Environment Canada
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Guidelines on the Use and Acceptability of Oil Spill Dispersants

Environmental Emergency Branch
Report EPS-1-EE-73-1
August 1973

GUIDELINES

ON THE

USE AND ACCEPTABILITY

OF

OIL SPILL DISPERSANTS

August 1973

SUMMARY

This report forms the foundation of Environment Canada Guidelines on the Use and Acceptability of Oil Spill Dispersants. A standard list of dispersants will be prepared on the basis of these guidelines. Only dispersants that satisfy the acceptability criteria shall be allowed for use and, except in the case of extreme emergency, only with the express permission of Environment Canada or delegated provincial authorities. Furthermore, in view of various provincial legislations, the use of dispersants is also subject to the requirements of the provinces concerned. Uses are limited to well specified spill situations and a detailed report on each use is required. Dispersants must contain no highly toxic compounds, must satisfy toxicity and biodegradability specifications, must be reasonably effective under conditions of use, and must be applied in a recommended manner. Procedures for having a dispersant placed on the standard list are presented here along with labelling requirements and recommended methods for determining toxicity, biodegradability and effectiveness. These guidelines will be revised as new knowledge becomes available.

RÉSUMÉ

Environnement Canada utilisera ce rapport comme base des règles qu'il doit établir sur l'emploi et l'admissibilité des agents de dispersion pour nappes de pétrole. Ces règles serviront à dresser une liste standard de dispersants. Seul l'emploi des dispersants qui satisferont les normes d'acceptabilité sera permis, et, sauf en cas d'extrême urgence, seulement par autorisation expresse du Ministère ou des autorités provinciales déléguées. En plus, l'emploi des dispersants doit être CONFORME [redacted] aux exigences des provinces concernées. Il ne sera permis d'utiliser les dispersants que lors de déversements bien déterminés et chaque utilisation fera l'objet d'un rapport détaillé. Les dispersants ne doivent contenir aucun produit hautement toxique, doivent être conformes aux règles de toxicité et de biodégradabilité, doivent être raisonnablement efficaces sur le terrain et doivent être employés selon les méthodes recommandées. Le texte indique comment faire la demande pour obtenir qu'un dispersant soit placé sur la liste standard, quelles sont les exigences quant à l'étiquetage et quelles sont les méthodes recommandées de mesure de la toxicité, de l'efficacité et de la biodégradabilité. Les règles seront révisées à la lumière des nouvelles connaissances.

ACKNOWLEDGEMENTS

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SECTION 1

INTRODUCTION

Oil spills on water present a potential danger to aquatic life and to human health and welfare. While spilled oil must be removed from the water whenever possible, in some well-defined situations its dispersal into the water may be desirable.

Dispersants are chemical agents that promote the dispersion (emulsification, solubilization) of oil in water upon the application of sufficient mixing energy. Coalescence and refloating of dispersed oil droplets may be prevented by the presence of the dispersant on their surface and by subsequent dilution in large volumes of water.

The incentives for dispersing oil in the water are:

(1) the rate of oil biodegradation is increased because of the increased surface-to-volume ratio;

(2) damage to waterfowl is averted because the oil is removed from the water surface;

(3) fire hazards may be reduced;

(4) the adhering of oil to most surfaces is reduced;

(5) the formation of tar-like residue is prevented.

The disadvantages of using dispersants include:

(1) the toxic effects of most dispersants on aquatic organisms;

(2) the increased surface-to-volume ratio increases the toxicity of the oil to aquatic organisms;

(3) the dispersed oil is difficult to remove from surface water supplies;

(4) the inability of many dispersants to effectively disperse floating oil such that the above advantages can be realized;

(5) the lack of knowledge concerning the fate of oil once it has been dispersed;

(6) application of dispersant on volatile oils may increase fire hazards.

Environment Canada recognizes the potential of chemical dispersion as a countermeasure technique for combating oil spills, but realizes that most marketed products have properties that can cause as much or more damage to the environment as could the oil if left untreated. Because of toxic effects, varying reports on performance and the present lack of knowledge on the fate of dispersed oil in the water, appropriate control of their use is warranted. The criteria presented here form the foundation for Guidelines on the Use and Acceptability of Oil Spill Dispersants. A list of dispersants that satisfies the acceptability criteria will be prepared on the basis of these guidelines.

These guidelines shall apply to all areas of jurisdiction of the Federal Government. In other cases, they should be considered as a recommendation in the formulation, coordination and implementation of guidelines for the effective use of dispersants in combating oil spills and for safeguarding the environment.

The Federal Contingency Plan to deal with oil spills is well coordinated with similar provincial plans and defines the involvement of various agencies in spill incidents. In each spill incident, the On-Scene-Commander heads a Response Team whose essential quality is its ability to act swiftly and effectively. Environment Canada recognizes that difficult calculations on effectiveness, temperature effects, toxic concentrations, movement and possible deliterious effects, etc., should be made before and not during an oil spill incident. These guidelines will assist On-Scene-Commanders and Response Teams in this respect.

It is realized at the present time that no laboratory method of estimating field effectiveness and toxicity is completely satisfactory. This is why Environment Canada stresses the importance of receiving detailed reports on all instances of dispersant use. Such reports will be most valuable in future decisions on dispersants, application methods, effectiveness and possible environmental effects.

Environment Canada wishes to encourage further development in dispersant formulations, application and mixing methods, and evaluation procedures, and will consider revising various aspects of parts of the present guidelines as new basic knowledge and technology become available.

SECTION 2

GUIDELINES FOR THE USE OF DISPERSANTS

2.1 General Requirements

(a) Except in the cases of extreme emergency as noted in 2.3 (a) below, chemical dispersants shall be used only with the express permission of Environment Canada or provincial authorities who are administering Section 33 of the Fisheries Act. Furthermore, in view of various provincial legislations, the use of dispersants is also subject to the requirements of the provinces concerned* (see item 2.6). Chemical dispersants can then be used under competent direction and in accordance with recommended techniques.

(b) The On-Scene-Commander**, in consultation with the appropriate agencies (see item 2.6), will determine the priorities of protection in each spill incident and the use of dispersants is subject to these priorities.

(c) Only the dispersants that satisfy the Acceptability Criteria as set forth in these guidelines shall be used.

(d) All uses of chemical dispersants must be formally documented, as outlined in these guidelines.

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* Under the Clean Water Act in Alberta, it is unlawful, even under extreme emergency, to use oil spill dispersants without permission of provincial authorities.

** In the context of these guidelines, the On-Scene-Commander is the person in charge of the clean-up operation and is not necessarily a government representative.

2.2 Restrictions on the Use of Dispersants

The use of dispersants will usually be avoided:

(a) in any waters containing major fish populations or large breeding or migration areas for species of fish or other aquatic life which may be damaged or reduced in market value by exposure to dispersants and/or dispersed oil;

(b) in any waters where such use may significantly affect surface water supplies;

(c) where eventual dilution of the dispersed oil is limited either because the water is quiescent or because the water volume is small relative to the spill size;

(d) on oils that have been deposited on sandy beaches or on shorelines with important flora and fauna;

(e) under conditions where the dispersant is ineffective, as determined by the effectiveness tests described in these guidelines, or in the judgement of the On-Scene-Commander.

2.3 Possible Conditions for the Use of Dispersants

Dispersants that satisfy the acceptability criteria may be considered for use:

(a) when their use will prevent or reduce hazard to human life or limb, or substantial hazard of fire to property. (Caution is advised in the case of spills of volatile oils where both the application of the dispersant and the subsequent agitation may increase fire hazards. Furthermore, it should be realized that in spills of volatile oils (e.g. No. 1 to No. 4 fuel oils), natural agitation by winds, waves and currents may rapidly disperse the oil);

(b) when their use will prevent or substantially reduce hazard to a significant population of waterfowl or marine mammals;

(c) when their use will prevent or substantially reduce significant damage to valuable property except in conditions:

(i) where such use is restricted as noted in 2.2 above and

(ii) where other methods for controlling and removing the oil are reasonably effective.

2.4 Recommended Application Procedures

It should be emphasized that dispersants may be used only when other methods are not reasonnably effective, as outlined in section 2.3. Furthermore, if approval for use has been granted, the following procedures must be followed:

(a) The Effectiveness Table for the dispersant shall be used as a guideline by the On-Scene-Commander to determine its applicability under the given field conditions.

(b) If the dispersant is considered to be effective, it shall be applied only if the proper equipment for application and mixing is available. Such equipment may include portable pump-eductor systems, portable pressure tank applicators, spray boats with spray booms, boats with propellers, other mixing devices, etc.

(c) After using a minimum quantity of dispersant, the On-Scene-Commander will evaluate its effectiveness. If the dispersant is found to be ineffective, dispersing will be discontinued.

(d) When using dispersant, only the strict minimum should be used to correct the condition for which approval was given.

2.5 Research and Development Purposes

For purposes of research and development, Environment Canada, in agreement with provincial authorities may authorize the use of dispersants in specified amounts and locations under controlled conditions irrespective of the provisions of these guidelines.

2.6 Application for Use of Dispersants

Within the Federal Government, the Environmental Protection Service is the coordinating agency responsible for giving approval for the use of oil spill dispersants. In each spill incident, excepting extreme emergencies, the On-Scene-Commander will contact the regional headquarters of the Environmental Protection Service regarding the potential use of dispersants. The Environmental Protection Service, through the Regional Environmental Emergency Teams, will be responsible for contacting the Fisheries and Marine Service and the Environmental Management Service. Dispersants will not be approved for use if any of these three services advise against such action. In cases of dispute or conflicting requirements, the responsibility for obtaining higher direction will rest with the Environmental Protection Service. The Environmental Protection Service will also establish contact with competent provincial authorities regarding the possible approval for dispersant use.

Appendix III gives a contact list for the Environmental Protection Service personnel across Canada.

2.7 Reports of Dispersant Uses

Environment Canada considers it essential that each dispersant use be well documented, and expects a detailed report from the On-Scene-Commander (or the responsible personnel in charge) on each instance of dispersant application. Such reports may assist later in surveys of spill areas, and will contribute to our knowledge of effectiveness and the possible ecological effects under various field conditions.

The reports need not be made in a specific form, but should contain as much information as possible and should generally follow the check list given here.

Check List of Required Information:

(a) Details of the Spill:

- Location
- Date and Time of Spill
- Source and Type of Oil
- Estimated Amount, Slick Area and Thickness
- Appearance of the Oil
- "Age" of the Oil

(b) Environmental Conditions:

- Air and Water Temperature
- Wind, Waves and Currents
- Water Depth

(c) Dispersant Application:

- Reason for Dispersant Use
- Type of Dispersant
- Amount Used
- Application and Mixing Methods (equipment, manpower, time)

(d) <u>Observations:</u>

- Estimated amount of oil left on the water surface
- Observations on the dispersed oil (spreading, disappearance, resurfacing, etc.)
- Any observations on possible effects of oil (waterfowl, fish, shorelines, vegetation).
- Any observations hours/days after the application.

Reports should be addressed to:

Regional Environmental Emergency Co-ordinator
Environmental Protection Service
Regional Office*

* Please refer to Appendix III for addresses of the various regional offices.

SECTION 3

ACCEPTABILITY CRITERIA

Environment Canada will publish a list of dispersants that satisfy the acceptability criteria which will serve as a guide to all organizations and spill-control cooperatives that will stockpile contingency supplies of dispersants. In oil spill incidents, only those dispersants listed will be acceptable for use and only then in accordance with these guidelines.

3.1 Prohibited Ingredients

(a) The dispersant shall not contain aromatic hydrocarbons, chlorinated hydrocarbons, nor heavy metals in concentrations greater than:

Total aromatic hydrocarbons: 3%
Total chlorinated hydrocarbons: 0.05 mg/litre
Mercury: 0.005 mg/litre
Cadmium: 0.01 mg/litre
Lead: 0.05 mg/litre

(b) The dispersant shall be non-corrosive to the storage containers and shall not contain any caustic alkali or free mineral acid.

3.2 Effectiveness

(a) Dispersion Stability

At least 60% on No. 2 fuel oil at a dispersant/oil ratio of 0.3 and at a temperature of 15^{o}C should remain in emulsion for six hours as determined by the Simulated Environmental Tank (S.E.T.) test procedure, using the average of results from at least 3 duplicate tests (see Appendix I).

(b) Effectiveness Using S.E.T.

Tests should be performed with the S.E.T. to determine the minimum dispersant/oil ratio needed to disperse 65% of the floating oil in a "10-minute" test. Using the average of results from at least 3 duplicate tests, this ratio should be obtained for:

(i) No. 2 fuel oil* at 5°C (41°F) and 15°C (59°F)

(ii) Crude oil at 5°C and 15°C

(iii) Medium Bunker fuel oil at 5°C and 15°C

(iv) Heavy Bunker fuel oil at 5°C and 15°C

For a given oil and temperature, if the dispersant cannot disperse 65% of the oil or if the dispersant/oil ratio is greater than unity, the dispersant is considered ineffective under these conditions. If the dispersant is ineffective for all the oils at both temperatures, it will not be approved. A table of test results (Effectiveness Table) should be constructed as shown in the example below, which considers a hypothetical dispersant:

TABLE I

EFFECTIVENESS TABLE

	Minimum Dispersant/Oil Ratio			
Oil / Temp.	No. 2 F.O.	Crude	Med. Bunker	Heavy Bunker
5°C	0.50	ineffective	ineffective	ineffective
15°C	0.25	1.0	ineffective	ineffective

(Minimum dispersant/oil ratio needed to disperse 65% of floating oil in a "10-minute" test, considering a hypothetical dispersant).

Here, the hypothetical dispersant is considered effective with No. 2 fuel oil at 5°C and 15°C and with crude oil at 15°C.

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* (i) No. 2 fuel oil as specified by ASTM D-396-67 and with an aromatic content of 15% by volume minimum.

(ii) Crude oil with gravity (°API): 30-43; and residuum (700°F): 30 volume percent minimum.

(iii) Medium Bunker fuel oil with viscosity (SSF at 122°F): 175-200

(iv) Heavy Bunker fuel oil with viscosity (SSF at 122°F): 250-300

(c) Other Effectiveness Tests

Environment Canada recognizes the difficulties in obtaining the above data and the limitations of the S.E.T. test, and will consider effectiveness data obtained by alternate methods. In such cases, experimental conditions and details must be well documented and Environment Canada must be satisfied that the data can be used as a meaningful measure of dispersing effectiveness under field conditions. These date must also be compared with the reference dispersant identified in Appendix I, item 3 (C).

3.3. Toxicity

(a) The dispersant shall have a 96-hour LC50 which is greater than 1000 mg/litre as measured by methods approved by Environment Canada (see Appendix II). (Other bioassay data obtained previously to these guidelines by other methods will be considered and accepted if they are judged satisfactory).

(b) A mixture of the dispersant and No. 2 fuel oil in the ratio of 1:1 shall have a 96-hour LC50 which is greater than 100 mg/litre, as measured by methods approved by Environment Canada (see Appendix II).

3.4 Degradability

The dispersant must be biodegradable as determined by a 20-day product degradation analysis, and the biodegradation products must not be more toxic than the dispersant itself as determined by a simple bioassay (see Appendix II, item 6).

3.5 Hazards to Handling Personnel

In the opinion of the Department of National Health and Welfare, if the dispersant constitutes a serious occupational hazard to health, it will not be approved.

3.6 Labelling Requirements

All containers of the dispersant must carry a label, in English and French, with the following information:

(a) the following statement:

EXCEPT IN THE CASE OF FIRE OR SAFETY HAZARDS, THIS OIL SPILL DISPERSANT MUST NOT BE USED ON WATER WITHOUT THE EXPRESS PERMISSION OF ENVIRONMENT CANADA AND COMPETENT PROVINCIAL AUTHORITIES

EXCEPTÉ EN CAS DE FEU OU DE SÉCURITÉ, CE DISPERSANT NE DOIT PAS ÊTRE EMPLOYÉ POUR DISPERSER DE L'HUILE SUR L'EAU SANS LA PERMISSION EXPRESSE D'ENVIRONNEMENT CANADA ET DES AUTORITÉS PROVINCIALES COMPÉTENTES

(b) name, brand or trademark, if any, under which the chemical is sold.
(c) name and address of the manufacturer and distributor.
(d) pour point in ^{o}C and (^{o}F).
(e) flammability notice, as required.
(f) special handling, storage and safety precautions, as required.
(g) date of production or production lot number.

3.7 Brochure Requirements

Each container must be accompanied by a pamphlet containing the following minimum information:

1. Information required on the label except item 3.6 (g).
2. Effectiveness Table. Describe recommended procedures for use with regard to water salinity, water temperature, types and ages of oils and identify recommended equipment to obtain the most efficient dispersion.
3. Hazards to handling personnel.
4. Toxicity level of the dispersant as determined by the Bioassay Test described in these guidelines.
5. Physical properties such as specific gravity, viscosity at $0^{o}C$ and $15^{o}C$,aqueous and organic miscibility and shelf life.

SECTION 4

PROCEDURES FOR HAVING A DISPERSANT PLACED ON THE STANDARD LIST

The information required by Environment Canada in order to place a specific chemical dispersant on the standard list is itemized in the following form:

Chief, Research and Development
Environmental Emergency Branch
Environmental Protection Service
Environment Canada
Ottawa, Ontario
KIA OH3

APPLICATION FOR HAVING A DISPERSANT PLACED ON THE STANDARD LIST

Date: ____________________

1. PRODUCT TRADE NAME: ____________________

   Name of Manufacturer: ____________________

   Address: ____________________

   Distributor in Canada: ____________________

   Address: ____________________

   Technical Representative: ____________________

   Address: ____________________

   Telephone: Area Code: ________ Number: ____________ Ext.: ____________

   Telex/TWX No.: ____________

2. COMPOSITION:

   Submit details of the composition of the product such as generic structural type (e.g. alkyl aryl sulphonate, ethoxylated alcohol, etc.) and other pertinent information.

   ____

   ____

   ____

   ____

   ____

   ____

3. ANALYTICAL DATA:

Indicate the laboratory test procedures and instruments used in the analyses for obtaining the following data:

Concentration in mg/litre of:

Lead: ____________________ Mercury: ____________________

Cadmium: ____________________ Total chlorinated hydrocarbons: ____________________

Total aromatic hydrocarbons: ____________________.

Solubility at 15°C in: fresh water ____________________

sea water ____________________

Colour (visual): ____________________ Flash Point (open cup): __________ °C

Pour Point: ____________________ °C Freezing Point: ____________________ °C

Specific Gravity: ______________ at __________ °C

Viscosity: ____________________ at __________ °C in ________ units

pH: ______________

4. HAZARD TO OPERATORS:

Inhalation (acute LC50 to __________) ______________

Skin Irritation or sensitivity concentration: ____________________

Eye Irritation: ______________

Sensory Threshold properties: ____________________

Hazardous gases produced in combustion: ______________

Chronic hazards: ____________________

5. PERFORMANCE EFFECTIVENESS:

(i) S.E.T. test procedure (attach all experimental data)

(a) Average (N = 3) percent of #2 Fuel Oil dispersed at 6 hours = ________%.

(b) Minimum dispersant/oil ratio needed to disperse 65% of floating oil after 10 minutes (complete table below)

	Minimum Dispersant/Oil Ratio			
oil type / temperature	#2 Fuel Oil	Crude	Medium Bunker	Heavy Bunker
5^oC				
15^oC				

EFFECTIVENESS TABLE

(ii) Other effectiveness data. Detailed descriptions of test procedures should be submitted with this application.

6. TOXICITY:

Submit details of the toxicity test as outlined in Appendix II.

(a) Dispersant Toxicity

Acute Toxicity to Aquatic Species

Species	Type of Water	Temperature, °C	96-hour LC50
______	______	______	______
______	______	______	______
______	______	______	______

(b) Oil/Dispersant Toxicity

The test shall be performed with No. 2 fuel oil.

Dispersant + No. 2 Fuel Oil in a dispersant/oil ratio of__________ and a test species of____________ has a 96-hour LC50 at________°C equal to___________mg. of mixture per litre.

7. DEGRADATION:

(a) Submit details of Product Degradation by chemical analysis over a 20-day period using the shake flask or other equivalent technique.

(b) The dispersant, which has been aged for 16 days with initial concentration that of the 96-hour LC50 shall not be lethal to more than half the test fish in a four-day exposure (submit details of test as outlined in Appendix II).

(c) Give shelf-life of dispersant in the sealed container.

8. METHODS OF USE:

Describe recommended procedures for use with regard to water salinity, water temperature, types and ages of oils and identify recommended equipment to obtain the most efficient dispersion.

9. PRODUCTION AND DISTRIBUTION:

Give address and telephone number of a central contact where inventory control of product across Canada is exercised on a 24 hr basis. If not feasible, give addresses and telephone numbers of all storage and distribution depots of product. If possible, submit information, on the scale of production that can be undertaken, and if necessary, maintained to meet emergency requirements.

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APPENDIX I

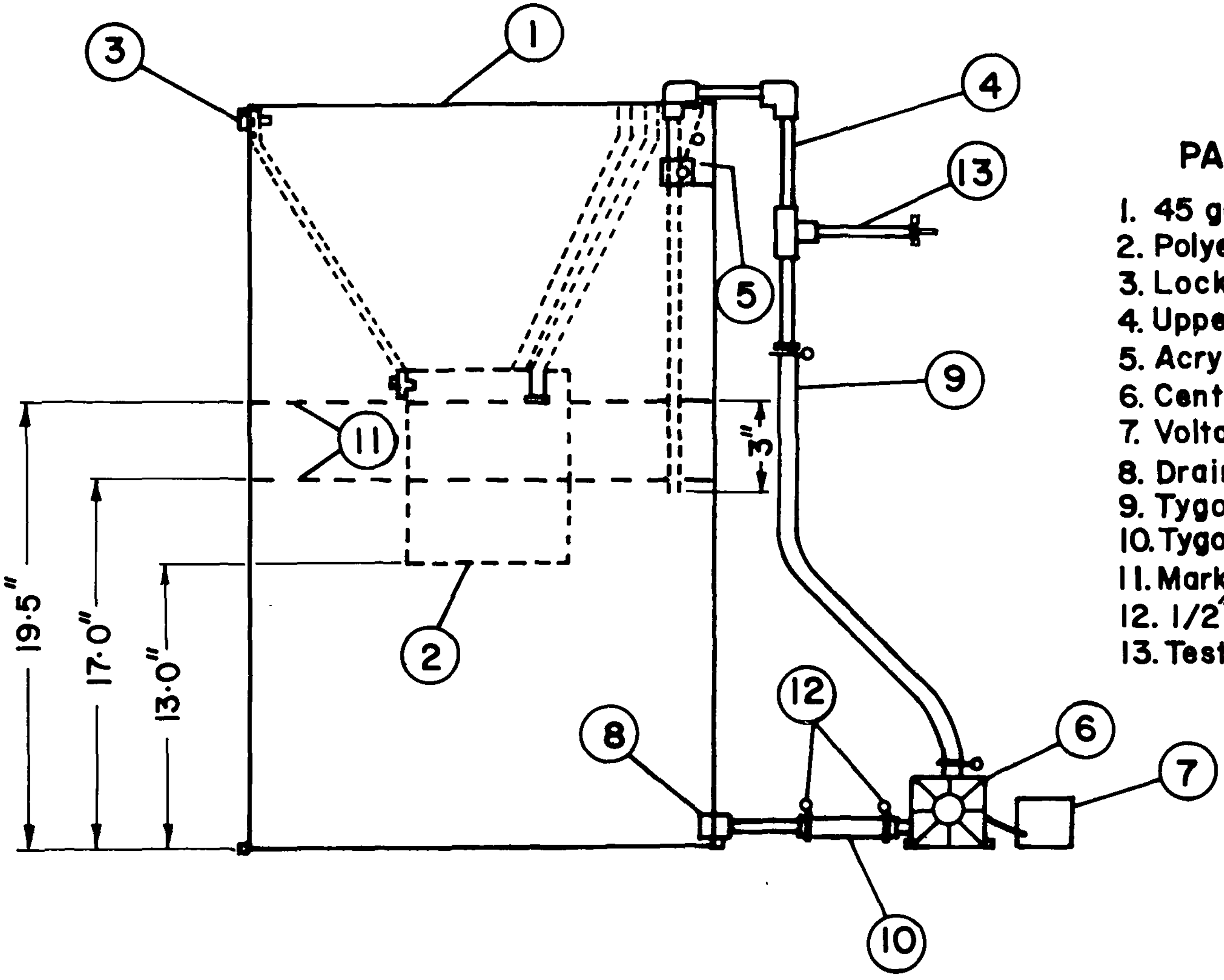
EFFECTIVENESS TEST

1. Introduction

Chemical dispersion of an oil slick is a complex process which is dependent on a number of variables existing under field conditions, for example: (a) chemical composition of the water; (b) physical and chemical properties of the oil; (c) water temperature; (d) length of time of atmospheric exposure; (e) wind, wave and current action; (f) type, quantity and manner of application of the dispersant; and (g) degree and type of agitation.

At present, no test is available which satisfactorily considers all these factors in the determination of a measure of dispersing effectiveness under field conditions. Nevertheless some data are needed to determine whether a particular dispersant will perform effectively under certain conditions. The modified Simulated Environmental Tank (S.E.T.) test described here, despite its limitations, is the best test presently available and is recommended until a superior method is developed.

The S.E.T. test was developed by the U.S. Navy to measure the oil-emulsifying capability of solvent bilge cleaning agents and was modified by the Federal Water Pollution Control Administration[18] and the California State Water Resources Control Board[7] for measuring the performance effectiveness of chemical dispersants. It involves the use of a "Simulated Environmental Tank" (see Figure 1). A sample of chemical dispersant is sprayed on an oil slick formed on the surface of water. The mixture is hosed and the water is recirculated continuously, samples being withdrawn at given intervals and analyzed to determine the percent oil dispersed.



PARTS LIST

1. 45 gallon steel drum
2. Polyethylene cylinder
3. Locking
4. Upper part of plumbing
5. Acrylic lock-unit for plumbing
6. Centrifugal recirculating pump
7. Voltage regulator
8. Drain
9. Tygon Tubing 3/4" I.D. x 2·5ft.
10. Tygon Tubing 3/4" I.D. x 0·6ins.
11. Marked levels at 19·5 & 17·0 ins.
12. 1/2" Screw clamps.
13. Test sample outlet

Fig. I SIMULATED ENVIRONMENTAL TANK

2. Comments and Modification of S.E.T. Test

The main advantages of the original and subsequently modified S.E.T. tests are its simplicity and its usefulness for relating results from various laboratories. The primary disadvantage has been the poor reproducibility of the experimental data. On the basis of work carried out by Environment Canada, several modifications have been made to the S.E.T. test and are incorporated in this effectiveness test[14]. The main changes are noted below:

a) More Useful Measure of Effectiveness

In previous S.E.T. test procedures, the dispersant/oil ratio is fixed and the percent dispersion is measured. In field operations, however, a fixed ratio is not used; rather an attempt is made to disperse all of the oil. Thus a more useful measure of effectiveness is the minimum dispersant/oil ratio needed to disperse a given percentage of oil. Here, this percentage is conservatively chosen to be 65% in view of the fact that the mixing powers available under field operations are greater than those used in the test.

b) Temperature Effect

The effect of temperature on performance effectiveness is not studied in the NAVY test. It is important to have some estimate of this effect. Accordingly, here the test procedure is extended to include the testing at two temperatures, $5 \pm 2^{o}C$ and $15 \pm 2^{o}C$. These temperatures refer to the water temperature at the beginning of each test.

Since the test will be kept simple, no temperature control is required and the temperature will change slightly as each experiment progresses.

c) Mixing of Dispersant with Oil

In the original S.E.T. test procedure, mixing of the dispersant and oil is accomplished by hosing through a 3/16" diameter nozzle. By this technique, much of the available energy is not utilized for mixing the oil and dispersant but rather is dissipated several inches beneath the water surface. In this effectiveness test, hosing is accomplished with the use of a standard garden spray nozzle. This greatly increases the surface mixing efficiency and the reproducibility of the results.

d) Other changes

Other changes have been made to the S.E.T. test such as a) painting the inside of the drum white for easier detection of oil droplets attached to the tank side, b) keeping the cylinder in the tank throughout the test and thus eliminating the problem of washing the cylinder when it is removed and increasing the effective hosing time, c) using a different type of spray bottle for applying the dispersant and d) modifying the filtering procedures for removing trace amounts of water from the chlorofom solutions.

Even with the aforementioned modifications, the following two limitations of the S.E.T. test should be noted:

a) Simulation of Mixing

Since the S.E.T. test was designed to measure the effectiveness of bilge cleaners, mixing is accomplished by water hosing under high pressure. During field operations, however, mixing of the oil slick and dispersant is best accomplished with the use of boat propellers or other high speed mechanical devices where the mixing power available is an order of magnitude greater. This available mixing energy is not considered in the S.E.T. test.

b) Pump effect

There is a high probability that the dispersed oil is further emulsified or broken up in the recirculating centrifugal pump. If this is the case, the simulation of field conditions is questionable.

3. Modified Simulated Environmental Tank Test

A brief outline of the modified S.E.T. test is given here. A more detailed description of the test equipment and procedures is available elsewhere(14).

(a) Materials for the S.E.T.

One 45-gallon steel drum, epoxy-painted white

Recirculating centrifugal pump, 400-450 gph at 2' head

Powerstat

½ inch I.D. PVC pipe, fittings, etc.

3/16 inch I.D. tygon tubing

150-ml polyethylene spray bottle with four 0.25 mm diameter holes drilled in lid.

25 ft. reinforced garden hose

garden spray valve (Aqua-Gun Melnor Ind.)

7½ inch diameter x 9 inch deep polyethylene cylinder (cut from 2 gallon polyethylene bottle) fitted with positioning device.

Clamps and miscellaneous parts

(b) Laboratory Equipment Required

Spectrophotometer and its accessories

100-ml volumetric flasks

500-ml separatory funnels

250-ml separatory funnel

glass funnel and filter paper

500 ml graduted cylinder

200 ml beaker

30 ml beaker

200 ml storage bottles with tightly sealed lids

1000 ml wash bottle

weight scale (200 ± .05 gm)

(c) Testing with a Reference Dispersant

In order to compare experimental procedures and results with other laboratories, it is necessary to perform experiments with a reference dispersant. Mix 2-ml of standard surfactant (IGEPAL CO 630, GAF Corp.) with 25-ml 2-methyl-2, 4-pentanediol (hexylene glycol). Then add this solution to 100 grams of test oil in a 250-ml separatory funnel and mix by agitating vigorously for 10 minutes. Then place on the surface of water and hose with water immediately. A sample should be collected after 10 minutes and analyzed as indicated below to obtain the dispersion value.

(d) Number of Tests

Experiments are to be performed with the dispersant and 4 test oils: * No. 2 fuel oil, crude oil and medium and heavy Bunker fuel oils. Each dispersant/oil combination shall be tested at 2 temperatures, 5°C and 15°C and at 3 ratios of dispersant to oil, 0.1, 0.30 and 1.0 by weight. The minimum dispersant/oil ratio needed to disperse 65% of the floating oil is obtained by interpolation techniques.

Stability of the dispersion is determined by analyzing samples withdrawn after 6 hours for the dispersant with No. 2 fuel oil at a dispersant/oil ratio of 0.30 and at an initial water temperature of 15 ±2°C. These two stability tests can be simple extensions of the "10-minute" experiments described above.

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* For specification of oils, see Section 3.2

(e) <u>Shortcuts</u>

For a given oil and temperature, if the dispersant cannot disperse 65% of the oil or if the dispersant/oil ratio needed is greater than unity the dispersant is considered to be ineffective under these conditions. If the tests are performed in a sequence moving from light to heavy oils, high to low temperatures and high to low ratios, considerable time and money can be saved. It is suggested that the set of experiments be started with No. 2 fuel oil at a dispersant/oil ratio of 1.0 and at a temperature of 15°C. If the dispersant cannot disperse 65% of the oil, it is considered ineffective at these conditions. It is also reasonable to assume that the dispersant is ineffective with the heavier oils and further tests are unnecessary. Similarly, if the dispersant is effective with No. 2 fuel oil at 5°C and 15°C, but not effective with the crude oil at 15°C, then tests with the crude oil at 5°C or with the Bunker fuel oils are not necessary. Because of the large variance associated with the S.E.T. test, it is necessary to perform at least 3 replications.

(f) <u>Test Procedures</u>

(1) Fill the 45-gallon drum to the 17-inch mark with water and bring the temperature to T°C (T = 5 ± 2 or 15 ± 2°C).

(2) Place the floating plastic cylinder in the centre of the tank so that it extends 4.5 inches below the surface of the water. The cylinder can be held in an upright position by means of clear acrylic strips fixed by nylon screws (14). Other techniques for positioning the cylinder, such as using flotation collars, laboratory clamps and strings are also satisfactory.

(3) With the recirculating pump off, carefully pour approximately 100 grams of the test oil into the centre of the "ring". The exact amount of oil should be determined by weighing the dispersant container before and after the addition of the oil.

(4) Allow 1 minute for the oil slick to stabilize and come to equilibrium with the water temperature.

(5) Add x grams of the dispersant (x = 10, 30, or 100) by spraying (using the polyethylene spray bottle) in a fine stream contacting as much oil as possible. The dispersant must be applied in less than 30 seconds. Weigh the bottle before and after the application of the dispersant to obtain the weight of the dispersant.

(6) Wait 10 minutes to allow the dispersant to contact the oil inside the ring.

(7) Agitate the oil-chemical mixture using tap water at 50 psi from a rubber or plastic hose, of $\frac{1}{2}$-inch inside diameter, fitted with a garden spray nozzle, fully opened. Agitation of the surface of the water in the tank should be continued until the volume in the tank has reached 19.5 inches. In some cases, the high foaming characteristics of some dispersants make it difficult to see the 19.5-inch mark. This problem can be overcome by timing the interval required to reach the 19.5-inch mark prior to the test and duplicating this interval in this stage of the test procedure.

(8) As soon as the hosing is completed, start the recirculation pump and continue throughout the test, at a rate of six gallons per minute.

(9) Ten minutes after the termination of hosing, withdraw a 500-ml sample to determine the amount of initial dispersion. Before taking the actual sample, the sampling line should be cleared first by collecting a few hundred mls. in a beaker. After taking the sample, return the collected oil-water mixture to the tank.

(10) Immediately after collection, transfer the 500-ml sample quantitatively to a separatory funnel and extract the dispersed oil with four 20-ml portions of chloroform and dilute to 100-ml with additional chloroform. Pour the chloroform solution over 2 grams of anhydrous sodium sulfate in a 200-ml glass storage bottle.

(11) Determine the concentration of oil at suitable wavelengths with a spectrophotometer. (A standard calibration curve should be prepared by plotting absorbence vs. oil concentration for the oils used by selecting a suitable wavelength.

(12) Before the calculations are performed, corrections must be made to the spectrophotometer readings. This provides for the sample blank of the dispersant and the natural detergent qualities of the test water. The sample blank for the chemical dispersant is determined by performing the above test with the dispersant in the absence of oil.

(g) Calculations

Calculate the percentage of oil dispersed as follows:

$$\% \text{ dispersion} = \frac{\frac{A}{B} \times C \times 100}{D}$$

where A = weight of oil in 500-ml sample (gm)

B = volume of sample (500-ml)

C = volume of water in S.E.T. (ml)

D = weight of oil used in the test (gm)

(h) Reporting

An Effectiveness Table (see Section 3.2 (b)) should be constructed. All data must be made available to Environment Canada.

APPENDIX II

BIOASSAY TEST

1. Introduction

The aim of the bioassay procedures is to establish a valid estimate of the acute lethal toxicity of dispersants and dispersant/oil mixtures. The test does not necessarily attempt to imitate the exact situation which might prevail in nature, but involves standard procedures and conditions which are easily reproducible.

These standard conditions involve the use of a standard species, a standard test water and temperature, and a specified test procedure. It is realized that it may be difficult or impossible in some instances to adhere to these conditions. Alternative non-standard conditions may be used in such circumstances. However, in case of uncertainty or dispute about the validity of the results, information based on the standard test must be used.

There are three materials to be tested: (1) the dispersant alone; (2) oil + the dispersant; and (3) a reference toxicant. For each of these, the median lethal concentration must be determined after four-day exposures. This value is merely a convenient reference point for expressing the acute lethal toxicity of a material to an agreed standard fish. Obviously, the "safe" concentration which would cause no sublethal effects, would be lower than the median lethal concentration, often much lower.

These bioassays are conducted on rainbow trout. Static tests are recommended instead of the more sophisticated continuous-flow tests, since they seem more realistic and probably more accurate for a material as difficult to handle as oil.

The methods below are derived in large part from five sources. The first is the standard bioassay method of the American Public Health Association and others[1], which is derived from a paper by Doudoroff and co-authors[9]. The second source is a series of review articles on bioassays by Sprague[25].

The third source is a proposed method for bioassay of dispersants and oil, by LaRoche *et al.*[15], while the fourth and fifth are publications by the Ontario, Ministry of the Environment[21] and the California State Water Resources Control Board[7].

2. Materials

(a) Oil and Dispersants

The oil to be used in the bioassay tests is No. 2 fuel oil (as specified by ASTM D-396-67 and with an aromatic content of 15% by volume minimum). The oil should be stored and transported in sealed small containers (about 100 to 200 ml) to prevent loss of volatiles. A fresh container should be used for each new set of tests. Dispersants should be kept sealed when in storage.

(b) Reference Toxicant

As described by LaRoche *et al.*[15], "the use of a reference toxicant is desirable as a link between the findings of different investigators, as an internal standard to compare the relative toxicities of substances, and as a measure of condition of test organisms...". Reagent grade dodecyl sodium sulphate (DSS, also known as sodium lauryl sulphate) is rapid, nonselective, and consistent in its toxicity to aquatic animals. Accordingly, the median lethal concentration of DSS is to be determined each time the LC50 is determined for a dispersant. Concentrations should be expressed as mg of DSS per litre of testwater. The median lethal concentration may be expected to be in the general range of 1 to 10 mg/l.

(c) Test Species

Rainbow trout (Salmo gairdneri Richardson) is specified here as the standard species for testing dispersants and oil. This species should always be used unless circumstances make it impossible or undesirable to do so. If a non-standard species is selected, it should preferably be a sensitive sport or commercial fish which is locally important. Unusually tolerant species should be avoided. The species should be adaptable to healthy life in the laboratory for some weeks, and readily available from a common source with similar past history. The young of any Canadian species of the family Salmonidae (salmon, trout, and char) would be best. There could also be special reasons for using an invertebrate species in the tests. This could be done, with the same limitations as described above for non-standard fish. The rationale behind these requirements is chiefly that use of a standard species allows meaningful comparison of results from different laboratories and for different dispersants. Rainbow trout is a desirable species to use since it seems fairly typical of most sensitive cool-water fish in its resistance to pollutants, and is widespread in Canada. Indeed it is now common throughout much of the world and is becoming widely used as a standard species in bioassays. Another non-standard species is the fathead minnow (Pimephales promelas Rafinesque) which also has a wide distribution in Canada, extending well up into the Northwest Territories.

(d) Size of Fish

Fish should be small, preferably less than 5 grams or 8 cm. The length of the largest fish in a set of tests must not be more than 1.5 times that of the smallest. It is permissible to use larger fish if desired, but the required amounts of test-water may become so large as to be impractical (see below). For this reason, it has sometimes been recommended (e.g. LaRoche *et al.*[15]) that fish should be smaller than 1.5 gm. For a given species, individuals should be of the same age and size range and should be obtained from the same source, for any single series of tests.

(e) Holding of Test Fish

The fish should be held and acclimated in tanks and accessories which are made of non-toxic material. Often used today are cylindrical fibreglass tanks with large bottom areas, and a central drain and standpipe taking water and debris from the bottom. Tanks should be exposed to an ordinary sequence of darkness and light, either from natural daylight or room lighting. Flowing water is desirable to carry away metabolic wastes, preferably at least one litre per minute for every kilogram of fish being held. Vigorous aeration with oil-free compressed air should be provided. The tank should contain one litre of water or more for every 10 grams of fish, and in addition the volume of water in the tank should ideally equal the total flow during two or three hours, which would mean 90% replacement of water in 4.5 or 7 hours (Sprague, 1969). Fish can be kept with less water than described above, but with increased risk of disease and stress problems. At least two weeks must be allowed for acclimation of fish to the dilution water and test temperature. Abrupt changes in quality of holding water must be avoided, especially extreme temperature changes. Fish should not be handled unnecessarily.

They should be transferred carefully, quickly, and as gently as possible to avoid placing the fish under unnecessary stress. Feeding during holding is important. The most desirable food is usually a specially designed and balanced fish food, for example, in the form of moist frozen pellets. Other kinds may be available from feed suppliers. A recipe for preparing food is given by LaRoche et al. and beef liver might be substituted for the clams which they specify. Daily or twice-daily feeding to immediate repletion during the 5-day work week is usually satisfactory, but very small fish may require several feedings of small quantities of food every day. Fish must not be fed for 48 hours prior to, or during bioassays. Disease should be virtually absent, and mortality should be negligible, if a stock of fish is to be considered suitable for bioassays. Weak fish will of course prejudice the assay so that the test-material will appear to be more dangerous. Only those fish which feed actively and appear to be healthy should be used in the bioassays and any individual injured or dropped while handling must be discarded. Do not use fish which show a group mortality greater than 5% during the 4-day period preceding the bioassay.

(f) Dilution Water

Any natural supply of clean, non-toxic water may be used for holding the fish. The best supply is usually water from a well, which is then aerated, possibly filtered, but not chlorinated. Surface water which is known to be clean may also be used. "Tap-water" is often poisonous to fish, and in particular, small traces of chlorine such as remain after carbon filtration, may cause mortality after long exposure. Copper or galvanized plumbing, brass fittings, or the plasticisers from molded fittings, may also cause toxicity and must be completely avoided.

The standard test-water is specified here to have a total hardness of 50 mg/l as $CaCO_3$ and a normal balance of dissolved salts . This may be obtained by diluting well water with distilled water. This standard water is prescribed since there may be effects of hardness on the toxicity of some dispersants. However, if circumstances make it impossible to make up such water, any natural water may be used as described above. Another alternative is to prepare synthetic dilution water. Suitable formulae for both hard and soft freshwaters have been published[6,21].

(g) Physico-chemical Conditions

The temperature should be 15°C (59°F) for holding and testing. Fluctuation of 2 degrees Celcius above or below this average is not critical during holding, and a maximum fluctuation of ± one degree from this average is acceptable during testing. The holding tanks should be aerated vigorously, and the dissolved oxygen should not drop as far as 1 mg/l below saturation. Test tanks must not be aerated. In fresh water, it is desirable that the dissolved oxygen in the test water should not drop below 8 mg/l, and it need not do so if a satisfactory volume of test water is used. If the oxygen drops below 6.5 mg/l (a "moderate" level of protection according to Doudoroff and Shumway, 1970), the test should be rejected and repeated with a greater volume of test water per gram of fish. The pH of the dilution water should normally be the stable one which prevails when the water is well aerated. The pH in fresh water tests should not vary more than 0.2 units, except as caused by the test-material. If it does cause undue change in pH, this must not be adjusted, but accepted as part of its toxicity. Total hardness must be measured in the dilution water at the start of each series of tests. In each tank, the temperature must be measured daily, and the pH and dissolved oxygen must be measured at least at the beginning and end of the test, and preferably daily.

3. Method for Setting up Bioassays

For each dispersant which is to be evaluated, there are three major sets of bioassays to be run, each with a different material;

(1) the reference toxicant DSS;
(2) the dispersant alone;
(3) the dispersant with No. 2 fuel oil.

In addition, a less complex bioassay of degradability must be performed.

The objective in each of the three sets of bioassays is to determine the lethal concentration for 50% of the individuals during a 4-day exposure, signified by the term 96-hour LC50. The abbreviation, used in many fields of biological testing, is numerically equal to the notations TL50 and TLm, sometimes used in work with fish.

(a) Exploratory Tests

Small-scale exploratory tests may be performed before the full-scale tests, to determine the general range of concentrations which should be tested. These preliminary tests could be very primitive, such as one or two fish in a large beaker of test water, observed over a day or two. The important thing would be to use a wide range of concentrations over intervals of an order of magnitude, such as 1, 10, 100, and 1000 mg/1.

(b) General Conditions

The usual principles of good scientific investigation apply. For example, a control test containing only dilution water must be run for each series. All the test-tanks including the control must have identical conditions, except for the concentration of the test-material. The selected concentrations should be formally randomized among the available test tanks, and the fish should be randomized among the tanks also. If more than one fish dies in the control test, this suggests unhealthy fish that are likely to be overly sensitive to wastes, residual toxicity from the container or lining, or toxicity from the dilution water itself. Any mortality in the control should be regarded as unsatisfactory, and anything greater than 20% mortality means that the series of tests must be rejected. The cause of the problem should be ascertained and eliminated before a new series is started.

(c) Number of Fish

It is desirable to have about 10 fish at each concentration. If the fish are large, it may be necessary to split them between two or more tanks at the same concentration, to get the recommended volumes of water per gram of fish. If that entails difficulties, it may be satisfactory to reduce the number of fish in each concentration to as low as five, but that is the absolute minimum. The confidence limits on the final LC50 will become wider as precision is reduced with fewer fish, and such imprecision could lead to rejection of the results by Environment Canada.

(d) Dispersant/Oil Ratios and Concentrations

The ratio of dispersant to No. 2 fuel oil should be unity. This high ratio is used to ensure that the oil is entirely dispersed in the test tank, and is not floating on the water surface during the bioassay. The concentration of the dispersant/oil mixture must be expressed as total mg. of oil plus dispersant per litre of testwater.

(e) Concentrations to be Tested

Several concentrations of each material should be tested, and should be chosen from a geometric or logarithmic series. The series in "Standard Methods" (APHA *et al*. (1)) is suitable. The first full test following the exploratory ones should be selected from adjacent members of the series 0.1, 0.32, 1.0, 3.2, 10, etc. After initial observations it might be desirable to start another test one step higher or lower in the series. Sometimes it might be desirable to get a finer gradation by following up with tests at intermediate concentrations in the same series: 0.1, 0.18, 0.32, 0.56, 1.0, 1.8, etc.

The final results from the bioassay must meet the following criterion:

For calculating the final value of LC50 at least three partial responses between 30 and 70% mortality inclusive must be used.

(f) Preparation of Test-Mixtures

For the reference toxicant, dodecyl sodium sulphate (DSS), the required weight of material may simply be dissolved in a beaker full of dilution water taken from the tank to be used, this may be poured into the tank, mixed well, and the fish added. For the dispersant and the dispersant/oil mixture, the following procedure is required. One litre of dilution water is taken from the appropriate tank. The required amount of oil (if any) and the required amount of dispersant should be weighed and added to the water. This should be homogenized in a high-speed laboratory blender. The mixture should be then added to the water in the test-tank and dispersed by hand-stirring. The laboratory stirrer in the test-tank should then be adjusted as described in the following sections. It is essential that the oil/dispersant mixture stay in emulsion during the bioassay. It will be necessary to determine, by trial-and-error, the required blending speed and time, and the stirring speed in the test-tank to accomplish this. After it is assured that negligible surfacing of oil is taking place (a 30-minute waiting period should be observed) the fish may be added to the tank.

(g) Test-Tanks and Volume of Testwater

The fish must be allowed reasonable scope to swim around freely in whatever containers are used. The best shape for tanks is oval or cylindrical. For these tests with dispersants and oils, the tanks must be freshly lined for each test with polyethylene or an equivalent liner. The testwater should be stirred during the experiment to maintain a homogeneous emulsion. A laboratory stirrer with a propeller blade should be employed, operated at a suitable r.p.m. and depth to ensure circulation with no vortexing.

The shaft and the blade should be surrounded by a shield of coarse screening to protect the fish. Alternatively, the fish could be contained in a rigid-frame cage, large enough to occupy most of the test-tank with the exception of the stirrer. The cage should be covered with coarse netting such as nylon or other non-toxic, non-absorbent material. Keeping the fish in a cage is almost obligatory if the testwater is dark-couloured or opaque, since the cage could be lifted to bring fish near the surface for observation of mortality, without undue disturbance. There must be at least 2 litres, and preferably 3 litres, of testwater per gram of fish, per day. For a 4-day test this means at least 8 litres per gm of fish, and preferably 12 litres. This should ensure that vithout aeration, the oxygen concentration would not be lowered more than 2 mg/l. Very small fish, for example of guppy size, would require several times as much testwater since their respiration rate would be higher per unit of biomass. The recommended volume of testwater should also prevent serious accumulations of metabolic wastes, and depletion of the toxicant by the fish.

4. Observations on Mortality

Although only the data from 96 hours are required for the formal statement of toxicity, daily inspections must be made for removal of dead fish and to check conditions in the test-tank. Beyond this, more frequent observations in the earlier part of the test yield a great deal of extra information, are usually beneficial to the investigator, and are therefore recommended.

The observation times should approximate a logarithmic series, and a convenient set might be as follows: 1, 2, 4, 8±1, 14±2, 24, 33±3, 48, 72, and 96 hours. If a test were started in mid-morning, most of these observations could be made during normal day-time working hours, the single exception being an observation late in the first evening.

Fish should be considered dead when there is no respiratory or other overt movement, and no response to gentle prodding. Dead fish should be removed as soon as observed, and wet weight recorded. The test should be terminated at 96 hours and the LC50 must be calculated.

5. Analysis of Results

(a) Plotting Results to Estimate LC50

The mortality observed in each test-tank at 96 hours is plotted against the concentration in the same tank. Concentration is on a logarithmic scale, and percent mortality is on a probability or "probit" scale (Fig. 2). Such graph paper can be purchased from standard sources. Since the probit scale never reaches zero or 100 percent mortality, any such points must be plotted with an arrow indicating their true position. A line is next fitted to the points by eye. Most consideration should be given to points between 30% and 70% mortality, and an effort made to minimize total vertical deviations of the line from the points. Only one successive zero percent and one successive 100 percent should be utilized, these to be the ones nearest the centre of the range of concentrations used. If there is doubt about the placement of the line, it should be rotated to a flatter slope since this acknowledges more variability in the data. The concentration causing 50%

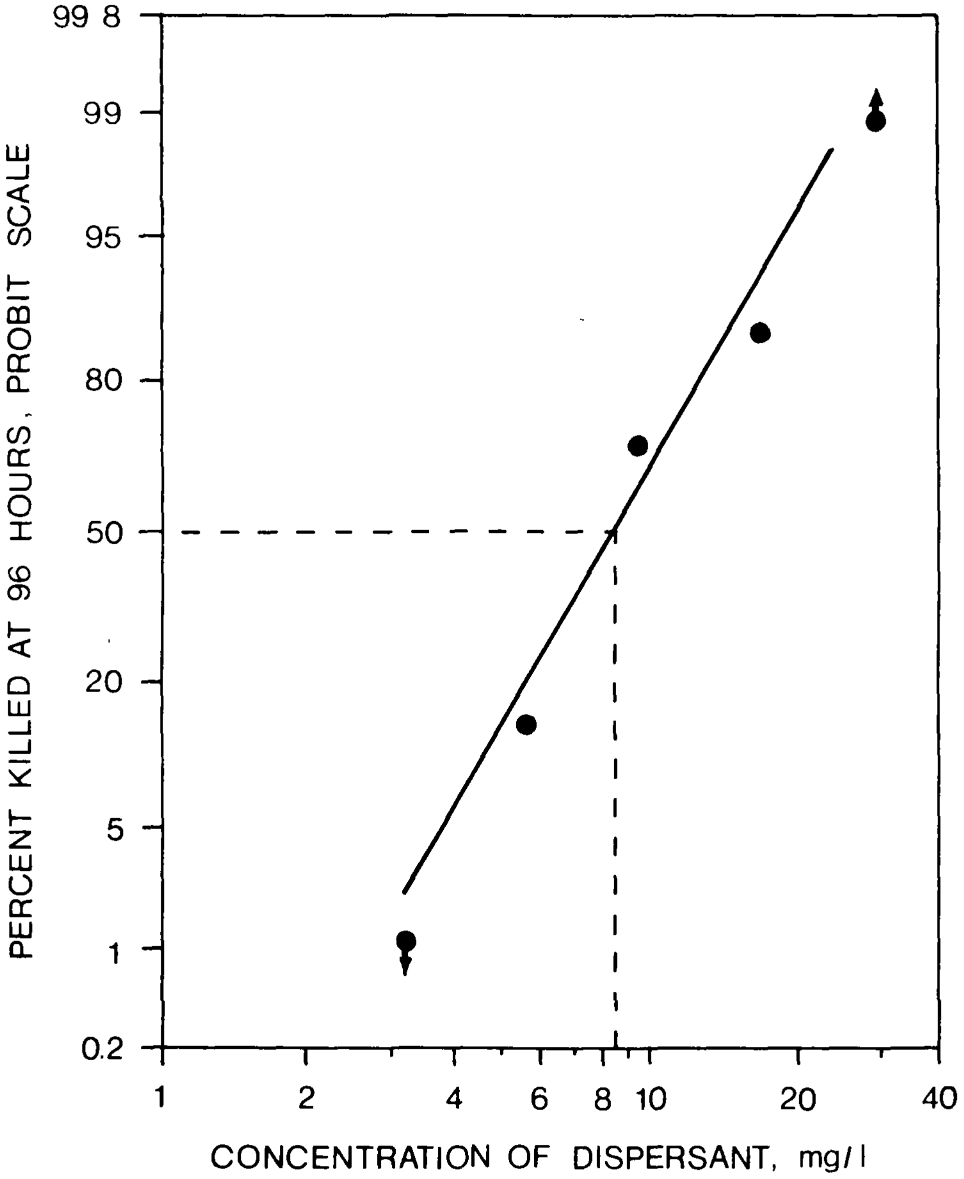

Fig. 2. Plotting results and estimating the LC50 for a hypothetical dispersant which is very toxic. The concentration lethal to half the fish is read from the eye-fitted line and is between 8 and 9 mg/1. A more common type of log-probit paper has a probit scale running from 2% to 98% mortality and is satisfactory.

mortality is read from the fitted line, and this is the provisional estimate of the 96-hour LC50.

(b) Confidence Limits of LC50

The confidence limits must be estimated and reported. The common methods of fitting lines and making such estimates are not valid, and special techniques are needed. One way to do this is by the simple nomographic techniques of Litchfield and Wilcoxon[16] which require about 10 to 15 minutes after some experience. Confidence limits of the LC50 may be estimated by the Litchfield-Wilcoxon method. These limits should be reported along with the LC50 and the "slope function" or "S".

Computer methods may also be used for probit analysis. Any suitable program based on the method of Finney[11] may be used. An example of such a program is BMDO3S in the BMD package (Dixon,[8]). Another example, in which the data are simply typed in at a terminal, is the program called PROBITANAL, available from the Institute of Computer Science at the University of Guelph. These programs compute the LC50, its confidence limits, and the slope of the probit line, which should all be reported. In addition, a graph like Fig. 2 must always be drawn, and inspected for reasonableness of the computed LC50. In some cases of unusual probit lines, the investigator's judgement will carry more weight than the automatically-computed LC50. Alternate methods of estimating the LC50 and its confidence limits are available and have been reviewed (Sprague, 1969).

(c) Optional Extra Analysis -- Constructing a Toxicity Curve

If the optional, more frequent observations of mortality have been made, the investigator will get earlier insight into his results if he constructs a toxicity curve similar to that in Fig. 3, as the tests proceed. To do this, it is merely necessary to make a graphic estimate of the LC50 (as in Fig. 2) for each observation time, such as 8 hours, for example. The LC50's at successive time intervals are plotted on logarithmic paper as in Fig. 3, as the exposures proceed. Including a copy of the toxicity curves with the report of results is optional but highly desirable for maximum comprehension of the results.

6. Degradability Test Procedure

The dispersant must be biodegradable as determined by performing a Product Degradation by chemical analysis over a 20-day period using the shake flask or similar technique[17].

Furthermore, the biodegradation products must not be more toxic than the dispersant itself. In order to assess this, a bioassay is to be performed on an aged sample. A concentration of dispersant which is equal to the 96-hour LC50 is made up in the standard way and held in a bioassay tank with stirrer. The testwater should be seeded with hydrocarbon-degrading bacteria, taken from such sources as oil refinery wastewater or slag, activated sludge or oil-soaked soil. If this is not possible, the testwater may be seeded with 50-ml samples of surface water from 2 or 3 local sources. A control test with no dispersant is started in the same way.

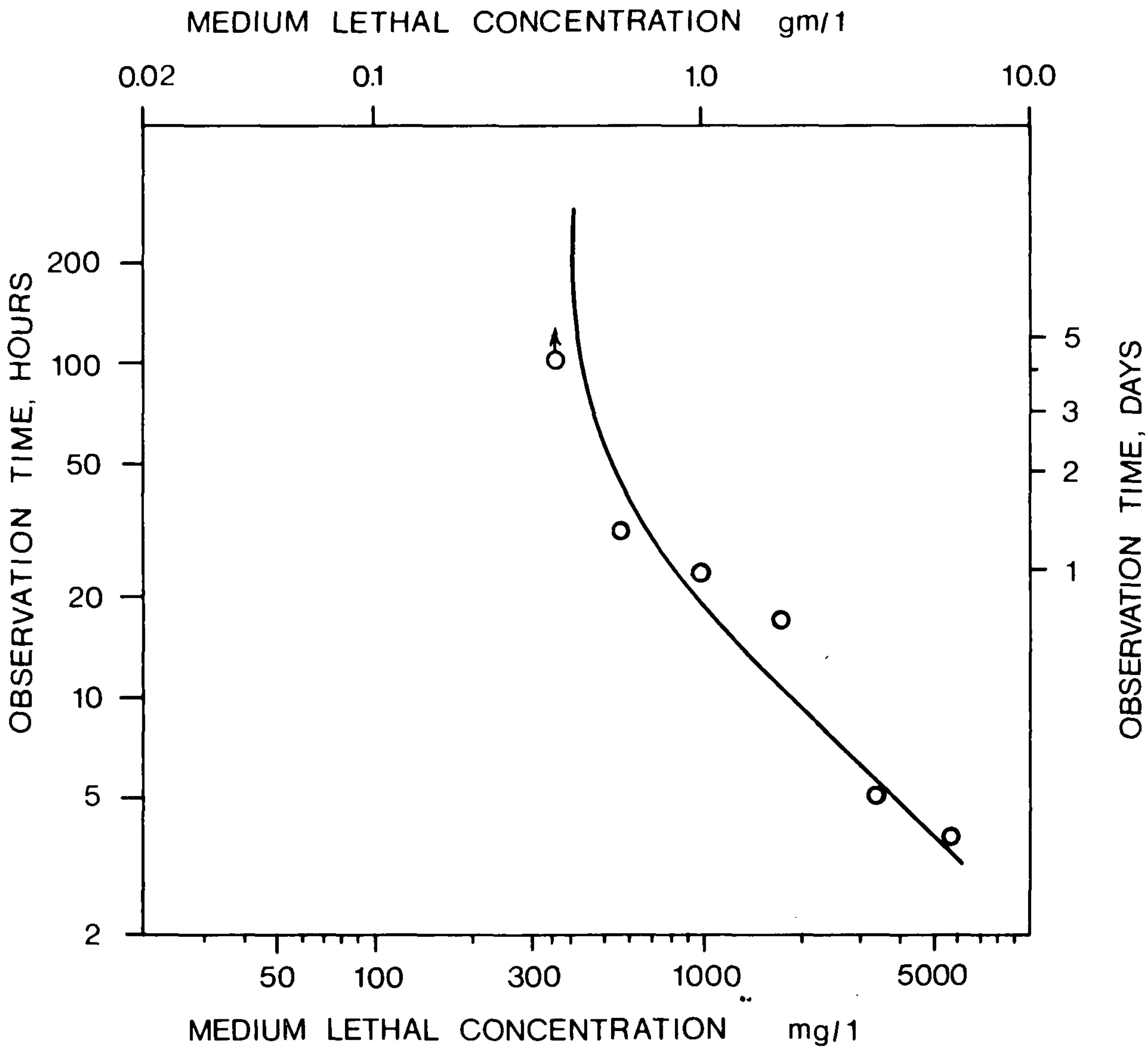

Fig. 3. Example of a toxicity curve for a hypothetical dispersant which is only moderately toxic.

At the end of the sixteenth day of holding, fish are introduced into the test tanks, and observed during the following four days. If mortality in the aged dispersant is no greater than in the control (presumably zero), then the dispersant has excellent degradation characteristics. If the aged dispersant kills more than the control test, but less than 50% of the test fish, it has satisfactory degradation characteristics. If more than half of the fish die in the aged dispersant, the degradation is not satisfactory, and the degree of degradation may be quantified by estimating the time to reach 50% mortality, and comparing it with the toxicity curve for fresh dispersant, if such a curve was made.

7. Criteria

The 96-hour LC50 for fresh dispersant using rainbow trout in standard water should not be less than 1000 mg/l. The 96-hour LC50 for the total amount of dispersant plus oil should not be less than 100 mg/l. Dispersant which has been aged for 16 days, with original concentration equal to the 96-hour LC50, should not be lethal to more than half the test-fish in a four-day exposure.

8. Reporting

(a) Fish

Species and source
Age and approximate acclimation history
Date obtained and dates of tests
Acclimation history in brief since fish were obtained
Feeding
Average of wet weights of all fish used in tests, and variability (range or standard deviation).

(b) Water

Source
Special treatment before use
Total hardness and pH
Formula used if synthetic water
Average and variability of daily or other scheduled measurements in test tanks and control tanks, for temperature, pH, and dissolved oxygen. (Separate any tanks with unusual readings).

Usual amount of testwater used in each tank.

(c) Equipment

Blender: describe type, blending speed, blending time

Test-Tank: describe arrangement including stirrer dimensions and stirring speed used.

(d) Test Results

Report the 96-hour LC50, its 95% confidence limits the slope of the probit line, and mortality in controls, for:

the reference toxicant, DSS;
the dispersant being tested; and
the dispersant with oil.

The toxicity curve may also be provided for each, if available.

In the Degradability Test, report (1) the details of the product degradation analysis; (2) the number of test fish and mortality in the aged dispersant and in the control; (3) details of the seeding technique; (4) time to reach 50% mortality if more than half died in the dispersant.

APPENDIX III

CONTACT LIST

Environmental Protection Service Contact List

Atlantic Region/Halifax

Regional Director	Dr. C.J. Edmonds P.O. Box 2406 Halifax, N.S.	Office: Home:	(902) 426-3593 426-2308 (902) 454-0444
Environmental Emergency Co-ordinator	Mr. H.T. Doane P.O. Box 2406 Halifax, N.S.	Office: Home:	(902) 426-6121 (902) 454-1974

Quebec Region/Montreal

Regional Director	Mr. G. Gauthier P.O. Box 1330 Station B Montreal 110, P.Q.	Office: Home:	(514) 283-7377 (514) 482-4299
Environmental Emergency Co-ordinator	Mr. Y. Plunier P.O. Box 1330 Station B Montreal 110, P.Q.	Emergency: Office: Home:	(514) 283-2333 (514) 283-2345 (514) 655-0563

Ontario Region/Burlington

Environmental Emergency Co-ordinator	Mr. N. Vanderkooy P.O. Box 5050 867 Lakeshore Road Burlington, Ontario L7R 4A6	Office: Home:	(416) 637-4705 637-4623 (416) 459-8610

Northwest Region/Edmonton

Regional Director	Mr. J.J. Eatock 10th Floor Imperial Oil Bldg. 10025 Jasper Ave. Edmonton, Alberta T5J 2X9	Office: Home:	(403) 425-4580 (403) 467-0452
Environmental Emergency Co-ordinator	Mr. R.K. Pettigrew Imperial Oil Bldg. 10025 Jasper Ave. Edmonton, Alberta T5J 2X9	Office: Home:	(403) 425-5128 425-5129 (403) 487-1529

Pacific Region/Vancouver

Regional Director	Mr. R.E. McLaren 1090 West Pender St. Vancouver, B.C.	Office: Home:	(604) 666-1064 (604) 922-5808
Environmental Emergency Co-ordinator	Mr. C.T. Hatfield 1090 West Pender St. Vancouver, B.C.	Office: Home:	(604) 666-1370 (604) 929-2608

National Headquarters/Ottawa

Manager, National Environmental Emergency Centre	Cdr. R.A. Beach 15th Floor Place Vincent Massey Ottawa, Ontario K1A OH3	Office: Home:	(819) 997-3742 (613) 224-1259
Assistant Manager, National Environmental Emergency Centre	Mr. C.S.L. McNeil 15th Floor Place Vincent Massey Ottawa, Ontario K1A OH3	Office: Home:	(819) 997-3878 (613) 836-3977