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OSEI Corporation Summary of the US EPA Regional Response

Team VII Testing of OSE II on Heavy Waste Oil

February 1st to March 8th, 2012

The US EPA RRT VII tested Oil Spill Eater II (OSE II) on a very heavy, viscous waste oil utilizing fresh water from two different sources: the Missouri River and the Blues Springs Lake. Heavy oil such as this generally forms a heavy emulsion and is difficult to break down based on its normal properties; however, waste oil will include additives/preservatives and potentially teflons from the refining process to help prevent the oil from breaking down in engines, and to add oil life so engines need fewer oil changes and have easier oil flow. Because of the additives, this type of oil is much harder to remediate than fresh crude oil.

There were eight 10-gallon aquariums utilized. Four were filled with Missouri River fresh water and four were filled with fresh water from Blue Springs Lake. Equal amounts of the heavy waste oil was poured onto the surface of each aquarium. Two aquariums with Missouri River fresh water had OSE II applied to them, and two aquariums

with Blue Springs Lake fresh water had OSE II applied. An aerator was placed in one of the Missouri River aquariums with OSE II, and an aerator was placed in one aquarium with Blue Springs Lake with OSE II.

The oil in the aquariums with OSE II turned a brownish color and thinned out until the layer of oil contained just a remnant of the oil. The OSE II aquariums showed a large reduction of the waste oil - 72.5% reduction on average in the Diesel Range Organics (DRO), and the Oil Range Organics (ORO) showed an average reduction of 73.5%. One aquarium showed a reduction just over 60%, however there was a large spike for the last samples. Usually anomalies such as this are not considered.

The 4 other aquariums where OSE II was not applied, showed very little reduction of oil in three of the aquariums, and the fourth aquarium, (Blue Springs Lake water and no mechanical aeration) showed slightly more reduction of the oil than the one with air. This is usually not the case. The container with air generally shows more reduction than one without.

This test of heavy waste oil with OSE II applied showed conclusive evidence that OSE II remediated the oil to CO₂ and water, and based on the rate of biodegradation of the oil, given 10 to 14 days more, there would have been no oil left at all. This test shows that OSE II complies with the Clean Water Act where it states a response method must permanently remove oil from the environment.

INTERESTING NOTE: A comparative analysis of Tank 2 (treated with OSE II and no oxygen added) and Tank 4 (no OSE II added and no oxygen added doing nothing), shows that the dissolved oxygen end point in Tank 2 was 1.5 and in Tank 4 was .7. This demonstrates that OSE II does not deplete oxygen in the water column any more than the results of doing nothing at all. Therefore, it is the oil itself that is depleting the oxygen.

PHOTOGRAPHS TAKEN THROUGHOUT THE TESTING PROCEDURE OF OSE II ON BLUE SPRINGS LAKE AND MISSOURI RIVER WATER FOR THE US EPA REGION VII

The pictures below are numbered in the order they were received from the US EPA RRT VII (Regional Response Team VII). There were two types of waters used in a set of 8 aquariums: water from Blue Springs Lake, and water from the Missouri River. Waste oil was applied to each aquarium. OSE II was applied to 4 of the aquariums. 4 of the aquariums were used as controls. Each set of 4 had two with air added and 2 without air added. Each tank was numbered.

Waste oil was used as the test oil in each aquarium

Missouri River water

Tank 1: OSE II treated with aeration

Tank 2: OSE II treated with no aeration

Tank 3: Control with aeration

Tank 4. Control with No aeration

Blue Springs Lake water

Tank 5: OSE II treated with aeration

Tank 6: OSE II treated with no aeration

Tank 7: Control with aeration

Tank 8: Control with No aeration

PICTORIAL REPRESENTATION AND WHAT IT MEANS:

All the aquariums started out with semi-cloudy water, which is in line with water obtained from a river or lake. Had the cloudiness been due to sediment, after a short time the sediment would have settled and the water would have cleared. Based on the photographs taken. Based on the pictures taken, it took a few days for the aquarium's water to clear that did not have OSE II applied to the surface oil, indicating that the turbidity (cloudiness) of the water was caused by microbes living in the water.

The aquariums where OSE II was applied, with or without aeration, had the cloudiness (turbidity) increased. That shows that large amounts of bacteria were grown (colonized) to digest the oil. The pictures show the oil turning colors and then thinning out in the aquariums that had OSE II applied to them, until only remnants of the oil remained. Had the test continued for a few more days, the oil would have been completely removed.

The test results showed that OSE II reduced the total parts hydrocarbons (TPH - sometimes called total petroleum hydrocarbons, i.e., the total amount of oil in the aquarium) to a marked extent. OSE II reduced the DRO (lighter ends of the oil) on average 72.5% and the ORO (heavier ends of the oil) by 73.5% on average. This correlates with the pictorial observations of large amounts of bacteria being colonized or populated by OSE II (creating temporarily cloudy water during the process) and subsequently digesting the oil to CO₂ and water, reducing the depth and size of the oil layer, and permanently removing it from the environment.

The Control aquariums (the "control" tanks in this test had only oil poured into them and no OSE II) showed the comparison of what occurs if you do nothing at all to clean up an oil spill. The control tanks started out slightly cloudy when the water was initially poured in, but, and as time went by, the control aquariums became less cloudy because the toxicity of the oil that was poured into them killed off virtually all of the indigenous microbes. This is consistent with what occurs when there is a spill in the environment: the toxicity of the oil destroys or kills off living organisms, especially single-celled organisms, thus, impacting the entire eco system. As the bacteria died off, the water slowly cleared in the tanks, which means that when a spill happens in the environment, the oil will just linger for a protracted period of time while the toxicity of the oil continues to adversely effect the eco system.

According to the test results, all of the control tanks showed very little reduction in oil. However, tank 3 (with air and no OSE II) and tank 7 (with no air and no OSE II) showed more reduction in the test numbers than is biochemically possible, based on the photographs taken. The test numbers from these 2 tanks do not track with the photographs. It is impossible to have the level of reductions in oil that were reported and have a tank that is as clear as what the photographs show.

The following are pictures in time sequence of the EPA test on OSE II in February and March of 2012. They compare the results of using OSE II over doing nothing at all on waste oil. The difference in results is compelling for the use of OSE II. OSE II reduced, on average, 2.3 percent of the oil each day during the EPA's test period. Therefore, given approximately 10 to 14 more days, the oil in these aquariums would have been completely eliminated. These are great remediation results produced by OSE II on a difficult oil to remediate.

Photos #98, 99, 0-6: The aquariums all started with slightly cloudy water which is to be expected from fresh water samples just pulled from lakes or rivers, due to the bacteria living in the water.



98,

99



0,1





4 MISSOURI RIVER
NOT TREATED W OSEII
NOT AERATED-CONTROL

2,3



5 BLUE SPRINGS LAKE
TREATED WITH OSEII
AERATED



6 BLUE SPRINGS LAKE
TREATED WITH OSEII
NOT AERATED

4,5



7 BLUE SPRINGS LAKE
NOT TREATED W OSEII
AERATED-CONTROL



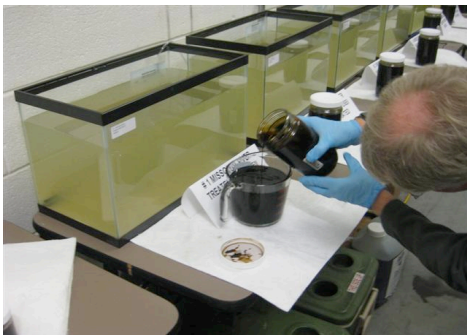
8 BLUE SPRINGS LAKE
NOT TREATED W OSEII
NOT AERATED-CONTROL

6,

The oil used for the test was a very heavy waste oil that tends to form emulsions, making it difficult to remediate. Waste oil generally contains preservatives and additives, including teflons, which are added during the refining process to prevent it from easily breaking down in an engine and to extend the life of the oil.

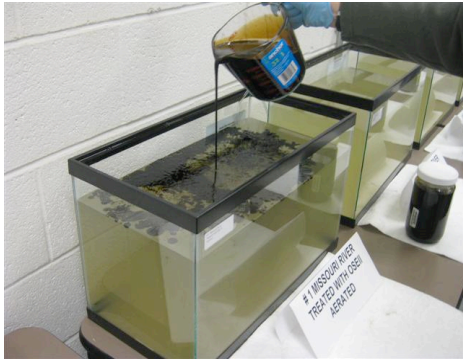


7,8



9,10





11,12



13 and 14 below show that every aquarium had oil added to it and the oil covered the entire surface of the water.



13,14



These photos show that the scientists carefully delineated between the aquariums as to which water environment was in that aquarium, whether or not it was being aerated, and which aquariums had OSE II added to them.

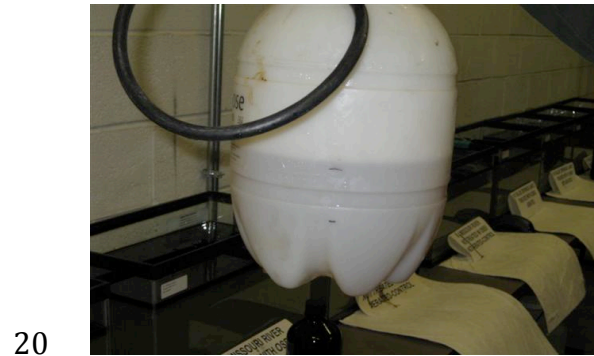


17,18



19,

This is the container in which OSE II was mixed with water at the standard 50 parts water to 1 part OSE II ratio.



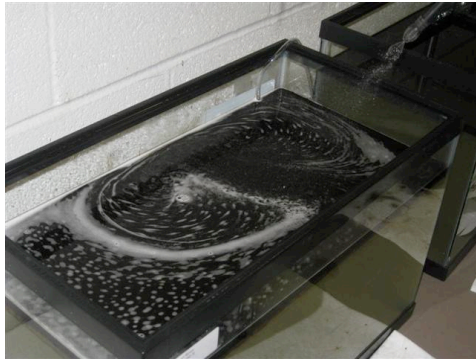
20

21 - 24 OSE II is sprayed on the oil in the aquariums.



21, 22





23, 24



25-29 - Upon application of OSE II you can see the immediate effects on the oil as OSE II starts the detoxification process. Detoxifying the oil as a first step is vital to protecting the indigenous bacteria so that they can begin the process of digesting it which eventually results in the defined end point of CO₂ and water.



25,26





5 BLUE SPRINGS LAKE
TREATED WITH OSEII

27,28



2 MISSOURI RIVER
TREATED WITH OSEII
NOT AERATED.



1 MISSOURI RIVER
TREATED WITH OSEII
AERATED

29,

30 - Note that the water in the tank where OSE II was not applied has not changed. This is because the oil is so toxic it is killing off the indigenous microbes so that there is no change happening to the oil.



31-53 The aquariums where there was aeration showed faster growth of bacteria than the non aerated; however, all the aquariums treated with OSE II show a marked increase in turbidity, or cloudiness, which means you have bacterial growth and the on-going digestion of the oil.



31,32





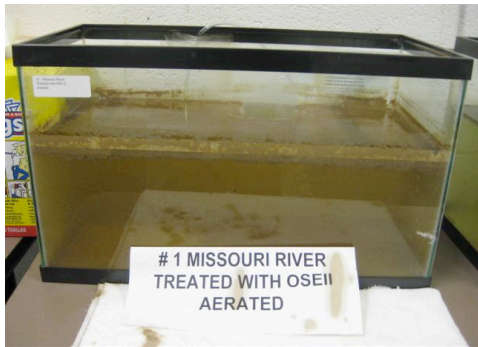
5 BLUE SPRINGS LAKE
TREATED WITH OSEII

33,34



7 BLUE SPRINGS LAKE
NOT TREATED W OSEII
AERATED-CONTROL

(pictures skipped to 50)



1 MISSOURI RIVER
TREATED WITH OSEII
AERATED

50,51



2 MISSOURI RIVER
TREATED WITH OSEII
NOT AERATED



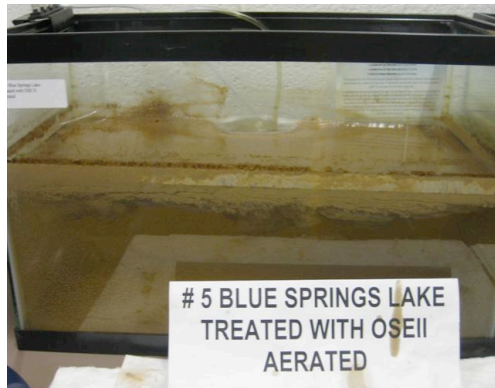
3 MISSOURI RIVER
NOT TREATED W OSEII
AERATED-CONTROL

52,53



4 MISSOURI RIVER
NOT TREATED W OSEII
NOT AERATED-CONTROL

54-71 The photos below include aquariums with Blue Springs Lake water, but even though this is lake water rather than river water, the same actions and reactions are occurring - the OSE II treated aquariums show the oil breaking down and becoming cloudy, and in the aquariums that are not treated with OSE II the water is becoming clear because the indigenous microbes are dying off due to the toxicity of the oil and the oil on the surface is unchanged.



54,55



56,57





1 MISSOURI RIVER
TREATED WITH OSEII
AERATED

58,59



2 MISSOURI RIVER
TREATED WITH OSEII
NOT AERATED



3 MISSOURI RIVER
NOT TREATED W OSEII
AERATED-CONTROL

60,61



4 MISSOURI RIVER
NOT TREATED W OSEII
NOT AERATED-CONTROL



62,63



64,65



66, and 67: These are simply showing these turning different colors as it is in different stages of breaking down.



66,67



Skipped 68,69



70,71



72-75 As the test time increased, the OSE II aquariums show less oil and more cloudiness, while the controls show about the same amount of oil and clearer water.



72,73



74,75



Pictures 76 treated with OSE II above, and picture 78 not treated with OSE II shows a dramatic difference in the oil layer and the cloudiness of the water.



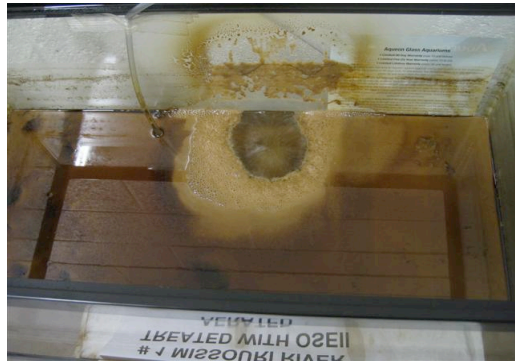
76,77



78,

NOTE: PHOTOGRAPHS 79 – 86 ARE TAKEN LOOKING DOWN INTO THE TOP OF THE TANK.

79 – The bacteria is remediating the oil (both heavier and lighter hydrocarbons), leaving a small circle of heavier oil outside still in the process of remediating, and a thin film that is left behind from the heavier hydrocarbons. The bacteria go after that as a last step in this tank's remediation process.



79

80-85 These photos show how nothing remediates when you do nothing.



80,

81 – This is very thin remnant of the waste oil that is in its last stages of being converted to CO2 and water as the manufacturer’s stated and predicted result of the application of OSE II.



81

82-85 These again show the results of doing nothing and leaving nature to its own devices to try to handle the toxic oil. These photos were taken from the top down into the tank, the black rim on top of the aquarium and the brick wall is being reflected by the black oil. oil



82,83





84,85

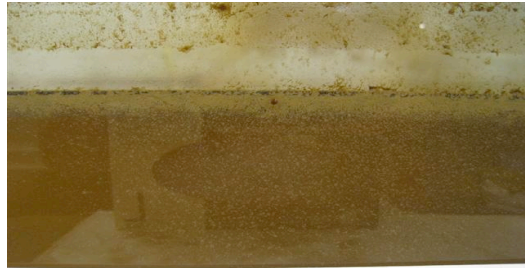


Pictures 79 and 81 treated with OSE II compared to pictures 80, 82 83 84, and 85 where OSE II was not applied shows a distinct difference from the top view of the oil. OSE II is reducing the amount of oil leaving the remnants of teflons and additives contained in the waste oil along with sediments. The control shows virtually no remediation.



86,

87 - 94 - These are side view pictures of the tanks, which show the oil layer reducing and the water being extremely turbid (cloudy), which shows that the bacteria is digesting the oil, as it remediates 100% of the oil to CO2 and water. Those that have not been treated with OSE II below (90 and 94) show no change in the oil layer, and because virtually all of the indigenous bacteria have been killed off by the oil, the water is completely clear.



87

1 MISSOURI RIVER
TREATED WITH OSE



2 MISSOURI RIVER
TREATED WITH OSE

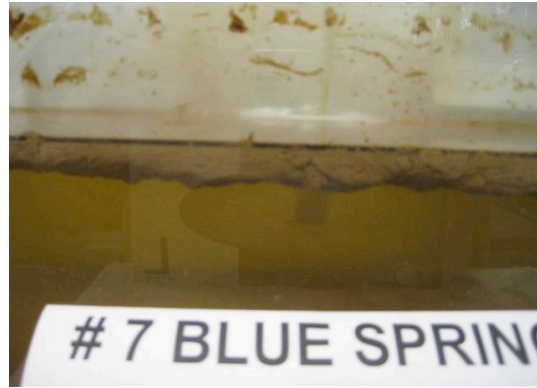


3 MISSOURI RIVER

88,89



90,91



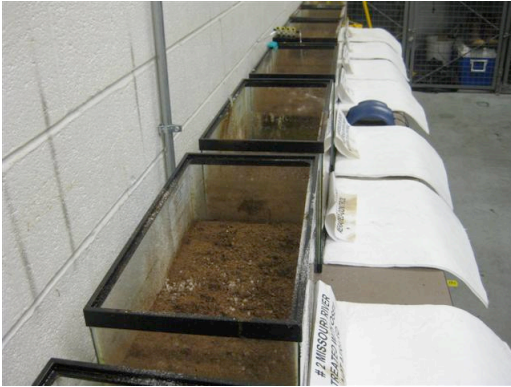
92,93



94,

95 shows tanks treated with OSE II with just remnants of the oil left after approximately 30 days. Had the test been allowed to continue for approximately 10 – 14 more days, per the rate of biodegradation, the oil would have been remediated by 100%.

95



Steven Pedigo

CEO OSEI CORP.

OIL SPILL EATER II TEST RESULTS

Tank 1 – Missouri River Water Treated With Oil Spill Eater II - Aerated

Date	Days after Start of Test	Readings with Water Quality Meter			Lab Results (mg/L)		Notes
		Temp (°C)	pH	DO (mg/L)	TPH-DRO	TPH-ORO	
2/1/12	0	9.2	8.8	13.7	22,400	50,100	³ / ₈ " black oil on top
2/2/12	1	21.1	8.3	8.5	23,000	56,200	No visible change
2/6/12	5	22.9	8.3	0.3	9,590	21,700	Water column cloudy
2/9/12	8	22.4	8.0	5.7	8,680	16,300	Water column cloudy, oil has turned brown
2/16/12	15	22.0	8.1	6.1	8,750	14,000	Water column cloudy, ¹ / ₂ " brown oil on top
2/23/12	22	21.8	7.7	5.8	4,280	9,420	Water column cloudy, ¹ / ₂ " brown oil on top
3/1/12	29	21.6	7.9	5.1	3,420	7,330	Water column cloudy, ⁷ / ₁₆ " brown oil on top
3/8/12	36	21.8	7.8	6.5	5,670	12,800	Water column cloudy, ³ / ₈ " brown oil on top

Tank 2 – Missouri River Water Treated With Oil Spill Eater II - Not Aerated

Date	Days after Start of Test	Readings with Water Quality Meter			Lab Results		Notes
		Temp (°C)	pH	DO (mg/L)	TPH-DRO	TPH-ORO	
2/1/12	0	8.2	8.6	13.9	22,400	50,100	3/8" black oil on top
2/2/12	1	20.3	8.4	8.1	27,100	65,900	No visible change
2/6/12	5	22.9	8.4	0.3	10,700	23,800	Water column cloudy
2/9/12	8	23.0	7.9	1.5	4,890	9,220	Water column cloudy, oil still black
2/16/12	15	22.6	7.5	1.3	8,580	16,800	Water column cloudy, 5/16" black oil on top
2/23/12	22	22.4	7.5	1.5	12,400	18,700	Water column cloudy, 1/4" black oil on top
3/1/12	29	22.2	7.4	0.6	4,860	10,000	Water column cloudy, 3/16" black oil on top
3/8/12	36	22.4	7.7	1.5	4,740	10,400	Water column cloudy, 1/8" black oil on top

OIL SPILL EATER II TEST RESULTS

Tank 3 – Missouri River Water Not Treated With Oil Spill Eater II - Aerated

Date	Days after Start of Test	Readings with Water Quality Meter			Lab Results		Notes
		Temp (°C)	pH	DO (mg/L)	TPH-DRO	TPH-ORO	
2/1/12	0	7.9	8.6	13.6	22,400	50,100	3/8" black oil on top
2/2/12	1	20.8	8.5	8.1	35,400	73,300	No visible change
2/6/12	5	22.7	8.1	5.0	10,800	22,100	Water column less cloudy than Tanks 1 & 2
2/9/12	8	22.4	8.2	7.1	2,750	5,160	Water column as before, oil has turned brown
2/16/12	15	21.8	8.2	7.4	7,180	14,100	Water column as before, 3/4" brown oil on top
2/23/12	22	21.6	7.9	6.6	2,700	7,210	Water column as before, 1 1/2" brown oil on top
3/1/12	29	21.3	8.3	6.4	5,140	10,000	Water column as before, 1 1/4" brown oil on top
3/8/12	36	21.5	8.1	6.6	14,800	33,300	Water column as before, 1 1/8" brown oil on top

Tank 4 – Missouri River Water Not Treated With Oil Spill Eater II - Not Aerated

Date	Days after Start of Test	Readings with Water Quality Meter			Lab Results		Notes
		Temp (°C)	pH	DO (mg/L)	TPH-DRO	TPH-ORO	
2/1/12	0	8.4	8.6	13.3	22,400	50,100	3/8" black oil on top
2/2/12	1	20.7	8.5	8.5	34,100	84,800	No visible change
2/6/12	5	23.0	7.9	0.3	15,100	33,100	Water column less cloudy than Tanks 1 & 2
2/9/12	8	23.2	7.8	1.2	10,200	19,200	Water column as before, oil still black
2/16/12	15	22.7	7.9	1.1	4,430	8,660	Water column as before, 5/16" black oil on top
2/23/12	22	22.5	7.9	1.2	12,500	27,900	Water column as before, 1/4" black oil on top
3/1/12	29	22.4	7.4	0.4	8,300	17,600	Water column as before, 3/16" black oil on top
3/8/12	36	22.5	7.8	0.7	20,100	45,000	Water column as before, 1/8" black oil on top

OIL SPILL EATER II TEST RESULTS

Tank 5 – Blue Springs Lake Water Treated With Oil Spill Eater II - Aerated

Date	Days after Start of Test	Readings with Water Quality Meter			Lab Results		Notes
		Temp (°C)	pH	DO (mg/L)	TPH-DRO	TPH-ORO	
2/1/12	0	8.5	8.7	13.4	17,300	38,800	3/8" black oil on top
2/2/12	1	20.4	8.5	8.6	36,800	89,700	No visible change
2/6/12	5	22.8	8.1	0.8	9,630	24,600	Water column cloudy
2/9/12	8	22.3	7.9	6.5	949	1,740	Water column cloudy, oil has turned brown
2/16/12	15	21.8	8.1	7.1	6,140	12,000	Water column cloudy, 3" brown oil on top
2/23/12	22	21.6	7.9	6.5	11.6	21.5	Water column cloudy, 2" brown oil on top
3/1/12	29	21.5	7.9	5.7	11,000	20,600	Water column cloudy, 1" brown oil on top
3/8/12	36	21.6	8.0	6.5	3,980	8,700	Water column cloudy, 9/16" brown oil on top

Tank 6 – Blue Springs Lake Water Treated With Oil Spill Eater II - Not Aerated

Date	Days after Start of Test	Readings with Water Quality Meter			Lab Results		Notes
		Temp	pH	DO	TPH-	TPH-	

		(°C)		(mg/L)	DRO	ORO	
2/1/12	0	8.4	8.6	13.1	17,300	38,800	3/8" black oil on top
2/2/12	1	20.3	8.4	8.9	25,300	52,700	No visible change
2/6/12	5	22.9	8.4	0.4	28,300	72,700	Water column cloudy
2/9/12	8	22.9	7.7	1.4	13,800	25,900	Water column cloudy, oil still black
2/16/12	15	22.5	8.0	1.5	4,620	7,470	Water column cloudy, 5/16" black oil on top
2/23/12	22	22.4	7.8	1.5	12,000	26,600	Water column cloudy, 1/4" black oil on top
3/1/12	29	22.2	7.3	0.5	9,220	20,100	Water column cloudy, 3/16" black oil on top
3/8/12	36	22.4	7.8	0.3	7,090	15,000	Water column cloudy, 1/8" black oil on top

OIL SPILL EATER II TEST RESULTS

Tank 7 – Blue Springs Lake Water Not Treated With Oil Spill Eater II - Aerated

Date	Days after Start of Test	Readings with Water Quality Meter			Lab Results		Notes
		Temp (°C)	pH	DO (mg/L)	TPH-DRO	TPH-ORO	
2/1/12	0	8.4	8.6	13.2	17,300	38,800	3/8" black oil on top
2/2/12	1	20.5	8.5	8.9	36,900	77,100	No visible change
2/6/12	5	22.8	8.2	5.5	30,800	61,900	Water column less cloudy than Tanks 5 & 6
2/9/12	8	22.0	7.9	7.3	1,540	2,820	Water column as before, oil has turned brown
2/16/12	15	21.6	8.7	8.2	NA	NA	Water column as before, 3/4" brown oil on top
2/23/12	22	21.5	8.4	7.5	1,690	3,810	Water column as before, 3/4" brown oil on top
3/1/12	29	21.4	8.0	6.4	16,500	35,200	Water column as before, 5/8" brown oil on top
3/8/12	36	21.5	8.4	6.6	16,000	35,900	Water column as before, 7/16" brown oil on top

NA Not analyzed (sample containers broken during shipment to laboratory).

Tank 8 – Blue Springs Lake Water Not Treated With Oil Spill Eater II - Not Aerated

Date	Days after Start of Test	Readings with Water Quality Meter			Lab Results		Notes
		Temp (°C)	pH	DO (mg/L)	TPH-DRO	TPH-ORO	
2/1/12	0	8.7	8.6	13.0	17,300	38,800	3/8" black oil on top
2/2/12	1	20.8	8.5	8.7	41,100	100,000	No visible change
2/6/12	5	22.8	8.0	4.3	39,300	83,900	Water column less cloudy than Tanks 5 & 6
2/9/12	8	23.0	7.9	1.8	11,500	21,400	Water column as before, oil still black
2/16/12	15	22.6	8.6	1.2	3,450	6,840	Water column as before, 5/16" black oil on top
2/23/12	22	22.4	8.4	1.3	4,450	9,430	Water column as before, 1/4" black oil on top
3/1/12	29	22.3	7.1	1.1	9,710	19,200	Water column as before, 3/16" black oil on top
3/8/12	36	22.8	8.1	0.8	8,060	17,000	Water column as before, 1/8" black oil on top

Notes:

On the initial day of the test (02/01/12, Day 0), a composite sample was collected from the tanks containing Missouri River water, and another composite sample was collected from tanks containing water from Blue Springs Lake. These samples were collected after oil had been added to the water, and before addition of any Oil Spill Eater II.

A second air pump was added on 02/08/12 to increase aeration, in an attempt to increase dissolved oxygen (DO) levels in the aerated tanks.

**Draft Proposal for Bench-Scale Testing of Oil Spill Eater II
Region 7 Regional Response Team Science and Technology Workgroup
January 30, 2012**

The Region 7 Regional Response Team (R7 RRT) has tasked the R7 RRT Science and Technology Workgroup (S&TW) to evaluate the oil spill control agent, Oil Spill Eater II (OSEII), manufactured by Oil Spill Eater International Corporation (OSEI) in Dallas, Texas, for pre-authorized use in Region 7. As part of this evaluation the S&TW, with the assistance of Tetra Tech EM Inc., under the Superfund Technical Assessment and Response Team (START) 3 contract, will conduct a bench-scale study to help evaluate the effectiveness of OSEII to clean up oil spills on surface water and soils.

Oil Spill Eater II is sold as a biodegradable, non-toxic, water soluble, liquid nutrient, which is intended to promote accelerated growth of indigenous bacteria that degrade petroleum compounds to constituents that present minimal threat to the environment (carbon dioxide, etc.). The product is applied to a spill by spraying the impacted area, at a concentration recommended by the manufacturer. The product is marketed as having been proven effective on spills of various petroleum materials, including gasoline, diesel fuel, crude oil, and refined oil. In addition, testing conducted by OSEI has indicated the product is not toxic to aquatic life (if used according to the manufacturer's directions).

For the bench-scale study to be conducted in EPA Region 7, Oil Spill Eater II will be applied to waste oil that has been added to containers of water collected from two surface water bodies in the Kansas City area—Lake Jacomo, a 970-acre lake about 15 miles east of downtown Kansas City, and the Missouri River. The testing will be performed in 10-gallon aquariums staged in a temperature-controlled environment at the EPA Region 7 warehouse in Kansas City, Missouri.

The testing procedure will be conducted in both aerated and non-aerated containers, to simulate turbulent and stagnant flow conditions. Containers of water collected from both Kansas City area surface water bodies with no added Oil Spill Eater II will be incorporated as controls for the study. Samples of the water in all containers will be analyzed for total petroleum hydrocarbons (TPH)-diesel range organics (DRO) and TPH-oil range organics (ORO) before application of the Oil Spill Eater II, and periodically thereafter to evaluate the rate of TPH degradation. Visual observations of conditions in the aquariums, including any obvious changes since the previous sampling, will also be recorded throughout the testing. Final water samples will be collected on the 35th day after introduction of the product, and the final TPH results will be evaluated to determine whether a threat to human health or the environment (presented by TPH-DRO and TPH-ORO) appears to remain. A summary report will be prepared by START following the bench-scale test. A description of the intended testing procedure is attached as Appendix A.

APPENDIX A
SUMMARY OF TESTING PROCEDURE

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1. Approximately 40 gallons of water will be collected from Lake Jacomo and the Missouri River (sources) in 55-gallon drums, using a submersible (or similar) pump.
2. Water from the drums will be transferred to eight labeled 10-gallon aquariums (four aquariums for water from each source).
3. Approximately 1 liter of waste oil will be added to water in all eight aquariums.
4. A calibrated, submersible probe (or equivalent) will be used to measure dissolved oxygen, pH, and temperature in each aquarium.
5. Samples of oil and water will be collected from each aquarium to determine initial TPH concentrations. These samples will be collected from the entire water column with a device such as a disposable Coliwasa, to ensure representative samples are obtained from each container. Each sample will consist of two 1-liter glass bottles for analysis of TPH-DRO and TPH-ORO.
6. A mixture of source water and Oil Spill Eater II will be prepared in pump-type garden sprayers (one sprayer for each source), at a ratio of 3 ounces of Oil Spill Eater II to 128 ounces of source water.
7. Approximately 1 liter of each Oil Spill Eater II mixture will be sprayed onto the oil surface in two aquariums containing its respective source water (total of four aquariums).
8. Plastic tubing from an air pump (bubbler) will be placed into four of the aquariums—one with Oil Spill Eater II and one without for each of the two sources. The air pump will be connected to a manifold, from which separate tubing will extend to the four aquariums. This will ensure similar flow rates for all aquariums. A summary of all eight aquariums follows:
 - a. Missouri River Water – Treated & Aerated
 - b. Missouri River Water – Treated & Non-aerated
 - c. Missouri River Water – Untreated & Aerated (control)
 - d. Missouri River Water – Untreated & Non-aerated (control)
 - e. Lake Jacomo Water – Treated & Aerated
 - f. Lake Jacomo Water – Treated & Non-aerated
 - g. Lake Jacomo Water – Untreated & Aerated (control)
 - h. Lake Jacomo Water – Untreated & Non-aerated (control)
9. Samples will be collected from all aquariums on the following seven days after the start of testing: 1, 3, 7, 14, 21, 28, and 35. These samples will be collected as described in Step 5 for the same analyses. Field parameters will also be measured before each sampling event, as described in Step 4.

Notes:

The aquariums will be covered with screens during the testing period to prevent foreign material from entering the containers.

Videotape footage and photographs will be taken to document all phase of the testing procedures.

All samples will be maintained at a temperature at or below 4 degrees Celsius and submitted to a START-contracted laboratory for analysis by EPA SW-846 Method 8015 (or equivalent).