GOM-SCHEMA: The impact of the Deepwater Horizon spill on historic shipwreck microbiomes in the northern Gulf of Mexico

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Shipwreck Corrosion - Hydrocarbon Exposure

SCHEMA

Microbiology -Archaeology

BOEM
Bureau of Ocean Energy Management

Cooperative Agreement M13AC00015
Interagency Agreement M13PG00020

Ewing Bank Shipwreck
July 2014

Co-PIs: Melanie Damour (BOEM), Lisa Fitzgerald (NRL), Chris Horrell (BSEE), Robert Church (Oceaneering)

Collaborators: , Jennifer Salerno (GMU); Brenda Little, Allen Reed, Yoko Furukawa, Ricky Ray, Jason Lee (NRL- Stennis); Samantha Joye (UGA), Dan Warren (Oceaneering),

Historic shipwrecks in the northern Gulf of Mexico

- Greater than 50 years old
- Protected under National Historic Preservation Act (NHPA)
  - Federal agencies must consider effects of permitted activities (energy exploration) on cultural resources
- Impacts from *Deepwater Horizon* spill not addressed by Natural Resource Damage Assessment, GOMRI, RESTORE Act funding, etc.)
- Cultural, maritime and wartime heritage spanning 500 years on seafloor in northern GOM
- "100% Non-Renewable Resource" Melanie Damour, BOEM Marine Archaeologist, Co-PI

Halo, tanker – WWII Casualty 1942
(150m)
~150 km from Macondo Well

U-166, German U-boat– WWII Casualty 1942
(1500m)
~8 km from Macondo Well

Anona, luxury steam yacht – freighter – sank 1944
(1300m)
~75 km from Macondo Well

Historic shipwrecks in the northern Gulf of Mexico

- Contemporary role in seafloor ecology
- Basis for artificial reef ecosystems
- Solid substrate for organism settlement
- Intact ecosystem in deep-sea (trophic complexity)
- Expand knowledge of life in the deep-sea
- Ideal location to monitor trophic effects of *Deepwater Horizon* spill over time

Ewing Bank wreck– 19th century merchant
(600m)
~150 km from Macondo Well

Mica wreck– 19th century sailing – hull split by oil pipeline
(800m)
~12 km from Macondo Well

Viosca Knoll wreck , 19th century – hull split by platform anchor
(600m)
~75 km from Macondo Well

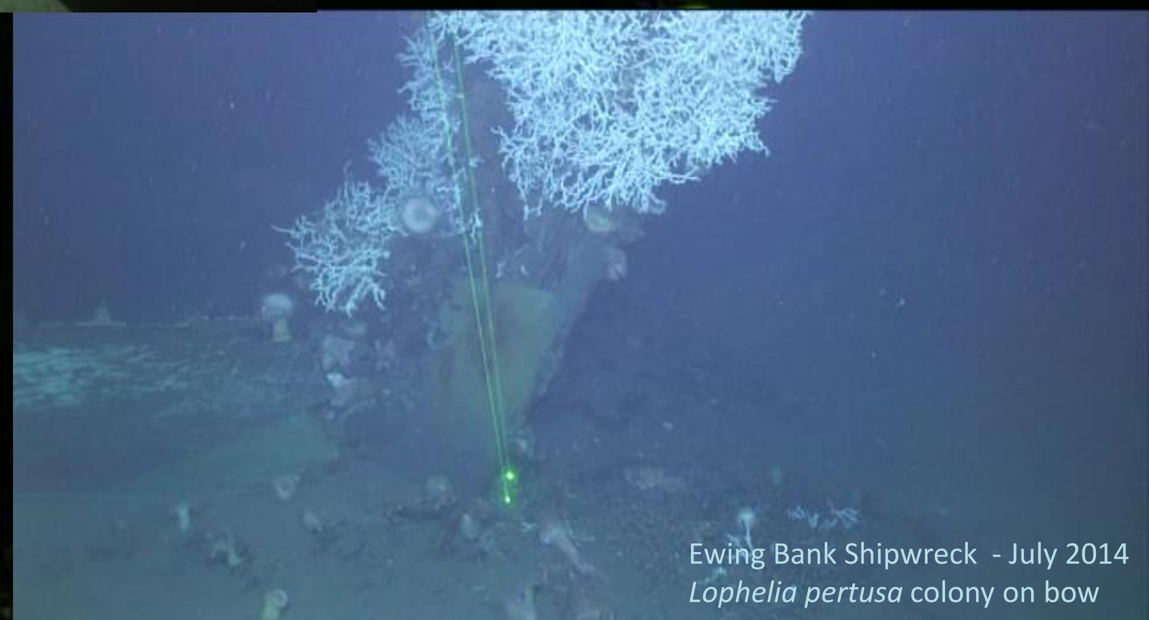
Deep sea shipwreck ecology

Viosca Knoll Shipwreck – July 2014
And the fantastic photo bombing fish
(video 16x speed)

Invisible Majority
Microorganisms

Foundation for life in the ocean:
Chemosynthesis
biofilms on wrecks allow other organisms to settle

Visible Diversity
Bivalves
Coral
Finfish
Tube worms
Etc….



Ewing Bank Shipwreck - July 2014
Lophelia pertusa colony on bow

Shipwreck Microbial Ecology

- All surfaces in the marine environment are immediately colonized by microorganisms

  → biofilms

- Biofilms establish chemical and physical conditions for recruitment and attachment of higher trophic levels (shipwreck >>reef)

Bow of the *Anona* shipwreck, SCHEMA study March 2014

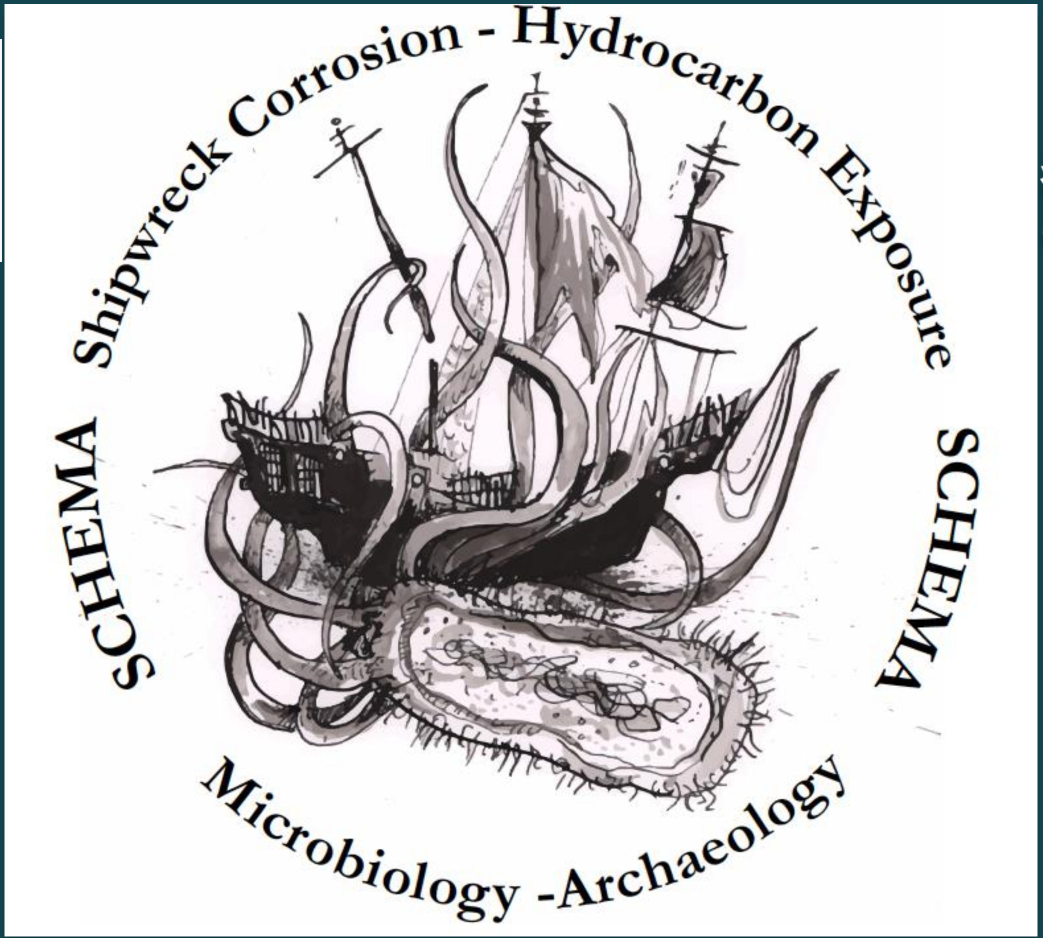
- Microorganisms – ubiquitous, metabolically diverse, short life spans
  - First to respond to contamination (biosensor)

- Spill effects may impact foundation of artificial reef ecosystem
- Spill effects may impact wreck preservation (microbial corrosion)

Shipwreck Microbial Ecology: time, place and (historic) context

NOPP National Oceanographic Partnership Program
Promoting Partnerships for the Future of Oceanography

BOEM
Bureau of Ocean Energy Management



The University of Southern Mississippi

BSEE
Bureau of Safety and Environmental Enforcement

George Mason University

The PAST Foundation
access through innovation

Naval Research Laboratory
Washington, DC

Master of Fine Arts
Science and Natural History Filmmaking
Montana State University

The University of Georgia

Deep Sea Systems International

LUMCON
Louisiana Universities Marine Consortium

C&C Technologies
Survey Services

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Christopher Horrell, BSEE
Robert Church, Oceaneering

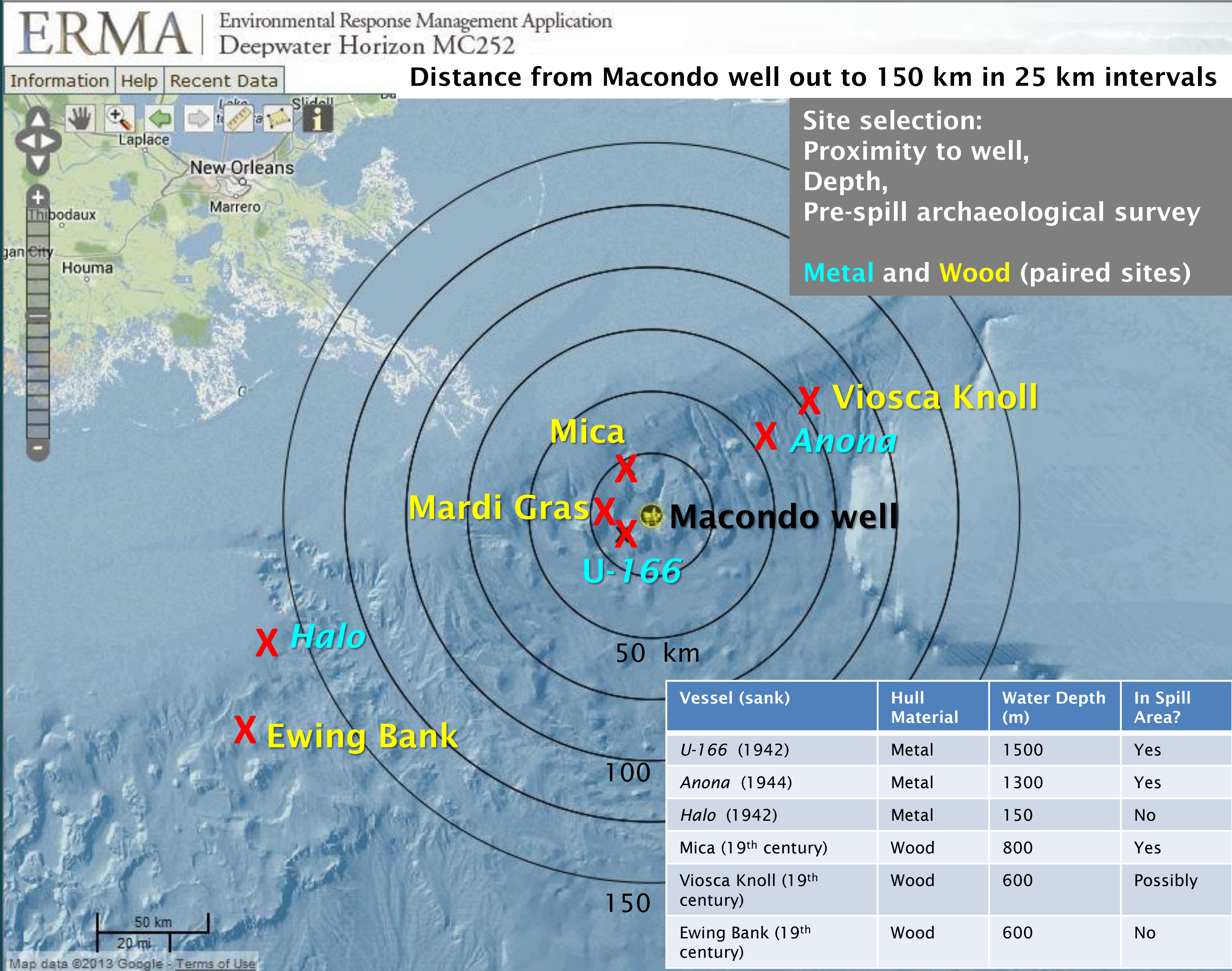
SCHEMA Purpose:

- Document the spill's prospective lasting effects on historic shipwrecks
- Impacts to shipwreck microbiomes through comparative study (impacted vs. non-impacted sites)
    - Sediment analysis
        - Microbiome composition
        - Microbiome function
        - Sediment geology
        - Sediment geochemistry
    - Biofilm Recruitment
- Role of microorganisms in metal corrosion to identify long-term (hull) impacts related to spill exposure
    - *In situ* biofilm experiments
    - Lab biofilm experiments
- Archaeological surveys
    - cm scale 3D imagery
    - Video/photo

Distance from Macondo well out to 150 km in 25 km intervals

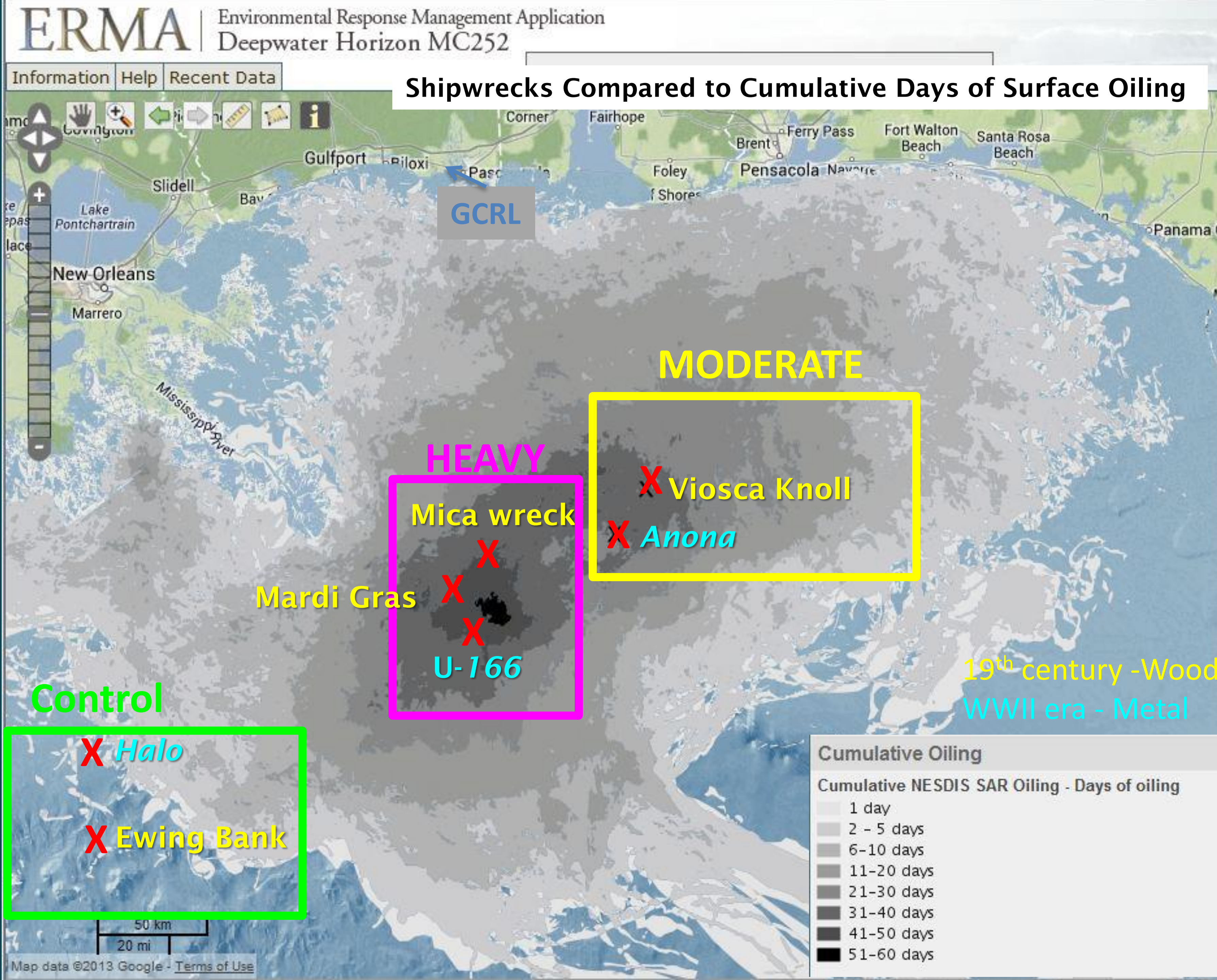
Site selection:
Proximity to well,
Depth,
Pre-spill archaeological survey

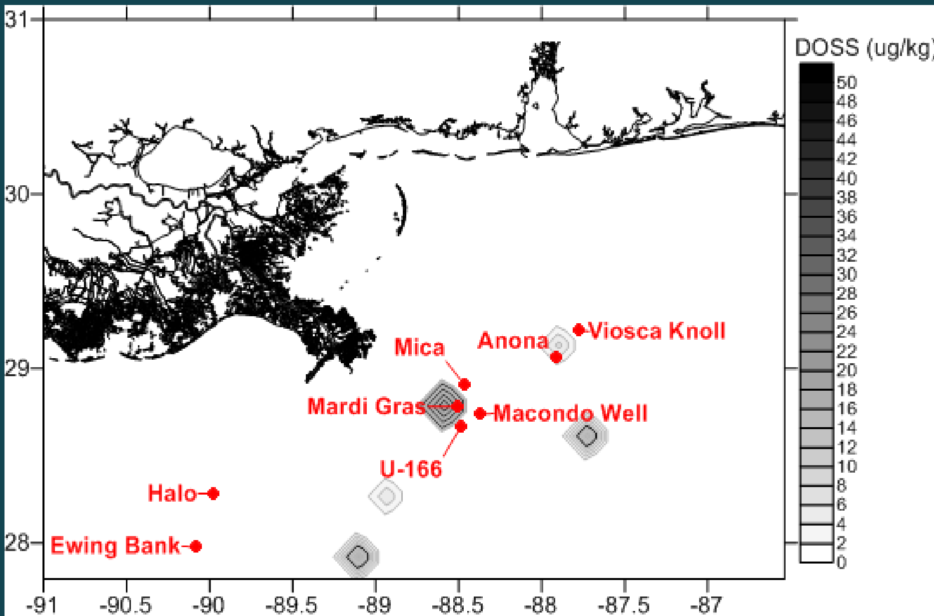
Metal and Wood (paired sites)

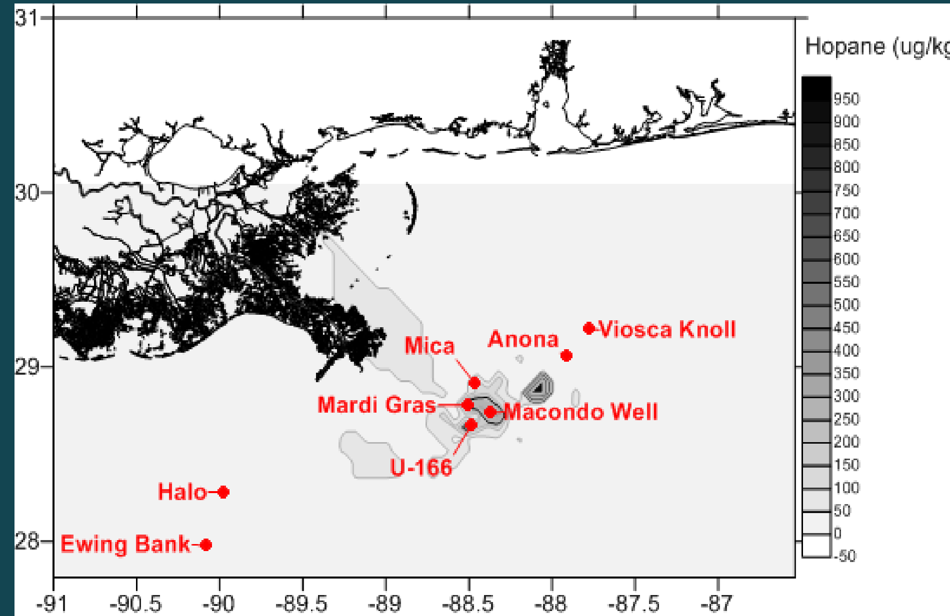


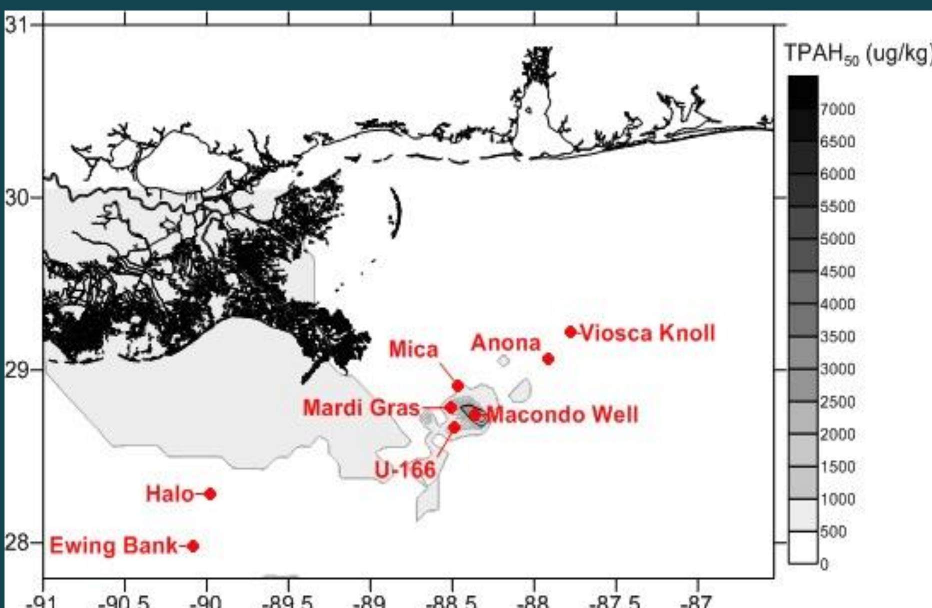
Vessel (sank)	Hull Material	Water Depth (m)	In Spill Area?
U-166 (1942)	Metal	1500	Yes
Anona (1944)	Metal	1300	Yes
Halo (1942)	Metal	150	No
Mica (19th century)	Wood	800	Yes
Viosca Knoll (19th century)	Wood	600	Possibly
Ewing Bank (19th century)	Wood	600	No

Shipwrecks Compared to Cumulative Days of Surface Oiling









NRDA Data set
2010-2014 sampling effort
~1200 sediment cores

2 sites (*U-166* and Mardi Gras)
in the 'Acute Footprint'

Hopane and PAH
Corexit (bis-(2-ethylhexyl) sulfosuccinate (DOSS))

Valentine et al., PNAS 111. 2014
Stout et al., Mar Pol Bul 114. 2017

Field Work

- R/V *Pelican* - LUMCON
- 2 ROV cruises – 2014
- *Global Explorer* ROV

March 2014

Video/photo surveys of wrecks

3D Laser Sonar Scanning

Deploy biofilm recruitment experiments

Coral collection (microbiome analysis)

July 2014

Sediment collection at wreck site and outside of debris field

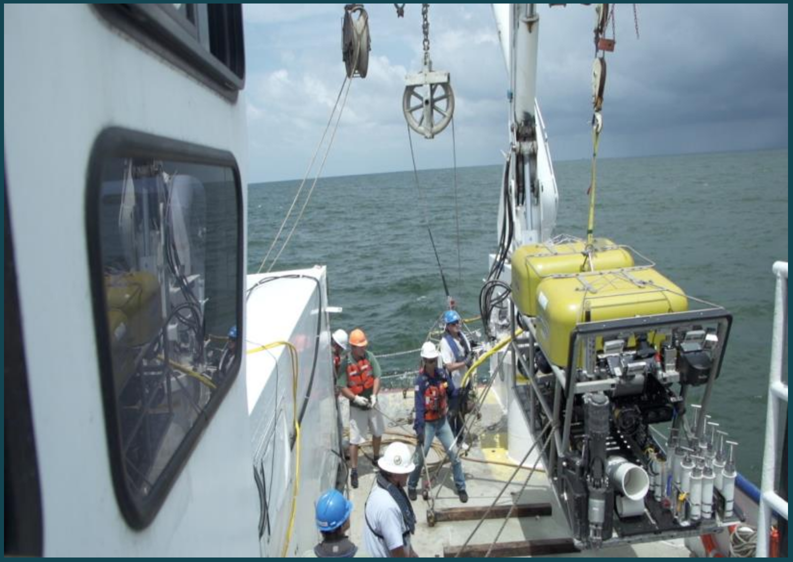
Recover biofilm recruitment experiments (1 of 2)

Water sampling (2m above wreck)

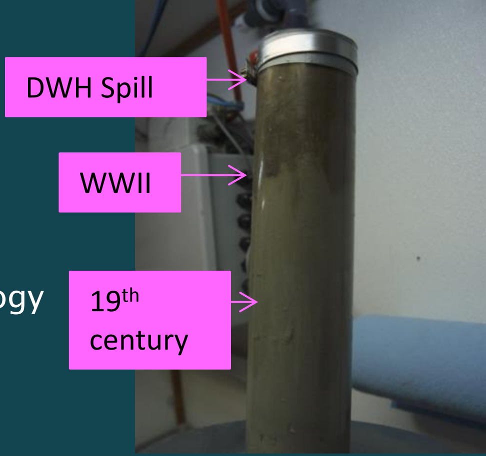
Four additional Cruises 2015 – 2017 (R/V Pelican and R/V Point Sur)

Multi-coring + ROV– 100% supported by NRL

Annual monitoring of spill recovery



Use sediment surrounding site to document effects

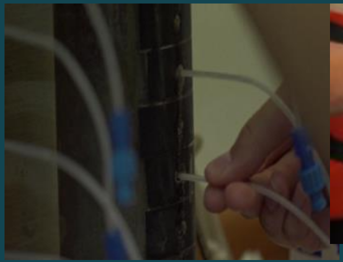


Sediment Deposition in northern GoM: **~ 0.07 cm/year**

20 cm core = ~300 years of history
Timeframe for when wrecks arrived on seafloor and DWH

Sampled at 2 cm intervals down core: profile geochemistry, physical properties, microbiology

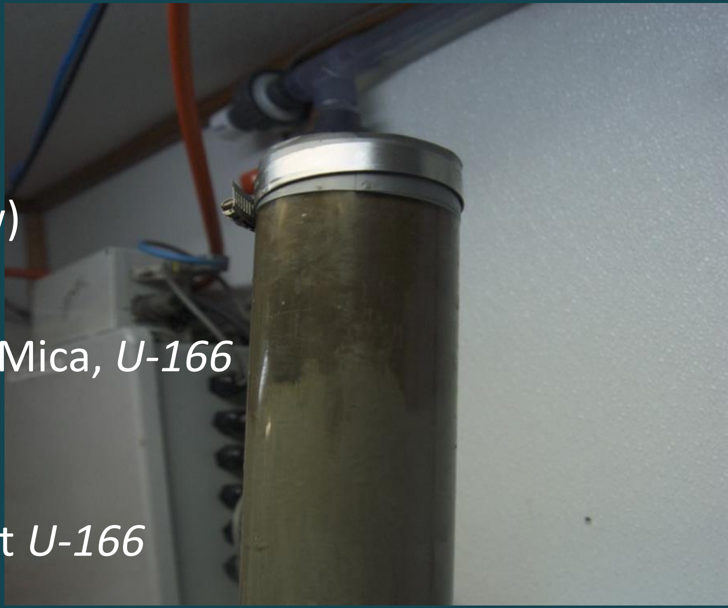
- Molecular Ecology
  - FastDNA Spin kit for Soil (MP Bio)
  - Illumina MiSeq Sequencing
  - V6 – V8 variable region of the 16S rRNA gene
    - B969F/BA1406R - Bacteria
    - A956F/A1401R – Archaea
    - ~30K sequences per sample; avg read length: 450 bp
- Bioinformatics
  - USEARCH – quality control
  - QIIME – taxonomic assignment
  - PICRUSt – explore functional gene potential
  - Primer E – statistical analysis



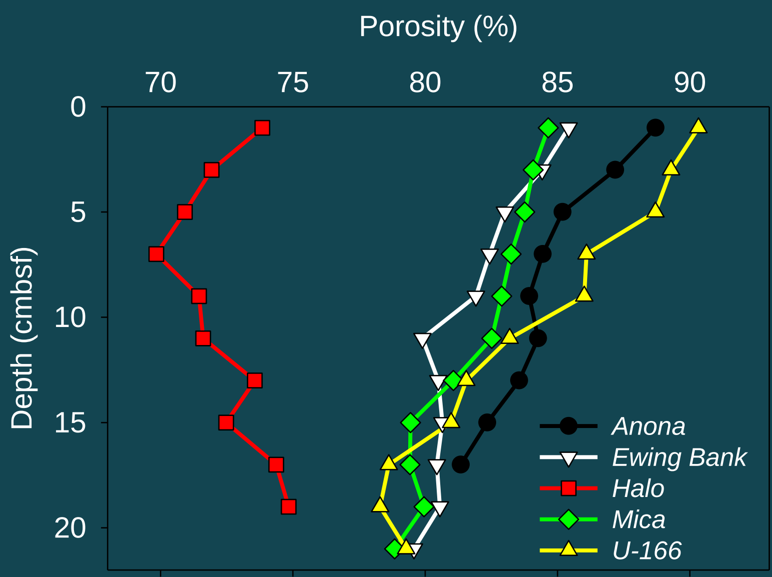
- Geochemical Analysis (UGA)
  - PAH + Total Pet Hydrocarbons (GC-MS)
  - Radiocarbon Natural Abundance
- Sediment Physical Properties (NRL)
  - Sedimentation Rate ^{210}Pb
  - Porosity

(video at 16x speed)

Sediment Geochemistry and Physical Properties



- Upper 4 cm of *U-166* & *Anona* – oil flocculent (oil snow)
- Upper 4 cm of *U-166* & *Anona - High Porosity*
- Long chain n-Alkane and TPH residue – surface/middle Mica, *U-166*
- Exceptional Sedimentation: *U-166* - 0.63 cm/year
  - No change in radiocarbon age from surface to bottom
- – Physical and Chemical Data provide evidence of oiling at *U-166*

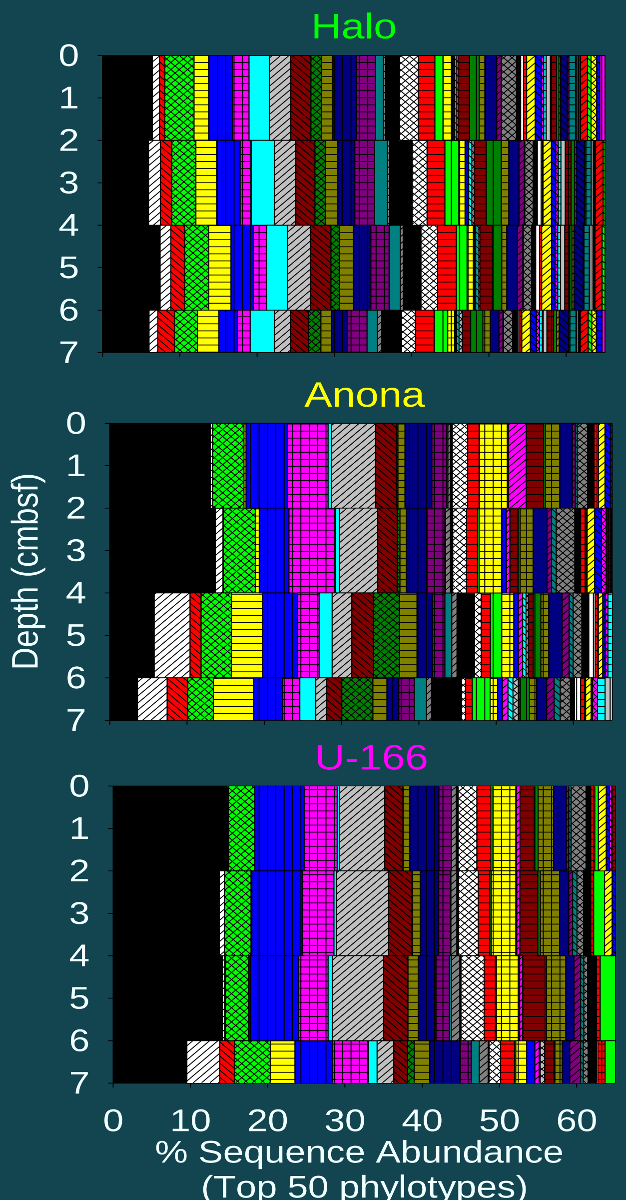


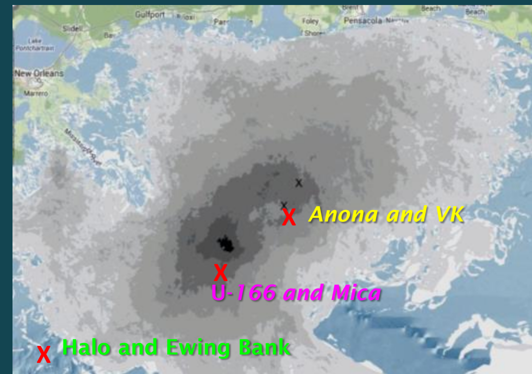
Data source: A. Reed, Y. Furukawa, NRL

Site		Depth (cmbsf)	^{14}C age, years BP	$\Delta^{14}C$	Sedimentation (cm/year)
Halo	Control	0-2	7340	-598.9	0.08
Halo	Control	18-20	**28660**	-971.8	
Ewing Bank	Control	0-2	2330	-251.6	0.12
Ewing Bank	Control	18-20	3630	-363.4	
Anona	Moderately	0-2	1270	-146.4	0.26
Anona	Moderately	16-18	2230	-242.7	
Mica	Heavily	0-2	1830	-204.1	0.14
Mica	Heavily	18-20	3160	-325.4	
U-166	Heavily	0-2	**2590**	-275.9	**0.63**
U-166	Heavily	18-20	**2540**	-271.5	

*Halo – tanker carrying crude oil when torpedoed. Hydrocarbon residue and radiocarbon age anomaly consistent with site formation timeframe

Bacterial Community Composition





Control sites- Highest Biodiversity

Biodiversity higher near wrecks vs. 100-200m away

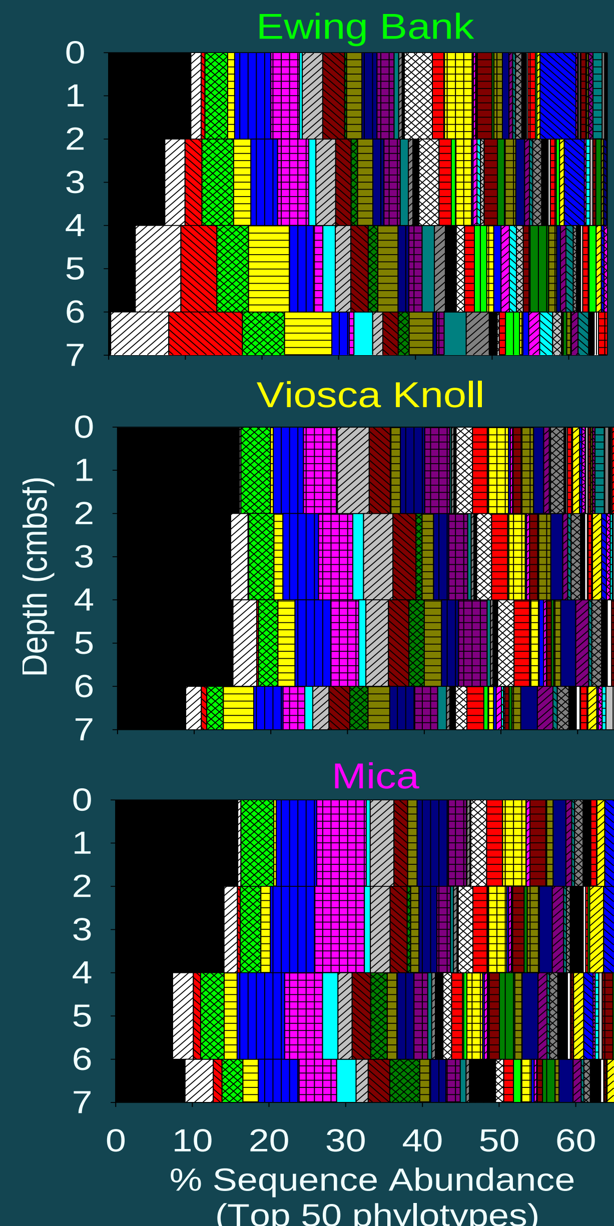
Heavily & Moderately impacted sites
Piscrickettsiaceae sp. (PAH degrader) ~16% sequences

No decline upper 6 cm – *U-166*, VK & Mardi Gras (not pictured)

U-166 and Mardi Gras – biodiversity same near and away from wrecks

Biological evidence of oiling at *U-166*

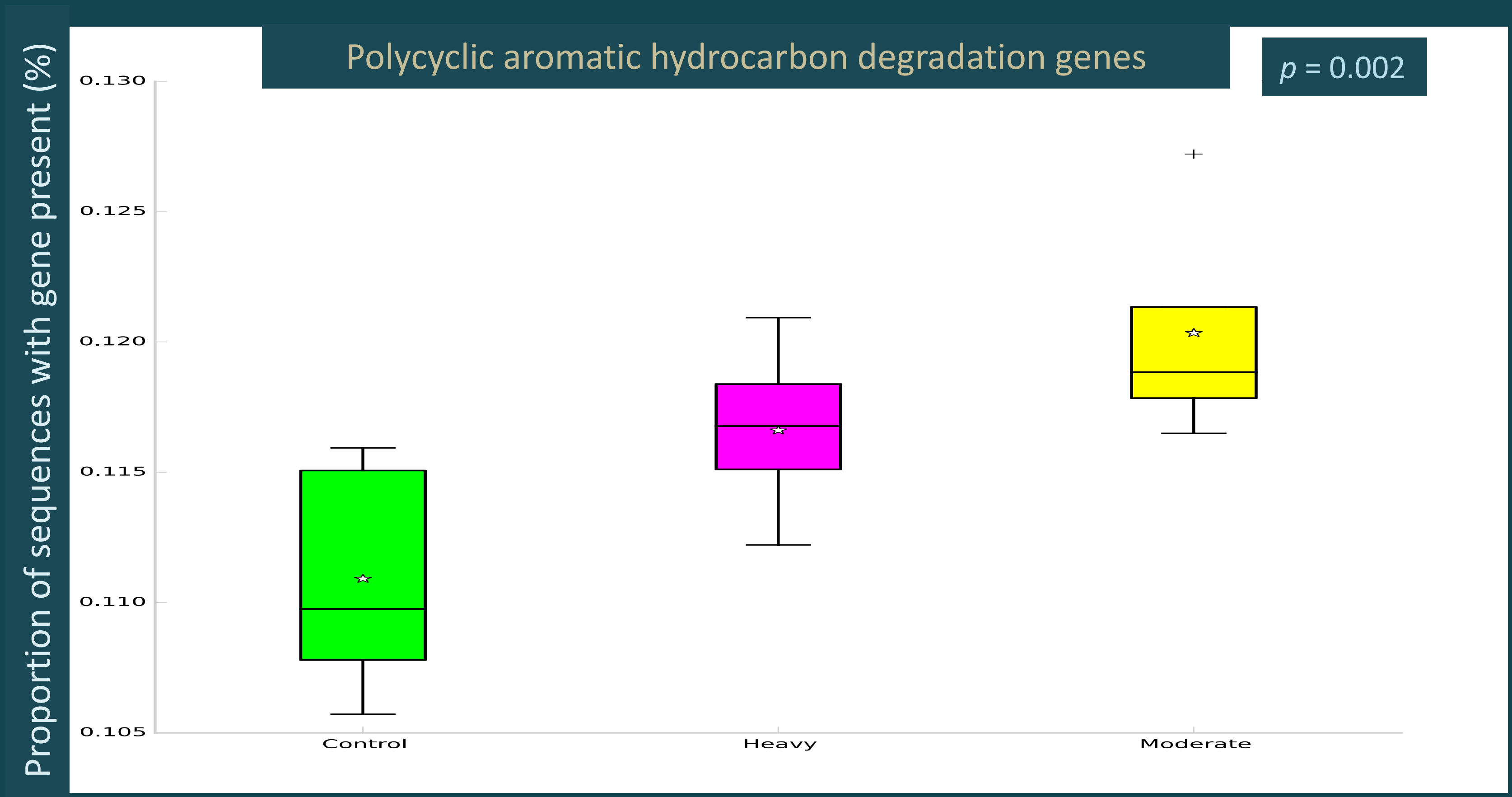
Observations continued through 2017



Community function at different site types

PICRUSt – Phylogenetic Investigation of Communities by Reconstruction of Unobserved States

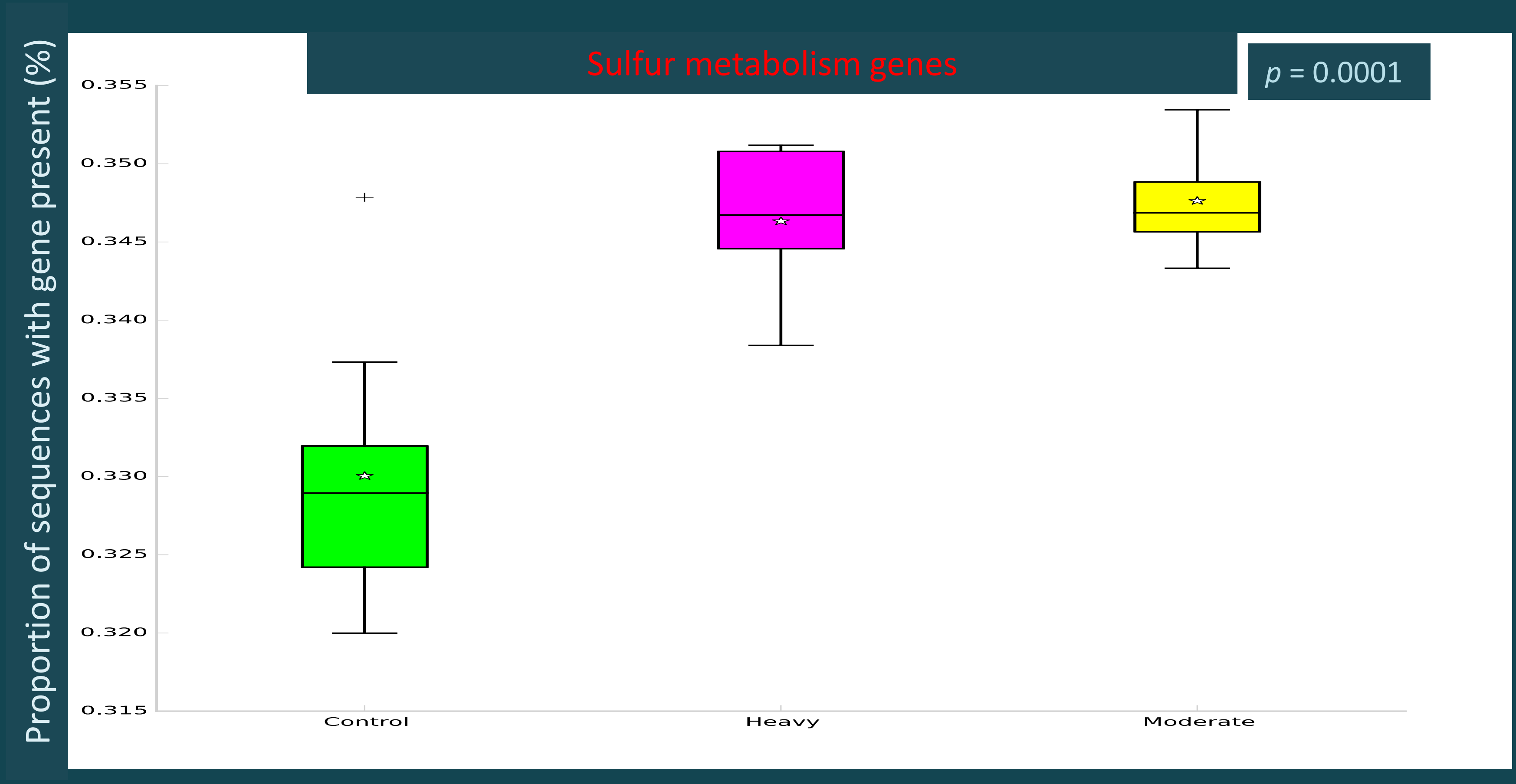
Predict functional composition of metagenome using 16S data and reference genome database



Control
Heavy
Moderate

PAH Degradation Genes elevated in moderate and heavy impacted sites

PICRUSt Predictions of FunctionalGenes



Control
Heavy
Moderate

Sulfur metabolism genes elevated in moderate and heavy impacted sites

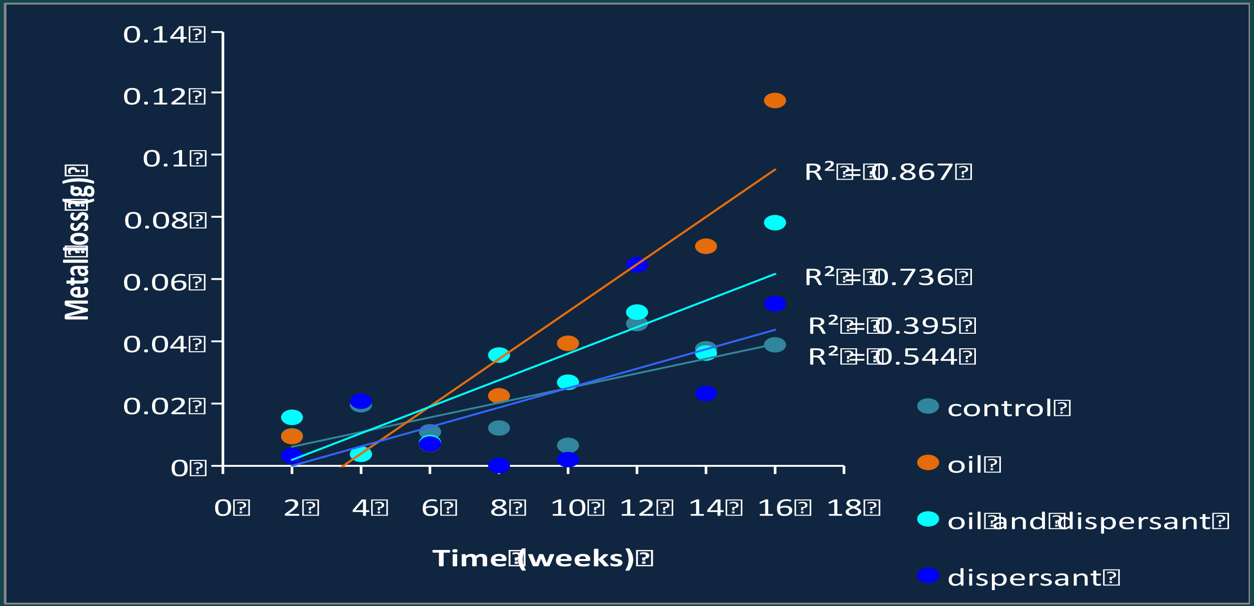
Spill contaminants and shipwreck preservation

– Sulfur Metabolism – Role in microbially-mediated metal corrosion

» Metabolites of sulfate reduction – chemically attack metal

– Laboratory Experiment:

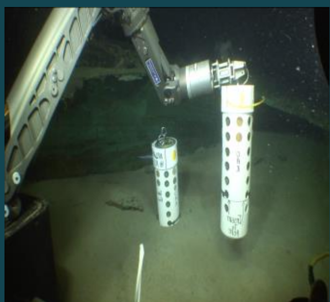
- Oil and dispersant introduced into seawater tanks
- Carbon steel disks sampled every 2 weeks for 4 months
- Monitored biofilm biodiversity and metal loss
  - Corexit introduction – Immediate and sustained reduction in biodiversity
  - Oil & Dispersant introduction – Increase in sulfur metabolism genes (metagenome analysis)
  - Increased metal loss in oil amended tanks



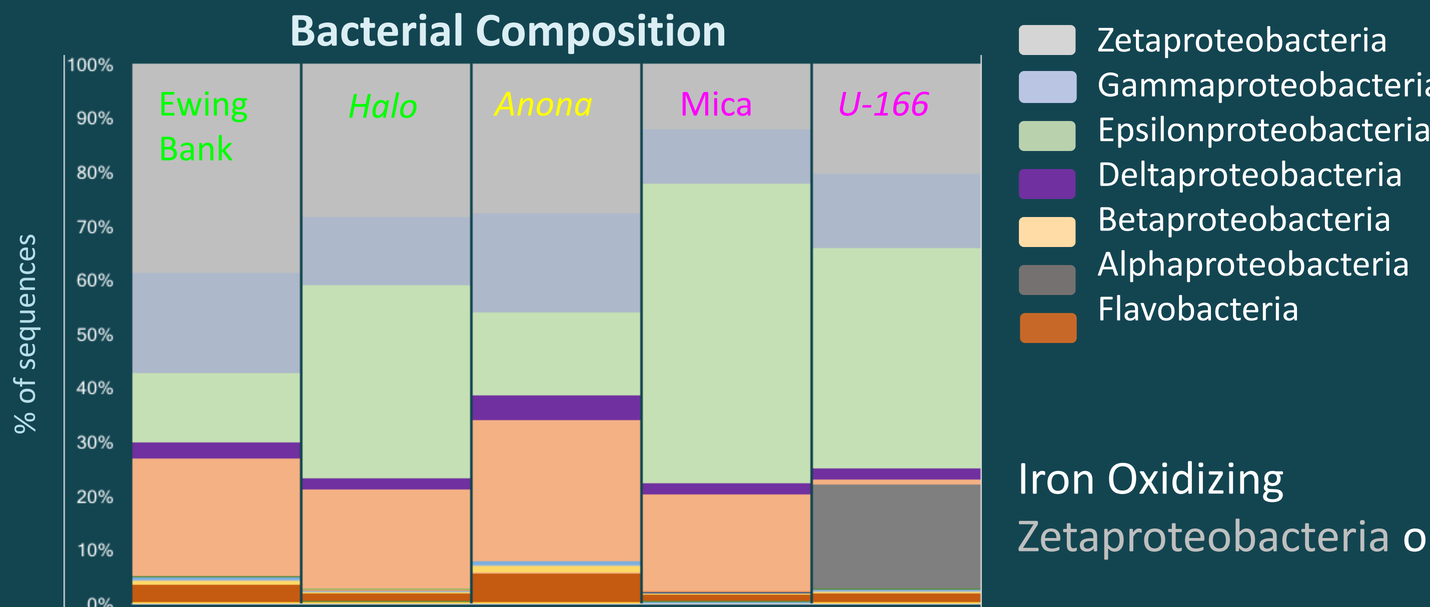
Corrosion products on carbon steel disk after 6 weeks in deep-sea water amended with oil

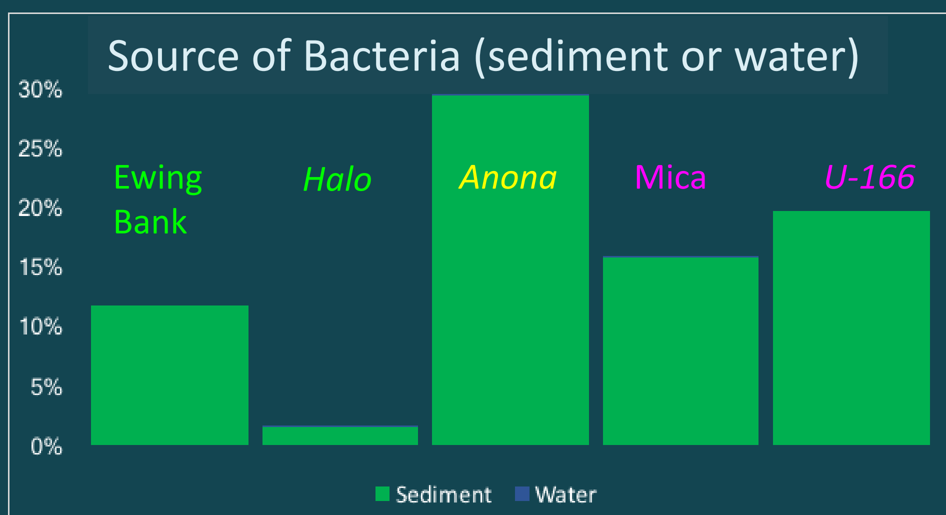
Biofilm Recruitment Experiment

Replicate steel disks on seafloor for 4 months - Within 2m of wreck



Biofilm recruitment Experiments placed on seafloor by ROV





Iron Oxidizing Zetaproteobacteria on steel

Source is sediment

Increased connectivity between sediment and wrecks at impacted sites

High sedimentation rates - ”fluffy” oil snow

Implications for preservation

Summary

- Three lines of evidence (geochemical, geological, microbiological) that historic shipwrecks were impacted by *Deepwater Horizon* Spill
- Depressed biodiversity in presence of dispersant
- Elevated sulfur metabolism & metal loss in presence of oil
- Sediment/wreck interactions – impacts biofilm formation

Outcomes

- Microbiomes detect spill effects AND shipwreck presence
- Tool to monitor historic and acute events on seafloor
- Interesting in context of 7 SCHEMA sites

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    - Interagency Agreement M13PG00020
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  - Deep Sea Systems, Int. (*Global Explorer* ROV)

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  - Melanie Damour, James Moore, Brian Jordan (BOEM)
  - Christopher Horrell (BSEE)
  - Jennifer Salerno (GMU)
  - Lisa Fitzgerald, Brenda Little, Yoko Furukawa, Allen Reed, Jason Lee, Ricky Ray, Tom Boyd (NRL)
  - Rob Church, Dan Warren (Oceaneering)

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