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86930000306 - 308

# Mobil Oil Corporation

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ATTN 8(d) Health and Safety Reporting Rule (Notification/Reporting)

Gentlemen

On behalf of Mobil and its manufacturing and processing establishments located in the United States and in accordance with Health and Safety Study Reporting Regulations for TSCA Section 8(d) in 40 CFR Part 716, we hereby submit the following studies

| | CAS NO. | LISTED SUBSTANCE | REPORT, PROJECT OR STUDY NO. | TEST NAME(S) |
|---|---|---|---|---|
| 86930000306 | 80-05-7 | 4,4-Isopropylidenediphenol | *65163 | Biodegradation |
| 86930000307 | 1330-20-7<br>100-41-4<br>108-88-3<br>95-63-6 | Xylenes<br>Ethyl Benzene<br>Toluene<br>Pseudocumene | *65363 | Biodegradation |
| 86930000308 | 102-71-6<br>111-42-2 | Ethanol,2,2,2-Nitrilotris<br>Diethanolamine | *65430 | Standard Ames Test |

* Studies involve a mixture containing the indicated TSCA Section 8(d) listed substance.

Two copies of each study have been provided with appendixes removed that contained underlying data in the form of laboratory notes and daily monitoring records One copy is complete with confidential information (e.g. employee names, product names and formulation information) identified by bracketing and the basis of the claim there noted. The second copy is for public disclosure with such confidential information removed.

If you should have any questions regarding the above information or any aspect of this matter, I can be contacted at the above address or telephone number (609) 737-5436.

Sincerely,

D E Wagner

D. E Wagner
Manager
Product Safety Coordination

DEW/dmg
Attachment
8D-06243 sam

**Mobil Environmental Health and Safety Department**
**Environmental and Health Sciences Laboratory**

| | |
|---|---|
| TO LIAISON: | |
| STUDY NO | 65163 |
| CRU NO | 92248 |
| SAMPLE NAME: | |
| STUDY TITLE: | Aerobic Biodegradation Study of |
| REQUESTOR: | MRDC, CRL and Paulsboro |

Results: The biodegradability of (CRU# 92248) was assessed, at an initial test material concentration of 10 mg carbon/L, using the EPA Shake Flask method (EPA 560/6-82-003, CG-2000) with an unacclimated sewage/soil inoculum. Using the unacclimated inoculum, this ultimate biodegradability test, which follows the conversion of test material carbon to $CO_2$ is equivalent to the OECD Modified Sturm Test (301B).

In 28 days, 83.6% of the carbon in was converted to $CO_2$. In the same period, 71.7% of the carbon in the positive control, Rapeseed Oil, was converted to $CO_2$.

met both criteria necessary for classification in the OECD biodegradability category of Ready Biodegradable To be placed in this category, greater than 60% of a test material's carbon must be converted to $CO_2$ in 28 days and, in addition, once 10% of the material has been converted to $CO_2$, the 60% mark must be reached in 10 days.

3/23/93
STUDY DIRECTOR DATE

3/30/93
SECTION MANAGER DATE

4/1/93
MANAGER DATE
ENVIRONMENTAL AND HEALTH
SCIENCES LABORATORY

DISTRIBUTION:

Liaison, Archives (Original), Database, Study Director/ Scientists

8693000003006

# Aerobic Biodegradation Study of

(Method is a modification of the EPA Shake Flask Test, EPA 560/6-82-003, CG-2000)

| | |
|---|---|
| STUDY NO | 55163 |
| MATERIAL TESTED: | |
| CRU NUMBER | 92248 |
| REQUESTOR DIVISION. | MRDC, CRL and Paulsboro |
| STUDY PERFORMED BY | Mobil Environmental and Health Sciences Laboratory, Pennington-Rocky Hill Road Pennington, New Jersey 08534 |
| EXPERIMENTAL START DATE: | 2/10/93 |
| EXPERIMENTAL TERMINATION DATE: | 3/11/93 |

2/18/93
PRINCIPAL INVESTIGATOR(S)
&

3/23/93
STUDY DIRECTOR
, Ph. D

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# SUMMARY

The biodegradability of (CRU# 92248) was assessed, at an initial test material concentration of 10 mg carbon/L, using the EPA Shake Flask method (EPA 560/6-82-003, CG-2000) with an unacclimated sewage/soil inoculum Using the unacclimated inoculum, this ultimate biodegradability test, which follows the conversion of test material carbon to $CO_2$, is equivalent to the OECD Modified Sturm Test (301B)

In 28 days, 83 6% of the carbon in was converted to $CO_2$ In the same time period, 71 7% of the carbon in the positive control, Rapeseed Oil, was converted to $CO_2$.

met both criteria necessary for classification in the OECD biodegradability category of Ready Biodegradable. To be placed in this category, greater than 60% of a test material's carbon must be converted to $CO_2$ in 28 days and, in addition, once 10% of the material has been converted to $CO_2$, the 60% mark must be reached in 10 days

# INTRODUCTION

In evaluating the toxicity of a test material, it is important to consider its persistence in the environment. This is the case because a highly toxic compound may be rapidly degraded in the environment and thus never have the opportunity to cause toxic effects in plants and animals while a slightly or moderately toxic compound may persist for months or years allowing time for toxic levels to be reached through bioaccumulation. The ultimate biodegradation of organic material leads to the production of carbon dioxide, water, and other mineral components. In the Shake Flask Test, the carbon dioxide is trapped in alkali and quantified by titration. To determine the percent of test material biodegraded to carbon dioxide, the experimental yield of carbon, as carbon dioxide from the test material, can be compared to the quantity of test material carbon originally added to the test system. No attempt was made during this study to determine the purity, homogeneity, or stability of the test material.

The Organization for Economic Cooperation and Development (OECD) has established a classification scheme by which a material can be categorized as ready biodegradable. A material placed in the ready biodegradable category must be rapidly and completely biodegraded in a variety of aerobic environments. The guidelines specify that in tests which measure organic carbon conversion to $CO_2$, as is the case for the Shake Flask Test, 60% of the test material carbon must be converted to $CO_2$ in 28 days. In addition, once biodegradation activity is initiated, defined as the point at which 10% of the test material carbon has been converted to $CO_2$, the 60% mark must be reached within 10 days.

It is important to note that the ready biodegradability criteria are very stringent and many materials that can be completely biodegraded aerobically in reasonable time frames will not meet these criteria. Such materials are classified as inherently biodegradable and should not be considered environmentally persistent.

# MATERIALS AND METHODS

Chemicals and Test Materials:

TEST MATERIAL
CRU No 92248

The test material was obtained from MRDC, CRL and Paulsboro The material is reported as 100% and no further determinations of the purity or homogeneity of this material were made. The storage time for of 2 years at room temperature was not exceeded

Acclimation and Test Medium Preparation:

The shake flask medium was prepared from the following stock solutions:

Solution 1 8 5 g of potassium phosphate monobasic, 21 75 g potassium phosphate dibasic, 50.3 g sodium phosphate dibasic heptahydrate, and 1 7 g ammonium chloride were dissolved in 500 mL of distilled water and were made to 1L.

Solution 2 22.5 g of magnesium sulfate heptahydrate were dissolved in distilled water and made to 1L with the same.

Solution 3: 27.5 g of calcium chloride were dissolved in distilled water and made to 1L with the same.

Solution 4 0 25 g of ferric chloride hexahydrate was dissolved in distilled water and made to 1L with the same.

Solution 5: 40.0 g of ammonium sulfate were dissolved in distilled water and made to 1L with the same.

Solution 6: 35 4 mg manganous chloride tetrahydrate, 57 2 mg boric acid, 42 8 mg zinc sulfate heptahydrate and 36 8 mg ammonium molybdate tetrahydrate were dissolved in distilled water and made to 1L with the same.

Fifteen liters of distilled water were added to a 25L carboy followed by 150 mL of solution 1 15 mL of solutions 2, 3, 5, and 6, and 60 mL solution 4 This medium is referred to as test medium.

The medium was mixed by inverting the carboy several times prior to dispensing.

Sewage Inoculum Preparation:

The sewage inoculum was prepared by collecting mixed liquor (approximately 1 liter) from an activated sludge treatment tank at the Pennington Municipal Wastewater Treatment Plant, Pennington, N.J. The sewage was collected the same day it was to be used and labeled as to date and time collected. The sewage was homogenized in a blender (approximately three

15 second pulses), transferred to an Erlenmeyer flask and aerated until ready for use. Three 5 mL aliquots of mixed liquor were dried in an oven at 103 ± 5°C for approximately one hour and the weight of dry solids was measured. Based on the solids measurement, enough mixed liquor was added to each test flask to achieve a solids concentration of 30 mg/L.

Soil Inoculum.

Soil #104 was obtained from a forested area within D&R State Park, N.J. This soil was kept in the cold (approximately 4°C) until used.

Test Material Preparation:

Solutions of the test and reference materials were prepared by adding an appropriate amount of the test material (or reference material) to a volumetric flask and dissolving it in methylene chloride. These solutions were added to the appropriate flasks to a final concentration of 10 mg/L carbon and the methylene chloride was allowed to volatilize under a gentle stream of air before the test medium was added.

Test Procedures

The test and reference materials were added to duplicate 2L Erlenmeyer flasks, as described above, followed by enough volume of test medium to yield a 1L final volume, after inoculum addition. Enough mixed liquor was added to each flask, to give a final dry sludge solids concentration of 30 mg/L. In addition 0.1 g of soil #104 (Appendix IV) was added to each flask followed by 1 mL of yeast extract solution (15 mg yeast extract per 100 mL of distilled water). The blank flasks were treated in an identical manner, except they received methylene chloride without an added substrate, as described above. The flasks were closed with neoprene stoppers containing a 10 mL KOH trap and an inlet and outlet port. The inlet port consisted of a glass tube which penetrated the stopper and extended well into the medium while the outlet port consisted of a glass tube which penetrated the stopper but ended well above the test medium. The flasks were then tightly stoppered and placed on a rotary shaker at 25±3°C and approximately 150 rpm.

Flask traps were sampled at 1-7 day intervals, depending on microbial activity. The KOH was removed from the trap by means of a 10 mL syringe and placed in a 50 mL Erlenmeyer flask. Using the same syringe, the trap was washed with approximately 10 mL of distilled water and the distilled water wash was placed in the same 50 mL Erlenmeyer flask as the 0.2N KOH. The trap contents and wash received 2 mL of a saturated solution of barium chloride and 0.1 mL of phenolphthalein indicator. The solution was then titrated to a colorless endpoint with 0.2N HCL. The trap was recharged with 10 mL of fresh KOH and inserted back into the flask. Prior to being placed on the shaker, the flask was flushed for several minutes with $CO_2$-free air. Air was bubbled through two consecutive flasks containing first, approximately 0.5L of 5N NaOH and second, approximately 1L of distilled water. This $CO_2$-free air was bubbled through the shake flask medium by disconnecting the inlet and outlet ports and connecting the inlet of the shake flask to the outlet port of the distilled water flask by means of tygon tubing. After aeration, the shake flask inlet port was disconnected and reconnected by means of tygon tubing to the outlet port of the shake flask.

One day prior to the final sampling, the medium was acidified with 1 mL of concentrated sulfuric acid and left on the shaker a minimum of 12 hours prior to sampling of the trap.

Data Analysis

Data analysis was based on the following equations.

$$2KOH + CO_2 <-> K_2CO_3 + H_2O$$

$$K_2CO_3 + BaCl_2 -> BaCO_3 \text{ (precipitate)} + 2KCl$$

As can be seen from the equations above, two moles of base are consumed for each mole of $CO_2$ trapped The mg of $CO_2$ trapped in the KOH are calculated from the following formula

(mL acid titrated blank - mL acid titrated test or reference material) * normality of acid * 1 mmole KOH/1 mmole HCl * 1 mmole carbon as $CO_2$/2 mmole KOH * mw carbon = mg C as $CO_2$.

(blank - test or reference material) * 0 2 * 6 = mg C as $CO_2$

Data Storage:

The original report and all supporting documents will be stored in the archives at EHSL

# RESULTS AND DISCUSSION

The biodegradability of (CRU# 92248) was assessed, at an initial test material concentration of 10 mg carbon/l, using the EPA Shake Flask method (EPA 560/6-82-003, CG-2000) with an unacclimated sewage/soil inoculum. Using the unacclimated inoculum, this ultimate biodegradability test, which follows the conversion of test material carbon to $CO_2$, is equivalent to the OECD Modified Sturm Test (301B).

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# REFERENCES

1 Premanufacture Testing of New Chemical Substances, Toxic Substances Control Act, Federal Register, Vol. 44(53), March 16, 1979, Method A 5 1

2 Aerobic Aquatic Biodegradation, Method CG-2000, Chemical Fate Testing Guidelines, Office of Toxic Substances, Office of Pesticides and Toxic Substances, U S. Environmental Protection Agency, August 1982, EPA 560/6-82-003