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Environmental Protection
Agency

Office of the Administrator
Science Advisory Board
Washington, D. C. 20460

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June 1989



Report of the Environmental Effects, Transport and Fate Committee

Review of the Alaskan Oil Spill Bioremediation Project



UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D. C. 20460

OFFICE OF
THE ADMINISTRATOR

June 16, 1989

The Honorable William K. Reilly
Administrator
U.S. Environmental Protection Agency
401 M. Street, S.W.
Washington, D.C. 20460

Dear Mr. Reilly:

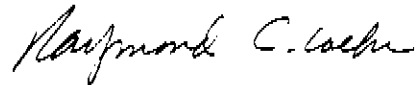
The Environmental Effects, Transport and Fate Committee of the Science Advisory Board has completed its review of the Office of Research and Development's (ORD's) "Research Plan for the Alaskan Oil Spill Bioremediation Project". The Committee congratulates ORD on its rapid response to this opportunity to field test bioremediation approaches.

This project, designed to provide data to demonstrate the potential use of bioremediation both as an emergency response tool for Prince William Sound and for future environmental remediation efforts, was evaluated for scientific and technical accuracy by the Committee and invited experts. The Committee supports ORD's effort to enhance bioremediation using addition of nutrients, but recommends that parallel efforts to augment bioremediation via inoculation with microorganisms undergo further laboratory investigation prior to field release. Additional recommendations included consultation with experts in field plot design to make sure that enhancement of biodegradation rates will be detected by the experiment, and a simplification of the battery of environmental effects measurements through association with an underlying rationale. A detailed presentation of these and other recommendations is provided in the attached report.

The Committee hopes that ORD will proceed with the nutrient enhancement experiment. They encourage EPA to go further to develop an active demonstration and implementation program based on bioremediation so that the research necessary for addressing such questions, and the technology for remediation will be defined before future spills occur.

The Board appreciates the opportunity to provide advice on this important issue and looks forward to receiving a response to the advice. In addition, we would appreciate receiving (for information) reports that result from the conduct of the research project.

Sincerely,



Dr. Raymond Loehr, Chairman
Science Advisory Board

Enclosure

cc: Dr. Donald Barnes
Dr. Erich Bretthauer
Dr. John Skinner
Dr. Hap Pritchard



UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

May 16, 1989

Dr. Raymond Loehr
Chairman, Science Advisory Board
c/o U.S. EPA
401 M Street, S.W.
Washington, D.C. 20460

OFFICE OF
THE ADMINISTRATOR

Dear Dr. Loehr:

We are pleased to transmit via this letter the advice of the Science Advisory Board's Environmental Effects, Transport and Fate Committee concerning the EPA's Alaskan Oil Spill Bioremediation Project.

This project was reviewed by the Committee and invited participants on May 15 and 16, 1989. The Committee evaluated the scientific adequacy of the project in light of its goal: determining if techniques for accelerating the hydrocarbon biodegradation rates of natural microbial communities can be used to help in the clean-up of the oil-contaminated Prince William Sound. The study is designed to provide data to support the use of bioremediation as part of the emergency response activities currently taking place at Prince William Sound, and will also allow for the effective use of biological treatment techniques for future environmental remediation. A detailed presentation of our views is contained in the attached report.

We appreciate the opportunity to provide advice on this important issue. The Committee would appreciate being involved in and informed of future SAB activities related to the long-term bioremediation of Prince William Sound.

Sincerely,

A handwritten signature in cursive script, appearing to read "Martin Alexander".

Dr. Martin Alexander
Chairman, Alaskan Bioremediation Protocol Review
Science Advisory Board

A handwritten signature in cursive script, appearing to read "Kenneth L. Dickson".

Dr. Kenneth Dickson
Chairman, Environmental Effects, Transport and Fate Committee
Science Advisory Board

CC: Dr. Erich Brettauer
Dr. John Skinner
Dr. Hap Pritchard

ABSTRACT

This report presents the conclusions and recommendations of the U.S. Environmental Protection Agency's Science Advisory Board summarizing a review of EPA's "Laboratory Plan for the Alaskan Oil Spill Bioremediation Project". This project was designed to provide data to demonstrate the potential use of bioremediation both as an emergency response tool for Prince William Sound, and for future environmental remediation efforts. The Board supports ORD's effort to enhance bioremediation using addition of nutrients, but recommends that parallel efforts to augment bioremediation via inoculation with microorganisms undergo further laboratory investigation prior to field release. Additional recommendations included consultation with experts in field plot design to make sure that enhancement of biodegradation rates will be detected by the experiment, and a simplification of the battery of environmental effects measurements through association with an underlying rationale.

Key Words: Prince William Sound; bioremediation; nutrient enhancement.

U.S. ENVIRONMENTAL PROTECTION AGENCY

NOTICE

This report has been written as a part of the activities of the Science Advisory Board, a public advisory group providing extramural scientific information and advice to the Administrator and other officials of the Environmental Protection Agency. The Board is structured to provide a balanced expert assessment of scientific matters related to problems facing the Agency. This report has not been reviewed for approval by the Agency; and hence, the contents of this report do not necessarily represent the views and policies of the Environmental Protection Agency or other agencies in Federal government. Mention of trade names or commercial produces does not constitute a recommendation for use.

U.S. ENVIRONMENTAL PROTECTION AGENCY
SCIENCE ADVISORY BOARD
ENVIRONMENTAL EFFECTS, TRANSPORT AND FATE COMMITTEE
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2.0 INTRODUCTION

2.1 Request for Science Advisory Board Review

President Bush asked the EPA Administrator to initiate research activities related to the recent Alaska oil spill. The Office of Research and Development (ORD) asked for Science Advisory Board (SAB) assistance with oversight of some of these activities.

The specific activity described herein consists of a review of a document developed by ORD entitled "Research Plan for the Alaskan Oil Spill Bioremediation Project". The objective of this project is to demonstrate the feasibility of accelerating the rate of biodegradation of oil spill residues on the shorelines of Prince William Sound, Alaska.

Due to the need to move rapidly, it was not possible to bring this request for review to the SAB's Executive Committee for approval. Instead, Dr. Raymond Loehr, Chairman of the SAB's Executive Committee met with other members of the Board, representatives of ORD, and SAB staff to consider the request and subsequently have the SAB accept the charge. The short-term effort on experimental bioremediation was discussed along with a longer-term effort on ecological recovery of Prince William Sound.

The SAB Staff Director and Dr. Loehr asked the Environmental Effects, Transport and Fate Committee (EET&FC), Chaired by Dr. Kenneth Dickson, to perform the review. The Committee agreed to review the bioremediation protocol with augmentation by experts as needed.

2.2 Charge

The Committee was charged with evaluating the scientific and technical adequacy of the protocol. Specifically, the Committee was asked to consider four questions: a) will this plan allow EPA to determine whether accelerating the rate of natural biodegradation is feasible, b) are the proposed assessments of ecological effects adequate, c) will information necessary to make decisions about utility of scale-up be generated by the plan, d) is the decision to exclude commercial, non-indigenous organisms from the protocol appropriate.

2.3 Committee Review Procedures

The Committee met on May 15 and 16, 1989, in St. Louis, Missouri. The document for review was provided to SAB members prior to the review. The proposed plan is attached as Appendix B. Briefings were provided to the Committee at the meeting by ORD staff. General and conceptual comments are provided in this report, while specific comments addressing the protocol are addressed in Appendix A.

Prior to the SAB Committee's review on May 15-16, 1989, ORD convened a separate Scientific Steering Committee independent of the SAB to provide information and guidance to the development of the bioremediation protocol. A workshop was held on April 17 and 18, 1989, to explore the feasibility of bioremediation, to develop a strategy for a small-scale demonstration, to prepare draft monitoring and assessment guidelines, to explore the ecological consequences of such a project, and to discuss long-term aspects that can be related to remediating future spills. Several follow-up meetings were held, site visits took place and information was gathered. The Scientific Steering Committee provided considerable guidance to the Agency in developing the protocols. A representative from this Steering Committee, Dr. Ronald Atlas, was present at the May meeting to provide information to the SAB Committee.

3.0 MAJOR CONCLUSIONS AND RECOMMENDATIONS

3.1 ORD's Rapid Response

The Committee congratulates ORD on its rapid response in generating a research plan to study the potential for enhancing biodegradation of spilled oil. ORD's action in convening appropriate EPA and non-EPA scientists via the Steering Committee mechanism is considered to be a useful approach. ORD has been presented with a unique opportunity to conduct important research. Bioremediation is recognized to show promise for emergency response and remediation, and a field test of the sort described will further clarify the utility of this approach. By developing the protocol under review, ORD has responded to the opportunity to conduct research that may accelerate the recovery of Prince William Sound and demonstrate the utility of bioremediation as a tool for future emergency response. ORD is also commended for seeking early input from the SAB, so that full benefit from guidance and oversight can be incorporated into the planning stages.

3.2 Support for the Concept

The Committee supports the conduct of the proposed bioremediation program. This support is, however, tempered by the qualifications presented herein. The program should be implemented not only for its potential value per se but also because it can serve as a case history, whether successful or not, for future actions. The program will provide a basis for considering bioremediation as a means for emergency response, as part of planning for clean-up efforts and for remediating inadvertent discharges.

3.3 Benefit Analysis

The Agency should conduct a preliminary analysis to document the possible benefit of the proposed research under best or worst case scenarios to establish a realistic idea of what can be accomplished. Nutrient or microorganism addition may have some impact on destroying petroleum fractions in the affected areas, but it is important to anticipate the potential rate enhancement that can be expected and to anticipate the effects of these enhanced rates of degradation on the ecosystem where damage has already been done. Preliminary calculations should be made based on information describing the site and known hydrocarbon degradation rates to establish the feasibility of this project. The minimal concentrations of added nitrogen and phosphorus that are known to enhance microbial degradation should be considered to set realistic bounds on what can be expected in such a bioremediation activity.

3.4 Consideration of Other Treatment Methods

The Committee suggested that other forms of treatment, such as addition of surfactants or emulsifying agents to enhance the availability of oil to microbes, be considered. In addition, the variety of means for fertilizer applications and for slow release of nitrogen and phosphorus from fertilizers should be further considered.

3.5 Measurability of Treatment Effects

The research protocol for bioremediation contains little information on plot design and experimental layout to statistically determine if fertilization or inoculation enhances biodegradation of oil on the beaches in Prince William Sound. The Committee is concerned that an adequate number of replicates of environmental samples will not be taken to allow for detection of differences between experimental treatment sites and reference sites.

The research protocol is based on the premise of detecting a 5% enhancement of biodegradation on fertilized beaches as compared to unfertilized beaches. In light of the variability that probably exists in the distribution of oil on the beaches, the heterogeneity in the beach composition, and the analytical variability, detection of this small degree of enhancement by fertilization will require a large number of replicate samples.

The Committee most strongly recommends that the project team immediately consult with a statistician who is fully versed in experimental layout and design for field testing. An estimate must immediately be made of the replication needed to detect a realistic difference between treated beaches and reference beaches. If the natural variability is as high as the Committee suspects, then a large number of replicate samples will be required. If this proves to be the case, it is recommended that the number of parameters to be measured be decreased and that sufficient replicates of the most important parameters be made to allow detection of reasonable differences between treatments.

Reference sites, as well as treatment sites, need to be adequately replicated. ORD may want to consider collecting samples from an additional test site that is not oil-contaminated but received nutrient addition. Inclusion of such a site will allow ORD to distinguish the effects of treatment alone and may better characterize the impacts that may result from nutrient addition itself.

3.5.1 Nutrient Loading

Natural nutrient loadings must be assessed to ensure that the effects of added nutrients are not confused or masked by natural conditions. In addition upwelling during the summer may create high natural levels of nitrogen and phosphorus, confounding the effects of fertilizer addition.

3.5.2 Hydrodynamics

The research plan lacks a consideration of expected mixing rates, likely dilution rates, etc., during the experimental period. The Committee strongly recommends that this readily correctable flaw be addressed by evaluating the importance of the hydrodynamics of the region. Considerations of hydrodynamics are important for several reasons:

a) These principles govern the potential for contact of the nutrients with the contaminated zone.

b) Hydrodynamics in and along the shore region may affect the concentration of the microorganisms in the oil-contaminated zone.

c) Repeated flooding of the shore region with water (e.g., via tides, waves, and run-off) may result in the mobilization of some of the entrapped oil and its release to the bay area.

3.6 Adequacy of Ecological Assessment

The proposed variety of measurements of ecological effects is too ambitious. The relationship between the questions being asked and the ecological endpoints to be measured was difficult to ascertain. A succinctly stated rationale for the ecological assessments would allow the development of a more focused approach. The following comments are provided to assist with simplifying the proposed measurements and building such a rationale.

The proposed protocol to measure the effect of fertilizer and inoculation will be severely confounded by biological responses known to occur as a result of oil spills, such as suppression of grazers, algal blooms, etc. Calculations of hydrodynamic dilution should be done, preferably by local aquatic scientists familiar with the area, to help predict the possibility of eutrophication. While the proposed experiment focuses primarily on beach effects, offshore effects should be considered in an ecological assessment of the onshore study. Moreover, water column assessments may be less variable and easier to analyze than benthic assessments, and will be just as relevant. The focus of the reviewed study seems to be on visible portions of the beaches, yet areas just below the low water tides are also likely to be affected.

The studies of mutagenicity and higher organisms are considered to be of less importance than those that reflect the activities of the microbial community, such as heterotrophic activity, and primary productivity. The importance of making measurements on the microbial community, a community that can respond to treatment in the short time that will characterize the experiment, was stressed along with the need for simplicity in design. Studies of the C(Carbon):N(Nitrogen) ratio are

considered to be more important than isotope studies which are difficult to perform, analyze, and interpret.

The possibility of stimulating algal growth should be considered via nutrient modeling, assays of algal response to various treatments in the laboratory, and in situ growth studies. Possible effects on infaunal communities and higher organisms (for example, mussels and macrophytic algae) should accompany the experiment under review via long-term monitoring programs. Such monitoring programs should address macroalgal abundance, gross species composition changes and residue uptake in mussels, along with other parameters.

In conclusion, the Committee stresses the need for selection of endpoints that will allow detection of possible responses to the experimental treatments. These responses must be distinguishable from the direct responses of the ecosystem to petroleum contamination.

3.7 Fertilizer Selection

The Committee agrees that there is clear evidence to support the view that nutrient addition may enhance bioremediation. However, inadequate attention has been given in the protocol to information available on fertilizer technology. More information should be sought on available slow release fertilizers which would probably enhance microbial growth most effectively and on application rates that are appropriate and feasible.

The methodology presently proposed using both oleophilic and commercial "slow-release" nutrient formulations is supported by the Committee. However, it is clear (and perhaps understandable given the rapid ORD response) that all options have not been considered. The Tennessee Valley Authority at Mussel Shoals, Alabama, has considerable information and expertise on slow release fertilizer formulations with different physical and chemical properties. This body of information should be considered in protocol development. Other delivery options need to be considered to the full extent possible, including the use of fertilizer spikes, coring equipment to implant fertilizers, and high pressure applications such as those used in asphaltic matrices to prevent rapid nutrient erosion from newly constructed roadways.

3.8 Organism Selection

The addition of microorganisms for bioremediation was considered by the Committee to be less feasible than the addition of nutrients. There is little convincing evidence to support this approach. In addition, limited attention was given to where the organisms would be obtained and how they would be cultured or applied. The Committee was not convinced that the added organisms would survive on the beach long enough to affect bioremediation.

The research plan does not address the characteristics of the organisms to be used for inoculation. The bacteria to be used should not be selected because of the ease of their cultivation in nitrogen- and phosphorus-rich liquid medium because these are not likely to be the organisms that will function in the area designated for inoculation. Enrichments should be established for oil degrading microorganisms which will be able to withstand the stresses at the test site and be able to grow under the conditions that prevail there. For intertidal, cobbled sites with nitrogen and phosphorus-poor water and oil adhering to the solids, the bacteria to be enriched probably should be those that are uniquely able to multiply at ambient temperatures, at very low nitrogen and phosphorus concentrations, and have adherence properties to allow them to attach to solids. They should also be able to withstand such possible intertidal stresses as varying salinity, high light intensity and possibly drying. Such microorganisms will not grow as readily in fermentors as do the species more commonly used for laboratory research purposes, but they are more likely to be beneficial in the target field situation.

The Committee recommends that laboratory studies be conducted to further investigate the possibility for bioremediating with added organisms, and suggests that the data so obtained be analyzed by ORD and reviewed by the Scientific Steering Committee, the SAB or other expert groups with no vested interest to provide guidance on the utility of possible scale-up for future activities. Since limited data are available to support the feasibility of bioremediating with added organisms, laboratory studies are a necessary precursor to field application.

3.9 Potential for Scale-Up

The research plan provides minimal scientific details to assess the potential of successful scale-up. The success or failure of bioremediation, assuming that laboratory studies will demonstrate enhanced biodegradation, will depend on the feasibility of scale-up. Therefore, it is strongly recommended that biodegradation rates be estimated and that simple hydrodynamics analyses be performed in order to assess the feasibility of scaling up the proposed approach. Such an analysis may reveal key factors for evaluation in preliminary experiments or in the proposed field studies. An effort should be made to design both laboratory and field-scale studies for maximizing information pertinent for process scale-up.

Site selection criteria should also be considered with potential for scale-up in mind. The criteria should ensure generalization of the results. The criteria given ensure a good experimental site but do not necessarily ensure a representative site. Snug Harbor's ability to represent other beaches with respect to hydrodynamics, sediment size, distribution of contamination, biota, etc. should be assessed.

3.10 Technical and Personnel Support

3.10.1 Redundancy in Analytical Capability

Decisions on whether the proposed treatments accelerate the degradation of petroleum hydrocarbons will be based on chemical analyses. These chemical determinations must be completed before a decision to "scale-up" is made. Any interruption in information flow from the analytical laboratories to the project officers will seriously jeopardize the successful completion of the experiment and the usefulness of the data. Therefore, sufficient redundancy should be built into the chemical analytical systems to compensate for inevitable equipment malfunctions. Not only should there be back-up gas chromatographs, but arrangements should be made with other laboratories to participate should major difficulties arise.

3.10.2 Detailed Analytical Chemistry to Determine Microbial Degradation Rates

The extent to which microbial communities are degrading the oil will be revealed by the disappearance of certain aromatic and aliphatic hydrocarbons relative to the control or reference sites. The critical and most sensitive step will be the accurate and precise determination of these substances in extracts of intertidal sediments (pebbles and cobbles). The proposed analytical protocol states that glass capillary gas chromatography with flame ionization detectors will be the major quantification tool. This appears appropriate since the composition of the oil mixture is known and analytical standards are available for many of the compounds in the aromatic and saturated fractions. The chromatography must, however, be as comprehensive as possible in order to maximize the potential of detecting compositional changes. The use of standard or "accepted" methods, developed for other purposes, may or may not be sufficient for this task. Consideration should be given to using analog to digital converters and data systems or computers to store complete instrument signals so that more detailed analyses can be performed if needed.

Portions of the final extracts should be stored in freezers for future chromatographic analyses should the necessity arise. EPA may want to consider providing samples to the National Institute for Standards and Technology for storage, analysis and comparison. A plan should also be developed for storing unanalyzed samples in appropriate freezers to maximize the amount of information that can be gained from this experiment.

3.10.3 Personnel

The Committee was not sure why local scientists and engineers are not being consulted by ORD, or being used more extensively to support the proposed projects. The reasons for involving local scientists and engineers are numerous and obvious.

Many areas of expertise have been represented in ORD's protocol development, and many relevant experts have been consulted. However, the omission of microbiologists with field experience, fertilizer technologists, engineers (e.g. from the Corps of Engineers), personnel from the University of Alaska, and, especially, statisticians, should be corrected.

4.0 CAUTIONS AND FUTURE DIRECTIONS

Many proprietary microbial preparations are being marketed, or attempts to market them are being made, with statements about their effectiveness and utility for the biodegradation of oils, greases, degreasing materials, PCBs, pesticides, and industrial chemicals. The effectiveness of most of these preparations has not been verified under conditions for which they are proposed for use. Validation and verification of the claims made by the inoculant manufacturers are not generally required. In the absence of demonstrated utility of these microbial preparations for biodegradation of target pollutants under conditions closely simulating the polluted area, the Agency should not use or encourage the use of any such inocula.

Instead, ORD should use the Exxon Valdez Oil Spill project and related research programs to initiate, together with appropriate program offices, a research demonstration and implementation program for the use of bioremediation as part of the Agency's emergency response plan for spills and inadvertent discharges of chemicals and mixtures. The long use of microbiological methods for the treatment of industrial and municipal wastes attests to the efficacy of biodegradation as a practical, low-cost, non-hazardous means for destroying chemicals. However, the ways that this technology can be applied in a timely manner to destroy chemicals that are inadvertently released have not been significantly addressed. The research necessary for addressing these aspects should be conducted and the technology for remediation should be well defined before future spills occur. The bioremediation program reviewed herein, whether successful or not, can serve as a case history for future research planning and technology development.

Appendix A: Specific Comments:

The design of the continuous flow experiments (page 15) should consider the appropriate residence time of the water that contacts the beach material. For example, one of the key scaling parameters is Q/V (in which Q is the water flow rate and V is the volume of the beach material to be treated). However, since the beach material is essentially a granular porous medium, the thickness of this layer should also be considered. One can, for example, rely on simple models of flow past a porous layer in order to ensure that the pertinent hydrodynamics are being considered. The hydrodynamic considerations involved in the design of the experiments where water movement is being considered do not appear to be well connected with the pertinent in-situ hydrodynamic factors.

Page 18 The statement "Distinction between dispersion and biodegradation will be assessed visually" is a gross oversimplification of the complex physical and biochemical processes that are taking place. A visual inspection cannot be used to distinguish between dispersion and biodegradation. It is not clear what visual parameters will be used in this assessment or whether they will be adequate for this purpose.

Page 19 Insufficient information is given to assess the proposed "additional tests" to determine the impact of tidal and weather extremes, freshwater inputs, and lateral mixing

Ammonium (page 19) is not a good tracer for hydrodynamics

Page 21 -
once current pattern is known, place control up-current from fertilized plots

page 23 continue experiment past 3-4 weeks to whenever the deadline for scale-up decisions to increase utility for future emergency response.

Nutrient release rates estimated in continuous flow systems (page 156) need better in situ velocity estimates or measurements with electromagnetic current meters (not film or wire).

Review EXXon data on oleophilic fertilizer stickiness, and penetration, (page 16) and toxicity.

Page 17 Oleophilic toxicity interaction in oil, synergistic effects

add infaunal species to be monitored to page 17

extrapolate uptake from fertilized plots

Appendix B: Excerpts from ORD's "Laboratory Plan for the Alaskan
Oil Spill Bioremediation Project"

BACKGROUND

The site of the Alaskan oil spill is a harsh and diverse environment with poor access. The shoreline, which is geologically young, ranges from vertical cliffs to boulders and pebbly beaches. High energy beaches are common with tides that vary from -4 to -1 m. In some areas, glacial and snow melt creates a strong freshwater signal.

The spilled oil has distributed over an estimated 1000 miles of shoreline. The distribution was primarily controlled by the prevailing winds and ocean currents which are typically from the northeast. Large Variations in the wind patterns and wave action has caused contamination of previously uncontaminated shoreline.

Major areas that have been contaminated include Knight Island, Eleanor Island, Smith Island, Green Island, and Naked Island. Knight Island, the most heavily impacted, has minimal flushing action in some bays and coves. It also has a considerable population of sea otters. Naked Island has extensive herring spawning areas and significant numbers of seabirds, and shorebirds. Presently, there is a substantial migration of birds which will be feeding on the beaches and intertidal areas.

Most of the floating oil in Prince William sound has disappeared leaving the beaches as the main point of contamination. The oil has settled into the fine beach gravel and covered rock surfaces and faces of vertical cliffs. Contamination occurs in and below the intertidal zone. An estimated 300 miles of contaminated shoreline are scheduled for cleaned up.

The oil itself has weathered and will continue to weather. An estimated 15-20% loss of the oil has been lost due to volatility. The residue is approximately 40 - 50% high molecular weight waxes and asphaltenes. On many beaches, the general condition is not that of a mousse but instead a black oily layer.

Presently, some beaches are being cleaned by a combination of flooding and the application of water under high and low pressure and /or high temperature. Vacuum extraction is being used to remove the released oil from the water surface. The cleaning process partially removes oil from the surface of rocks and beaches but does not effectively remove oil down in the fine grained gravel or the cobble. The extent of physical treatment is dependent upon the degree of contamination.

Table 1. Ratios of Alkane Peaks

SAMPLE	N-C ₁₇	Pristane	N-C ₁₈	Phytane	N-C ₁₇ /Pristane	N-C ₁₈ /Phytan
Calibration						
Fresh PB Crude	3,000	1,730	2,560	1,260	1.7	2.0
CH ₂ Cl ₂ Blank						
Surface Control	<.0025	.0053 ^a	<.0025	<.0025	<0.47	--
6" Depth Control	<.0025	.0056 ^a	<.0025	<.0025	<0.45	--
NW Bay Surface Elenore Is	12.9	8.75	12.9	6.55	1/5	1.9
NW Bay 6" Depth	1.63	1.18	1.44	.870	1.4	1.7
NW Bay 18" Depth	.0435	.108	.0331	.0765	.40	.43
Seal Island	29.0	17.9	25.8	12.4	1.6	2.1
Smith Island	.605	.403	.545	.281	1.5	1.9
Disk Island	12.5	15.6	13.0	12.6	.80	1.0
Fresh Oiled Rock	1,840	1,290	1,840	1,070	1.4	1.7
Weathered Oiled Rock	1,980	1,110	2,280	1,150	1.8	2.0

^a - possible biogenic input

PROJECT PLAN

I. Preliminary Studies

The overall project is composed of two parallel studies. The first will consist of a field study to evaluate the use of different nutrient additions to enhance the biodegradation of the contaminating oil. The second involves a smaller scale field study to evaluate the use of adding microbial cultures to enhance the degradation rate. Preliminary data will be gathered to assess initial field and application conditions. This will include:

- * Survey of the geomorphology, oceanography, and oil contamination to determine if appropriate sites for the demonstration projects were available.
- * Chemical characterization of the weathered oil taken from selected sites in Prince William Sound.
- * Collection of information on the characteristics and availability of slow release and oleophilic fertilizers.

Results from initial studies are summarized below.

A. Beach Survey

During the first trip to Prince William Sound (PWS) on 04/26/89 - 05/02/89, the EPA Bioremediation task force members surveyed most of the impacted beaches using small boats, float planes and helicopters. Descriptive assessments of geomorphology, tidal action and extent of contamination were made. Protected beaches that had moderate oil contamination over a long stretch of either coarse gravel, pebble and/or cobble were examined. Homogeneity of the beach areas in terms of geomorphology and oil contamination was also considered.

Oil contamination can be described in two ways:

1.) Primary contamination. The color of the oil is black. The oil slick was present near the beach for an extended period of time and oil covers all or most of the intertidal zone. Visual penetration of the oil into the gravel was 4 - 18 cm in moderately impacted areas. Oil was also blown on the beach during a storm. These beaches are characterized by a relatively thin layer of oil, mostly above the high tide mark.

Additional beaches of fine gravel had little or no signs of contamination on the surface. However, several millimeters below the surface oil was visible. Such beaches usually contained larger rocks with visible oil coverage.

2.) Secondary contamination. The color of the oil is mostly brown. Coloration is due to mousse formation on the sea before beaches were impacted. A relatively small number of beaches at P.W.S. show mousse contamination. It is more pronounced along the Kenai Peninsula South West of Seward.

For the nutrient addition demonstration project the gravel/cobble stone beaches impacted by oil covering the tidal zone where oil is on and or below the surface seemed most appropriate. These beaches are both extremely important in terms of biological habitats to shorebirds, crustaceans and fish larvae and fry, and are also the most difficult to physically clean. Physical treatment of pressurized water will change the natural stratification of such beaches, and may result in extensive erosion. Chemical treatment could cause further damage to marine organisms living along the shoreline, below the tide zone, which survived the initial toxicity of the oil.

Based on these surveys the location of Snug Harbor was chosen as the area for further consideration of the demonstration projects. This area had ample protected beaches of the proper geomorphology and relative uniform oil contamination to a moderate extent. It was also readily accessible with plane or helicopter and was probably an area that would be cleaned by Exxon later in the summer.

B. Chemical Composition of Weathered Oil

Beach material from several contaminated areas was sampled and the material extracted with methylene chloride. Extracts were evaporated and the residue was weighed and brought up in a specified volume of pentane. The solutions were analyzed by capillary gas chromatography directly or fractionated into aliphatic and aromatic fractions and chromatographed. Details of the analytical methods are given in Appendix II.

Samples analyzed:

- * Eleanor Island, Northwest Bay, surface (0 - 4"); oil impacted and control beach material.
- * Eleanor Island, Northwest Bay, depth at 6"; oil impacted and control beach material.
- * Eleanor Island, Northwest Bay, depth at 18", oil impacted beach material only.
- * Seal Island, surface (0 - 2"), post initial physical cleaning.
- * Smith Island, surface (0 - 2"); oil impacted beach material.

- * Disk Island, surface (0 -2"); oil impacted beach material.
- * Disk Island, fresh-looking oiled rock.
- * Disk Island, weathered-looking oiled rock.

All control samples were taken several meters above the impacted area. Gas chromatographs for some of the analyses are shown in Appendix II.

Analysis of the results showed a typical envelop of weathered oil with hydrocarbons below C11-12 missing. A large quantity of biodegradable hydrocarbons, C13-C28, were present in these chromatograms. The presence of these compounds in the oil suggests that it can be biodegraded by the naturally occurring bacterial population. The fractionated samples showed relative small quantities of aromatic hydrocarbons and loss of hydrocarbons up to the methyl naphthalenes.

Both controls showed some peaks with low retention times. It is assumed that these peaks are not crude oil related compounds, and are probably of biogenic source. Beach material from impacted surface and 6" depth looked similar. A sample from the 18" depth showed much lower concentrations of hydrocarbons. However, many peaks were present and a distinct pattern of the oil is seen. Visually, at that specific location, the oil was seen at 4-6" depth only.

The collected from Seal Island showed a low degree of weathering with a significant amount of C-10 n-alkane present. A much more weathered sample was apparent from the Disc Island rock that visually appeared weathered.

Table 1 gives the calculated ratios of C-17/pristane and C-18/phytane for each of the samples taken. Except for the Disk Island surface sample, little biodegradation of the oil is evident.

C. Fertilizer Information

Information on the characteristics of selected slow release water soluble and oleophilic fertilizers are listed as follows:

IBDU FERTILIZER/NUTRIENT FORMULATION - A 24/4/12 (N-P-K) fertilizer that is formulated to give both an immediate and sustained response. 100% of the nitrogen is derived from ammonium phosphates, urea, and isobutyldiene diurea, with a minimum of 45% from water insoluble isobutyldiene diurea. Available phosphoric acid is derived from potassium sulfate and potassium magnesium phosphate. Iron is derived from ferrous sulfate. When used on turf, water soluble nitrogen response will become evident in approximately one week, while isobutyldiene

diurea will begin release in 3-4 weeks and continue for a minimum of 12 weeks. Information on cost is pending.

SPIKES - Nutrients can also be supplied using off-the-shelf tree food spikes. The spikes can be obtained in various formulations, such as 14-5-5 (N-P-K) or 16-10-9. Phosphorus is present as phosphoric acid (P_2O_5) and potassium is present as potash (K_2O). The spikes can be implanted underneath the exposed cobblestone at various locations in and above the tidal zone or attached to stakes or placed in mesh bags, which can then be secured to the beach either in holes, trenches, or weighted down by rocks. It's cost is approximately \$1.45 per pound (case price, 16-10-9 formulation).

OLEOPHILIC FERTILIZER FORMULATIONS/INIPOL EAP 22 - A proprietary mixture of nutrients encapsulated by oleic acid (the external phase) designed and originally produced by Elf Aquitaine (France) for tertiary oil recovery. This fertilizer (nutrient) formulation belongs to a category of oleophilic nutrients, in that the vehicle (oleic acid - surfactant) renders the nutrients to become suitably attached to the oil phase and thus prevents them from becoming solubilized in aqueous phase and subsequently washed out. Its appearance is a clear liquid with a specific gravity of 0.996, a viscosity of 250 cSt, a pour point of 11 C, and a flash point of >100 C. Its cost is approximately \$1.50 per pound.

INORGANIC SOURCES - Several inorganic sources of fertilizer are available, such as ammonium phosphate, ammonium nitrate, and the slightly soluble magnesium ammonium phosphate. Reasons for using these materials are simplicity, no known toxicity at the intended concentrations, and no additional carbon source. Combination with urea is possible. These inorganic compounds, which are relatively fast release, can be combined with the oleophilic fertilizer to supplement the amount of N and P in the formulation.

The biodegradation of oil has been extensively studied over the last 20 years. As a result, the microbiological fate of oil in the aquatic environments is well understood. Studies have also shown that oil degradation can occur in cold water environments (Atlas et al., 1977). Because of this data base, The EPA Office of Research and Development convened a workshop of national and international scientists involved in oil biodegradation research and asked them about the possibility of some type of oil bioremediation in connection with the Prince William Sound oil spill. The objectives of that workshop and the list of the attendees is given in Appendix I. Several bioremediation options became apparent as a result of the workshop discussions. This included the following options for accelerating oil degradation:

- * The addition of nitrogen and phosphorus nutrients.
- * Inoculation with commercial or enriched indigenous microorganisms.
- * Alteration of site characteristics by mechanical mixing.
- * Increasing availability of the oil by the use emulsifying agents.
- * One or more of the above in combination.

It was the consensus opinion of the workshop participants that it was worth performing demonstration projects on the first two options, nutrient addition and inoculation. Decisions for scale up would be based on the success of the demonstration projects. Nutrient addition appeared to give the greatest chances for success for this season. It was the general conclusion that bioremediation would be most effective if it was coupled with ongoing cleanup operations in Prince William Sound. Specifically, bioremediation could possibly be quite effective in removing oil from the beaches that is not be removed by the current physical washing procedure; that is, a process whereby oil under the rocks and down in the gravel beach pore water could be removed.

Pursuant to these conclusions, the EPA Biosystems Technology Development Scientific Steering Committee has developed the following implementation plan.

PROJECT MANAGEMENT

This project will be managed by the EPA office of Research and Development. A team of scientists will carry out, under EPA's direction, this on-site project in Prince William Sound. A research plan will be developed and peer reviewed prior to initiation of the project. Figure 1 is a summary of the project management structure.

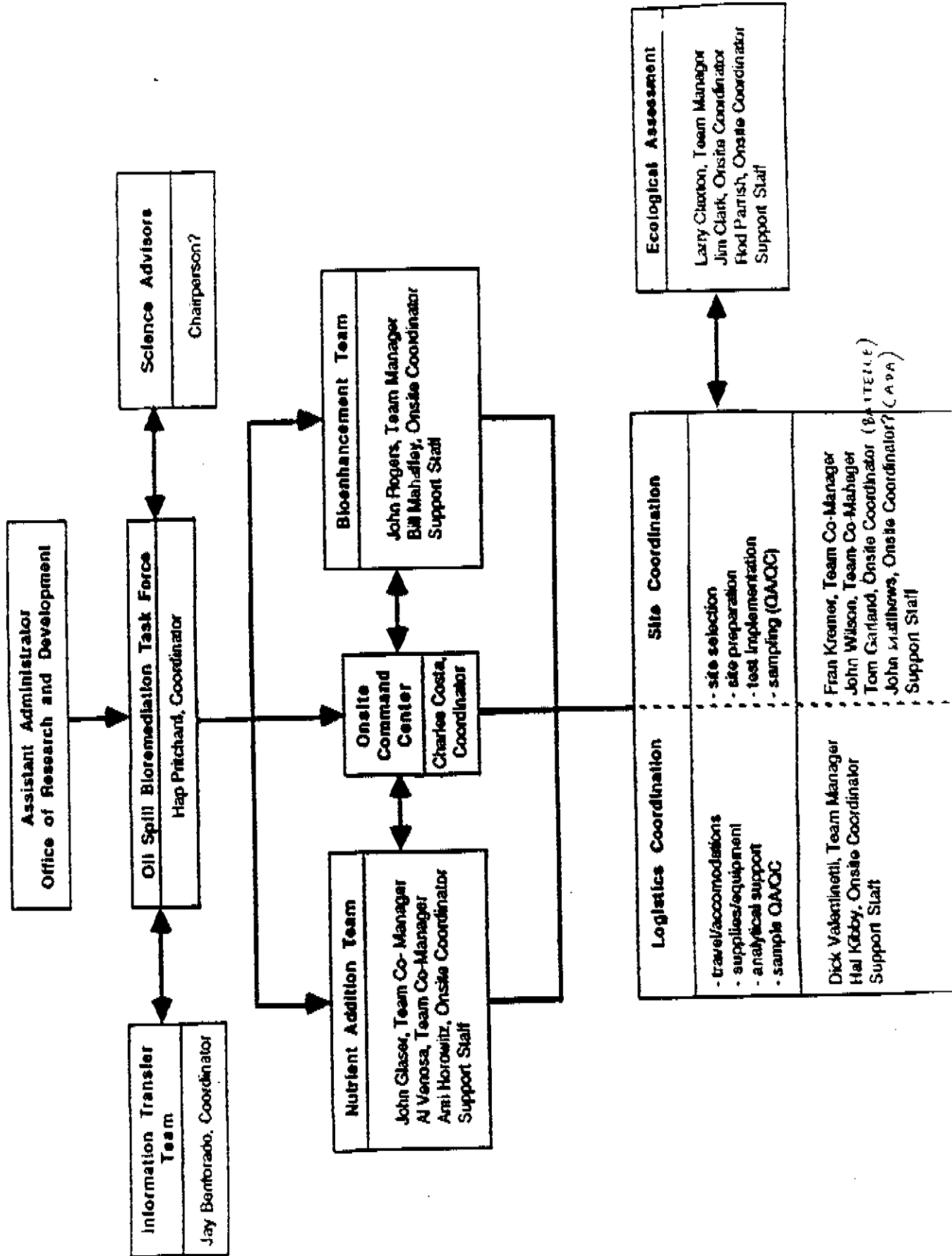
The project will be implemented over a four month period from May 1, 1989 to September 30, 1989. A time line for this implementation is given in Figure 2. For the nutrient addition study, the schedule will be to obtain information on success as soon as possible; we expect this to occur early in the summer. If success is apparent within this time period, it will allow scale up operations to be implemented in time to affect oil cleanup during the summer season of 1989 when temperatures are reasonable for biodegradation activities on the beaches.

The bioenhancement study will also be implemented in early May starting with a series of laboratory studies. Information and microbial cultures are planned to be available for testing in the field in early June. If scale up becomes a possibility, immediate action will be undertaken to mobilize reactors for culturing of large quantities of the bacteria. Inoculations on a larger scale could possibly commence in July.

A scientific meeting to report and discuss the results of the project will be held in late September. A full documentation of the project will be prepared and released in the Fall, 1989.

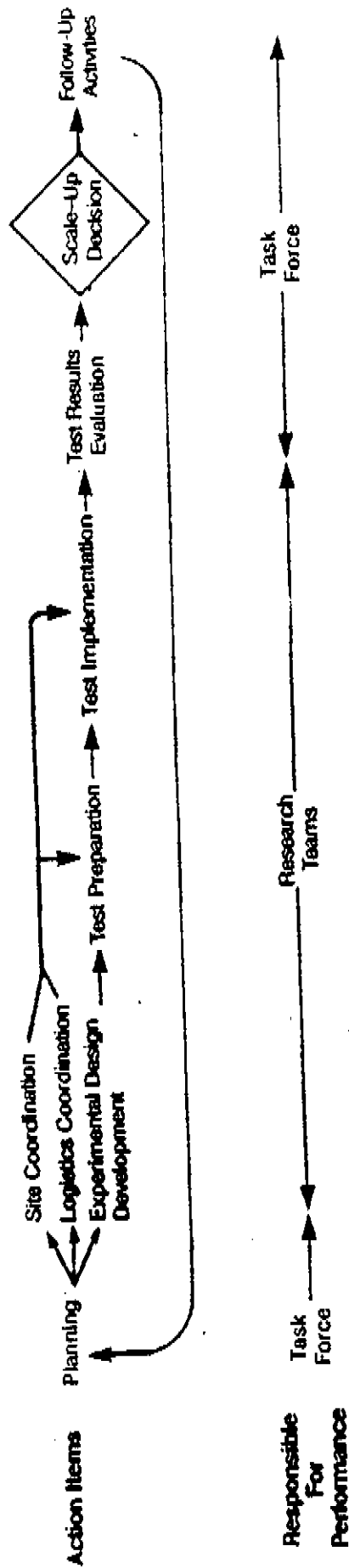
FIGURE 1

Proposed Coordination for EPA Oil Spill Bioremediation



FIGURE

Proposed Plan of Action Flow Diagram

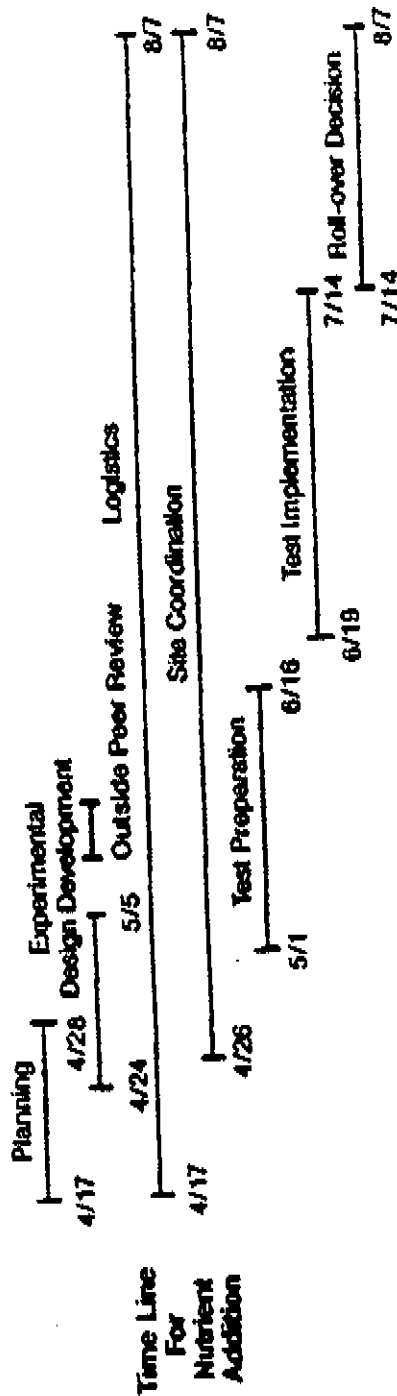


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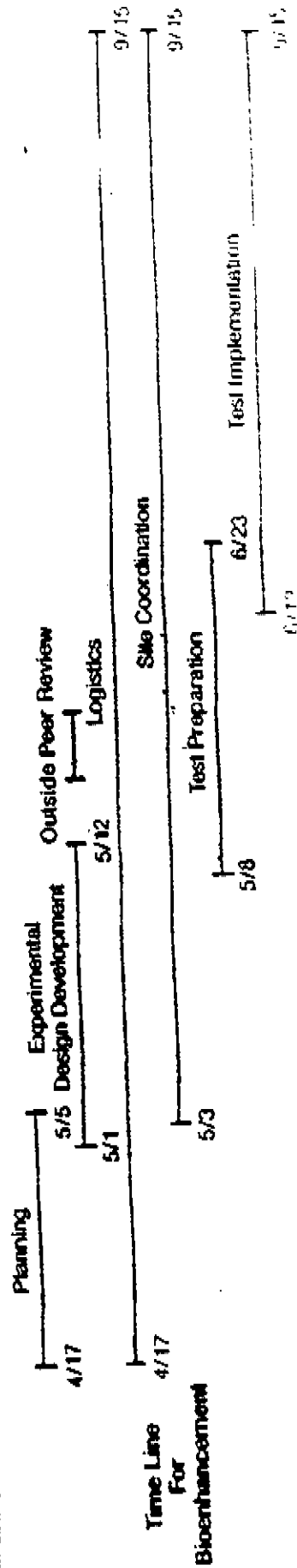
Task Force

Research Teams

Task Force



Time Line For Nutrient Addition



Time Line For Bioenhancement