

CytoCulture Louisiana Research Team

Integrated Shoreline Remediation Strategies

Gulf Coast Marsh Clean Up Effort with UCB

Proposed Team, Concepts and Scope of Work



June 14, 2010

Integrated Shoreline Remediation Strategy for Testing in Louisiana

Operate a Remediation Response Team to Respond to Oiled Shorelines using Field-Proven Integrated Technologies

- Identify, test & evaluate remediation technologies in field plots
- Match technologies & data to oiled Areas of Impact (AOI's)
- Obtain state & federal permits, product approvals, consensus
- Integrate appropriate remediation technologies for each AOI
- Coordinate technology vendors, application protocols, logistics
- Integrate sampling protocols, GIS grid systems, laboratories
- Coordinate state & federal agency oversight & participation
- Supervise shoreline clean-up implementation, technical support
- Monitor process, sample collection, retrieval & lab analysis

Documentation & Reporting

Evaluation of Integrated Shoreline Remediation Technologies

- Interpret field test plot data for individual & combined remediation methods: physical, chemical & biological
- Coordinate data sharing with state & federal agencies participating in field trials of integrated technologies
- Report to Agencies on field trial evaluations with technology recommendations specific to types of AOI's
- At each AOI slated for treatments, collect baseline data, samples during treatments & monitor shorelines weekly
- Report to Agencies with visual, chemical & microbiological data on a monthly basis thereafter

Remediation Evaluation Project Team

To Test & Integrate Field-Proven Technologies

- ***CytoCulture*** International, Inc. / ***Rapid Energy Services***, LLC
Bioremediation expertise, microbiological lab, pilot test facility
in Lafayette, LA within 15 min of University of Louisiana
- **REC-Royal Engineers & Consultants, LLC** Lafayette, LA
Environmental Management (EM), engineers & scientists
- ***Southern Petroleum Laboratories***, Lafayette, LA
Analytical chemistry, sample retrieval services
- ***Hydro Environmental Technologies***, Baton Rouge, LA
Sampling sediments & water, remediation work force

Project Team Key Personnel

to Identify, Test & Integrate Remediation Methods

Lead researchers, engineers, analytical staff & management

- CytoCulture: Randall von Wedel, Ph.D. – Biochemist
- Rapid Energy Services: David Trahan, Chemist
- Royal Engineering: Mitch Andrus, P.E., Greg Scott, R.G.
- Southern Petroleum Laboratories - team
- Hydro Environmental Service - team

Project Team Facilities

Dedicated to Integrated Remedial Response Gulf Region

- Royal Engineering management, research, consulting, GIS & drafting service facilities in New Orleans & Lafayette
- CytoCulture-Rapid Energy wet laboratory and pilot testing facilities in Lafayette; chemical blending facilities, support
- CytoCulture environmental microbiology services lab in San Francisco, CA to monitor biodegradation parameters
- Southern Petroleum Laboratories analytical chemistry labs & sample handling infrastructure: Lafayette & Gulf region
- Hydro Environmental work force, vehicle & vessel fleet, sampling & remediation services across Gulf region

Louisiana University Researchers

Scientists and laboratories committed to Team collaborations

- **Andrew Nyman, Ph.D. & Vanessa Tobias, Ph.D.:** Marsh habitat, shoreline cleaning strategies, effects of cleaners
LSU College of Agriculture / LSU Ag Center Research
- **Mark Zappi, Ph.D., P.E. –** Shoreline bioremediation
University of Louisiana, Prof. & Dean of Engineering
- **Ramesh Kolluru, Ph.D., U. L. Prof. Engineering &**
Exec. Dir. National Incident Management Systems &
Advanced Technologies; U.L. Assist.VP Grad. Research
- **Ehab Meselhe, Ph.D., U.L. Prof. Engineering:** Wetlands
Hydrology & Modeling including research for Corps

Additional Academic Collaborators

- Paul Klerks, Ph.D. UL: Toxicology of dispersants in wetlands
- Donald Hayes, Ph.D. UL Engineering: Wetland oil spill mitigation; collaborator with Dr. Mark Zappi

Additional LA researchers are being interviewed at this time.

Potential Collaborations with University of California, Berkeley

- Thomas Azwell, UCB Environmental Science, Policy & Management along with Dr. John Coates, UCB
- Will Stringfellow, Ph.D., Lawrence Berkeley Laboratory, Department of Energy and Environment
- Angus McGrath, Ph.D. (UCB doctorate), Stantec, Inc.

Remediation Scenarios by Habitat

To Field-test & Document selected remediation strategies for:

- Marsh vegetation – fringing edges of marsh exposed to oil
- Wetlands & mudflats - tidal areas penetrated by oil slicks
- Waterways & harbors – breakwaters, bulkheads, docks, piers
- Sand beaches – coastal shore, outer marsh & barrier islands
- Oiled ships & work vessels – clean oiled hulls in water

Integration of Remediation Strategies

Examples of oil mitigation methods to evaluate and field test:

- Oil collection and recovery methods for sensitive shorelines
- Oil skimming & bulk oil recovery methods for open water
- Oil-water separation equipment and instrumentation
- Shoreline cleaners to 'lift and float' bulk oil from surfaces
- Surfactant & nutrient formulations to speed biodegradation
- Biological absorbent technologies to degrade residuals
- Slow release peroxide formulations to oxygenate water
- Direct oxygenation by floating electrochemical devices
- Monitoring DO, pH, temp & degradation parameters by satellite using floating remote, solar-powered devices

Technologies under Consideration

Chemical treatments listed on NCP Schedule of Products

- Oil skimmers designed for shallow water, limited access and suitable for manual deployment from skiffs in marsh
- Oil-water separator boxes suitable for placement in shallow water adjacent to fringing marsh to collect oil
- Shoreline cleaners that facilitate recovery of stranded oil rather than emulsifying into the water column; bio-product basis
- Mild surfactant-nutrient solutions used to rinse off residual hydrocarbons, improve oxygen penetration to sediments and enhance natural biodegradation of remaining contaminants
- Absorbent boom made from dry peat moss as a biologically active material to biodegrade residual oil post marsh cleaning
- Automated oxygenation and remote monitoring devices

Proteus-CytoCulture-RES Team

Observations of Oiled Marsh

- May 19 Field Trip on 24-ft fishing boat to collect samples
- Collected 'control' sediment & soil samples from clean beaches; clean seawater from open areas free of oil
- Collected oil scum from degraded, emulsified oil slick 7 nm south of Pass a Loutre lighthouse; photographed
- Observed oil slick penetrating marsh 0.5 nm NE of the Pass a Loutre lighthouse, just south of old North Pass
- Photographed and video-taped oil emulsion moving through cane vegetation & entrapment within small bay
- Collected samples of oil emulsion & impacted marsh water
- Samples provided to LSU, UL and CytoCulture in CA

Location of Oiled Marsh May 19



Oiled marsh at Pass a Loutre to the E of the Mississippi

Oiled Marsh May 19 Sample Area

One of the First Wetlands Impacted by Oil



Fringing marsh cane penetrated with oil emulsion

Oiled Marsh May 19 Sample Area



Inner bay of marsh saturated with black oil

Oiled Marsh May 19 Sample Area



Trailing edge of emulsified oil slick moving in

Collecting Oil Emulsion Samples



Oil Emulsion Samples for Microbiological & Chemical Analysis



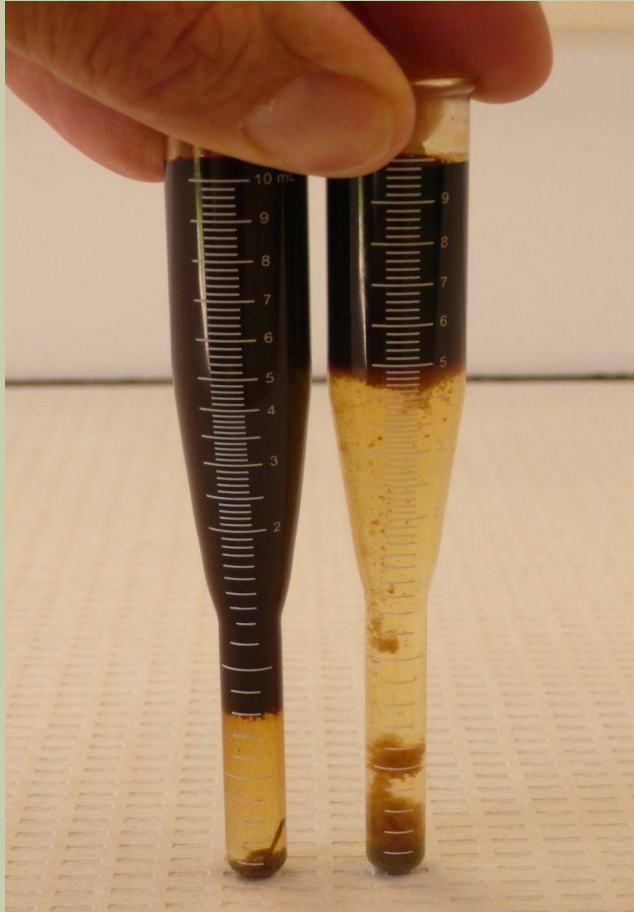
Samples analyzed by CytoCulture & LSU

Rapid Energy Services Wet Lab

Lafayette, Louisiana



Breaking oil emulsion with CytoSol



Left: warmed oil emulsion after low speed centrifugation 37 deg C

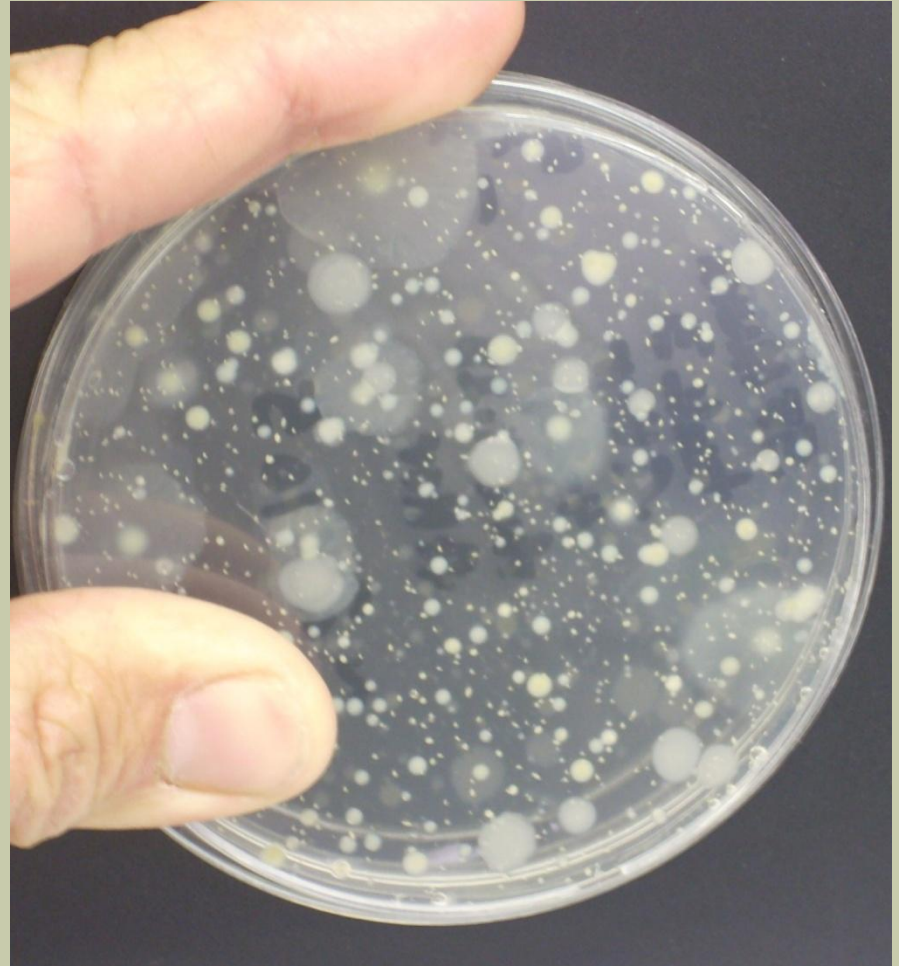
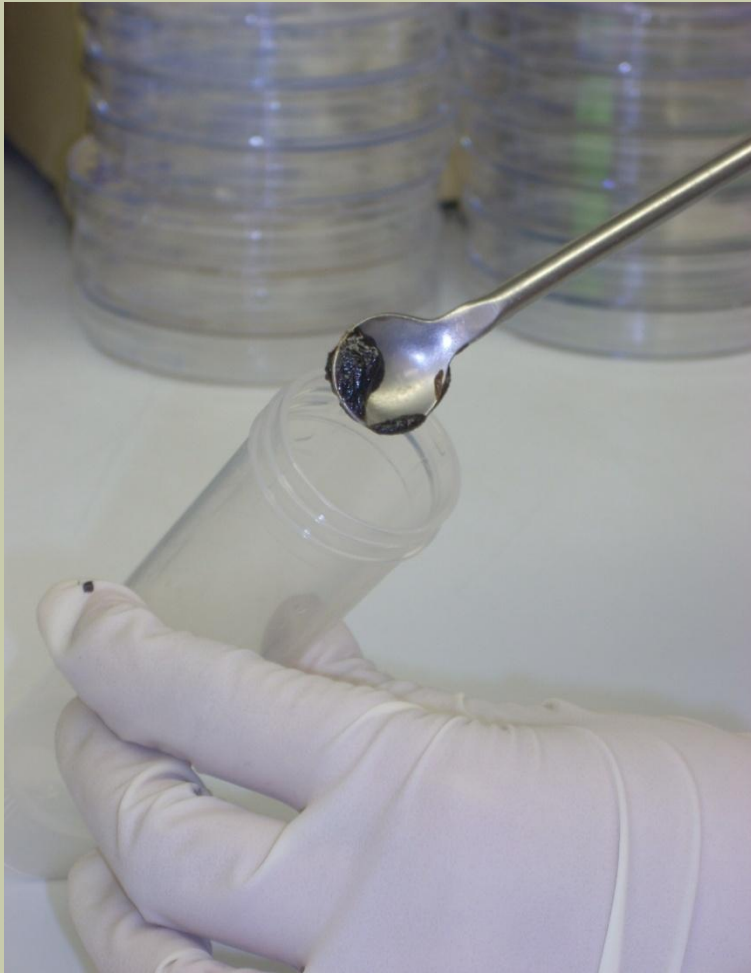
Right: warmed oil emulsion treated with CytoSol (20% to 80% emulsion) and centrifuged. Emulsion contains 62% water, 0.4% organic debris & ca. 37% liquid oil. Other samples had as little as 18% liquid oil in emulsion.

CytoCulture's Environmental Microbiology Lab Monitors Oil-Degrading Bacteria



Aerobic & Anaerobic Hydrocarbon-Degraders

Bacteria Enumeration for HC-Degraders



Emulsion samples have high populations of oil-eating bacteria

CytoCulture's Lab Data for AEROBIC Bacteria Populations Associated with Oiled Marsh May 19

Samples of clean 'control' water & shoreline vs. oil emulsion
Aerobic bacteria counts in 'colony forming units' per g or ml

	<u>Heterotrophs</u>	<u>HC-Degraders</u>
Clean seawater offshore	1×10^7	6×10^5
Clean marsh beach sand	2×10^4	1×10^2
Clean marsh peat detritus	2×10^5	2×10^4
Oil Emulsion Scum offshore	2×10^8	3×10^7
Oil Emulsion at oiled marsh	3×10^5	6×10^2
Oily water from oiled marsh	3×10^5	4×10^4

PENDING Data for ANAEROBIC Bacteria Populations Associated with Oiled Marsh May 19

Samples of clean 'control' water & shoreline vs. oil emulsion
Anaerobic bacteria counts in 'colony forming units' per g or ml

	<u>Heterotrophs</u>	<u>HC-Degraders</u>
Clean seawater offshore	3×10^5	1×10^5
Clean marsh beach sand	3×10^7	2×10^6
Clean marsh peat detritus	6×10^7	3×10^7
Oil Emulsion Scum offshore	7×10^7	6×10^7
Oil Emulsion at oiled marsh	2×10^5	9×10^5
Oily water from oiled marsh	2×10^5	2×10^5

Evaluating Oil Emulsion in Pools at Rapid Energy's Pilot Test Facility



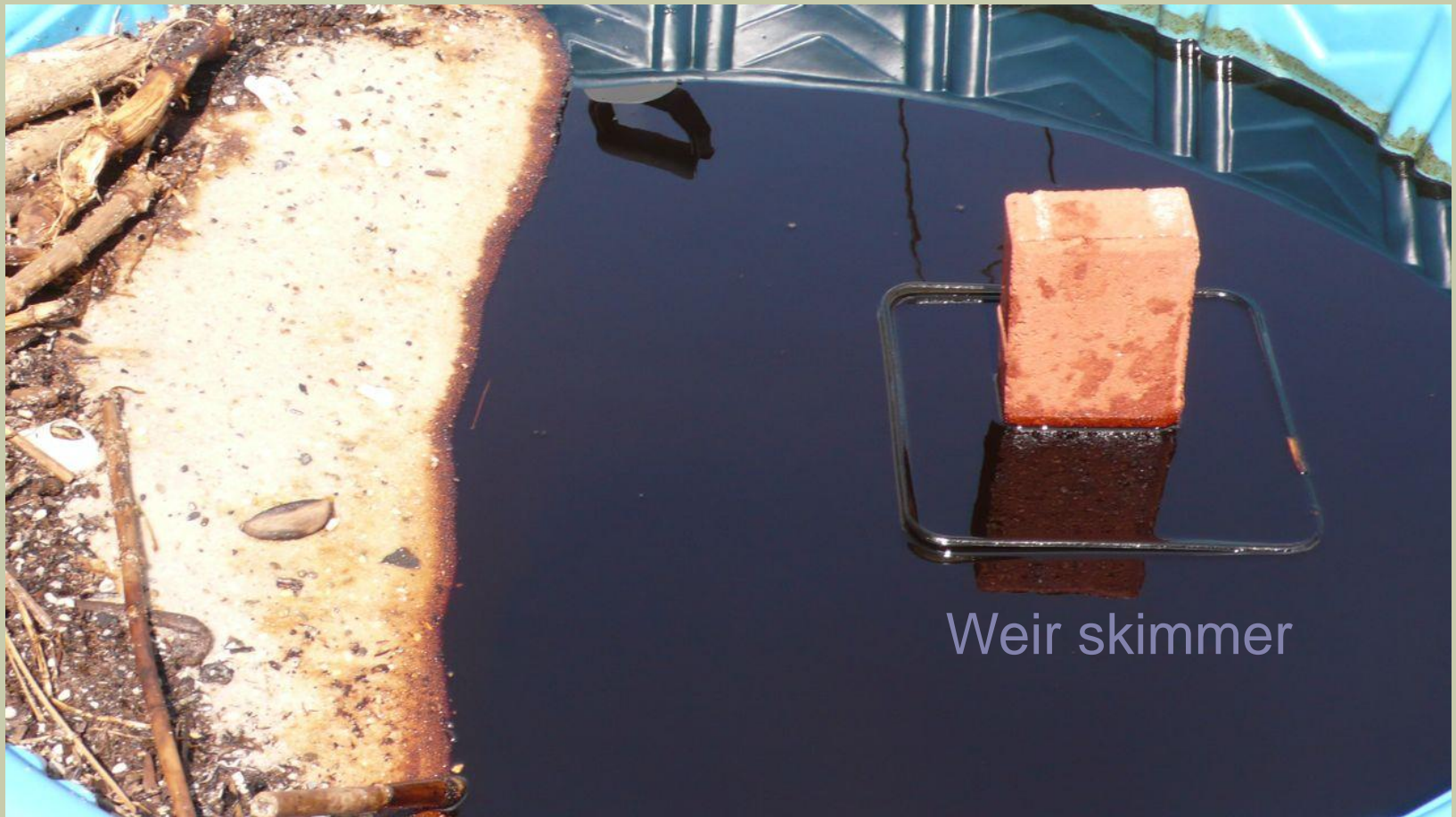
Simulating oiled marsh beach with emulsion

Mesocosm study of the oil emulsion impacting a Louisiana marsh beach



See www.cytosolbiosolvent.com/videos/ for pilot tests

Oiled Beach with Simulated Skimming by a Weir Skimmer Machine



Weir skimmer

See www.cytosolbiosolvent.com/videos/ for pilot tests

May 22 Pilot Test Initial Conclusions

Simulating oiled marsh beach using emulsion collected May 19

Oil observed & collected at impacted marsh near Pass a Loutre

- Highly degraded by sun, seawater, bacteria & evaporation
- Apparently emulsified with surfactants by BP
- Low level of ‘adhesiveness’ (not adhering much to substrates)
- High level of ‘cohesiveness’ (sticks together, forms globules)
- Neutral density: light winds will roll emulsion under water
- Easy to collect by mechanical skimmers or absorbent pads
- Stranded emulsion on beach responds well to surface washing agents: CytoSol Biosolvent followed by Proteus-168 rinse

Summary

Integrated Shoreline Remediation Strategy – next steps

- CytoCulture Remediation Team is in place & ready to respond to Agencies/BP to proceed with technology testing
- Infrastructure in place: laboratories, pilot test facilities, vessels, vehicles, containment systems, products staged
- Technology vendors under consideration are on board
- Pilot & laboratory testing of oil emulsion are underway
- University researchers in Louisiana await funding to set up independent assessments of proposed field trial protocols
- State and federal agencies appear interested in approach, but challenging to penetrate the Incident Command Center

Contact Info for Research Team

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