil will mean that at equilibrium, distribution will be mainly to air and soil, with much less than 1.0% being present in water (European Chemicals Bureau, 2000).

Photodegradation:
The direct aqueous photolysis of an organic molecule occurs when it absorbs sufficient light energy to result in a structural transformation. Only light energy at wavelengths between 290 and 750 nm can result in photochemical transformations in the environment, although absorption is not always sufficient for a chemical to undergo photochemical degradation. In general, many component molecules in crude oils will undergo direct photolysis. Saturated hydrocarbons (paraffins and naphthenics), olefins with one double bond, and single ring aromatics, which constitute the majority of these components, do not absorb appreciable light energy above 290 nm. However, to a limited extent, some degradation of PAH molecules in crude oil in the environment may occur as the result of photo-oxidative processes, although PAHs bound to sediments are reported to be less susceptible to photo-oxidation. The persistence of PAHs in sediments may be due to lack of light for photo-oxidation. Therefore, this fate process will not contribute to a measurable degradative removal of chemical components in this category from the environment.

Atmospheric oxidation as a result of hydroxyl radical attack is not direct photochemical degradation, but rather indirect degradation. AOPs can be calculated using a computer model. Indirect photolysis can be estimated using models accepted by the US EPA and other authorities. An estimation method accepted by the US EPA includes the calculation of atmospheric oxidation potential (AOP). In air, the volatile hydrocarbon molecules in crude oils will undergo reaction with photosensitized oxygen in the form of hydroxyl radicals (OH-) (Atkinson, 1990). The computer program AOPWIN (atmospheric oxidation program for Microsoft Windows 1), used by the US EPA OPPTS (Office of Pollution

Prevention and Toxic Substances), calculates a chemical half-life based on an overall OH- reaction rate constant, a 12-hr day, and a given OH- concentration.

Atmospheric oxidation half-life values for typical hydrocarbon constituents of crude oils (European Chemicals Bureau, 2000):

Constituent	Half-life (days)
benzene	6.5
n-butane	3.2
n-hexane	1.4
toluene	1.3
cyclohexane	1.1
n-decane	0.69
n-tetradecane	0.42
naphthalene	0.37

Summary: Insufficient data are available to characterize the atmospheric oxidation potential of chemical components found in materials in this category. Therefore, representative components for this category are identified and their AOP values calculated. AOPWIN version 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_3, based on a 12-hour day at 25^0C. Hydrocarbons of carbon number greater than C20 will have little or no tendency to partition to air. **No additional modeling is necessary.**

Stability in Water:
Summary: 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 alkyl halides, amides, carbamates, carboxylic acid esters and lactones, epoxides, phosphate esters, and sulfonic acid esters (Harris, 1982). The majority of chemical components in crude oils are hydrocarbons, which are not included in these chemical groups, and they are not subject to hydrolysis reactions with water. **No additional testing or modeling is necessary.**

Chemical Transport and Distribution in the Environment (Fugacity Modeling):
A widely used fugacity model is the EQC (Equilibrium Criterion) model. Level 1 is a steady state, equilibrium model that utilizes the input of basic chemical properties including molecular weight, vapor pressure, and water solubility to calculate distribution within a standardized regional environment (Mackay et al., 1992). Transport media are air, soil, water, biota, suspended sediment and sediment. Values representing the calculated range of distribution to environmental media of representative hydrocarbon components found in crude oils will be presented in a robust summary. Results for a range of percent distribution of typical hydrocarbons found in crude oils are shown in tabular form as follows, but a more exhaustive analysis of crude oil hydrocarbon partitioning is presented in the robust summary section.

	%Air	%Water	%Soil	%Sediment	%Susp. matter	%Biota
Propane	100.0	0	0	0	0	0
n-eicosane	0	0	97.7	2.2	0.1	0
benzene	98.8	1.1	0.1	0	0	0

Summary: Fugacity modeling for those constituents in crude oils indicates that, at steady state, the lower molecular weight components will mainly partition to air, with a maximum of about 1% of mono-aromatic hydrocarbons partitioning to water. As the molecular weights increase, there is less tendency for the hydrocarbons to partition to air, the environmental distribution being preferentially to soil and sediment. Collectively, the wide molecular weight range of the hydrocarbons in crude oil will mean that at equilibrium, distribution will be mainly to air and soil, with much less than 1.0% being present in water. These data are adequate to define environmental distribution of crude oil components. **No additional modeling is necessary for this endpoint.**

Biodegradation: Crude oil contains hydrocarbons of well-defined generic types that are biodegraded at different rates. n-Alkanes are readily degraded in seawater, since many Microorganisms can utilize them. Branched-chain or iso-alkanes are less readily biodegraded, but they do ultimately biodegrade. The degradation of cycloalkanes has not been extensively studied, but the ring structure is more resistant to biodegradation, and degrades more slowly. Aromatic hydrocarbons are also resistant to biodegradation, but a few microorganisms are able to utilize them. High molecular weight compounds, the tars and asphaltenes, show little to no degradation, and are persistent.

Few data are available on the biodegradation of samples of crude oils obtained under laboratory conditions. However, extensive research on oil degradation in marine environments indicates that virtually all kinds of oil are susceptible to microbial oxidation. The rate of oxidation is influenced by microbial characteristics, and environmental factors such as available nutrients, oxygen, temperature and degree of dispersion. Crude oil has been found to be equally well degraded in sea water and in fresh water, with the availability of nitrogen and phosphorous containing compounds especially important in determining rate of degradability. Adapted microorganisms are often found in ocean areas where crude oil spills are common. Zobell (1969) has calculated that where an adapted microbial population is available in well-aerated seawater at 20 to 30°C, the rate of crude oil oxidation ranges from 0.02 to 0.2 g of oil oxidized/m^2/day. The same author found experimentally that complete oxidation of 1.0 mg of hydrocarbon requires between 3 and 4 g of oxygen, i.e. it has a BOD of 3 to 4 mg oxygen/mg. Since the oxygen content of seawater is between 6 and 11 mg/liter, depending on salinity and temperature, this means that about 320,000 liters of seawater is required to oxidize one liter of crude oil. Five day respirometric tests run both in fresh water and in salt water at 30°C using a Kuwait crude oil resulted in 15% and 3% biodegradation, respectively (Bridie, A.L. and Bos, J., 1971). Biodegradation rates for crude oils will vary considerably, but in standard 28-day studies, none would be expected to be readily biodegradable. However, the evidence from

spillages and from natural seepages is that most of the non-volatile constituents of crude oil are inherently biodegradable, but that some of the highest molecular weight components are persistent in water (CONCAWE, 2001).

Summary: A technical discussion of hydrocarbon degradation of crude oils and representative component molecules based on reviews of available literature will be incorporated in a robust summary format. **No additional testing is proposed.**

EVALUATION OF EXISTING ECOTOXICITY DATA AND PROPOSED TESTING

The HPV Chemical Test Program includes acute toxicity endpoints for a freshwater fish and invertebrate, and toxicity to a freshwater alga. The materials in the Crude Oil Category produce a similar range of toxicity for these three endpoints based on results of comparable studies using standard test methods and exposure solution preparation procedures. This is because crude oils are complex combinations of relatively similar series of homologous chemicals. Therefore, their short-termed toxicities are expected to fall within the range of toxicity demonstrated by the materials summarized in this test plan. Experimental studies of acute aquatic toxicity show values for crude oil samples in the range 10 to over 100 mg/l, which were evaluated as dispersions of the whole oil in water or as water accommodated fractions (WAFs) (CONCAWE 2001). These values are in accordance with the predicted aquatic toxicity of crude oils based on their hydrocarbon composition. This is expected because the majority of constituents in crude oil are neutral organic hydrocarbons, whose mode of action is non-polar narcosis, which is brought about by disruption of biological membrane function. Acute toxicity is attributed to those water-soluble hydrocarbon components that are saturates (paraffins or cyclics) and mono- and di-aromatics (Peterson, 1994; van Wezel and Opperhuizen, 1995; CONCAWE, 1996; Di Toro, et al., 2000). The differences between toxicities (i.e., LC/LL50, EC/EL50) can be explained by the differences between the target tissue-partitioning behaviors of the individual chemicals (Verbruggen et al., 2000).

Aquatic Toxicity

For the assessment of eco-toxicity of poorly water soluble mixtures of hydrocarbons as found in petroleum products, it is now generally accepted that results should be expressed in terms of the "loading rate" (OECD 2000). The "loading rate" may be defined as the amount of the product that is equilibrated with the aqueous test medium, and the aqueous phase at equilibrium is termed the water accommodated fraction (WAF) for the loading rate (OECD 2000). Toxicological endpoints such as the LL50 or EL50 are used to express the loading rate of the product that is lethal or produces a specific effect to 50% of the test organisms. Studies in which the results are expressed in terms of the measured concentrations of hydrocarbons in dilutions of "water soluble fractions (WSF)" do not allow the eco-toxicity of a product to be expressed in terms of the amount of that product required to produce a particular effect and, therefore, such results are not comparable to results obtained with other substances.

In spite of the variety of fish and invertebrate species that have been tested under a range of different laboratory methodologies, some consensus on sensitivity of aquatic species and different life stages can been made. In general, the larvae of fish and crustaceans appear to be most susceptible to the water-soluble fraction of crude oil. Exposures of plankton and algae have indicated that certain species of diatoms and green algae are inhibited, whereas micro-flagellates are less sensitive. For the most part, mollusks and most inter-tidal worm species appear to be tolerant of oil contamination (McAuliffe, 1966).

For the Crude Oil HPV Category, robust summaries were prepared for six acute eco-toxicity studies with a fish, an invertebrate, and an alga on samples of two crude oils. All exposure solutions were prepared as water accommodated fractions. Fish LL50 values were 21 and 41 mg/L, invertebrate EL50 values were 2.7 and 4.1, and alga LL50 values based upon germination of spores were 122 and 528 mg/L, while alga EL50 values based upon growth were 122 and 311 mg/L (EMBSI 2001a-e, 2002).

These summarized studies are supported by published data on aquatic toxicity of crude oils that show a wide variety in the response of organisms to oil exposures. Some of this variability may be due to using different methodologies such as WAFs and WSFs, as noted above, but overall, referenced data generally follows the information provided in the robust summaries. The following information summarizes published reliable representative aquatic toxicity data of crude oils.

LC50 values for rainbow trout of 350 and 310 mg/L were reported for nominal concentrations of Forties crude oil prepared as dispersions (Westlake, 1991). Although the test dispersions were prepared in closed jars, tests were run in open vessels, and significant evaporative losses of lower molecular weight hydrocarbons may have occurred. Therefore these data may reflect an underestimation of the acute toxicity. Similar open-system tests exposing fish species to either dispersions of crude oil and water, or to water soluble fractions of crude oil, indicate LC50 values as a function of loading rate ranged from 3.7 g/L to greater than 80 g/L (Anderson, et al., 1974). Since significant evaporative losses of lower molecular weight hydrocarbons may have occurred, the measured LC50 values potentially underestimate the true eco-toxicity. Forty-eight hour LC50 values for rainbow trout exposed to water-soluble fractions (WSF) of Norman Wells crude oil were 10.4 mg/l (open vessel) and 11.6 mg/l (closed vessel), based on measurements of dissolved hydrocarbons made at the beginning and end of the 48-hour period (Lockhart, et al., 1987).

The acute toxicity of nineteen crude oils to brown shrimp was assessed using whole-oil dispersions prepared by continuous stirring and gave LC50 values in the range 32 to 140 μl/L, equivalent to approximately 27 to 120 mg/L (Franklin and Lloyd, 1982). In tests with *Daphnia magna*, dispersions of Forties crude oil in water gave EC50 values of 43 and 51 mg/L (Westlake, 1991). Although the test dispersions were prepared in closed jars, tests were run in open vessels, significant evaporative losses of lower molecular weight hydrocarbons may have occurred and hence the measured EC50 values underestimate the true eco-toxicity. Lethal loading rate concentrations LL50 (96h) of 618 mg/l and 39

mg/L were determined for the test species, *Mysodopsis bahia,* and the kelp forest mysid, *Holmesimysis costata*, when exposed to water accommodated fractions of Kuwait crude oil (Bragin, et al., 1994).

Algal tests with *Anabaena doliolum* were run using WSF of Assam crude oil and using whole crude oil in equilibrium with the algal suspension medium. The WSF method gave a mean 15-day EC50 of 9.06 mg/l, and the direct loading (or whole oil) method gave a 15-day EC50 of 5.73 mg/l, both based on measured dissolved hydrocarbon concentrations in the aqueous phase (Gaur and Singh, 1989).

Significant effects were found at both 40% WSF (17ppm oil) and 50% WSF (21ppm oil) of Ekofisk crude oil on the development of the eggs of two sea urchin species, Strongylocentrotus pallidus and S. droebachiensis. At these levels, the larvae filled with degenerating cells, and differentiation of the intestine and skeletal growth were inhibited compared with the control larvae (Falk-Petersen, 1979).

Other risks to aquatic species, semi-aquatic birds, and sea mammals include physical fouling of plumage, fur, gills etc, by floating oil product. This results in loss of buoyancy, insulation and smothering of inter-tidal animals. Ingestion of oil resulting from attempts by animals to clean contaminated body parts may result in severe enteritis and toxicity.

Spills in freshwater environments have been shown to adversely affect the diversity and abundance of the aquatic macro-invertebrate community, with the observed effects associated with oil sorption and substrate coating (Poulton, et al., 1997; Poulton, et al., 1998). Recovery of such communities in some habitats may be rapid (e.g., riffle areas of streams/rivers), while impacts to backwater areas may persist for months. Ultimately, the type of crude oil and the local conditions and habitats will dictate the potential and extent to which crude oil persists and cause effects in the environment.

Summary: A range of measurements of aquatic toxicity has been obtained in laboratory studies of crude oils. Variability in results may be related in part to the source of the crude oil, or it may reflect different approaches to testing. However, those studies using dispersions of whole oil, employing water soluble fractions, and water accommodated fractions have generally given LC50 or EC50 values in the range 10-100 mg/L or greater when expressed in terms of oil loading rate. **No additional testing is proposed.**

TABLE 4. MATRIX OF AVAILABLE ADEQUATE DATA AND PROPOSED TESTING FOR THE PRIMARY TEST MATERIALS

	Petroleum Crude Oil CAS #8002-05-9
Melting Point	Adequate
Boiling Point	Adequate
Vapor Pressure	Adequate
Partition Coefficient	Adequate
Water Solubility	Adequate
Photodegradation	Adequate
Stability in Water	Adequate
Transport and Distribution	Adequate
Biodegradation	Adequate
Acute Toxicity to Fish	Adequate
Acute Toxicity to Aquatic Invertebrates	Adequate
Toxicity to Algae	Adequate
Acute Toxicity	Adequate
Repeated Dose	Adequate
Genotoxicity, in vitro	Adequate
Genotoxicity, in vivo	Adequate
Repro/Developmental	**Test [2 samples]**

Adequate Indicates adequate existing data.
Test Indicates proposed testing

TEST PROPOSAL

This study plan proposes a Combined Repeat Dose Toxicity Study with Reproductive/Developmental toxicity Screening Test (OECD protocol 422) for a selected light crude (high paraffinic, low aromatic, $>33^0$API) and heavy crude (low paraffinic, high aromatic, $<28^0$API) oil sample by the dermal route. Dermal and oral exposures have been shown to induce similar systemic effects (e.g. hematologic changes, liver enlargement and thymic atrophy) with light and heavy crude oils (Leighton, 1990; Khan et al., 1987; Feuston et al., 1997a,b). Dermal exposure has been selected as the most likely route of human exposure. Analytical characterization will be performed on the light and heavy crude oils selected for mammalian testing. Environmental effects of petroleum crude are well defined from laboratory studies and monitoring at spill sites and no further testing is proposed.

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1984a Acute oral: Arab Light Crude Study #40961; Beryl Crude Oil Study #40951; Mid-continent Crude Oil Study #40971.

1984a Acute dermal: Arab Light Crude Study #40962; Beryl Crude Oil Study #40952, Mid-continent Crude oil Study #40972.

1985 Eye irritation: Arab Light Crude Study #40963; Beryl Crude Oil Study #40953, Mid-continent Crude Oil Study #40973.

1985 Skin irritation: Arab Light Crude Study #40964; Beryl Crude Oil Study #40954, Mid-continent Crude Oil Study #40974.

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1991a Delayed contact hypersensitivity study in guinea pigs [Buehler sensitization test] of Lost Hills Light Crude Oil. Study #63841.

1991b Delayed contact hypersensitivity study in guinea pigs [Buehler sensitization test] of Belridge Heavy Crude Oil. Study #63853.

Repeat Dose toxicity

1992a 13-week dermal administration of Lost Hills Light to rats. Study #63834.

1992b 13-week dermal administration of Belridge Heavy to rats. Study #63846.

In vitro Genetic Toxicity

1984b. A modified Ames pre-incubation assay for the determination of specific mutagenicity of the DMSO extract of Arab Light Crude Oil Study #40965; Mid-continent Crude Oil Study #40975; Beryl Crude Oil Study #40955.

1990b. A modified Ames pre-incubation assay for the determination of specific mutagenicity of the DMSO extract of Belridge Heavy Crude Oil # Study #63850; Lost Hills Light Crude Oil Study #63838.

1991c. Metaphase analysis of Chinese Hamster Ovary (CHO) cells treated *in vitro* with a DMSO extract of Lost Hills Light Crude Oil Study #63840.

1991d. Metaphase analysis of Chinese Hamster Ovary (CHO) cells treated *in vitro* with a DMSO extract of Belridge Heavy Crude Oil Study #63852.

In vivo Genetic Toxicity

1991e. Micronucleus assay of bone marrow cells from rats treated via dermal administration of Belridge Heavy Crude Oil Study # 63847.

1990c. Micronucleus assay of bone marrow cells from rats treated via dermal administration of Lost Hills Light Crude Oil Study # 63835.

Developmental Toxicity

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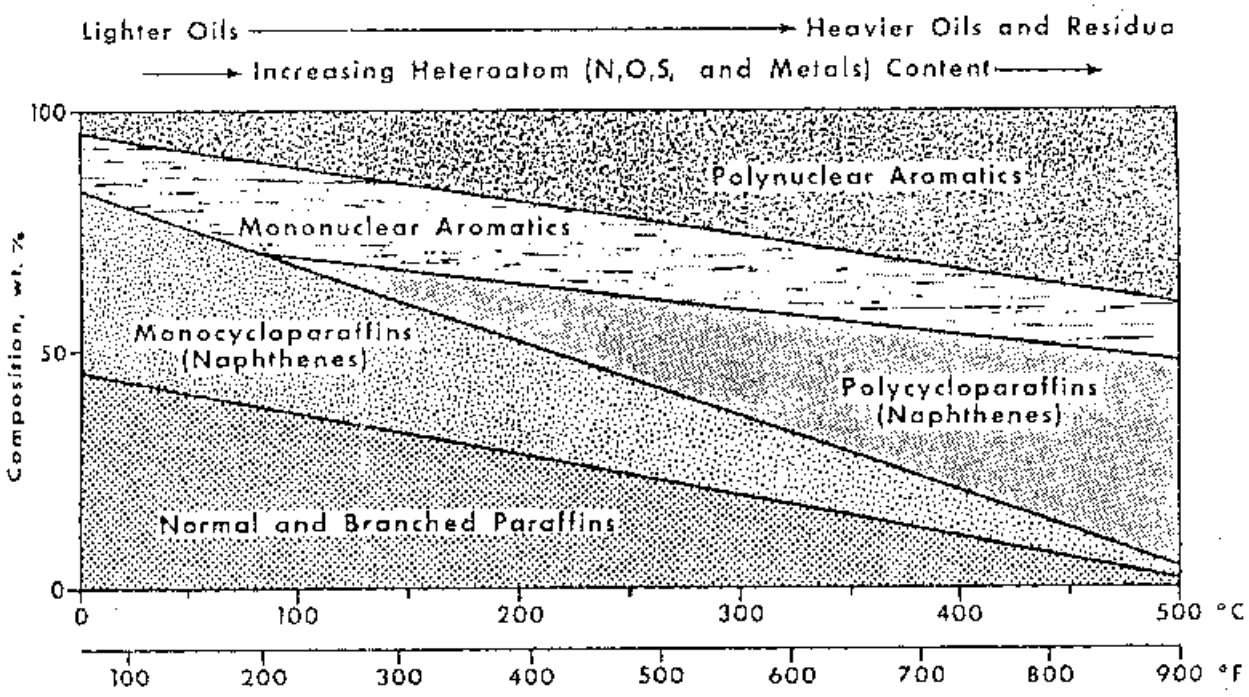
APPENDIX 1; Petroleum Chemistry and Crude Composition

The hydrocarbons that comprise crude oil – paraffins, naphthenes (cycloparaffins) and aromatics – share some structural features but differ in the ratio of hydrogen to carbon atoms and how those atoms are arranged. Olefins are not present in crude oils and are formed from rearrangement of atoms during the cracking process to produce gasoline-blending streams. Paraffins occur in higher concentrations in lower boiling fractions of crude oil while the concentration of naphthenes (cycloparaffins) and aromatics increase at higher boiling ranges (Fig A1-1)



Feedstock composition represented by the distribution of chemical types.

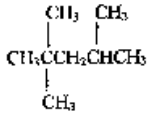
Mobil, 1997

Hydrocarbon molecules in crude oil may include from 1 to more than 50 carbon atoms at room temperature. When isolated, hydrocarbons containing 1-4 carbon atoms are gases, those with 5-19 carbon atoms are usually liquid, and those with 40 or more carbon atoms are solids.

Paraffins: C_nH_{2n+2} where n= number of carbon atoms.

Carbons are joined by single bonds (e.g. butane, $CH_3CH_2CH_2CH_3$). Paraffins with 4 or more C atoms may have 2 or more structural arrangements or structural isomers, for example:

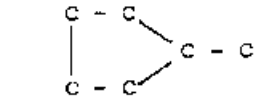
normal octane, $CH_3CH_2CH_2CH_2CH_2CH_2CH_2CH_3$ or isooctane



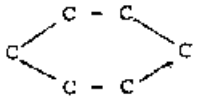
Normal paraffins occur in most crude oils but in varying total concentrations (King, 1983). As a rule, crude oils of older geological age contain higher quantities of n-paraffins. Occurrences relative to other hydrocarbon classes decrease as the boiling point of fractions distilled from crude oil increases. C_{12}-C_{16} n-paraffins have been identified as accelerators of skin carcinogenesis. Branched (iso) paraffins are found throughout the boiling range but do diminish with increasing boiling point. Certain lower molecular weight branched paraffins are capable of producing kidney damage in male rats (light hydrocarbon nephropathy).

Naphthenes: Cycloparaffins in gasoline have 5 or 6 carbon atoms arranged in a ring and belong to either a cyclopentane or cyclohexane series, for example:

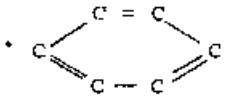
C_6H_{12} Methylcyclopentane

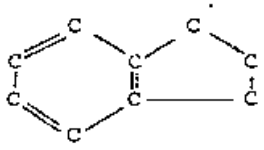


C_6H_{12} Cyclohexane



Cycloparaffins constitute a substantial proportion of petroleum with 5- to 6-membered ring structure being the predominant type. Most individual cycloparaffins that have been isolated are in the boiling range of gasoline and kerosene. The cycloparaffin portion of lubricant fractions is a complex mixture of non-condensed and condensed 5- and 6-member rings. Polycycloparaffins may act as inhibitors in skin carcinogenesis.

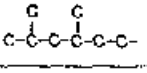
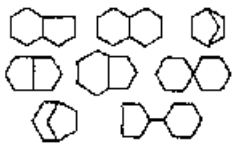
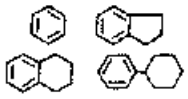
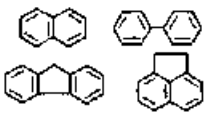
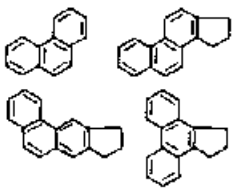
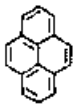
Aromatics: Some carbon atoms are arranged in a ring joined by aromatic bonds. for example: benzene, C_6H_6 .  In polycyclic aromatics, some carbons are shared by 2 or more rings, for example, indane, C_9H_{10}



Aromatic hydrocarbon types appear to be present in the same relative proportions in different crude oils. Where several possibilities for alkyl substitution exist, the predominant isomers are generally those containing substituents with the smallest number of carbon atoms. In heavier, lubricant type fractions, mixed aromatic-cycloparaffin hydrocarbons

predominate, as mono-, di-, or trinuclear aromatic-cycloparaffin hydrocarbons. Certain polynuclear aromatics are associated with systemic toxicity and skin cancer.
Resins and asphaltenes are high molecular wt fractions (500-10,000)_containing N, S, and oxygen found in the residuum/bottoms of crude oils. They have high polarity, low solubility and limited bioavailability and toxicity. They may constitute from 10% in light paraffinic oil to up to 60% in heavy crude oils.
Much of the compositional information described above was derived from the extensive analysis of a Ponca Oklahoma crude, performed under the sponsorship of the American Petroleum Institute. Table A1-1 summarizes these findings (King, 1983).

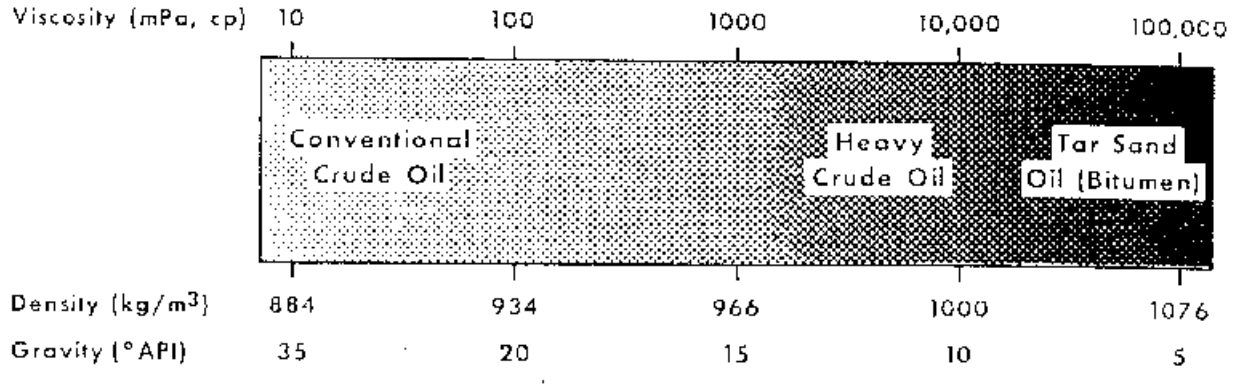
TABLE A1-1: Types of Hydrocarbons Isolated from Ponca Crude

HYDROCARBON TYPE	BASIC STRUCTURES	NUMBER ISOLATED	FRACTIONS WHERE FOUND
Paraffins			
n-Paraffins	C-C-C-C-C-C-	33	All Distillates
Branched Paraffins		41	Gasoline, Light Gas Oil
Cycloparaffins			
Monocycloparaffins		39	Gasoline, Kerosine
Bicycloparaffins		29	Gasoline, Kerosine
Tricycloparaffins		3	Gasoline, Kerosine
Aromatics			
Mononuclear Aromatics		48	Gasoline, Kerosine, Light Gas Oil
Dinuclear Aromatics		60	Kerosine, Light Gas Oil
Trinuclear Aromatics		10	Heavy Gas Oil-Light Lubricant
Tetranuclear Aromatics		3	Heavy Gas Oil-Light Lubricant

APPENDIX 2: Crude Product Potential

Crude oils are classified by viscosity, density and API gravity. API gravity was developed as a means to identify the gasoline production potential of a crude oil; the higher the API gravity, the more valuable the crude. Fig. A2-1 illustrated classification of crude oil by this density-gravity method.

Fig. A2-1



Type of Crude	Characteristics
1. Conventional or "light" crude	Density-gravity range less that 934kg/m^3 (>33^{0}API)
2. "Heavy" crude oil	Density-gravity range from 1000kg/m^3 to more than 934kg/m^3 (10^{0}API to <28^{0}API) Maximum viscosity of 10,000mPa.s(cp)
3. "Extra-heavy" crude oil; may also include atmospheric residua. (b.p.>340^{0}C; >650^{0}F)	Density-gravity greater than 1000kg/m^3 (<10^{0}API) Maximum viscosity of 10,000mPa.s(cp)
4. Tar sand bitumen [before upgrade]or natural asphalt; may also include vacuum residua. (b.p.>510^{0}C; >950^{0}F)	Density-gravity greater than 1000kg/m^3 (<10^{0}API) Viscosity greater than 10,000mPa.s(cp)

Mackerer and Biggs, AIHCE, 1996; Platts, 2003

Heavier crude oils have higher density-gravity values and higher viscosity, with lower API gravity, making them less suitable for gasoline stocks but better candidates for lubricant and heavy fuel production. Fig. A2-2 shows yield comparisons for 4 typical crude oils.

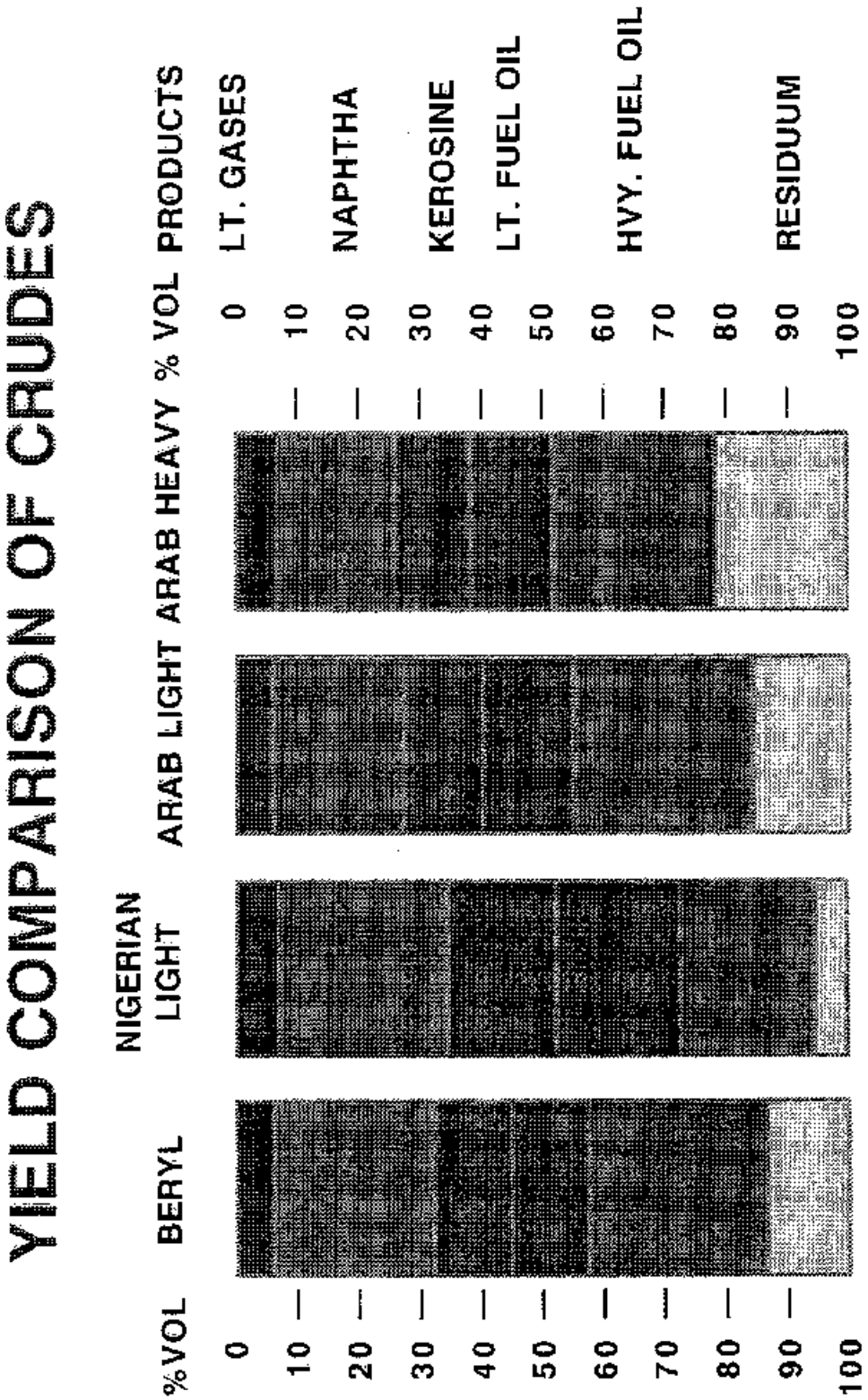

YIELD COMPARISON OF CRUDES
%VOL
BERYL
NIGERIAN LIGHT
ARAB LIGHT
ARAB HEAVY
% VOL
PRODUCTS
LT. GASES
NAPHTHA
KEROSINE
LT. FUEL OIL
HVY. FUEL OIL
RESIDUUM
0
10
20
30
40
50
60
70
80
90
100

Table 2.1 BRIEF SUMMARY OF THE HISTORY OF REFINING PROCESSING

YEAR	PROCESS NAME	PROCESS PURPOSE	BYPRODUCTS, ETC.
1862	Atmospheric Distillation	Produce Kerosine	Naphtha, Tar, etc.
1870	Vacuum Distillation	Lubricants (original) Cracking feedstocks (1930's)	Asphalt, Residual Coker Feedstocks
1913	Thermal Cracking	Increase Gasoline	Residual, Bunker Fuel
1916	Sweetening	Reduce Sulfur	
1930	Thermal Reformation	Improve Octane Number	Residual
1932	Hydrogenation	Remove Sulfur	Sulfur
1932	Coking	Produce Light Products	Coke
1933	Solvent Extraction	Improve Lubricant Viscosity Index	Aromatics
1935	Solvent Dewaxing	Improve Pour Point	Waxes
1935	Cat. Polymerization	Improve Gasoline Yield & Octane No.	Petrochem Feedstocks
1937	Catalytic Cracking	Higher Octane Gasoline	Petrochem Feedstocks
1939	Visbreaking	Reduce Viscosity	Increased Distillate, Tar
1940	Alkylation	Increase Gasoline Octane & Yield	High Octane Aviation Gasoline
1940	Isomerization	Produce Alkylation Feedstock	Naphtha
1950	Deasphalting	Increase Cracking Feedstock	Asphalt
1952	Catalytic Reforming	Convert Low Quality Naphtha	Aromatics
1954	Hydrodesulfurization	Remove Sulfur	Sulfur
1956	Inhibitor Sweetening	Remove Mercaptans	
1957	Cat. Isomerization	Convert to Molecules w/High Oct. No.	Alkylation Feedstocks
1960	Hydrocracking	Improve Quality & Reduce Sulfur	Alkylation Feedstocks
1961	Fluid Cat. Cracking	Increase Gasoline Yield	Petrochem Feedstocks
1974	Catalytic Dewaxing	Improve Pour Point	
1975	Resid. Hydrocracking	Increase Gasoline Yield from Residual	

Table A2-1 summarizes the history of petroleum refining. Since the first refinery was established in 1862, processes have been developed and continually improved to maximize the yield and efficiency of production of high quality fuels, lubricants and petrochemicals from petroleum crude, and concomitantly to minimize or eliminate hazardous components, waste products and environmental contaminants.

ROBUST SUMMARY
OF INFORMATION ON

Substance: **CRUDE OIL**

CAS No. **8002-05-9**

Summary prepared by: American Petroleum Institute

Creation date: **FEBRUARY 22, 2002**

Printing date: **NOVEMBER 20, 2003**

Date of last Update: **NOVEMBER 15, 2003**

Number of Pages: **77**

NB. Reliability of data included in this summary has been assessed using the approach described by Klimisch, et al.

Klimisch, H. J., Andreae, M. and Tillman, U, (1997)
A systematic approach for evaluating the quality of experimental toxicological and ecotoxicological data.
Regulatory Toxicology and Pharmacology 25, 1-5.

1.1.1 GENERAL SUBSTANCE INFORMATION

Substance type : Petroleum product
Physical status : Liquid

Remark : The CAS definition for Petroleum (Crude oil) is:

> "A complex combination of hydrocarbons. It consists predominantly of aliphatic, alicyclic and aromatic hydrocarbons. It may also contain small amounts of nitrogen, oxygen and sulfur compounds. This category encompasses light, medium, and heavy petroleums, as well as the oils extracted from tar sands".
>
> Hydrocarbonaceous materials requiring major chemical changes for their recovery or conversion to petroleum refinery feedstocks such as crude shale oils, upgrade shale oils and liquid coal fuels are not included in this definition.

Crude oil contains hydrocarbons in the carbon number range from C1 to C60+. It also contains organometallic complexes, notably of sulfur and vanadium, and dissolved gases such as hydrogen sulfide. Crude oils range from thin, light colored oils consisting mainly of gasoline-quality stock to heavy, thick tar-like materials.
An "average crude oil has the following general composition:

Carbon	84%
Hydrogen	14%
Sulfur	1-3%
Nitrogen	1%
Oxygen	1%
Minerals and salts	0.1%

The chemical composition of crude oils can vary tremendously from different producing regions and even from within a particular formation.

Examples of compositions of various whole crudes are shown in the following table

Crude source	**Paraffins % vol.**	**Naphthenes % vol**	**Aromatics % vol**	**Sulfur % wt**	**API gravity (°API)**
Light crudes					
Saudi light	63	18	19	2.0	34
South Louisiana	79	45	19	0	35
Nigerian light	37	54	9	0.1	36
North sea Brent	50	34	16	0.4	37
Beryl					

47	34	19	0.4	37
Lost Hills				
Non-Aromatics 50%		50	0.9	
Heavy crudes				
Prudhoe Bay				
27	36	28	0.9	28
Saudi Heavy				
60	20	15	2.1	28
Venezuela Heavy				
35	53	12	2.3	24
Belridge Heavy				
Non-aromatics 37%		63	1.1	-
Mid-range crudes				
Kuwait				
63	20	24	2.4	31
Venezuela Light				
52	34	14	1.5	30
USA West Texas Sour				
46	32	22	1.9	32

Crude oils may be categorized in either of several different ways e.g.

Paraffinic vs naphthenic.
Crude oils contain both paraffinic and naphthenic hydrocarbons but if there is a preponderance of paraffinic hydrocarbons present, the crude oil is referred to as a paraffinic crude. These crudes would be rich in straight and branched chain paraffins. Conversely a crude in which naphthenic hydrocarbons are predominant is referred to as a naphthenic crude. These crudes contain mainly naphthenic and aromatic hydrocarbons

Sweet vs sour
Crude oils may be referred to as either sweet or sour depending on the level of hydrogen sulfide present. A sweet crude has very little H_2S whereas a sour crude has larger quantities of H_2S present.

Light vs heavy
Crude oils may be divided into Light and Heavy crudes on the basis of their gravity.
The API gravity is determined as:

$$°API = \frac{141.5}{\text{Specific gravity}} - 131.5$$

Crude oils with gravity > 33°API are considered as light crudes. Such crudes with a high percentage composition of hydrogen are usually more suitable for processing for gasoline production. Heavy crudes, ie those with gravity < 28°API tend to contain more asphaltenes and are usually rich in aromatics. These heavy crudes require more steps in their processing.

Information in this robust summary is presented for light and heavy crudes since this categorization distinguishes between crudes with a high paraffinic content (Light crudes) and those with a high aromatic/naphthenic content (Heavy crudes). This represents the extremes of the ranges of crudes available.

1.13 REVIEWS

Memo : IARC Review

Remark : IARC reviewed the evidence for carcinogenicity of crude oil to man and animals and published the result in 1989.
IARC concluded that:

There is inadequate evidence for the carcinogenicity in humans of crude oil

There is limited evidence for the carcinogenicity in experimental animals of crude oil

The overall evaluation was

Crude oil is not classifiable as to its carcinogenicity to humans (Group 3)

(38)

2.1 MELTING POINT

Value	:	-30 - 30 °C
Sublimation	:	no
Method	:	ASTM D97
GLP	:	no data
Remark	:	The figures quoted are a typical range for the drop point as measured by a standard oil industry procedure. For some low wax crudes, pour points below -30 °C are obtained.
Reliability	:	(1) valid without restriction

(1) (16)

2.2 BOILING POINT

Value	:	-1 - 565 °C at 1013 hPa
Decomposition	:	yes
Method	:	ASTM D86
GLP	:	no data
Remark	:	The method measures the distillation range of petroleum products. The figures quoted represent the approximate range for crude oils and are based on (a) the boiling point of n-butane for the lower value and (b) an upper limit quoted by Speight. In practice, atmospheric distillation of crude oil is not practiced above 275-300 °C, to avoid thermal decomposition of the residue. The residue is normally vacuum distilled in a subsequent operation.
Reliability	:	(1) valid without restriction

(1) (16) (77)

2.4 VAPOUR PRESSURE

GLP	:	no data
Test substance	:	Crude oil
Remark	:	Vapor pressure measurements were provided for 10 petroleum crude oils originating from various locations throughout the world. The source of these data are from Environment Canada Environmental Technology Center, a government-maintained database (Jokuty et al., 2002). Data cited by OGJ may be considered a secondary source, but the data cited by ESD and EETD were measured data sponsored by those agencies using a standardized method for measuring vapor pressure of petroleum products[1]. As such, these 10 measurements provide a body of data adequate for use in the U.S. EPA HPV program and specific for the physical/chemical endpoint of vapor pressure. (1) ASTM D323, Standard Test Method for vapor Pressure of Petroleum Products (Reid Method)
Result	:	Vapor Pressure, kPa See the following table and Remarks section Temperature °C Not stated Decomposition Not stated

Vapor Pressures, kPa:

Product Name	Origin	Reid Vapor Pressure Value, kPa	Reference
Alaska North Slope	Alaska USA	19	ESD 91
Arabian Medium	Saudi Arabia	22.1	OGJ 99
Alif	Yemen	45	OGJ 99
Amna	Libya	27	OGJ 99
Ashtart	Tunisia	13	OGJ 99
Atkinson	Beaufort Sea, Canada	6	ESD 91
Alberta Sweet Mixed Blend	Alberta, Canada	19	EETD 84
Abu Al Bu Khoush	United Arab Emirates	24	OGJ 99
Beryl	North Sea, UK	36	OGJ 99
Bombay High	India	33	OGJ 99

Reliability : (2) valid with restrictions
Data obtained from a government (Environment Canada) database who sponsored the vapor pressure measurements cited by ESD and EETD.
(20) (21) (39) (74)

2.5 PARTITION COEFFICIENT

Log pow : 2 - 6
Method : Calculated

Remark : The calculation was done by the CLOGP Version 3.5 program (calculation of Log partition coefficient octanol/water). The figures represent the spread of calculated and/or measured values for typical hydrocarbon components of crude oil. Calculated values for higher molecular weight hydrocarbons will be above 6, but such values are notional, since no correlation has been established between calculated and experimental values.
Reliability : (1) valid without restriction
(16) (45)

2.6.1 SOLUBILITY IN DIFFERENT MEDIA

Solubility in : Water
Method : Preparation of water soluble fraction
Year : 1990
GLP : no
Test substance : Crude oil

Method : Individual saturated crude oil solutions were prepared by adding approximately 10 ml of the respective oil to 50-100 ml of water in 125 ml separatory funnels. Funnels were gently shaken for at least 48 hrs either with a magnetic stir bar or with a wrist action shaker, then placed in a temperature bath for at least 48 hr prior to analysis. Solubility at 5, 20 and 22 (±2C) °C was determined in both double distilled water and salt water (3% NaCl). The effect of water to oil ratio on the solubility of crude oil components was determined by injecting oil into sealed vials completely filled with water, and mixed at low speed for an equilibration period of 20 days. Analysis was determined by purge-and-trap/GC; solid sorbent

extraction with HPLC and fluorescence analysis.

Result : Fluorescence/HPLC analysis was inadequate in quantification of hydrocarbon composition.
Purge and trap/GC analysis results based on total benzene, toluene, ethyl benzene+xylenes (combined concentration) and naphthalenes (BTE+XN as mg/l) are reported along with viscosity (vis=c.p @ 20C) and density (d=g/cm³ @ 20°C) for each oil at respective temperatures (°C) in distilled and saltwater (22° reported as ± 2°C)

	BTE+XN (mg/l)	
Oil	**Distilled water**	**Saltwater**
Norman Wells 4.41 vis; 0.832 d		
	30 (5) 29-33 (20)	25.5 (5)
	31.8-33.5 (22)	20 (22)
4.7 vis; 0.84 d	33 (20)	14.8 (5)
Alberta: 4.98 vis; 0.835 d	25.02(22)	
Swan Hills: (no vis. density)	35.1 (22)	
Prudhoe Bay: 34.9 vis; 0.901 d	29.01 (22)	
Lago Medio: 41.1 vis; 0.872 d	23.66-25.5 (22)	16.47 (22)
Kopanoar: 17.5 vis; 0.9 d	10.42 (22)	
Murban: 4.37 vis; 0.824 d	28.62 (22)	
Hibernia B 33.2 vis; 0.837d		16.92 (22)
Hibernia J (no vis., density)		7.75 (22)
Mobil A (no vis., density)	29.6 (22)	
Mobil B (no vis., density)	58 (22)	

Results for oil-water ratio testing were not quantified, but general observations were stated as follows: concentrations of the water soluble fractions decreases as the water-to oil ratio increases and the composition of the water soluble fraction changes as the ratio changes. At low water to oil ratios, the WSF is composed predominantly (80%) of BTEX. As the water to oil ratio increases, these compounds account for a smaller proportion of the dissolved compounds. At a water/oil ratio of 10000, these compounds account for only 15-30% of the total WSF.

Conclusion : Limited detail is provided for exact amounts of crude oil used for preparing aqueous solutions, nor is there any information regarding the composition of each of the crude oils tested, either as hydrocarbon type or inorganic components (such as sulfur). Also, no information on the GC calibration standard composition used to identify and quantify soluble components in the equilibrated aqueous -oil solutions is provided. Individual components of complex petroleum substances have specific and differing solubilities. 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.

Reliability : (2) valid with restrictions

(14) (15) (18) (19) (47) (75)

2.14 ADDITIONAL REMARKS

Memo : CONCAWE

Remark : Petroleum is a natural organic material consisting mainly of hydrocarbons. It occurs in both the gaseous and liquid states in geological traps. The liquid phase, after being freed from dissolved gas and any associated salt water, is known as crude oil. All the information presented in this Data Set relates to crude oil.
Crude oil is not a uniform substance since its physical and chemical properties vary from oilfield to oilfield and can even vary within wells at the same oilfield. At one extreme, crude oil is a light, mobile, straw-colored liquid containing a large proportion of hydrocarbons which are readily distilled at atmospheric pressure. At the other extreme, crude oil is a highly viscous, semi-solid, black substance from which little can be distilled at atmospheric pressure before thermal decomposition occurs.
The most consistent property of crude oils is their relatively small range of elemental composition, as the ranges in the following table show:

element	composition (wt%)
carbon	83.9 - 86.8
hydrogen	11.0 - 14.0
sulfur	0.06 - 8.0
nitrogen	0.02 - 1.7
oxygen	0.08 - 1.82
metals	0.00 - 0.14

Crude oils are normally characterized in terms of three properties; density, viscosity and sulfur content. Crude oils are identified as either light (specific gravity <0.82), or medium (specific gravity 0.82 to 0.97), or heavy (specific gravity > 0.97). The viscosity is an expression of the mobility of the crude oil. The sulfur content has a marked influence on the refinery procedures to which the crude oil, and in particular its derivatives, will be subjected in order to produce acceptable products.
Crude oils are also characterized in terms of their chemical composition, specifically on the predominance of the hydrocarbon types that are present. Modern practice tends to recognize two main types of crude, namely paraffinic and naphthenic. Paraffinic crude oils are rich in straight-chain and branched-chain alkanes, whereas in naphthenic crudes the main constituents are cycloparaffins and aromatic hydrocarbons. However, this is a simplified picture, as many crude oils fall between or outside these two types.

Source : The technical information has been compiled by the Oil Companies' European Organization for Environmental Health Protection (CONCAWE), based at Madouplein-1, B-1210 Brussel, Belgium, and this organization holds copies of the reference articles cited in this data set

Reliability : (2) valid with restrictions

(5)

3.1.1 PHOTODEGRADATION

Type : air
Light source : Sun light
INDIRECT PHOTOLYSIS
Sensitizer : OH
Conc. of sensitizer : 1000000 molecule/cm³
Method : Calculated according to Atkinson, 1990

Remark : Atkinson gives data which enables half lives to be calculated for the degradation of hydrocarbons in contact with hydroxyl radicals in the troposphere, under the influence of sunlight. Values for typical hydrocarbon constituents of crude oils are as follows:

Constituent	Half-life (days)
benzene	6.5
n-butane	3.2
n-hexane	1.4
toluene	1.3
cyclohexane	1.1
n-decane	0.69
n-tetradecane	0.42
naphthalene	0.37

Hydrocarbons of carbon number greater than C20 will have little or no tendency to partition to air (see Sub-chapter3.2.2).

Reliability : (2) valid with restrictions

(3) (16)

3.1.2 STABILITY IN WATER

GLP : no

Remark : 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 alkyl halides, amides, carbamates, carboxylic acid esters and lactones, epoxides, phosphate esters and sulfonic acid esters. The chemical components found in the materials that comprise the crude oil category are hydrocarbons that are not subject to hydrolysis because they lack functional groups that hydrolyze.

Reliability : (1) valid without restriction

(36)

3.3.2 DISTRIBUTION

Media : air - biota - sediment(s) - soil - water
Method : Calculation according Mackay, Level I
Year : 1981

Remark : Distribution has been calculated according to Mackay Level I using the parameters defined in a paper by van der Zandt and van Leeuwen. The lower molecular weight components will mainly partition to air, with a

maximum of about 1% of mono-aromatic hydrocarbons partitioning to water. As the molecular weights increase, there is less tendency for the hydrocarbons to partition to air, the environmental distribution being preferentially to soil and sediment. Collectively, the wide molecular weight range of the hydrocarbons in crude oil will mean that at equilibrium, distribution will be mainly to air and soil, with much less than 1.0% being present in water.

Result : Results for percent distribution of typical hydrocarbons found in crude oils are shown in tabular form as follows:

air	water	soil	sediment	susp.matter	biota
propane					
100.0	0.0	0.0	0.0	0.0	0.0
n-butane					
100.0	0.0	0.0	0.0	0.0	0.0
n-hexane					
100.0	0.0	0.0	0.0	0.0	0.0
n-octane					
99.9	0.0	0.1	0.0	0.0	0.0
n-decane					
99.1	0.0	0.9	0.0	0.0	0.0
n-tetradecane					
76.7	0.0	22.8	0.5	0.0	0.0
n-eicosane					
0.0	0.0	97.7	2.2	0.1	0.0
cyclohexane					
99.9	0.0	0.1	0.0	0.0	0.0
benzene					
98.8	1.1	0.1	0.0	0.0	0.0
toluene					
98.4	1.0	0.6	0.0	0.0	0.0
p-xylene					
97.8	1.0	1.2	0.0	0.0	0.0
o-xylene					
97.4	1.2	1.4	0.0	0.0	0.0
ethylbenzene					
98.2	0.8	1.0	0.0	0.0	0.0
n-butylbenzene					
91.0	0.4	8.5	0.2	0.0	0.0

Reliability : (1) valid without restriction

(16) (46)

3.6 BOD5, COD OR BOD5/COD RATIO

BOD5
Method : Theoretical Oxygen Demand (TOD) was calculated
Year : 1971
GLP : no data

Method : Tests were run at 30 °C in fresh water using a respirometric procedure with crude oil at 50 and 70 mg/l. Prior to the test, the crude was "topped" at up to 100 °C to remove light ends, which comprised about 10% of the material

Result : The BOD/TOD ratio after 10 days was 0.3 at 50 mg/l and 0.04 at 70 mg/l. With the addition of nutrients, using ammonia at up to 4.6 mg/l of nitrogen and phosphate at up to 15 mg/l of phosphorus, the BOD/TOD ratio increased to a maximum of 0.3 after 5 days , and 0.34 after 10 days. In further studies, run at 10 °C in sea water in the presence of nutrients, the

degradation rate was found to be lowered by a factor of 2 to 3 below that at 30 °C. The reference quotes the BOD5/TOD ratio, but does not give the individual values for BOD5 and TOD.

Reliability : (2) valid with restrictions

(7) (16)

3.8 ADDITIONAL REMARKS

Memo : General

Remark : The world production of crude oil per year is of the order of 3 billion tonnes per year, of which about half is transported by sea. In his 1989 publication, Clark states that the best estimate of petroleum hydrocarbons entering the sea per year is about 3 million tonnes, of which about one million tonnes is attributable to crude oil. Such pollution arises from the cleaning of oil tanker compartments, offshore oil production operations, discharge of coastal refinery effluent and spillage from oil tankers. Crude oil also enters the oceans by natural seepage from undersea locations. Particular attention has focused on major oil tanker spillages, notably involving the Torrey Canyon in 1969, the Amoco Cadiz in 1978 and the Exxon Valdez in 1990. As a result, the processes determining the fate of oil in seawater are reasonably well understood and have been reviewed by Atlas and Bartha. Initially, the oil spreads out as a film on the sea surface as a result of wind and wave action. The more volatile, lower molecular weight hydrocarbons are lost by evaporation. Polar compounds and the mono-aromatic hydrocarbons have an appreciable water solubility and are taken into solution. A key ancillary process is that of emulsification, since crude oil has a natural tendency to form emulsions in sea water. Such emulsions are usually of the oil-in-water type, but may also be of the water-in-oil type. The latter are often of the intractable 'chocolate mousse' type. Significant amounts of crude oil, particularly the higher molecular weight compounds, sink naturally, rolling along the ocean bottom picking up sand and shells and forming tarry balls which are resistant to degradation by any method. Hydrocarbons may also reach the bottom sediments by sorption onto suspended particles which ultimately settle on the sea floor. Spilt oil also undergoes chemical changes, particularly oxidation by free radical mechanisms initiated by sunlight.
The initial products of such reactions are hydroperoxides, and these in turn form compounds such as alcohols, acids and aldehydes, many of which have an appreciable water solubility. Polymerization also occurs to yield intractable tarry materials.
The bulk of spilt crude oil is biodegraded by the micro-organisms present in sea water. Emulsification to form oil-in-water emulsions yields small particles of crude oil that are biodegraded by bacteria, yeasts, fungi and actinomycetes. Many factors influence the rate of biodegradation, in particular temperature, dissolved oxygen concentration and the availability of nitrogen and phosphorus nutrients. Adapted micro-organisms are often found in ocean areas where crude oil spills are common. Zobell has calculated that where an adapted microbial population is available in well-aerated sea water at 20 to 30 °C, the rate of crude oil oxidation ranges from 0.02 to 0.2 g of oil oxidized/m^2/day. The same author found experimentally that complete oxidation of 1.0 mg of hydrocarbon requires between 3 and 4 g of oxygen, i.e. it has a BOD of 3 to 4 mg oxygen/mg. Since the oxygen content of sea water is between 6 and 11 mg/liter, depending on salinity and temperature, this means that about 320 000 liters of sea water is required to oxidize one liter of crude oil. Crude oil

contains hydrocarbons of well-defined generic types that are biodegraded at different rates. n-Alkanes are readily degraded in sea water, since many micro-organisms can utilize them. Branched-chain or iso-alkanes are less readily biodegraded but they do ultimately biodegrade. The degradation of cycloalkanes has not been extensively studied, but the ring structure is resistant to biodegradation. Aromatic hydrocarbons are also resistant to biodegradation, but a few micro-organisms are able to utilize them. High molecular weight compounds, the tars and asphaltenes, degrade very slowly.

(4) (12) (16) (17) (80) (81)

4.1 ACUTE/PROLONGED TOXICITY TO FISH

Type	:	semistatic
Species	:	Oncorhynchus mykiss (Fish, fresh water)
Exposure period	:	96 hour(s)
Unit	:	mg/l
Limit test	:	no
Analytical monitoring	:	yes
Method	:	OECD Guide-line 203 "Fish, Acute Toxicity Test"
Year	:	2001
GLP	:	yes
Test substance	:	CAS 8002-05-9 Crude oil - 0.5% paraffinic

Method : LL_{50} at 96 hr calculated using the Trimmed Spearman-Karber Method (Hamilton, et al, 1977)

Result : Mortality (no. of deaths/treatment) at 96 hrs: 0, 0, 0, 0, 5 and 10, respectively for control, 1.4, 3.2, 8.5, 21 and 50 mg/l treatments.
96-hr LL_{50} = 21 mg/l, 95% C.I: 16-28 mg/l (as nominal loading rate)
96-hour No Observed Effect Loading (NOEL) was 8.5 mg/l (observed.)
Results are quoted in terms of 50% Lethal Loading (LL_{50}), the loading rate of test substance resulting in 50% mortality of the test species exposed to the WAF.

Measured BTEX (mg/l) in test treatment (mg/l load)

Day	Control	1.4	3.2	8.5	21	50
0(new)	ND	0.129	0.382	0.835	1.94	4.73
1 (old)	ND	0.086	0.243	0.665	1.60	4.04
3 (new)	0.005	0.148	0.301	0.973	1.86	NA
4 (old)	0.005	0.127	0.255	0.813	1.70	NA

ND=not detected, NA=not analyzed due to 100% mortality
The Practical Quantitation Limit (PQL) was approximately 0.0035 mg/l (3.5ng/ml) which corresponds to the concentration of the lowest standard analyzed.

Test condition : Test solutions were prepared as water accommodated fractions (WAF). The control and dilution water was a reconstituted moderately hard water aerated prior to use. Water quality analyses for the dilution water was as follows:
alkalinity = 72 mg/l as $CaCO_3$
hardness = 96 mg/l (as $CaCO_3$)
specific conductance = 320 µmhos
pH = 7.7
dissolved oxygen = 8.6 ppm.
Based on results of range-finding tests for a similar petroleum product, measured loading rates of 1.4, 3.2, 8.5, 21 and 50 mg/l of crude oil to water were used to prepare test solutions for definitive toxicity tests. Test substance, added volumetrically, was mixed for each individual treatment in dilution water for 24 hours in 12-liter stoppered containers with approximately 20% vortex and less than 10% headspace volume. The WAFs were allowed to settle and cool to test temperature for 1-2 hours prior to drawing off the aqueous solutions for testing. Fish were 85 days old at test initiation and were obtained from Pierce Associates Inc., West Buxton, ME, Lot 454. Loading of fish body mass to treatment was 0.551 g fish per liter of aqueous solution, mean length at termination was 4.1 cm (sd=0.2), and mean weight was 0.496 g (sd=0.078). Test vessels were 4-liter glass aspirator bottles with Teflon® covered neoprene

stoppers. Two 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 at least 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 BTEX. Mean water temperature range was 13.8 (sd=0.1) °C. Test photoperiod was 16 hrs. light and 8 hr dark, dissolved oxygen measurements ranged from 6.5 to 8.8 ppm, pH values between 6.8 and 7.8.

Reliability : (1) valid without restriction

(22)

Type : semistatic
Species : Oncorhynchus mykiss (Fish, fresh water)
Exposure period : 96 hour(s)
Unit : mg/l
Limit test : no
Analytical monitoring : yes
Method : OECD Guide-line 203 "Fish, Acute Toxicity Test"
Year : 2001
GLP : yes
Test substance : CAS 8002-05-9 Crude oil 3% paraffinic

Method : LL_{50} at 96 hr calculated using the Trimmed Spearman-Karber Method (Hamilton, et al, 1977)

Result : Mortality (no. of deaths/treatment) at 96 hrs: 0, 0, 0, 0, 5 and 10, respectively for control, 2.7, 6.8, 16, 40 and 109 mg/l treatments.
96-hr LL_{50} = 41 mg/l, 95% C.I: 30-55 mg/l (as nominal loading rate)
96-hour No Observed Effect Loading (NOEL) was 16 mg/l (observed.)
Results are quoted in terms of 50% Lethal Loading (LL_{50}), the loading rate of test substance resulting in 50% mortality of the test species exposed to the WAF.

Measured BTEX (mg/l) in test treatment (mg/l load)

Day	Control	2.7	6.8	16	40	109
0 (new)	ND	0.093	0.366	0.541	1.36	2.02
1 (old)	0.050	0.066	0.184	0.411	0.857	1.84
3 (new)	ND	0.096	0.272	0.585	1.28	2.01
4 (old)	0.031	0.084	0.221	0.484	1.03	NA

ND=not detected, NA=not analyzed due to 100% mortality
Guideline/protocol deviations: DO level was 53%, not 60% of saturation.

Test condition : Test solutions were prepared as water accommodated fractions (WAF). The control and dilution water was a reconstituted moderately hard water aerated prior to use. Water quality analyses for the dilution water was as follows:
alkalinity = 78 mg/l as $CaCO_3$
hardness = 105 mg/l (as $CaCO_3$)
specific conductance = 324 µmhos
pH = 7.6
dissolved oxygen = 8.4 ppm.
Nominal loading rates of 0, 3.8, 16 and 74 mg/l were used to prepare test solutions for range-finding toxicity tests.
Based on results of range-finding tests, measured loading rates of 0, 2.7, 6.8, 16, 40 and 109 mg/l of crude oil to water were used to prepare test solutions for definitive toxicity tests. Test substance, added volumetrically, was mixed for each individual treatment in dilution water for 24 hours in 13-

liter stoppered containers with approximately 20% vortex and less than 10% headspace volume. The WAFs were allowed to settle and cool to test temperature for 1-2 hours prior to drawing off the aqueous solutions for testing. Fish were 85 days old at test initiation and were obtained from Pierce Associates Inc., West Buxton, ME, Lot 454. Loading of fish body mass to treatment was 0.4 g fish per liter of aqueous solution, mean length at termination was 3.9 cm (sd=0.2), and mean weight was 0.466 g (sd=0.034). Test vessels were 4-liter glass aspirator bottles with Teflon® covered neoprene stoppers. Two 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 at least 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 BTEX. Water temperature range was 14.3 to 14.9 °C. Test photoperiod was 16 hrs. light and 8 hr dark, dissolved oxygen measurements ranged from 5.4 to 8.8 ppm, pH values between 6.8 and 7.8.

Reliability : (1) valid without restriction

(27)

Type	:	static
Species	:	Cyprinodon variegatus (Fish, estuary, marine)
Exposure period	:	96 hour(s)
Unit	:	mg/l
LC50	:	2900 - 80000 measured/nominal
Analytical monitoring	:	yes
Method	:	Procedure as detailed in paper by Anderson (see Reference)
Year	:	1974
GLP	:	no data
Test substance	:	South Louisiana and Kuwait crude oils

Method : The test species was the Sheepshead Minnow. Dispersions of two crude oil samples in water were prepared by shaking the constituents together vigorously for 5 minutes on a shaker platform. Fish tests were run at 5 concentrations. Figures quoted are for the loading rates.

Remark : These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. Since significant evaporative losses of lower hydrocarbons may have occurred, the LC_{50} values are expected to be lower than those cited. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '3', not reliable.

Result : The results were as follows:

For Kuwait crude:
48-hour LC_{50} = 80 000+ mg/l
96-hour LC_{50} = 80 000+ mg/l
For South Louisiana crude:
48-hour LC_{50} = 33 000 mg/l
96-hour LC_{50} = 29 000 mg/l

Parallel tests run with water-soluble fractions (WSF) failed to produce any meaningful results.

Reliability : (3) invalid

(2)

Type	:	static
Species	:	Fundulus similis (Fish, estuary, marine)
Exposure period	:	96 hour(s)
Unit	:	mg/l
LC50	:	6000 - 14800 measured/nominal
Method	:	Procedure as detailed in paper by Anderson (see Reference)
Year	:	1974
GLP	:	yes
Test substance	:	South Louisiana and Kuwait crude oils
Method	:	The test species was the Longnose Killifish. Dispersions of two crude oil samples in water were prepared by shaking the constituents together vigorously for 5 minutes on a shaker platform. Fish tests were run at 5 concentrations. Figures quoted are for the loading rates.
Remark	:	These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. Since significant evaporative losses of lower hydrocarbons may have occurred, the LC_{50} values are expected to be lower than those cited. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '3', not reliable.
Result	:	The results were as follows: For Kuwait crude: 48-hour LC_{50} = 14 800 mg/l 96-hour LC_{50} = 14 800 mg/l For South Louisiana crude: 48-hour LC_{50} = 6000 mg/l 96-hour LC_{50} = 6000 mg/l
Reliability	:	(3) invalid

(2)

Type	:	static
Species	:	Menidia beryllina (Fish, estuary, marine)
Exposure period	:	96 hour(s)
Unit	:	mg/l
LC50	:	3700 - 9400 measured/nominal
Analytical monitoring	:	no
Method	:	Procedure as detailed in paper by Anderson (see Reference)
Year	:	1974
GLP	:	yes
Test substance	:	South Louisiana and Kuwait crude oils
Method	:	The test species was the Tidewater Silverside. Dispersions of two crude oil samples in water were prepared by shaking the constituents together vigorously for 5 minutes on a shaker platform. Fish tests were run at 5 concentrations. Figures quoted are for the loading rates.
Remark	:	These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. Since significant evaporative losses of lower hydrocarbons may have occurred, the LC_{50} values are expected to be lower than those cited. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '3', not reliable.
Result	:	The results were as follows: For Kuwait crude: 48-hour LC_{50} = 15 000 mg/l, 96-hour LC_{50} = 9400 mg/l

For South Louisiana crude:
48-hour LC_{50} = 5000 mg/l
96-hour LC_{50} = 3700 mg/l

Source : CONCAWE Bruxelles
Reliability : (3) invalid

(2)

Type : static
Species : Salmo gairdneri (Fish, estuary, fresh water)
Exposure period : 48 hour(s)
Unit : mg/l
Analytical monitoring : yes
Method : Procedure as detailed in paper by Lockhart, Danell and Murray (see Reference)
Year : 1987
GLP : yes
Test substance : Norman Wells crude oil

Method : The test species was the Rainbow Trout. A water-soluble fraction (WSF) of the test substance was prepared by adding crude oil to water at a concentration of 12.5 ml/l and stirring for 2 hours. After settling for 72 hours, groups of 5 fish were exposed to solutions containing 20, 30, 40 and 50% WSF. Three series of tests were run:
(a) with closed vessels
(b) with open vessels, and
(c) with aerated vessels.

Remark : These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '2', reliable with restrictions.

Result : The results were as follows:
48-hour LC_{50} (open vessel) = 10.4 mg/l
48-hour LC_{50} (closed vessel) = 11.6 mg/l
based on measurements of dissolved hydrocarbons made at the beginning and end of the 48-hour period. No fish died in the aerated vessels, and hydrocarbons were undetectable in solution at the 48-hour time point in these studies.

Source : CONCAWE Bruxelles
Reliability : (2) valid with restrictions

(44)

Type : static
Species : Salmo gairdneri (Fish, estuary, fresh water)
Exposure period : 96 hour(s)
Unit : mg/l
LC50 : 291 measured/nominal
Analytical monitoring : no
Method : procedure as detailed in paper by Westlake (see Reference)
Year : 1991
GLP : yes
Test substance : Forties crude oil

Method : The test species was the Rainbow Trout. Dispersions of oil in water at 8 concentrations in the range 57 to 1330 mg/l were prepared by shaking the constituents together in closed jars for 5 minutes using a reciprocating table. Fish tests were run in open vessels.

Remark	:	These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. Since significant evaporative losses of lower hydrocarbons may have occurred, the LC_{50} values are expected to be lower than those cited. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '3', not reliable.
Result	:	The results were as follows: 24-hour LC_{50} = 756 mg/l 96-hour LC_{50} = 291 mg/l 168-hour LC_{50} = 133 mg/l As the tests were run in open vessels, significant evaporative losses of lower hydrocarbons will have occurred and hence the measured LC_{50} values underestimate the true ecotoxicity.
Source	:	CONCAWE Bruxelles
Reliability	:	(3) invalid

(78)

Type	:	static
Species	:	Salmo gairdneri (Fish, estuary, fresh water)
Unit	:	mg/l
LC50	:	258 measured/nominal
Analytical monitoring	:	no
Method	:	procedure as detailed in paper by Westlake (see Reference)
GLP	:	yes
Test substance	:	Forties crude oil
Method	:	The test species was the Rainbow Trout. Crude oil was added to water at 8 concentrations in the range 57 to 1330 mg/l in open vessels fitted with shielded propeller stirrers. A constant rate of stirring was maintained during the 7-day exposure period to the fish.
Remark	:	These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. Since significant evaporative losses of lower hydrocarbons may have occurred, the LC_{50} values are expected to be lower than those cited. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '3', not reliable.
Result	:	The results were as follows: 24-hour LC_{50} = 557 mg/l 96-hour LC_{50} = 258 mg/l 168-hour LC_{50} = 253 mg/l As the tests were run in open vessels, significant evaporative losses of lower hydrocarbons will have occurred and hence the measured LC_{50} values underestimate the true ecotoxicity.
Source	:	CONCAWE Bruxelles
Reliability	:	(3) invalid

(78)

4.2 ACUTE TOXICITY TO AQUATIC INVERTEBRATES

Species	:	Crangon crangon (Crustacea)
Exposure period	:	96 hour(s)
Unit	:	mg/l
LC50	:	27 - 110 measured/nominal

Analytical monitoring	:	no
Method	:	Procedure as detailed in paper by Franklin and Lloyd (see Reference)
Year	:	1982
GLP	:	no data
Test substance	:	Samples of 8 crude oils
Method	:	The test species was the Brown Shrimp. Groups of 20 shrimps were exposed to nominal concentrations in the range 17 to 3400 mg/l of each crude oil in sea water. After addition of the crude oil, the solutions were stirred at a constant rate using a shielded stirrer. Crude oil solutions were renewed after 48 hours. Mortalities were recorded daily.
Remark	:	These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. Since no analytical determinations were reported, these data are ranked by API crude oil task group as '2', reliable with restrictions.
Result	:	The lowest LC_{50} value was obtained for Brent crude and the highest for Thistle crude. Other North Sea crude oils tested were from the Argyll, Auk, Beryl, Claymore, Ekofisk, Forties, Montrose, Murchison and Piper fields.
Source	:	CONCAWE Bruxelles
Reliability	:	(2) valid with restrictions

(32)

Species	:	Crangon crangon (Crustacea)
Exposure period	:	96 hour(s)
Unit	:	mg/l
LC50	:	41 - 119 measured/nominal
Analytical monitoring	:	no
Method	:	Procedure as detailed in paper by Franklin and Lloyd (see Reference)
Year	:	1982
GLP	:	no data
Test substance	:	Samples of 8 crude oils
Method	:	The test species was the Brown Shrimp. Groups of 20 shrimps were exposed to nominal concentrations in the range 17 to 3400 mg/l of each crude oil in sea water. After addition of the crude oil, the solutions were stirred at a constant rate using a shielded stirrer. Crude oil solutions were renewed after 48 hours. Mortalities were recorded daily.
Remark	:	These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. Since no analytical determinations were reported, these data are ranked by API crude oil task group as '2', reliable with restrictions.
Result	:	The lowest LC_{50} value was obtained for Abu Dhabi crude and the highest for Iranian Light crude. Other crude oils tested were from Libya, Saudi Arabia, Nigeria, Kuwait, Iraq and Iran (heavy crude).
Source	:	CONCAWE Bruxelles
Reliability	:	(2) valid with restrictions

(33)

Species	:	Daphnia magna (Crustacea)
Exposure period	:	24 hour(s)
Unit	:	mg/l
EC50	:	36 measured/nominal
Analytical monitoring	:	no
Method	:	OECD Guide-line 202
Year	:	1981

GLP	:	yes
Test substance	:	Forties crude oil
Method	:	Dispersions of oil in water at 5 concentrations in the range 10 to 100 mg/l were prepared by shaking the constituents together in closed jars for 5 minutes using a reciprocating table. Groups of 10 daphnids were exposed to the resulting solutions. Tests were run in open vessels.
Remark	:	These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. Since significant evaporative losses of lower hydrocarbons may have occurred, the LC_{50} values are expected to be lower than those cited. These data are ranked by API crude oil task group as '3', not reliable.
Result	:	The results were as follows: 24-hour EC_{50} = 36 mg/l. As the tests were run in open vessels, significant evaporative losses of lower hydrocarbons will have occurred and hence the measured EC_{50} value underestimates the true ecotoxicity.
Source	:	CONCAWE Bruxelles
Reliability	:	(3) invalid

(78)

Species	:	Daphnia magna (Crustacea)
Exposure period	:	24 hour(s)
Unit	:	mg/l
EC50	:	= 42 measured/nominal
Analytical monitoring	:	no
Method	:	OECD Guide-line 202
Year	:	1981
GLP	:	yes
Test substance	:	Forties crude oil
Method	:	Crude oil was added to water at 8 concentrations in the range 10 to 100 mg/l in open vessels fitted with shielded propeller stirrers. A constant rate of stirring was maintained during the 24-hour exposure period to groups of 10 daphnids.
Remark	:	These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. Since significant evaporative losses of lower hydrocarbons may have occurred, the LC_{50} values are expected to be lower than those cited. These data are ranked by API crude oil task group as '3', not reliable.
Result	:	The results were as follows: 24-hour EC_{50} = 42 mg/l. As the tests were run in open vessels, significant evaporative losses of lower hydrocarbons will have occurred and hence the measured EC_{50} value underestimates the true ecotoxicity.
Source	:	CONCAWE Bruxelles
Reliability	:	(3) invalid

(78)

Type	:	
Species	:	Mysidopsis bahia (Crustacea)
Exposure period	:	96 hour(s)
Unit	:	mg/l
LL50	:	618 measured/nominal
Analytical monitoring	:	yes
Method	:	40 CFR Part 797.1930, Mysid Shrimp Acute Test
Year	:	1993

GLP	:	yes
Test substance	:	Kuwait crude oil
Method	:	Tests were carried out in polypropylene containers using water accommodated fractions prepared by physical dispersion. These were renewed daily and kept tightly covered with teflon sheets. Loading rates of 340, 695, 1550, 3470 and 7740 mg/l were used in this study. The test species was the estuarine mysid, Mysodopsis bahia.
Remark	:	These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '1', reliable without restrictions.
Result	:	A lethal loading rate concentration LL_{50} (96h) of 618 mg/l was calculated from data presented in the original publication after consultation with the main author. Weight percent distribution for the crude oil components with BP up to 425 °F are: paraffins: 68.56%; aromatics: 15.69 and naphthenes: 11.86%.
Source	:	CONCAWE Bruxelles
Reliability	:	(1) valid without restriction

Species	:	Holmesiysis costata
Exposure period	:	96 hour(s)
Unit	:	mg/l
LL50	:	39.5 measured/nominal
Analytical monitoring	:	yes
Method	:	40 CFR Part 797.1930, Mysid Shrimp Acute Test
Year	:	1993
GLP	:	yes
Test substance	:	Kuwait crude oil
Method	:	Tests were carried out in polypropylene containers using water accommodated fractions prepared by physical dispersion. These were renewed daily, and kept tightly covered with teflon sheets. Loading rates of 11.6, 34.3, 99.1, 284 and 862 mg/l were used in this study. The test species was the kelp forest mysid, Holmesimysis costata.
Remark	:	These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '1', reliable without restrictions.
Result	:	A lethal loading rate concentration, LL_{50} (96h) of 39.5 mg/l was calculated from data presented in the original publication after consultation with the main author. Weight percent distribution for the crude oil components with BP up to 425 °F are: paraffins: 68.56%; aromatics: 15.69 and naphthenes: 11.86%.
Source	:	CONCAWE Bruxelles
Reliability	:	(1) valid without restriction

(6)

Type	:	semistatic
Species	:	Kelp Forest Mysid (Holmesymis costata)
Exposure period	:	96 hour(s)
Unit	:	mg/l
Limit Test	:	no
Analytical monitoring	:	yes

Method : USEPA 600 4-90-027
Year : 2002
GLP : yes
Test substance : CAS 8002-05-9 Crude oil, 0.5% paraffinic

Method : LL_{50} at 96 hr calculated maximum likelihood analysis based on D.J. Finney, 1971

Result : Mortality (no. of deaths/treatment) at 96 hrs: 0, 2, 2, 1, 15 and 20, respectively for control, 0.14, 0.28, 1.4, 3.5, and 11 mg/l treatments.
96-hr LL_{50} = 2.7 mg/l, 95% C.I: 2.2-3.6 mg/l (as actual loading rate)
Results are quoted in terms of 50% Lethal Loading (LL_{50}), the loading rate of test substance resulting in 50% mortality of the test species exposed to the WAF.

Measured BTEX (mg/l) in test treatment (mg/l load)

Day	Control	0.14	0.28	1.4	3.5	11
0 (new)	ND	ND	ND	0.049	0.167	0.500
1 (old)	ND	ND	ND	0.0118	0.053	0.239
3 (new)	0.047	ND	0.003	0.0586	0.203	NA
4 (old)	ND	ND	ND	0.017	0.0939	NA

ND=not detected, NA=not analyzed due to 100% mortality
The practical quantitation limit was 0.003 mg/l. Measured soluble concentrations of BTEX may have been reduced by the presence of brine shrimp in the test chambers

Test condition : Test species: Juvenile Mysids used in the test were 1day old and were obtained from A.K. Siewers, Santa Cruz, CA, USA; the Mysids were acclimated for 3 days under laboratory conditions in seawater supplied by the organism vendor and fed from in-house cultures of Artemia salina nauplii that were less than 24 hours old. Test organisms were fed brine shrimp once per day during the study.

Test System: Individual water accommodated fractions (WAFs) were prepared for each test level. New test solutions were prepared for each 24-hour interval throughout the duration of the test. The control and dilution water was natural seawater collected from Manasquan Inlet, NJ, (NJDEP designated collection site) with a salinity of 33% and passed through 0.45 um filters prior to use.

Based on results of range-finding tests for a similar petroleum product, actual loading rates of 0, 0.14, 0.28, 1.4, 3.5 and 11 mg/l of crude oil to water were used to prepare test solutions for definitive toxicity tests. Test substance, added volumetrically, was mixed for each individual treatment in dilution water for 24 hours in appropriate-sized, stoppered containers with approximately 20% vortex and less than 10% headspace volume. The WAFs were allowed to settle for 1-2 hours prior to drawing off the aqueous solutions for testing. A new WAF was prepared 24 hours prior to each renewal. Water quality (temperature, pH, salinity, and dissolved oxygen) measurements were recorded daily on the new solutions. Temperature, pH, and dissolved oxygen measurements were recorded on the old solutions (composite of replicates).

Test conditions: Two replicate chambers per treatment were tested. Each replicate contained ten mysids. Replicate chambers were 500 ml glass jars with Teflon screw lids, containing approximately 450 ml of test solution with no headspace. All test and control solutions were replenished daily. Renewals were accomplished by transferring the mysids to fresh solution in a second set of test chambers on days 1, 2 and 3.

Light: 16-hour light per day and a daylight intensity that ranged from 494 to 720 Lux.

Test temperature: 11.9°C (S.D.=0.2).

Water chemistry:
Dissolved oxygen ranged from 6.4 - 7.0 mg/l
pH ranged from 8.1 - 8.5
salinity ranged from 32 - 34 ppt.

Freshly prepared and old WAF test solutions were analyzed by GC-FID for concentrations of BTEX.

Reliability : (1) valid without restriction
3 (26)

Type : semistatic
Species : Kelp Forest Mysid (Holmesymis costata)
Exposure period : 96 hour(s)
Unit : mg/l
Limit Test : no
Analytical monitoring : yes
Method : USEPA 600 4-90-027
Year : 2001
GLP : yes
Test substance : CAS 8002-05-9 Crude oil, 3% paraffinic

Method : LL_{50} at 96 hr calculated maximum likelihood analysis based on D.J. Finney, 1971

Result : Mortality (no. of deaths/treatment) at 96 hrs: 1, 2, 3, 6, 20 and 20, respectively for control, 0.6, 1.7, 3.6, 8.3 and 21 mg/l treatments.
96-hr LL_{50} = 4.1 mg/l, 95% C.I: 3.3-5.3 mg/l (as actual loading rate)
Results are quoted in terms of 50% Lethal Loading (LL_{50}), the loading rate of test substance resulting in 50% mortality of the test species exposed to the WAF.

Measured BTEX (mg/l) in test treatment (mg/l load)

Day	Control	0.6	1.7	3.6	8.3	21
0 (new)	ND	ND	ND	ND	0.072	0.247
1 (old)	ND	ND	ND	ND	ND	0.057
3 (new)	ND	ND	ND	0.019	NA	NA
4 (old)	ND	ND	ND	0.023	NA	NA

ND=not detected, NA=not analyzed due to 100% mortality
The practical quantitation limit was 0.003 mg/l. Measured soluble concentrations of BTEX may have been reduced by the presence of brine shrimp in the test chambers

Test condition : Test species: Juvenile Mysids used in the test were 1day old and were obtained from A.K. Siewers, Santa Cruz, CA, USA; the Mysids were acclimated for 3 days under laboratory conditions in seawater supplied by the organism vendor and fed from in-house cultures of Artemia salina nauplii that were less than 24 hours old. Test organisms were fed brine shrimp once per day during the study.

Test System: Individual water accommodated fractions (WAFs) were prepared for each test level. New test solutions were prepared for each 24-hour interval throughout the duration of the test. The control and dilution water was natural seawater collected from Manasquan Inlet, NJ,

(NJDEP designated collection site) with a salinity of 33% and passed through 0.45 um filters prior to use.

Nominal loading rates of 0, 1.8, 19 and 105 mg/l were used to prepare test solutions for range-finding toxicity tests.
Based on results of range-finding tests, actual loading rates of 0, 0.6, 1.7, 3.6, 8.3 and 21 mg/l of crude oil to water were used to prepare test solutions for definitive toxicity tests. Test substance, added volumetrically, was mixed for each individual treatment in dilution water for 24 hours in appropriate-sized, stoppered containers with approximately 20% vortex and less than 10% headspace volume. The WAFs were allowed to settle for 1-2 hours prior to drawing off the aqueous solutions for testing. A new WAF was prepared 24 hours prior to each renewal. Water quality (temperature, pH, salinity, and dissolved oxygen) measurements were recorded daily on the new solutions. Temperature, pH, and dissolved oxygen measurements were recorded on the old solutions (composite of replicates).

Test conditions: Two replicate chambers per treatment were tested. Each replicate contained ten mysids. Replicate chambers were 500 ml glass jars with Teflon® screw lids, containing approximately 450 ml of test solution with no headspace. All test and control solutions were replenished daily. Renewals were accomplished by transferring the mysids to fresh solution in a second set of test chambers on days 1, 2 and 3.
Light: 16-hour light per day and a daylight intensity that ranged from 494 to 720 Lux.
Test temperature: 12.3°C (S.D.=0.1).
Water chemistry:
Dissolved oxygen ranged from 6.4 - 7.0 mg/l
pH ranged from 8.1 - 8.5
salinity ranged from 32 – 33 ppt.
Freshly prepared and old WAF test solutions were analyzed by GC-FID for concentrations of BTEX.

Reliability : (1) valid without restriction

(25)

4.3 TOXICITY TO AQUATIC PLANTS E.G. ALGAE

Species	:	Anabaena doloilum (Algae)
Endpoint	:	growth rate
Exposure period	:	15 day(s)
Unit	:	mg/l
Analytical monitoring	:	yes
Method	:	Procedure as detailed by Gaur and Singh (see Reference)
Year	:	1989
GLP	:	no data
Test substance	:	Assam crude oil

Method : Tests were run using:
(a) a water-soluble fraction (WSF) of the test substance, and
(b) the test substance in equilibrium with the algal suspension medium.
To prepare the WSF, crude oil was added to the sterilized medium in the ratio 1:20, and was stirred in closed bottles for 12 hours. After a 4-hour separation period the aqueous phase was separated and diluted for the tests. In the direct loading tests, crude oil was applied to absorbent pads and these were held in the culture suspension. Analysis was done by gas

chromatography, following solvent extraction of the aqueous phase using n-pentane.

Remark : These data are included to provide supporting evidence of the expected cumulative toxicity of the soluble components in crude oil. These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '1', reliable without restrictions.

Result : The WSF method gave a mean 15-day EC_{50} of 9.06 mg/l, and the direct loading (or whole oil) method gave a 15-day EC_{50} of 5.73 mg/l, both based on measured dissolved hydrocarbon concentrations in the aqueous phase.

Source : CONCAWE Bruxelles

Reliability : (1) valid without restriction

(34)

Species : Macrocystis pyrifera
Exposure period : 48 hour(s)
Unit : mg/l
Limit test : no
Analytical monitoring : yes
Year : 2001
GLP : yes
Test substance : CAS 8002-05-9 Crude oil - 0.5% paraffinic

Method : The method used was described by Chapman, G.A., D.L. Denton and J.M. Lazorchak. (1995).

Statistical method
Lethal Loading and Effect Loading: Norberg-King, T.J., A Linear Interpolation Method for Sublethal Toxicity: The Inhibition Concentration (ICp) Approach (Version 2.0). July 1993. U.S. Environmental Protection Agency, Environmental Research Laboratory, Duluth MN.

Result : 48 hour results, based on mg of test substance per liter of natural seawater

Germination LL_{50} (mg/l)	**Tube Length EL_{50} (mg/l)**	**Germination NOEL (mg/l)**	**Germination LOEL (mg/l)**	**Tube Length NOEL (mg/l)**	**Tube Length LOEL (mg/l)**
122	122	52	146	18	52

Analytical results

Nominal (mg/l)	**Measured Concentration (mg/l as BTEX)**	
	Day 0	Day 2
Control	none detected	none detected
18	0.542	0.478
52	1.82	1.23
146	5.91	3.27
415	15.2	5.88
1185	20.0	11.3

Practical Quantitation Limit (PQL) was approximately 0.0035 mg/l (3.5ng/ml)

Test condition : Individual test treatment solutions were prepared as Water Accommodated Fractions (WAFs). Actual loading levels of test substance in natural seawater (dilution water) were 18 mg/l, 52 mg/l, 146 mg/l, 415 mg/l and 1185 mg/l. A control treatment consisting of natural seawater with no test

substance was also prepared. The WAFs were prepared by adding the appropriate amount of test substance to natural seawater in glass aspirator bottles and stirring on magnetic stirplates for 24 hours. Natural seawater was filtered through a 0.45 µm filter, then a 0.2 µm filter (collected from Manasquan Inlet, NJ, NJDEP designated collection site).
The salinity of the seawater was 34 ± 2 ppt.
Ammonia (NH_3 + NH_4+) = 0 mg/l
Nitrite (NO_2-) = <0.3 mg/l
Nitrate (N) = 1 mg/l.
The mixtures were allowed to settle and cool to test temperature for 1.25 hour before removing the aqueous portions (WAFs) for testing. Samples of the WAFs prepared at each loading level were taken on Day 0 and at termination from each treatment (composite of replicates). The samples were analyzed for benzene, toluene, ethylbenzene and o-xylene (BTEX), all hydrocarbons detected in the entire chromatographic run were quantified versus the BTEX standards and the reported results reflect the sum of all quantified compounds. Samples were taken with no headspace in volatile organic analysis (VOA) vials.
Test temperature was 14.0 (sd=0.3) °C.

The kelp sporophylls (reproductive blades) were received from A. K. Siewers, Santa Cruz, CA 95060, packed on ice at 7.5°C, the day after they were harvested. The sporophylls were rinsed with 0.2 µm filtered seawater at 14°C and blotted dry with paper towels. They were then desiccated at 14°C for one hour. The sporophylls were rinsed again with 0.2µm filtered seawater at 14°C. Approximately 20 of the sporophylls were placed into a one liter glass beaker containing 800 ml of 0.2 µm filtered seawater at 14°C. The blades were left in the beaker for 30 minutes while the zoospores released. The blades were removed and the spore solution was allowed to settle for 20 minutes. 250 ml was poured from the top of the beaker, this was the spore stock. The zoospores were released from the sporophylls and inoculated into test chambers within a two hour period. The zoospore release was initiated within 24 hours of sporophyll harvest.

Five replicate chambers for each dose treatment consisted of 500 ml size glass jars sealed with Teflon® lined screw type lids, containing approximately 450 ml of test solution to minimize headspace and prevent loss of volatile components.
A pre-cleaned 2.5 cm x 7.6 cm glass slide was placed in the bottom of each jar prior to adding test solution. The initial concentration was approximately 7.5 x 10^3 zoospores/ml in each replicate chamber. The spore stock density was determined as follows: 1 ml of the stock and 1 ml of glacial acetic acid were added to 8 ml of 0.2 µm filtered seawater in a graduated cylinder. The solution was mixed and counts were performed using a hemacytometer at 100x. Each test chamber, containing approximately 7500 spores, was labeled to show the study number, loading level, replicate and randomization number. Each test and control chamber were observed for germination of the gametophyte spores and length of the embryonic gametophyte germination tube after approximately 48 hrs. The following procedure was used. The glass slide was removed from the jar and the bottom of the slide was dried with a paper towel. A 24 x 50 mm cover slideslip was placed onto the slide and the edges were dabbed. Germination was considered successful if a germ tube was present on the settled zoospore. Germination was considered unsuccessful if no germ tube was visible.
400x magnification was used to differentiate germination and measure the tubes. A spore had to have a protuberance that extended at least one

spore diameter from the edge of the spore to be considered germinated. The first 100 spores, which were easily identifiable, were counted while moving the slide across the field of view of the microscope. Measurements were made by moving the slide and choosing the first 10 spores with straight germ tubes within the same focal plane of the microscope that were closest to the scale in the ocular micrometer. The ocular micrometer was calibrated with a stage micrometer at 400x to assure accurate measurements. The micrometer was calibrated from 3 to 25 µm. The total length of the tube from the edge of the original spore membrane was measured.

Water quality (temperature, pH, dissolved oxygen, and salinity) measurements were made on each treatment on Day 0 and at termination (composite of replicates). BTEX total concentration at termination was at least 80% of the initial concentration.

The study was deemed acceptable. Mean control germination was > 70% in the controls. Mean germination tube length was > 10 µm in the controls.

Reliability : (1) valid without restriction

(10) (24)

Species : Marine algae (Macrocystit pyrifera)
Exposure period : 48 hour(s)
Unit : mg/l
Limit test : no
Analytical monitoring : yes
Method : Chapman, G.A., D.L. Denton and J.M. Lazorchak. 1995
Year : 2001
GLP : yes
Test substance : CAS 8002-05-9 Crude oil 3% paraffinic

Method : Lethal Loading and Effect Loading: Norberg-King, T.J., A Linear Interpolation Method for Sublethal Toxicity: The Inhibition Concentration (ICp) Approach (Version 2.0). July 1993. U.S. Environmental Protection Agency, Environmental Research Laboratory, Duluth MN.

Result : 48 hour results, based on mg of test substance per liter of natural seawater (95%CI)

Germination LL_{50}	Tube Length EL_{50}	Germination NOEL	Germination LOEL	Tube Length NOEL	Tube Length LOEL
528 (497-558)	311 (257-403)	53	130	23	53

Analytical results

Nominal (mg/l)	Measured Concentration (mg/l as BTEX) Day 0	Day 2
Control	none detected	none detected
23	0.047	none detected
53	0.586	0.039
130	1.09	0.428
323	3.35	1.92
802	5.03	4.43

Practical Quantitation Limit (PQL) was approximately 0.0035 mg/l (3.5ng/ml)
Spore concentration was 1000 spore/ml, instead of 7500 spore/ml, as

required. However total spore number per replicate was adequate sample size for determination of end points.

Test condition : Individual test treatment solutions were prepared as Water Accommodated Fractions (WAFs). Actual loading levels of test substance in natural seawater (dilution water) were 23 mg/l, 53 mg/l, 130 mg/l, 323 mg/l and 802 mg/l. A control treatment consisting of natural seawater with no test substance was also prepared. The WAFs were prepared by adding the appropriate amount of test substance to natural seawater in glass aspirator bottles and stirring on magnetic stirplates for 24 hours. Natural seawater was filtered through a 0.45 µm filter, then a 0.2 µm filter (collected from Manasquan Inlet, NJ, NJDEP designated collection site). The salinity of the seawater was 34 ± 2 ppt. The mixtures were allowed to settle and cool to test temperature for 1 hour before removing the aqueous portions (WAFs) for testing. Samples of the WAFs prepared at each loading level were taken on Day 0 and at termination from each treatment (composite of replicates). The samples were analyzed for benzene, toluene, ethylbenzene and o-xylene (BTEX), all hydrocarbons detected in the entire chromatographic run were quantified versus the BTEX standards and the reported results reflect the sum of all quantified compounds. Samples were taken with no headspace in volatile organic analysis (VOA) vials. Mean test temperature was 14.1 (sd = 0.3) °C.

The kelp sporophylls (reproductive blades) were received from A. K. Siewers, Santa Cruz, CA 95060, packed on ice at 12.2°C, the day after they were harvested. The sporophylls were rinsed with 0.2 µm filtered seawater at 14°C and blotted dry with paper towels. They were then desiccated at 14°C for one hour. The sporophylls were rinsed again with 0.2µm filtered seawater at 14°C. Approximately 20 of the sporophylls were placed into a one liter glass beaker containing 800 ml of 0.2 µm filtered seawater at 14°C. The blades were left in the beaker for 30 minutes while the zoospores released. The blades were removed and the spore solution was allowed to settle for 25 minutes. 250 ml was poured from the top of the beaker, this was the spore stock. The zoospores were released from the sporophylls and inoculated into test chambers within a two hour period. The zoospore release was initiated within 24 hours of sporophyll harvest. Five replicate chambers for each dose treatment consisted of 500 ml size glass jars sealed with Teflon® lined screw type lids, containing approximately 450 ml of test solution. A pre-cleaned 2.5 cm x 7.6 cm glass slide was placed in the bottom of each jar prior to adding test solution. The initial concentration was approximately 1.0×10^3 zoospores/ml in each replicate chamber. The spore stock density was determined as follows: 1 ml of the stock and 1 ml of glacial acetic acid were added to 8 ml of 0.2 µm filtered seawater in a graduated cylinder. The solution was mixed and counts were performed using a hemacytometer at 100x. Each test chamber, containing approximately 1000 spores, was labeled to show the study number, loading level, replicate and randomization number. Each test and control chamber was observed for germination of the gametophyte spores and length of the embryonic gametophyte germination tube.
The following procedure was used. The glass slide was removed from the jar, and the bottom of the slide was dried with a paper towel. A 24 x 50 mm cover slideslip was placed onto the slide and the edges were dabbed. Germination was considered successful if a germ tube was present on the settled zoospore. Germination was considered unsuccessful if no germ tube was visible. 400x magnification was used to differentiate germination and measure the tubes. A spore had to have a protuberance that extended at least one spore diameter from the edge of the spore to be considered germinated. The first 100 spores, which were easily identifiable, were

counted while moving the slide across the field of view of the microscope. Measurements were made by moving the slide and choosing the first 10 spores with straight germ tubes within the same focal plane of the microscope that were closest to the scale in the ocular micrometer. The ocular micrometer was calibrated with a stage micrometer at 400x to assure accurate measurements. The micrometer was calibrated from 3 to 25 µm. The total length of the tube from the edge of the original spore membrane was measured. Water quality (temperature, pH, dissolved oxygen, and salinity) measurements were made on each treatment on Day 0 and at termination (composite of replicates). BTEX total concentration at termination was at least 80% of the initial concentration. The study was deemed acceptable. Mean control germination was > 70% in the controls. Mean germination tube length was > 10 um in the controls.

Reliability : (1) valid without restriction

(10) (23)

4.9 ADDITIONAL REMARKS

Memo : Effect of crude and refined oil on fresh and sea water organisms

Remark : Burks has extensively reviewed the effects of crude and refined oils on organisms found in fresh and sea water. He noted that where spillages occur the non-mobile species suffer the greatest mortality, whereas fish species can often escape from the affected region. The extent of the initial mortality depends on the chemical nature of the oil, the location, and the physical conditions, particularly the temperature and wind velocity. Most affected freshwater and marine communities recover from the effects of an oil spill within a year. The occurrence of biogenic hydrocarbons in the world's oceans is well recorded. They have the characteristic isoprenoid structure, and measurements made in water columns indicate a background concentration of 1.0 to 10 µl/l. The higher molecular weight materials are dispersed as particles, with the highest concentrations of about 20 µl/l occurring in the top 3 mm layer of water.
A wide variation in the response of organisms to oil exposures has been noted. The larvae of fish and crustaceans appear to be most susceptible to the water-soluble fraction of crude oil. Exposures of plankton and algae have indicated that certain species of diatoms and green algae are inhibited, whereas microflagellates are not.
For the most part, molluscs and most intertidal worm species appear to be tolerant of oil contamination.

These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '4', reliability not assigned

Source : CONCAWE Bruxelles

Reliability : (4) not assignable

(8)

Memo : Effect of crude oil on Lobster larvae

Remark : The effects of emulsified South Louisiana crude oil on the development of American Lobster, Homarus americanus, was investigated during the development of the first four larval stages over a 15-day period. Hatched larvae were exposed to concentrations of 0, 0.1 and 1.0 ppm crude oil, six times per day for periods ranging from 0.8 to 5.6 minutes, using a flow-through system. The tests showed that 1.0 ppm crude oil was a sub-lethal

concentration for the lobster larvae and that no significant effects were found at 0.1 ppm. At 0.1 ppm crude oil, the survival rate of the larvae was comparable with that of the controls, but at 1.0 ppm the survival value was about 50% that of the control larvae. Other effects observed at 1.0 crude oil included: (a) lethargy, reduced feeding and lack of the characteristic aggression of the control animals, (b) an increase in development time from 12 to 15 days, and (c) a change in the pigmentation of the larvae from the normal pale blue, almost transparent, color to a sharp red appearance.

These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '4', reliability not assigned

Source : CONCAWE Bruxelles
Reliability : (4) not assignable

(31)

Memo : Effect of crude oil on Sea urchin eggs

Remark : Falk-Petersen has studied the effects of the water-soluble fraction (WSF) of Ekofisk crude oil on the development of the eggs of two sea urchin species, Strongylocentrotus pallidus and S. droebachiensis. The WSF was prepared by shaking Ekofisk crude oil and sea water in the ratio 1:9 for 5 minutes, allowing to separate for 20 hours, and using the aqueous phase for the studies.
Fertilized eggs were exposed to dilutions of the WSF for up to 9 days at 3 to 5 °C. Embryos and larvae were examined regularly by scanning and transmission electron microscopy after fixing in 2% OsO_4 in sea water. Concentrations of 30% WSF, corresponding to 13 ppm dissolved oil, did not impair either development or the ultrastructure of the stages from egg to pluteus. However, significant effects on development were found at both 40% WSF (17 ppm oil) and 50% WSF (21 ppm oil). At these levels, the larvae filled with degenerating cells, and differentiation of the intestine and skeletal growth were inhibited compared with the control larvae.

These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '4', reliability not assigned

Source : CONCAWE Bruxelles
Reliability : (4) not assignable

(28)

Memo : Effect of crude oil on eggs

Remark : Crude oil from 4 locations was applied externally at different loadings to batches of 30 fertilized eggs of mallard ducks on day 3 of incubation at 37.5 °C. Eggs were candled each day to determine mortality, and dead embryos examined for abnormalities. On day 18 of incubation, surviving embryos were examined for external malformations. The observed LD50 values for crude oils from Kuwait, Prudhoe Bay, South Louisiana and Texas were 2.2, 8.3, 1.3 and 5.5 µl/egg, respectively. The South Louisiana and Texas crudes both produced significantly reduced growth, both above and below the LD_{50} value; preformed embryos were also found with both these crudes.

These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '4',

reliability not assigned

Reliability : (4) not assignable

(37)

Memo : Poorly water soluble mixtures

Remark : For the assessment of the ecotoxicity of poorly water soluble mixtures of hydrocarbons as found in petroleum products, it is now generally accepted that results should be expressed in terms of the "loading rate". The "loading rate" may be defined as the amount of the product which must be equilibrated with the aqueous test medium in order to produce a specified level of effect. Studies in which the results are expressed in terms of the measured concentrations of hydrocarbons in dilutions of "water soluble fractions (WSF)" do not allow the ecotoxicity of a product to be expressed in terms of the amount of that product required to produce a particular effect and, therefore, such results are not comparable to results obtained with other substances.

These data are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9. These data are ranked by API crude oil task group as '1', reliable without restrictions.

Source : CONCAWE Bruxelles

Reliability : (1) valid without restriction

(13) (35) (79)

5.1.1 ACUTE ORAL TOXICITY

Type	:	LD_{50}
Value	:	> 5000 mg/kg bw
Species	:	rat
Strain	:	Sprague-Dawley
Sex	:	male/female
Number of animals	:	5
Vehicle	:	None, undiluted
Doses	:	Single dose: 5000 mg/kg
Year	:	1984
GLP	:	no data
Test substance	:	Beryl crude (Light crude oil)

Method : Groups of five male and five female Sprague-Dawley rats were dosed once by oral gavage with the test material at a dose level of 5 g/kg. The animals were observed frequently on the day of treatment and daily thereafter for 14 days. The animals were weighed on the day of treatment and again 7 and 14 days later.

Result : There were no deaths following treatment.
During the first week post dosing lacrimation and a discharge covering the perineum (genital area) were observed.
At termination of the study all animals appeared healthy and had gained weight.
The oral LD_{50} was judged to be greater than 5 g/kg.

Reliability : (2) valid with restrictions
Not clear whether this study was carried out to GLP and few experimental data given. Nevertheless, the study is sufficient to demonstrate an LD_{50} of greater than 5 g/kg.

(55)

Type : LD_{50}

Result : The results of other acute toxicity studies which have been reported are as follows:

Sample	LD_{50}	Clinical signs
Lost Hills Light (Mobil consolidated report)		
	>5 g/kg	Soft stool
		Urogenital discharge
		Decreased fecal output
		Anal discharge
MCSL Crude (midcontinent) Mobil study No. 40971		
	>5 g/kg	Decreased activity
		Hunching
		Discharge/perineal staining
Arab Light (Mobil study No. 40961)		
	>5 g/kg	Discharge/perineal staining
Belridge heavy (Mobil summarized data)		
	>5 g/kg	Mild gastrointestinal effects

Smith et al (1981) also demonstrated that three different crude oils (Crude

type not specified) were non-toxic in male mice as follows:

Wilmington Crude oil	>16 g/kg
Recluse crude oil	>16 g/kg
Mixed petroleum crudes	>10 g/kg

(54) (56) (63) (73) (76)

5.1.3 ACUTE DERMAL TOXICITY

Type	:	LD_{50}
Value	:	> 2000 mg/kg bw
Species	:	rabbit
Strain	:	New Zealand white
Sex	:	male/female
Number of animals	:	3
Vehicle	:	None, undiluted
Doses	:	Single dose: 2000 g/kg
Year	:	1984
GLP	:	no data
Test substance	:	Beryl Crude (Light crude oil)
Method	:	The skin was clipped from the trunks of three male and three female New Zealand White rabbits prior to treatment with test material. The skin of the backs of three animals (2 male, 1 female) was abraded and the skin of the remaining animals was left intact. The liquid, undiluted test material was then applied as a single dose of 2 g/kg to the shorn back of all six animals. The test site was covered with a gauze and an occlusive wrap. The animals were also fitted with Elizabethan collars to prevent chewing of the occlusive covering and ingestion of test material. 24 hours after application of the test material the occlusive dressing was removed and any surplus test material was removed from the skin by gently wiping with cotton moistened with physiological saline. The animals were observed frequently on the day of treatment and daily thereafter for 14 days. Two hours after wiping the residual material from the skin (26 hours after treatment), the areas of application were assessed for skin irritation. Further evaluations for skin irritation were also made on the 3rd and 7th days following treatment.
Result	:	There were no deaths in this study. A few animals had soft stool/diarrhea during the observation period, however, all animals were normal at study termination. At the end of the study five of the six animals had gained weight and one had lost a small amount of weight. [No actual body weight data are provided in the report]. There was no evidence of systemic toxicity during the study although there was some skin reaction at the site of application. Following 24 hours of skin contact, varying degrees (slight, moderate) irritation were observed at the 26 and 72 hour readings. At the end of the first week, the skin response was barely perceptible or absent. The dermal LD_{50} was judged to be greater than 2 g/kg
Reliability	:	(2) valid with restrictions Not clear whether this study was carried out to GLP and few experimental data given.

(52)

Type : LD_{50}

Result : Additionally, acute dermal LD_{50}s have also been reported for three other light crude oils.
The results are as follows:

Sample	LD_{50}	Clinical signs
Lost Hills Light (Mobil Study No. 63831)		
	>2 g/kg	Soft stool
		Decreased food consumption
		Decreased fecal output
MCSL Crude (midcontinent) Mobil study No. 40972		
	>2 g/kg	Diarrhea
		Nasal discharge
Arab Light (Mobil study No. 40962)		
	>2 g/kg	No signs of toxicity
Belridge Heavy (Mobil summarized data)		
	>2 g/kg	Mild gastrointestinal effects

(51) (53) (63) (73)

5.2.1 SKIN IRRITATION

Species : rabbit
Concentration : undiluted
Exposure : Occlusive
Exposure time : 24 hour(s)
Number of animals : 6
Vehicle : Undiluted
Year : 1985
GLP : no data
Test substance : Lost Hills Light Crude

Method : Prior to test the hair was clipped from the backs of six New Zealand White rabbits (sex not specified). Three one inch square sites (anterior, mid-dorsal and posterior) on the right side of each animal were lightly abraded. Three similarly orientated sites were left intact on the left side of the animal, thus providing six test sites on each rabbit.
0.5 ml of test material was applied to each test site. The material applied to the anterior and mid-dorsal sites was covered with a Webril patch and these were occluded. The dorsal sites remained open. The rabbits were fitted with collars to prevent ingestion of the test material and chewing of the occluded patches.
Four hours after treatment, the anterior sites were unwrapped. The intact site was immediately examined for evidence of corrosive effects and both sites were then gently wiped with a gauze. Thirty minutes later, the skin was evaluated for irritation. The sites were evaluated again 24 hours after patch removal. 48 hours after application, the 4 hour occluded intact site was re assessed for corrosion. Both sites were scored for irritation 4 hours later. The 4 hour occluded sites were again re-evaluated for irritation on day 3.
24 hours after treatment, the two mid-dorsal sites were unwrapped and

along with the posterior uncovered sites were wiped with cotton. The four sites were evaluated for irritation 2 hours later, again approximately 72 hours later and finally on the 7th day after treatment.

Result : 4 Hour occluded sites (DOT, OECD methods)

Mean values (24, 48 & 72 hours) for erythema and edema at the intact sites were 1.69 and 1.3 respectively.
The initial response of the skin to the test material was slight, with little difference in response between intact or abraded sites.

Actual scores were:

	Intact		Abraded	
	Erythema	Edema	Erythema	Edema
4.5 hrs	1.0	1.2	1.0	1.3
28 hrs	1.2	1.0	1.0	1.0
52 hrs	1.7	1.3	1.7	1.3
76 hrs	1.8	1.5	1.8	1.7
7 days	0.8	0.7	0.5	0.7
10 days	0	0.3	0	0

24 Hour occluded sites (FHSA method)

Actual scores were:

	Intact		Abraded	
	Erythema	Edema	Erythema	Edema
26 hrs	1.7	1.7	1.7	1.7
72 hrs	2.3	1.5	2.2	1.7
7 days	1.2	0.7	0.8	0.8
10 days	0.2	0	0.2	0.2

Non-occluded sites

Actual scores were:

	Intact		Abraded	
	Erythema	Edema	Erythema	Edema
26 hrs	1.7	1.2	1.8	1.3
72 hrs	2.8	1.5	2.7	1.3
7 days	1.3	0.8	1.3	0.8
10 days	0.3	0	0.2	0

Conclusion : The classification according to various methods/criteria are summarized as follows:

Guideline	Score	Rating
DOT Corrosion	Negative	Non-corrosive
EEC (4hr occluded)	Erythema 1.6 Edema 1.3	Non-irritant
OSHA PII (4 h occl.)	2.8	Non-irritant
FHSA PII (24 h occl.)	3.6	Non-irritant

Reliability : (1) valid without restriction

(63)

Species	:	rabbit
Concentration	:	undiluted
Exposure	:	Occlusive
Exposure time	:	24 hour(s)
Number of animals	:	6
Vehicle	:	Non, undiluted
Year	:	1984
GLP	:	no data
Test substance	:	Arab Light crude

Method :

Prior to test the hair was clipped from the backs of six New Zealand White rabbits (sex not specified). Three one inch square sites (anterior, mid-dorsal and posterior) on the right side of each animal were lightly abraded. Three similarly orientated sites were left intact on the left side of the animal, thus providing six test sites on each rabbit.
0.5 ml of test material was applied to each test site. The material applied to the anterior and mid-dorsal sites was covered with a Webril patch and these were occluded. The dorsal sites remained open. The rabbits were fitted with collars to prevent ingestion of the test material and chewing of the occluded patches.
Four hours after treatment, the anterior sites were unwrapped. The intact site was immediately examined for evidence of corrosive effects and both sites were then gently wiped with a gauze. Thirty minutes later, the skin was evaluated for irritation. The sites were evaluated again 24 hours after patch removal. 48 hours after application, the 4 hour occluded intact site was re assessed for corrosion. Both sites were scored for irritation 4 hours later. The 4 hour occluded sites were again re-evaluated for irritation on day 3.
24 hours after treatment, the two mid-dorsal sites were unwrapped and along with the posterior uncovered sites were wiped with cotton. The four sites were evaluated for irritation 2 hours later, again approximately 72 hours later and finally on the 7th day after treatment.

Result :

4 Hour Occluded (OECD Method, DOT)

Mean values (24, 48 & 72 hours) for erythema and edema at the intact sites were 0.9 and 0.1 respectively.
The initial response of the skin to the test material was slight. Within 72 hours after treatment, most sites had recovered, with little difference in response between intact or abraded sites.

<u>24 Hour Occluded</u> (FHSA method)
The day following application of the test material a moderate erythema was observed. This subsided during the subsequent days such that there was only barely perceptible irritation at the end of the first week. Abrasion had no effect on either the initial response or the recovery.

<u>Non Occluded</u>
Uncovered treated sites had moderate erythema 24 hours after application of the test material. This response diminished and was virtually absent after one week.
Abrasion had no effect on either the initial response or the recovery.

Reliability :

(2) valid with restrictions
A textual description of the results only was available. No actual data were provided in the report.

(60)

Species : rabbit
Concentration : undiluted
Exposure : Occlusive
Exposure time : 4 hour(s)
PDII : 1.9
Result : not irritating
Classification : not irritating
Year : 1997
Test substance : Belridge heavy crude

Result : Summarized data for Belridge Heavy crude oil are:

Guideline	Score	Rating
DOT Corrosion	Negative	Non-corrosive
EEC (4hr occluded)	Erythema 0.6 Edema 0.8	Non-irritant
OSHA PII (4 h occl.)	1.4	Non-irritant
FHSA PII (24 h occl.)	2.1	Non-irritant

Reliability : (2) valid with restrictions
Data only available in tabular form, but studies were part of a series of studies using same methods, all of which were reliability 1.

(73)

5.2.2 EYE IRRITATION

Species : rabbit
Concentration : undiluted
Dose : .1 ml
Comment : not rinsed
Number of animals : 6
Year : 1985
Test substance : Beryl Crude (Light crude oil)

Method : 0.1 ml of undiluted test material was instilled into one eye of each of six New Zealand White rabbits. The eyes were not washed and were evaluated for irritation at 1, 24, 48 and 72 hours after treatment. Fluorescein was used to aid the evaluation of the 72 hour reading. Treated eyes that stained positively with fluorescein were examined again 7 and 14 days after treatment.

Result : The mean irritation scores at each of the observation times were:

	1 hr	24 hr	48 hr	72 hr
Cornea	0	0	0	0
Iris	0	0	0	0
Conjunctivae	4.0	1.7	1.3	1.0

The test material was judged to be non-irritating to the rabbit eye.

Reliability : (2) valid with restrictions
The only reservation is that it is not clear whether this study was carried out according to GLP. Otherwise the study and reporting are sound.

(58)

Species : rabbit

Result : Eye irritation studies have been reported for three other samples of light crude oil.
There were no effects on either the iris or cornea. The only effects recorded were on the conjunctivae.
The mean irritation scores (conjunctivae only) are as follows:

Crude type	Result 1 hr	24 hr	48 hr	72 hr	Ref
MCSL (Midcontinent)	2.3	0.3	0	0.7	Mobil 40973
Lost Hills Light	8.0	3.7	2.7	1.7	Mobil 63832
Arab Light	5.3	1.3	0.7	0.3	Mobil 40963
Belridge Heavy	0.9	0.8	0.9	Mobil 1997 (57) (59) (63) (73)	

5.3 SENSITIZATION

Type : Buehler Test
Species : guinea pig
Concentration : 1st: Induction 15 % occlusive epicutaneous
2nd: Challenge 15 % occlusive epicutaneous
3rd: Challenge 10 % occlusive epicutaneous
Number of animals : 20
Vehicle : Mineral oil
Result : not sensitizing
Year : 1991
GLP : yes
Test substance : Lost Hills Light Crude

Method : Concentrations of test material used in this sensitization study were determined in a pre screening study.

Induction phase
0.4 ml of a 15% w/w concentration of test material in mineral oil was applied using a Hill Top Chamber with a 25 mm Webril swatch to the shorn backs of 10 male and 10 female Guinea pigs. The patch was occluded for six hours. The patches were applied to the same site once per week for 3 weeks. After each 6-hour exposure period, the patches were removed and the skin wiped with gauze moistened with mineral oil. The positive control material (DNCB) was applied at a concentration of 0.05% w/w in 70% ethanol to 5 male and 5 female Guinea pigs. Treatment of the positive control animals was the same as for the test animals, except that the skin was wiped with saline after patch removal.

Challenge
Challenge patch application was performed 14 days after the last induction dose had been applied. Dual challenge patches were applied to fresh application sites of previously shorn skin of the animals. The test material

was applied at concentrations of 10 and 15% w/w in mineral oil. The patches were then occluded for 6 hours.
On the day following challenge patch application, the skin was depilated and 2 hours later and scored for signs of sensitization. The sites were examined after a further 48 hours but this time without depilation.
Naive and positive control animals were challenged with DNCB at a concentration of 0.05% in acetone.

Result : The results were as follows:

	No. responding	**Mean erythema score**
Test material (10% concentration)		
24 hrs induced	3/19	0.2
24 hrs control	0/10	0
48 hrs induced	3/19	0.3
48 hrs control	2/10	0.2
Test material (15% concentration)		
24 hrs induced	2/19	0.2
24 hrs control	2/10	0.2
48 hrs induced	1/19	0.1
48 hrs control	1/10	0.1
DNCB positive control		
24 hrs induced	5/5	3.4
24 hrs control	0/5	0
48 hrs induced	5/5	2.6
48 hrs control	0/5	0

Although two test animals responded, the test material was judged to be non-sensitizing, since the test criterion required an mean erythema score of +2 for a positive response.

Reliability : (1) valid without restriction

(66)

Type : Buehler Test
Concentration : 1st: Induction 15 %
2nd: Challenge 10 %
3rd: Challenge 15 %
Result : not sensitizing
Year : 1991
Test substance : Belridge heavy crude

Result : A similar Buehler test of Belridge heavy crude oil was also negative.
Reliability : (2) valid with restrictions

(65)

5.4 REPEATED DOSE TOXICITY

Type	:	Sub-chronic
Species	:	rat
Sex	:	male/female
Strain	:	Sprague-Dawley
Route of admin.	:	dermal
Frequency of treatm.	:	5 days/week for 13 weeks
Doses	:	30, 125 and 500 mg/kg/day
Control group	:	yes, concurrent no treatment
Year	:	1992
GLP	:	no data
Test substance	:	Two crude oils

Method :

Two separate but identical studies are reported, one for each of two crude oils (Crude I and Crude II). The methods used were identical for both studies.
Undiluted test material was applied to the shorn unoccluded skin of groups of ten male and ten female Sprague-Dawley rats at doses of 30, 125 and 500 mg/kg/day. Application was once daily, five times each week for 13 weeks. Groups of ten rats of each sex served as untreated controls. Each animal was fitted with an Elizabethan collar to prevent ingestion of the applied test material. At the end of each week residual test material was wiped from the backs of the animals. Animals were observed regularly for clinical signs of toxicity and body weights were recorded weekly. The animals were sacrificed during week 14 of the study after fasting overnight. All animals were necropsied and blood samples were taken for a range of hematological and serum chemistry determinations. A range of organs were weighed and tissues examined histologically.
An additional two groups of ten males were treated at a dose level of 0 and 500 mg/kg/day for an evaluation of male reproductive health.
For these animals testes weight and cauda epididymis weights were recorded. Additionally, the number of sperm and % normal sperm in the cauda were recorded as well as the number of spermatids in the testis.

Result :

No animals died in either of the studies and there were no clinical signs of systemic toxicity attributable to either crude oil. Both crude oils caused the same minimal skin irritation (flaking) at the exposure site.

<u>Body weights</u>
Apart from the 500 mg/kg/day group for Crude II that gained less weight than the controls (217g compared to 247g over the course of the study). All other body weight measurements were similar to the respective controls.

Hematology
Significant changes relative to their respective controls only occurred in the highest dose group animals (500 mg/kg/day). The % increases (+) or decreases (-) that occurred in these dose groups are shown in the following table. [Changes are shown to the nearest whole number]

Parameter	Crude I		Crude II	
	Males	Females	Males	Females
RBC	-6%	NSD	-8%	-7%
Hemoglobin	-6%	NSD	-11%	-8%
Hematocrit	-5%	NSD	-9%	-8%
Platelets	NSD	NSD	-19%	NSD

NSD No significant difference

Serum chemistry
Although more parameters were affected in the high dose Crude II females, there was no consistent pattern of change that could be attributed to the two crude oils. The differences %+ or %- are shown in the table below.

	Dose group (mg/kg/day)					
	Crude I			Crude II		
	30	125	500	30	125	500
MALES						
Calcium			-7%			
Glucose		+13%	+13%			
Urea nitrogen			+53%			
Uric acid						-27%
FEMALES						
ALT						-18%
Cholesterol					+41%	+60%
Glucose	+16%		+20%			
Potassium			-11%			-13%
Urea nitrogen						+31%
Uric acid						-27%

Necropsy findings
There were no treatment-related observations at necropsy. With the exception of the liver and thymus, there were no absolute or relative organ weight changes when compared to controls.
The organ weight differences that were observed were confined to the 125 and 500 mg/kg/day groups and are summarized below.

Organ wt		Crude I		Crude II	
		125	500	125	500
MALES					
Liver	(absolute)		+20%		+22%
	(relative)		+22%	+18%	+33%
Thymus	(absolute)				-41%
FEMALES					
Liver	(absolute)		+13%		+24%
	(relative)		+12%		+31%
Thymus	(absolute)				-35%
	(relative)				-27%

Histopathology
Changes in the skin included hyperplasia and an associated dermal inflammatory cell infiltration. In general the effects were slightly more severe in the animals treated with Crude I. All male and female animals exposed to Crude I were affected whereas at the lowest dose level 8/10 male and 6/10 females exposed to Crude II were affected.
Other histopathological findings were associated with the bone marrow, Liver, thymus and thyroid although effects generally were greater with crude II than with crude I. The findings are tabulated below

Crude I

Bone marrow	No effects in either sex at any dose level
Liver	Multifocal, mononuclear cell infiltration in 3 males and 2 females at 500 mg/kg/day. Multifocal hepatocellular vacuolation in 3 females at 500 mg/kg/day only
Thymus	Atrophy in one male and two females at 500mg/kg/day only
Thyroid	Hypertrophy and hyperplasia of follicular epithelium in some males and females at all dose levels

Crude II

	Bone marrow Increased cellularity in 2 males at each dose level (6 males in highest dose group) and focal necrosis in 2 males in the 50 mg/kg/day group. In the females increased cellularity was observed in 9/10 animals
Liver	Hepatocellular vacuolation in one male and one female in the 500 mg/kg/day group. Mononuclear cell infiltration was also observed but in only one male in the highest dose group
Thymus	Atrophy was observed in six males and 7 females in the 500 mg/g/day group.
Thyroid	Hypertrophy and hyperplasia was seen in a few animals at all dose levels.

The spermatozoa/spermatid evaluations that were conducted on the separate 500 mg/kg/day group of animals exposed to crude I did not reveal any effects when compared to a control.

Test substance : Two crude oils were examined in the paper by Feuston et al
The characteristics of the crude oils was reported as:

Chemical class	**Crude I**	**Crude II**
Nonaromatics	50.0	37.3
<3-Ring PAH	35.3	41.7
3- to 5- Ring PAH	10.2	15.7
3- Ring PAH	5.9	8.1
4- Ring PAH	2.2	4.0
5- Ring PAH	2.1	3.6
Sulfur-PAC	2.4	2.9

Nitrogen (N)-PAC	5.4	8.4
Nonbasic N-PAC	3.8	5.9
Total	103.3	106.0

From information available in the Mobil report of study No 63834 and other laboratory data, it is clear that Crude I is Lost Hills Light Crude and Crude II is Belridge heavy crude oil.

(30) (72)

Type : Sub-acute
Species : mouse
Sex : male/female
Strain : CD-1
Route of admin. : gavage
Exposure period : 5 days
Frequency of treatm. : Daily for five days
Doses : 2, 4, 8, 12 & 16 ml/kg/day
Control group : yes, concurrent no treatment
Year : 1990
GLP : no data
Test substance : Prudhoe Bay Crude oil (Heavy crude oil)

Method : Two pilot studies were conducted with groups of 5 male and 5 female mice. In these studies Prudhoe Bay crude oil was administered by gavage at doses of 0, 5 and 10 ml/kg daily for five days. No other experimental details are given. However, the results led the author to conduct four further follow-up studies and these are described below.

Experiment 1 (Three crudes, each at a single dose level)
One of three crude oils or Bunker C oil was administered by gavage, daily at a single dose level of 10 ml/kg for 5 days to groups of 10 male mice. A further group of 10 male mice were intubated each day but no material was administered; these animals served as controls.

Experiment 2 (One crude, single dose level)
Groups of 10 male mice were given either Prudhoe Bay crude oil or mineral oil USP or corn oil at a dose of 10 ml/kg/day for five days. A separate group of 10 controls were intubated but no material was administered. This study was to compare the effect of one crude oil with the effects of two oils expected to be non-toxic.

Experiment 3 (One crude, five dose levels)
Prudhoe bay crude oil was administered to 10 or 11 male mice at dose levels of 2, 4, 8, 12 or 16 ml/kg/day. A further group of 10 male mice were sham treated and served as controls.

A fourth experiment was also carried out which studied the effect of crude oil exposure on vitamin E and selenium-deficient animals. This study is not summarized here.

In all experiments 1-3 the animals were weighed on the day they were first dosed and again prior to necropsy. The differences in weights were recorded as change in body weight during the studies. Necropsy was 24 hours after the 5th dose had been administered.
At necropsy, blood samples were taken for determination of blood cell counts, packed cell volume (PCV), hemoglobin and red cell indices

(MCHC). Blood smears were stained with methylene blue to detect the presence of Heinz bodies in red cells. Liver was removed and weighed whereas the spleen and thymus were weighed after fixation. Portions of all major organs were fixed for subsequent histological examination.

Data were analyzed by analysis of variance and regression procedures of the SAS programs (SAS Institute Inc., Cary, NC)

Result **:** Although several studies were reported, the only studies summarized here are study A in which several crude oils are tested at a single dose level and study C in which PBCO is examined at five different dose levels. Neither information on oils other than crude oil or on the effect of Vitamin E and selenium-depletion are included in this robust summary.

In the pilot studies it was reported that the oils were distasteful to the mice and that after the first dose, administration of test material was difficult. Several mice from each group died from inhalation of oil that occurred during the oral dosing. The author stated that the pilot studies demonstrated a dose related decrease (8-11%) in PCV, a reduction in body weight gain, a 74% increase in liver weight per unit body weight and a 66% reduction in thymus weight. No data are given but this was the reason given for the further studies that were carried out.

Although the studies carried out were separate, the author reported the results together and these are summarized below.

Hematology

It was reported (but no data presented) that in experiment A none of the oils tested resulted in significant changes in PCV, number of red blood cells or whole blood hemoglobin. The following table summarizes the hematological results obtained with PBCO only.

Study	No of mice	Daily dose PBCO (ml/kg/day)	PCV	MCHC (g/l)
A	10	0	0.43±0.02	336±22
	8	10	0.42±0.04	328±10
C	11	0	0.43±0.02	328±11
	8	12	0.40±0.03	348±3*

* p<0.05, analysis of variance with Tukey's multiple range procedure

Body and organ weights

The organ and body weight changes are summarized in the following tables

Study A comparison of three different crude oils

	Treatment group		
Control	PBCO	SLCO	ALCO
Group size			
10	8	6	7
Change in body wt (g)			
+1.74±0.88	-0.33±1.5	-0.77±2.36*	-1.43±2.28*
Liver wt (g)			
2.15±0.2	3.03±0.54*	2.73±0.5	2.66±0.43*

Liver/body wt (g/g)			
0.07±0	0.11±0.01*	0.10±0.01*	0.09±0.01*
Spleen/body wt (g/g)			
3.52±0.61	2.46±0.24*	2.49±0.29*	2.17±0.66*
Thymus/body wt (g/g)			
2.26±0.46	0.85±0.33*	1.01±0.41*	1.12±0.46*

* P<0.05 Analysis of variance with Tukey's multiple range procedure

Study C: PBCO at 5 dose levels

Body wt change (g)	Liver wt (g)	Organ/body wt (g/g) liver	spleen	thymus
Control group (10 mice)				
+3.14±1.33	1.95±.18	.06±.01	3.9±.49	2.7±.29
2 ml/kg/day (10 mice)				
-0.88±2.57	2.32±.22	.09±0	2.65±.62	1.61±.49*
4 ml/kg/day (9 mice)				
-0.86±1.24	2.38±.33*	.09±.01*	2.57±.23*	1.35±.32*
8 ml/kg/day (8 mice)				
-1.66±2.03*	2.58±.48*	.1±.01*	2.31±.67*	1.03±.4*
12 ml/kg/day (9 mice)				
-1.66±1.71*	2.69±.27*	.1±.01*	2.2±.44*	.71±.22*
16 ml/kg/day (7 mice)				
-3.29±1.4*	2.15±.35	.1±.01*	2.26±.87*	.65±.15*
Coefficient of determination (R^2)				
0.49***	0.08**	0.48***	0.31***	0.64***

* P<0.05 Analysis of variance with Tukey's multiple range procedure
** P<0.03 Analysis of variance in regression
*** P<0.001 Analysis of variance in regression

Morphology

There were no gross abnormalities at necropsy except a reduction in the size of thymus glands in those animals that had received crude oil. Liver, kidney, spleen, lung and thymus from groups of 5 mice that had received 0, 5 or 10 ml PBCO for 5 days in the pilot study were examined histologically. The thymus glands of mice in the 10 ml/kg/day group had very thin cortices and reduced densities of lymphocytes in the remaining cortex compared to controls. Hepatocytes from these same animals had uniformly dense cytoplasm while control mouse hepatocytes had large areas of rarified cytoplasm typical of normal glycogen-replete mouse livers. The histology of the thymus glands of the 5 ml/kg/day group was intermediate between the controls and the 10 ml/kg/day group.
There were no histological lesions in the other tissues examined.
Overall, the results demonstrated minor hematological change, liver enlargement and atrophy of spleen and thymus.

Test substance : The following crude oils were examined:

Prudhoe Bay crude oil (PBCO) [a heavy crude]
South Louisiana crude oil (SLCO) [a light crude]
Arabian Light crude oil (ALCO)
In addition a bunker C oil was examined
A USP grade mineral oil and a corn oil were also included in the studies.

Reliability : (2) valid with restrictions
These studies were probably not carried out according to GLP and the studies did not include measurement of all the parameters normally measured in repeat dose studies. Although the results are of limited value they do nevertheless add some information on the effect of the repeated exposure of mice to crude oil.

(41)

5.5 GENETIC TOXICITY 'IN VITRO'

Type : Modified Ames Assay
System of testing : Ames test of a DMSO extract of test substance
Test concentration : 1 to 50µl/plate
Metabolic activation : with
Year : 1984
GLP : no data
Test substance : Beryl Crude (LIght crude oil)

Method : DMSO extracts were prepared by mixing 2 ml test material with 3 ml cyclohexane to homogeneity. 10 ml DMSO was then added and mixed thoroughly. The mixture was vortexed every 5 minutes for a total of 30 minutes. After 30 minutes the mixture was centrifuged and the DMSO layer (extract) removed and stored for testing.

The extract was only tested with metabolic activation in Salmonella typhimurium strain TA 98 at the following doses: 1, 3, 5, 7, 10, 15, 25 and 50 µl/plate. Additionally the DMSO extract of a carcinogenic oil was also tested in the same manner.
Positive control chemicals were: 2-aminoanthracene (2 µg)and benzoa(a)pyrene (5 µg).
The DMSO extract of a refrigerator oil was used as negative control (50 µl).
The metabolic activation mixture was derived from Araclor-induced hamster liver and this was use at eight times the standard concentration (0.4 ml rather than 0.05 ml).

The test material and control substances were added to tubes at the doses shown above and were incubated for 20 minutes with Salmonella broth culture. Colonies of histidine prototrophs were counted 48 hours after plate incubation.

Assay acceptance criteria

A linear dose response curve for mutagenicity must be obtained before the test material can be ranked for potency

Spontaneous and solvent control reversion rates for TA98 must fall between 20 and 50 revertants /plate

The slope of the dose response curve for the positive control carcinogenic oil must fall between 1.5 - 4.5 netrevertants/plate

Provided the above criteria are met the net revertants per plate is plotted

against dose and the best initial straight line response is determined by regression analysis. Test materials producing slopes less than 0.5 net revertants/µl are considered inactive.

Result : The No. revertants for each dose level of Beryl crude are as follows (NB data for the carcinogenic oil are not shown):

Dose (µl)	No. of revertants
0 (Spontaneous)	43
0 (solvent control)	39
50	113
25	85
15	74
10	67
7	61
5	57
3	47
1	48
2-AA control	540
BaP control	366
Refrigerator oil	43

The slope for the Beryl crude was determined to be 2.5
The slope for the carcinogenic oil was 3.7

Remark : Similar studies were conducted for Arab light, MCSL, Lost Hills light and Belridge Heavy crudes.
The results were:

Crude	Slope	Reference
Arab light	3.8	Mobil study 40965
MCSL crude	1.5	Mobil study 40975
Belridge Heavy	1.7	Mobil study 663850
Lost Hills light	0	Mobil study 63838

Conclusion : A modified Ames assay conducted on five different crudes demonstrated that all but one (Lost Hills light) were mutagenic. The Lost Hills Light did not demonstrate any mutagenic activity.

Reliability : (2) valid with restrictions
It is not clear whether the studies were carried out according to GLP. Furthermore, the assay was designed as a screen for carcinogenic activity However, the results are useful to identify the mutagenic potential of crude oils.

(48) (49) (50) (61) (62)

Type : Cytogenetic assay
System of testing : Chinese Hamster Ovary cells
Test concentration : 1 to 20 µl DMSO extract/ml culture medium
Metabolic activation : with
Result : negative
Year : 1991
GLP : no data
Test substance : Lost Hills Light crude

Method : Preparation of DMSO extract
10 ml DMSO was added to approximately 2 ml test substance.
The tube containing the mixture was vortexed every 5 minutes for one minute. After 30 minutes, the mixture was centrifuged and the DMSO layer

(Extract) was removed for testing.

Control substances:
Solvent control: DMSO
Negative control: Untreated flasks
Positive controls: Cyclophosphamide monohydrate at concentration of 10 µg/ml

Duplicate cultures were used - one set for metaphase analysis and the other for a determination of cytotoxicity (Mitotic Index). All cultures were conducted with metabolic activation.

CHO cells were exposed for two hours to test material at concentrations of 1, 2.5, 5, 10, 15 and 20 µl/ml. After two hours, treatment was terminated and the cultured cells were washed and re-fed with complete culture medium. Approximately 16 hours later, colcicine was added and two hours later the cells were harvested as described below.

Metaphase analysis
Metaphase cells were collected by tapping a flask to release the loosely attached mitotic cells into culture medium and pelleting them by centrifugation. The cells were resuspended and then fixed in methanol:acetic acid (3:1) and stored refrigerated until slide preparation. A minimum of 4 slides was made for each culture flask. 100 cells were examined microscopically per flask and were scored for structural chromosomal aberrations. Although gaps were recorded they were not used in the analysis for chromosomal aberration since their significance is questionable.

Cytotoxicity assay (Mitotic Index determination)
For this portion of the assay all cells in each flask were collected.
The medium in each flask was replaced and 5 ml of trypsin was added and after incubation for approximately 5 minutes the flasks were examined to ensure that the cells had rounded up. The cells were then released into the trypsin solution mechanically and the resulting suspension was centrifuged in the presence of medium. The centrifuged cells were swelled in hypotonic solution and were fixed in methanol:acetic acid as above. A minimum of 2 slides per flask were prepared for the determination of mitotic index. The slides were stained with Giemsa and at least 1000 nuclei per flask were scored as either mitotic or interphasic.

$$\text{The MI} = \frac{\text{No mitotic cells}}{\text{Total No of cells scored}} \times 100$$

Criteria for a positive or negative response
A test substance is considered to have elicited a positive response if at least one concentration shows a statistically significant increase in the proportion of cells with aberrations and a significant positive dose-response exists.
Biological significance of the result is also taken into account in determining the response.

Result :

Determination of Mitotic Index (MI)

There was not a dose related decrease in MI at treatments that were not toxic. The MI at each of the dose concentrations of Lost Hills Light crude (LHL)was:

Treatment	Mitotic Index
20 µl/ml extract of LHL	no cells found
15 µl/ml extract of LHL	no cells found
10 µl/ml extract of LHL	0.6
5 µl/ml extract of LHL	10.7
2.5 µl/ml extract of LHL	8.4
1 µl/ml extract of LHL	12.2
20 µl/ml DMSO	7.6
Negative control	10.4

Metaphase analysis

The concentrations analyzed for chromosomal aberrations were 5, 2.5 and 1.0 µg/ml.

There was no increase in the proportion of cells with structural chromosome aberrations (gaps excluded). Nor was a dose response observed. The data are:

Treatment	No of cells with one or more aberrations (100 cells examined)
5 µl/ml extract of LHL	2
2.5 µl/ml extract of LHL2	
1 µl/ml extract of LHL	1
DMSO (solvent control	2
Negative control	2
CP (positive control)	28

Reliability : (1) valid without restriction

Despite the fact that whether the study was carried out to GLP is not clear, it is, nevertheless, reliable and valid.

(70)

Type : Cytogenetic assay
System of testing : Chinese Hamster Ovary cells
Test concentration : 1 to 20 µl DMSO extract/ml culture medium
Metabolic activation : with
Result : negative
Year : 1991
GLP : no data
Test substance : Belridge Heavy crude

Method :

Preparation of DMSO extract

10 ml DMSO was added to approximately 2 ml test substance. The tube containing the mixture was vortexed every 5 minutes for one minute. After 30 minutes, the mixture was centrifuged and the DMSO layer (Extract) was removed for testing.

Control substances:
Solvent control: DMSO

Negative control: Untreated flasks
Positive controls: Cyclophosphamide monohydrate at concentration of 10 µg/ml

Duplicate cultures were used - one set for metaphase analysis and the other for a determination of cytotoxicity (Mitotic Index). All cultures were conducted with metabolic activation.

CHO cells were exposed for two hours to test material at concentrations of 1, 2.5, 5, 10, 15 and 20 µl/ml. After two hours, treatment was terminated and the cultured cells were washed and re-fed with complete culture medium. Approximately 16 hours later, colcicine was added and two hours later the cells were harvested as described below.

Metaphase analysis
Metaphase cells were collected by tapping a flask to release the loosely attached mitotic cells into culture medium and pelleting them by centrifugation. The cells were resuspended and then fixed in methanol:acetic acid (3:1) and stored refrigerated until slide preparation. A minimum of 4 slides was made for each culture flask. 100 cells were examined microscopically per flask and were scored for structural chromosomal aberrations. Although gaps were recorded they were not used in the analysis for chromosomal aberration since their significance is questionable.

Cytotoxicity assay (Mitotic Index determination)
For this portion of the assay all cells in each flask were collected. The medium in each flask was replaced and 5 ml of trypsin was added and after incubation for approximately 5 minutes the flasks were examined to ensure that the cells had rounded up. The cells were then released into the trypsin solution mechanically and the resulting suspension was centrifuged in the presence of medium. The centrifuged cells were swelled in hypotonic solution and were fixed in methanol:acetic acid as above. A minimum of 2 slides per flask were prepared for the determination of mitotic index. The slides were stained with Giemsa and at least 1000 nuclei per flask were scored as either mitotic or interphasic.

$$\text{The MI} = \frac{\text{No mitotic cells}}{\text{Total No of cells scored}} \times 100$$

Criteria for a positive or negative response
A test substance is considered to have elicited a positive response if at least one concentration shows a statistically significant increase in the proportion of cells with aberrations and a significant positive dose-response exists. Biological significance of the result is also taken into account in determining the result.

Result : Determination of Mitotic Index (MI)
There was not a dose related decrease in MI at treatments that were not toxic. The MI at each of the dose concentrations of Belridge Heavy crude (BH)was:

Treatment	**Mitotic Index**
20 µl/ml extract of BH	no cells found
15 µl/ml extract of BH	no cells found
10 µl/ml extract of BH	no cells found
5 µl/ml extract of BH	11.2
2.5 µl/ml extract of BH	10.2

1 µl/ml extract of BH	13.1
20 µl/ml DMSO	7.6
Negative control	10.4

Metaphase analysis
The concentrations analyzed for chromosomal aberrations were 5, 2.5 and 1.0 µg/ml. There was no increase in the proportion of cells with structural chromosome aberrations (gaps excluded). Nor was a dose response observed. The data are:

Treatment	**No of cells with one or more aberrations (100 cells examined)**
5 µl/ml extract of LHL	1
2.5 µl/ml extract of LHL	2
1 µl/ml extract of LHL	1
DMSO (solvent control	2
Negative control	2
CP (positive control)	28

Reliability : (1) valid without restriction
Despite the fact that whether the study was carried out to GLP is not clear, it is, nevertheless, reliable and valid.

(69)

Type : Ames test
Metabolic activation : with and without
Year : 1981
GLP : no data
Test substance : South Louisiana crude (Light crude oil)

Method : The authors summarized the protocol in the following table:

Test strains
Salmonella typhimurium strains TA-1535, TA-1537, TA-1538, TA-98 and TA-100

Liver homogenate
S-9 from male Charles River CD rat liver 500 mg Aroclor/kg for 5 days

Procedure
Treatment without activation: 2 ml top agar, 0.1 ml dissolved test chemical, 0.1 ml bacterial culture (10^8 cells)
Treatment with activation: 2 ml top agar, 0.1 ml dissolved test chemical, 0.1 ml bacterial culture (10^8 cells) plus 0.5 ml S-9 mix contains per milliliter 0.3 ml S-9 (1 g tissue + 3 ml 0.15M KCl), 8 mM Mg Cl_2, 33 mM KCl, 5mM glucose-6-phosphate, 4 mM NADP, and 100 mM sodium phosphate (pH 7.4).
Incubation period: 48 hr

Design: Preliminary toxicity determination
Duplicate plates/point
Five test concentrations and solvent and positive controls
Duplicate experiments

Solvent Dimethyl sulfoxide

Data analysis
Mutagenic: statistically significant. Increase in total revertant colony number (P= 0.01) and dose response (P= 0.01)

Scoring

	Revertant/µg	Induced frequency/ Spontaneous frequency
(-)	<0.01	-*
(+)	0.1 to 0.01	<5
(++)	>0.1	>5

* No greater than response

Result : The results are given in the following table

Sample	Mutagenic activity (with activation)			
	TA-1537	TA-1538	TA-98	TA-100
Benzo[a]pyrene	++	++	++	++
South Louisiana crude	-	-	-	+

It is not clear from the report whether any activity was detected in the assay without metabolic activation.

Reliability : (4) not assignable
The study is not fully reported (data for the assay without metabolic activation are not given)

(9)

Type : Ames test
Metabolic activation : with and without
Result : negative
Year : 1982
GLP : no data
Test substance : Wilmington crude

Method : Salmonella typhimurium strains TA 98 and TA 100 were used in this study. The test sample was suspended in Tween 80 prior to testing. Each sample was weighed in a glass vial and then an equal weight of Tween 80 was added. The oil and detergent were stirred until thoroughly homogenized. Distilled water was then added dropwise with continuous stirring until a stable, homogenous suspension was obtained. The concentration of the mixture was then adjusted to 10% sample - 10% Tween 80 (w/v) by the addition of more distilled water.
The assay was carried out in triplicate with and without metabolic activation by S9 liver homogenate that was prepared from Aroclor-induced Sprague-Dawley rats. The S9 homogenate contained 13 mg of protein/ml and was stored at -70°C. S9 homogenate was used at a concentration of 10% (v/v) in the assay.
BaP was used as the positive control.
Negative control plates for determination of the spontaneous reversion rate were treated with 400µl or less of 10% Tween 80, a non-toxic and non-mutagenic dose.

No other experimental details are given in the paper, but reference is made to the papers by Ames that describe the methodology.

Result : Addition of up to 400µl Tween 80 per plate did not alter the spontaneous

reversion rates of either TA 98 or TA 100. Tween 80 had no effect on the mutagenicity of BaP, which induced similar numbers of revertants in TA 98 with S9 activation whether dissolved in DMSO or Tween 80.
The crude oil sample was inactive in all bacterial mutagenicity tests.

Reliability : (4) not assignable
The numbers of revertants were not given. The result was simply stated by the authors.

(43)

Type : Sister chromatid exchange assay
System of testing : Human lymphocytes
Year : 1982
GLP : no data
Test substance : Wilmington crude oil

Method : Heparinized blood was obtained from a healthy 25-year old male. [The method for culture of the lymphocytes was not described but reference was given which describes the method].
Benzo(a)pyrene (BaP) and N-methyl-N'-nitro-N-nitroso guanidine (MNNG) were used as positive control substances.

The test sample was suspended in Tween 80 prior to testing.
Each sample was weighed in a glass vial and then an equal weight of Tween 80 was added. The oil and detergent were stirred until thoroughly homogenized. Distilled water was then added dropwise with continuous stirring until a stable, homogenous suspension was obtained. The concentration of the mixture was then adjusted to 1% sample - 1% Tween 80 (w/v) by the addition of more distilled water. Lymphocyte cultures were treated 18 hours after initiation by
(a) continuous exposure to test chemical without exogenous activation or
(b) by exposure for 2.5 hours with activation by S9.
[procedure described elsewhere by Stetka and Wolff (1976) for metabolic activation].
In tests with S9, cells were collected by centrifugation at 150 x g for 10 minutes, resuspended in 5 ml of McCoy's 5A medium containing only antibiotics and treated with microliter quantities of test material or solvent. An aliquot of 0.25 ml of S9 mix containing 10% S9 (v/v) was added to each culture. Cells were incubated for 2.5 hours with occasional mixing. The cells were collected, washed once in medium with 5% fetal bovine serum and resuspended in 8 ml of culture medium containing BrdUrd for further incubation. In experiments testing MNNG, lymphocytes that had been cultured for 18 hours were suspended in medium without serum, S9 or BrdUrd, treated for 2 hours with MNNG in acetone or acetone alone (controls), washed as above, resuspended in complete medium with BrdUrd and incubated further. At 68 hours after the cultures were initiated, Colcemid (0.01µg/ml) was added; 4 hours later, the cells were harvested by centrifugation, subjected to hypotonic shock for 7 minutes in 0.075M KCl, fixed in 3 changes of cold methanol-acetic acid (3:1) and spread on chilled, wet microslides that had been pre-cleaned in 95% ethanol. The preparations were stained by the Hoechst 33258-black light-Giemsa method and numbers of SCE were counted in 25 metaphase cells containing 40-48 chromosomes

Result : Tween 80 was not cytotoxic and did not induce increases in the numbers of SCE in cultures of lymphocytes treated with 100 ppm Tween 80 for 54 hours without activation or for 2.5 hours with S9 activation.
MNNG, a strong alkylating agent, induced a greater than 5-fold increase in

SCE at a concentration of only 0.15 ppm.
The response to BaP in the lymphocyte cultures indicated that the S9 metabolic system was functional. Although increases in SCE of only 50% over the number in control cultures were induced, these increases are statistically significant (P<0.001). In the presence of S9 mix, only 1/16 the amount of BaP was required to induce the same number of SCE observed without S9, indicating that metabolism had occurred. However, 0.5µg/ml of BaP with S9 mix was toxic to these cultures, as indicated by poor cell morphology and a reduced number of cells in mitosis.
Wilmington crude did not induce an increase in SCE at any dose concentration with or without S9 activation.
The data are summarized in the following table.

Treatment	Dose (ppm)	No. of cells	SCE/cell ±S.E.
24 hours exposure, without S9			
Tween 80	0	25	8.4±0.5
	100	25	9.7±0.6
B(a)P	0	25	7.7±0.6
	4	25	10.8±1.2
	8	25	15.1±0.9**
Wilmington crude oil			
	40	25	10.7±1.2
	50	25	9.9±0.8
2.5 hours exposure, with S9			
Tween 80	40	25	9.6±0.0
	100	25	8.0±0.5
B(a)P	0	25	10.9±1.9
	0.2	25	13.4±0.8**
	0.5	10	15.9±1.2**
	1.0	0	Toxic
MNNG	0	25	8.7±0.4
	0.015	23	11.7±0.5**
	0.05	23	28.3±1.3**
	0.15	21	48.5±2.2**
Wilmington crude oil			
	20	25	8.8±0.7
	30	25	6.6±0.5

** P<0.001

Reliability : (2) valid with restrictions
Complete description of study not given in publication. Some methods details published elsewhere

(43)

5.6 GENETIC TOXICITY 'IN VIVO'

Type	:	Micronucleus assay
Species	:	rat
Sex	:	male/female
Strain	:	Sprague-Dawley
Route of admin.	:	dermal
Exposure period	:	13 weeks
Doses	:	30, 125 and 500 mg/kg/day
Result	:	negative
Year	:	1990
GLP	:	yes
Test substance	:	Lost Hills Light crude

Method : The animals used in this study were taken from a 13 week repeated dermal administration study (Study 63834).
Bone marrow was removed from the femurs of five male and five female rats from each dose group (0, 30, 125 and 500 mg/kg/day).
A nearly pure erythrocyte fraction was obtained and two slides prepared for each animal. 1000 PCEs (polychromatic erythrocyte) and 1000 NCEs (normochromatic erythrocyte) were scored to determine the percentage of micronucleated erythrocytes. 1000 erythrocytes were scored to determine the ratio of PCEs to NCEs.

Result : The results are given in the following table. Standard deviations have not been included in the table. However, statistical analyses were conducted and it was found that there were no significant differences between treated and control animals.
It was concluded that the test material was not cytotoxic to red blood cell formation and furthermore did not increase the formation of micronucleated PCEs or NCEs in the bone marrow.

Dose mg/kg	Sex	No of animals	PCE/NCE	MNPCEs (%)	MNNCEs (%)
0	F	5	1.34	0.1	0
0	M	5	0.79	0.02	0
0	M+F	10	1.06	0.06	0
30	F	5	1.26	0.04	0
30	M	5	0.68	0	0.02
30	M+F	10	0.97	0.02	0.01
125	F	5	1.18	0	0
125	M	5	1.17	0.06	0
125	M+F	10	1.17	0.03	0
500	F	5	1.25	0.02	0
500	M	5	0.83	0.04	0
500	M+F	10	1.04	0.03	0

PCE Polychromatic erythrocytes
NCE Normochromatic erythrocytes
%MNPCEs % Micronucleated PCEs
%MNNCEs % Micronucleated normochromatic erythrocytes

Remark : Two further studies have been reported.

1. Belridge heavy crude.
   The results of this study were the same as those for the study on Lost Hills Light described above. (Mobil study 63847)

2. Wilmington crude
   Results in ICR Swiss mice showed that Wilmington crude oil had relatively little capacity to cause chromosome breakage and/or non-disjunction. (Lockard et al, 1982)

Reliability : (1) valid without restriction
(43) (64) (71) (72)

Type : Sister chromatid exchange assay
Species : mouse
Sex : male
Strain : ICR
Route of admin. : i.p.
Doses : 1.8, 3.6 &7.2 g/kg
Year : 1982
GLP : no data
Test substance : Wilmington crude

Method : The assay was carried out in groups of 3 male 10-14 week old Sch:ICR mice.
The mice were lightly anesthetized with ether and a 50 mg tablet of BrdUrd was inserted beneath the skin on the dorsal side of the neck of each animal. After 1 hour, treatment was given by a single i.p. injection at doses of 1.8, 3.6 and 7.2 g/kg (equivalent to 0.8, 0.4 and 0.2 times the approximate LD50).
Control groups of mice were injected with B(a)P at doses of 0.16, 0.08 and 0.04 g/kg, Cyclophosphamide at doses of 0.01 and 0.005 g/kg in distilled water. Trioctanoin and distilled water were also given to negative solvent controls.
22 hours after treatment, animals were injected i.p. with Colchicin (10 mg/kg). 2 hours later the mice were sacrificed by cervical dislocation and their femurs were removed. The bone marrow cells were flushed out with saline. Pooled cells from the 2 femurs of each animal were treated with hypotonic KCl for 30 minutes, were fixed and were spread on slides. The slides were stained and the numbers of SCE counted in 25 metaphase cells from each animal, each metaphase having 36-42 chromosomes.

Result : Control animals injected with the solvent trioctanoin had a mean of 5.3 SCE/cell. A slight but significant increase in SCE was found in animals treated with the highest dose only of Wilmington crude oil, but the response was not dose related.
B(a)P and cyclophosphamide induced dose-related increases in SCE.
The actual results are shown in the following table.

Treatment	Dose	No. survivors/ No. treated	No. cells examined	SCE/ cell±S.E.
Trioctanoin	0.5 ml/ mouse	7/8	175	5.3±0.4
B(a)P	0.16 g/kg	3/3	75	9.7±0.6***
	0.08	3/3	75	8.5±0.5***
	0.04	3/3	75	7.8±0.5***
Cyclophosphamide				
	0.01 g/kg	3/3	75	19.5±0.6***
	0.005	3/3	75	14.6±0.6***
Wilmington crude oil				
	7.2 g/kg	3/3	75	6.6±0.4**
	3.6	3/3	50	5.0±0.3
	1.8	3/3	75	6.4±0.4

** P<0.05
*** P<0.001

Reliability : (2) valid with restrictions
Probably not to GLP and method description is not comprehensive.

(43)

5.7 CARCINOGENICITY

Species : mouse
Sex : male
Strain : C3H
Route of admin. : dermal
Exposure period : 18 months
Frequency of treatm. : Twice weekly
Doses : 50 mg per application
Result : positive
Control group : yes
GLP : no data
Test substance : Two crudes C & D

Method : 50 mg undiluted test material was applied to the shorn interscapular region of groups of 50 male mice. Application was twice weekly for 18 months or until grossly-observable cancer was found. Each animal was observed weekly for the appearance of tumors. The percentage of animals developing tumors and the time to appearance of first tumor were recorded. No distinction was made between histologically benign or malignant lesions.

Result : The % tumors and average latencies for the two crudes C & D are shown below.

	% Tumors*	Latency**
Crude C	33	76
Crude D	56	64

* Based on Final Effective Number ** Average latency in weeks

Test substance : Two crudes were included in this study. They were characterized as follows:

	Crude C	Crude D
Sulfur (wt%)	0.21	2.54
Distillation yield (vol%)		
Int-120°F	0.0	3.3
120-350	2.5	18.3
350-550	41.9	19.5
550-700	21.0	12.2
700-1070	31.2	26.8
1070 btms	3.4	19.9
Composition of 550-700°F		
Fraction (wt%)	5.1	33.3
Monocycloparaffins	8.1	11.5
Polycycloparaffins	46.6	19.5
Total saturated hydrocarbons	59.8	64.3
Mononuclear aromatics	21.5	11.0
Di- and Trinuclear aromatics	14.4	21.8
Polynuclear aromatics	1.0	1.8
Total aromatic hydrocarbons	36.9	34.6
Resins	3.3	1.1
Asphaltenes	0.0	0.0

Reliability : (2) valid with restrictions

(42)

Species : mouse
Sex : male/female
Strain : C3H
Route of admin. : dermal
Exposure period : 105 weeks
Frequency of treatm. : Three times weekly
Doses : 25 mg per application
Control group : yes
Year : 1988
GLP : no data
Test substance : Heavy crude oil, San Joaquin Valley

Method : The study reported by Clarke et al was a comparison of the carcinogenicity of shale and petroleum crude oil and of several distillate streams derived from the two crude oils. Only the details relating to petroleum crude and the respective positive and negative controls are included in this summary. The crude oil, positive and negative control materials were applied three times weekly at a dose of 25 mg/application to 25 male and 25 female mice. The materials were applied to the shorn dorsal thoracic region which was shaved as necessary during the study. Dosing was continued for up to 105 weeks.
Animals were observed twice daily for overt signs of toxicity, moribundity and mortality. Animals were weighed weekly for the first 13 weeks and every two weeks thereafter. Animals were palpated weekly for external and internal masses.

Every animal was subjected to a complete necropsy. All organs and the remainder of the carcass were fixed and stored. Body weights and weights of liver, kidneys, brain and gonads were recorded. A microscopic examination was made of histological slides prepared from skin from a

control site, skin from treated site, kidney, liver, lungs, gonads, urinary bladder, spleen, sternal bone marrow and any other organ considered abnormal at necropsy.

Result : Survival was affected by treatment with petroleum crude oil.
After one year, survival was approximately 70% and by week 70 survival was approximately 50%. There were few survivors at 105 weeks. This is in contrast to the negative control group with 50% survival at the end of the study. Dermal irritation at the test site first appeared at 271 days and males developed irritation earlier than the females [no details given]. Necrosis occurred and was characterized as loss of integrity of the skin with visible cracking, separation and sloughing of skin often revealing underlying mesenchymal tissue.

There was no indication of toxic or oncogenic effects on internal organs.

The authors reported the occurrence of reactive hyperplasia in the spleen and bone marrrow and this was characterized by increased populations of lymphocytes, plasma cells and neutrophils or granulocyte precursors and was attributed to the inflammation, necrosis and other tissue alterations occurring at the test site. [There is no indication in the publication whether this finding occurred in the group treated with petroleum crude oil.]

The tumor incidences that were recorded are shown in the following table:

	Crude oil	**B(a)P**	**Control**
Incidence (%)	37/44 (84)	49/49	0/46
Latency (weeks)	62±13	28±4	
Squamous cell carcinoma	29 (66)	49 (100)	0
Fibrosarcoma	7 (16)	0	0

Test substance : The San Joaquin Valley crude oil is a heavy crude with the following characteristics:

Gravity (°API)	10.7
Boiling range (°F)	410-1071
Nitrogen (ppm wt)	8150
Sulfur (ppm wt)	10,510

Due to its high viscosity, the crude oil was administered as a 2:1 dilution in mineral oil.
Mineral oil USP was used as a negative control
B(a)P (0.15% w/v in mineral oil) was used as a positive control

Reliability : (2) valid with restrictions
Few experimental details are given in this publication. Despite this, the information may be used in the overall assessment of the carcinogenic potential of crude oil

(11)

5.8.2 DEVELOPMENTAL TOXICITY/TERATOGENICITY

Species	:	rat
Sex	:	female
Strain	:	Sprague-Dawley
Route of admin.	:	gavage
Frequency of treatm.	:	Various
Year	:	1987
GLP	:	no data
Test substance	:	Prudhoe Bay crude oil (Heavy crude oil)

Method :

Presumed pregnant female Sprague-Dawley rats were used for this series of studies.

Prudhoe Bay crude oil was administered by gavage according to the following schedules

1. As a single dose (5 ml/kg) on either day 3, 6, 11, 15 or 17 of gestation.
2. Daily from gestation days 6-17 incl at a dose level of 1.0 or 2.0 ml/kg.
3. As a single dose on day six of gestation at either 2, 5, 7 or 10 ml/kg.

Respective controls received equivalent amounts of saline.

On day 18 of pregnancy, animals were sacrificed and the numbers and position of implantations, resorptions and dead fetuses were recorded. The live fetuses were removed, weighed and inspected for gross external abnormalities with the aid of a dissection microscope. Skeletal examinations and examination of soft internal tissues were not carried out in this preliminary study.
Student's t-test was used for comparing control and experimental results. P values were derived from a 2-tailed table of Student's values for t. The level of significance chosen was $P<0.05$.

Result :

There were no maternal deaths following oral administration of PBCO although at high doses the animals exhibited signs of intoxication (crouching, indicative of acute irritation).
Maternal body weight changes were significantly less in animals that had been treated with PBCO at early days of gestation, in particular those animals given PBCO on day 6 of gestation only.
The body weight data are shown in the following tables. Note that although the author provided data on mean and standard deviation, for simplicity this summary only contains mean values (SDs are not shown). Statistically significant differences are indicated.
Body wt changes for dams given a single (5 ml/kg) dose of PBCO. Figures in parenthesis indicate group size

Treatment	**Body weight change (g on days indicated)**				
Day	**0-6**	**6-11**	**11-15**	**15-18**	**0-18**
Control (11)	30.4	32.6	28	30	121
3 (9)	32.75	28.25	23.5*	31	115.22
6 (10)	28.8	22.4*	24.2*	27.8	103.2*
11 (10)	34.8	35.6	16*	35.2	122.6
15 (10)	32.5	33.5	28	35.3	131.5
17 (10)	31.67	34	27.33	31.67	124.67

* Significant from control ($P<0.05$)

Effect of different doses of PBCO on dam body weight changes
No. of animals shown in parenthesis

Treatment	Body weight change (g on days indicated)				
	0-6	6-11	11-15	15-18	0-18
Control (8)	27.33	25.67	27.33	32	112.33
Dose (ml/kg) given on day 6 only					
2 (8)	27.5	25	23*	27.5	1.3
5 (10)	26.21	18.08*	19.5*	26.92*	91.22*
7 (8)	26.67	17.33*	15.35*	26.92*	87.67*
10 (8)	30.67	14.33*	11*	20*	75.67*
Dose given on days 6-17 daily					
1 (10)	31	21*	23.5*	22*	97.5*
2 (10)	27.5	16.5	13*	17*	75*

* Significant from control (P<0.05)

The author concluded that the reduced body weight gains were probably due to the high resorption rates that occurred.

Effects on fetal viability and development
The results of the effect of administering PBCO as a single dose of 5 ml/kg at different stages of gestation are summarized below:

Values shown are mean±SD
* significant from control (p<0.05)

No. implants per dam	Resorptions/ dead fetuses (%)	No. live fetuses	Fetal weight (g)	Fetal crown-rump length (mm)
Day 0				
13.5±2.39	4.68±.93	13±1.24	1.267±.09	22.2±1.44
Day 3				
14.5±1.92	10.77±1.02*	11.25±1.5	1.19±.08*	20±1.73*
Day 6				
3.23±1.43	15.48±1.32*	11.02±1.49*	1.2±.09*	1.16±1.38
Day 11				
2.9±1.26	8.22±1.08*	11.4±.89*	1.287±.11	21.56±1.37
Day 15				
12.72±1.63	5.02±1.19	12.5±.71	1.313±.083	22.65±1.33
Day 17				
13±1.8	3.62±.89	12.33±1.15	1.304±.124	23.01±1.41

These results demonstrate that the incidence of resorptions and dead fetuses was increased in the animals given PBCO during the early days of pregnancy whereas those given PBCO during the later stages of pregnancy were unaffected. The greatest effect was on day 6 of gestation.

Repeated daily administration of low doses (1 or 2 ml/kg) on days 6 through 7 showed an additive effect and at a dose level of 2 ml/kg/day resulted in up to 40% fetal deaths. These data are shown in the following table.

The effect of different doses of PBCO

Values shown are mean ± SD
* significant from control (p<0.05)

No. implants per dam	Resorptions/ dead fetuses (%)	No. live fetuses	Fetal weight (g)	Fetal crown-rump length (mm)
Dose (ml/kg)				
0 ml/kg				
13.95±2.23	3.96±0.87	13.72±1.56	.309±.091	21.62±1.17
Day 6				
2 ml/kg				
14±2.63	6.84±1.21	13.85±2.33	1.326±.119	21.78±1.54
5 ml/kg				
13.26±1.95	12.67±1.18*	11.73±1.47*	1.216±.09*	21.54±1.27
7 ml/kg				
12.93±2.35	13.23±1.35*	11.68±1.46*	1.2±.109*	21.28±.89
10 ml/kg				
14.12±2.12	22.86±1.98*	10.5±1.59*	1.192±.108*	0.73±1.19
Day 6-17 (daily)				
1 ml/kg				
13.92±2.46	17.5±2.02*	10±1.95*	1.184±.107	0.16±1.34*
2 ml/kg				
12.93±1.98	43.8±1.92*	8.72±1.34*	1.182±.097*	.96±1.31*

In conclusion, administration of PBCO to pregnant females resulted in an increased incidence of resorptions, increased fetal death and decreased fetal body weight. These effects occurred at doses which were maternally toxic.

Reliability : (2) valid with restrictions
Probably not conducted to GLP. Study was a preliminary study and was limited in scope. Only information on embryo and fetotoxicity was derived in this study.

(40)

Species : rat
Sex : female
Strain : Sprague-Dawley
Route of admin. : dermal
Frequency of treatm. : Daily
Duration of test : Days 0-19 of gestation
Doses : 30, 125 and 500 mg/kg/day
Control group : yes, concurrent no treatment
Year : 1991
GLP : no data
Test substance : Belridge Heavy Crude

Method : Groups of twelve presumed-pregnant Sprague-Dawley rats were distributed into six groups as shown below. The test material was applied to the shorn dorsal skin once daily at the doses shown from day 0 to day 19 of gestation. The animals were shaved weekly and were fitted with collars to prevent ingestion of the test material.

Group	Dose level
Prenatal groups	
1	Sham control
2	30 mg/kg/day
3	125 mg/kg/day
4	500 mg/kg/day
Postnatal groups	
5	Sham control
6	500 mg/kg/day

Animals were observed at least once daily throughout gestation until sacrifice for signs of pathosis, abortion, premature delivery, dystocia and/or death. Dams and their litters were observed postpartum days 0 through 4. On day 0 postpartum, pups were examined for external malformations and variations and were observed daily for the presence of milk in their stomachs.
Body weights and food consumption of all prenatal group females were recorded at regular intervals throughout gestation and for the post natal groups throughout gestation and body weights only on postpartum days 0 and 4.
On day 20 of gestation all prenatal group animals were sacrificed. The abdominal cavity was exposed and the reproductive organs examined. Following removal of the uterus and ovaries the remains were subjected to macroscopic examination and the liver and thymus were weighed.
The uterus and ovaries of each animal were examined grossly. The no. of corpora lutea per ovary was recorded. Ovaries of non pregnant females were examined and then discarded. The uterine contents of each pregnant animal were exposed and the number and location of all implantations were recorded. The uterus of each female that appeared non-gravid was pressed between two glass slides and examined grossly for evidence of implantation. Blood samples were collected at the time of sacrifice and these were used for a range of serum chemistry determinations.
Each live fetus was weighed and its sex determined and examined for external anomalies. After gross examination the fetuses were equally distributed into two groups. One group was processed for an evaluation of visceral anomalies and the other group for skeletal anomalies.
Females in the postnatal groups with surviving offspring were sacrificed on post partum day 4 and the abdominal cavity was exposed for a gross examination of the reproductive organs. The uterus was also examined and then discarded. All organs were examied macroscopically and the thymus and liver were weighed.

Remark : This study was also reported in the open literature by Feuston et al.

Result : Clinical observations

Treatment related clinical observations in the prenatal and postnatal groups consisted of skin irritation in the 500 mg/kg/day group. This included erythema, edema, scabs and open sores at the treatment site. There was also a red vaginal discharge in animals in this group. One high dose prenatal female had excessive vaginal discharge and, after being found moribund, was sacrificed on gestation day 14. Uterine examination revealed that all but two fetuses had been resorbed.
Body weights and food intakes of the 500 mg/kg/day prenatal groups were reduced compared to controls (reduced by approx 31%). Other dose groups were unaffected.

Necropsy findings

With the exception of an approximately 11% increase in relative liver

weight in the 500 mg/kg/day group, no other organ or relative organ weight changes were recorded. [The authors comment that the observed increase in relative liver weight was possibly attributable to the decreased body weight of this group].
A prominent vascular pattern of the liver was observed in one female of the 125 mg/kg/day group and two animals in the highest dose group.

Serum chemistry
The only observation was a 38% reduction of total bilirubin in the prenatal 500 mg/kg/day group.

Reproductive and fetal evaluations
Effects only occurred in the highest dose group (500 mg/kg/day). These included

an increase in the mean number/percent resorptions (5.3 compared to 1.3 and 35% compared to 7.8% respectively) and a corresponding decrease in litter size (10.8 compared to 14.5).

A decrease in mean fetal body weights for all viable fetuses (3.4 compared to 3.7).

Male fetuses in this dose group seemed to be more affected than females.
Fetal skeletal anomalies recorded were limited to incomplete ossification of the nasal bones and caudal centra.
There were no treatment related visceral anomalies.

Post partum observations
During the post partum period there were no clinical observations of note except some persistent skin irritation. There were no findings at necropsy.

Litter data
Two females in the 500 mg/kg/day group had no viable offspring; their litters were totally resorbed. The incidence of pup mortality was two times greater in the group treated with crude oil during the lactation period. Mean duration of gestation was unaffected, but it was noted that the majority of treated females delivered either in the afternoon of day 22 or morning of day 23 whereas the controls delivered in the morning of day 22. [The biological significance of this observation is uncertain.]

Reliability : (1) valid without restriction

(29) (67)

Species	:	rat
Sex	:	female
Strain	:	Sprague-Dawley
Route of admin.	:	dermal
Frequency of treatm.	:	Daily
Duration of test	:	Days 0-19 of gestation
Doses	:	125, 500 and 2000 mg/kg/day
Control group	:	yes, concurrent no treatment
NOAEL maternal tox.	:	125 mg/kg bw
NOAEL teratogen.	:	500 mg/kg bw
Year	:	1991
GLP	:	no data
Test substance	:	Lost Hills Light

Method : Groups of twelve presumed-pregnant Sprague-Dawley rats were distributed into six groups as shown below. The test material was applied

to the shorn dorsal skin once daily at the doses shown from day 0 to day 19 of gestation. The animals were shaved weekly and were fitted with collars to prevent ingestion of the test material.

Group	Dose level
Prenatal groups	
1	Sham control
2	125 mg/kg/day
3	500 mg/kg/day
4	2000 mg/kg/day
Postnatal groups	
5	Sham control
6	20000 mg/kg/day

Animals were observed at least once daily throughout gestation until sacrifice for signs of pathosis, abortion, premature delivery, dystocia and/or death. Dams and their litters were observed postpartum days 0 through 4. On day 0 postpartum, pups were examined for external malformations and variations and were observed daily for the presence of milk in their stomachs.
Body weights and food consumption of all prenatal group females were recorded at regular intervals throughout gestation. For the post natal groups body weights and food intakes were recorded throughout gestation but only body weights were recorded on postpartum days 0 and 4.
On day 20 of gestation all prenatal group animals were sacrificed. The abdominal cavity was exposed and the reproductive organs examined. Following removal of the uterus and ovaries the remains were subjected to macroscopic examination and the liver and thymus were weighed.
The uterus and ovaries of each animal were examined grossly. The number of corpora lutea per ovary was recorded. Ovaries of non-pregnant females were examined and then discarded. The uterine contents of each pregnant animal were exposed and the number and location of all implantations were recorded. The uterus of each female that appeared non-gravid was pressed between two glass slides and examined grossly for evidence of implantation. Blood samples were collected at the time of sacrifice and these were used for a range of serum chemistry determinations.
Each live fetus was weighed and its sex determined and examined for external anomalies. After gross examination the fetuses were equally distributed into two groups. One group was processed for an evaluation of visceral anomalies and the other group for skeletal anomalies.
Females in the postnatal groups with surviving offspring were sacrificed on post partum day 4 and the abdominal cavity was exposed for a gross examination of the reproductive organs. After examination, the uterus was discarded. All organs were examined macroscopically and the thymus and liver were weighed.

Remark : This study was also reported in the open literature by Feuston et al.

Result : There were only few clinical observations that were considered to be treatment-related and these were confined to the high dose group (2000 mg/kg/day). The observations consisted of a red vaginal discharge, and one female in this group was pale in color. Slight skin irritation occurred in a few animals but it is not clear whether this was compound-related or self inflicted because the animals attempted to groom themselves, despite the fact that they had been fitted with collars. It is possible also that because of this some ingestion of test material may have occurred.
Animals in the 500 and 2000 mg/kg/day groups gained less weight than the controls over the gestation period (80% and 60% of controls respectively).

Food consumption for these two groups was also less than controls for the first 10 days of gestation but was similar to controls thereafter.
At necropsy, it was noted that there were more animals in the 2000 mg/kg/day group with a small thymus. Absolute and relative thymus weights were reduced in this group by approximately 50%. A reduction in absolute thymus weight was also recorded for the 500 mg/kg/day group, but the difference was not statistically significant.
Liver weights (absolute and relative) were increased in the 500 and 2000 mg/kg group, but only the relative organ weights were statistically significant (11 and 18% increases for the 500 and 2000mg groups respectively).
Serum chemistry differences were only observed in the 500 and 2000 mg/kg/day groups These changes were as follows:

Parameter	**Dose group (mg/kg/day)**	
	500	**2000**
AST		+27%
ALT		+37%
Alkaline phosphatase		+64%
Cholesterol		+22%
Triglycerides	-42%	-62%
Total bilirubin	-35%	-49%
A/G ratio		+15%
Phosphorus		+20%
SDH		+67%

All other serum chemistry measurements were unaffected by treatment.

Reproductive evaluations
The only effect was a significant increase in mean number/% resorptions (6-fold) and a corresponding decrease in litter size (38%) in the 200 mg/kg/day group.

Fetal evaluations
Mean fetal body weights were reduced by approximately 13% in the 200 mg/kg/day group.
There was an increase in incomplete ossification of various skeletal structures which included nasal bones and vertebrae but no increases in visceral abnormalities. The data are shown below:
Date are shown as fetal incidence/litter incidence

Anomoly	**Dose group (mg/kg/day)**			
	0	**125**	**500**	**2000**
Ossification variants				
nasal bones	1.0/8.3	15b/55	33b/82b	96b/100b
Thoracic centra	4/33	5.5/36	8.5/55	15a/78
Caudal centra	1/8.3	9.9a/36	3.7/36	15b/67b
Sternebrae <2	2/8.3	6.6/36	7.3/36	23b/67a
Visceral malformations				
Right-sided esophagus	0/0	-	0/0	4.1/22

a P<0.05
b P<0.01

In the prenatal group, the litter effects were;

Three females had no viable offspring, their litters were totally

resorbed

Two females had their entire litters die by postpartum day 3

There was a significant increase in the number of stillborn pups (7 compared to 2 for controls)

Liveborn pups weighed less than the controls (-8% and -17% on days 0 and 4 respectively).

Conclusion : Lost Hills Light crude was shown to be maternally toxic at 500 and 2000 mg/kg/day and developmentally toxic at 2000 mg/kg/day.

Reliability : (1) valid without restriction

(29) (68)

5.8.3 TOXICITY TO REPRODUCTION, OTHER STUDIES

Type : Study for sperm abnormality
In vitro/in vivo : In vivo
Species : mouse
Sex : male
Strain : other: B6C3F1/Hap hybrid
Route of admin. : i.p.
Frequency of treatm. : Once daily for 5 days
Duration of test : 40 days
Doses : 1.0 & 2.1 g/kg
Control group : yes
Year : 1982
GLP : no data
Test substance : Wilmington crude

Method : Groups of five 10-12 week old male mice were injected i.p. daily for 5 days with test samples in trioctanoin at doses of 1 and 2.1 g/kg. Control mice were injected with trioctanoin (5x0.25 ml) or BaP (5x0.05 or 1x0.07g). After 35 days, the animals were sacrificed, the cauda epididymes were removed, combined and minced in 0.9% NaCl or Tyrode's solution and the sperm were stained by adding eosin Y solution. The preparations were spread on slides and air-dried. The percentage of abnormal sperm was determined in 500 sperm from each animal. For control of bias in scoring, positive and negative control slides from a previous experiment were included at a ratio of 1 bias control slide to 5 new slides.

Result : The data from the study are given in the following table and show that treatment with Wilmington crud did not cause an increase in the percentage of abnormal sperm.

Sample	Dose	No. survivors/ no. treated	Abnormal sperm (%±SD)
Trioctanoin			
	5x0.25 ml	10/10	4.5±1.7
Wilmington crude			
	5x2.1 g/kg	5/5	6.1±2.1
	5x1.0 g/kg	5/5	5.4±1.3
BaP	5x0.05 g/kg	10/10	19.7±16.6

(43)

5.9 SPECIFIC INVESTIGATIONS

Endpoint : Mechanistic Studies
Type : Initiation/promotion assay
Species : mouse
Sex : male
Strain : Charles River (ChR-CD)
Route of admin. : Dermal
No. of animals : 30
Vehicle : undiluted
Exposure period : 180 day(s)
Frequency of treatm. : See method
Doses : See method
Control group : yes
Year : 1981
GLP : no data
Test substance : South Louisiana crude

Method : The authors summarized the protocol in the following table.

Animals: Charles River (ChR-CD) male mice Six weeks old
Toe clipped for identification
Backs shaved free of hair

Group size 30 mice per group

Housing 2 mice per cage

Diet Water and laboratory chow ad libitum

Observation Daily

Clipping of hair on the back Weekly, as needed

Treatments
Initiation: 50µl material as received
Promotion: 2 weeks after initiation treatment
2.5µg phorbolmyristate acetate (PMA)/0.1ml acetone
Three times a week for 180 days

Parameters measured
Body weight, recorded weekly
Time to development of first tumor
Tumors charted monthly
Mortality

Result :

Sample	No. mice with tumors lasting >30 days	Av No. tumors/ tumor-bearing mouse	No. days to first mouse with tumor
Benzo[a]pyrene positive control			
	26	6.0	48
South Louisiana Crude			
	5	1.4	71
Toluene and PMA			

	4	1.5	111
PMA alone			
	3	1.0	92

The authors concluded that the South Louisiana crude oil was not an initiator.

Reliability : (4) not assignable
The study was a preliminary screen and was not fully reported. The data are however are useful in assessing the tumor initiating activity of crude oil. (9)

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Study No. 63841
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B005 SUBST_MASTER_TAB
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* encompasses light, medium, and heavy petroleums, as well as the oils
* extracted from tar sands. Hydrocarbonaceous materials requiring major
* chemical changes for their recovery or conversion to petroleum refinery
* feedstocks such as crude shale oils, upgraded shale oils and liquid coal
* fuels are not included in this definition.
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F101 Robust summary of information on crude oil
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F007 22-02-2002
F008 Robust summary of information on crude oil
F009 A35-01
EOR
F001 37
F002 1
F003 Crude oil
F012 Y
F010 09-10-2003
F004 12031538
F005 22-02-2002
F006 12031538
F007 22-02-2002
F008 Crude Oil

F009 A35-01
EOB
C
B101 GI_GENERAL_INFORM_TAB
F001 37
F002 1
F003 13-11-2003
F004 IUC4
F010 A04-06
F011 A19-03
EOB
C
B126 GI_ADD_REVIEWS_TAB
F001 37
F002 1
F003 06-10-2003
F004 IUC4
F007 IARC Review
EOB
C
B201 PC_MELTING_TAB
F001 37
F002 1
F003 23-02-2003
F004 IUC4
F015 A36-002
F007 A02-03
F008 -30
F009 30
F010 A30-02
F011 A30-02
F012 P01-03: ASTM D97
F014 A03-02
EOB
C
B202 PC_BOILING_TAB
F001 37
F002 1
F003 23-02-2003
F004 IUC4
F016 A36-002
F007 A02-03
F008 -1
F009 565
F010 1013
F011 P02-01
F012 A30-03
F013 P03-03: ASTM D86
F015 A03-02
EOB
C
B204 PC_VAPOUR_TAB
F001 37

F002 1
F003 09-10-2003
F004 IUC4
F015 A36-003
F014 A03-02
F018 A01-03: Crude oil
EOB
C
B205 PC_PARTITION_TAB
F001 37
F002 1
F003 23-02-2003
F004 IUC4
F014 A36-002
F007 A02-03
F008 2
F009 6
F011 P07-04
EOB
C
B206 PC_WATER_SOL_TAB
F001 37
F002 1
F003 30-09-2003
F004 IUC4
F023 A36-003
F020 P09-03: Preparation of water soluble fraction
F021 1990
F022 A03-01
F025 A01-03: Crude oil
F030 C14-001
EOB
C
B212 PC_OTHER_TAB
F001 37
F002 1
F003 23-02-2003
F004 IUC4
F008 A36-003
F009 CONCAWE
EOB
C
B301 EN_PHOTODEGRADATION_TAB
F001 37
F002 1
F003 23-02-2003
F004 IUC4
F045 A36-003
F008 F01-01
F009 F02-05: according to Atkinson, 1990
F011 F03-01
F034 F06-03
F035 1000000

F036 F07-02
EOB
C
B302 EN_STABILITY_IN_WATER_TAB
F001 37
F002 1
F003 13-11-2003
F004 IUC4
F040 A36-002
F039 A03-01
EOB
C
B306 EN_DISTRIBUTION_TAB
F001 37
F002 1
F003 23-02-2003
F004 IUC4
F010 A36-002
F007 F24-02
F008 F23-01
F009 1981
EOB
C
B309 EN_BOD_COD_TAB
F001 37
F002 1
F003 23-02-2003
F004 IUC4
F022 A36-003
F007 F32-03: Theoretical Oxygen Demand (TOD) was calculated
F008 1971
F009 A03-02
EOB
C
B311 EN_OTHER_TAB
F001 37
F002 2
F003 13-11-2003
F004 IUC4
F009 General
EOB
C
B401 EC_FISHTOX_TAB
F001 37
F002 1
F003 09-10-2003
F004 IUC4
F033 A36-002
F007 A01-03: CAS 8002-05-9 Crude oil - 0.5% paraffinic
F008 E01-04
F009 E02-0101
F010 E03-03
F011 2001

F012 96
F013 E04-02
F014 E05-02
F031 A03-03
F032 A03-03
F050 C47-001
EOR
F001 37
F002 2
F003 09-10-2003
F004 IUC4
F033 A36-002
F007 A01-03: CAS 8002-05-9 Crude oil 3% paraffinic
F008 E01-04
F009 E02-0101
F010 E03-03
F011 2001
F012 96
F013 E04-02
F014 E05-02
F031 A03-03
F032 A03-03
F050 C47-001
EOR
F001 37
F002 3
F003 09-10-2003
F004 IUC4
F033 A36-004
F007 A01-03: Forties crude oil
F008 E01-05
F009 E02-0139
F010 E03-05: procedure as detailed in paper by Westlake (see Reference)
F014 E05-02
F021 A02-03
F022 258
F031 A03-01
F032 A03-03
F045 E35-02
EOR
F001 37
F002 4
F003 09-10-2003
F004 IUC4
F033 A36-004
F007 A01-03: Forties crude oil
F008 E01-05
F009 E02-0139
F010 E03-05: procedure as detailed in paper by Westlake (see Reference)
F011 1991
F012 96
F013 E04-02
F014 E05-02

F021 A02-03
F022 291
F031 A03-01
F032 A03-03
F045 E35-02
EOR
F001 37
F002 5
F003 09-10-2003
F004 IUC4
F033 A36-004
F007 A01-03: South Louisiana and Kuwait crude oils
F008 E01-05
F009 E02-0030
F010 E03-05: Procedure as detailed in paper by Anderson (see Reference)
F011 1974
F012 96
F013 E04-02
F014 E05-02
F021 A02-03
F022 2900
F023 80000
F031 A03-03
F032 A03-02
F045 E35-02
EOR
F001 37
F002 6
F003 09-10-2003
F004 IUC4
F033 A36-004
F007 A01-03: South Louisiana and Kuwait crude oils
F008 E01-05
F009 E02-0047
F010 E03-05: Procedure as detailed in paper by Anderson (see Reference)
F011 1974
F012 96
F013 E04-02
F014 E05-02
F021 A02-03
F022 6000
F023 14800
F032 A03-03
F045 E35-02
EOR
F001 37
F002 7
F003 09-10-2003
F004 IUC4
F033 A36-004
F007 A01-03: South Louisiana and Kuwait crude oils
F008 E01-05
F009 E02-0082

F010 E03-05: Procedure as detailed in paper by Anderson (see Reference)
F011 1974
F012 96
F013 E04-02
F014 E05-02
F021 A02-03
F022 3700
F023 9400
F031 A03-01
F032 A03-03
F045 E35-02
EOR
F001 37
F002 8
F003 09-10-2003
F004 IUC4
F033 A36-003
F007 A01-03: Norman Wells crude oil
F008 E01-05
F009 E02-0139
F010 E03-05: Procedure as detailed in paper by Lockhart, Danell and Murray
* (see Reference)
F011 1987
F012 48
F013 E04-02
F014 E05-02
F031 A03-03
F032 A03-03
EOB
C
B402 EC_DAPHNIATOX_TAB
F001 37
F002 1

F003 29-09-2003
F004 IUC4
F032 A36-002
F007 A01-03: CAS 8002-05-9 Crude oil, 0.5% paraffinic
F008 E06-0034: Kelp Forest Mysid (Holmesymis costata)
F009 E07-04: USEPA 600 4-90-027
F010 2002
F011 96
F012 E04-02
F013 E05-02
F030 A03-03
F031 A03-03
F042 E01-04
F050 C47-001
EOR
F001 37
F002 2
F003 17-03-2003
F004 IUC4

F032 A36-002
F007 A01-03: CAS 8002-05-9 Crude oil, 3% paraffinic
F008 E06-0034: Kelp Forest Mysid (Holmesymis costata)
F009 E07-04: USEPA 600 4-90-027
F010 2001
F011 96
F012 E04-02
F013 E05-02
F030 A03-03
F031 A03-03
F042 E01-04
F050 C47-001
EOR
F001 37
F002 3
F003 09-10-2003
F004 IUC4
F032 A36-003
F007 A01-03: Samples of 8 crude oils
F008 E06-0006
F009 E07-04: Procedure as detailed in paper by Franklin and Lloyd (see
* Reference)
F010 1982
F011 96
F012 E04-02
F013 E05-02
F026 LC50
F027 A02-03
F028 27
F029 110
F030 A03-01
F031 A03-02
F047 E35-02
EOR
F001 37
F002 4
F003 10-10-2003
F004 IUC4
F032 A36-003
F007 A01-03: Samples of 8 crude oils
F008 E06-0006
F009 E07-04: Procedure as detailed in paper by Franklin and Lloyd (see
* Reference)
F010 1982
F011 96
F012 E04-02
F013 E05-02
F026 LC50
F027 A02-03
F028 41
F029 119
F030 A03-01
F031 A03-02

F047 E35-02
EOR
F001 37
F002 5
F003 09-10-2003
F004 IUC4
F032 A36-004
F007 A01-03: Forties crude oil
F008 E06-0010
F009 E07-03
F010 1981
F011 24
F012 E04-02
F013 E05-02
F020 A02-03
F021 36
F030 A03-01
F031 A03-03
F045 E35-02
EOR
F001 37
F002 6
F003 09-10-2003
F004 IUC4
F032 A36-004
F007 A01-03: Forties crude oil
F008 E06-0010
F009 E07-03
F010 1981
F011 24
F012 E04-02
F013 E05-02
F020 A02-03
F021 42
F030 A03-01
F031 A03-03
F045 E35-02
EOR
F001 37
F002 7
F003 09-10-2003
F004 IUC4
F032 A36-002
F007 A01-03: Kuwait crude oil
F008 E06-0033: Holmesiysis costata
F009 E07-04: 40 CFR Part 797.1930, Mysid Shrimp Acute Test
F010 1993
F011 96
F012 E04-02
F013 E05-02
F026 LL50
F027 A02-03
F028 39.5

F030 A03-03
F031 A03-03
F047 E35-02
EOR
F001 37
F002 8
F003 09-10-2003
F004 IUC4
F032 A36-002
F007 A01-03: Kuwait crude oil
F008 E06-0023
F009 E07-04: 40 CFR Part 797.1930, Mysid Shrimp Acute Test
F010 1993
F011 96
F012 E04-02
F013 E05-02
F026 LL50
F027 A02-03
F028 618
F030 A03-03
F031 A03-03
F047 E35-02
EOB
C
B403 EC_ALGAETOX_TAB
F001 37
F002 1
F003 10-10-2003
F004 IUC4
F036 A36-002
F007 A01-03: CAS 8002-05-9 Crude oil - 0.5% paraffinic
F008 E08-0063: Macrocystis pyrifera
F010 2001
F012 48
F013 E04-02
F014 E05-02
F034 A03-03
F035 A03-03
F054 C47-001
EOR
F001 37
F002 2
F003 09-10-2003
F004 IUC4
F036 A36-002
F007 A01-03: CAS 8002-05-9 Crude oil 3% paraffinic
F008 E08-0063: Marine algae (Macrocystit pyrifera)
F009 E09-04: Chapman, G.A., D.L. Denton and J.M. Lazorchak. 1995
F010 2001
F012 48
F013 E04-02
F014 E05-02
F034 A03-03

F035 A03-03
F054 C47-001
EOR
F001 37
F002 3
F003 09-10-2003
F004 IUC4
F036 A36-002
F007 A01-03: Assam crude oil
F008 E08-0003
F009 E09-04: Procedure as detailed by Gaur and Singh (see Reference)
F010 1989
F011 E10-02
F012 15
F013 E04-01
F014 E05-02
F034 A03-03
F035 A03-02
EOB
C
B412 EC_OTHER_TAB
F001 37
F002 1
F003 10-10-2003
F004 IUC4
F008 A36-002
F009 Poorly water soluble mixtures
EOR
F001 37
F002 2
F003 10-10-2003
F004 IUC4
F008 A36-005
F009 Effect of crude oil on eggs
EOR
F001 37
F002 3
F003 01-10-2003
F004 IUC4
F008 A36-005
F009 Effect of crude oil on Lobster larvae
EOR
F001 37
F002 4
F003 09-10-2003
F004 IUC4
F008 A36-005
F009 Effect of crude and refined oil on fresh and sea water organisms
EOR
F001 37
F002 5
F003 01-10-2003
F004 IUC4

F008 A36-005
F009 Effect of crude oil on Sea urchin eggs
EOB
C
B501 TO_ACUTE_ORAL_TAB
F001 37
F002 1
F003 29-09-2003
F004 IUC4
F017 A36-003
F007 A01-03: Beryl crude (Light crude oil)
F008 T01-03
F009 T02-24
F011 1984
F012 A02-04
F014 5000
F015 T04-01
F016 A03-02
F019 T24-03
F020 5
F021 T52-003: None, undiluted
F022 T23-42
F023 Single dose: 5000 mg/kg
EOR
F001 37
F002 3
F003 28-09-2003
F004 IUC4
F008 T01-03
EOB
C
B503 TO_ACUTE_DERMAL_TAB
F001 37
F002 1
F003 29-09-2003
F004 IUC4
F017 A36-003
F007 A01-03: Beryl Crude (Light crude oil)
F008 T01-03
F009 T02-23
F011 1984
F012 A02-04
F014 2000
F015 T04-01
F016 A03-02
F019 T24-03
F020 3
F021 T52-003: None, undiluted
F022 T23-31
F023 Single dose: 2000 g/kg
EOR
F001 37
F002 2

F003 29-09-2003
F004 IUC4
F008 T01-03
EOB
C
B505 TO_SKIN_IRRITATION_TAB
F001 37
F002 1
F003 03-11-2003
F004 IUC4
F014 A36-002
F007 A01-03: Lost Hills Light Crude
F008 T02-23
F010 1985
F013 A03-02
F017 T49-001
F018 T50-001
F019 24
F020 T55-001
F021 6
F023 T52-003: Undiluted
EOR
F001 37
F002 2
F003 09-10-2003
F004 IUC4
F014 A36-003
F007 A01-03: Arab Light crude
F008 T02-23
F010 1984
F013 A03-02
F017 T49-001
F018 T50-001
F019 24
F020 T55-001
F021 6
F023 T52-003: Non, undiluted
EOR
F001 37
F002 3
F003 28-09-2003
F004 IUC4
F014 A36-003
F007 A01-03: Belridge heavy crude
F008 T02-23
F010 1997
F011 T15-04
F012 T46-06
F017 T49-001
F018 T50-001
F019 4
F020 T55-001
F022 1.9

EOB
C
B506 TO_EYE_IRRITATION_TAB
F001 37
F002 1
F003 22-05-2003
F004 IUC4
F014 A36-003
F007 A01-03: Beryl Crude (Light crude oil)
F008 T02-23
F010 1985
F017 T49-001
F018 .1
F019 T56-001
F022 6
F023 T51-002
EOR
F001 37
F002 2
F003 28-09-2003
F004 IUC4
F008 T02-23
EOB
C
B507 TO_SENSITIZATION_TAB
F001 37
F002 1
F003 29-09-2003
F004 IUC4
F015 A36-002
F007 A01-03: Lost Hills Light Crude
F008 T18-01
F009 T02-10
F011 1991
F013 T21-02
F014 A03-03
F017 20
F018 T53-001
F019 15
F020 T49-002
F021 T54-002
F022 T53-002
F023 15
F024 T49-002
F025 T54-002
F026 T53-002
F027 10
F028 T49-002
F029 T54-002
F030 T52-003: Mineral oil
EOR
F001 37
F002 2

F003 28-09-2003
F004 IUC4
F015 A36-003
F007 A01-03: Belridge heavy crude
F008 T18-01
F011 1991
F013 T21-02
F018 T53-001
F019 15
F020 T49-002
F022 T53-002
F023 10
F024 T49-002
F026 T53-002
F027 15
F028 T49-002
EOB
C
B508 TO_REPEATED_DOSE_TAB
F001 37
F002 1
F003 29-09-2003
F004 IUC4
F007 A01-03:Two crude oils
F008 T02-24
F009 T23-42
F010 T24-03
F011 T25-01
F013 1992
F015 5 days/week for 13 weeks
F017 30, 125 and 500 mg/kg/day
F018 T27-04
F029 A03-02
F032 C07-002
EOR
F001 37
F002 2
F003 29-09-2003
F004 IUC4
F030 A36-003
F007 A01-03: Prudhoe Bay Crude oil (Heavy crude oil)
F008 T02-18
F009 T23-10
F010 T24-03
F011 T25-03
F013 1990
F014 5 days
F015 Daily for five days
F017 2, 4, 8, 12 & 16 ml/kg/day
F018 T27-04
F029 A03-02
F032 C07-001
EOB

C
B509 TO_GENETIC_IN_VITRO_TAB
F001 37
F002 1
F003 09-10-2003
F004 IUC4
F016 A36-003
F017 1
F007 A01-03: Beryl Crude (LIght crude oil)
F008 T30-19: Modified Ames Assay
F009 T31-18
F010 1984
F011 Ames test of a DMSO extract of test substance
F012 T32-02
F014 A03-02
F015 1 to 50µl/plate
EOR
F001 37
F002 2
F003 09-10-2003
F004 IUC4
F016 A36-002
F017 2
F007 A01-03: Lost Hills Light crude
F008 T30-06
F010 1991
F011 Chinese Hamster Ovary cells
F012 T32-02
F013 T33-02
F014 A03-02
F015 1 to 20 µl DMSO extract/ml culture medium
EOR
F001 37
F002 3
F003 09-10-2003
F004 IUC4
F016 A36-002
F017 3
F007 A01-03: Belridge Heavy crude
F008 T30-06
F010 1991
F011 Chinese Hamster Ovary cells
F012 T32-02
F013 T33-02
F014 A03-02
F015 1 to 20 µl DMSO extract/ml culture medium
EOR
F001 37
F002 4
F003 29-09-2003
F004 IUC4
F016 A36-005
F017 4

F007 A01-03: South Louisiana crude (Light crude oil)
F008 T30-01
F010 1981
F012 T32-03
F014 A03-02
EOR
F001 37
F002 5
F003 09-10-2003
F004 IUC4
F016 A36-005
F017 5
F007 A01-03: Wilmington crude
F008 T30-01
F010 1982
F012 T32-03
F013 T33-02
F014 A03-02
EOR
F001 37
F002 6
F003 09-10-2003
F004 IUC4
F016 A36-003
F017 6
F007 A01-03: Wilmington crude oil
F008 T30-15
F010 1982
F011 Human lymphocytes
F014 A03-02
EOB
C
B510 TO_GENETIC_IN_VIVO_TAB
F001 37
F002 1
F003 29-09-2003
F004 IUC4
F018 A36-002
F007 A01-03: Lost Hills Light crude
F008 T34-07
F009 T02-24
F010 T23-42
F012 1990
F013 T24-03
F014 T25-01
F015 13 weeks
F016 30, 125 and 500 mg/kg/day
F017 A03-03
F020 T33-02
EOR
F001 37
F002 2
F003 09-10-2003

F004 IUC4
F018 A36-003
F007 A01-03: Wilmington crude
F008 T34-09
F009 T02-18
F010 T23-22
F012 1982
F013 T24-02
F014 T25-05
F016 1.8, 3.6 &7.2 g/kg
F017 A03-02
EOB
C
B511 TO_CARCINOGENICITY_TAB
F001 37
F002 1
F003 29-09-2003
F004 IUC4
F020 A36-003
F007 A01-03: Two crudes C & D
F008 T02-18
F009 T23-07
F010 T24-02
F011 T38-01
F014 18 months
F015 Twice weekly
F017 50 mg per application
F018 T27-07
F019 A03-02
F022 T33-03
EOR
F001 37
F002 2
F003 29-09-2003
F004 IUC4
F020 A36-003
F007 A01-03: Heavy crude oil, San Joaquin Valley
F008 T02-18
F009 T23-07
F010 T24-03
F011 T38-01
F013 1988
F014 105 weeks
F015 Three times weekly
F017 25 mg per application
F018 T27-07
F019 A03-02
EOB
C
B513 TO_DEVELOPMENTAL_TAB
F001 37
F002 1
F003 29-09-2003

F004 IUC4
F030 A36-002
F007 A01-03: Belridge Heavy Crude
F008 T02-24
F009 T23-42
F010 T24-01
F011 T25-01
F013 1991
F014 Days 0-19 of gestation
F016 Daily
F017 30, 125 and 500 mg/kg/day
F018 T27-04
F029 A03-02
EOR
F001 37
F002 2
F003 09-10-2003
F004 IUC4
F030 A36-002
F007 A01-03: Lost Hills Light
F008 T02-24
F009 T23-42
F010 T24-01
F011 T25-01
F013 1991
F014 Days 0-19 of gestation
F016 Daily
F017 125, 500 and 2000 mg/kg/day
F018 T27-04
F019 A02-03
F020 125
F022 T43-02
F023 A02-03
F024 500
F026 T43-02
F029 A03-02
EOR
F001 37
F002 3
F003 09-10-2003
F004 IUC4
F030 A36-003
F007 A01-03: Prudhoe Bay crude oil (Heavy crude oil)
F008 T02-24
F009 T23-42
F010 T24-01
F011 T25-03
F013 1987
F016 Various
F029 A03-02
EOB
C
B018 TO_REPRODUCTION_OTHER_TAB

F001 37
F002 1
F003 12-02-2003
F004 IUC4
F007 C37-001: Study for sperm abnormality
F008 C08-002
F009 T02-18
F010 T23-48: B6C3F1/Hap hybrid
F011 T24-02
F012 T25-05
F014 Once daily for 5 days
F015 40 days
F016 A01-03: Wilmington crude
F017 1.0 & 2.1 g/kg
F018 T27-07
F020 1982
F021 A03-02
EOB
C
B019 TO_SPEC_INVEST_TAB
F001 37
F002 1
F003 09-10-2003
F004 IUC4
F005 A36-005
F007 C11-004
F010 C40-001: Initiation/promotion assay
F011 A01-03: South Louisiana crude
F012 T02-18
F013 T23-48: Charles River (ChR-CD)
F014 T24-02
F015 Dermal
F016 30
F017 T52-003: undiluted
F018 180
F019 C35-003
F020 See method
F021 See method
F024 1981
F025 A03-02
F027 C53-006
EOB
C
B601 TEXT_TAB
F002 37
F010 1.1.1
F004 1
F005 RM
F006 The CAS definition for Petroleum (Crude oil) is:
**
** "A complex combination of hydrocarbons. It consists
** predominantly of aliphatic, alicyclic and aromatic
** hydrocarbons. It may also contain small amounts of

** nitrogen, oxygen and sulfur comp
F007 The CAS definition for Petroleum (Crude oil) is:
**
** "A complex combination of hydrocarbons. It consists
** predominantly of aliphatic, alicyclic and aromatic
** hydrocarbons. It may also contain small amounts of
** nitrogen, oxygen and sulfur compounds. This category
encompasses
* light, medium, and heavy petroleums, as well as the oils extracted
* from tar sands".
**
** Hydrocarbonaceous materials requiring major chemical
changes for
* their recovery or conversion to petroleum refinery feedstocks such as
* crude shale oils, upgrade shale oils and liquid coal
fuels are not
* included in this definition.
**
**
** Crude oil contains hydrocarbons in the carbon number range
** from C1 to C60+. It also contains organometallic complexes,
** notably of sulfur and vanadium, and dissolved gases such as hydrogen
* sulfide.
** Crude oils range from thin, light colored oils consisting
** mainly of gasoline-quality stock to heavy, thick tar-like
** materials.
** An "average crude oil has the following general composition:
** Carbon 84%
** Hydrogen 14%
** Sulfur 1-3%
** Nitrogen 1%
** Oxygen 1%
** Minerals and salts 0.1%
**
**
** The chemical composition of crude oils can vary tremendously from
* different producing regions and even from within a particular formation.
**
** Examples of compositions of various whole crudes are shown
** in the following table
**
** Crude Paraffins Naphthenes Aromatics Sulfur API
** source % vol. % vol. % vol. % wt. gravity
** (°API)
**
** Light crudes
** Saudi light 63 18 19 20 34
** South Louisiana
** 79 45 19 0 35
** Nigerian light
** 37 54 9 0.1 36
** North sea Brent
** 50 34 16 0.4 37

** Beryl
** 47 34 19 0.4 37
** Lost Hills
** Non-Aromatics 50% 50 0.9
**
** Heavy crudes
** Prudhoe Bay 27 36 28 0.9 28
** Saudi Heavy 60 20 15 2.1 28
** Venezuela Heavy
** 35 53 12 2.3 24
** Belridge Heavy
** Non-aromatics 37% 63 1.1 -
**
** Mid-range crudes
** Kuwait 63 20 24 2.4 31
** Venezuela Light
** 52 34 14 1.5 30
** USA West Texas Sour
** 46 32 22 1.9 32
**
**
** Crude oils may be categorized in either of several different ways e.g.
**
** Paraffinic vs naphthenic.
** Crude oils contain both paraffinic and naphthenic
** hydrocarbons but if there is a preponderance of parafffinic
** hydrocarbons present, the crude oil is referred to as a
** paraffinic crude. These crudes would be rich in straight and branched
* chain paraffins. Conversely a crude in which
** naphthenic hydrocarbons are predominant is referred to as a
** naphthenic crude. These crudes contain mainly naphthenic and aromatic
* hydrocarbons
**
** Sweet vs sour
** Crude oils may be referred to as either sweet or sour
** depending on the level of hydrogen sulfide present. A sweet crude has
* very little H2S whereas a sour crude has larger quantities of H2S present.
**
** Light vs heavy
**
** Crude oils may be divided into Light and Heavy crudes on the basis of
* their gravity.
** The API gravity is determined as:
** °API= 141.5/Specific gravity - 131.5
**
** Crude oils with gravity >33°API are considered as light
** crudes. Such crudes with a high percentage composition of
** hydrogen are usually more suitable for processing for
** gasoline production.
** Heavy crudes, ie those with gravity <28°API tend to contain
** more asphaltenes and are usually rich in aromatics. These
** heavy crudes require more steps in their processing.
**

**
** Information in this robust summary is presented for light
** and heavy crudes since this categorization distinguishes
** between crudes with a high paraffinic content (Light crudes) and those
* with a high aromatic/naphthenic content (Heavy crudes). This represents
* the extremes of the ranges of crudes available.
F020 4489
EOR
F002 37
F010 1.13
F004 1
F005 RE
F006 International Agency for Cancer (1989)
** Occupational exposures in petroleum refining; crude oil and
** major petroleum fuels
** IARC monograph 45, IARC, Lyon, France
F007 International Agency for Cancer (1989)
** Occupational exposures in petroleum refining; crude oil and
** major petroleum fuels
** IARC monograph 45, IARC, Lyon, France
F020 4490
EOR
F002 37
F010 1.13
F004 1
F005 RM
F006 IARC reviewed the evidence for carcinogenicity of crude oil
** to man and animals and published the result in 1989.
** IARC concluded that:
** There is inadequate evidence for the carcinogenicity in humans of crude
* oil
**
** There is limited evidence
F007 IARC reviewed the evidence for carcinogenicity of crude oil
** to man and animals and published the result in 1989.
** IARC concluded that:
** There is inadequate evidence for the carcinogenicity in humans of crude
* oil
**
** There is limited evidence for the carcinogenicity in experimental
* animals of crude oil
**
** The overall evaluation was
** Crude oil is not classifiable as to its carcinogenicity to humans
* (Group 3)
F020 4491
EOR
F002 37
F010 2.1
F004 1
F005 RE
F006 American Society for Testing and Materials (ASTM), (1991)
** Annual book of ASTM standards, Section 5, Petroleum

** products, Lubricants and Fossil Fuels
** ASTM, Philadelphia, Pa
F007 American Society for Testing and Materials (ASTM), (1991)
** Annual book of ASTM standards, Section 5, Petroleum
** products, Lubricants and Fossil Fuels
** ASTM, Philadelphia, Pa
F020 4492
EOR
F002 37
F010 2.1
F004 1
F005 RE
F006 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F007 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F020 4493
EOR
F002 37
F010 2.1
F004 1
F005 RM
F006 The figures quoted are a typical range for the drop point as measured by
* a standard oil industry procedure. For some low wax crudes, pour points
* below -30 °C are obtained.
F007 The figures quoted are a typical range for the drop point as measured by
* a standard oil industry procedure. For some low wax crudes, pour points
* below -30 °C are obtained.
F020 4494
EOR
F002 37
F010 2.14
F004 1
F005 RE
F006 Banks, R.E. and King, P.J. (1986)
** Chemistry and Physics of Petroleum.
** In: Modern Petroleum Technology, Part I, 5th edition,
** Hobson, G.D. (Ed), John Wiley, Chichester.
F007 Banks, R.E. and King, P.J. (1986)
** Chemistry and Physics of Petroleum.
** In: Modern Petroleum Technology, Part I, 5th edition,
** Hobson, G.D. (Ed), John Wiley, Chichester.
F020 4495
EOR
F002 37
F010 2.14
F004 1
F005 RM
F006 Petroleum is a natural organic material consisting mainly of
** hydrocarbons. It occurs in both the gaseous and liquid
** states in geological traps. The liquid phase, after being
** freed from dissolved gas and any associated salt water, is
** known

F007 Petroleum is a natural organic material consisting mainly of
** hydrocarbons. It occurs in both the gaseous and liquid
** states in geological traps. The liquid phase, after being
** freed from dissolved gas and any associated salt water, is
** known as crude oil. All the information presented in this
** Data Set relates to crude oil.
** Crude oil is not a uniform substance since its physical and
** chemical properties vary from oilfield to oilfield and can
** even vary within wells at the same oilfield. At one
** extreme, crude oil is a light, mobile, straw-colored liquid
** containing a large proportion of hydrocarbons which are
** readily distilled at atmospheric pressure. At the other
** extreme, crude oil is a highly viscous, semi-solid, black
** substance from which little can be distilled at atmospheric
** pressure before thermal decomposition occurs.
** The most consistent property of crude oils is their
** relatively small range of elemental composition, as the
** ranges in the following table show:
** element composition (wt%)
** carbon 83.9 - 86.8
** hydrogen 11.0 - 14.0
** sulfur 0.06 - 8.0
** nitrogen 0.02 - 1.7
** oxygen 0.08 - 1.82
** metals 0.00 - 0.14
**
** Crude oils are normally characterized in terms of three
** properties; density, viscosity and sulfur content. Crude
** oils are identified as either light (specific gravity
** <0.82), or medium (specific gravity 0.82 to 0.97), or heavy
** (specific gravity > 0.97). The viscosity is an expression
** of the mobility of the crude oil. The sulfur content has a
** marked influence on the refinery procedures to which the
** crude oil, and in particular its derivatives, will be
** subjected in order to produce acceptable products.
** Crude oils are also characterized in terms of their chemical composition,
* specifically on the predominance of the hydrocarbon types that are
* present. Modern practice tends to recognize two main types of crude,
* namely paraffinic and naphthenic. Paraffinic crude oils are rich in
* straight-chain and branched-chain alkanes, whereas in
** naphthenic crudes the main constituents are cycloparaffins
** and aromatic hydrocarbons. However, this is a simplified
** picture, as many crude oils fall between or outside these
** two types.
F020 4496
EOR
F002 37
F010 2.14
F004 1
F005 SO
F006 The technical information has been compiled by the Oil
** Companies' European Organization for Environmental Health
** Protection (CONCAWE), based at Madouplein-1, B-1210

** Brussel, Belgium, and this organization holds copies of the
** reference artic
F007 The technical information has been compiled by the Oil
** Companies' European Organization for Environmental Health
** Protection (CONCAWE), based at Madouplein-1, B-1210
** Brussel, Belgium, and this organization holds copies of the
** reference articles cited in this data set
F008 IUC4
F009 09-10-2003
F020 4497
EOR
F002 37
F010 2.2
F004 1
F005 RE
F006 American Society for Testing and Materials (ASTM), (1991)
** Annual book of ASTM standards, Section 5, Petroleum
** products, Lubricants and Fossil Fuels
** ASTM, Philadelphia, Pa
F007 American Society for Testing and Materials (ASTM), (1991)
** Annual book of ASTM standards, Section 5, Petroleum
** products, Lubricants and Fossil Fuels
** ASTM, Philadelphia, Pa
F020 4498
EOR
F002 37
F010 2.2
F004 1
F005 RE
F006 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F007 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F020 4499
EOR
F002 37
F010 2.2
F004 1
F005 RE
F006 Speight, J. G. (1990)
** Fuel science and technology handbook
** Marcel Dekker Inc., New York and Basel
F007 Speight, J. G. (1990)
** Fuel science and technology handbook
** Marcel Dekker Inc., New York and Basel
F020 4500
EOR
F002 37
F010 2.2
F004 1
F005 RM
F006 The method measures the distillation range of petroleum
** products. The figures quoted represent the approximate range

** for crude oils and are based on (a) the boiling point of
** n-butane for the lower value and (b) an upper limit quoted
** by Spei
F007 The method measures the distillation range of petroleum
** products. The figures quoted represent the approximate range
** for crude oils and are based on (a) the boiling point of
** n-butane for the lower value and (b) an upper limit quoted
** by Speight. In practice, atmospheric distillation of crude
** oil is not practiced above 275-300 °C, to avoid thermal
** decomposition of the residue. The residue is normally vacuum
** distilled in a subsequent operation.
F020 4501
EOR
F002 37
F010 2.4
F004 1
F005 RE
F006 Emergency Sciences Division (ESD) (1991)
** Experimental data, Environment Canada, Ottawa, ON.
F007 Emergency Sciences Division (ESD) (1991)
** Experimental data, Environment Canada, Ottawa, ON.
F008 IUC4
F009 13-11-2003
F020 4506
EOR
F002 37
F010 2.4
F004 1
F005 RE
F006 Environmental Emergencies Technology Division (EETD) (1984) Experimental
* data, Environment Canada, Ottawa, ON.
F007 Environmental Emergencies Technology Division (EETD) (1984) Experimental
* data, Environment Canada, Ottawa, ON.
F008 IUC4
F009 13-11-2003
F020 4508
EOR
F002 37
F010 2.4
F004 1
F005 RE
F006 Jokuty, P. S., Whiticar, Z., Fingas, M., Fieldhouse, B., Lambert, P. and
* Mullin, J. (2002)
** Properties of crude oils and oil products.
** Environmental protection service, Environment Canada, Ottowa, Ontario.
** Internet version: 2002 via http://w
F007 Jokuty, P. S., Whiticar, Z., Fingas, M., Fieldhouse, B., Lambert, P. and
* Mullin, J. (2002)
** Properties of crude oils and oil products.
** Environmental protection service, Environment Canada, Ottowa, Ontario.
** Internet version: 2002 via http://www.etcenter.org/spills
F020 4505
EOR

F002 37
F010 2.4
F004 1
F005 RE
F006 Oil and Gas Journal Databook (OGJ) (1999)
** Tulsa, OK.
F007 Oil and Gas Journal Databook (OGJ) (1999)
** Tulsa, OK.
F008 IUC4
F009 13-11-2003
F020 4507
EOR
F002 37
F010 2.4
F004 1
F005 RL
F006 Reliable with restrictions. Data obtained from a government (Environment
* Canada) database who sponsored the vapor pressure measurements cited by
* ESD and EETD.
F007 Reliable with restrictions. Data obtained from a government (Environment
* Canada) database who sponsored the vapor pressure measurements cited by
* ESD and EETD.
F020 4504
EOR
F002 37
F010 2.4
F004 1
F005 RM
F006 Vapor pressure measurements were provided for 10 petroleum crude oils
* originating from various locations throughout the world. The source of
* these data are from Environmet Canada Environmetal Technology Ceneter, a
* government-maintained da
F007 Vapor pressure measurements were provided for 10 petroleum crude oils
* originating from various locations throughout the world. The source of
* these data are from Environmet Canada Environmetal Technology Ceneter, a
* government-maintained database (Jokuty et al., 2002). Data cited by OGJ
* may be considered a secondary source, but the data cited by ESD and EETD
* were measured data sponsored by those agencies using a standardized
* method for measuring vapor pressure of petroleum products (1). As such,
* these 10 measurements provide a body of data adequate for use in the
* U.S. EPA HPV program and specific for the physical/chemical endpoint of
* vapor pressure.
**
** (1) ASTM D323, Standard Test Method for vapor Pressure of Petroleum
* Products (Reid Method)
F020 4503
EOR
F002 37
F010 2.4
F004 1
F005 RS
F006 Vapor Pressure, kPa See the following table and Remarks section
** Temperature °C Not stated

** Decomposition Not stated
**
** Vapor Pressures, kPa:
**
** Reid Vapor
** Product Pressure
** Name Origin Value, kPa Reference
** Alaska North Slope Alaska
F007 Vapor Pressure, kPa See the following table and Remarks section
** Temperature °C Not stated
** Decomposition Not stated
**
** Vapor Pressures, kPa:
**
** Reid Vapor
** Product Pressure
** Name Origin Value, kPa Reference
** Alaska North Slope Alaska USA 19 ESD 91
** Arabian Medium Saudi Arabia 22.1 OGJ 99
** Alif Yemen 45 OGJ 99
** Amna Libya 27 OGJ 99
** Ashtart Tunisia 13 OGJ 99
** Atkinson
** Beaufort Sea, Canada 6 ESD 91
** Alberta Sweet Mixed Blend
** Alberta, Canada 19 EETD 84
** Abu Al Bu Khoush
** United Arab Emirates 24 OGJ 99
** Beryl North Sea, UK 36 OGJ 99
** Bombay High India 33 OGJ 99
**
F020 4502
EOR
F002 37
F010 2.5
F004 1
F005 RE
F006 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F007 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F020 4509
EOR
F002 37
F010 2.5
F004 1
F005 RE
F006 Log Pow Database (1993)
** Pomona College
F007 Log Pow Database (1993)
** Pomona College
F020 4510
EOR

F002 37
F010 2.5
F004 1
F005 RM
F006 The calculation was done by the CLOGP Version 3.5 program
** (calculation of Log partition coefficient octanol/water).
** The figures represent the spread of calculated and/or
** measured values for typical hydrocarbon components of crude
** oil. Calcu
F007 The calculation was done by the CLOGP Version 3.5 program
** (calculation of Log partition coefficient octanol/water).
** The figures represent the spread of calculated and/or
** measured values for typical hydrocarbon components of crude
** oil. Calculated values for higher molecular weight
** hydrocarbons will be above 6, but such values are notional,
** since no correlation has been established between calculated
** and experimental values.
F020 4511
EOR
F002 37
F010 2.6.1
F004 1
F005 CL
F006 Limited detail is provided for exact amounts of crude oil used for
* preparing aqueous solutions, nor is there any information regarding the
* composition of each of the crude oils tested, either as hydrocarbon type
* or inorganic components (suc
F007 Limited detail is provided for exact amounts of crude oil used for
* preparing aqueous solutions, nor is there any information regarding the
* composition of each of the crude oils tested, either as hydrocarbon type
* or inorganic components (such as sulfur). Also, no information on the GC
* calibration standard composition used to identify and quantify soluble
* components in the equilibrated aqueous -oil solutions is provided.
* Individual components of complex petroleum substances have specific and
* differing solubilities. 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.
F020 4512
EOR
F002 37
F010 2.6.1
F004 1
F005 ME
F006 Individual saturated crude oil solutions were prepared by adding

* approximately 10 ml of the respective oil to 50-100 ml of water in 125 ml
* separatory funnels. Funnels were gently shaken for at least 48 hrs
* either with a magnetic stir bar o
F007 Individual saturated crude oil solutions were prepared by adding
* approximately 10 ml of the respective oil to 50-100 ml of water in 125 ml
* separatory funnels. Funnels were gently shaken for at least 48 hrs
* either with a magnetic stir bar or with a wrist action shaker, then
* placed in a temperature bath for at least 48 hr prior to analysis.
* Solubility at 5, 20 and 22 (±2C) °C was determined in both double
* distilled water and salt water (3% NaCl). The effect of water to oil
* ratio on the solubility of crude oil components was determined by
* injecting oil into sealed vials completely filled with water, and mixed
* at low speed for an equilibration period of 20 days. Analysis was
* determined by purge-and-trap/GC; solid sorbent extraction with HPLC and
* fluorescence analysis.
F020 4514
EOR
F002 37
F010 2.6.1
F004 1
F005 RE
F006 CONCAWE (1992)
** Ecotoxicological Testing Of Petroleum Products:
** Test Methodology.
** Report 92/56
** Brussels.
F007 CONCAWE (1992)
** Ecotoxicological Testing Of Petroleum Products:
** Test Methodology.
** Report 92/56
** Brussels.
F008 IUC4
F009 30-09-2003
F020 4516
EOR
F002 37
F010 2.6.1
F004 1
F005 RE
F006 CONCAWE (1996)
** Environmental risk assessment of petroleum substances:
** the hydrocarbon block method.
** Report 96/52
** Brussels.
F007 CONCAWE (1996)
** Environmental risk assessment of petroleum substances:
** the hydrocarbon block method.
** Report 96/52
** Brussels.
F020 4515
EOR
F002 37
F010 2.6.1

F004 1
F005 RE
F006 ECETOC (1996)
** Aquatic Toxicity Testing of Sparingly Soluble, Volatile and Unstable
* Substances.
** Monograph 26.
F007 ECETOC (1996)
** Aquatic Toxicity Testing of Sparingly Soluble, Volatile and Unstable
* Substances.
** Monograph 26.
F020 4517
EOR
F002 37
F010 2.6.1
F004 1
F005 RE
F006 ECETOC (1998)
** QSARS in the Assessment of the Environmental Fate and Effects of
* Chemicals.
** Technical Report No. 74.
F007 ECETOC (1998)
** QSARS in the Assessment of the Environmental Fate and Effects of
* Chemicals.
** Technical Report No. 74.
F020 4518
EOR
F002 37
F010 2.6.1
F004 1
F005 RE
F006 McAuliffe, C. (1966)
** Solubility in water of paraffin, cycloparaffin, olefin, acetylene,
* cycloolefin and aromatic hydrocarbons.
** J. Phys. Chem. vol. 70, No 4, pp 1267-1275
F007 McAuliffe, C. (1966)
** Solubility in water of paraffin, cycloparaffin, olefin, acetylene,
* cycloolefin and aromatic hydrocarbons.
** J. Phys. Chem. vol. 70, No 4, pp 1267-1275
F020 5445
EOR
F002 37
F010 2.6.1
F004 1
F005 RE
F006 Shiu, et al. Bridie, A.L. and Bos, J. (1990)
** The Water Solubility of Crude Oils and Petroleum Products. Oil & Chemical
* Pollution 7, 1990, 57-84.
F007 Shiu, et al. Bridie, A.L. and Bos, J. (1990)
** The Water Solubility of Crude Oils and Petroleum Products. Oil & Chemical
* Pollution 7, 1990, 57-84.
F008 IUC4
F009 13-11-2003
F020 4519

EOR
F002 37
F010 2.6.1
F004 1
F005 RS
F006 Fluorescence/HPLC analysis was inadequate in quantification of
* hydrocarbon composition.
** Purge and trap/GC analysis results based on total benzene, toluene, ethyl
* benzene+xylenes (combined concentration) and naphthalenes (BTE+XN as
* mg/L) a
F007 Fluorescence/HPLC analysis was inadequate in quantification of
* hydrocarbon composition.
** Purge and trap/GC analysis results based on total benzene, toluene, ethyl
* benzene+xylenes (combined concentration) and naphthalenes (BTE+XN as
* mg/L) are reported along with viscosity (vis=c.p @ 20C) and density
* (d=g/cm³ @ 20°C) for each oil at respective temperatures (°C) in
* distilled and saltwater (22° reported as ± 2°C)
**
* BTE+XN (mg/l)

** Oil	Distilled water	Saltwater
** Norman Wells		
** 4.41 vis; 0.832 d		
**	30 (5)	25.5 (5)
**	29-33 (20)	
**	31.8-33.5 (22)	20 (22)
** 4.7 vis; 0.84 d	33 (20)	14.8 (5)
** Alberta: 4.98 vis; 0.835 d	25.02(22)	
** Swan Hills: (no vis. density)	35.1 (22)	
** Prudhoe Bay: 34.9 vis; 0.901 d	29.01 (22)	
** Lago Medio: 41.1 vis; 0.872 d	23.66-25.5 (22)	16.47 (22)
** Kopanoar: 17.5 vis; 0.9 d	10.42 (22)	
** Murban: 4.37 vis; 0.824 d	28.62 (22)	
** Hibernia B 33.2 vis; 0.837d		16.92 (22)
** Hibernia J (no vis., density)		7.75 (22)
** Mobil A (no vis., density)	29.6 (22)	
** Mobil B (no vis., density)	58 (22)	

**
** Results for oil-water ratio testing were not quantified, but general
* observations were stated as follows: concentrations of the water soluble
* fractions decreases as the water-to oil ratio increases and the
* composition of the water soluble fraction changes as the ratio changes.
* At low water to oil ratios, the WSF is composed predominantly (80%) of
* BTEX. As the water to oil ratio increases, these compounds account for a
* smaller proportion of the dissolved compounds. At a water/oil ratio of
* 10000, these compounds account for only 15-30% of the total WSF.
F020 4513

EOR
F002 37
F010 3.1.1
F004 1
F005 RE
F006 Atkinson, R. (1990)
** Gas-phase tropospheric chemistry of organic compounds: a
** review
** Atmos. Environ. vol 24A, pp 1-41
F007 Atkinson, R. (1990)
** Gas-phase tropospheric chemistry of organic compounds: a
** review
** Atmos. Environ. vol 24A, pp 1-41
F020 4520
EOR
F002 37
F010 3.1.1
F004 1
F005 RE
F006 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F007 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F020 4521
EOR
F002 37
F010 3.1.1
F004 1
F005 RM
F006 Atkinson gives data which enables half lives to be
** calculated for the degradation of hydrocarbons in contact
** with hydroxyl radicals in the troposphere, under the
** influence of sunlight. Values for typical hydrocarbon
** constituents of crude o
F007 Atkinson gives data which enables half lives to be
** calculated for the degradation of hydrocarbons in contact
** with hydroxyl radicals in the troposphere, under the
** influence of sunlight. Values for typical hydrocarbon
** constituents of crude oils are as follows:
** Constituent Half-life
** (days)
** benzene 6.5
** n-butane 3.2
** n-hexane 1.4
** toluene 1.3
** cyclohexane 1.1
** n-decane 0.69
** n-tetradecane 0.42
** naphthalene 0.37
** Hydrocarbons of carbon number greater than C20 will have
** little or no tendency to partition to air (see
** Sub-chapter3.2.2).
F020 4522

EOR
F002 37
F010 3.1.2
F004 1
F005 RE
F006 Harris, J. C. (1982)
** Rate of hydrolysis
** In: Handbook of chemical property estimation methods,
** Environmental behaviour of organic chemicals
** Chapter 7
** Lyman, W. J., Reehl,W. F. and Rosenblatt, D. H. (Eds)
** McGraw-Hill, New York
F007 Harris, J. C. (1982)
** Rate of hydrolysis
** In: Handbook of chemical property estimation methods,
** Environmental behaviour of organic chemicals
** Chapter 7
** Lyman, W. J., Reehl,W. F. and Rosenblatt, D. H. (Eds)
** McGraw-Hill, New York
F020 5443
EOR
F002 37
F010 3.1.2
F004 1
F005 RM
F006 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 alkyl
* halides, amides, carbamates, car
F007 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 alkyl
* halides, amides, carbamates, carboxylic acid esters and lactones,
* epoxides, phosphate esters and sulfonic acid esters. The chemical
* components found in the materials that comprise the crude oil category
* are hydrocarbons that are not subject to hydrolysis because they lack
* functional groups that hydrolyze.
F020 5442
EOR
F002 37
F010 3.3.2
F004 1
F005 RE
F006 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F007 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F020 4523
EOR
F002 37
F010 3.3.2
F004 1
F005 RE

F006 Mackay, D., Patterson,S. and Shiu, W.Y. (1992)
** Generic models for evaluating the regional fate of chemicals
** Chemosphere Vol 24, pp 695-717
F007 Mackay, D., Patterson,S. and Shiu, W.Y. (1992)
** Generic models for evaluating the regional fate of chemicals
** Chemosphere Vol 24, pp 695-717
F020 4524
EOR
F002 37
F010 3.3.2
F004 1
F005 RM
F006 Distribution has been calculated according to Mackay Level I
** using the parameters defined in a paper by van der Zandt and
** van Leeuwen.
** The lower molecular weight components will mainly partition
** to air, with a maximum of about 1% of mono-ar
F007 Distribution has been calculated according to Mackay Level I
** using the parameters defined in a paper by van der Zandt and
** van Leeuwen.
** The lower molecular weight components will mainly partition
** to air, with a maximum of about 1% of mono-aromatic
** hydrocarbons partitioning to water. As the molecular
** weights increase, there is less tendency for the
** hydrocarbons to partition to air, the environmental
** distribution being preferentially to soil and sediment.
** Collectively, the wide molecular weight range of the
** hydrocarbons in crude oil will mean that at equilibrium,
** distribution will be mainly to air and soil, with much less
** than 1.0% being present in water.
F020 4525
EOR
F002 37
F010 3.3.2
F004 1
F005 RS
F006 Results for percent distribution of typical hydrocarbons
** found in crude oils are shown in tabular form as follows:
** air water soil sediment susp.matter biota
** propane
** 100.0 0.0 0.0 0.0 0.0
F007 Results for percent distribution of typical hydrocarbons
** found in crude oils are shown in tabular form as follows:
** air water soil sediment susp.matter biota
** propane
** 100.0 0.0 0.0 0.0 0.0 0.0
** n-butane
** 100.0 0.0 0.0 0.0 0.0 0.0
** n-hexane
** 100.0 0.0 0.0 0.0 0.0 0.0
** n-octane
** 99.9 0.0 0.1 0.0 0.0 0.0
** n-decane

** 99.1 0.0 0.9 0.0 0.0 0.0
** n-tetradecane
** 76.7 0.0 22.8 0.5 0.0 0.0
** n-eicosane
** 0.0 0.0 97.7 2.2 0.1 0.0
** cyclohexane
** 99.9 0.0 0.1 0.0 0.0 0.0
** benzene
** 98.8 1.1 0.1 0.0 0.0 0.0
** toluene
** 98.4 1.0 0.6 0.0 0.0 0.0
** p-xylene
** 97.8 1.0 1.2 0.0 0.0 0.0
** o-xylene
** 97.4 1.2 1.4 0.0 0.0 0.0
** ethylbenzene
** 98.2 0.8 1.0 0.0 0.0 0.0
** n-butylbenzene
** 91.0 0.4 8.5 0.2 0.0 0.0
F020 4526
EOR
F002 37
F010 3.6
F004 1
F005 ME
F006 Tests were run at 30 °C in fresh water using a respirometric
** procedure with crude oil at 50 and 70 mg/l. Prior to the
** test, the crude was "topped" at up to 100 °C to remove light
** ends, which comprised about 10% of the material
F007 Tests were run at 30 °C in fresh water using a respirometric
** procedure with crude oil at 50 and 70 mg/l. Prior to the
** test, the crude was "topped" at up to 100 °C to remove light
** ends, which comprised about 10% of the material
F020 4527
EOR
F002 37
F010 3.6
F004 1
F005 RE
F006 Bridie, A. L. and Bos,J. (1971)
** Biological degradation of mineral oil in sea water.
** J. Inst. Pet. Vol 57, pp 270-277
F007 Bridie, A. L. and Bos,J. (1971)
** Biological degradation of mineral oil in sea water.
** J. Inst. Pet. Vol 57, pp 270-277
F008 IUC4
F009 28-08-2003
F020 4528
EOR
F002 37
F010 3.6
F004 1
F005 RE

F006 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F007 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F020 4529
EOR
F002 37
F010 3.6
F004 1
F005 RS
F006 The BOD/TOD ratio after 10 days was 0.3 at 50 mg/l and 0.04
** at 70 mg/l.
** With the addition of nutrients, using ammonia at up to 4.6
** mg/l of nitrogen and phosphate at up to 15 mg/l of
** phosphorus, the BOD/TOD ratio increased to a maximum of 0.
F007 The BOD/TOD ratio after 10 days was 0.3 at 50 mg/l and 0.04
** at 70 mg/l.
** With the addition of nutrients, using ammonia at up to 4.6
** mg/l of nitrogen and phosphate at up to 15 mg/l of
** phosphorus, the BOD/TOD ratio increased to a maximum of 0.3
** after 5 days , and 0.34 after 10 days. In further studies,
** run at 10 °C in sea water in the presence of nutrients, the
** degradation rate was found to be lowered by a factor of 2 to
** 3 below that at 30 °C. The reference quotes the BOD5/TOD
** ratio, but does not give the individual values for BOD5 and
** TOD.
F020 4530
EOR
F002 37
F010 3.8
F004 2
F005 RE
F006 Atlas, R.M. and Bartha, R. (1973)
** Fate and effects of polluting petroleum in the marine
** environment.
** In: Residue Reviews of Pesticides and Other Contaminants in
** the TotalEnvironment, vol. 49, pp. 49-85
F007 Atlas, R.M. and Bartha, R. (1973)
** Fate and effects of polluting petroleum in the marine
** environment.
** In: Residue Reviews of Pesticides and Other Contaminants in
** the TotalEnvironment, vol. 49, pp. 49-85
F020 4533
EOR
F002 37
F010 3.8
F004 2
F005 RE
F006 Clark, R.B., (1989)
** Marine Pollution.
** 2nd Edition. Clarendon Press, Oxford, pp. 33-64
F007 Clark, R.B., (1989)
** Marine Pollution.

** 2nd Edition. Clarendon Press, Oxford, pp. 33-64
F020 4534
EOR
F002 37
F010 3.8
F004 2
F005 RE
F006 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F007 CONCAWE (2000)
** ECB IUCLID dataset for crude oil
F020 4535
EOR
F002 37
F010 3.8
F004 2
F005 RE
F006 CONCAWE (2001)
** Environmental classification of petroleum substances - summary data and
* rationale
** Report No. 01/54
** CONCAWE, Brussels
F007 CONCAWE (2001)
** Environmental classification of petroleum substances - summary data and
* rationale
** Report No. 01/54
** CONCAWE, Brussels
F020 5444
EOR
F002 37
F010 3.8
F004 2
F005 RE
F006 Zobell, C.E. (1963)
** The occurrence, effects and fate of oilpolluting the sea,
** Int. J. Air Wat. Poll., vol. 7, pp. 173-198
F007 Zobell, C.E. (1963)
** The occurrence, effects and fate of oilpolluting the sea,
** Int. J. Air Wat. Poll., vol. 7, pp. 173-198
F020 4536
EOR
F002 37
F010 3.8
F004 2
F005 RE
F006 Zobell, C.E. (1969)
** Microbial modification of crude oil in the sea.
** In: Joint Conference on Prevention and Control of Oil
** Spills.
** Proceedings of API-FWPCA Meeting, December15-17, New York.
** American Petroleum Institute publication, pp.
F007 Zobell, C.E. (1969)
** Microbial modification of crude oil in the sea.

** In: Joint Conference on Prevention and Control of Oil
** Spills.
** Proceedings of API-FWPCA Meeting, December15-17, New York.
** American Petroleum Institute publication, pp. 317ff
F020 4537
EOR
F002 37
F010 3.8
F004 2
F005 RM
F006 The world production of crude oil per year is of the order
** of 3 billion tonnes per year, of which about half is
** transported by sea. In his 1989 publication, Clark states
** that the best estimate of petroleum hydrocarbons entering the sea per
F007 The world production of crude oil per year is of the order
** of 3 billion tonnes per year, of which about half is
** transported by sea. In his 1989 publication, Clark states
** that the best estimate of petroleum hydrocarbons entering the sea per
* year is about 3 million tonnes, of which about one million tonnes is
* attributable to crude oil. Such pollution arises from the cleaning of
* oil tanker compartments, offshore oil production operations, discharge of
* coastal refinery effluent and spillage from oil tankers. Crude oil also
* enters the oceans by natural seepage from undersea locations.
** Particular attention has focused on major oil tanker
** spillages, notably involving the Torrey Canyon in 1969, the
** Amoco Cadiz in 1978 and the Exxon Valdez in 1990. As a
** result, the processes determining the fate of oil in
** seawater are reasonably well understood and have been
** reviewed by Atlas and Bartha.
** Initially, the oil spreads out as a film on the sea surface
** as a result of wind and wave action. The more volatile,
** lower molecular weight hydrocarbons are lost by evaporation.
** Polar compounds and the mono-aromatic hydrocarbons have an
** appreciable water solubility and are taken into solution. A key
* ancillary process is that of emulsification, since
** crude oil has a natural tendency to form emulsions in sea
** water. Such emulsions are usually of the oil-in-water type, but may also
* be of the water-in-oil type. The latter are often of the intractable
* 'chocolate mousse' type.
** Significant amounts of crude oil, particularly the higher
** molecular weight compounds, sink naturally, rolling along
** the ocean bottom picking up sand and shells and forming
** tarry balls which are resistant to degradation by any
** method. Hydrocarbons may also reach the bottom sediments by sorption
* onto suspended particles which ultimately settle on the sea floor.
** Spilt oil also undergoes chemical changes, particularly
** oxidation by free radical mechanisms initiated by sunlight.
** The initial products of such reactions are hydroperoxides,
** and these in turn form compounds such as alcohols, acids and aldehydes,
* many of which have an appreciable water
** solubility. Polymerization also occurs to yield intractable tarry
* materials.
** The bulk of spilt crude oil is biodegraded by the

** micro-organisms present in sea water. Emulsification to
** form oil-in-water emulsions yields small particles of crude
** oil that are biodegraded by bacteria, yeasts, fungi and
** actinomycetes. Many factors influence the rate of
** biodegradation, in particular temperature, dissolved oxygen
** concentration and the availability of nitrogen and
** phosphorus nutrients. Adapted micro-organisms are often
** found in ocean areas where crude oil spills are common.
** Zobell has calculated that where an adapted microbial
** population is available in well-aerated sea water at 20 to
** 30 °C, the rate of crude oil oxidation ranges from 0.02 to
** 0.2 g of oil oxidized/m²/day. The same author found
** experimentally that complete oxidation of 1.0 mg of
** hydrocarbon requires between 3 and 4 g of oxygen, i.e. it
** has a BOD of 3 to 4 mg oxygen/mg. Since the oxygen content
** of sea water is between 6 and 11 mg/liter, depending on
** salinity and temperature, this means that about 320 000
** liters of sea water is required to oxidize one liter of
** crude oil.
** Crude oil contains hydrocarbons of well-defined generic
** types that are biodegraded at different rates. n-Alkanes
** are readily degraded in sea water, since many
** micro-organisms can utilize them. Branched-chain or
** iso-alkanes are less readily biodegraded but they do
** ultimately biodegrade. The degradation of cycloalkanes has
** not been extensively studied, but the ring structure is
** resistant to biodegradation. Aromatic hydrocarbons are also resistant to
* biodegradation, but a few micro-organisms are able to utilize them. High
* molecular weight compounds, the tars and asphaltenes, degrade very slowly.
F020 4538
EOR
F002 37
F010 4.1
F004 1
F005 ME
F006 LL 50 at 96 hr calculated using the Trimmed Spearman-Karber
** Method (Hamilton, et al, 1977)
F007 LL 50 at 96 hr calculated using the Trimmed Spearman-Karber
** Method (Hamilton, et al, 1977)
F020 4539
EOR
F002 37
F010 4.1
F004 1
F005 RE
F006 ExxonMobil Biomedical Sciences Inc. (2002)
** Fish - acute toxicity test: Study No. 121258, test substance
** MRD-01-02. 2002.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.
F007 ExxonMobil Biomedical Sciences Inc. (2002)
** Fish - acute toxicity test: Study No. 121258, test substance
** MRD-01-02. 2002.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.

F020 4540
EOR
F002 37
F010 4.1
F004 1
F005 RS
F006 Mortality (no. of deaths/treatment) at 96 hrs: 0, 0, 0, 0, 5
** and 10, respectively for control, 1.4, 3.2, 8.5, 21 and 50
** mg/l treatments.
** 96-hr LL50 = 21 mg/l, 95% C.I: 16-28 mg/l (as nominal
** loading rate)
** 96-hour No Observed Effect Loadin
F007 Mortality (no. of deaths/treatment) at 96 hrs: 0, 0, 0, 0, 5
** and 10, respectively for control, 1.4, 3.2, 8.5, 21 and 50
** mg/l treatments.
** 96-hr LL50 = 21 mg/l, 95% C.I: 16-28 mg/l (as nominal
** loading rate)
** 96-hour No Observed Effect Loading (NOEL) was 8.5 mg/l
** (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.
** Measured BTEX (mg/l) in test treatment (mg/l load)

** Day	Control	1.4	3.2	8.5	21	50
** 0(new)	ND	0.129	0.382	0.835	1.94	4.73
** 1 (old) ND	0.086	0.243	0.665	1.60	4.04	
** 3 (new) 0.005	0.148	0.301	0.973	1.86	NA	
** 4 (old) 0.005	0.127	0.255	0.813	1.70	NA	

**
** ND=not detected, NA=not analyzed due to 100% mortality
** The Practical Quantitation Limit (PQL) was approximately
** 0.0035 mg/l (3.5ng/ml) which corresponds to the
** concentration of the lowest standard analyzed.
F020 4541
EOR
F002 37
F010 4.1
F004 1
F005 TC
F006 Test solutions were prepared as water accommodated fractions
** (WAF). The control and dilution water was a reconstituted
** moderately hard water aerated prior to use. Water quality
** analyses for the dilution water was as follows: alkalinity =
** 72
F007 Test solutions were prepared as water accommodated fractions
** (WAF). The control and dilution water was a reconstituted
** moderately hard water aerated prior to use. Water quality
** analyses for the dilution water was as follows: alkalinity =
** 72 mg/l as CaCO3 , hardness =96 mg/l (as CaCO3), specific
** conductance = 320 µmhos, pH = 7.7 and dissolved oxygen = 8.6
** ppm.
** Based on results of range-finding tests for a similar
** petroleum product, measured loading rates of 1.4, 3.2, 8.5,

** 21 and 50 mg/l of crude oil to water were used to prepare
** test solutions for definitive toxicity tests. Test
** substance, added volumetrically, was mixed for each
** individual treatment in dilution water for 24 hours in
** 12-liter stoppered containers with approximately 20% vortex
** and less than 10% headspace volume. The WAFs were allowed to
** settle and cool to test temperature for 1-2 hours prior to
** drawing off the aqueous solutions for testing. Fish were 85
** days old at test initiation and were obtained from Pierce
** Associates Inc., West Buxton, ME, Lot 454. Loading of fish
** body mass to treatment was 0.551 g fish per liter of aqueous
** solution, mean length at termination was 4.1 cm (sd=0.2),
** and mean weight was 0.496 g (sd=0.078). Test vessels were
** 4-liter glass aspirator bottles with Teflon® covered
** neoprene stoppers. Two 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 at least
** 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 BTEX. Mean water temperature range was
** 13.8 (sd=0.1) °C. Test photoperiod was 16 hrs. light and 8
** hr dark, dissolved oxygen measurements ranged from 6.5 to
** 8.8 ppm, pH values between 6.8 and 7.8.
F020 4542
EOR
F002 37
F010 4.1
F004 2
F005 ME
F006 LL 50 at 96 hr calculated using the Trimmed Spearman-Karber
** Method (Hamilton, et al, 1977)
F007 LL 50 at 96 hr calculated using the Trimmed Spearman-Karber
** Method (Hamilton, et al, 1977)
F020 4543
EOR
F002 37
F010 4.1
F004 2
F005 RE
F006 ExxonMobil Biomedical Sciences Inc. (2002)
** Fish - acute toxicity test: Study No. 101158, test substance
** MRD-01-01.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.
F007 ExxonMobil Biomedical Sciences Inc. (2002)
** Fish - acute toxicity test: Study No. 101158, test substance
** MRD-01-01.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.
F020 4544
EOR
F002 37

F010 4.1
F004 2
F005 RS
F006 Mortality (no. of deaths/treatment) at 96 hrs: 0, 0, 0, 0, 5
** and 10, respectively for control, 2.7, 6.8, 16, 40 and 109
** mg/l treatments.
** 96-hr LL50 = 41 mg/l, 95% C.I: 30-55 mg/l (as nominal
** loading rate)
** 96-hour No Observed Effect Loadin
F007 Mortality (no. of deaths/treatment) at 96 hrs: 0, 0, 0, 0, 5
** and 10, respectively for control, 2.7, 6.8, 16, 40 and 109
** mg/l treatments.
** 96-hr LL50 = 41 mg/l, 95% C.I: 30-55 mg/l (as nominal
** loading rate)
** 96-hour No Observed Effect Loading (NOEL) was 16 mg/l
** (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.
** Measured BTEX (mg/l) in test treatment
** (mg/l load)
** Day Control 2.7 6.8 16 40 109
** 0 (new) ND 0.093 0.366 0.541 1.36 2.02
** 1 (old) 0.050 0.066 0.184 0.411 0.857 1.84
** 3 (new) ND 0.096 0.272 0.585 1.28 2.01
** 4 (old) 0.031 0.084 0.221 0.484 1.03 NA
**
** ND=not detected, NA=not analyzed due to 100% mortality
** Guideline/protocol deviations: DO level was 53%, not 60% of
** saturation.
F020 4545
EOR
F002 37
F010 4.1
F004 2
F005 TC
F006 Test solutions were prepared as water accommodated fractions
** (WAF). The control and dilution water was a reconstituted
** moderately hard water aerated prior to use. Water quality
** analyses for the dilution water was as follows: alkalinity =
** 78
F007 Test solutions were prepared as water accommodated fractions
** (WAF). The control and dilution water was a reconstituted
** moderately hard water aerated prior to use. Water quality
** analyses for the dilution water was as follows: alkalinity =
** 78 mg/l as CaCO3 , hardness =105 mg/l (as CaCO3), specific
** conductance = 324 µmhos, pH = 7.6 and dissolved oxygen = 8.4
** ppm.
** Nominal loading rates of 0, 3.8, 16 and 74 mg/l were used
** to prepare test solutions for range-finding toxicity tests.
** Based on results of range-finding tests, measured loading
** rates of 0, 2.7, 6.8, 16, 40 and 109 mg/l of crude oil to
** water were used to prepare test solutions for definitive

** toxicity tests. Test substance, added volumetrically, was
** mixed for each individual treatment in dilution water for 24
** hours in 13-liter stoppered containers with approximately
** 20% vortex and less than 10% headspace volume. The WAFs were
** allowed to settle and cool to test temperature for 1-2 hours
** prior to drawing off the aqueous solutions for testing. Fish
** were 85 days old at test initiation and were obtained from
** Pierce Associates Inc., West Buxton, ME, Lot 454. Loading of
** fish body mass to treatment was 0.4 g fish per liter of
** aqueous solution, mean length at termination was 3.9 cm
** (sd=0.2), and mean weight was 0.466 g (sd=0.034). Test
** vessels were 4-liter glass aspirator bottles with Teflon®
** covered neoprene stoppers. Two 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 at least
** 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 BTEX. Water temperature range was 14.3
** to 14.9 °C. Test photoperiod was 16 hrs. light and 8 hr
** dark, dissolved oxygen measurements ranged from 5.4 to 8.8
** ppm, pH values between 6.8 and 7.8.
F020 4546
EOR
F002 37
F010 4.1
F004 3
F005 ME
F006 The test species was the Rainbow Trout.
** Crude oil was added to water at 8 concentrations in the range 57 to 1330
* mg/l in open vessels fitted with shielded propeller stirrers. A constant
* rate of stirring was maintained during the 7-day expo
F007 The test species was the Rainbow Trout.
** Crude oil was added to water at 8 concentrations in the range 57 to 1330
* mg/l in open vessels fitted with shielded propeller stirrers. A constant
* rate of stirring was maintained during the 7-day exposure period to the
* fish.
F020 4547
EOR
F002 37
F010 4.1
F004 3
F005 RE
F006 Westlake, G.E.(1991
** Aquatic test media preparation for low solubility materials, Proceedings
* of the Society of Environmental Toxicology and Chemistry (SETAC)
* Conference on Environmental Sciences and Sustainable Development,
* University of Sh
F007 Westlake, G.E.(1991
** Aquatic test media preparation for low solubility materials, Proceedings
* of the Society of Environmental Toxicology and Chemistry (SETAC)

* Conference on Environmental Sciences and Sustainable Development,
* University of Sheffield, April 1991.
F020 4549
EOR
F002 37
F010 4.1
F004 3
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. Since
* significant evaporative losses of lower hydrocarbons may have occurred,
* the LC50 values are expected to
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. Since
* significant evaporative losses of lower hydrocarbons may have occurred,
* the LC50 values are expected to be lower than those cited. These data are
* also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by API crude oil task group as '3', not reliable.
F020 4550
EOR
F002 37
F010 4.1
F004 3
F005 RS
F006 The results were as follows:
**
** 24-hour LC50 = 557 mg/l
** 96-hour LC50 = 258 mg/l
** 168-hour LC50 = 253 mg/l
**
** As the tests were run in open vessels, significant
** evaporative losses of lower hydrocarbons will have occurred
** and h
F007 The results were as follows:
**
** 24-hour LC50 = 557 mg/l
** 96-hour LC50 = 258 mg/l
** 168-hour LC50 = 253 mg/l
**
** As the tests were run in open vessels, significant
** evaporative losses of lower hydrocarbons will have occurred
** and hence the measured LC50 values underestimate the true
** ecotoxicity.
F020 4548
EOR
F002 37
F010 4.1
F004 3
F005 SO
F006 CONCAWE Bruxelles
F007 CONCAWE Bruxelles
F008 IUC4
F009 09-10-2003

F020 4571
EOR
F002 37
F010 4.1
F004 4
F005 ME
F006 The test species was the Rainbow Trout. Dispersions of oil in water at 8
* concentrations in the range 57 to 1330 mg/l were prepared by shaking the
* constituents together in closed jars for 5 minutes using a reciprocating
* table. Fish tests we
F007 The test species was the Rainbow Trout. Dispersions of oil in water at 8
* concentrations in the range 57 to 1330 mg/l were prepared by shaking the
* constituents together in closed jars for 5 minutes using a reciprocating
* table. Fish tests were run in open vessels.
F020 4551
EOR
F002 37
F010 4.1
F004 4
F005 RE
F006 Westlake, G.E.(1991
** Aquatic test media preparation for low solubility materials, Proceedings
* of the Society of Environmental Toxicology and Chemistry (SETAC)
* Conference on Environmental Sciences and Sustainable Development,
* University of Sh
F007 Westlake, G.E.(1991
** Aquatic test media preparation for low solubility materials, Proceedings
* of the Society of Environmental Toxicology and Chemistry (SETAC)
* Conference on Environmental Sciences and Sustainable Development,
* University of Sheffield, April 1991.
F020 4554
EOR
F002 37
F010 4.1
F004 4
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. Since
* significant evaporative losses of lower hydrocarbons may have occurred,
* the LC50 values are expected to
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. Since
* significant evaporative losses of lower hydrocarbons may have occurred,
* the LC50 values are expected to be lower than those cited. These data are
* also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by API crude oil task group as '3', not reliable.
F020 4553
EOR
F002 37
F010 4.1
F004 4
F005 RS
F006 The results were as follows:

**
** 24-hour LC50 = 756 mg/l
** 96-hour LC50 = 291 mg/l
** 168-hour LC50 = 133 mg/l
** As the tests were run in open vessels, significant
** evaporative losses of lower hydrocarbons will have occurred
** and he
F007 The results were as follows:
**
** 24-hour LC50 = 756 mg/l
** 96-hour LC50 = 291 mg/l
** 168-hour LC50 = 133 mg/l
** As the tests were run in open vessels, significant
** evaporative losses of lower hydrocarbons will have occurred
** and hence the measured LC50 values underestimate the true
** ecotoxicity.
F020 4552
EOR
F002 37
F010 4.1
F004 4
F005 SO
F006 CONCAWE Bruxelles
F007 CONCAWE Bruxelles
F008 IUC4
F009 09-10-2003
F020 4572
EOR
F002 37
F010 4.1
F004 5
F005 ME
F006 The test species was the Sheepshead Minnow. Dispersions of two crude oil
* samples in water were prepared by shaking the constituents together
* vigorously for 5 minutes on a shaker platform. Fish tests were run at 5
** concentrations. Figures q
F007 The test species was the Sheepshead Minnow. Dispersions of two crude oil
* samples in water were prepared by shaking the constituents together
* vigorously for 5 minutes on a shaker platform. Fish tests were run at 5
** concentrations. Figures quoted are for the loading rates.
F020 4555
EOR
F002 37
F010 4.1
F004 5
F005 RE
F006 Anderson, J.W. et al.(1974)
** Characteristics of dispersions and water-soluble extracts of crude and
* refined oils, and their toxicity to estuarine crustaceans and fish, Mar.
* Biol.,vol.27, pp. 75-88.
F007 Anderson, J.W. et al.(1974)
** Characteristics of dispersions and water-soluble extracts of crude and
* refined oils, and their toxicity to estuarine crustaceans and fish, Mar.

* Biol.,vol.27, pp. 75-88.
F008 IUC4
F009 13-11-2003
F020 4556
EOR
F002 37
F010 4.1
F004 5
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. Since
* significant evaporative losses of lower hydrocarbons may have occurred,
* the LC50 values are expected to
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. Since
* significant evaporative losses of lower hydrocarbons may have occurred,
* the LC50 values are expected to be lower than those cited. These data are
* also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by API crude oil task group as '3', not reliable.
F020 4558
EOR
F002 37
F010 4.1
F004 5
F005 RS
F006 The results were as follows:
** For Kuwait crude:
** 48-hour LC50 = 80 000+ mg/l
** 96-hour LC50 = 80 000+ mg/l
** For South Louisiana crude:
** 48-hour LC50 = 33 000 mg/l
** 96-hour LC50 = 29 000 mg/l
**
** Parallel tests run with
F007 The results were as follows:
** For Kuwait crude:
** 48-hour LC50 = 80 000+ mg/l
** 96-hour LC50 = 80 000+ mg/l
** For South Louisiana crude:
** 48-hour LC50 = 33 000 mg/l
** 96-hour LC50 = 29 000 mg/l
**
** Parallel tests run with water-soluble fractions (WSF) failed to produce
* any meaningful results.
F020 4557
EOR
F002 37
F010 4.1
F004 6
F005 ME
F006 The test species was the Longnose Killifish. Dispersions of two crude oil
* samples in water were prepared by shaking the constituents together
* vigorously for 5 minutes on a shaker platform. Fish tests were run at 5

* concentrations. Figures
F007 The test species was the Longnose Killifish. Dispersions of two crude oil
* samples in water were prepared by shaking the constituents together
* vigorously for 5 minutes on a shaker platform. Fish tests were run at 5
* concentrations. Figures quoted are for the loading rates.
F020 4560
EOR
F002 37
F010 4.1
F004 6
F005 RE
F006 Anderson, J.W. et al.(1974)
** Characteristics of dispersions and water-soluble extracts of crude and
* refined oils, and their toxicity to estuarine crustaceans and fish, Mar.
* Biol.,vol.27, pp. 75-88.
F007 Anderson, J.W. et al.(1974)
** Characteristics of dispersions and water-soluble extracts of crude and
* refined oils, and their toxicity to estuarine crustaceans and fish, Mar.
* Biol.,vol.27, pp. 75-88.
F008 IUC4
F009 13-11-2003
F020 4562
EOR
F002 37
F010 4.1
F004 6
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. Since
* significant evaporative losses of lower hydrocarbons may have occurred,
* the LC50 values are expected to
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. Since
* significant evaporative losses of lower hydrocarbons may have occurred,
* the LC50 values are expected to be lower than those cited. These data are
* also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by API crude oil task group as '3', not reliable.
F020 4561
EOR
F002 37
F010 4.1
F004 6
F005 RS
F006 The results were as follows:
** For Kuwait crude:
** 48-hour LC50 = 14 800 mg/l
** 96-hour LC50 = 14 800 mg/l
** For South Louisiana crude:
** 48-hour LC50 = 6000 mg/l
** 96-hour LC50 = 6000 mg/l
F007 The results were as follows:
** For Kuwait crude:
** 48-hour LC50 = 14 800 mg/l

** 96-hour LC50 = 14 800 mg/l
** For South Louisiana crude:
** 48-hour LC50 = 6000 mg/l
** 96-hour LC50 = 6000 mg/l
F020 4559
EOR
F002 37
F010 4.1
F004 7
F005 ME
F006 The test species was the Tidewater Silverside. Dispersions of two crude
* oil samples in water were prepared by shaking the constituents together
* vigorously for 5 minutes on a shaker platform. Fish tests were run at 5
** concentrations. Figure
F007 The test species was the Tidewater Silverside. Dispersions of two crude
* oil samples in water were prepared by shaking the constituents together
* vigorously for 5 minutes on a shaker platform. Fish tests were run at 5
** concentrations. Figures quoted are for the loading rates.
F020 4563
EOR
F002 37
F010 4.1
F004 7
F005 RE
F006 Anderson, J.W. et al.(1974)
** Characteristics of dispersions and water-soluble extracts of crude and
* refined oils, and their toxicity to estuarine crustaceans and fish, Mar.
* Biol.,vol.27, pp. 75-88.
F007 Anderson, J.W. et al.(1974)
** Characteristics of dispersions and water-soluble extracts of crude and
* refined oils, and their toxicity to estuarine crustaceans and fish, Mar.
* Biol.,vol.27, pp. 75-88.
F008 IUC4
F009 13-11-2003
F020 4565
EOR
F002 37
F010 4.1
F004 7
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. Since
* significant evaporative losses of lower hydrocarbons may have occurred,
* the LC50 values are expected to
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. Since
* significant evaporative losses of lower hydrocarbons may have occurred,
* the LC50 values are expected to be lower than those cited. These data are
* also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by API crude oil task group as '3', not reliable.
F020 4566
EOR
F002 37

F010 4.1
F004 7
F005 RS
F006 The results were as follows:
** For Kuwait crude: 48-hour LC50 = 15 000 mg/l, 96-hour LC50 = 9400 mg/l
** For South Louisiana crude: 48-hour LC50 = 5000 mg/l
** 96-hour LC50 = 3700 mg/l
F007 The results were as follows:
** For Kuwait crude: 48-hour LC50 = 15 000 mg/l, 96-hour LC50 = 9400 mg/l
** For South Louisiana crude: 48-hour LC50 = 5000 mg/l
** 96-hour LC50 = 3700 mg/l
F020 4564
EOR
F002 37
F010 4.1
F004 7
F005 SO
F006 CONCAWE Bruxelles
F007 CONCAWE Bruxelles
F008 IUC4
F009 09-10-2003
F020 4573
EOR
F002 37
F010 4.1
F004 8
F005 ME
F006 The test species was the Rainbow Trout. A water-soluble fraction (WSF) of
* the test substance was prepared by adding crude oil to water at a
* concentration of 12.5 ml/l and stirring for 2 hours. After settling for
* 72 hours, groups of 5 fish
F007 The test species was the Rainbow Trout. A water-soluble fraction (WSF) of
* the test substance was prepared by adding crude oil to water at a
* concentration of 12.5 ml/l and stirring for 2 hours. After settling for
* 72 hours, groups of 5 fish were exposed to solutions containing 20, 30,
* 40 and 50% WSF. Three series of tests were run:
** (a) with closed vessels
** (b) with open vessels, and
** (c) with aerated vessels.
F020 4567
EOR
F002 37
F010 4.1
F004 8
F005 RE
F006 Lockhart, W.L., Danell, R.W. and Murray, D.A. (1987)
** Acute toxicity bioassays with petroleum products: influence of exposure
* conditions.
** In: Oil in Freshwater: Chemistry, Biology, Counter-measure Technology,
* Vandermeulen, J.H. and Hrud
F007 Lockhart, W.L., Danell, R.W. and Murray, D.A. (1987)
** Acute toxicity bioassays with petroleum products: influence of exposure
* conditions.

** In: Oil in Freshwater: Chemistry, Biology, Counter-measure Technology,
* Vandermeulen, J.H. and Hrudey, S.E. (Eds), pp. 335-344, Pergamon Press.
F008 IUC4
F009 13-11-2003
F020 4569
EOR
F002 37
F010 4.1
F004 8
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by API crude oil task group as '2', reliable with
* restrictions.
F020 4570
EOR
F002 37
F010 4.1
F004 8
F005 RS
F006 The results were as follows:
** 48-hour LC50 (open vessel) = 10.4 mg/l
** 48-hour LC50 (closed vessel) = 11.6 mg/l
**
** based on measurements of dissolved hydrocarbons made at the
** beginning and end of the 48-hour period. No fish died
F007 The results were as follows:
** 48-hour LC50 (open vessel) = 10.4 mg/l
** 48-hour LC50 (closed vessel) = 11.6 mg/l
**
** based on measurements of dissolved hydrocarbons made at the
** beginning and end of the 48-hour period. No fish died in the aerated
* vessels, and hydrocarbons were undetectable in solution at the 48-hour
* time point in these studies.
F020 4568
EOR
F002 37
F010 4.1
F004 8
F005 SO
F006 CONCAWE Bruxelles
F007 CONCAWE Bruxelles
F008 IUC4
F009 09-10-2003
F020 4574
EOR
F002 37
F010 4.2

F004 1
F005 ME
F006 LL 50 at 96 hr calculated maximum likelihood analysis based
** on D.J. Finney, 1971
F007 LL 50 at 96 hr calculated maximum likelihood analysis based
** on D.J. Finney, 1971
F020 4575
EOR
F002 37
F010 4.2
F004 1
F005 RE
F006 ExxonMobil Biomedical Sciences Inc. (2002)
** Mysid - acute toxicity test: Study No. 121260, test
** substance MRD-01-02. 2002.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.
F007 ExxonMobil Biomedical Sciences Inc. (2002)
** Mysid - acute toxicity test: Study No. 121260, test
** substance MRD-01-02. 2002.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.
F020 4576
EOR
F002 37
F010 4.2
F004 1
F005 RS
F006 Mortality (no. of deaths/treatment) at 96 hrs: 0, 2, 2, 1,
** 15 and 20, respectively for control, 0.14, 0.28, 1.4, 3.5,
** and 11 mg/l treatments.
** 96-hr LL50 = 2.7 mg/l, 95% C.I: 2.2-3.6 mg/l (as actual
** loading rate)
** Results are quoted in term
F007 Mortality (no. of deaths/treatment) at 96 hrs: 0, 2, 2, 1,
** 15 and 20, respectively for control, 0.14, 0.28, 1.4, 3.5,
** and 11 mg/l treatments.
** 96-hr LL50 = 2.7 mg/l, 95% C.I: 2.2-3.6 mg/l (as actual
** loading rate)
** 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.
** Measured BTEX (mg/l) in test treatment (mg/l load)
** Day Control 0.14 0.28 1.4 3.5 11
** 0 (new) ND ND ND 0.049 0.167 0.500
** 1 (old) ND ND ND 0.0118 0.053 0.239
** 3 (new) 0.047 ND 0.003 0.0586 0.203 NA
** 4 (old) ND ND ND 0.017 0.0939 NA
**
** ND=not detected, NA=not analyzed due to 100% mortality
** The practical quantitation limit was 0.003 mg/l. Measured
** soluble concentrations of BTEX may have been reduced by the
** presence of brine shrimp in the test chambers
F020 4577
EOR

F002 37
F010 4.2
F004 1
F005 TC
F006 Test species: Juvenile Mysids used in the test were 1day old and were
* obtained from A.K. Siewers, Santa Cruz, CA, USA; the Mysids were
* acclimated for 3 days under laboratory
** conditions in seawater supplied by the organism vendor and
** fed fro
F007 Test species: Juvenile Mysids used in the test were 1day old and were
* obtained from A.K. Siewers, Santa Cruz, CA, USA; the Mysids were
* acclimated for 3 days under laboratory
** conditions in seawater supplied by the organism vendor and
** fed from in-house cultures of Artemia salina nauplii that
** were less than 24 hours old. Test organisms were fed brine
** shrimp once per day during the study.
**
** Test System: Individual water accommodated fractions (WAFs)
** were prepared for each test level. New test solutions were
** prepared for each 24-hour interval throughout the duration
** of the test. The control and dilution water was natural
** seawater collected from Manasquan Inlet, NJ, (NJDEP
** designated collection site) with a salinity of 33% and
** passed through 0.45 um filters prior to use.
**
** Based on results of range-finding tests for a similar
** petroleum product, actual loading rates of 0, 0.14, 0.28,
** 1.4, 3.5 and 11 mg/l of crude oil to water were used to
** prepare test solutions for definitive toxicity tests. Test
** substance, added volumetrically, was mixed for each
** individual treatment in dilution water for 24 hours in
** appropriate-sized, stoppered containers with approximately
** 20% vortex and less than 10% headspace volume. The WAFs were allowed to
* settle for 1-2 hours prior to drawing off the aqueous solutions for
* testing. A new WAF was prepared 24 hours prior to each renewal. Water
* quality (temperature, pH, salinity, and dissolved oxygen) measurements
* were recorded daily on the new solutions. Temperature, pH, and dissolved
* oxygen measurements were recorded on the old solutions (composite of
* replicates).
**
** Test conditions: Two replicate chambers per treatment were
** tested. Each replicate contained ten mysids. Replicate
** chambers were 500 ml glass jars with Teflon screw lids,
** containing approximately 450 ml of test solution with no
** headspace. All test and control solutions were replenished
** daily. Renewals were accomplished by transferring the mysids to fresh
* solution in a second set of test chambers on days 1, 2 and 3.
**
** Light: 16-hour light per day and a daylight intensity that
** ranged from 494 to 720 Lux.
**
** Test temperature: 11.9°C (S.D.=0.2).
**

** Water chemistry: Dissolved oxygen ranged from 6.4 - 7.0
** mg/l; pH ranged from 8.1 - 8.5; salinity ranged from 32 - 34 ppt.
**
** Freshly prepared and old WAF test solutions were analyzed by GC-FID for
* concentrations of BTEX.
F020 4578
EOR
F002 37
F010 4.2
F004 2
F005 ME
F006 LL 50 at 96 hr calculated maximum likelihood analysis based
** on D.J. Finney, 1971
F007 LL 50 at 96 hr calculated maximum likelihood analysis based
** on D.J. Finney, 1971
F020 4579
EOR
F002 37
F010 4.2
F004 2
F005 RE
F006 ExxonMobil Biomedical Sciences Inc. (2002)
** Mysid - acute toxicity test: Study No. 101160, test
** substance MRD-01-01.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.
F007 ExxonMobil Biomedical Sciences Inc. (2002)
** Mysid - acute toxicity test: Study No. 101160, test
** substance MRD-01-01.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.
F020 4580
EOR
F002 37
F010 4.2
F004 2
F005 RS
F006 Mortality (no. of deaths/treatment) at 96 hrs: 1, 2, 3, 6,
** 20 and 20, respectively for control, 0.6, 1.7, 3.6, 8.3 and
** 21 mg/l treatments.
** 96-hr LL50 = 4.1 mg/l, 95% C.I: 3.3-5.3 mg/l (as actual
** loading rate)
** Results are quoted in terms o
F007 Mortality (no. of deaths/treatment) at 96 hrs: 1, 2, 3, 6,
** 20 and 20, respectively for control, 0.6, 1.7, 3.6, 8.3 and
** 21 mg/l treatments.
** 96-hr LL50 = 4.1 mg/l, 95% C.I: 3.3-5.3 mg/l (as actual
** loading rate)
** 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.
** Measured BTEX (mg/l) in test treatment (mg/l load)
** Day Control 0.6 1.7 3.6 8.3 21
** 0 (new) ND ND ND ND 0.072 0.247
** 1 (old) ND ND ND ND ND 0.057

** 3 (new) ND ND ND 0.019 NA NA
** 4 (old) ND ND ND 0.023 NA NA
**
** ND=not detected, NA=not analyzed due to 100% mortality
** The practical quantitation limit was 0.003 mg/l. Measured
** soluble concentrations of BTEX may have been reduced by the
** presence of brine shrimp in the test chambers
F020 4581
EOR
F002 37
F010 4.2
F004 2
F005 TC
F006 Test species: Juvenile Mysids used in the test were 1day old
** and were obtained from A.K. Siewers, Santa Cruz, CA, USA;
** the Mysids were acclimated for 3 days under laboratory
** conditions in seawater supplied by the organism vendor and
** fed fro
F007 Test species: Juvenile Mysids used in the test were 1day old
** and were obtained from A.K. Siewers, Santa Cruz, CA, USA;
** the Mysids were acclimated for 3 days under laboratory
** conditions in seawater supplied by the organism vendor and
** fed from in-house cultures of Artemia salina nauplii that
** were less than 24 hours old. Test organisms were fed brine
** shrimp once per day during the study.
**
** Test System: Individual water accommodated fractions (WAFs)
** were prepared for each test level. New test solutions were
** prepared for each 24-hour interval throughout the duration
** of the test. The control and dilution water was natural
** seawater collected from Manasquan Inlet, NJ, (NJDEP
** designated collection site) with a salinity of 33% and
** passed through 0.45 um filters prior to use.
**
** Nominal loading rates of 0, 1.8, 19 and 105 mg/l were used
** to prepare test solutions for range-finding toxicity tests.
** Based on results of range-finding tests, actual loading
** rates of 0, 0.6, 1.7, 3.6, 8.3 and 21 mg/l of crude oil to
** water were used to prepare test solutions for definitive
** toxicity tests. Test substance, added volumetrically, was
** mixed for each individual treatment in dilution water for 24
** hours in appropriate-sized, stoppered containers with
** approximately 20% vortex and less than 10% headspace volume.
** The WAFs were allowed to settle for 1-2 hours prior to
** drawing off the aqueous solutions for testing. A new WAF was
** prepared 24 hours prior to each renewal. Water quality
** (temperature, pH, salinity, and dissolved oxygen)
** measurements were recorded daily on the new solutions.
** Temperature, pH, and dissolved oxygen measurements were
** recorded on the old solutions (composite of replicates).
**
** Test conditions: Two replicate chambers per treatment were
** tested. Each replicate contained ten mysids. Replicate

** chambers were 500 ml glass jars with Teflon® screw lids,
** containing approximately 450 ml of test solution with no
** headspace. All test and control solutions were replenished
** daily. Renewals were accomplished by transferring the mysids
** to fresh solution in a second set of test chambers on days
** 1, 2 and 3.
** Light: 16-hour light per day and a daylight intensity that
** ranged from 494 to 720 Lux.
** Test temperature: 12.3°C (S.D.=0.1).
** Water chemistry: Dissolved oxygen ranged from 6.4 - 7.0
** mg/l; pH ranged from 8.1 - 8.5; salinity ranged from 32 - 33
** ppt.
** Freshly prepared and old WAF test solutions were analyzed by
** GC-FID for concentrations of BTEX.
F020 4582
EOR
F002 37
F010 4.2
F004 3
F005 ME
F006 The test species was the Brown Shrimp. Groups of 20 shrimps were exposed
* to nominal concentrations in the range 17 to 3400 mg/l of each crude oil
* in sea water. After addition of the crude oil, the solutions were
* stirred at a constant rate
F007 The test species was the Brown Shrimp. Groups of 20 shrimps were exposed
* to nominal concentrations in the range 17 to 3400 mg/l of each crude oil
* in sea water. After addition of the crude oil, the solutions were
* stirred at a constant rate using a shielded stirrer. Crude oil solutions
* were renewed after 48 hours. Mortalities were recorded daily.
F020 4583
EOR
F002 37
F010 4.2
F004 3
F005 RE
F006 Franklin, F.L. and Lloyd, R.(1982)
** The toxicity of twenty-five oils in relation to the MAFF dispersant
* tests,
** Fisheries Research Technical Report No. 70, 1982
** Ministry of Agriculture, Fisheries and Food, Lowestoft.
F007 Franklin, F.L. and Lloyd, R.(1982)
** The toxicity of twenty-five oils in relation to the MAFF dispersant
* tests,
** Fisheries Research Technical Report No. 70, 1982
** Ministry of Agriculture, Fisheries and Food, Lowestoft.
F008 IUC4
F009 13-11-2003
F020 4585
EOR
F002 37
F010 4.2
F004 3
F005 RM

F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* Since no analytical dete
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* Since no analytical determinations were reported, these data are ranked
* by API crude oil task group as '2', reliable with restrictions.
F020 4586
EOR
F002 37
F010 4.2
F004 3
F005 RS
F006 The lowest LC50 value was obtained for Brent crude and the highest for
* Thistle crude. Other North Sea crude oils tested were from the Argyll,
* Auk, Beryl, Claymore, Ekofisk, Forties, Montrose, Murchison and Piper
* fields.
F007 The lowest LC50 value was obtained for Brent crude and the highest for
* Thistle crude. Other North Sea crude oils tested were from the Argyll,
* Auk, Beryl, Claymore, Ekofisk, Forties, Montrose, Murchison and Piper
* fields.
F020 4584
EOR
F002 37
F010 4.2
F004 3
F005 SO
F006 CONCAWE Bruxelles
**
**
F007 CONCAWE Bruxelles
**
**
F008 IUC4
F009 09-10-2003
F020 4606
EOR
F002 37
F010 4.2
F004 4
F005 ME
F006 The test species was the Brown Shrimp. Groups of 20 shrimps were exposed
* to nominal concentrations in the range 17 to 3400 mg/l of each crude oil
* in sea water. After addition of the crude oil, the solutions were
* stirred at a constant rate
F007 The test species was the Brown Shrimp. Groups of 20 shrimps were exposed
* to nominal concentrations in the range 17 to 3400 mg/l of each crude oil
* in sea water. After addition of the crude oil, the solutions were
* stirred at a constant rate using a shielded stirrer. Crude oil solutions
* were renewed after 48 hours. Mortalities were recorded daily.
F020 4587

EOR
F002 37
F010 4.2
F004 4
F005 RE
F006 Franklin, F.L. and Lloyd, R.(1982)
** The toxicity of twenty-five oils in relation to the MAFF dispersant
* tests,
** Ministry of Agriculture, Fisheries and Food, Lowestoft. Fisheries
* Research Technical Report No. 70, 1982.
F007 Franklin, F.L. and Lloyd, R.(1982)
** The toxicity of twenty-five oils in relation to the MAFF dispersant
* tests,
** Ministry of Agriculture, Fisheries and Food, Lowestoft. Fisheries
* Research Technical Report No. 70, 1982.
F020 4590
EOR
F002 37
F010 4.2
F004 4
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* Since no analytical dete
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* Since no analytical determinations were reported, these data are ranked
* by API crude oil task group as '2', reliable with restrictions.
F020 4589
EOR
F002 37
F010 4.2
F004 4
F005 RS
F006 The lowest LC50 value was obtained for Abu Dhabi crude and the highest
* for Iranian Light crude. Other crude oils tested were from Libya, Saudi
* Arabia, Nigeria, Kuwait, Iraq and Iran (heavy crude).
F007 The lowest LC50 value was obtained for Abu Dhabi crude and the highest
* for Iranian Light crude. Other crude oils tested were from Libya, Saudi
* Arabia, Nigeria, Kuwait, Iraq and Iran (heavy crude).
F020 4588
EOR
F002 37
F010 4.2
F004 4
F005 SO
F006 CONCAWE Bruxelles
F007 CONCAWE Bruxelles
F008 IUC4
F009 09-10-2003
F020 4607

EOR
F002 37
F010 4.2
F004 5
F005 ME
F006 Dispersions of oil in water at 5 concentrations in the range 10 to 100
* mg/l were prepared by shaking the constituents together in closed jars
* for 5 minutes using a
** reciprocating table. Groups of 10 daphnids were exposed to the resulting
* so
F007 Dispersions of oil in water at 5 concentrations in the range 10 to 100
* mg/l were prepared by shaking the constituents together in closed jars
* for 5 minutes using a
** reciprocating table. Groups of 10 daphnids were exposed to the resulting
* solutions. Tests were run in open vessels.
F020 4591
EOR
F002 37
F010 4.2
F004 5
F005 RE
F006 Westlake, G.E.(1991
** Aquatic test media preparation for low solubility materials, Proceedings
* of the Society of Environmental Toxicology and Chemistry (SETAC)
* Conference on Environmental Sciences and Sustainable Development,
* University of Sh
F007 Westlake, G.E.(1991
** Aquatic test media preparation for low solubility materials, Proceedings
* of the Society of Environmental Toxicology and Chemistry (SETAC)
* Conference on Environmental Sciences and Sustainable Development,
* University of Sheffield, April 1991.
F020 4593
EOR
F002 37
F010 4.2
F004 5
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* Since significant evapor
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* Since significant evaporative losses of lower hydrocarbons may have
* occurred, the LC50 values are expected to be lower than those cited.
* These data are ranked by API crude oil task group as '3', not reliable.
F020 4594
EOR
F002 37
F010 4.2
F004 5
F005 RS

F006 The results were as follows: 24-hour EC50 = 36 mg/l. As the tests were
* run in open vessels, significant evaporative losses of lower hydrocarbons
* will have occurred
** and hence the measured EC50 value underestimates the true ecotoxicity.
F007 The results were as follows: 24-hour EC50 = 36 mg/l. As the tests were
* run in open vessels, significant evaporative losses of lower hydrocarbons
* will have occurred
** and hence the measured EC50 value underestimates the true ecotoxicity.
F020 4592
EOR
F002 37
F010 4.2
F004 5
F005 SO
F006 CONCAWE Bruxelles
F007 CONCAWE Bruxelles
F008 IUC4
F009 09-10-2003
F020 4608
EOR
F002 37
F010 4.2
F004 6
F005 ME
F006 Crude oil was added to water at 8 concentrations in the range 10 to 100
* mg/l in open vessels fitted with shielded propeller stirrers. A constant
* rate of stirring was
** maintained during the 24-hour exposure period to groups of 10 daphnids.
**
F007 Crude oil was added to water at 8 concentrations in the range 10 to 100
* mg/l in open vessels fitted with shielded propeller stirrers. A constant
* rate of stirring was
** maintained during the 24-hour exposure period to groups of 10 daphnids.
**
F020 4597
EOR
F002 37
F010 4.2
F004 6
F005 RE
F006 Westlake, G.E.(1991
** Aquatic test media preparation for low solubility materials, Proceedings
* of the Society of Environmental Toxicology and Chemistry (SETAC)
* Conference on Environmental Sciences and Sustainable Development,
* University of Sh
F007 Westlake, G.E.(1991
** Aquatic test media preparation for low solubility materials, Proceedings
* of the Society of Environmental Toxicology and Chemistry (SETAC)
* Conference on Environmental Sciences and Sustainable Development,
* University of Sheffield, April 1991.
F020 4596
EOR
F002 37

F010 4.2
F004 6
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* Since significant evapor
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* Since significant evaporative losses of lower hydrocarbons may have
* occurred, the LC50 values are expected to be lower than those cited.
* These data are ranked by API crude oil task group as '3', not reliable.
F020 4595
EOR
F002 37
F010 4.2
F004 6
F005 RS
F006 The results were as follows: 24-hour EC50 = 42 mg/l. As the tests were
* run in open vessels, significant evaporative losses of lower hydrocarbons
* will have occurred
** and hence the measured EC50 value underestimates the true ecotoxicity.
F007 The results were as follows: 24-hour EC50 = 42 mg/l. As the tests were
* run in open vessels, significant evaporative losses of lower hydrocarbons
* will have occurred
** and hence the measured EC50 value underestimates the true ecotoxicity.
F020 4598
EOR
F002 37
F010 4.2
F004 6
F005 SO
F006 CONCAWE Bruxelles
F007 CONCAWE Bruxelles
F008 IUC4
F009 09-10-2003
F020 4609
EOR
F002 37
F010 4.2
F004 7
F005 ME
F006 Tests were carried out in polypropylene containers using water
* accommodated fractions prepared by physical dispersion. These were
* renewed daily, and kept tightly covered with teflon sheets. Loading rates
* of 11.6, 34.3, 99.1, 284 and 862 mg/
F007 Tests were carried out in polypropylene containers using water
* accommodated fractions prepared by physical dispersion. These were
* renewed daily, and kept tightly covered with teflon sheets. Loading rates
* of 11.6, 34.3, 99.1, 284 and 862 mg/l were used in this study. The test
* species was the kelp forest mysid, Holmesimysis costata.
**

F020 4599
EOR
F002 37
F010 4.2
F004 7
F005 RE
F006 Bragin, G.E., Clark, J.R. and Pace, C.B.(1994)
** Comparison of physically and chemically dispersed crude oil toxicity
* under continuous and spiked exposure scenarios, Marine Spill Response
* Corporation, Washington D.C., MSRC Technical Report Se
F007 Bragin, G.E., Clark, J.R. and Pace, C.B.(1994)
** Comparison of physically and chemically dispersed crude oil toxicity
* under continuous and spiked exposure scenarios, Marine Spill Response
* Corporation, Washington D.C., MSRC Technical Report Series 94-015, 45p.
F008 IUC4
F009 13-11-2003
F020 4601
EOR
F002 37
F010 4.2
F004 7
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by API crude oil task group as '1', reliable
* without restrictions.
F020 4602
EOR
F002 37
F010 4.2
F004 7
F005 RS
F006 A lethal loading rate concentration, LL50 (96h) of 39.5 mg/l was
* calculated from data presented in the original publication after
* consultation with the main author. Weight percent distribution for the
* crude oil components with BP up to 425
F007 A lethal loading rate concentration, LL50 (96h) of 39.5 mg/l was
* calculated from data presented in the original publication after
* consultation with the main author. Weight percent distribution for the
* crude oil components with BP up to 425 °F are: paraffins: 68.56%;
* aromatics: 15.69 and naphthenes: 11.86%.
F020 4600
EOR
F002 37
F010 4.2
F004 7
F005 SO
F006 CONCAWE Bruxelles

**
F007 CONCAWE Bruxelles
**
F008 IUC4
F009 09-10-2003
F020 4610
EOR
F002 37
F010 4.2
F004 8
F005 ME
F006 Tests were carried out in polypropylene containers using water
* accommodated fractions prepared by physical dispersion. These were
* renewed daily and kept tightly covered with teflon sheets. Loading rates
* of 340, 695, 1550, 3470 and 7740 mg/
F007 Tests were carried out in polypropylene containers using water
* accommodated fractions prepared by physical dispersion. These were
* renewed daily and kept tightly covered with teflon sheets. Loading rates
* of 340, 695, 1550, 3470 and 7740 mg/l were used in this study. The test
* species was the estuarine mysid, Mysodopsis bahia.
**
F020 4603
EOR
F002 37
F010 4.2
F004 8
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by API crude oil task group as '1', reliable
* without restrictions.
F020 4605
EOR
F002 37
F010 4.2
F004 8
F005 RS
F006 A lethal loading rate concentration LL50 (96h) of 618 mg/l was calculated
* from data presented in the original publication after consultation with
* the main author. . Weight percent distribution for the crude oil
* components with BP up to 425
F007 A lethal loading rate concentration LL50 (96h) of 618 mg/l was calculated
* from data presented in the original publication after consultation with
* the main author. . Weight percent distribution for the crude oil
* components with BP up to 425 °F are: paraffins: 68.56%; aromatics: 15.69
* and naphthenes: 11.86%.
F020 4604
EOR

F002 37
F010 4.2
F004 8
F005 SO
F006 CONCAWE Bruxelles
F007 CONCAWE Bruxelles
F008 IUC4
F009 09-10-2003
F020 4611
EOR
F002 37
F010 4.3
F004 1
F005 ME
F006 The method used was described by Chapman, G.A., D.L. Denton
** and J.M. Lazorchak. (1995).
**
** Statistical method
** Lethal Loading and Effect Loading: Norberg-King, T.J., A
** Linear Interpolation Method for Sublethal Toxicity: The
** Inhibition Concent
F007 The method used was described by Chapman, G.A., D.L. Denton
** and J.M. Lazorchak. (1995).
**
** Statistical method
** Lethal Loading and Effect Loading: Norberg-King, T.J., A
** Linear Interpolation Method for Sublethal Toxicity: The
** Inhibition Concentration (ICp) Approach (Version 2.0). July
** 1993. U.S. Environmental Protection Agency, Environmental
** Research Laboratory, Duluth MN.
F020 4612
EOR
F002 37
F010 4.3
F004 1
F005 RE
F006 Chapman, G.A., Denton, D.L. and Lazorchak, J.M. (1995)
** Short-Term Methods for Estimating the Chronic Toxicity of
** Effluents and Receiving Waters to West Coast Marine and
** Estuarine Organisms. EPA-600/R-95-136.
** US Environmental Protection Age
F007 Chapman, G.A., Denton, D.L. and Lazorchak, J.M. (1995)
** Short-Term Methods for Estimating the Chronic Toxicity of
** Effluents and Receiving Waters to West Coast Marine and
** Estuarine Organisms. EPA-600/R-95-136.
** US Environmental Protection Agency, Cincinnati, OH.
F008 IUC4
F009 28-08-2003
F020 4613
EOR
F002 37
F010 4.3
F004 1

F005 RE
F006 ExxonMobil Biomedical Sciences Inc. (2002)
** Giant Kelp, Germination and Growth Test: Study No. 121274,
** test substance MRD-01-02. 2002.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.
F007 ExxonMobil Biomedical Sciences Inc. (2002)
** Giant Kelp, Germination and Growth Test: Study No. 121274,
** test substance MRD-01-02. 2002.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.
F020 4614
EOR
F002 37
F010 4.3
F004 1
F005 RS
F006 48 hour results, based on mg of test substance per liter of
** natural seawater
**

** Germination	Tube Length	Germination	Tube Length			
** LL50	EL50		NOEL	LOEL	NOEL	LOEL
** (mg/l)	(mg/l)		(mg/l)	(mg/l)	(mg/l) (mg/l)	
** 122	122	52	146	18	52	

**
F007 48 hour results, based on mg of test substance per liter of
** natural seawater
**

** Germination	Tube Length	Germination	Tube Length			
** LL50	EL50		NOEL	LOEL	NOEL	LOEL
** (mg/l)	(mg/l)		(mg/l)	(mg/l)	(mg/l) (mg/l)	
** 122	122	52	146	18	52	

**
** Analytical results
**
** Nominal (mg/l) Measured Concentration (mg/l as BTEX)
**

**	Day 0	Day 2
** Control	none detected	none detected
** 18	0.542	0.478
** 52	1.82	1.23
** 146	5.91	3.27
** 415	15.2	5.88
** 1185	20.0	11.3

**
** Practical Quantitation Limit (PQL) was approximately 0.0035
** mg/l (3.5ng/ml)
F020 4615
EOR
F002 37
F010 4.3
F004 1
F005 TC
F006 Individual test treatment solutions were prepared as Water
** Accommodated Fractions (WAFs). Actual loading levels of test substance in

* natural seawater (dilution water) were 18 mg/l, 52 mg/l, 146 mg/l, 415
* mg/l and 1185 mg/l. A control
** treat
F007 Individual test treatment solutions were prepared as Water
** Accommodated Fractions (WAFs). Actual loading levels of test substance in
* natural seawater (dilution water) were 18 mg/l, 52 mg/l, 146 mg/l, 415
* mg/l and 1185 mg/l. A control
** treatment consisting of natural seawater with no test
** substance was also prepared. The WAFs were prepared by
** adding the appropriate amount of test substance to natural
** seawater in glass aspirator bottles and stirring on magnetic stirplates
* for 24 hours. Natural seawater was filtered through a 0.45 µm filter,
* then a 0.2 µm filter (collected from Manasquan Inlet, NJ, NJDEP
* designated collection site).
** The salinity of the seawater was 34 ± 2 ppt.
** Ammonia (NH3 + NH4+) = 0 mg/l
** Nitrite (NO2-) = <0.3 mg/l
** Nitrate (N) = 1 mg/l.
** The mixtures were allowed to settle and cool to test
** temperature for 1.25 hour before removing the aqueous
** portions (WAFs) for testing. Samples of the WAFs prepared at each loading
* level were taken on Day 0 and at termination from each treatment
* (composite of replicates). The samples were analyzed for benzene,
* toluene, ethylbenzene and o-xylene (BTEX), all hydrocarbons detected in
* the entire chromatographic run were quantified versus the BTEX standards
* and the reported results reflect the sum of all quantified compounds.
* Samples were taken with no headspace in volatile organic analysis (VOA)
* vials.
** Test temperature was 14.0 (sd=0.3) °C.
**
** The kelp sporophylls (reproductive blades) were received
** from A. K. Siewers, Santa Cruz, CA 95060, packed on ice at
** 7.5°C, the day after they were harvested. The sporophylls
** were rinsed with 0.2 µm filtered seawater at 14°C and
** blotted dry with paper towels. They were then desiccated at 14°C for one
* hour. The sporophylls were rinsed again with 0.2µm filtered seawater at
* 14°C. Approximately 20 of the sporophylls were placed into a one liter
* glass beaker
** containing 800 ml of 0.2 µm filtered seawater at 14°C. The
** blades were left in the beaker for 30 minutes while the
** zoospores released. The blades were removed and the spore
** solution was allowed to settle for 20 minutes. 250 ml was
** poured from the top of the beaker, this was the spore stock.
** The zoospores were released from the sporophylls and
** inoculated into test chambers within a two hour period. The zoospore
* release was initiated within 24 hours of sporophyll harvest.
**
** Five replicate chambers for each dose treatment consisted of 500 ml size
* glass jars sealed with Teflon® lined screw type lids, containing
* approximately 450 ml of test solution to minimize headspace and prevent
* loss of volatile components.
** A pre-cleaned 2.5 cm x 7.6 cm glass slide was placed in the
** bottom of each jar prior to adding test solution. The

** initial concentration was approximately 7.5 x 103
** zoospores/ml in each replicate chamber. The spore stock
** density was determined as follows: 1 ml of the stock and 1
** ml of glacial acetic acid were added to 8 ml of 0.2 µm
** filtered seawater in a graduated cylinder. The solution
** was mixed and counts were performed using a hemacytometer at 100x. Each
* test chamber, containing approximately 7500
** spores, was labeled to show the study number, loading level, replicate
* and randomization number. Each test and control chamber were observed for
* germination of the gametophyte spores and length of the embryonic
* gametophyte germination tube after approximately 48 hrs. The following
* procedure was used. The glass slide was removed from the jar and the
* bottom of the slide was dried with a paper towel. A 24 x 50 mm cover
* slideslip was placed onto the slide and the edges were dabbed.
* Germination was considered successful if a germ tube was present on the
* settled zoospore. Germination was considered unsuccessful if no germ
* tube was visible.
** 400x magnification was used to differentiate germination and measure the
* tubes. A spore had to have a protuberance that extended at least one
* spore diameter from the edge of the spore to be considered germinated.
* The first 100 spores, which were easily identifiable, were counted while
* moving the slide across the field of view of the microscope.
** Measurements were made by moving the slide and choosing the
** first 10 spores with straight germ tubes within the same
** focal plane of the microscope that were closest to the scale in the
* ocular micrometer. The ocular micrometer was
** calibrated with a stage micrometer at 400x to assure
** accurate measurements. The micrometer was calibrated from 3 to 25 µm.
* The total length of the tube from the edge of the original spore membrane
* was measured.
**
** Water quality (temperature, pH, dissolved oxygen, and
** salinity) measurements were made on each treatment on Day 0
** and at termination (composite of replicates). BTEX total
** concentration at termination was at least 80% of the initial
* concentration.
**
** The study was deemed acceptable. Mean control germination
** was > 70% in the controls. Mean germination tube length was > 10 µm in
* the controls.
F020 4616
EOR
F002 37
F010 4.3
F004 2
F005 ME
F006 Lethal Loading and Effect Loading: Norberg-King, T.J., A
** Linear Interpolation Method for Sublethal Toxicity: The
** Inhibition Concentration (ICp) Approach (Version 2.0). July
** 1993. U.S. Environmental Protection Agency, Environmental
** Research
F007 Lethal Loading and Effect Loading: Norberg-King, T.J., A
** Linear Interpolation Method for Sublethal Toxicity: The

** Inhibition Concentration (ICp) Approach (Version 2.0). July
** 1993. U.S. Environmental Protection Agency, Environmental
** Research Laboratory, Duluth MN.
F020 4617
EOR
F002 37
F010 4.3
F004 2
F005 RE
F006 Chapman, G.A., Denton, D.L. and Lazorchak, J.M. (1995)
** Short-Term Methods for Estimating the Chronic Toxicity of
** Effluents and Receiving Waters to West Coast Marine and
** Estuarine Organisms. EPA-600/R-95-136.
** US Environmental Protection Age
F007 Chapman, G.A., Denton, D.L. and Lazorchak, J.M. (1995)
** Short-Term Methods for Estimating the Chronic Toxicity of
** Effluents and Receiving Waters to West Coast Marine and
** Estuarine Organisms. EPA-600/R-95-136.
** US Environmental Protection Agency, Cincinnati, OH.
F008 IUC4
F009 28-08-2003
F020 4618
EOR
F002 37
F010 4.3
F004 2
F005 RE
F006 ExxonMobil Biomedical Sciences Inc. (2002)
** Giant Kelp, Germination and Growth Test: Study No. 101174,
** test substance MRD-01-01.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.
F007 ExxonMobil Biomedical Sciences Inc. (2002)
** Giant Kelp, Germination and Growth Test: Study No. 101174,
** test substance MRD-01-01.
** ExxonMobil Biomedical Sciences Inc. Annandale, NJ.
F020 4619
EOR
F002 37
F010 4.3
F004 2
F005 RS
F006 48 hour results, based on mg of test substance per liter of
** natural seawater (95%CI)
**
** Germination Tube Length Germination Tube Length LL50 EL50
* NOEL LOEL NOELLOEL
** 528 311 53 130 23 53 (497
F007 48 hour results, based on mg of test substance per liter of
** natural seawater (95%CI)
**
** Germination Tube Length Germination Tube Length LL50 EL50
* NOEL LOEL NOELLOEL
** 528 311 53 130 23 53 (497-558)

* (257-403)
**
** Analytical results
** Nominal (mg/l) Measured Concentration (mg/l as BTEX)
**
** Day 0 Day 2
** Control none detected none detected
** 23 0.047 none detected
** 53 0.586 0.039
** 130 1.09 0.428
** 323 3.35 1.92
** 802 5.03 4.43
**
** Practical Quantitation Limit (PQL) was approximately 0.0035
** mg/l (3.5ng/ml)
**
**
**
** Spore concentration was 1000 spore/ml, instead of 7500
** spore/ml, as required. However total spore number per
** replicate was adequate sample size for determination of end
** points.
F020 4620
EOR
F002 37
F010 4.3
F004 2
F005 TC
F006 Individual test treatment solutions were prepared as Water
** Accommodated Fractions (WAFs). Actual loading levels of test
** substance in natural seawater (dilution water) were 23 mg/l,
** 53 mg/l, 130 mg/l, 323 mg/l and 802 mg/l. A control
** treatme
F007 Individual test treatment solutions were prepared as Water
** Accommodated Fractions (WAFs). Actual loading levels of test
** substance in natural seawater (dilution water) were 23 mg/l,
** 53 mg/l, 130 mg/l, 323 mg/l and 802 mg/l. A control
** treatment consisting of natural seawater with no test
** substance was also prepared. The WAFs were prepared by
** adding the appropriate amount of test substance to natural
** seawater in glass aspirator bottles and stirring on
** magnetic stirplates for 24 hours. Natural seawater was
** filtered through a 0.45 µm filter, then a 0.2 µm filter
** (collected from Manasquan Inlet, NJ, NJDEP designated
** collection site). The salinity of the seawater was 34 ± 2
** ppt. The mixtures were allowed to settle and cool to test
** temperature for 1 hour before removing the aqueous portions
** (WAFs) for testing. Samples of the WAFs prepared at each
** loading level were taken on Day 0 and at termination from
** each treatment (composite of replicates). The samples were
** analyzed for benzene, toluene, ethylbenzene and o-xylene
** (BTEX), all hydrocarbons detected in the entire
** chromatographic run were quantified versus the BTEX

** standards and the reported results reflect the sum of all
** quantified compounds. Samples were taken with no headspace
** in volatile organic analysis (VOA) vials.
** Mean test temperature was 14.1 (sd=0.3) °C.
**
** The kelp sporophylls (reproductive blades) were received
** from A. K. Siewers, Santa Cruz, CA 95060, packed on ice at
** 12.2°C, the day after they were harvested. The sporophylls
** were rinsed with 0.2 µm filtered seawater at 14°C and
** blotted dry with paper towels. They were then desiccated at
** 14°C for one hour. The sporophylls were rinsed again with
** 0.2µm filtered seawater at 14°C. Approximately 20 of the
** sporophylls were placed into a one liter glass beaker
** containing 800 ml of 0.2 µm filtered seawater at 14°C. The
** blades were left in the beaker for 30 minutes while the
** zoospores released. The blades were removed and the spore
** solution was allowed to settle for 25 minutes. 250 ml was
** poured from the top of the beaker, this was the spore stock.
** The zoospores were released from the sporophylls and
** inoculated into test chambers within a two hour period. The
** zoospore release was initiated within 24 hours of sporophyll
** harvest.
** Five replicate chambers for each dose treatment consisted of
** 500 ml size glass jars sealed with Teflon® lined screw type
** lids, containing approximately 450 ml of test solution. A
** pre-cleaned 2.5 cm x 7.6 cm glass slide was placed in the
** bottom of each jar prior to adding test solution. The
** initial concentration was approximately 1.0×10^3
** zoospores/ml in each replicate chamber. The spore stock
** density was determined as follows: 1 ml of the stock and 1
** ml of glacial acetic acid were added to 8 ml of 0.2 µm
** filtered seawater in a graduated cylinder. The solution
** was mixed and counts were performed using a hemacytometer at
** 100x. Each test chamber, containing approximately 1000
** spores, was labeled to show the study number, loading level,
** replicate and randomization number. Each test and control
** chamber was observed for germination of the gametophyte
** spores and length of the embryonic gametophyte germination
** tube. The following procedure was used. The glass slide
** was removed from the jar, and the bottom of the slide was
** dried with a paper towel. A 24 x 50 mm cover slideslip was
** placed onto the slide and the edges were dabbed.
** Germination was considered successful if a germ tube was
** present on the settled zoospore. Germination was considered
** unsuccessful if no germ tube was visible. 400x
** magnification was used to differentiate germination and
** measure the tubes. A spore had to have a protuberance that
** extended at least one spore diameter from the edge of the
** spore to be considered germinated. The first 100 spores,
** which were easily identifiable, were counted while moving
** the slide across the field of view of the microscope.
** Measurements were made by moving the slide and choosing the
** first 10 spores with straight germ tubes within the same

** focal plane of the microscope that were closest to the scale
** in the ocular micrometer. The ocular micrometer was
** calibrated with a stage micrometer at 400x to assure
** accurate measurements. The micrometer was calibrated from 3
** to 25 µm. The total length of the tube from the edge of the
** original spore membrane was measured.
** Water quality (temperature, pH, dissolved oxygen, and
** salinity) measurements were made on each treatment on Day 0
** and at termination (composite of replicates). BTEX total
** concentration at termination was at least 80% of the initial
** concentration.
** The study was deemed acceptable. Mean control germination
** was > 70% in the controls. Mean germination tube length was
** > 10 um in the controls.
F020 4621
EOR
F002 37
F010 4.3
F004 3
F005 ME
F006 Tests were run using:
** (a) a water-soluble fraction (WSF) of the test
** substance, and
** (b) the test substance in equilibrium with the
** algal suspension medium.
** To prepare the WSF, crude oil was added to the sterilized medium in
F007 Tests were run using:
** (a) a water-soluble fraction (WSF) of the test
** substance, and
** (b) the test substance in equilibrium with the
** algal suspension medium.
** To prepare the WSF, crude oil was added to the sterilized medium in the
* ratio 1:20, and was stirred in closed bottles for 12 hours. After a
* 4-hour separation period the aqueous
** phase was separated and diluted for the tests. In the direct loading
* tests, crude oil was applied to absorbent pads and these were held in the
* culture suspension. Analysis was done by gas chromatography, following
* solvent extraction of the aqueous phase using n-pentane.
F020 4622
EOR
F002 37
F010 4.3
F004 3
F005 RE
F006 Gaur, J.P. and Singh, A.K. (1989)
** Comparative studies on the toxicity of petroleum oils and their aqueous
* extracts towards Anabaena doliolum,
** Proc. Indian Acad. Sci. (Plant Sci.), vol. 99, pp. 459-466,
F007 Gaur, J.P. and Singh, A.K. (1989)
** Comparative studies on the toxicity of petroleum oils and their aqueous
* extracts towards Anabaena doliolum,
** Proc. Indian Acad. Sci. (Plant Sci.), vol. 99, pp. 459-466,
F020 4624

EOR
F002 37
F010 4.3
F004 3
F005 RM
F006 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by
F007 These data are included to provide supporting evidence of the expected
* cumulative toxicity of the soluble components in crude oil. These data
* are also cited in the European Chemicals Bureau IUCLID for CAS 8002-05-9.
* These data are ranked by API crude oil task group as '1', reliable
* without restrictions.
F020 4625
EOR
F002 37
F010 4.3
F004 3
F005 RS
F006 The WSF method gave a mean 15-day EC50 of 9.06 mg/l, and
** the direct loading (or whole oil) method gave a 15-day EC50
** of 5.73 mg/l, both based on measured dissolved hydrocarbon
** concentrations in the aqueous phase.
**
F007 The WSF method gave a mean 15-day EC50 of 9.06 mg/l, and
** the direct loading (or whole oil) method gave a 15-day EC50
** of 5.73 mg/l, both based on measured dissolved hydrocarbon
** concentrations in the aqueous phase.
**
F020 4623
EOR
F002 37
F010 4.3
F004 3
F005 SO
F006 CONCAWE Bruxelles
**
F007 CONCAWE Bruxelles
**
F008 IUC4
F009 09-10-2003
F020 4626
EOR
F002 37
F010 4.9
F004 1
F005 RE
F006 CONCAWE (1992)
** Ecotoxicological testing of petroleum products: test methodology
** Report No. 92/56
** CONCAWE, Brussels.
F007 CONCAWE (1992)

** Ecotoxicological testing of petroleum products: test methodology
** Report No. 92/56
** CONCAWE, Brussels.
F008 IUC4
F009 13-11-2003
F020 4628
EOR
F002 37
F010 4.9
F004 1
F005 RE
F006 GESAMP (1989)
** Reports and Studies No. 35, Annex 9, Advice for aquatic toxicity testing
* of substances or of mixtures containing compounds of low solubility
** International Maritime Organization (IMO), London
F007 GESAMP (1989)
** Reports and Studies No. 35, Annex 9, Advice for aquatic toxicity testing
* of substances or of mixtures containing compounds of low solubility
** International Maritime Organization (IMO), London
F008 IUC4
F009 13-11-2003
F020 4629
EOR
F002 37
F010 4.9
F004 1
F005 RE
F006 Whitehouse, P. and Mallet, M. (1983)
** Aquatic toxicity testing for notification of new substances. An advisory
* document for dealing with "difficult" substances
** Water Research Centre (WRC), Medmenham UK.
F007 Whitehouse, P. and Mallet, M. (1983)
** Aquatic toxicity testing for notification of new substances. An advisory
* document for dealing with "difficult" substances
** Water Research Centre (WRC), Medmenham UK.
F008 IUC4
F009 13-11-2003
F020 4630
EOR
F002 37
F010 4.9
F004 1
F005 RM
F006 For the assessment of the ecotoxicity of poorly water soluble mixtures of
* hydrocarbons as found in petroleum products, it is now generally accepted
* that results should be expressed in terms of the "loading rate". The
* "loading rate" may be
F007 For the assessment of the ecotoxicity of poorly water soluble mixtures of
* hydrocarbons as found in petroleum products, it is now generally accepted
* that results should be expressed in terms of the "loading rate". The
* "loading rate" may be defined as the amount of the product which must be
* equilibrated with the aqueous test medium in order to produce a specified
* level of effect. Studies in which the results are expressed in terms of

* the measured concentrations of hydrocarbons in dilutions of "water
* soluble fractions (WSF)" do not allow the ecotoxicity of a product to be
* expressed in terms of the amount of that product required to produce a
* particular effect and, therefore, such results are not comparable to
* results
** obtained with other substances.
F020 4627
EOR
F002 37
F010 4.9
F004 1
F005 RM
F006 These data are also cited in the European Chemicals Bureau IUCLID for CAS
* 8002-05-9. These data are ranked by API crude oil task group as '1',
* reliable without restrictions.
F007 These data are also cited in the European Chemicals Bureau IUCLID for CAS
* 8002-05-9. These data are ranked by API crude oil task group as '1',
* reliable without restrictions.
F020 4631
EOR
F002 37
F010 4.9
F004 1
F005 SO
F006 CONCAWE Bruxelles
**
F007 CONCAWE Bruxelles
**
F008 IUC4
F009 09-10-2003
F020 4644
EOR
F002 37
F010 4.9
F004 2
F005 RE
F006 Hoffman, D.J. and Albers, P.H. (1984)
** Evaluation of potential embryotoxicity and teratogenicity of 42
* herbicides, insecticides and petroleum contaminants to mallard eggs
** Arch. Environ. Contam. Toxicol., vol. 13, pp. 15-27.
F007 Hoffman, D.J. and Albers, P.H. (1984)
** Evaluation of potential embryotoxicity and teratogenicity of 42
* herbicides, insecticides and petroleum contaminants to mallard eggs
** Arch. Environ. Contam. Toxicol., vol. 13, pp. 15-27.
F008 IUC4
F009 13-11-2003
F020 4633
EOR
F002 37
F010 4.9
F004 2
F005 RM
F006 Crude oil from 4 locations was applied externally at different loadings

* to batches of 30 fertilized eggs of mallard ducks on day 3 of incubation
* at 37.5 °C. Eggs were candled each day to determine mortality, and dead
* embryos examined for ab
F007 Crude oil from 4 locations was applied externally at different loadings
* to batches of 30 fertilized eggs of mallard ducks on day 3 of incubation
* at 37.5 °C. Eggs were candled each day to determine mortality, and dead
* embryos examined for abnormalities. On day 18 of incubation, surviving
* embryos were examined for external malformations. The observed LD50
* values for crude oils from Kuwait, Prudhoe Bay, South Louisiana and Texas
* were 2.2, 8.3, 1.3 and 5.5 µl/egg, respectively. The South Louisiana and
* Texas crudes both produced significantly
** reduced growth, both above and below the LD50 value; preformed embryos
* were also found with both these crudes.
F020 4632
EOR
F002 37
F010 4.9
F004 2
F005 RM
F006 These data are also cited in the European Chemicals Bureau IUCLID for CAS
* 8002-05-9. These data are ranked by API crude oil task group as '4',
* reliability not assigned
F007 These data are also cited in the European Chemicals Bureau IUCLID for CAS
* 8002-05-9. These data are ranked by API crude oil task group as '4',
* reliability not assigned
F020 4634
EOR
F002 37
F010 4.9
F004 3
F005 RE
F006 Forns, J.M. (1977)
** The effects of crude oil on larvae of lobster, Homarus americanus.
** In: Proceedings of the API-EPA-US Coast Guard 1977 Oil Spill Conference
* - Prevention Behaviour, Control, Clean-up.
** American Petroleum Institute, Publ
F007 Forns, J.M. (1977)
** The effects of crude oil on larvae of lobster, Homarus americanus.
** In: Proceedings of the API-EPA-US Coast Guard 1977 Oil Spill Conference
* - Prevention Behaviour, Control, Clean-up.
** American Petroleum Institute, Publication No. 4284, pp. 569-573.
F008 IUC4
F009 13-11-2003
F020 4636
EOR
F002 37
F010 4.9
F004 3
F005 RM
F006 The effects of emulsified South Louisiana crude oil on the development of
* American Lobster, Homarus americanus, was investigated during the
* development of the first four larval stages over a 15-day period.
* Hatched larvae were exposed to co

F007 The effects of emulsified South Louisiana crude oil on the development of
* American Lobster, Homarus americanus, was investigated during the
* development of the first four larval stages over a 15-day period.
* Hatched larvae were exposed to concentrations of 0, 0.1 and 1.0 ppm crude
* oil, six times per day for periods ranging from 0.8 to 5.6 minutes, using
* a flow-through system. The tests showed that 1.0 ppm crude oil was a
* sub-lethal concentration for the lobster larvae and that no significant
* effects were found at 0.1 ppm. At 0.1 ppm crude oil, the survival rate
* of the larvae was comparable with that of the controls, but at 1.0 ppm
* the survival value was about 50% that of the control larvae. Other
* effects observed at 1.0 crude oil included: (a) lethargy, reduced
* feeding and lack of the characteristic aggression of the control
* animals, (b) an increase in development time from 12 to 15 days, and (c)
* a change in the pigmentation of the larvae from the normal pale blue,
* almost transparent, color to a sharp red appearance.
F020 4635
EOR
F002 37
F010 4.9
F004 3
F005 RM
F006 These data are also cited in the European Chemicals Bureau IUCLID for CAS
* 8002-05-9. These data are ranked by API crude oil task group as '4',
* reliability not assigned
F007 These data are also cited in the European Chemicals Bureau IUCLID for CAS
* 8002-05-9. These data are ranked by API crude oil task group as '4',
* reliability not assigned
F020 4637
EOR
F002 37
F010 4.9
F004 3
F005 SO
F006 CONCAWE Bruxelles
**
F007 CONCAWE Bruxelles
**
F008 IUC4
F009 09-10-2003
F020 4645
EOR
F002 37
F010 4.9
F004 4
F005 RE
F006 Burks, S.L. (1980)
** Effects of oil on aquatic organisms. A review of selected literature
** American Petroleum Institute, Publication No. 29-30299, API, Washington DC
F007 Burks, S.L. (1980)
** Effects of oil on aquatic organisms. A review of selected literature
** American Petroleum Institute, Publication No. 29-30299, API, Washington DC
F008 IUC4
F009 13-11-2003

F020 4639
EOR
F002 37
F010 4.9
F004 4
F005 RM
F006 Burks has extensively reviewed the effects of crude and refined oils on
* organisms found in fresh and sea water. He noted that where spillages
* occur the non-mobile species suffer the greatest mortality, whereas fish
* species can often escap
F007 Burks has extensively reviewed the effects of crude and refined oils on
* organisms found in fresh and sea water. He noted that where spillages
* occur the non-mobile species suffer the greatest mortality, whereas fish
* species can often escape from the affected region. The extent of the
* initial mortality depends on the chemical nature of the oil, the
* location, and the physical conditions, particularly the temperature and
* wind velocity. Most affected freshwater and marine communities recover
* from the effects of an oil spill within a year. The occurrence of
* biogenic hydrocarbons in the world's oceans is well recorded. They have
* the characteristic isoprenoid structure, and measurements made in water
* columns indicate a background concentration of 1.0 to 10 µl/l. The
* higher molecular weight materials are dispersed as particles, with the
* highest concentrations of about 20 µl/l occurring in the top 3 mm layer
* of water.
** A wide variation in the response of organisms to oil exposures has been
* noted. The larvae of fish and crustaceans appear to be most susceptible
* to the water-soluble fraction of crude oil. Exposures of plankton and
* algae have indicated that certain species of diatoms and green algae are
* inhibited, whereas microflagellates are not.
** For the most part, molluscs and most intertidal worm species appear to be
* tolerant of oil contamination.
F020 4638
EOR
F002 37
F010 4.9
F004 4
F005 RM
F006 These data are also cited in the European Chemicals Bureau IUCLID for CAS
* 8002-05-9. These data are ranked by API crude oil task group as '4',
* reliability not assigned
F007 These data are also cited in the European Chemicals Bureau IUCLID for CAS
* 8002-05-9. These data are ranked by API crude oil task group as '4',
* reliability not assigned
F020 4640
EOR
F002 37
F010 4.9
F004 4
F005 SO
F006 CONCAWE Bruxelles
**
F007 CONCAWE Bruxelles
**

F008 IUC4
F009 09-10-2003
F020 4646
EOR
F002 37
F010 4.9
F004 5
F005 RE
F006 Falk-Petersen, I-B.,(1979)
** Toxic effects of aqueous extracts of Ekofisk crude oil, crude oil
* fractions and commercial oil products on the development of sea urchin
* eggs
** Sarsia, vol.64, pp. 161-169.
F007 Falk-Petersen, I-B.,(1979)
** Toxic effects of aqueous extracts of Ekofisk crude oil, crude oil
* fractions and commercial oil products on the development of sea urchin
* eggs
** Sarsia, vol.64, pp. 161-169.
F008 IUC4
F009 13-11-2003
F020 4642
EOR
F002 37
F010 4.9
F004 5
F005 RM
F006 Falk-Petersen has studied the effects of the water-soluble fraction (WSF)
* of Ekofisk crude oil on the development of the eggs of two sea urchin
* species, Strongylocentrotus pallidus and S. droebachiensis. The WSF was
* prepared by shaking Ekof
F007 Falk-Petersen has studied the effects of the water-soluble fraction (WSF)
* of Ekofisk crude oil on the development of the eggs of two sea urchin
* species, Strongylocentrotus pallidus and S. droebachiensis. The WSF was
* prepared by shaking Ekofisk crude oil and sea water in the ratio 1:9 for
* 5 minutes, allowing to separate for 20 hours, and using the aqueous phase
* for the studies.
** Fertilized eggs were exposed to dilutions of the WSF for up to 9 days at
* 3 to 5 °C. Embryos and larvae were examined regularly by scanning and
* transmission electron microscopy after fixing in 2% OsO4 in sea water.
** Concentrations of 30% WSF, corresponding to 13 ppm dissolved oil, did not
* impair either development or the ultrastructure of the stages from egg to
* pluteus. However, significant effects on development were found at both
* 40%
** WSF (17 ppm oil) and 50% WSF (21 ppm oil). At these levels, the larvae
* filled with degenerating cells, and differentiation of the intestine and
* skeletal growth were inhibited compared with the control larvae.
F020 4641
EOR
F002 37
F010 4.9
F004 5
F005 RM
F006 These data are also cited in the European Chemicals Bureau IUCLID for CAS

* 8002-05-9. These data are ranked by API crude oil task group as '4',
* reliability not assigned
F007 These data are also cited in the European Chemicals Bureau IUCLID for CAS
* 8002-05-9. These data are ranked by API crude oil task group as '4',
* reliability not assigned
F020 4643
EOR
F002 37
F010 4.9
F004 5
F005 SO
F006 CONCAWE Bruxelles
**
F007 CONCAWE Bruxelles
**
F008 IUC4
F009 09-10-2003
F020 4647
EOR
F002 37
F010 5.1.1
F004 1
F005 ME
F006 Groups of five male and five female Sprague-Dawley rats were dosed once
* by oral gavage with the test material at a dose level of 5 g/kg.
** The animals were observed frequently on the day of treatment and daily
* thereafter for 14 days.
** The anim
F007 Groups of five male and five female Sprague-Dawley rats were dosed once
* by oral gavage with the test material at a dose level of 5 g/kg.
** The animals were observed frequently on the day of treatment and daily
* thereafter for 14 days.
** The animals were weighed on the day of treatment and again 7 and 14 days
* later.
F020 4648
EOR
F002 37
F010 5.1.1
F004 1
F005 RE
F006 Mobil (1984)
** The acute oral toxicity of beryl crude in albino rats
** MEHSL Study No. 40951
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1984)
** The acute oral toxicity of beryl crude in albino rats
** MEHSL Study No. 40951
** Mobil Environmental and Health Science Laboratory
F020 4649
EOR
F002 37
F010 5.1.1
F004 1

F005 RL
F006 Not clear whether this study was carried out to GLP and few
** experimental data given.
** Nevertheless, the study is sufficient to demonstrate an LD50 of greater
* than 5 g/kg.
F007 Not clear whether this study was carried out to GLP and few
** experimental data given.
** Nevertheless, the study is sufficient to demonstrate an LD50 of greater
* than 5 g/kg.
F008 IUC4
F020 4650
EOR
F002 37
F010 5.1.1
F004 1
F005 RS
F006 There were no deaths following treatment.
** During the first week post dosing lacrimation and a
** discharge covering the perineum (genital area) were
** observed.
** At termination of the study all animals appeared healthy and
** had gained weight.
** The
F007 There were no deaths following treatment.
** During the first week post dosing lacrimation and a
** discharge covering the perineum (genital area) were
** observed.
** At termination of the study all animals appeared healthy and
** had gained weight.
** The oral LD50 was judged to be greater than 5 g/kg.
F008 IUC4
F020 4651
EOR
F002 37
F010 5.1.1
F004 3
F005 RE
F006 Mobil (1984)
** The acute oral toxicity of Arab light crude in Albino rats
** Study No. 40961
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1984)
** The acute oral toxicity of Arab light crude in Albino rats
** Study No. 40961
** Mobil Environmental and Health Science Laboratory
F020 4652
EOR
F002 37
F010 5.1.1
F004 3
F005 RE
F006 Mobil (1984)
** The acute oral toxicity of MCSL crude (Midcontinent) in

** albino rats
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1984)
** The acute oral toxicity of MCSL crude (Midcontinent) in
** albino rats
** Mobil Environmental and Health Science Laboratory
F020 4653
EOR
F002 37
F010 5.1.1
F004 3
F005 RE
F006 Mobil (1990)
** Consolidated acute test report on Lost Hills light crude
** Study Nos. 63830, 63831, 63832, 63833
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1990)
** Consolidated acute test report on Lost Hills light crude
** Study Nos. 63830, 63831, 63832, 63833
** Mobil Environmental and Health Science Laboratory
F020 4654
EOR
F002 37
F010 5.1.1
F004 3
F005 RE
F006 Mobil (1997)
** Toxicological and compositional data on petroleum refinery heavy streams
** Mobil Environmental and Health Sciences Laboratory
F007 Mobil (1997)
** Toxicological and compositional data on petroleum refinery heavy streams
** Mobil Environmental and Health Sciences Laboratory
F020 4657
EOR
F002 37
F010 5.1.1
F004 3
F005 RE
F006 Smith, L. H., Haschek, W. M. and Witschi, H. (1981)
** Acute toxicity of selected crude and refined shale oil and
** petroleum-derived substances.
** In Greist, W. H.. et al (Eds)
** Health Investigation of Oil Shale develpment pp 141-160.
** Ann Arbor Sc
F007 Smith, L. H., Haschek, W. M. and Witschi, H. (1981)
** Acute toxicity of selected crude and refined shale oil and
** petroleum-derived substances.
** In Greist, W. H.. et al (Eds)
** Health Investigation of Oil Shale develpment pp 141-160.
** Ann Arbor Science, Mich.
F020 4655
EOR
F002 37

F010 5.1.1
F004 3
F005 RS
F006 The results of other acute toxicity studies which have been reported are
* as follows:
**
** Sample LD50 Clinical signs
** Light crudes
** Lost Hills Light (Mobil consolidated report)
** >5 g/kg Soft stool
** Urogenital discharge
** Decreased feca
F007 The results of other acute toxicity studies which have been reported are
* as follows:
**
** Sample LD50 Clinical signs
** Light crudes
** Lost Hills Light (Mobil consolidated report)
** >5 g/kg Soft stool
** Urogenital discharge
** Decreased fecal output
** Anal discharge
**
** MCSL Crude (midcontinent) Mobil study No. 40971
** >5 g/kg Decreased activity
** Hunching
** Discharge/perineal staining
** Arab Light (Mobil study No. 40961)
** >5 g/kg Discharge/perineal staining
** Belridge heavy (Mobil summarized data)
** >5 g/kg Mild gastrointestinal
effects
**
** Smith et al (1981) also demonstrated that three different
** crude oils (Crude type not specified) were non-toxic in male mice as
* follows:
**
** Wilmington Crude oil >16 g/kg
** Recluse crude oil >16 g/kg
** Mixed petroleum crudes >10 g/kg
F020 4656
EOR
F002 37
F010 5.1.3
F004 1
F005 ME
F006 The skin was clipped from the trunks of three male and three female New
* Zealand White rabbits prior to treatment with test material. The skin of
* the backs of three animals (2 male, 1 female) was abraded and the skin of
* the remaining animals
F007 The skin was clipped from the trunks of three male and three female New
* Zealand White rabbits prior to treatment with test material. The skin of
* the backs of three animals (2 male, 1 female) was abraded and the skin of

* the remaining animals was left intact. The liquid, undiluted test
* material was then applied as a single dose of 2 g/kg to the shorn back of
* all six animals.
** The test site was covered with a gauze and an occlusive
** wrap. The animals were also fitted with Elizabethan collars
** to prevent chewing of the occlusive covering and ingestion
** of test material.
** 24 hours after application of the test material the occlusive dressing
* was removed and any surplus test material was removed from the skin by
* gently wiping with cotton moistened with physiological saline.
** The animals were observed frequently on the day of treatment and daily
* thereafter for 14 days.
**
** Two hours after wiping the residual material from the skin
** (26 hours after treatment), the areas of application were
** assessed for skin irritation. Further evaluations for skin
** irritation were also made on the 3rd and 7th days following
** treatment.
F008 IUC4
F020 4658
EOR
F002 37
F010 5.1.3
F004 1
F005 RE
F006 Mobil (1984)
** The acute dermal toxicity of Beryl crude in albino rabbits
** Study No. 40952
** Mobil Environmental and Health Sciences laboratory
F007 Mobil (1984)
** The acute dermal toxicity of Beryl crude in albino rabbits
** Study No. 40952
** Mobil Environmental and Health Sciences laboratory
F008 IUC4
F020 4659
EOR
F002 37
F010 5.1.3
F004 1
F005 RL
F006 Not clear whether this study was carried out to GLP and few
** experimental data given.
F007 Not clear whether this study was carried out to GLP and few
** experimental data given.
F008 IUC4
F020 4660
EOR
F002 37
F010 5.1.3
F004 1
F005 RS
F006 There were no deaths in this study.
** A few animals had soft stool/diarrhea during the observation period,

* however, all animals were normal at study termination.
** At the end of the study five of the six animals had gained
** weight and one had lo
F007 There were no deaths in this study.
** A few animals had soft stool/diarrhea during the observation period,
* however, all animals were normal at study termination.
** At the end of the study five of the six animals had gained
** weight and one had lost a small amount of weight. [No actual body weight
* data are provided in the report].
**
** There was no evidence of systemic toxicity during the study
** although there was some skin reaction at the site of
** application.
** Following 24 hours of skin contact, varying degrees (slight, moderate)
* irritation were observed at the 26 and 72 hour readings. At the end of
* the first week, the skin response was barely perceptible or absent.
** The dermal LD50 was judged to be greater than 2 g/kg
F008 IUC4
F020 4661
EOR
F002 37
F010 5.1.3
F004 2
F005 RE
F006 Mobil (1984)
** The acute dermal toxicity of Arab light crude in albino
** rabbits
** Study No. 40962
** Mobil Environmental and Health Sciences laboratory
F007 Mobil (1984)
** The acute dermal toxicity of Arab light crude in albino
** rabbits
** Study No. 40962
** Mobil Environmental and Health Sciences laboratory
F020 4663
EOR
F002 37
F010 5.1.3
F004 2
F005 RE
F006 Mobil (1984)
** The acute dermal toxicity of MCSL crude (Midcontinent) in
** albino rabbits
** Study No. 40972
** Mobil Environmental and Health Sciences laboratory
F007 Mobil (1984)
** The acute dermal toxicity of MCSL crude (Midcontinent) in
** albino rabbits
** Study No. 40972
** Mobil Environmental and Health Sciences laboratory
F020 4662
EOR
F002 37

F010 5.1.3
F004 2
F005 RE
F006 Mobil (1990)
** Consolidated acute test report on Lost Hills light crude
** Study Nos. 63830, 63831, 63832, 63833
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1990)
** Consolidated acute test report on Lost Hills light crude
** Study Nos. 63830, 63831, 63832, 63833
** Mobil Environmental and Health Science Laboratory
F020 4664
EOR
F002 37
F010 5.1.3
F004 2
F005 RE
F006 Mobil (1997)
** Toxicological and compositional data on petroleum refinery heavy streams
** Mobil Environmental and Health Sciences Laboratory
F007 Mobil (1997)
** Toxicological and compositional data on petroleum refinery heavy streams
** Mobil Environmental and Health Sciences Laboratory
F020 4666
EOR
F002 37
F010 5.1.3
F004 2
F005 RS
F006 Additionally, acute dermal LD50s have also been reported for three other
* light crude oils.
** The results are as follows:
**
** Sample LD50 Clinical signs
** Lost Hills Light (Mobil Study No. 63831)
** >2 g/kg Soft stool
** Decreased food consump
F007 Additionally, acute dermal LD50s have also been reported for three other
* light crude oils.
** The results are as follows:
**
** Sample LD50 Clinical signs
** Lost Hills Light (Mobil Study No. 63831)
** >2 g/kg Soft stool
** Decreased food consumption
** Decreased fecal output
**
** MCSL Crude (midcontinent) Mobil study No. 40972
** >2 g/kg Diarrhea
** Nasal discharge
**
** Arab Light (Mobil study No. 40962)
** >2 g/kg No signs of toxicity

**
** Belridge Heavy (Mobil summarized data)
** >2 g/kg Mild gastrointestinal
effects
F020 4665
EOR
F002 37
F010 5.2.1
F004 1
F005 CL
F006 The classification according to various methods/criteria are summarized
* as follows:
**
** Guideline Score Rating

**
** DOT Corrosion Negative Non-corrosive
** EEC (4hr occluded) Erythema 1.6 Non-irritant
** Edema 1.3
**
** OSHA PII (4 h occl.) 2.8 Non-i
F007 The classification according to various methods/criteria are summarized
* as follows:
**
** Guideline Score Rating
**
** DOT Corrosion Negative Non-corrosive
** EEC (4hr occluded) Erythema 1.6 Non-irritant
** Edema 1.3
**
** OSHA PII (4 h occl.) 2.8 Non-irritant
** FHSA PII (24 h occl.) 3.6 Non-irritant
F008 IUC4
F020 4667
EOR
F002 37
F010 5.2.1
F004 1
F005 ME
F006 Prior to test the hair was clipped from the backs of six New Zealand
* White rabbits (sex not specified). Three one inch square sites
* (anterior, mid-dorsal and posterior) on the right side of each animal
* were lightly abraded. Three
** similarly
F007 Prior to test the hair was clipped from the backs of six New Zealand
* White rabbits (sex not specified). Three one inch square sites
* (anterior, mid-dorsal and posterior) on the right side of each animal
* were lightly abraded. Three
** similarly orientated sites were left intact on the left side of the
* animal, thus providing six test sites on each rabbit.
** 0.5 ml of test material was applied to each test site. The
** material applied to the anterior and mid-dorsal sites was
** covered with a Webril patch and these were occluded. The

** dorsal sites remained open. The rabbits were fitted with
** collars to prevent ingestion of the test material an chewing of the
* occluded patches.
**
** Four hours after treatment, the anterior sites were
** unwrapped. The intact site was immediately examined for
** evidence of corrosive effects and both sites were then gently wiped with
* a gauze. Thirty minutes later, the skin was evaluated for irritation. The
* sites were evaluated again 24 hours after patch removal. 48 hours after
* application, the 4 hour occluded intact site was re assessed for
* corrosion. Both sites were scored for irritation 4 hours later. The 4
* hour occluded sites were again re-evaluated for irritation on day 3.
**
** 24 hours after treatment, the two mid-dorsal sites were
** unwrapped and along with the posterior uncovered sites were
** wiped with cotton. The four sites were evaluated for
** irritation 2 hours later, again approximately 72 hours later and finally
* on the 7th day after treatment.
F008 IUC4
F020 4668
EOR
F002 37
F010 5.2.1
F004 1
F005 RE
F006 Mobil (1990)
** Consolidated acute test report on Lost Hills light crude
** Study Nos. 63830, 63831, 63832, 63833
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1990)
** Consolidated acute test report on Lost Hills light crude
** Study Nos. 63830, 63831, 63832, 63833
** Mobil Environmental and Health Science Laboratory
F008 IUC4
F009 03-10-2002
F020 4669
EOR
F002 37
F010 5.2.1
F004 1
F005 RS
F006 4 Hour occluded sites (DOT, OECD methods)
**
** Mean values (24, 48 & 72 hours) for erythema and edema at
** the intact sites were 1.69 and 1.3 respectively.
** The initial response of the skin to the test material was
** slight, with little difference
F007 4 Hour occluded sites (DOT, OECD methods)
**
** Mean values (24, 48 & 72 hours) for erythema and edema at
** the intact sites were 1.69 and 1.3 respectively.
** The initial response of the skin to the test material was
** slight, with little difference in response between intact or

** abraded sites.
**
** Actual scores were:
**

**		Intact		Abraded	
**		Erythema	Edema	Erythema	Edema
**	4.5 hrs	1.0	1.2	1.0	1.3
**	28 hrs	1.2	1.0	1.0	1.0
**	52 hrs	1.7	1.3	1.7	1.3
**	76 hrs	1.8	1.5	1.8	1.7
**	7 days	0.8	0.7	0.5	0.7
**	10 days	0	0.3	0	0

**
** 24 Hour occluded sites (FHSA method)
**
** Actual scores were:
**

**		Intact		Abraded	
**		Erythema	Edema	Erythema	Edema
**	26 hrs	1.7	1.7	1.7	1.7
**	72 hrs	2.3	1.5	2.2	1.7
**	7 days	1.2	0.7	0.8	0.8
**	10 days	0.2	0	0.2	0.2

**
**
**
** Non-occluded sites
**
** Actual scores were:
**

**		Intact		Abraded	
**		Erythema	Edema	Erythema	Edema
**	26 hrs	1.7	1.2	1.8	1.3
**	72 hrs	2.8	1.5	2.7	1.3
**	7 days	1.3	0.8	1.3	0.8
**	10 days	0.3	0	0.2	0

F008 IUC4
F020 4670
EOR
F002 37
F010 5.2.1
F004 2
F005 ME
F006 Prior to test the hair was clipped from the backs of six New Zealand
* White rabbits (sex not specified). Three one inch square sites
* (anterior, mid-dorsal and posterior) on the right side of each animal
* were lightly abraded. Three
** similarly
F007 Prior to test the hair was clipped from the backs of six New Zealand
* White rabbits (sex not specified). Three one inch square sites
* (anterior, mid-dorsal and posterior) on the right side of each animal
* were lightly abraded. Three
** similarly orientated sites were left intact on the left side of the

* animal, thus providing six test sites on each rabbit.
** 0.5 ml of test material was applied to each test site. The
** material applied to the anterior and mid-dorsal sites was
** covered with a Webril patch and these were occluded. The
** dorsal sites remained open. The rabbits were fitted with
** collars to prevent ingestion of the test material and chewing of the
* occluded patches.
**
** Four hours after treatment, the anterior sites were
** unwrapped. The intact site was immediately examined for
** evidence of corrosive effects and both sites were then gently wiped with
* a gauze. Thirty minutes later, the skin was evaluated for irritation. The
* sites were evaluated again 24 hours after patch removal. 48 hours after
* application, the 4 hour occluded intact site was re assessed for
* corrosion. Both sites were scored for irritation 4 hours later. The 4
* hour occluded sites were again re-evaluated for irritation on day 3.

**
** 24 hours after treatment, the two mid-dorsal sites were
** unwrapped and along with the posterior uncovered sites were
** wiped with cotton. The four sites were evaluated for
** irritation 2 hours later, again approximately 72 hours later and finally
* on the 7th day after treatment.
F008 IUC4
F020 4671
EOR
F002 37
F010 5.2.1
F004 2
F005 RE
F006 Mobil (1985)
** Skin irritation by Arab light crude after single
** applications, occluded and non-occluded, on albino rabbits
** Study No. 40964
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1985)
** Skin irritation by Arab light crude after single
** applications, occluded and non-occluded, on albino rabbits
** Study No. 40964
** Mobil Environmental and Health Science Laboratory
F008 IUC4
F020 4672
EOR
F002 37
F010 5.2.1
F004 2
F005 RL
F006 A textual description of the results only was available. No
** actual data were provided in the report.
F007 A textual description of the results only was available. No
** actual data were provided in the report.
F008 IUC4
F020 4673

EOR
F002 37
F010 5.2.1
F004 2
F005 RS
F006 4 Hour Occluded (OECD Method, DOT)
**
** Mean values (24, 48 & 72 hours) for erythema and edema at
** the intact sites were 0.9 and 0.1 respectively.
** The initial response of the skin to the test material was
** slight. Within 72 hours after treatment
F007 4 Hour Occluded (OECD Method, DOT)
**
** Mean values (24, 48 & 72 hours) for erythema and edema at
** the intact sites were 0.9 and 0.1 respectively.
** The initial response of the skin to the test material was
** slight. Within 72 hours after treatment, most sites had
** recovered, with little difference in response between intact or abraded
* sites.
**
** 24 Hour Occluded (FHSA method)
**
** The day following application of the test material a
** moderate erythema was observed. This subsided during the
** subsequent days such that there was only barely perceptible
** irritation at the end of the first week. Abrasion had no
** effect on either the initial response or the recovery.
**
** Non Occluded
**
** Uncovered treated sites had moderate erythema 24 hours after application
* of the test material. This response diminished and was virtually absent
* after one week.
** Abrasion had no effect on either the initial response or the recovery.
F008 IUC4
F020 4674
EOR
F002 37
F010 5.2.1
F004 3
F005 RE
F006 Mobil (1997)
** Toxicological and compositional data on petroleum refinery heavy streams
** Mobil Environmental and Health Sciences Laboratory
F007 Mobil (1997)
** Toxicological and compositional data on petroleum refinery heavy streams
** Mobil Environmental and Health Sciences Laboratory
F020 4676
EOR
F002 37
F010 5.2.1
F004 3
F005 RL

F006 Data only available in tabular form, but studies were part of a series of
* studies using same methods, all of which were reliability 1.
F007 Data only available in tabular form, but studies were part of a series of
* studies using same methods, all of which were reliability 1.
F020 4677
EOR
F002 37
F010 5.2.1
F004 3
F005 RS
F006 Summarized data for Belridge Heavy crude oil are:
**

** Guideline		Score	Rating
** DOT Corrosion		Negative	Non-corrosive
** EEC (4hr occluded)	Erythema 0.6		Non-irritant
**		Edema 0.8	
** OSHA PII (4 h occl.)	1.4		Non-irritant
** FHSA PII (24 h occl.)	2.1		

F007 Summarized data for Belridge Heavy crude oil are:
**

** Guideline		Score	Rating
** DOT Corrosion		Negative	Non-corrosive
** EEC (4hr occluded)	Erythema 0.6		Non-irritant
**		Edema 0.8	
** OSHA PII (4 h occl.)	1.4		Non-irritant
** FHSA PII (24 h occl.)	2.1		Non-irritant

F020 4675
EOR
F002 37
F010 5.2.2
F004 1
F005 ME
F006 0.1 ml of undiluted test material was instilled into one eye
** of each of six New Zealand White rabbits.
** The eyes were not washed and were evaluated for irritation
** at 1, 24, 48 and 72 hours after treatment. Fluorescein was
** used to aid the eva
F007 0.1 ml of undiluted test material was instilled into one eye
** of each of six New Zealand White rabbits.
** The eyes were not washed and were evaluated for irritation
** at 1, 24, 48 and 72 hours after treatment. Fluorescein was
** used to aid the evaluation of the 72 hour reading.
** Treated eyes that stained positively with fluorescein were
** examined again 7 and 14 days after treatment.
F008 IUC4
F020 4678
EOR
F002 37
F010 5.2.2

F004 1
F005 RE
F006 Mobil (1985)
** Primary eye irritation of Beryl crude in albino rabbits
** Study No. 40953
** Mobil Environmental and Health Sciences Laboratory
F007 Mobil (1985)
** Primary eye irritation of Beryl crude in albino rabbits
** Study No. 40953
** Mobil Environmental and Health Sciences Laboratory
F008 IUC4
F009 03-10-2002
F020 4679
EOR
F002 37
F010 5.2.2
F004 1
F005 RL
F006 The only reservation is that it is not clear whether this
** study was carried out according to GLP.
** Otherwise the study and reporting are sound.
F007 The only reservation is that it is not clear whether this
** study was carried out according to GLP.
** Otherwise the study and reporting are sound.
F008 IUC4
F020 4680
EOR
F002 37
F010 5.2.2
F004 1
F005 RS
F006 The mean irritation scores at each of the observation times
** were:
**

**	1 hr	24 hr	48 hr	72 hr
** Cornea	0	0	0	0
** Iris	0	0	0	0
** Conjunctivae	4.0	1.7	1.3	1.0

**
** The test material was judged to be non-irritating to the
** rabbit eye.
F007 The mean irritation scores at each of the observation times
** were:
**

**	1 hr	24 hr	48 hr	72 hr
** Cornea	0	0	0	0
** Iris	0	0	0	0
** Conjunctivae	4.0	1.7	1.3	1.0

**
** The test material was judged to be non-irritating to the
** rabbit eye.
F008 IUC4
F020 4681

EOR
F002 37
F010 5.2.2
F004 2
F005 RE
F006 Mobil (1985)
** Primary eye irritation of Arab light crude in albino rabbits
** Study N0. 40963
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1985)
** Primary eye irritation of Arab light crude in albino rabbits
** Study N0. 40963
** Mobil Environmental and Health Science Laboratory
F008 IUC4
F020 4682
EOR
F002 37
F010 5.2.2
F004 2
F005 RE
F006 Mobil (1985)
** Primary eye irritation of MSCL Crude (Midcontinent) in
** albino rabbits.
** Study No. 40973
** Mobil Environmental and Health Sciences Laboratory
F007 Mobil (1985)
** Primary eye irritation of MSCL Crude (Midcontinent) in
** albino rabbits.
** Study No. 40973
** Mobil Environmental and Health Sciences Laboratory
F008 IUC4
F020 4683
EOR
F002 37
F010 5.2.2
F004 2
F005 RE
F006 Mobil (1990)
** Consolidated acute test report on Lost Hills light crude
** Study Nos. 63830, 63831, 63832, 63833
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1990)
** Consolidated acute test report on Lost Hills light crude
** Study Nos. 63830, 63831, 63832, 63833
** Mobil Environmental and Health Science Laboratory
F008 IUC4
F009 03-10-2002
F020 4684
EOR
F002 37
F010 5.2.2
F004 2
F005 RE

F006 Mobil (1997)
** Toxicological and compositional data on petroleum refinery heavy streams
** Mobil Environmental and Health Sciences Laboratory
F007 Mobil (1997)
** Toxicological and compositional data on petroleum refinery heavy streams
** Mobil Environmental and Health Sciences Laboratory
F020 4686
EOR
F002 37
F010 5.2.2
F004 2
F005 RS
F006 Eye irritation studies have been reported for three other
** samples of light crude oil.
** There were no effects on either the iris or cornea. The only effects
* recorded were on the conjunctivae.
** The mean irritation scores (conjunctivae only) are
F007 Eye irritation studies have been reported for three other
** samples of light crude oil.
** There were no effects on either the iris or cornea. The only effects
* recorded were on the conjunctivae.
** The mean irritation scores (conjunctivae only) are as
** follows:
**

** Crude type		Result			Ref
**	1 hr	24 hr	48 hr	72 hr	
** MCSL (Midcontinent)					
**	2.3	0.3	0	0.7	Mobil 40973
**					
** Lost Hills Light					
**	8.0	3.7	2.7	1.7	Mobil 63832
**					
** Arab Light					
**	5.3	1.3	0.7	0.3	Mobil 40963
** Belridge Heavy		0.9	0.8	0.9	Mobil 1997

F020 4685
EOR
F002 37
F010 5.3
F004 1
F005 ME
F006 Concentrations of test material used in this sensitization
** study were determined in a pre screening study.
**
** Induction phase
** 0.4 ml of a 15% w/w concentration of test material in
** mineral oil was applied using a Hill Top Chamber with a 25
** mm
F007 Concentrations of test material used in this sensitization
** study were determined in a pre screening study.
**
** Induction phase
** 0.4 ml of a 15% w/w concentration of test material in

** mineral oil was applied using a Hill Top Chamber with a 25
** mm Webril swatch to the shorn backs of 10 male and 10 female Guinea pigs.
* The patch was occluded for six hours.
** The patches were applied to the same site once per week for
** 3 weeks. After each 6-hour exposure period, he patches were
** removed and the skin wiped with gauze moistened with mineral oil.
** The positive control material (DNCB) was applied at a
** concentration of 0.05% w/w in 70% ethanol to 5 male and 5
** female Guinea pigs. Treatment of the positive control
** animals was the same as for the test animals, except that
** the skin was wiped with saline after patch removal.
**
** Challenge
** Challenge patch application was performed 14 days after the
** last induction dose had been applied.
** Dual challenge patches were applied to fresh application
** sites of previously shorn skin of the animals. The test
** material was applied at concentrations of 10 and 15% w/w in
** mineral oil. The patches were then occluded for 6 hours.
** On the day following challenge patch application, the skin
** was depilated and 2 hours later and scored for signs of
** sensitization. The sites were examined after a further 48
** hours but this time without depilation.
** Naive and positive control animals were challenged with DNCB at a
* concentration of 0.05% in acetone.
F008 IUC4
F020 4687
EOR
F002 37
F010 5.3
F004 1
F005 RE
F006 Mobil (1991)
** Delayed contact hypersensitivity study in Guinea pigs
** (Buehler sensitization test) of Lost Hills light crude oil
** Study No. 63841
** Environmental and Health Sciences Laboratory
F007 Mobil (1991)
** Delayed contact hypersensitivity study in Guinea pigs
** (Buehler sensitization test) of Lost Hills light crude oil
** Study No. 63841
** Environmental and Health Sciences Laboratory
F008 IUC4
F020 4688
EOR
F002 37
F010 5.3
F004 1
F005 RS
F006 The results were as follows:

**	No. responding	Mean erythema score
** Test material (10% concentration)		

**	24 hrs induced	3/19	0.2
**	24 hrs control	0/10	0
**			
**	48 hrs induced	3/19	0.3
**	48 hrs control	2/10	0.2

** Test material (15% concentrat
F007 The results were as follows:

**		No. responding	Mean erythema score
** Test material (10% concentration)			
**	24 hrs induced	3/19	0.2
**	24 hrs control	0/10	0
**			
**	48 hrs induced	3/19	0.3
**	48 hrs control	2/10	0.2
**			
** Test material (15% concentration)			
**	24 hrs induced	2/19	0.2
**	24 hrs control	2/10	0.2
**			
**	48 hrs induced	1/19	0.1
**	48 hrs control	1/10	0.1
**			
** DNCB positive control			
**	24 hrs induced	5/5	3.4
**	24 hrs control	0/5	0
**			
**	48 hrs induced	5/5	2.6
**	48 hrs control	0/5	0

** Although two test animals responded, the test material was
** judged to be non-sensitizing, since the test criterion
** required an mean erythema score of +2 for a positive
** response.
F008 IUC4
F020 4689
EOR
F002 37
F010 5.3
F004 2
F005 RE
F006 Mobil (1991)
** Delayed contact hypersensitivity study in Guinea pigs
** (Buehler sensitization test) of Belridge Heavy crude oil
** Study No. 63853
** Environmental and Health Sciences Laboratory
F007 Mobil (1991)
** Delayed contact hypersensitivity study in Guinea pigs
** (Buehler sensitization test) of Belridge Heavy crude oil
** Study No. 63853
** Environmental and Health Sciences Laboratory
F020 4691

EOR
F002 37
F010 5.3
F004 2
F005 RS
F006 A similar Buehler test of Belridge heavy crude oil was also
** negative.
F007 A similar Buehler test of Belridge heavy crude oil was also
** negative.
F020 4690
EOR
F002 37
F010 5.4
F004 1
F005 ME
F006 Two separate but identical studies are reported, one for
** each of two crude oils (Crude I and Crude II). The methods
** used were identical for both studies.
**
** Undiluted test material was applied to the shorn unoccluded
** skin of groups of ten mal
F007 Two separate but identical studies are reported, one for
** each of two crude oils (Crude I and Crude II). The methods
** used were identical for both studies.
**
** Undiluted test material was applied to the shorn unoccluded
** skin of groups of ten male and ten female Sprague-Dawley
** rats at doses of 30, 125 and 500 mg/kg/day.
** Application was once daily, five times each week for 13
** weeks. Groups of ten rats of each sex served as untreated
** controls.
** Each animal was fitted with an Elizabethan collar to prevent ingestion of
* the applied test material.
** At the end of each week residual test material was wiped
** from the backs of the animals.
** Animals were observed regularly for clinical signs of
** toxicity and body weights were recorded weekly.
** The animals were sacrificed during week 14 of the study
** after fasting overnight.
** All animals were necropsied and blood samples were taken for a range of
* hematological and serum chemistry determinations.
** A range of organs were weighed and tissues examined
** histologically.
**
** An additional two groups of ten males were treated at a
** dose level of 0 and 500 mg/kg/day for an evaluation of male
** reproductive health.
** For these animals testes weight and cauda epididymis weights were
* recorded. Additionally, the number of sperm and % normal sperm in the
* cauda were recorded as well as the number of spermatids in the testis.
F008 IUC4
F020 4692
EOR

F002 37
F010 5.4
F004 1
F005 RE
F006 Feuston, M. H., Mackerer, C. M., Schreiner, C. A. and
** Hamilton, C. E. (1997)
** Systemic toxicity of dermally applied crude oils in rats
** J. Tox. Env. Health, Vol 51, pp 387-399
F007 Feuston, M. H., Mackerer, C. M., Schreiner, C. A. and
** Hamilton, C. E. (1997)
** Systemic toxicity of dermally applied crude oils in rats
** J. Tox. Env. Health, Vol 51, pp 387-399
F008 IUC4
F020 4693
EOR
F002 37
F010 5.4
F004 1
F005 RE
F006 Mobil (1992)
** Thirteen-week dermal administration of Lost Hills Light to
** rats
** Study No. 63834
** Environmental Health and Sciences Laboratory
F007 Mobil (1992)
** Thirteen-week dermal administration of Lost Hills Light to
** rats
** Study No. 63834
** Environmental Health and Sciences Laboratory
F008 IUC4
F020 4694
EOR
F002 37
F010 5.4
F004 1
F005 RS
F006 No animals died in either of the studies and there were no
** clinical signs of systemic toxicity attributable to either
** crude oil. Both crude oils caused the same minimal skin
** irritation (flaking) at the exposure site.
**
** Body weights
** Apart fro
F007 No animals died in either of the studies and there were no
** clinical signs of systemic toxicity attributable to either
** crude oil. Both crude oils caused the same minimal skin
** irritation (flaking) at the exposure site.
**
** Body weights
** Apart from the 500 mg/kg/day group for Crude II that gained
** less weight than the controls (217g compared to 247g over
** the course of the study) all other body weight measurements
** were similar to the respective controls.

**

** Hematology

** Significant changes relative to their respective controls

** only occurred in the highest dose group animals (500

** mg/kg/day). The % increases (+) or decreases (-) that

** occurred in these dose groups are shown in the following

** table. [Changes are shown to the nearest whole number]

**

** Parameter	Crude I		Crude II	
**	Males	Females	Males	Females
** RBC	-6%	NSD	-8%	-7%
** Hemoglobin	-6%	NSD	-11%	-8%
** Hematocrit	-5%	NSD	-9%	-8%

** Platelets	NSD	NSD	-19%	NSD

**

** NSD No significant difference

**

** Serum chemistry

** Although more parameters were affected in the high dose

** Crude II females, there was no consistent pattern of change

** that could be attributed to the two crude oils. The

** differences %+ or %- are shown in the table below.

**

**	Dose group (mg/kg/day)					
**	Crude I			Crude II		
**	30	125	500	30	125	500
** MALES						
**						
** Calcium			-7%			
** Glucose		+13%	+13%			
** Urea nitrogen			+53%			
** Uric acid						-27%
**						
** FEMALES						
**						
** ALT						-18%
** Cholesterol					+41%	+60%
** Glucose	+16%		+20%			
** Potassium			-11%			-13%
** Urea nitrogen						+31%
** Uric acid						-27%

**

** Necropsy findings

** There were no treatment-related observations at necropsy.

** With the exception of the liver and thymus, there were no

** absolute or relative organ weight changes when compared to

** controls.

** The organ weight differences that were observed were

** confined to the 125 and 500 mg/kg/day groups and are

** summarized below.

**

** Organ	Crude I	Crude II

** wt		125	500	125	500
**					
** MALES					
**					
** Liver	(absolute)	+20%		+22%	
**	(relative)	+22%	+18%	+33%	
** Thymus	(absolute)			-41%	
**					
** FEMALES					
**					
** Liver	(absolute)	+13%		+24%	
**	(relative)	+12%		+31%	
** Thymus	(absolute)			-35%	
**	(relative)			-27%	

** Histopathology
**
** Changes in the skin included hyperplasia and an associated
** dermal inflammatory cell infiltration. In general the
** effects were slightly more severe in the animals treated
** with Crude I. All male and female animals exposed to Crude I
** were affected whereas at the lowest dose level 8/10 male and 6/10 females
* exposed to Crude II were affected.
**
** Other histopathological findings were associated with the
** bone marrow, Liver, thymus and thyroid although effects
** generally were greater with crude II than with crude I.
** The findings are tabulated below
**
** Crude I
**
** Bone marrow No effects in either sex at any dose level
**
** Liver Multifocal, mononuclear cell infiltration in 3 males and 2
* females at 500 mg/kg/day.
** Multifocal hepatocellular vacuolation in 3 females at 500 mg/kg/day
* only
**
** Thymus Atrophy in one male and two females at 500
** mg/kg/day only
**
** Thyroid Hypertrophy and hyperplasia of follicular
** epithelium in some males and females at all dose levels
**
** Crude II
**
** Bone marrow Increased cellularity in 2 males at each dose level (6
* males in highest dose group) and focal necrosis in 2 males in the 50
* mg/kg/day group.
** In the females increased cellularity was

observed in 9/10 animals
**
** Liver Hepatocellular vacuolation in one
male and one female in the
* 500 mg/kg/day group.
** Mononuclear cell infiltration was also
observed but in only one
* male in the highest dose group
**
** Thymus Atrophy was observed in six males and 7
females in the 500
* mg/g/day group.
**
** Thyroid Hypertrophy and hyperplasia was seen in a
few animals at all
* dose levels.
**
** The spermatozoa/spermatid evaluations that were conducted on the separate
* 500 mg/kg/day group of animals exposed to crude I did not reveal any
* effects when compared to a control.
F008 IUC4
F020 4695
EOR
F002 37
F010 5.4
F004 1
F005 TS
F006 Two crude oils were examined in the paper by Feuston et al
** The characteristics of the crude oils was reported as:
**
** Chemical class Crude I Crude II
**
** Nonaromatics 50.0 37.3
** <3-Ring PAH 35.3 41.7
** 3- to 5- Ring PAH 10.2 15.7
** 3- Ring PAH 5.9
F007 Two crude oils were examined in the paper by Feuston et al
** The characteristics of the crude oils was reported as:
**
** Chemical class Crude I Crude II
**
** Nonaromatics 50.0 37.3
** <3-Ring PAH 35.3 41.7
** 3- to 5- Ring PAH 10.2 15.7
** 3- Ring PAH 5.9 8.1
** 4- Ring PAH 2.2 4.0
** 5- Ring PAH 2.1 3.6
** Sulfur-PAC 2.4 2.9
** Nitrogen (N)-PAC 5.4 8.4
** Nonbasic N-PAC 3.8 5.9
**
** Total 103.3 106.0
**

** From information available in the Mobil report of study No
** 63834 and other laboratory data, it is clear that Crude I is Lost Hills
* Light Crude and Crude II is Belridge heavy crude oil.
F008 IUC4
F020 4696
EOR
F002 37
F010 5.4
F004 2
F005 ME
F006 Two pilot studies were conducted with groups of 5 male and 5 female mice.
* In these studies Prudhoe Bay crude oil was
** administered by gavage at doses of 0, 5 and 10 ml/kg daily
** for five days. No other experimental details are given.
** However,
F007 Two pilot studies were conducted with groups of 5 male and 5 female mice.
* In these studies Prudhoe Bay crude oil was
** administered by gavage at doses of 0, 5 and 10 ml/kg daily
** for five days. No other experimental details are given.
** However, the results led the author to conduct four further
** follow-up studies and these are described below.
**
** Experiment 1 (Three crudes, each at a single dose level)

** One of three crude oils or Bunker C oil was administered by
** gavage, daily at a single dose level of 10 ml/kg for 5 days
** to groups of 10 male mice. A further group of 10 male mice
** were intubated each day but no material was administered;
** these animals served as controls.
**
** Experiment 2 (One crude, single dose level)
** Groups of 10 male mice were given either Prudhoe Bay crude
** oil or mineral oil USP or corn oil at a dose of
** 10 ml/kg/day for five days. A separate group of 10 controls were
* intubated but no material was administered. This study was to compare the
* effect of one crude oil with the effects of two oils expected to be
* non-toxic.
**
** Experiment 3 (One crude, five dose levels)
** Prudhoe bay crude oil was administered to 10 or 11 male mice at dose
* levels of 2, 4, 8, 12 or 16 ml/kg/day. A further group of 10 male mice
* wee sham treated and served as controls.
**
** A fourth experiment was also carried out which studied the
** effect of crude oil exposure on vitamin E and
** selenium-deficient animals. This study is not summarized
** here.
**
** In all experiments 1-3 the animals were weighed on the day
** they were first dosed and again prior to necropsy. The
** differences in weights were recorded as change in body
** weight during the studies. Necropsy was 24 hours after the
** 5th dose had been administered.

** At necropsy, blood samples were taken for determination of
** blood cell counts, packed cell volume (PCV), hemoglobin and red cell
* indices (MCHC). Blood smears were stained with
** methylene blue to detect the presence of Heinz bodies in red cells.
** Liver was removed and weighed whereas the spleen and thymus
** were weighed after fixation. Portions of all major organs
** were fixed for subsequent histological examination.
**
** Data were analyzed by analysis of variance and regression
** procedures of the SAS programs (SAS Institute Inc., Cary,
** NC)
F008 IUC4
F020 4697
EOR
F002 37
F010 5.4
F004 2
F005 RE
F006 Leighton, F. A. (1990)
** The systemic toxicity of Prudhoe Bay crude and other
** petroleum oils to CD-1 mice.
** Arch. Environ. Contam. Toxicol Vol 19, pp257-262
F007 Leighton, F. A. (1990)
** The systemic toxicity of Prudhoe Bay crude and other
** petroleum oils to CD-1 mice.
** Arch. Environ. Contam. Toxicol Vol 19, pp257-262
F008 IUC4
F020 4698
EOR
F002 37
F010 5.4
F004 2
F005 RL
F006 These studies were probably not carried out according to GLP
** and the studies did not include measurement of all the
** parameters normally measured in repeat dose studies.
** Although the results are of limited value they do
** nevertheless add some
F007 These studies were probably not carried out according to GLP
** and the studies did not include measurement of all the
** parameters normally measured in repeat dose studies.
** Although the results are of limited value they do
** nevertheless add some information on the effect of the
** repeated exposure of mice to crude oil.
F008 IUC4
F020 4699
EOR
F002 37
F010 5.4
F004 2
F005 RS
F006 Although several studies were reported, the only studies
** summarized here are study A in which several crude oils are

** tested at a single dose level and study C in which PBCO is
** examined at five different dose levels.
**
** Neither information on
F007 Although several studies were reported, the only studies
** summarized here are study A in which several crude oils are
** tested at a single dose level and study C in which PBCO is
** examined at five different dose levels.
**
** Neither information on oils other than crude oil or on the
** effect of Vitamin E and selenium-depletion are included in
** this robust summary.
**
** In the pilot studies it was reported that the oils were
** distasteful to the mice and that after the first dose,
** administration of test material was difficult. Several mice
** from each group died from inhalation of oil that occurred
** during the oral dosing.
** The author stated that the pilot studies demonstrated a dose related
* decrease (8-11%) in PCV, a reduction in body weight gain, a 74% increase
* in liver weight per unit body weight and a 66% reduction in thymus
* weight. No data are given but this was the reason given for the further
* studies that were carried out.
**
** Although the studies carried out were separate, the author
** reported the results together and these are summarized
** below.
**
** Hematology
** It was reported (but no data presented) that in experiment A none of the
* oils tested resulted in significant changes in PCV, number of red blood
* cells or whole blood hemoglobin.
** The following table summarizes the hematological results
** obtained with PBCO only.
**

** Study	No of mice	Daily dose PBCO (ml/kg/day)	PCV	MCHC (g/l)
** A	10	0	0.43±0.02	336±22
**	8	10	0.42±0.04	328±10
** C	11	0	0.43±0.02	328±11
**	8	12	0.40±0.03	348±3*

**
** * p<0.05, analysis of variance with Tukey's multiple range procedure
**
** Body and organ weights
** The organ and body weight changes are summarized in the
** following tables
** Study A comparison of three different crude oils

**	Treatment group			
** Control	PBCO	SLCO	ALCO	

**

** Group size

** 10 8 6 7

**

** Change in body wt (g)

** +1.74±0.88 -0.33±1.5 -0.77±2.36* -1.43±2.28*

**

** Liver wt (g)

** 2.15±0.2 3.03±0.54* 2.73±0.5 2.66±0.43*

**

** Liver/body wt (g/g)

** 0.07±0 0.11±0.01* 0.10±0.01* 0.09±0.01*

**

** Spleen/body wt (g/g)

** 3.52±0.61 2.46±0.24* 2.49±0.29* 2.17±0.66*

**

** Thymus/body wt (g/g)

** 2.26±0.46 0.85±0.33* 1.01±0.41* 1.12±0.46*

**

** * P<0.05 Analysis of variance with Tukey's multiple range procedure

**

**

** Study C: PBCO at 5 dose levels

**

**	Body wt change (g)	Liver wt (g)	Organ/body wt (g/g) liver	spleen	thymus
**	Control group (10 mice)				
**	+3.14±1.33	1.95±.18	.06±.01	3.9±.49	2.7±.29
**	2 ml/kg/day (10 mice)				
**	-0.88±2.57	2.32±.22	.09±0	2.65±.62	1.61±.49*
**	4 ml/kg/day (9 mice)				
**	-0.86±1.24	2.38±.33*	.09±.01*	2.57±.23*	1.35±.32*
**	8 ml/kg/day (8 mice)				
**	-1.66±2.03*	2.58±.48*	.1±.01*	2.31±.67*	1.03±.4*
**	12 ml/kg/day (9 mice)				
**	-1.66±1.71*	2.69±.27*	.1±.01*	2.2±.44*	.71±.22*
**	16 ml/kg/day (7 mice)				
**	-3.29±1.4*	2.15±.35	.1±.01*	2.26±.87*	.65±.15*
**	Coefficient of determination (R^2)				
**	0.49***	0.08**	0.48***	0.31***	0.64***

**

** * P<0.05 Analysis of variance with Tukey's multiple range procedure

** ** P<0.03 Analysis of variance in regression

** *** P<0.001 Analysis of variance in regression

**
** Morphology
** There were no gross abnormalities at necropsy except a
** reduction in the size of thymus glands in those animals that had received
* crude oil.
** Liver, kidney, spleen, lung and thymus from groups of 5 mice that had
* received 0, 5 or 10 ml PBCO for 5 days in the pilot study were examined
* histologically. The thymus glands of mice in the 10 ml/kg/day group had
* very thin cortices and reduced densities of lymphocytes in the remaining
* cortex compared to controls.
** Hepatocytes from these same animals had uniformly dense
** cytoplasm while control mouse hepatocytes had large areas of rarified
* cytoplasm typical of normal glycogen-replete mouse livers. The histology
* of the thymus glands of the 5
** ml/kg/day group was intermediate between the controls and
** the 10 ml/kg/day group.
**
** There were no histological lesions in the other tissues
** examined.
**
** Overall, the results demonstrated minor hematological
** change, liver enlargement and atrophy of spleen and thymus.
F008 IUC4
F020 4700
EOR
F002 37
F010 5.4
F004 2
F005 TS
F006 The following crude oils were examined:
** Prudhoe Bay crude oil (PBCO) [a heavy crude]
** South Louisiana crude oil (SLCO) [a light crude]
** Arabian Light crude oil (ALCO)
** In addition a bunker C oil was examined
** A USP grade mineral oil and a corn
F007 The following crude oils were examined:
** Prudhoe Bay crude oil (PBCO) [a heavy crude]
** South Louisiana crude oil (SLCO) [a light crude]
** Arabian Light crude oil (ALCO)
** In addition a bunker C oil was examined
** A USP grade mineral oil and a corn oil were also included in the studies.
F008 IUC4
F020 4701
EOR
F002 37
F010 5.5
F004 1
F005 CL
F006 A modified Ames assay conducted on five different crudes
** demonstrated that all but one (Lost Hills light) were
** mutagenic. The Lost Hills Light did not demonstrate any
** mutagenic activity.
F007 A modified Ames assay conducted on five different crudes

** demonstrated that all but one (Lost Hills light) were
** mutagenic. The Lost Hills Light did not demonstrate any
** mutagenic activity.
F008 IUC4
F020 4702
EOR
F002 37
F010 5.5
F004 1
F005 ME
F006 DMSO extracts were prepared by mixing 2 ml test material
** with 3 ml cyclohexane to homogeneity. 10 ml DMSO was then
** added and mixed thoroughly. The mixture was vortexed every 5
** minutes for a total of 30 minutes. After 30 minutes the
** mixture
F007 DMSO extracts were prepared by mixing 2 ml test material
** with 3 ml cyclohexane to homogeneity. 10 ml DMSO was then
** added and mixed thoroughly. The mixture was vortexed every 5
** minutes for a total of 30 minutes. After 30 minutes the
** mixture was centrifuged and the DMSO layer (extract) removed
** and stored for testing.
**
** The extract was only tested with metabolic activation in
** Salmonella typhimurium strain TA 98 at the following doses:
** 1, 3, 5, 7, 10, 15, 25 and 50 µl/plate. Additionally the
** DMSO extract of a carcinogenic oil was also tested in the
** same manner.
** Positive control chemicals were: 2-aminoanthracene (2 ug)and
** benzoa(a)pyrene (5 ug).
** The DMSO extract of a refrigerator oil was use as negative
** control (50 ul).
** The metabolic activation mixture was derived from
** Araclor-induced hamster liver and this was use at eight
** times the standard concentration (0.4 ml rather than 0.05
** ml).
**
** The test material and control substances were added to tubes
** at the doses shown above and were incubated for 20 minutes
** with Salmonella broth culture. Colonies of histidine
** prototrophs were counted 48 hours after plate incubation.
**
** Assay acceptance criteria
** A linear dose response curve for mutagenicity must be
** obtained before the test material can be ranked for
** potency
**
** Spontaneous and solvent control reversion rates for TA98
** must fall between 20 and 50 revertants /plate
**
** The slope of the dose response curve for the positive
** control carcinogenic oil must fall between 1.5 - 4.5 net
** revertants/plate
**

** Provided the above criteria are met the net revertants per
** plate is plotted against dose and the best initial straight
** line response is determined by regression analysis.
** Test materials producing slopes less than 0.5 net
** revertants/µl are considered inactive.
F008 IUC4
F020 4703
EOR
F002 37
F010 5.5
F004 1
F005 RE
F006 Mobil (1984)
** A modified Ames pre-incubation assay for the determination
** of specific mutagenicity of the DMSO extract of Arab light
** crude
** Study No. 40965
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1984)
** A modified Ames pre-incubation assay for the determination
** of specific mutagenicity of the DMSO extract of Arab light
** crude
** Study No. 40965
** Mobil Environmental and Health Science Laboratory
F008 IUC4
F020 4704
EOR
F002 37
F010 5.5
F004 1
F005 RE
F006 Mobil (1984)
** A modified Ames pre-incubation assay for the determination
** of specific mutagenicity of the DMSO extract of MCSL crude
** (Midcontinent)
** Study No. 40975
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1984)
** A modified Ames pre-incubation assay for the determination
** of specific mutagenicity of the DMSO extract of MCSL crude
** (Midcontinent)
** Study No. 40975
** Mobil Environmental and Health Science Laboratory
F008 IUC4
F020 4705
EOR
F002 37
F010 5.5
F004 1
F005 RE
F006 Mobil (1984)
** A modified Ames pre-incubation mutagenesis assay for
** determination of specific mutagenicity of the DMSO extract

** of Beryl crude
** Mobil Health and Environmental Science Laboratory Study
** 40955
F007 Mobil (1984)
** A modified Ames pre-incubation mutagenesis assay for
** determination of specific mutagenicity of the DMSO extract
** of Beryl crude
** Mobil Health and Environmental Science Laboratory Study
** 40955
F008 IUC4
F020 4706
EOR
F002 37
F010 5.5
F004 1
F005 RE
F006 Mobil (1990)
** A modified Ames pre-incubation mutagenesis assay for
** determination of specific mutagenicity of the DMSO extract
** of Belridge Heavy
** Study No. 63850
** Mobil Health and Environmental Science Laboratory
F007 Mobil (1990)
** A modified Ames pre-incubation mutagenesis assay for
** determination of specific mutagenicity of the DMSO extract
** of Belridge Heavy
** Study No. 63850
** Mobil Health and Environmental Science Laboratory
F008 IUC4
F020 4707
EOR
F002 37
F010 5.5
F004 1
F005 RE
F006 Mobil (1990)
** A modified Ames pre-incubation mutagenesis assay for
** determination of specific mutagenicity of the DMSO extract
** of Lost Hills Light
** Study No. 63838
** Mobil Health and Environmental Science Laboratory
F007 Mobil (1990)
** A modified Ames pre-incubation mutagenesis assay for
** determination of specific mutagenicity of the DMSO extract
** of Lost Hills Light
** Study No. 63838
** Mobil Health and Environmental Science Laboratory
F008 IUC4
F020 4708
EOR
F002 37
F010 5.5
F004 1

F005 RL
F006 It is not clear whether the studies were carried out
** according to GLP. Furthermore, the assay was designed as a
** screen for carcinogenic activity However, the results are
** useful to identify the mutagenic potential of crude oils.
F007 It is not clear whether the studies were carried out
** according to GLP. Furthermore, the assay was designed as a
** screen for carcinogenic activity However, the results are
** useful to identify the mutagenic potential of crude oils.
F008 IUC4
F020 4709
EOR
F002 37
F010 5.5
F004 1
F005 RM
F006 Similar studies were conducted for Arab light, MCSL, Lost
** Hills light and Belridge Heavy crudes.
** The results were:
**

	Crude		Slope	Reference
**	Arab light	3.8		Mobil study 40965
**	MCSL crude	1.5		Mobil study 40975
**	Belridge Heavy		1.7	Mobil st

F007 Similar studies were conducted for Arab light, MCSL, Lost
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**

	Crude		Slope	Reference
**	Arab light	3.8		Mobil study 40965
**	MCSL crude	1.5		Mobil study 40975
**	Belridge Heavy		1.7	Mobil study 663850
**	Lost Hills light	0		Mobil study 63838

F008 IUC4
F020 4710
EOR
F002 37
F010 5.5
F004 1
F005 RS
F006 The No. revertants for each dose level of Beryl crude are as
** follows (NB data for the carcinogenic oil are not shown):
**

	Dose (µl)	No. of revertants
**	0 (Spontaneous)	43
**	0 (solvent control)	39
**	50	113
**	25	85
**	15	74
**	10	67
**	7	61

F007 The No. revertants for each dose level of Beryl crude are as
** follows (NB data for the carcinogenic oil are not shown):

**

**	Dose (µl)	No. of revertants	
**	0 (Spontaneous)		43
**	0 (solvent control)	39	
**	50		113
**	25		85
**	15		74
**	10		67
**	7		61
**	5		57
**	3		47
**	1		48
**	2-AA control	540	
**	BaP control	366	
**	Refrigerator oil	43	

**
** The slope for the Beryl crude was determined to be 2.5
** The slope for the carcinogenic oil was 3.7
F008 IUC4
F020 4711
EOR
F002 37
F010 5.5
F004 2
F005 ME
F006 Preparation of DMSO extract
** 10 ml DMSO was added to approximately 2 ml test substance.
** The tube containing the mixture was vortexed every 5 minutes for one
* minute. After 30 minutes, the mixture was
** centrifuged and the DMSO layer (Extract) w
F007 Preparation of DMSO extract
** 10 ml DMSO was added to approximately 2 ml test substance.
** The tube containing the mixture was vortexed every 5 minutes for one
* minute. After 30 minutes, the mixture was
** centrifuged and the DMSO layer (Extract) was removed for
** testing.
**
** Control substances:
** Solvent control: DMSO
** Negative control: Untreated flasks
** Positive controls: Cyclophosphamide monohydrate at
** concentration of 10 µg/ml
**
** Duplicate cultures were used - one set for metaphase
** analysis and the other for a determination of cytotoxicity
** (Mitotic Index). All cultures were conducted with metabolic
** activation.
**
** CHO cells were exposed for two hours to test material at
** concentrations of 1, 2.5, 5, 10, 15 and 20 µl/ml.
** After two hours, treatment was terminated and the cultured
** cells were washed and re-fed with complete culture medium.
** Approximately 16 hours later, colcicine was added and two

** hours later the cells were harvested as described below.
**
** Metaphase analysis
** Metaphase cells were collected by tapping a flask to release the loosely
* attached mitotic cells into culture medium and pelleting them by
* centrifugation. The cells were resuspended and then fixed in
* methanol:acetic acid (3:1) and stored refrigerated until slide
* preparation.
** A minimum of 4 slides was made for each culture flask.
** 100 cells were examined microscopically per flask and were
** scored for structural chromosomal aberrations. Although gaps were
* recorded they were not used in the analysis for
** chromosomal aberration since their significance is
** questionable.
**
** Cytotoxicity assay (Mitotic Index determination)
** For this portion of the assay all cells in each flask were
** collected.
** The medium in each flask was replaced and 5 ml of trypsin
** was added and after incubation for approximately 5 minutes
** the flasks were examined to ensure that the cells had
** rounded up.
** The cells were then released into the trypsin solution
** mechanically and the resulting suspension was centrifuged in the presence
* of medium. The centrifuged cells were swelled in hypotonic solution and
* were fixed in methanol:acetic acid as above.
** A minimum of 2 slides per flask were prepared for the
** determination of mitotic index.
** The slides were stained with Giemsa and at least 1000 nuclei per flask
* were scored as either mitotic or interphasic.
**
** The MI = No mitotic cells x 100/Total No of cells scored.
**
** Criteria for a positive or negative response
** A test substance is considered to have elicited a positive
** response if at least one concentration shows a statistically significant
* increase in the proportion of cells with aberrations and a significant
* positive dose-response exists.
** Biological significance of the result is also taken into
** account in determining the response.
F008 IUC4
F020 4712
EOR
F002 37
F010 5.5
F004 2
F005 RE
F006 Mobil (1991)
** Metaphase analysis of Chinese hamster ovary (CHO) cells
** treated in-vitro with a DMSO extract of Lost Hills Light
** Study 63840
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1991)

** Metaphase analysis of Chinese hamster ovary (CHO) cells
** treated in-vitro with a DMSO extract of Lost Hills Light
** Study 63840
** Mobil Environmental and Health Science Laboratory
F008 IUC4
F020 4713
EOR
F002 37
F010 5.5
F004 2
F005 RL
F006 Despite the fact that whether the study was carried out to
** GLP is not clear, it is, nevertheless, reliable and valid.
F007 Despite the fact that whether the study was carried out to
** GLP is not clear, it is, nevertheless, reliable and valid.
F008 IUC4
F020 4714
EOR
F002 37
F010 5.5
F004 2
F005 RS
F006 Determination of Mitotic Index (MI)
**
** There was not a dose related decrease in MI at treatments
** that were not toxic. The MI at each of the dose
** concentrations of Lost Hills Light crude (LHL)was:
**
** Treatment Mitotic Index
** 20 µl/ml extract o
F007 Determination of Mitotic Index (MI)
**
** There was not a dose related decrease in MI at treatments
** that were not toxic. The MI at each of the dose
** concentrations of Lost Hills Light crude (LHL)was:
**
** Treatment Mitotic Index
** 20 µl/ml extract of LHL no cells found
** 15 µl/ml extract of LHL no cells found
** 10 µl/ml extract of LHL 0.6
** 5 µl/ml extract of LHL 10.7
** 2.5 µl/ml extract of LHL 8.4
** 1 µl/ml extract of LHL 12.2
**
** 20 µl/ml DMSO 7.6
**
** Negative control 10.4
**
** Metaphase analysis
**
** The concentrations analyzed for chromosomal aberrations were 5, 2.5 and
* 1.0 µg/ml.
** There was no increase in the proportion of cells with

** structural chromosome aberrations (gaps excluded). Nor was a dose
* response observed.
** The data are:
**

** Treatment		No of cells with one or more aberrations (100 cells examined)
** 5 µl/ml extract of LHL		2
** 2.5 µl/ml extract of LHL	2	
** 1 µl/ml extract of LHL		1
** DMSO (solvent control		2
** Negative control	2	
** CP (positive control)	28	

F008 IUC4
F020 4715
EOR
F002 37
F010 5.5
F004 3
F005 ME
F006
** Preparation of DMSO extract
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* minute. After 30 minutes, the mixture was
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F007
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* minute. After 30 minutes, the mixture was
** centrifuged and the DMSO layer (Extract) was removed for
** testing.
**
** Control substances:
** Solvent control: DMSO
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** Positive controls: Cyclophosphamide monohydrate at
** concentration of 10 µg/ml
**
** Duplicate cultures were used - one set for metaphase
** analysis and the other for a determination of cytotoxicity
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** activation.
**
** CHO cells were exposed for two hours to test material at
** concentrations of 1, 2.5, 5, 10, 15 and 20 µl/ml.
** After two hours, treatment was terminated and the cultured
** cells were washed and re-fed with complete culture medium.

** Approximately 16 hours later, colcicine was added and two
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* attached mitotic cells into culture medium and pelleting them by
* centrifugation. The cells were resuspended and then fixed in
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** For this portion of the assay all cells in each flask were
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** The cells were then released into the trypsin solution
** mechanically and the resulting suspension was centrifuged in the presence
* of medium. The centrifuged cells were swelled in hypotonic solution and
* were fixed in methanol:acetic acid as above.
** A minimum of 2 slides per flask were prepared for the
** determination of mitotic index.
** The slides were stained with Giemsa and at least 1000 nuclei per flask
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**
** The MI = No mitotic cells x 100/Total No of cells scored.
**
** Criteria for a positive or negative response
** A test substance is considered to have elicited a positive
** response if at least one concentration shows a statistically
** significant increase in the proportion of cells with
** aberrations and a significant positive dose-response exists.
** Biological significance of the result is also taken into
** account in determining the result.
F008 IUC4
F020 4716
EOR
F002 37
F010 5.5
F004 3
F005 RE
F006 Mobil (1991)
** Metaphase analysis of Chinese hamster ovary (CHO) cells
** treated in-vitro with a DMSO extract of Belridge Heavy
** Study 63852
** Mobil Environmental and Health Science Laboratory

F007 Mobil (1991)
** Metaphase analysis of Chinese hamster ovary (CHO) cells
** treated in-vitro with a DMSO extract of Belridge Heavy
** Study 63852
** Mobil Environmental and Health Science Laboratory
F008 IUC4
F020 4717
EOR
F002 37
F010 5.5
F004 3
F005 RL
F006 Despite the fact that whether the study was carried out to
** GLP is not clear, it is, nevertheless, reliable and valid.
F007 Despite the fact that whether the study was carried out to
** GLP is not clear, it is, nevertheless, reliable and valid.
F008 IUC4
F020 4718
EOR
F002 37
F010 5.5
F004 3
F005 RS
F006 Determination of Mitotic Index (MI)
**
** There was not a dose related decrease in MI at treatments
** that were not toxic. The MI at each of the dose
** concentrations of Belridge Heavy crude (BH)was:
**
** Treatment Mitotic Index
** 20 µl/ml extract of B
F007 Determination of Mitotic Index (MI)
**
** There was not a dose related decrease in MI at treatments
** that were not toxic. The MI at each of the dose
** concentrations of Belridge Heavy crude (BH)was:
**
** Treatment Mitotic Index
** 20 µl/ml extract of BH no cells found
** 15 µl/ml extract of BH no cells found
** 10 µl/ml extract of BH no cells found
** 5 µl/ml extract of BH 11.2
** 2.5 µl/ml extract of BH 10.2
** 1 µl/ml extract of BH 13.1
**
** 20 µl/ml DMSO 7.6
**
** Negative control 10.4
**
** Metaphase analysis
**
** The concentrations analyzed for chromosomal aberrations were
** 5, 2.5 and 1.0 µg/ml.

** There was no increase in the proportion of cells with
** structural chromosome aberrations (gaps excluded). Nor was a dose
* response observed.
** The data are:
**

** Treatment	No of cells with one or more aberrations (100 cells examined)
** 5 µl/ml extract of LHL	1
** 2.5 µl/ml extract of LHL	2
** 1 µl/ml extract of LHL	1
** DMSO (solvent control	2
** Negative control	2
** CP (positive control)	28

F008 IUC4
F020 4719
EOR
F002 37
F010 5.5
F004 4
F005 ME
F006 The authors summarized the protocol in the following table:
**
** Test strains
** Salmonella typhimurium strains TA-1535, TA-1537, TA-1538,
** TA-98 and TA-100
**
** Liver homogenate

** S-9 from male Charles River CD rat liver
** 500 mg Aroclor/kg for 5 days
**
** Pr
F007 The authors summarized the protocol in the following table:
**
** Test strains
** Salmonella typhimurium strains TA-1535, TA-1537, TA-1538,
** TA-98 and TA-100
**
** Liver homogenate
** S-9 from male Charles River CD rat liver
** 500 mg Aroclor/kg for 5 days
**
** Procedure
** Treatment without activation: 2 ml top agar, 0.1 ml
** dissolved test chemical, 0.1 ml bacterial culture (108
** cells)
**
** Treatment with activation: 2 ml top agar, 0.1 ml dissolved

** test chemical, 0.1 ml bacterial culture (108 cells) plus 0.5
** ml S-9 mix contains per milliliter 0.3 ml S-9 (1 g tissue +
** 3 ml 0.15M KCl), 8 mM Mg Cl2, 33 mM KCl, 5mM
** glucose-6-phosphate, 4 mM NADP, and 100 mM sodium phosphate
** (pH 7.4).
**
** Incubation period: 48 hr
**
** Design: Preliminary toxicity determination
** Duplicate plates/point
** Five test concentrations and solvent and positive
** controls
** Duplicate experiments
**
** Solvent
** Dimethyl sulfoxide
**
** Data analysis
** Mutagenic: statistically significant. Increase in total
** revertant colony number (P</= 0.01) and dose response
** (P</=0.01)
**
** Scoring

**			Induced frequency/ Spontaneous frequency
**		Revertant/µg	
**	(-)	<0.01	-*
**	(+)	0.1 to 0.01	<5
**	(++)	>0.1	>5

**
** * No greater than response
F008 IUC4
F020 4720
EOR
F002 37
F010 5.5
F004 4
F005 RE
F006 Calkins, W. H., Deye, J. F., Hartgrove, R. W., King, C. F.
** and Krahn, D. F. (1981)
** Synthetic crude oils from coal: Mutagenicity and
** tumor-initiating screening tests.
** In Mahlum, D. D. et al (Eds)
** Coal conversion and the environment pp 461-47
F007 Calkins, W. H., Deye, J. F., Hartgrove, R. W., King, C. F.
** and Krahn, D. F. (1981)
** Synthetic crude oils from coal: Mutagenicity and
** tumor-initiating screening tests.
** In Mahlum, D. D. et al (Eds)
** Coal conversion and the environment pp 461-470
** U. S. Dept. of Energy, Washington D. C.
F008 IUC4

F020 4721
EOR
F002 37
F010 5.5
F004 4
F005 RL
F006 The study is not fully reported (data for the assay without
** metabolic activation are not given)
F007 The study is not fully reported (data for the assay without
** metabolic activation are not given)
F008 IUC4
F020 4722
EOR
F002 37
F010 5.5
F004 4
F005 RS
F006 The results are given in the following table
**
**

** Sample	Mutagenic activity (with activation)			
**	TA-1537	TA-1538	TA-98	TA-100
** Benzo[a]pyrene	++	++	++	++
** South Louisiana crude	-	-	-	+

**
** It is not clear from the report whether any activity
F007 The results are given in the following table
**
**

** Sample	Mutagenic activity (with activation)			
**	TA-1537	TA-1538	TA-98	TA-100
** Benzo[a]pyrene	++	++	++	++
** South Louisiana crude	-	-	-	+

**
** It is not clear from the report whether any activity was
** detected in the assay without metabolic activation.
F008 IUC4
F020 4723
EOR
F002 37
F010 5.5
F004 5
F005 ME
F006 Salmonella typhimurium strains TA 98 and TA 100 were used in this study.
** The test sample was suspended in Tween 80 prior to testing.
** Each sample was weighed in a glass vial and then an equal
** weight of Tween 80 was added. The oil and deterge
F007 Salmonella typhimurium strains TA 98 and TA 100 were used in this study.
** The test sample was suspended in Tween 80 prior to testing.
** Each sample was weighed in a glass vial and then an equal
** weight of Tween 80 was added. The oil and detergent were

** stirred until thoroughly homogenized. Distilled water was
** then added dropwise with continuous stirring until a stable, homogenous
* suspension was obtained. The concentration of the mixture was then
* adjusted to 10% sample - 10% Tween 80 (w/v) by the addition of more
* distilled water.
** The assay was carried out in triplicate with and without
** metabolic activation by S9 liver homogenate that was
** prepared from Aroclor-induced Sprague-Dawley rats. The S9
** homogenate contained 13 mg of protein/ml and was stored at
** -70°C. S9 homogenate was used at a concentration of 10%
** (v/v) in the assay.
** BaP was used as the positive control.
** Negative control plates for determination of the spontaneous reversion
* rate were treated with 400µl or less of 10% Tween 80, a non-toxic and
* non-mutagenic dose.
**
** No other experimental details are given in the paper, but
** reference is made to the papers by Ames that describe the
** methodology.
F008 IUC4
F020 4724
EOR
F002 37
F010 5.5
F004 5
F005 RE
F006 Lockard, J. M., Prater, J. W., Viau, C. J., Enoch, H. G. and
** Sabharwal. P. S. (1982)
** Comparative study of the genotoxic properties of Eastern and
** Western U. S. shale oils, crude petroleum and coal-derived
** oil.
** Mutation Research Vol 102, pp
F007 Lockard, J. M., Prater, J. W., Viau, C. J., Enoch, H. G. and
** Sabharwal. P. S. (1982)
** Comparative study of the genotoxic properties of Eastern and
** Western U. S. shale oils, crude petroleum and coal-derived
** oil.
** Mutation Research Vol 102, pp 221-235
F008 IUC4
F009 28-08-2003
F020 4725
EOR
F002 37
F010 5.5
F004 5
F005 RL
F006 The numbers of revertants were not given. The result was
** simply stated by the authors.
F007 The numbers of revertants were not given. The result was
** simply stated by the authors.
F008 IUC4
F020 4726
EOR

F002 37
F010 5.5
F004 5
F005 RS
F006 Addition of up to 400µl Tween 80 per plate did not alter the
** spontaneous reversion rates of either TA 98 or TA 100. Tween
** 80 had no effect on the mutagenicity of BaP, which induced
** similar numbers of revertants in TA 98 with S9 activation
** w
F007 Addition of up to 400µl Tween 80 per plate did not alter the
** spontaneous reversion rates of either TA 98 or TA 100. Tween
** 80 had no effect on the mutagenicity of BaP, which induced
** similar numbers of revertants in TA 98 with S9 activation
** whether dissolved in DMSO or Tween 80.
** The crude oil sample was inactive in all bacterial
** mutagenicity tests.
F008 IUC4
F020 4727
EOR
F002 37
F010 5.5
F004 6
F005 ME
F006 Heparinized blood was obtained from a healthy 25-year old
** male. [The method for culture of the lymphocytes was not
** described but reference was given which describes the
** method].
** Benzo(a)pyrene (BaP) and N-methyl-N'-nitro-N-nitroso
** guanidine
F007 Heparinized blood was obtained from a healthy 25-year old
** male. [The method for culture of the lymphocytes was not
** described but reference was given which describes the
** method].
** Benzo(a)pyrene (BaP) and N-methyl-N'-nitro-N-nitroso
** guanidine (MNNG) were used as positive control substances.
**
** The test sample was suspended in Tween 80 prior to testing.
** Each sample was weighed in a glass vial and then an equal
** weight of Tween 80 was added. The oil and detergent were
** stirred until thoroughly homogenized. Distilled water was
** then added dropwise with continuous stirring until a stable, homogenous
* suspension was obtained. The concentration of the mixture was then
* adjusted to 1% sample - 1% Tween 80 (w/v) by the addition of more
* distilled water.
** Lymphocyte cultures were treated 18 hours after initiation
** by (a) continuous exposure to test chemical without
** exogenous activation or
** (b) by exposure for 2.5 hours with activation by S9.
** [procedure described elsewhere by Stetka and Wolff (1976)
** for metabolic activation].
** In tests with S9, cells were collected by centrifugation at
** 150 x g for 10 minutes, resuspended in 5 ml of McCoy's 5A
** medium containing only antibiotics and treated with

** microliter quantities of test material or solvent. An
** aliquot of 0.25 ml of S9 mix containing 10% S9 (v/v)
** was added to each culture. Cells were incubated for 2.5
** hours with occasional mixing. The cells were collected,
** washed once in medium with 5% fetal bovine serum and
** resuspended in 8 ml of culture medium containing BrdUrd for
** further incubation. In experiments testing MNNG, lymphocytes
** that had been cultured for 18 hours were suspended in
** medium without serum, S9 or BrdUrd, treated for 2 hours with MNNG in
* acetone or acetone alone (controls), washed as
** above, resuspended in complete medium with BrdUrd and
** incubated further. At 68 hours after the cultures were
** initiated, Colcemid (0.01µg/ml) was added; 4 hours later,
** the cells were harvested by centrifugation, subjected to
** hypotonic shock for 7 minutes in 0.075M KCl, fixed in 3
** changes of cold methanol-acetic acid (3:1) and spread on
** chilled, wet microslides that had been pre-cleaned in 95%
** ethanol. The preparations were stained by the Hoechst
** 33258-black light-Giemsa method and numbers of SCE were
** counted in 25 metaphase cells containing 40-48 chromosomes
** for each treatment.
F008 IUC4
F020 4728
EOR
F002 37
F010 5.5
F004 6
F005 RE
F006 Lockard, J. M., Prater, J. W., Viau, C. J., Enoch, H. G. and
** Sabharwal. P. S. (1982)
** Comparative study of the genotoxic properties of Eastern and
** Western U. S. shale oils, crude petroleum and coal-derived
** oil.
** Mutation Research Vol 102, pp
F007 Lockard, J. M., Prater, J. W., Viau, C. J., Enoch, H. G. and
** Sabharwal. P. S. (1982)
** Comparative study of the genotoxic properties of Eastern and
** Western U. S. shale oils, crude petroleum and coal-derived
** oil.
** Mutation Research Vol 102, pp 221-235
F008 IUC4
F009 28-08-2003
F020 4729
EOR
F002 37
F010 5.5
F004 6
F005 RL
F006 Complete description of study not given in publication. Some
** methods details published elsewhere
F007 Complete description of study not given in publication. Some
** methods details published elsewhere
F008 IUC4

F020 4730
EOR
F002 37
F010 5.5
F004 6
F005 RS
F006 Tween 80 was not cytotoxic and did not induce increases in
** the numbers of SCE in cultures of lymphocytes treated with
** 100 ppm Tween 80 for 54 hours without activation or for 2.5
** hours with S9 activation.
** MNNG, a strong alkylating agent, ind
F007 Tween 80 was not cytotoxic and did not induce increases in
** the numbers of SCE in cultures of lymphocytes treated with
** 100 ppm Tween 80 for 54 hours without activation or for 2.5
** hours with S9 activation.
** MNNG, a strong alkylating agent, induced a greater than
** 5-fold increase in SCE at a concentration of only 0.15 ppm.
** The response to BaP in the lymphocyte cultures indicated
** that the S9 metabolic system was functional. Although
** increases in SCE of only 50% over the number in control
** cultures were induced, these increases are statistically
** significant (P<0.001). In the presence of S9 mix, only 1/16
** the amount of BaP was required to induce the same number of
** SCE observed without S9, indicating that metabolism had
** occurred. However, 0.5µg/ml of BaP with S9 mix was toxic to
** these cultures, as indicated by poor cell morphology and a
** reduced number of cells in mitosis.
** Wilmington crude did not induce an increase in SCE at any
** dose concentration with or without S9 activation.
** The data are summarized in the following table.
**

** Treatment	Dose (ppm)	No. of cells	SCE/cell ±S.E.
** 24 hours exposure, without S9			
** Tween 80	0	25	8.4±0.5
**	100	25	9.7±0.6
** B(a)P	0	25	7.7±0.6
**	4	25	10.8±1.2
**	8	25	15.1±0.9**
** Wilmington crude oil	40	25	10.7±1.2
**	50	25	9.9±0.8
** 2.5 hours exposure, with S9			
** Tween 80	40	25	9.6±0.0
**	100	25	8.0±0.5
** B(a)P	0	25	10.9±1.9
**	0.2	25	13.4±0.8**
**	0.5	10	15.9±1.2**

**		1.0	0	Toxic
**				
**	MNNG	0	25	8.7±0.4
**		0.015	23	11.7±0.5**
**		0.05	23	28.3±1.3**
**		0.15	21	48.5±2.2**
**				
**	Wilmington			
**	crude oil	20	25	8.8±0.7
**		30	25	6.6±0.5
**				
**	**	P<0.001		

F008 IUC4
F020 4731
EOR
F002 37
F010 5.6
F004 1
F005 ME
F006 The animals used in this study were taken from a 13 week
** repeated dermal administration study (Study 63834).
** Bone marrow was removed from the femurs of five male and
** five female rats from each dose group (0, 30, 125 and 500
** mg/kg/day).
** A ne
F007 The animals used in this study were taken from a 13 week
** repeated dermal administration study (Study 63834).
** Bone marrow was removed from the femurs of five male and
** five female rats from each dose group (0, 30, 125 and 500
** mg/kg/day).
** A nearly pure erythrocyte fraction was obtained and two
** slides prepared for each animal.
** 1000 PCEs (polychromatic erythrocyte) and 1000 NCEs
** (normochromatic erythrocyte) were scored to determine the
** percentage of micronucleated erythrocytes. 1000 erythrocytes
** were scored to determine the ratio of PCEs to NCEs.
F008 IUC4
F020 4732
EOR
F002 37
F010 5.6
F004 1
F005 RE
F006 Lockard, J. M., Prater, J. W., Viau, C. J., Enoch, H. G. and
** Sabharwal. P. S. (1982)
** Comparative study of the genotoxic properties of Eastern and
** Western U. S. shale oils, crude petroleum and coal-derived
** oil.
** Mutation Research Vol 102, pp
F007 Lockard, J. M., Prater, J. W., Viau, C. J., Enoch, H. G. and
** Sabharwal. P. S. (1982)
** Comparative study of the genotoxic properties of Eastern and
** Western U. S. shale oils, crude petroleum and coal-derived

** oil.
** Mutation Research Vol 102, pp 221-235
F008 IUC4
F009 28-08-2003
F020 4733
EOR
F002 37
F010 5.6
F004 1
F005 RE
F006 Mobil (1990)
** Micronucleus assay of bone marrow red blood cells from rats
** treated via dermal administration of Lost Hills Light
** Study 63835
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1990)
** Micronucleus assay of bone marrow red blood cells from rats
** treated via dermal administration of Lost Hills Light
** Study 63835
** Mobil Environmental and Health Science Laboratory
F008 IUC4
F020 4734
EOR
F002 37
F010 5.6
F004 1
F005 RE
F006 Mobil (1991)
** Micronucleus assay of bone marrow cells from rats
** treated via dermal administration of Belridge Heavy
** Study 63847
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1991)
** Micronucleus assay of bone marrow cells from rats
** treated via dermal administration of Belridge Heavy
** Study 63847
** Mobil Environmental and Health Science Laboratory
F008 IUC4
F020 4735
EOR
F002 37
F010 5.6
F004 1
F005 RE
F006 Mobil (1992)
** Thirteen-week dermal administration of Lost Hills Light to
** rats
** Study No. 63834
** Environmental Health and Sciences Laboratory
F007 Mobil (1992)
** Thirteen-week dermal administration of Lost Hills Light to
** rats
** Study No. 63834

** Environmental Health and Sciences Laboratory
F008 IUC4
F020 4736
EOR
F002 37
F010 5.6
F004 1
F005 RM
F006 Two further studies have been reported.
**
** 1. Belridge heavy crude.
** The results of this study were the same as those for the study on Lost
* Hills Light described above. (Mobil study 63847)
**
** 2. Wilmington crude
** Results in ICR Swiss mice s
F007 Two further studies have been reported.
**
** 1. Belridge heavy crude.
** The results of this study were the same as those for the study on Lost
* Hills Light described above. (Mobil study 63847)
**
** 2. Wilmington crude
** Results in ICR Swiss mice showed that Wilmington crude oil had
* relatively little capacity to cause chromosome breakage and/or
* non-disjunction. (Lockard et al, 1982)
F008 IUC4
F020 4737
EOR
F002 37
F010 5.6
F004 1
F005 RS
F006 The results are given in the following table. Standard
** deviations have not been included in the table. However,
** statistical analyses were conducted and it was found that
** there were no significant differences between treated and
** control anim
F007 The results are given in the following table. Standard
** deviations have not been included in the table. However,
** statistical analyses were conducted and it was found that
** there were no significant differences between treated and
** control animals.
**
** It was concluded that the test material was not cytotoxic to
** red blood cell formation and furthermore did not increase
** the formation of micronucleated PCEs or NCEs in the bone
** marrow.
**

** Dose mg/kg	Sex	No of animals	PCE/NCE	MNPCEs (%)	MNNCEs (%)
** 0	F	5	1.34	0.1	0

**	0	M	5	0.79	0.02	0
**	0	M+F	10	1.06	0.06	0
**						
**	30	F	5	1.26	0.04	0
**	30	M	5	0.68	0	0.02
**	30	M+F	10	0.97	0.02	0.01
**						
**	125	F	5	1.18	0	0
**	125	M	5	1.17	0.06	0
**	125	M+F	10	1.17	0.03	0
**						
**	500	F	5	1.25	0.02	0
**	500	M	5	0.83	0.04	0
**	500	M+F	10	1.04	0.03	0
**						
**		PCE	Polychromatic erythrocytes			
**		NCE	Normochromatic erythrocytes			
**		%MNPCEs	% Micronucleated PCEs			
**		%MNNCEs	% Micronucleated normochromatic erythrocytes			

F008 IUC4
F020 4738
EOR
F002 37
F010 5.6
F004 2
F005 ME
F006 The assay was carried out in groups of 3 male 10-14 week old Sch:ICR mice.
** The mice were lightly anesthetized with ether and a 50 mg
** tablet of BrdUrd was inserted beneath the skin on the dorsal side of the
* neck of each animal. After 1 hour,
F007 The assay was carried out in groups of 3 male 10-14 week old Sch:ICR mice.
** The mice were lightly anesthetized with ether and a 50 mg
** tablet of BrdUrd was inserted beneath the skin on the dorsal side of the
* neck of each animal. After 1 hour, treatment was given by a single i.p.
* injection at doses of 1.8, 3.6 and 7.2 g/kg (equivalent to 0.8, 0.4 and
* 0.2 times the approximate LD50).
** Control groups of mice were injected with B(a)P at doses of
** 0.16, 0.08 and 0.04 g/kg, Cyclophosphamide at doses of 0.01 and 0.005
* g/kg in distilled water. Trioctanoin and distilled water were also given
* to negative solvent controls.
** 22 hours after treatment, animals were injected i.p. with
** Colchicin (10 mg/kg). 2 hours later the mice were sacrificed by cervical
* dislocation and their femurs were removed. The bone marrow cells were
* flushed out with saline. Pooled cells from the 2 femurs of each animal
* were treated with hypotonic KCl for 30 minutes, were fixed and were
* spread on slides.
** The slides were stained and the numbers of SCE counted in 25 metaphase
* cells from each animal, each metaphase having
** 36-42 chromosomes.
F008 IUC4
F020 4739
EOR
F002 37

F010 5.6
F004 2
F005 RE
F006 Lockard, J. M., Prater, J. W., Viau, C. J., Enoch, H. G. and
** Sabharwal. P. S. (1982)
** Comparative study of the genotoxic properties of Eastern and
** Western U. S. shale oils, crude petroleum and coal-derived
** oil.
** Mutation Research Vol 102, pp
F007 Lockard, J. M., Prater, J. W., Viau, C. J., Enoch, H. G. and
** Sabharwal. P. S. (1982)
** Comparative study of the genotoxic properties of Eastern and
** Western U. S. shale oils, crude petroleum and coal-derived
** oil.
** Mutation Research Vol 102, pp 221-235
F008 IUC4
F009 28-08-2003
F020 4740
EOR
F002 37
F010 5.6
F004 2
F005 RL
F006 Probably not to GLP and method description is not
** comprehensive.
F007 Probably not to GLP and method description is not
** comprehensive.
F008 IUC4
F020 4741
EOR
F002 37
F010 5.6
F004 2
F005 RS
F006 Control animals injected with the solvent trioctanoin had a
** mean of 5.3 SCE/cell. A slight but significant increase in
** SCE was found in animals treated with the highest dose only
** of Wilmington crude oil, but the response was not dose
** relate
F007 Control animals injected with the solvent trioctanoin had a
** mean of 5.3 SCE/cell. A slight but significant increase in
** SCE was found in animals treated with the highest dose only
** of Wilmington crude oil, but the response was not dose
** related.
** B(a)P and cyclophosphamide induced dose-related increases in SCE.
** The actual results are shown in the following table.
**
** Treatment Dose No. survivors/ No. cells SCE/
** No. treated examined cell±S.E.
**
** Trioctanoin 0.5 ml/
** mouse 7/8 175 5.3±0.4
**

** B(a)P 0.16 g/kg 3/3 75 9.7±0.6***
** 0.08 3/3 75 8.5±0.5***
** 0.04 3/3 75 7.8±0.5***
**
** Cyclophosphamide
** 0.01 g/kg 3/3 75 19.5±0.6***
** 0.005 3/3 75 14.6±0.6***
**
** Wilmington crude oil
** 7.2 g/kg 3/3 75 6.6±0.4**
** 3.6 3/3 50 5.0±0.3
** 1.8 3/3 75 6.4±0.4
**
** ** P<0.05
** *** P<0.001
F008 IUC4
F020 4742
EOR
F002 37
F010 5.7
F004 1
F005 ME
F006 50 mg undiluted test material was applied to the shorn
** interscapular region of groups of 50 male mice.
** Application was twice weekly for 18 months or until
** grossly-observable cancer was found.
** Each animal was observed weekly for the appearan
F007 50 mg undiluted test material was applied to the shorn
** interscapular region of groups of 50 male mice.
** Application was twice weekly for 18 months or until
** grossly-observable cancer was found.
** Each animal was observed weekly for the appearance of
** tumors. The percentage of animals developing tumors and the
** time to appearance of first tumor were recorded.
** No distinction was made between histologically benign or
** malignant lesions.
F008 IUC4
F020 4743
EOR
F002 37
F010 5.7
F004 1
F005 RE
F006 Lewis, S. C., King, R. W., Cragg, S. T. and Hilman, D. W.
** (1984)
** Skin carcinogenesis potential of petroleum hydrocarbons:
** II Crude oil, distillate fractions and chemical class
** subfractions
** In Applied Toxicology of Petroleum Hydrocarbons
** Vol
F007 Lewis, S. C., King, R. W., Cragg, S. T. and Hilman, D. W.
** (1984)
** Skin carcinogenesis potential of petroleum hydrocarbons:

** II Crude oil, distillate fractions and chemical class
** subfractions
** In Applied Toxicology of Petroleum Hydrocarbons
** Vol VI of Advances in Modern Environmental Toxicology, M.
** Mehlman, Ed. Princeton Scientific Publishers, Princeton H.J.
** pp 139-150
F008 IUC4
F020 4744
EOR
F002 37
F010 5.7
F004 1
F005 RS
F006 The % tumors and average latencies for the two crudes C & D
** are shown below.
**

**		% Tumors*	Latency**
**	Crude C	33	76
**	Crude D	56	64

**
** * Based on Final Effective Number
** ** Average latency in weeks
F007 The % tumors and average latencies for the two crudes C & D
** are shown below.
**

**		% Tumors*	Latency**
**	Crude C	33	76
**	Crude D	56	64

**
** * Based on Final Effective Number
** ** Average latency in weeks
F008 IUC4
F020 4745
EOR
F002 37
F010 5.7
F004 1
F005 TS
F006 Two crudes were included in this study. They were
** characterized as follows:
**

**		Crude C	Crude D
**	Sulfur (wt%)	0.21	2.54
**	Distillation yield (vol%)		
**	Int-120°F	0.0	3.3
**	120-350	2.5	18.3
**	350-550	41.9	19.5
**	550-700	21.0	12.2
**	700-		

F007 Two crudes were included in this study. They were
** characterized as follows:
**

**		Crude C	Crude D

** Sulfur (wt%)		0.21		2.54
** Distillation yield (vol%)				
** Int-120°F		0.0		3.3
** 120-350		2.5		18.3
** 350-550		41.9		19.5
** 550-700		21.0		12.2
** 700-1070		31.2		26.8
** 1070 btms		3.4		19.9
**				
** Composition of 550-700°F				
** Fraction (wt%)		5.1		33.3
** Monocycloparaffins	8.1		11.5	
** Polycycloparaffins	46.6		19.5	
** Total saturated hydrocarbons	59.8		64.3	
** Mononuclear aromatics		21.5		11.0
** Di- and Trinuclear aromatics	14.4		21.8	
** Polynuclear aromatics		1.0		1.8
** Total aromatic hydrocarbons	36.9		34.6	
** Resins		3.3		1.1
** Asphaltenes		0.0		0.0

F008 IUC4
F020 4746
EOR
F002 37
F010 5.7
F004 2
F005 ME
F006 The study reported by Clarke et al was a comparison of the
** carcinogenicity of shale and petroleum crude oil and of
** several distillate streams derived from the two crude oils.
** Only the details relating to petroleum crude and the
** respective p
F007 The study reported by Clarke et al was a comparison of the
** carcinogenicity of shale and petroleum crude oil and of
** several distillate streams derived from the two crude oils.
** Only the details relating to petroleum crude and the
** respective positive and negative controls are included in
** this summary.
**
** The crude oil, positive and negative control materials were
** applied three times weekly at a dose of 25 mg/application to 25 male and
* 25 female mice.
** The materials were applied to the shorn dorsal thoracic
** region which was shaved as necessary during the study.
** Dosing was continued for up to 105 weeks.
** Animals were observed twice daily for overt signs of
** toxicity, moribundity and mortality. Animals were weighed
** weekly for the first 13 weeks and every two weeks
** thereafter. Animals were palpated weekly for external and
** internal masses.
**
** Every animal was subjected to a complete necropsy. All
** organs and the remainder of the carcass were fixed and

** stored. Body weights and weights of liver, kidneys, brain
** and gonads were recorded. A microscopic examination was made of
* histological slides prepared from skin from a control site, skin from
* treated site, kidney, liver, lungs, gonads, urinary bladder, spleen,
* sternal bone marrow and any other organ considered abnormal at necropsy.
F008 IUC4
F020 4747
EOR
F002 37
F010 5.7
F004 2
F005 RE
F006 Clark, C. R., Walter, M. K., Ferguson, P. W. and Katchen, M.
** (1988)
** Comparative dermal carcinogenesis of shale and
** petroleum-derived distillates
** Toxicol. and Ind. Health Vol 4, No. 1, pp 11-22
F007 Clark, C. R., Walter, M. K., Ferguson, P. W. and Katchen, M.
** (1988)
** Comparative dermal carcinogenesis of shale and
** petroleum-derived distillates
** Toxicol. and Ind. Health Vol 4, No. 1, pp 11-22
F008 IUC4
F020 4748
EOR
F002 37
F010 5.7
F004 2
F005 RL
F006 Few experimental details are given in this publication.
** Despite this, the information may be used in the overall
** assessment of the carcinogenic potential of crude oil
F007 Few experimental details are given in this publication.
** Despite this, the information may be used in the overall
** assessment of the carcinogenic potential of crude oil
F008 IUC4
F020 4749
EOR
F002 37
F010 5.7
F004 2
F005 RS
F006 Survival was affected by treatment with petroleum crude oil.
** After one year, survival was approximately 70% and by week
** 70 survival was approximately 50%. There were few survivors
** at 105 weeks. This is in contrast to the negative control
** gr
F007 Survival was affected by treatment with petroleum crude oil.
** After one year, survival was approximately 70% and by week
** 70 survival was approximately 50%. There were few survivors
** at 105 weeks. This is in contrast to the negative control
** group with 50% survival at the end of the study.
** Dermal irritation at the test site first appeared at 271

** days and males developed irritation earlier than the females [no details
* given].
** Necrosis occurred and was characterized as loss of integrity of the skin
* with visible cracking, separation and sloughing of skin often revealing
* underlying mesenchymal tissue.
**
** There was no indication of toxic or oncogenic effects on
** internal organs.
**
** The authors reported the occurrence of reactive hyperplasia
** in the spleen and bone marrrow and this was characterized by increased
* populations of lymphocytes, plasma cells and
** neutrophils or granulocyte precursors and was attributed to
** the inflammation, necrosis and other tissue alterations
** occurring at the test site. [There is no indication in the
** publication whether this finding occurred in the group
** treated with petroleum crude oil.]
**
** The tumor incidences that were recorded are shown in the
** following table:

**	Crude oil	B(a)P	Control		
** Incidence (%)	37/44 (84)	49/49	0/46		
** Latency (weeks)	62±13		28±4		
** Squamous cell carcinoma	29 (66)		49 (100)	0	
** Fibrosarcoma	7 (16)		0		0

F008 IUC4
F020 4750
EOR
F002 37
F010 5.7
F004 2
F005 TS
F006 The San Joaquin Valley crude oil is a heavy crude with the
** following characteristics:

** Gravity (°API)		10.7
** Boiling range (°F)	410-1071	
** Nitrogen (ppm wt)	8150	
** Sulfur (ppm wt)		10,510

**
** Due to its high viscosity, the crude oil was administered
F007 The San Joaquin Valley crude oil is a heavy crude with the
** following characteristics:

** Gravity (°API)		10.7
** Boiling range (°F)	410-1071	
** Nitrogen (ppm wt)	8150	
** Sulfur (ppm wt)		10,510

**
** Due to its high viscosity, the crude oil was administered as
** a 2:1 dilution in mineral oil.
** Mineral oil USP was used as a negative control
** B(a)P (0.15% w/v in mineral oil) was used as a positive
** control
F008 IUC4

F020 4751
EOR
F002 37
F010 5.8.2
F004 1
F005 ME
F006 Groups of twelve presumed-pregnant Sprague-Dawley rats were
** distributed into six groups as shown below. The test
** material was applied to the shorn dorsal skin once daily at
** the doses shown from day 0 to day 19 of gestation. The
** animals were
F007 Groups of twelve presumed-pregnant Sprague-Dawley rats were
** distributed into six groups as shown below. The test
** material was applied to the shorn dorsal skin once daily at
** the doses shown from day 0 to day 19 of gestation. The
** animals were shaved weekly and were fitted with collars to
** prevent ingestion of the test material.
**
** Group Dose level
** Prenatal groups
** 1 Sham control
** 2 30 mg/kg/day
** 3 125 mg/kg/day
** 4 500 mg/kg/day
** Postnatal groups
** 5 Sham control
** 6 500 mg/kg/day
**
** Animals were observed at least once daily throughout
** gestation until sacrifice for signs of pathosis, abortion,
** premature delivery, dystocia and/or death.
** dams and their litters were observed postpartum days 0
** through 4. On day 0 postpartum, pups were examined for
** external malformations and variations and were observed
** daily for the presence of milk in their stomachs.
**
** Body weights and food consumption of all prenatal group
** females were recorded at regular intervals throughout
** gestation and for the post natal groups throughout gestation and body
* weights only on postpartum days 0 and 4.
**
** On day 20 of gestation all prenatal group animals were
** sacrificed. The abdominal cavity was exposed and the
** reproductive organs examined. Following removal of the
** uterus and ovaries the remains were subjected to macroscopic examination
* and the liver and thymus were weighed.
**
** The uterus and ovaries of each animal were examined grossly.
** The no. of corpora lutea per ovary was recorded. Ovaries of
** non pregnant females were examined and then discarded.
** The uterine contents of each pregnant animal were exposed
** and the number and location of all implantations were
** recorded. The uterus of each female that appeared non-gravid

** was pressed between two glass slides and examined grossly
** for evidence of implantation.
** Blood samples were collected at the time of sacrifice and
** these were used for a range of serum chemistry
** determinations.
**
** Each live fetus was weighed and its sex determined and
** examined for external anomalies. After gross examination the fetuses were
* equally distributed into two groups. One group was processed for an
* evaluation of visceral anomalies and the other group for skeletal
* anomalies.
**
** Females in the postnatal groups with surviving offspring
** were sacrificed on post partum day 4 and the abdominal
** cavity was exposed for a gross examination of the
** reproductive organs. The uterus was also examined and then
** discarded. All organs were examied macroscopically and the
** thymus and liver were weighed.
F008 IUC4
F020 4754
EOR
F002 37
F010 5.8.2
F004 1
F005 RE
F006 Feuston, M. H., Hamilton, C. E., Schreiner, C. A. and
** Mackerer, C. R. (1997)
** Developmental toxicity of dermally applied crude oils in
** rats
** J. Tox and Env. Health, Vol 52, pp 79-93
F007 Feuston, M. H., Hamilton, C. E., Schreiner, C. A. and
** Mackerer, C. R. (1997)
** Developmental toxicity of dermally applied crude oils in
** rats
** J. Tox and Env. Health, Vol 52, pp 79-93
F008 IUC4
F020 4755
EOR
F002 37
F010 5.8.2
F004 1
F005 RE
F006 Mobil (1991)
** Developmental toxicity study in rats exposed dermally to
** Belridge heavy
** Study No. 63848
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1991)
** Developmental toxicity study in rats exposed dermally to
** Belridge heavy
** Study No. 63848
** Mobil Environmental and Health Science Laboratory
F008 IUC4

F020 4756
EOR
F002 37
F010 5.8.2
F004 1
F005 RM
F006 This study was also reported in the open literature by
** Feuston et al.
F007 This study was also reported in the open literature by
** Feuston et al.
F008 IUC4
F020 4757
EOR
F002 37
F010 5.8.2
F004 1
F005 RS
F006 Clinical observations
** Treatment related clinical observations in the prenatal and
** postnatal groups consisted of skin irritation in the 500
** mg/kg/day group. This included erythema, edema, scabs and open sores at
* the treatment site. There was
F007 Clinical observations
** Treatment related clinical observations in the prenatal and
** postnatal groups consisted of skin irritation in the 500
** mg/kg/day group. This included erythema, edema, scabs and open sores at
* the treatment site. There was also a red vaginal discharge in animals in
* this group. One high dose prenatal female had excessive vaginal discharge
* and, after being found moribund, was sacrificed on gestation day 14.
* Uterine examination revealed that all but two fetuses had been resorbed.
**
** Body weights and food intakes of the 500 mg/kg/day prenatal
** groups were reduced compared to controls (reduced by approx
** 31%). Other dose groups were unaffected.
**
** Necropsy findings
** With the exception of an approximately 11% increase in
** relative liver weight in the 500 mg/kg/day group, no other
** organ or relative organ weight changes were recorded. [The
** authors comment that the observed increase in relative liver weight was
* possibly attributable to the decreased body weight of this group.
** A prominent vascular pattern of the liver was observed in
** one female of the 125 mg/kg/day group and two animals in the highest dose
* group.
**
** Serum chemistry
** The only observation was a 38% reduction of total bilirubin
** in the prenatal 500 mg/kg/day group.
**
** Reproductive and fetal evaluations
** Effects only occurred in the highest dose group (500
** mg/kg/day). These included
** an increase in the mean number/percent resorptions (5.3 compared to 1.3

* and 35% compared to 7.8% respectively) and a corresponding decrease in
* litter size (10.8 compared to 14.5).
**
** A decrease in mean fetal body weights for all viable fetuses (3.4
* compared to 3.7).
**
** Male fetuses in this dose group seemed to be more affected
** than females. Fetal skeletal anomalies recorded were limited to
* incomplete ossification of the nasal bones and caudal centra.
** There were no treatment related visceral anomalies.
**
**
** Post partum observations
** During the post partum period there were no clinical
** observations of note except some persistent skin irritation.
** There were no findings at necropsy.
**
**
** Litter data
** Two females in the 500 mg/kg/day group had no viable
** offspring; their litters were totally resorbed.
** The incidence of pup mortality was two times greater in the
** group treated with crude oil during the lactation period.
** Mean duration of gestation was unaffected, but it was noted
** that the majority of treated females delivered either in the afternoon of
* day 22 or morning of day 23 whereas the
** controls delivered in the morning of day 22. [The biological significance
* of this observation is uncertain.]
F008 IUC4
F020 4758
EOR
F002 37
F010 5.8.2
F004 2
F005 CL
F006 Lost Hills Light crude was shown to be maternally toxic at
** 500 and 2000 mg/kg/day and developmentally toxic at 2000
** mg/kg/day.
F007 Lost Hills Light crude was shown to be maternally toxic at
** 500 and 2000 mg/kg/day and developmentally toxic at 2000
** mg/kg/day.
F008 IUC4
F020 4759
EOR
F002 37
F010 5.8.2
F004 2
F005 ME
F006 Groups of twelve presumed-pregnant Sprague-Dawley rats were
** distributed into six groups as shown below. The test
** material was applied to the shorn dorsal skin once daily at
** the doses shown from day 0 to day 19 of gestation. The

** animals were
F007 Groups of twelve presumed-pregnant Sprague-Dawley rats were
** distributed into six groups as shown below. The test
** material was applied to the shorn dorsal skin once daily at
** the doses shown from day 0 to day 19 of gestation. The
** animals were shaved weekly and were fitted with collars to
** prevent ingestion of the test material.
**
** Group Dose level
** Prenatal groups
** 1 Sham control
** 2 125 mg/kg/day
** 3 500 mg/kg/day
** 4 2000 mg/kg/day
** Postnatal groups
** 5 Sham control
** 6 20000 mg/kg/day
**
** Animals were observed at least once daily throughout
** gestation until sacrifice for signs of pathosis, abortion,
** premature delivery, dystocia and/or death.
** Dams and their litters were observed postpartum days 0
** through 4. On day 0 postpartum, pups were examined for
** external malformations and variations and were observed
** daily for the presence of milk in their stomachs.
**
** Body weights and food consumption of all prenatal group
** females were recorded at regular intervals throughout
** gestation. For the post natal groups body weights and food
** intakes were recorded throughout gestation
** but only body weights were recorded on postpartum days 0 and
** 4.
**
** On day 20 of gestation all prenatal group animals were
** sacrificed. The abdominal cavity was exposed and the
** reproductive organs examined. Following removal of the
** uterus and ovaries the remains were subjected to macroscopic
** examination and the liver and thymus were weighed.
**
** The uterus and ovaries of each animal were examined grossly.
** The number of corpora lutea per ovary was recorded. Ovaries
** of non-pregnant females were examined and then discarded.
** The uterine contents of each pregnant animal were exposed
** and the number and location of all implantations were
** recorded. The uterus of each female that appeared non-gravid
** was pressed between two glass slides and examined grossly
** for evidence of implantation.
** Blood samples were collected at the time of sacrifice and
** these were used for a range of serum chemistry
** determinations.
**
** Each live fetus was weighed and its sex determined and
** examined for external anomalies. After gross examination the

** fetuses were equally distributed into two groups. One group
** was processed for an evaluation of visceral anomalies and
** the other group for skeletal anomalies.
**
** Females in the postnatal groups with surviving offspring
** were sacrificed on post partum day 4 and the abdominal
** cavity was exposed for a gross examination of the
** reproductive organs. After examination, the uterus was
** discarded. All organs were examined macroscopically and the
** thymus and liver were weighed.
F008 IUC4
F020 4760
EOR
F002 37
F010 5.8.2
F004 2
F005 RE
F006 Feuston, M. H., Hamilton, C. E., Schreiner, C. A. and
** Mackerer, C. R. (1997)
** Developmental toxicity of dermally applied crude oils in
** rats
** J. Tox and Env. Health, Vol 52, pp 79-93
F007 Feuston, M. H., Hamilton, C. E., Schreiner, C. A. and
** Mackerer, C. R. (1997)
** Developmental toxicity of dermally applied crude oils in
** rats
** J. Tox and Env. Health, Vol 52, pp 79-93
F008 IUC4
F020 4761
EOR
F002 37
F010 5.8.2
F004 2
F005 RE
F006 Mobil (1991)
** Developmental toxicity study in rats exposed dermally to
** Lost Hills Light
** Study No. 63836
** Mobil Environmental and Health Science Laboratory
F007 Mobil (1991)
** Developmental toxicity study in rats exposed dermally to
** Lost Hills Light
** Study No. 63836
** Mobil Environmental and Health Science Laboratory
F008 IUC4
F020 4762
EOR
F002 37
F010 5.8.2
F004 2
F005 RM
F006 This study was also reported in the open literature by
** Feuston et al.

F007 This study was also reported in the open literature by
** Feuston et al.
F008 IUC4
F020 4763
EOR
F002 37
F010 5.8.2
F004 2
F005 RS
F006 There were only few clinical observations that were
** considered to be treatment-related and these were confined
** to the high dose group (2000 mg/kg/day). The observations
** consisted of a red vaginal discharge, and one female in this
** group was
F007 There were only few clinical observations that were
** considered to be treatment-related and these were confined
** to the high dose group (2000 mg/kg/day). The observations
** consisted of a red vaginal discharge, and one female in this
** group was pale in color.
** Slight skin irritation occurred in a few animals but it is
** not clear whether this was compound-related or self
** inflicted because the animals attempted to groom themselves,
** despite the fact that they had been fitted with collars. It
** is possible also that because of this some ingestion of test
** material may have occurred.
** Animals in the 500 and 2000 mg/kg/day groups gained less
** weight than the controls over the gestation period (80% and
** 60% of controls respectively). Food consumption for these
** two groups was also less than controls for the first 10 days
** of gestation but was similar to controls thereafter.
**
** At necropsy, it was noted that there were more animals in
** the 2000 mg/kg/day group with a small thymus. Absolute and
** relative thymus weights were reduced in this group by
** approximately 50%. A reduction in absolute thymus weight was
** also recorded for the 500 mg/kg/day group, but the
** difference was not statistically significant.
** Liver weights (absolute and relative) were increased in the
** 500 and 2000 mg/kg group, but only the relative organ
** weights were statistically significant (11 and 18% increases
** for the 500 and 2000mg groups respectively).
**
** Serum chemistry differences were only observed in the 500
** and 2000 mg/kg/day groups These changes were as follows:
**

** Parameter	Dose group (mg/kg/day)	
**	500	2000
** AST		+27%
** ALT		+37%
** Alkaline phosphatase	+64%	
** Cholesterol		+22%
** Triglycerides	-42%	-62%
** Total bilirubin	-35%	-49%

** A/G ratio +15%
** Phos[horus +20%
** SDH +67%
**
** All other serum chemistry measurements were unaffected by
** treatment.
**
** Reproductive evaluations
** The only effect was a significant increase in mean number/%
** resorptions (6-fold) and a corresponding decrease in litter
** size (38%) in the 200 mg/kg/day group.
**
** Fetal evaluations
** Mean fetal body weights were reduced by approximately 13% in
** the 200 mg/kg/day group.
** There was an increase in incomplete ossification of various
** skeletal structures which included nasal bones and vertebrae
** but no increases in visceral abnormalities.
** The data are shown below:
** Date are shown as fetal incidence/litter incidence
**

** Anomoly	Dose group (mg/kg/day)			
**	0	125	500	2000
** Ossification variants				
** nasal bones	1.0/8.3	15b/55	33b/82b	96b/100b
** Thoracic centra	4/33	5.5/36	8.5/55	15a/78
** Caudal centra	1/8.3	9.9a/36	3.7/36	15b/67b
** Sternebrae <2	2/8.3	6.6/36	7.3/36	23b/67a
** Visceral malformations				
** Right-sided esophagus	0/0	-	0/0	4.1/22

** a P<0.05
** b P<0.01
**
** In the prenatal group, the litter effects were;
**
** Three females had no viable offspring, their litters were
** totally resorbed
**
** Two females had their entire litters die by postpartum day
** 3
**
** There was a significant increase in the number of
** stillborn pups (7 compared to 2 for controls)
**
** Liveborn pups weighed less than the controls (-8% and -
** 17% on days 0 and 4 respectively).
F008 IUC4
F020 4764
EOR
F002 37
F010 5.8.2

F004 3
F005 ME
F006 Presumed pregnant female Sprague-Dawley rats were used for
** this series of studies.
**
** Prudhoe Bay crude oil was administered by gavage according
** to the following schedules
** 1. As a single dose (5 ml/kg) on either day 3, 6, 11, 15 or 17 of g
F007 Presumed pregnant female Sprague-Dawley rats were used for
** this series of studies.
**
** Prudhoe Bay crude oil was administered by gavage according
** to the following schedules
** 1. As a single dose (5 ml/kg) on either day 3, 6, 11, 15 or 17 of
* gestation.
** 2. Daily from gestation days 6-17 incl at a dose level of 1.0 or 2.0
* ml/kg.
** 3. As a single dose on day six of gestation at either 2, 5, 7 or 10
* ml/kg.
** Respective controls received equivalent amounts of saline.
**
** On day 18 of pregnancy, animals were sacrificed and the
** numbers and position of implantations, resorptions and dead
** fetuses were recorded. The live fetuses were removed,
** weighed and inspected for gross external abnormalities with
** the aid of a dissection microscope. Skeletal examinations
** and examination of soft internal tissues were not carried
** out in this preliminary study.
** Student's t-test was used for comparing control and
** experimental results. P values were derived from a 2-tailed
** table of Student's values for t. The level of significance
** chosen was P<0.05.
F008 IUC4
F020 4765
EOR
F002 37
F010 5.8.2
F004 3
F005 RE
F006 Khan, S., Martin, M., Payne, J. F. and Rahimulta, D. (1987)
** Embryotoxic evaluation of a Prudhoe Bay crude oil in rats
** Toxicology Letters Vol 38, pp 109-114
F007 Khan, S., Martin, M., Payne, J. F. and Rahimulta, D. (1987)
** Embryotoxic evaluation of a Prudhoe Bay crude oil in rats
** Toxicology Letters Vol 38, pp 109-114
F008 IUC4
F020 4766
EOR
F002 37
F010 5.8.2
F004 3
F005 RL
F006 Probably not conducted to GLP.

** Study was a preliminary study and was limited in scope.
** Only information on embryo and fetotoxicity was derived in
** this study.
F007 Probably not conducted to GLP.
** Study was a preliminary study and was limited in scope.
** Only information on embryo and fetotoxicity was derived in
** this study.
F008 IUC4
F020 4767
EOR
F002 37
F010 5.8.2
F004 3
F005 RS
F006 There were no maternal deaths following oral administration
** of PBCO although at high doses the animals exhibited signs
** of intoxication (crouching, indicative of acute irritation).
**
** Maternal body weight changes were significantly less in
** ani
F007 There were no maternal deaths following oral administration
** of PBCO although at high doses the animals exhibited signs
** of intoxication (crouching, indicative of acute irritation).
**
** Maternal body weight changes were significantly less in
** animals that had been treated with PBCO at early days of
** gestation, in particular those animals given PBCO on day 6
** of gestation only.
** The body weight data are shown in the following tables.
** Note that although the author provided data on mean and
** standard deviation, for simplicity this summary only
** contains mean values (SDs are not shown). Statistically
** significant differences are indicated.
**
** Body wt changes for dams given a single (5 ml/kg) dose of
** PBCO. Figures in parenthesis indicate group size
**

** Treatment	Body weight change (g on days indicated)				
** Day	0-6	6-11	11-15	15-18	0-18
** Control (11)	30.4	32.6	28	30	121
** 3 (9)	32.75	28.25	23.5*	31	115.22
** 6 (10)	28.8	22.4*	24.2*	27.8	103.2*
** 11 (10)	34.8	35.6	16*	35.2	122.6
** 15 (10)	32.5	33.5	28	35.3	131.5
** 17 (10)	31.67	34	27.33	31.67	124.67

**
** * Significant from control (P<0.05)
**
** Effect of different doses of PBCO on dam body weight changes
** No. of animals shown in parenthesis

** Treatment	Body weight change (g on days indicated)				
**	0-6	6-11	11-15	15-18	0-18
** Control (8)	27.33	25.67	27.33	32	112.33

**
** Dose (ml/kg) given on day 6 only
** 2 (8) 27.5 25 23* 27.5 1.3
** 5 (10) 26.21 18.08* 19.5* 26.92* 91.22*
** 7 (8) 26.67 17.33* 15.35* 26.92* 87.67*
** 10 (8) 30.67 14.33* 11* 20* 75.67*
**
** Dose given on days 6-17 daily
** 1 (10) 31 21* 23.5* 22* 97.5*
** 2 (10) 27.5 16.5 13* 17* 75*
**
** * Significant from control (P<0.05)
**
** The author concluded that the reduced body weight gains were
** probably due to the high resorption rates that occurred.
**
** Effects on fetal viability and development
**
** The results of the effect of administering PBCO as a single
** dose of 5 ml/kg at different stages of gestation are
** summarized below:
**
** Values shown are mean±SD
** * significant from control (p<0.05)
**
** No. Resorp- No. Fetal Fetal Crown-
** implants tions/ live weight rump length per dam dead
* fetuses (g) (mm)
** fetuses
** Day 0
** 13.5±2.39 4.68±.93 13±1.24 1.267±.09 22.2±1.44
** Day 3
** 14.5±1.92 10.77±1.02* 11.25±1.5 1.19±.08* 20±1.73*
** Day 6
** 3.23±1.43 15.48±1.32* 11.02±1.49* 1.2±.09* 1.16±1.38
** Day 11
** 2.9±1.26 8.22±1.08* 11.4±.89* 1.287±.11 21.56±1.37
** Day 15
** 12.72±1.63 5.02±1.19 12.5±.71 1.313±.083 22.65±1.33
** Day 17
** 13±1.8 3.62±.89 12.33±1.15 1.304±.124 23.01±1.41
**
** These results demonstrate that the incidence of resorptions
** and dead fetuses was increased in the animals given PBCO
** during the early days of pregnancy whereas those given PBCO
** during the later stages of pregnancy were unaffected. The
** greatest effect was on day 6 of gestation.
**
** Repeated daily administration of low doses (1 or 2 ml/kg) on days 6
* through 7 showed an additive effect and at a dose
** level of 2 ml/kg/day resulted in up to 40% fetal deaths.
** These data are shown in the following table.
**

**
** The effect of different doses of PBCO
**
** Values shown are mean ± SD
** * significant from control (p<0.05)
**
** No. implants per dam | Resorptions/ dead fetuses (%) | No. live fetuses | Fetal weight (g) | Fetal Crown-rump length (mm)
** Dose (ml/kg)
** 0 13.95±2.23 3.96±0.87 13.72±1.56 1.309±.091 21.62±1.17
**
** Day 6
** 2 14±2.63 6.84±1.21 13.85±2.33 1.326±.119 21.78±1.54
** 5 13.26±1.95 12.67±1.18* 11.73±1.47* 1.216±.09* 21.54±1.27
** 7 12.93±2.35 13.23±1.35* 11.68±1.46* 1.2±.109* 21.28±.89
** 10 14.12±2.12 22.86±1.98* 10.5±1.59* 1.192±.108* 0.73±1.19
**
** Day 6-17 (daily)
** 1 13.92±2.46 17.5±2.02* 10±1.95* 1.184±.107 0.16±1.34*
** 2 12.93±1.98 43.8±1.92* 8.72±1.34* 1.182±.097* .96±1.31*
**
** In conclusion, administration of PBCO to pregnant females
** resulted in an increased incidence of resorptions, increased fetal death
* and decreased fetal body weight. These effects occurred at doses which
* were maternally toxic.
F008 IUC4
F020 4768
EOR
F002 37
F010 5.8.3
F004 1
F005 ME
F006 Groups of five 10-12 week old male mice were injected i.p.
** daily for 5 days with test samples in trioctanoin at doses
** of 1 and 2.1 g/kg. Control mice were injected with
** trioctanoin (5x0.25 ml) or BaP (5x0.05 or 1x0.07g).
** After 35 days, the
F007 Groups of five 10-12 week old male mice were injected i.p.
** daily for 5 days with test samples in trioctanoin at doses
** of 1 and 2.1 g/kg. Control mice were injected with
** trioctanoin (5x0.25 ml) or BaP (5x0.05 or 1x0.07g).
** After 35 days, the animals were sacrificed, the cauda
** epididymes were removed, combined and minced in 0.9% NaCl or
** Tyrode's solution and the sperm were stained by adding eosin
** Y solution. The preparations were spread on slides and
** air-dried. The percentage of abnormal sperm was determined
** in 500 sperm from each animal. For control of bias in
** scoring, positive and negative control slides from a
** previous experiment were included at a ratio of 1 bias
** control slide to 5 new slides.

F008 IUC4
F020 4769
EOR
F002 37
F010 5.8.3
F004 1
F005 RE
F006 Lockard, J. M., Prater, J. W., Viau, C. J., Enoch, H. G. and
** Sabharwal. P. S. (1982)
** Comparative study of the genotoxic properties of Eastern and
** Western U. S. shale oils, crude petroleum and coal-derived
** oil.
** Mutation Research Vol 102, pp
F007 Lockard, J. M., Prater, J. W., Viau, C. J., Enoch, H. G. and
** Sabharwal. P. S. (1982)
** Comparative study of the genotoxic properties of Eastern and
** Western U. S. shale oils, crude petroleum and coal-derived
** oil.
** Mutation Research Vol 102, pp 221-235
F008 IUC4
F009 28-08-2003
F020 4770
EOR
F002 37
F010 5.8.3
F004 1
F005 RS
F006 The data from the study are given in the following table and
** show that treatment with Wilmington crud did not cause an
** increase in the percentage of abnormal sperm.
**
**
** Sample Dose No. survivors/ Abnormal sperm
** no. treated (%±SD)
**
** Triocta
F007 The data from the study are given in the following table and
** show that treatment with Wilmington crud did not cause an
** increase in the percentage of abnormal sperm.
**
**
** Sample Dose No. survivors/ Abnormal sperm
** no. treated (%±SD)
**
** Trioctanoin
** 5x0.25 ml 10/10 4.5±1.7
**
** Wilmington crude
** 5x2.1 g/kg 5/5 6.1±2.1
** 5x1.0 g/kg 5/5 5.4±1.3
**
** BaP 5x0.05 g/kg 10/10 19.7±16.6
F008 IUC4

F020 4771
EOR
F002 37
F010 5.9
F004 1
F005 ME
F006 The authors summarized the protocol in the following table.
**
** Animals
** Charles River (ChR-CD) male mice
** Six weeks old
** Toe clipped for identification
** Backs shaved free of hair
**
** Group size
** 30 mice per group
**
** Housing
** 2 mice per cage
**
** Diet
** Water
F007 The authors summarized the protocol in the following table.
**
** Animals
** Charles River (ChR-CD) male mice
** Six weeks old
** Toe clipped for identification
** Backs shaved free of hair
**
** Group size
** 30 mice per group
**
** Housing
** 2 mice per cage
**
** Diet
** Water and laboratory chow ad libitum
**
** Observation
** Daily
**
** Clipping of hair on the back
** Weekly, as needed
**
** Treatments
** Initiation: 50µl material as received
** Promotion: 2 weeks after initiation treatment
** 2.5µg phorbolmyristate acetate (PMA)/0.1ml acetone
** Three times a week for 180 days
**
** Parameters measured
** Body weight, recorded weekly

** Time to development of first tumor
** Tumors charted monthly
** Mortality
F008 IUC4
F020 4772
EOR
F002 37
F010 5.9
F004 1
F005 RE
F006 Calkins, W. H., Deye, J. F., Hartgrove, R. W., King, C. F.
** and Krahn, D. F. (1981)
** Synthetic crude oils from coal: Mutagenicity and
** tumor-initiating screening tests.
** In Mahlum, D. D. et al (Eds)
** Coal conversion and the environment pp 461-47
F007 Calkins, W. H., Deye, J. F., Hartgrove, R. W., King, C. F.
** and Krahn, D. F. (1981)
** Synthetic crude oils from coal: Mutagenicity and
** tumor-initiating screening tests.
** In Mahlum, D. D. et al (Eds)
** Coal conversion and the environment pp 461-470
** U. S. Dept. of Energy, Washington D. C.
F008 IUC4
F020 4773
EOR
F002 37
F010 5.9
F004 1
F005 RL
F006 The study was a preliminary screen and was not fully
** reported. The data are however are useful in assessing the
** tumor initiating activity of crude oil.
F007 The study was a preliminary screen and was not fully
** reported. The data are however are useful in assessing the
** tumor initiating activity of crude oil.
F008 IUC4
F020 4774
EOR
F002 37

F010 5.9
F004 1
F005 RS

F006 Sample	No. mice with tumors lasting >30 days	Av No. tumors/ tumor-bearing mouse	No. days to first mouse with tumor
** Benzo[a]pyrene positive control			
**	26	6.0	48
** South Louisiana Crude			

** 5 1.4 71
** Toluene and PMA
** 4 1.5 111
** PMA
F007 Sample No. mice Av No. No. days
** with tumors/ to first
** tumors tumor- mouse with
** lasting bearing tumor
** >30 days mouse
**
** Benzo[a]pyrene positive control
** 26 6.0 48
** South Louisiana Crude
** 5 1.4 71
** Toluene and PMA
** 4 1.5 111
** PMA alone
** 3 1.0 92
**
** The authors concluded that the South Louisiana crude oil was
** not an initiator.
F008 IUC4
F020 4775
EOB
C
X