Project Title: Microcosm Study on the Biodegradability of Lingering Oil in Prince William Sound 19 Years after the Exxon Valdez Oil Spill —Submitted under the BAA

Project Period: June 1, 2008 to May 31, 2009

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Abstract: This proposal will provide important information that would help evaluate the persistence of the lingering oil in many of the Prince William Sound beaches affected by the 1989 Exxon Valdez oil spill. Because biodegradation of oil occurs at the oil-water interface, limitations occurring in the vicinity of that interface are hypothesized to be the primary reason for the lingering oil. The likely sources of limitation include: (1) environmental limitations (such as low nutrient concentrations and/or low oxygen) that would limit biodegradation, and (2) the lack of bioavailability of the oil due to its weathering or the existence of an impenetrable "skin" on the oiled sediment. This study proposes to investigate the biodegradability of the lingering oil collected from several sites still showing signs of oil in the subsurface. It will answer important questions about the biodegradability of the oil that has undergone weathering for 19 years. The laboratory study described in this proposal will provide evidence that could support decisions on whether to bioremediate the remaining oil contaminating the subsurface at selected sites in PWS. A complementary tracer study is currently ongoing in PWS to establish and understand the hydrodynamic properties of the PWS beaches that would allow the addition of nutrients, and possibly oxygen, for biostimulating the lingering oil. Results from both studies combined will provide sufficient support to aid the Exxon Valdez Oil Spill Trustee Council in making a decision regarding the propriety of undertaking an investigation of the applicability of bioremediation in the field.

Funding for FY '08: $166,729, excluding 9% G&A requested by Trustee Council
FY '08 Cost-Sharing by U.S. EPA: $2,043
Total FY '08 requested including 9% G&A: $181,735

Funding for FY '09: $332,989, excluding 9% G&A requested by Trustee Council
FY '09 Cost Sharing by U.S. EPA: $14,508
Total FY '09 requested including 9% G&A: 354,238

TOTAL REQUESTED: $491,718 excluding 9% G&A and EPA Cost Share
Cost-Sharing by U.S. EPA: $16,551
TOTAL including EPA cost share: **$491,718**
TOTAL including 9% G&A (1.09 * $491,718): $535,973

1.0 NEED FOR THE PROJECT

This proposal addresses the request by the Exxon Valdez Oil Spill Trustee Council (EVOSTC) for proposals that investigate the physical processes that affect the lingering oil in the subsurface intertidal sediments on some beaches in Prince William Sound (PWS). The proposed research will investigate the biodegradability of the lingering oil to determine to what extent in-place treatment of lingering oil by the addition of nutrients and/or oxygen is feasible. *In-situ* bioremediation was identified by Michel *et al.* (2006) as one of two feasible alternatives for removing the lingering oil.

2.0 PROBLEM STATEMENT AND HYPOTHESES

In 2001 and 2004, scientists from the National Oceanic and Atmospheric Administration (NOAA) conducted geospatial surveys of lingering oil in PWS and found that about 11 hectares of shoreline remain contaminated with nearly 56,000 kg of subsurface oil from the 1989 Exxon Valdez oil spill (Short et al., 2004). Much of the lingering oil was present in subsurface sediments in the middle intertidal zone of the contaminated beaches. Although substantial weathering had occurred, the researchers concluded high concentrations of toxic and mutagenic contaminants were present, suggesting that this lingering oil poses an ongoing threat to organisms that encounter it. The residues were classified by Short et al. (2004) using the standard descriptive terms adopted for PWS, which are LOR, MOR, and HOR, corresponding to low, moderate, and high oil residue, respectively.

One possible explanation for the lingering oil is limitation in biodegradation rates caused by lack of nutrients and/or oxygen in the intertidal zones of certain beaches. The potential for limitation of the oil biodegradation rate by transport of certain critical nutrients, such as nitrogen, phosphorus, and oxygen, has its basis in the stoichiometric requirement for these materials to support biodegradation of petroleum hydrocarbons (Atlas and Bartha, 1972; Atlas, 1981; Wrenn et al., 2006). Demand for these nutrients is directly proportional to the concentration of oil in the contaminated sediments. Kinetic limitations due to low nutrient concentrations could also play a part in the oil lingering for such a long time in the subsurface. Previous research has shown that nutrient concentrations between about 2 to 10 mg N/L are required to support maximum oil biodegradation rates (Boufadel et al., 1999; Du et al., 1999). A 1994 field study in Delaware Bay (Venosa et al., 1996) demonstrated conclusively that nitrogen concentrations of 3 to 6 mg N/L in the interstitial pore water significantly ($p < 0.05$) stimulated hydrocarbon biodegradation by 2- to 3-fold over natural attenuation where the average interstitial nutrient concentrations averaged 0.8 mg N/L. These reported minimum nutrient concentrations are far higher than the average concentrations of 0.2 mg/L observed in PWS (Bragg et al., 1994). Exxon-Mobil (hereafter Exxon) scientists also reported an increase in the biodegradation rate of the Exxon Valdez oil (EVO) in PWS beaches when the pore water nitrogen increased from about 0.2 mg N/L to 1 mg N/L due to nutrient addition (Bragg et al., 1994).

Temperature and pH could also affect rates of biodegradation of oil. In particular, biodegradation of oil is expected to be highest in summer due to the relatively warmer temperature of water in the Sound. The literature has also shown that, during the biodegradation of poorly-soluble hydrocarbons, the degrading microorganisms act at the oil-water interface (Rosenberg and Rosenberg, 1981; Watkinson and Morgan, 1990; Jimenez and Bartha, 1996). Thus, one needs to account for the bioavailability of oil. That is, the biodegradation rate could be limited by the mass-transfer rate of the biodegradable components of oil, such as the polycyclic aromatic hydrocarbons (PAHs), to the oil-water interface (Rosenberg et al., 1992; Nicol et al., 1994) where the uptake by microorganisms occurs.

At the 2007Arctic Marine Oilspill Program (AMOP) Conference in British Columbia, Atlas and Bragg (2007) developed and presented a bioremediation index based on the degree of weathering of oil normalized to conserved biomarkers such as hopanes, stigmastanes, cholestanes, and others. They argued that, if the degree of weathering of oil normalized to one of the conserved biomarkers is 70% or more, then further attempts to bioremediate it would be futile since the weathering is so extensive that additional effort would not be justified. The Atlas-Bragg index does not fully account for the PAH content of the lingering oil in PWS. Sufficient PAHs appear to be present in some of the PWS sites to justify further study. This microcosm study is designed to address the extent to which the lingering oil is biodegradable given varying degrees of weathering. Based on recent surveys conducted by Short (personal communication, 2008), there appear to be a moderate number of sites where the PAHs in the lingering oil are less than 70% degraded according to the Atlas-Bragg model. This project directly addresses this concern.

3.0 OBJECTIVES

The objective of this project is to measure the biodegradability of the lingering oil in PWS 19 years following the original spill incident under conditions where nutrients and oxygen are not limiting (compared to the condition where nutrients but not oxygen are limiting). This laboratory study will provide evidence to support a recommendation concerning bioremediation of the remaining oil through the addition of nutrients and/or oxygen in the subsurface at selected sites in PWS. A tracer study is currently ongoing in PWS to establish the hydrodynamic properties of the PWS beaches for allowing the addition of nutrients for biostimulation of the lingering oil, in the event that the oil is found from this new investigation to be biodegradable. So, the data from the tracer study combined with the biodegradability of the lingering EVO from this project are expected to provide a strong foundation upon which the Trustee Council can make an informed decision regarding field application of bioremediation through the addition of nutrients and possibly oxygen.

4.0 RELEVANCE TO THE 1994 RESTORATION PLAN GOALS

The proposed research will investigate the potential limitations to biodegradation that may account for the persistence of *Exxon Valdez* oil in the subsurface sediments of beaches in PWS. A comprehensive microcosm study is proposed to determine the degree to which oiled sediment collected from PWS sites is amenable to biostimulation, the process whereby addition of nutrients stimulates native microbial populations to consume the remaining oil faster than natural attenuation. Another project, begun last summer, is underway to determine, among other things, *in-situ* concentrations of nitrogen and phosphorus. This project addresses this issue in two ways: (1) it is assumed that the N and P levels in PWS are similar to the extremely low levels reported by Bragg *et al*. (1994); this will be verified by the current field study of Boufadel *et al*., and (2) N and P will be added to some of the microcosms at non-limiting (i.e., excess) concentrations and compared to other microcosms having the natural concentrations of N and P in the seawater feeding the microcosms (natural attenuation). This information will assist in future remediation efforts because it will aid decisions on whether and where such efforts are appropriate and because the remediation technology that is selected must address the most important limiting factors.

The results of this investigation will provide needed information on the biodegradability of lingering oil in PWS to determine whether bioremediation of PWS beaches through nutrient addition is worth investigating further. If the factors that are responsible for the persistence of the subsurface oil can be controlled through engineered manipulation of subsurface conditions, this research may lead to development of a comprehensive bioremediation plan that will restore habitats that are adversely impacted by the lingering oil. The benefits of this research to the evaluation and implementation of bioremediation in PWS is consistent with the EVOSTC's objective of determining whether remediation of specific shorelines would protect or restore injured resources. Bioremediation would be the preferred remedial alternative for contaminated beaches in PWS because it would avoid significant habitat disturbance. So, the exposure of sensitive species to oil would not be expected to increase during remedial operations. The primary beneficiaries of this research would be natural resources in PWS that have not yet fully recovered from the *Exxon Valdez* oil spill due to exposure to the lingering oil and the human communities that depend on these resources for their livelihood and quality of life.

5.0 RATIONALE FOR A MICROCOSM STUDY

After examining the gas chromatography/mass spectrometry (GC/MS) data from the LOR, MOR, and HOR samples collected by the NOAA researchers (Short *et al*., 2004), the PAH concentrations appear to be high enough in the samples reported to justify conducting this microcosm study despite Atlas and Bragg's (2007) assertion that their bioremediation index was calculated at 70, signifying that further bioremediation effort is unjustified. This was especially true for the MOR and HOR samples. The average % remaining C_2-chrysene normalized 3-ring PAH concentrations of 17 LOR samples remaining in 2004 compared to day-11 oil was 45%. For the 14 MOR samples, the average % remaining was 161% (partially skewed by a few high results due to the variability of oil concentrations in samples measured), while for the 4 HOR samples, the % remaining was 69%. These concentrations are high enough to be a cause for concern in terms of long-lasting effects on the ecosystem in some of the contaminated PWS sites (see Zhu *et al*., 2001 and 2004, for a discussion of bioassays such as the solid-phase Microtox^{TM} assay, the algal solid phase assay, *Cladoceran* and amphipod survival assays, and gastropod survival assays, which have all been used to demonstrate remaining toxicity of oiled sediments). If a microcosm study is not conducted on the existing lingering oil, we will never know for certain if a full-scale bioremediation strategy for removing the oil has any potential at all of working. The data obtained from this project will fill an extremely important data gap and will complement the current Boufadel *et al.* study on Limiting Factors. The latter will provide the hydrodynamic and other physical limiting factors information that is crucial for developing a rationale for a follow-on bioremediation effort. The microcosm study will provide the microbiological information needed to estimate the degree to which the lingering oil in PWS is biodegradable. Thus, the data from both projects together are expected to answer the question as much as is physically and biologically possible.

6.0 OVERALL APPROACH

6.1 Collection of beach substrate and seawater.

Samples of beach substrate will be collected by a field crew in the summer of 2008. The samples will be collected from representative sites in PWS contaminated with oil residues of varying weathering states in accordance with the Atlas-Bragg model. The plan is to collect 50-liter samples of contaminated beach substrate from three different sites, tentatively identified as KN114A (Atlas-Bragg index of 76%), SM006B (Atlas-Bragg index of 60%), and PWS 3A4 on

Eleanor Island (Atlas-Bragg index of 30%) plus 20 liters of uncontaminated substrate from the PWS 3A4 site. Note that the lower the Atlas-Bragg Index, the greater is the expectation of achieving biostimulation. A minimum of five (5) substrate samples from each site will be collected, composited into 1 large sample, and mixed in the field by the crew. In addition, 50-liter samples (70 liters for the PWS 3A4 site) of seawater will also be collected at each site to serve as the seawater source for the lab microcosm studies. This plan requires three independent beach sites. The Contractor lab will set up triplicate microcosms of each treatment (as outlined below in Table 1) from each site to enable estimates of repeatability error (within-site error) in addition to reproducibility error (among-sites error).

We also considered setting up a separate treatment that would be fed freshwater instead of saltwater. The purpose would be to accommodate a freshwater bacterial community that could hypothetically have better oil degrading properties than saltwater organisms. This is because in the on-going Boufadel *et al*. "Limiting Factors" study, a preliminary finding showed that less oil was found on one beach where freshwater rather than saltwater was prevalent, suggesting that the freshwater microorganisms might have degraded the contaminating oil over the years better than those present in the saltwater portion of the beach. However, after considering this idea, we abandoned it for several reasons: (1) in our experience over the years, oil degrading communities derived from saltwater environments have in all cases exhibited significantly better oil degrading capabilities than freshwater organisms; (2) other explanations for the observation above are also possible (including washout of oil by the flowing freshwater or the possibility that that particular portion of the beach might not ever have been significantly contaminated by oil); and (3) even if there were a difference between freshwater and saltwater biodegradation, the result would not affect the approach to bioremediation in any follow-on field study. It would merely affect the biodegradation rate observations and not provide an incremental increase in beneficially useful information in terms of cleanup at the field scale.

As oil weathers in the field, the oil exposed to water and air will often form a crust similar to asphalt pavement surrounding the contamination and minimizing mass transfer between the oil and the external environment. Field sampling of contaminated substrate, its homogenization, and its placement into laboratory microcosms will admittedly disrupt these crusts that may have resulted in the persistence of the oil after 18 years in the subsurface. This crusting may have reduced the bioavailability of biodegradable contaminants to the hydrocarbon-degrading microbial populations at the oil/water interface. Therefore, the proposed bioremediation research performed in laboratory microcosms is expected to reveal a greater rate and extent of biodegradability than would normally be observed in a field demonstration without disruption of the substrate. Nevertheless, expected results from the proposed research will provide data on maximum attainable bioremediation based on unlimited nutrients and oxygen. This information would be very valuable for decision makers in strategizing the approach to further remediation.

6.2 Preparation and Sampling of Microcosms

Enough microcosms will be set up in triplicate to accommodate a total of six (6) sacrificial sampling events (corresponding tentatively to Days 0, 14, 28, 56, 112, and 168) for seven treatments [natural attenuation (NA) controls and biostimulation treatments for each of the 3 oiled sites and a "positive" control for the PWS 3A4 site]. Table 1 describes the treatments and allocation of replicates. The NA controls will consist of KN114A, SM006B, and PWS 3A4 microcosms containing seawater but with no nutrient addition. This will provide an estimate of natural biodegradability in the presence of nutrients naturally present at the site without any stimulation by external nutrient addition and will be the control against which to compare the biostimulation treatment. No killed controls are planned because the NA treatments are expected to result in very limited biodegradation due to the limited background concentrations of N and P. Thus, killed controls are not expected to provide any more useful information than the NA treatments.

The positive controls will consist of a third series of microcosms from the PWS 3A4 site to which will be added crude oil known to be biodegradable [Alaska North Slope (ANS) 521, which has been previously distilled at 521 °F to mimic ANS crude oil that has weathered for about two weeks]. Substrate for these positive control microcosms will consist of clean, uncontaminated material collected the same way as the other samples. However, prior to filling the microcosms in the lab with this material, this sample of substrate will be contaminated artificially with ANS 521 oil at an approximate concentration of 5 g/kg, which is close to the concentration found in many areas containing lingering oil. These microcosms, which will be supplied with excess nutrients just like the biostimulation treatment, will be sacrificed at the same times as the other treatments above. The source of oil degraders for all microcosms will be the natural PWS seawater and the beach substrate. The purpose of the positive controls is to determine whether some other characteristic of the water, oil, or sediments (such as lack of availability of oil or oil that is too weathered for effective treatment) would prevent the growth of these organisms, assuming a degradable hydrocarbon substrate is present.

Table 1 summarizes the experimental setup showing in each cell the number of microcosms planned for that cell. Day 0 measurements of hydrocarbons are not needed for the biostimulation microcosms because they are identical to the NA microcosms from the standpoint of the starting oil levels. This design requires a total of 117 experimental units (EUs). The Contractor lab will set up as shown in Table 1.

Table 1. Summary of experimental microcosm setup showing the no. of experimental units in each treatment and sampling time.

Sampling Day	KN114A		SM006B		Oiled PWS 3A4		Unoiled PWS 3A4
	*NA	Nutrients	*NA	Nutrients	*NA	Nutrients	Nutrients + Biodegradable Oil Added
0	3		3		3		3
14	3	3	3	3	3	3	3
28	3	3	3	3	3	3	3
56	3	3	3	3	3	3	3
112	3	3	3	3	3	3	3
168	3	3	3	3	3	3	3
Total	18	15	18	15	18	15	18

*NA = natural attenuation (no nutrients)

The field crew will preserve the four sediment and seawater samples by packing them in ice and shipping them in coolers via Federal Express to the receiving lab in Cincinnati. When the lab receives the beach substrate samples, the samples will be split to accommodate triplicate sets of NA and biostimulation microcosms. The seawater from each site will serve as the test water for that particular site's microcosms. Upon receipt of the substrate and seawater samples, the researchers will immediately set up 117 microcosms as shown in Table 1 above. Both the seawater and the sediment will serve as the source of the natural inoculum. A representative sample of each substrate will be extracted with dichloromethane (DCM) to remove adhered oil, and the extracted sample will be subjected to particle size distribution analysis and porosity determination. These analyses will be used to assess the magnitude of contaminated surface area and ease of transport of nutrients and oxygen.

The oiled beach material in PWS is located in the intertidal zone where submergence is intermittent. To partially simulate this, each microcosm will need to have a separate reservoir as a source of intermittently recirculating seawater. A microcosm (see Figure 1) will consist of one flat-bottomed, sterile glass I-Chem jar equipped with 2 glass tubes, one extending to the bottom of the jar and serving as a water withdrawal line, and the other serving as a water return line that delivers water to the top of the I-Chem jar above the water line and against the side. The withdrawn water is pumped via a peristaltic pump to an open glass reservoir (another I-Chem jar) physically situated above the glass I-Chem jar serving as the microcosm and equipped with a siphon. When the level of the water in the upper reservoir reaches the siphon level, water will be returned by gravity to the lower jar. The flow rate of the withdrawn water will be set to achieve approximately 6 cycles per day (once every 4 hours). This recirculation mechanism is intended to simulate intermittent submergence in the intertidal zone and to provide sufficient reoxygenation to prevent anoxic conditions. Because the upper reservoir will be open to the atmosphere, oxygen will be readily available to dissolve into the recirculating seawater, maintaining aerobic conditions (experience has confirmed that such is the case). Equally important, the continually recirculating seawater will also provide gentle mixing without disrupting the integrity of oil coverage. The bottom of each microcosm will be charged with 200 mL of 1-mm diameter clean glass beads to minimize short circuiting of recirculated seawater and prevent carryover of finer substrate particles into the recirculation system. The glass beads will be overlain with 800 mL of contaminated substrate. The entire microcosm will be filled with seawater to a depth of 2 cm above the substrate level during times of submergence. All microcosms and reservoirs will be wrapped in aluminum foil to prevent algal growth.

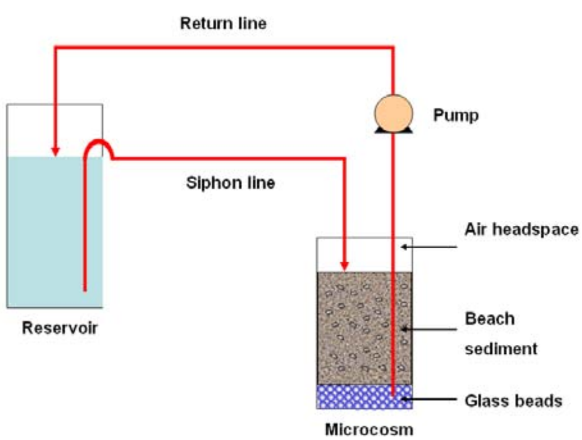

Figure 1. Schematic diagram of microcosm assembly.

6.3 Nitrogen and phosphorus measurements.

Nitrogen in the form of KNO_3 and phosphorus in the form of sodium tripolyphosphate will be supplied to each biostimulation microcosm and each positive control microcosm (but not the NA microcosms) at concentrations that will result in 10 mg nitrate-N/L and 1 mg P/L, respectively. Aqueous phase nitrate will be analyzed once per week in 3 randomly selected nutrient-amended microcosms to assess the degree to which nutrients will need to be re-supplied. It is assumed that biodegradation will not be limited if the nitrate-N is maintained at approximately 5-10 mg/L (Venosa and Zhu, 2005). If it falls below 5 mg/L, sufficient sterile KNO_3 will be added to bring the level back to 10 mg/L. Since the microcosms are closed, it will be unnecessary to ever add additional phosphorus. Periodic replacement of evaporated seawater will be accomplished by adding sterile distilled water (to prevent accumulation of salts).

The concentrations of nitrate and phosphate in water samples from the microcosms will be measured using ion exchange chromatography. Ammonium will be analyzed by a specific ion electrode. The water samples will be filtered through 0.45-μm membrane filters and stored frozen in clean high-density polyethylene bottles for later analysis. The detection limits for these methods are 20 μg N/L for ammonia, 0.1 μg N/L for nitrate, and 0.7 μg P/L for phosphate.

6.4 Sacrificing Microcosms

At Day 0, 12 microcosms designated in Table 1 will be sacrificed by extracting the substrate and the seawater with DCM. The extracts from these samples will be used to analyze Day-0 alkanes and PAHs by GC/MS operating in the selected ion monitoring mode (SIM), a method equivalent to EPA SW-846, Method 8270C. In addition, the extracts will also be analyzed with a GC/flame ionization detector (GC/FID) to determine total petroleum hydrocarbons (area under the curve). Alkanes will include normal and branched aliphatics ranging in carbon number from C_{12} to C_{35} plus pristane and phytane. Aromatics will include 2-, 3-, 4-, and >4-ring PAH compounds and their alkylated homologs (C_{0-4} -naphthalenes, C_{0-3} -fluorenes, C_{0-3} -dibenzothiophenes, C_{0-4} -phenanthrenes/anthracenes, C_{0-4} -napthbenzothiophenes, C_{0-2} -pyrenes, C_{0-4} -chrysenes, and selected pyrogenic PAHs). At each subsequent sampling event, 21 more microcosms (including the associated reservoirs and tubing) will be sacrificed and extracted similarly with DCM. After extraction, the mass of substrate will be measured by drying and weighing the samples on a top-loading balance. All analyte concentrations will be reported both in terms of mg/g dry weight substrate and normalized to hopane (mg analyte/mg hopane). All experimental units will be housed in a constant temperature room with the temperature set at 15 °C to mimic the average summer water temperature in PWS to which the beaches are exposed.

7.0 SCHEDULE

7.1 Project Milestones

The objective is to conduct a microcosm study to determine if the lingering oil in selected PWS sites is sufficiently biodegradable to warrant a field bioremediation effort.

- Boat chartered for collection of samples in PWS by June 1, 2008.
- Ordering supplies and lab setup of microcosms completed by July 1, 2008.
- Samples collected and shipped to Cincinnati by July 31, 2008.
- Microcosm study to begin when samples are received.

- Sacrificial sampling and analysis of microcosms for remaining oil at selected time intervals as written in the body of the proposal.
- Monthly reports from contractor to EPA.
- Quarterly reports to the Trustee Council.
- Experimental phase ends February 28, 2009.
- Final report completed by June 1, 2009.

7.2 Measurable Project Tasks

FY 08, Third Quarter (April 1, 2008 – June 30, 2008)

- June 5, 2008: Notification of funding by Trustee Council.
- June 15, 2008: All equipment and supplies ordered, quality assurance plan written
- July 1: Support boat and captain identified and contracted.

FY 08, Fourth Quarter (July 1, 2008 – September 30, 2008)

- July 3: Samples collected and shipped to Cincinnati.
- July 10: Microcosm study begins when samples arrive at Contractor's facility, Time 0 samples sacrificed.

FY 09, First Quarter (October 1, 2008 – December 31, 2008)

- October 1: Continue scheduled sacrificial sampling of microcosms.

FY 09, Second Quarter (January 1, 2009 – March 31, 2009)

- February 28: Final sampling event completed.

FY 09, Third Quarter (April 1, 2009 -- June 30, 2009)

- April 30: All chemical analyses completed.
- June 1: Final report draft written and submitted to Trustee Council.

FY 09, Fourth Quarter (July 1, 2009 -- September 30, 2009)

- August 31: submit a manuscript to a peer-reviewed journal after review by the team members.
- Circulate draft final report for comments.
- September 30: Submit final report to trustee council.

8.0 RESPONSIVENESS TO KEY TRUSTEE COUNCIL STRATEGIES

8.1 Community Involvement

How will affected communities be informed about the project and be given an opportunity to provide their input? Since this is a laboratory study and not a field study, it will not be necessary for local communities to provide their input.

How will research findings and other project information be communicated to local communities? Fact sheets and other summary materials will be written and made available to local communities and environmental coordinators. The Alaska Department of Environmental Conservation (ADEC) has staff experienced in risk communications with the public in Alaska. Reliance will be made on ADEC's resources for public communication as well as those available to other Alaskan collaborators.

To what extent will local hire be used for the acquisition of such things as vessels, technicians, and equipment? Our needs include a boat, captain, and crew plus at least two field technicians to help with setting up, sampling, and disassembly at the completion of the project. All these people will be local hires.

To what extent will traditional and local knowledge be incorporated into the project? We are relying not only on collaboration with two other research teams (J. Michel and J. Short, and Boufadel *et al*.) involving finding the lingering oil and determining the limiting factors that affect oil persistence, but also on the knowledge of the local people who have their own unique experience with the location of the oil and the layout of the area. We will solicit this knowledge through the EVOSTC outreach program.

8.2 Resource Management Applications: A Team Collaboration

This microcosm study is but one aspect of a series of studies being conducted in PWS to determine the cause of the lingering oil 19 years after the Exxon Valdez spill. The Boufadel *et al*. "Limiting Factors" study, headed up by our colleague from Temple University, Dr. Michel C. Boufadel, was designed to conduct a tracer study to learn about the hydrodynamics of various PWS beaches so that nutrients and possibly oxygen can be applied to stimulate oil degrading microbes to metabolize the weathered oil hydrocarbons. If the microcosm study proposed herein is successful in demonstrating that the oil in samples from various sites in PWS is still biodegradable, this finding coupled with the successful determination that environmental limitations have played a major role in explaining why oil has lingered in PWS beaches for 19 years (the Boufadel *et al*. "Limiting Factors" study) will provide quantitative evidence that bioremediation is a feasible strategy for removing the lingering oil cost-effectively and eventually restoring the affected areas for a full recovery of the biological resources that have been negatively impacted by the spill. Before implementing any restoration/remediation effort, it is critical to understand not only the limiting factors that cause oil persistence, but also to determine if the oil that is still persistent is not sufficiently weathered to eliminate the need for further treatment. Both aspects are equally important for recommending further restoration measures. That is why we selected a range of weathering levels to be tested in this study. This will be an important outcome for all area resource managers and communities, who want the lingering oil removed.

In Venosa *et al*. (1996), we investigated the biodegradation of an experimental oil spill on a beach in Delaware. More recently, we have developed a model with design nomographs (a two-dimensional diagram designed to allow the approximate graphical computation of a function) that describe how much and how often nutrients need to be applied (and the method of applying them) to maintain a level that would result in maximum biostimulation. The methodology requires knowledge of the hydrodynamics of groundwater and tidal flow on shorelines, and we have a unique knowledge of these important factors and interactions. Drs. Jacqui Michel and Jeff Short have conducted and still are conducting extensive research in PWS in regards to geomorphology and mapping levels of contamination lingering in PWS beaches, and we will be relying on their expertise to guide us in finding the best locations to obtain the samples needed for the microcosm study. In fact, the work conducted by Drs. Boufadel, Short, Michel, and Venosa in separate but related studies (both in the lab and in the field) is a multidisciplinary team effort aimed at answering the question of the biological treatability of the PWS lingering oil. Thus, due to our combined expertise and experience, we are confident that the proposed microcosm investigation in combination with the field tracer study and the additional survey of selected sites in PWS will answer the important questions raised. After completing these studies, we expect to be able to make a recommendation regarding a field application of nutrients and/or oxygen for the purpose of stimulating the biodegradation of remaining oil. We can claim this because we already know how to implement nutrient application methodology on exposed and sheltered marine shorelines, having done extensive research in the past in the field and the laboratory (wave tanks) and through hydrodynamic modeling (Li et al., 2007).

9.0 PUBLICATIONS AND REPORTS

The annual report for FY09 will be submitted on May 15, 2009. The draft final report will be submitted to the Trustee Council by June 1, 2009. The revised final report will be submitted by August 15, 2009. At least one peer-reviewed journal publication is expected from this study. It will address the biodegradability of the lingering oil found on PWS beaches and help determine if further cleanup is justified.

10.0 REFERENCES

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11.0 DATA MANAGEMENT AND QA/QC STATMENT

11.1 Study Design

The objective of this research is to determine the biodegradability of residual oil contamination in intertidal beaches of PWS that remain contaminated with oil from the Exxon Valdez oil spill. This will involve collecting oiled sediment samples and seawater from selected beach sites, homogenizing the substrate samples, shipping them under ice to Cincinnati, and setting up a series of microcosms for the study of natural attenuation and biostimulation of the residual oil.

The data collected in support of this objective will include measurement of oil using both GC/MS for specific hydrocarbons and GC/FID for TPH and nutrients with time. Due to the heterogeneity of each microcosm, sub-sampling of the microcosms is not possible. Therefore, triplicate microcosms will be sacrificed at appropriate sampling events and the entire contents will be extracted for hydrocarbon analysis. All resolved hydrocarbons will be normalized to hopane to help reduce expected heterogeneity and to allow the hopane to serve as a conservative biomarker. Aqueous phase nitrate will be analyzed once per week in three randomly selected nutrient-amended microcosms to assess the degree to which nutrients will need to be re-supplied. The experiment is a factorial design with three replicates of all treatments.

11.2 Sample Handling, Preservation, and Storage

At the specified time points, the microcosms will be sacrificed and extracted with DCM. The serum bottles containing the DCM extracts will be weighed and then stored at 5 °C until time of analysis. Prior to preparing the vials for the instrument, the weight of the serum bottles will be checked for loss on storage. If the weights are consistent, the integrity of the solution can be assumed. If not, the cause will be investigated. Nutrient samples will be filtered through 0.45 μm syringe filter immediately after collection. If the samples are not analyzed within 24 hours, they will be stored in a freezer.

11.3 Calibration Procedures

All hydrocarbon measurements will be made from DCM extracts of the substrate. The GC/FID will be calibrated using ANS 521 oil diluted in DCM to generate an external five point calibration curve for TPH. The GC/MS used to measure specific hydrocarbons will be calibrated using commercially available standards to generate an external five or six point calibration curve. The relative response factor (RRF) for each compound is calculated relative to its corresponding deuterated internal standard. The relative standard deviation (RSD%) of the RRFs for each compound should be less than 25%. If it is not, the instrument will be recalibrated prior to analysis of samples.

During the extraction procedure, a surrogate mixture containing seven deuterated components will be added to evaluate the extraction efficiency. An internal standard solution with nine deuterated components will be used to evaluate GC/MS response. Prior to use, the GC/MS tune conditions will be verified using a DFTPP check standard to check the mass measuring accuracy of the MS, the resolution sensitivity, the baseline threshold, and the ion abundance ranges.

All measurements will be within the calibrated concentration range. If the measured concentration of any critical compound is above the calibration range, the sample will be diluted and reanalyzed to quantify that particular compound within the linear calibration range. If the measured concentration of any critical compound is below the calibrated concentration range, the extract may be concentrated and reanalyzed.

A check standard will be analyzed every 10 samples to ensure the instrument is still operating properly. For the GC/MS, if more than 5 of the nearly 80 compounds are not within ± 25% of the expected value, the samples will be reanalyzed. For both hydrocarbon analyses, an analytical duplicate will be analyzed every 10 samples and blanks will be analyzed to ensure there are no interfering peaks.

Nitrate and phosphate analyses will be performed by ion exchange chromatography. External five point calibration curves will be generated using commercially available standard solutions. Check standards will be measured every 10 samples. One sample will be analyzed in duplicate every 10 samples. Matrix interferences will be evaluated by spiking at least one sample out of every 20 with a known mid-range concentration of the analyte. One reagent blank will be analyzed per 20 samples.

Ammonia will be analyzed using an ion specific probe. An external five point calibration curve will be generated using standards that have been prepared in artificial seawater to eliminate the matrix effect of the high ionic strength. Check standards, duplicates and matrix spikes will be similar to the other nutrient analytes.

11.4 Data Reduction and Reporting

GC/MS data files are generated by MSChemStation for each injection. Data files contain total ion chromatograms and selected ion chromatograms. Calibration curves are generated within MSChemStation software, and all data files are calculated against the calibration curve by MSChemStation. Concentrations of all compounds and recovery standards and response area for internal standards are added to a ChemStation database that can be opened in Excel. Data verification will be done by cross-checking between analysts for 10% of raw data and its reduction process. Similarly FID Chemstation generates similar data files for every injection.

At the termination of the microcosm study, GC/MS and GC/FID results will be collated on Excel spreadsheets for data analysis. All hydrocarbon data will be normalized to hopane, but the non-normalized data will also be reported on the same spreadsheet. First order biodegradation rate coefficients for each individual compound and the sum of all alkanes and

PAHs will be calculated using non-linear regression analysis for each treatment at each site. Analyses of variance (ANOVAs) will be conducted at each sampling event to ascertain statistical differences among treatments. Where differences exist, a Least Significant Difference (LSD) analysis will be conducted to determine mean separation. Non-linear regression slopes and intercepts will be analyzed for statistical differences among treatments. Similar treatment will be given to the GC/FID results.

ABBREVIATED CURRICULUM VITAE

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PROFESSIONAL EXPERIENCE

- Program Manager, Oil Spill Research Program, 1990 to present, U.S. EPA, Remediation and Restoration Branch (and predecessor branches), Land Remediation and Pollution Control Division (LRPCD), National Risk Management Research Laboratory (NRMRL), Cincinnati
- Team leader, 1989 Alaska Oil Spill Bioremediation Project involved with assessing effectiveness of nutrient formulations in the field and in 1990 led an independent analysis of commercial inocula in multiple field plots
- Program Co-manager, Biosystems Technology Development Steering Committee, 1988 to 1997, U.S. EPA, Remediation and Restoration Branch (and predecessor branches), LRPCD, NRMRL, Cincinnati
- Program Manager, Sludge Pathogen Program, 1986 to 1990, U.S. EPA, Sludge Technology Section, Municipal Wastewater Branch, Water and Hazardous Waste Treatment Research Division, Risk Reduction Engineering Laboratory, Cincinnati
- Program Manager, Municipal Wastewater Disinfection Program, 1975 to 1986, U.S. EPA, Wastewater Treatment Division, Water Engineering Research Laboratory, Cincinnati
- Research Microbiologist, 1968-1975, U.S. EPA and predecessor agencies, Municipal Wastewater Disinfection Program
- Research Fellow, 1967-1968, University of Cincinnati, Department of Civil and Environmental Engineering, Cincinnati.
- Part-Time Clinical Bacteriologist, 1966-1968, Central Bacteriology Laboratory, Cincinnati General Hospital (now University Hospital).

EDUCATION

- Doctor of Philosophy, Environmental Science, 1980, Department of Civil and Environmental Engineering, University of Cincinnati.
- Master of Science, Environmental Engineering, 1968, Department of Civil and Environmental Engineering, University of Cincinnati.
- Bachelor of Science, Microbiology, 1967, College of Arts and Sciences, University of Cincinnati.

RESEARCH INTERESTS AND EXPERTISE

- Development of alternative wastewater disinfection technologies for rendering POTW effluents safe for human exposure
- Pathogenic microbiological impact of spreading biosolids to land
- Development of protocols for testing the effectiveness of commercial bioremediation products for biodegrading crude oil or refined products in seawater, freshwater, beach sediments, wetlands, and soils.
- Development of methods for microbiological and chemical analysis of oil spill bioremediation activities.
- Development of improved scientific and practical understanding of the mechanisms of biodegradation of petroleum hydrocarbons, especially polycyclic aromatic hydrocarbons.
- Development of engineering strategies for bioremediating hydrocarbons contaminating the environment.
- Design and development of pilot- and field-scale research leading to the scientifically valid assessment of the effectiveness of bioremediation technologies for oil spill cleanup on marine and freshwater shorelines, wetlands, and terrestrial environments.
- Development of protocols for testing the effectiveness of chemical oil dispersants and surface-washing agents for use in regulatory rule-making.
- Pilot-scale (wave tank) testing of dispersant effectiveness as a function of energy dissipation rate.
- Development of strategies for treating spills of biofuels (various blends of biodiesel and ethanol/gasoline).
- Bioremediation of oxygenates such as MtBE, tBA, tBF, EtBE, DIPE, DEE, tAME, etc. in the presence and absence of ethanol and/or BTEX.
- Development of membrane bioreactor technology for the treatment of recalcitrant compounds.
- Development and assessment of flow diversion technology to completely contain LNAPL hot-zones for *ex-situ* and/or *in-situ* treatment, protecting the groundwater aquifer for downstream human consumption.

PROFESSIONAL ASSOCIATIONS/COMMITTEES

- American Society for Microbiology
- Society for Industrial Microbiology
  - Awards Committee (1993-1999)
- Water Environment Federation
  - Chairman, Disinfection Committee, 1987-1989
  - Research Committee, Water Pollution Control Federation, 1983-1988.
- Chairman, Pathogen Equivalency Committee, U.S. EPA-Cincinnati, 1990-1991
- Member of National Environmental Technology Applications Center (NETAC) panel of experts on effectiveness of bioremediation agents (1990 to 1998)
- Member of International Working Group on Guidelines for Decision Making and Implementation of Bioremediation on Shorelines, 1995 to 2003

- Participating member of Science Advisory Group for the European Commission's Project on a Field Evaluation of Oil Spill Bioremediation Strategies for Salt Marshes and Mudflats, 1995 to 2004
- Member of Science Advisory Board of the Texas General Land Office-Sponsored San Jacinto Oil Spill Bioremediation Project, 1994 to 1996
- Member of Science Advisory Board of the Texas General Land Office-Sponsored Coastal Oilspill Simulation System, 1994 to 1996
- Exhibit Chairman for the International Oil Spill Conference, 1997 to present
- Member of the Technical Advisory Committee for the New York State Center for Hazardous Waste Management at the State University of New York at Buffalo, 1995 to 1998
- Chairman of Oil Spill Subcommittee of the Bioremediation Action Committee, 1996 to 2000
- Member of the Louisiana Applied Oil Spill Research and Development (OSRADP) Advisory Panel, 1998 to present
- Member of the Science Advisory Committee for the Integrated Petroleum Environmental Consortium, 1999 to 2007
- Member of the Remediation Technologies Development Forum Phytoremediation Action Team for Hydrocarbons in Soil, 1998 to 2005
- Served as a member of M.S. committee for numerous graduate students at the University of Cincinnati (Mr. Todd Herrington, Mr. Kevin Strohmeier, Mr. Salil Pradhan, Mr. Jaydeep Purandare, Ms. Xiaoming Du, Mr. Chang-Hoon Ahn, Ms. Nita Naik, Mr. Christopher Luedeker, Mr. Mohamed Yasin, Mr. Ali Midella)
- Served or serving as a member of Ph.D. committee for several graduate students at the University of Cincinnati (Dr. Maher Zein, Dr. Michel Boufadel, Dr. Jiayang Cheng, Dr. Arturo A. Burbano, Dr. Marie Sedran, Dr. Amy Pruden, Dr. Karen M. Koran, Mr. Qiang Zhang, Mr. Seungjoon Chung, Ms. Darin Salam, Mr. Jai-Ho Cho, Mr. Pablo Campo-Moreno)

WITHIN AGENCY WORK GROUPS

- Lectured on bioremediation every year since 2001 at the Annual On-Scene Coordinators Training sponsored by the U.S. EPA’s Oil Program Center and Office of Emergency Management.
- Developed scientifically sound protocols for screening effective dispersant and bioremediation agent products for the Oil Spill program office. Protocols will be used in new regulatory rule-making guidelines.
- Wrote the ORD oil spill multi-year plan in coordination with the Oil Program Center.
- Helped write the Leaking Underground Storage Tank multi-year research plan for ORD.
- Served as Chairman of NRMRL’s Technical Qualifications Board (TQB) for GS 14 and 15 scientists in 2003 and Vice-Chairman in 2002. More recently (2008), served on TQB panel for two ST positions in ORD.

PROFESSIONAL SERVICE

Frequent Reviewer for:

- Environmental Science and Technology
- Journal of Industrial Microbiology and Biotechnology (editorial board)
- Bioremediation Journal (editorial board)
- Applied and Environmental Microbiology
- Journal of Environmental Engineering, American Society of Civil Engineers
- Canadian Journal of Microbiology
- Biodegradation
- Environmental Technology
- Journal of Water, Air, and Soil Pollution
- Journal of Hazardous Materials
- Enzyme and Microbial Technology
- Marine Pollution Bulletin
- Pedosphere
- Chemosphere
- Arctic Marine Oilspill Program
- Freshwater Spills Symposium Proceedings
- Marine Chemistry
- Journal of Environmental Quality

HONORS AND AWARDS

- 1990, Awarded Gold Medal, U.S. Environmental Protection Agency, for exemplary leadership in the Alaska Oil Spill Bioremediation Project.
- 1990 to 2007, Received Outstanding rating for continued superior performance on his job.
- May 1991 and April 1996, Nominated for Federal Employee of the Year Award by the U.S. EPA Risk Reduction Engineering Laboratory and NRMRL, respectively.
- 1992, Awarded Scientific and Technological Achievement Reward (STAR) for a published paper on the Alaskan Oil Spill Project.
- 1992, Awarded Scientific and Technological Achievement Reward (STAR) for a published paper on reduction in bacterial densities in wastewater solids.
- 1992, Awarded a Scientific and Technological Achievement Award (STAA) for a published paper on the Alaskan Oil Spill Project.
- 1998, Awarded Bronze Medal, U.S. Environmental Protection Agency, for continued superior performance as a member of EPA's Biosystems Committee.
- 2003, Awarded a Level 3 Scientific and Technological Achievement Award (STAA) for a published paper on the first definitive field study of hydrocarbon biodegradation in a freshwater wetland where significant oil penetration into the sediment has taken place.
- 2004, Awarded Bronze Medal, U.S. Environmental Protection Agency, for significant achievements in working with program offices to promote the use of strong science in Agency decisions.

- 2006, Scientific and Technological Achievement Award, Honorable Mention for manuscript entitled "Laboratory evaluation of oil spill bioremediation products in salt and freshwater systems."
- 2007, Awarded a plaque for exceptional/outstanding ORD technical assistance to the regions or program offices as part of the Leaking Underground Storage Tank Management Team.

PATENTS

2004, Co-developed an *ex-situ* membrane bioreactor (Biomass Concentrator Reactor, Patent No. 6,821,425) for treating groundwater contaminated with gasoline hydrocarbons and MtBE and for use in municipal and industrial wastewater treatment, phosphate removal, and drinking water treatment.

RECENT PUBLICATIONS

Peer-Reviewed Journal Articles

Boufadel, M.C., H.L. Li, M.T. Suidan, and A.D. **Venosa**. 2007. "Tracer Studies in a Laboratory Beach Subjected to Waves," Journal of Environmental Engineering-ASCE, 133(7): 722-732.

Boufadel, M.C., M.T. Suidan, A.D. **Venosa.** 2006. "Tracer Studies in Laboratory Beach Simulating Tidal Influences," Journal of Environmental Engineering-ASCE, 132(6): 616-623.

Campo, P., G.A. Sorial, M.T. Suidan and A.D. **Venosa**. 2006. "Statistical Evaluation of an Analytical GC/MS Method for the Determination of Long Chain Fatty Acids," Talanta, 68: 888-894.

Campo-Moreno, P, Y. Zhao, M.T. Suidan, and A.D. **Venosa**. 2007. "Biodegradation Kinetics and Toxicity of Vegetable Oil Triacylglycerols under Aerobic Conditions," Chemosphere, 68(11): 2054-2062.

Cho, J.H., M.M. Zein, M.T. Suidan, and A.D. **Venosa**. 2007. "Biodegradability of Alkylates as a Sole Carbon Source in the Presence of Ethanol or BTEX," Chemosphere, 68(2): 266-273.

Garcia-Blanco, S., A.D. **Venosa**, M.T. Suidan, K. Lee, S. Cobanli. 2006. "Biostimulation for the Treatment of an Oil-Contaminated Coastal Salt Marsh." Biodegradation, 18, 1 (2007): 1-15.

Haines, J.R., E.J. Kleiner, K.A. McClellan, K.M. Koran, E.L. Holder, D.W. King, and A.D. **Venosa**. 2005. "Laboratory evaluation of oil spill bioremediation products in salt and freshwater systems." J. Ind. Microbiol. Biotech. 32:171-185.

Haines, J.R., K.M. Koran, E.L. Holder, and A.D. **Venosa**. 2003. "Protocol for laboratory testing of crude-oil bioremediation products." J. Ind. Microbiol. Biotechnol. 30:107-113.

Kaku, V.J., M.C. Boufadel, and A.D. **Venosa**. 2006 . "Flow Dynamics in Eccentrically Rotating Flasks Used for Dispersant Effectiveness Testing." Environmental Fluid Mechanics, 6: 385–406, 2006

Kaku, V.J., M.C. Boufadel, and A.D. **Venosa**. 2006. Evaluation of Mixing Energy in Laboratory Flasks Used for Dispersant Effectiveness Testing. ASCE J. Environmental Eng. Div., 132(1):93-101.

Lee, K. A.D. **Venosa**, M.T. Suidan, C.W. Greer, G. Wohlgeschaffen, C. Cobanli, G.H. Tremblay, J. Gauthier, and K.G. Doe. 2002. "Monitoring recovery of a crude oil-contaminated saltmarsh following in-situ remediation treatments." In: C.A. Brebbia (ed.), Coastal Environment: Environmental Problems in Coastal Regions IV, WIT Press, Southampton, UK.

Li, Z., B.A. Wrenn, and A.D. **Venosa**. 2005. "Anaerobic biodegradation of vegetable oil and its metabolic intermediates in oil-enriched freshwater sediments." Biodegradation 16:341-352.

Li, Z., B.A. Wrenn, and A.D. **Venosa**. 2005. "Effect of iron on the sensitivity of hydrogen, acetate, and butyrate metabolism to inhibition by long-chain fatty acids in vegetable-oil-enriched freshwater sediments." Water Research 39(13): 3109-3119.

Li, Z. and B.A. Wrenn. 2004. "Effects of ferric hydroxide on the anaerobic biodegradation kinetics and toxicity of vegetable oil in freshwater sediments." Water Research 38:3859-3868.

Lin, Q., I.A. Mendelssohn, M.T. Suidan, K. Lee, and A.D. **Venosa**. 2002. "The dose-response relationship between No. 2 fuel oil and the growth of the salt marsh grass, Spartina alterniflora." Marine Poll. Bulletin, 44:897-902.

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Pruden, A., M. Sedran, M.T. Suidan, and A.D. **Venosa**. 2003. Biodegradation of MTBE and BTEX in an Aerobic Fluidized Bed Reactor. Water Sci. & Technol. 47(9):123-128.

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LIST OF COLLABORATORS OVER LAST 48 MONTHS

K. Lee (Fisheries and Oceans Canada, Dartmouth, NS)
Z. Li (Fisheries and Oceans Canada, Dartmouth, NS)
P. Kepkay (Fisheries and Oceans Canada, Dartmouth, NS)
M. Boufadel (Temple University, Philadelphia, PA)
B. Wrenn (Washington University, St. Louis)
M. Suidan (University of Cincinnati, Cincinnati, OH)
J. Michel (Research Planning, Inc., Columbia, SC)
J. Mullen (U.S. Minerals Management Service, Herndon, VA)
J. Short (NOAA, Auke Bay, AK)
J. Weaver (U.S. EPA, Athens, GA).

EXXON VALDEZ OIL SPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 07 - FY 09

Budget Category:	Proposed FY 08	Proposed FY 09	Proposed FY 010		TOTAL PROPOSED
Personnel	$33,528.5	$172,718.0	$0.0		$206,246.5
Travel	$0.0	$3,000.0	$0.0		$3,000.0
Contractual	$36,900.0	$3,000.0	$0.0		$39,900.0
Commodities	$18,000.0	$28,000.0	$0.0		$46,000.0
Equipment	$46,800.0	$0.0	$0.0		$46,800.0
Subtotal	$135,228.5	$206,718.0	$0.0		$341,946.5
Indirect (rate will vary by contractor)	$31,501.0	$118,271.0			$149,772.0
Net Requested Funds	$166,729.5	$324,989.0			
General Administration (9% of Subtotal)	$15,005.7	$29,249.0	$0.0		$44,254.7
Project Total	$181,735.2	$354,238.0	$0.0		$535,973.2
Other Resources:	$2,043.0	$14,508.0			

Cost-Sharing by U.S. EPA

FY 08

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Name of Contractor: Pegasus Technical Services

EXXON VALDEZ OIL SPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 07 - FY 09

Personnel Costs:				Months	Monthly		Personnel
	Name	Description		Budgeted	Costs	Overtime	Sum
	Dr. Makram T. Suidan			1.5	5282.0		7,923.0
	Mr. Daniel Radigan			1.5	4189.0		6,283.5
	Dr. Pablo Campo-Moreno			1.5	7107.0		10,660.5
	Mr. Seungjoon Chung			1.5	3025.0		4,537.5
	Ms. Melody Pennington			1.5	706.0		1,059.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
		Subtotal		7.5	20309.0	0.0	
						Personnel Total	$30,463.5

Travel Costs:		Ticket	Round	Total	Daily	Travel
	Description	Price	Trips	Days	Per Diem	Sum
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
					Travel Total	$0.0

FY 08

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Name of Contractor: Pegasus Technical Services

EXXON VALDEZ **OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 07 - FY 09

Contractual Costs:	Contractual
Description	Sum
Subaward (RPI, Inc) for sampling in PWS	36,900.0
Contractual Total	$36,900.0

Commodities Costs:	Commodities
Description	Sum
Materials & Supplies	18,000.0
Commodities Total	$18,000.0

FY 08

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Name of Contractor: Pegasus Technical Services

New Equipment Purchases:	Number	Unit	Equipment
Description	of Units	Price	Sum
Multichannel peristaltic pumps for microcosms	18	2600.0	46,800.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
		New Equipment Total	$46,800.0

Existing Equipment Usage:	Number	
Description	of Units	

FY 08

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Proposer: Dr. Albert D. Venosa - EPA

EXXON VALDEZ OIL SPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 07 - FY 09

Personnel Costs:				Months	Monthly		Personnel
	Name	Description		Budgeted	Costs	Overtime	Sum
	Dr. Albert D. Venosa			1.5	2043.0		3,064.5
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
		Subtotal		1.5	2043.0	0.0	
						Personnel Total	$3,065.0

Travel Costs:		Ticket	Round	Total	Daily	Travel
	Description	Price	Trips	Days	Per Diem	Sum
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
					Travel Total	$0.0

FY 08

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Proposer: Dr. Albert D. Venosa - EPA

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 07 - FY 09

Contractual Costs:	Contractual
Description	Sum
Contractual Total	$0.0

Commodities Costs:	Commodities
Description	Sum
Commodities Total	$0.0

FY 08

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Proposer: Dr. Albert D. Venosa - EPA

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 07 - FY 09

New Equipment Purchases: Description	Number of Units	Unit Price	Equipment Sum
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
Indicate replacement equipment with an R.		**New Equipment Total**	$0.0

Existing Equipment Usage: Description	Number of Units

FY 09

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Name of Contractor: Pegasus Technical Services

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 07 - FY 09

Personnel Costs:			Months	Monthly		Personnel
Name	Description		Budgeted	Costs	Overtime	Sum
Dr. Makram T. Suidan			10.5	3579.4		37,583.7
Mr. Daniel Radigan			10.5	1427.2		14,985.6
Dr. Pablo Campo-Moreno			10.5	6421.4		67,424.7
Mr. Seungjoon Chung			10.5	2528.5		26,549.3
Ms. Melody Pennington			10.5	419.7		4,406.9
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
	Subtotal		52.5	14376.2	0.0	
					Personnel Total	$150,951.0

Travel Costs:	Ticket	Round	Total	Daily	Travel
Description	Price	Trips	Days	Per Diem	Sum
Dr. Makram T. Suidan					1,500.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
				Travel Total	$1,500.0

FY 09

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Name of Contractor: Pegasus Technical Services

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 07 - FY 09

Contractual Costs:	Contractual
Description	Sum
Subaward (QA for Shaw Environmental, Inc.)	3,000.0
Contractual Total	$3,000.0

Commodities Costs:	Commodities
Description	Sum
Materials & Supplies	28,000.0
Commodities Total	$28,000.0

FY 09

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Name of Contractor: Pegasus Technical Services

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 07 - FY 09

New Equipment Purchases: Description	Number of Units	Unit Price	Equipment Sum
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
		New Equipment Total	$0.0

Existing Equipment Usage: Description	Number of Units	

FY 09

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Proposer: Dr. Albert D. Venosa - EPA

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 07 - FY 09

Personnel Costs:				Months	Monthly		Personnel
	Name	Description		Budgeted	Costs	Overtime	Sum
	Dr. Albert D. Venosa			10.5	2073.0		21,766.5
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
		Subtotal		10.5	2073.0	0.0	
						Personnel Total	$21,767.0

Travel Costs:		Ticket	Round	Total	Daily	Travel
	Description	Price	Trips	Days	Per Diem	Sum
	Dr. Albert D. Venosa					1,500.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
					Travel Total	$1,500.0

FY 09

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Proposer: Dr. Albert D. Venosa - EPA

Contractual Costs:	Contractual Sum
Description	
Contractual Total	$0.0

Commodities Costs:	Commodities Sum
Description	
Commodities Total	$0.0

FY 09

Project Number:
Project Title: Microcosm Study on the Biodegradability of Lingering Oil in PWS
Proposer: Dr. Albert D. Venosa - EPA

New Equipment Purchases:	Number	Unit	Equipment
Description	of Units	Price	Sum
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
Indicate replacement equipment with an R.		**New Equipment Total**	$0.0

Existing Equipment Usage:	Number	
Description	of Units	

FY 10

Project Number:
Project Title:
Contractor:

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 07 - FY 09

Personnel Costs:				Months	Monthly		Personnel
	Name	Description		Budgeted	Costs	Overtime	Sum
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
		Subtotal		0.0	0.0	0.0	
						Personnel Total	$0.0

Travel Costs:		Ticket	Round	Total	Daily	Travel
	Description	Price	Trips	Days	Per Diem	Sum
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
					Travel Total	$0.0

FY 10

Project Number:
Project Title:
Contractor:

EXXON VALDEZ OIL SPILL TRUSTEE COUNCIL
DETAILED BUDGET FORM FY 07 - FY 09

Contractual Costs:	Contractual
Description	Sum
Contractual Total	$0.0

Commodities Costs:	Commodities
Description	Sum
Commodities Total	$0.0

FY 10

Project Number:
Project Title:
Contractor:

New Equipment Purchases: Description	Number of Units	Unit Price	Equipment Sum
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
Indicate replacement equipment with an R.		**New Equipment Total**	$0.0

Existing Equipment Usage: Description	Number of Units	

FY 10

Project Number:
Project Title:
Proposer:

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 07 - FY 09

Personnel Costs:				Months	Monthly		Personnel
	Name	Description		Budgeted	Costs	Overtime	Sum
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
		Subtotal		0.0	0.0	0.0	
						Personnel Total	$0.0

Travel Costs:		Ticket	Round	Total	Daily	Travel
	Description	Price	Trips	Days	Per Diem	Sum
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
					Travel Total	$0.0

FY 10

Project Number:
Project Title:
Proposer:

Contractual Costs:	Contractual
Description	Sum
Contractual Total	$0.0

Commodities Costs:	Commodities
Description	Sum
Commodities Total	$0.0

FY 10

Project Number:
Project Title:
Proposer:

***EXXON VALDEZ* OIL SPILL TRUSTEE COUNCIL**
DETAILED BUDGET FORM FY 07 - FY 09

New Equipment Purchases: Description	Number of Units	Unit Price	Equipment Sum
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
Indicate replacement equipment with an R.		**New Equipment Total**	$0.0

Existing Equipment Usage: Description	Number of Units	