

# Archaea composition in the Northern Gulf of Mexico

Cynthia Kane

Mentor: Dr. Richard Snyder

Location: Pensacola, Florida



# About me

- ◆ Barnard College
- ◆ Rising Junior
- ◆ Biology Major

# About Dr. Snyder



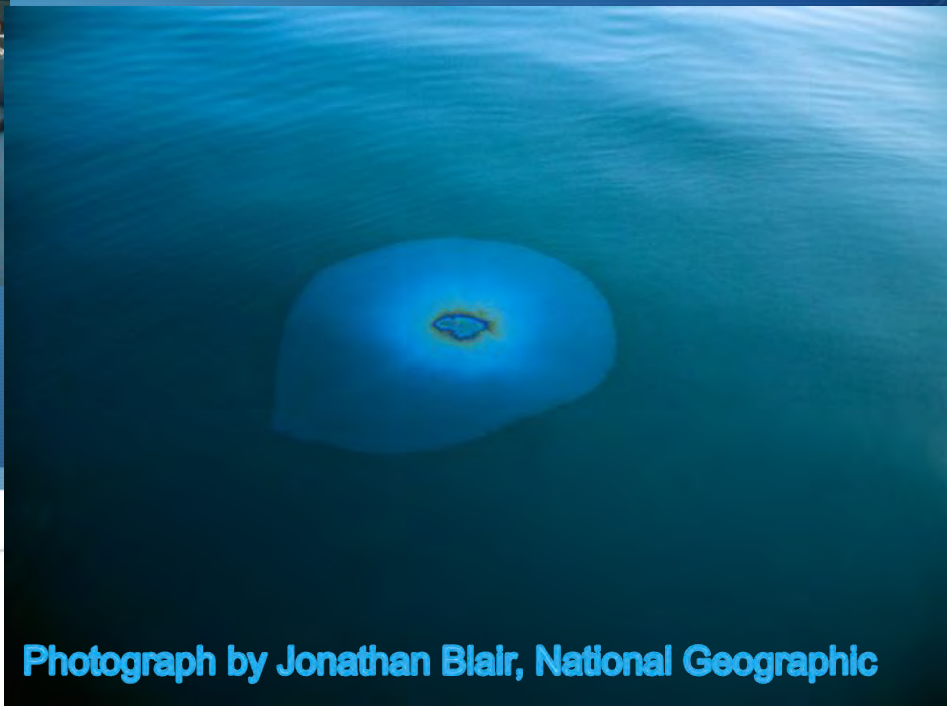
- ◆ Research Interests: Microbial and Macro Ecology, Pollution and Water Quality, Oceanography
- ◆ Current Research: Microbial oceanography in the NE Gulf of Mexico, coastal plant community analysis

# BP Oil Spill

**R/V Bellows 15-18 June 2010**

Photo credit:

Captain Larry Braun, R/V Bellows

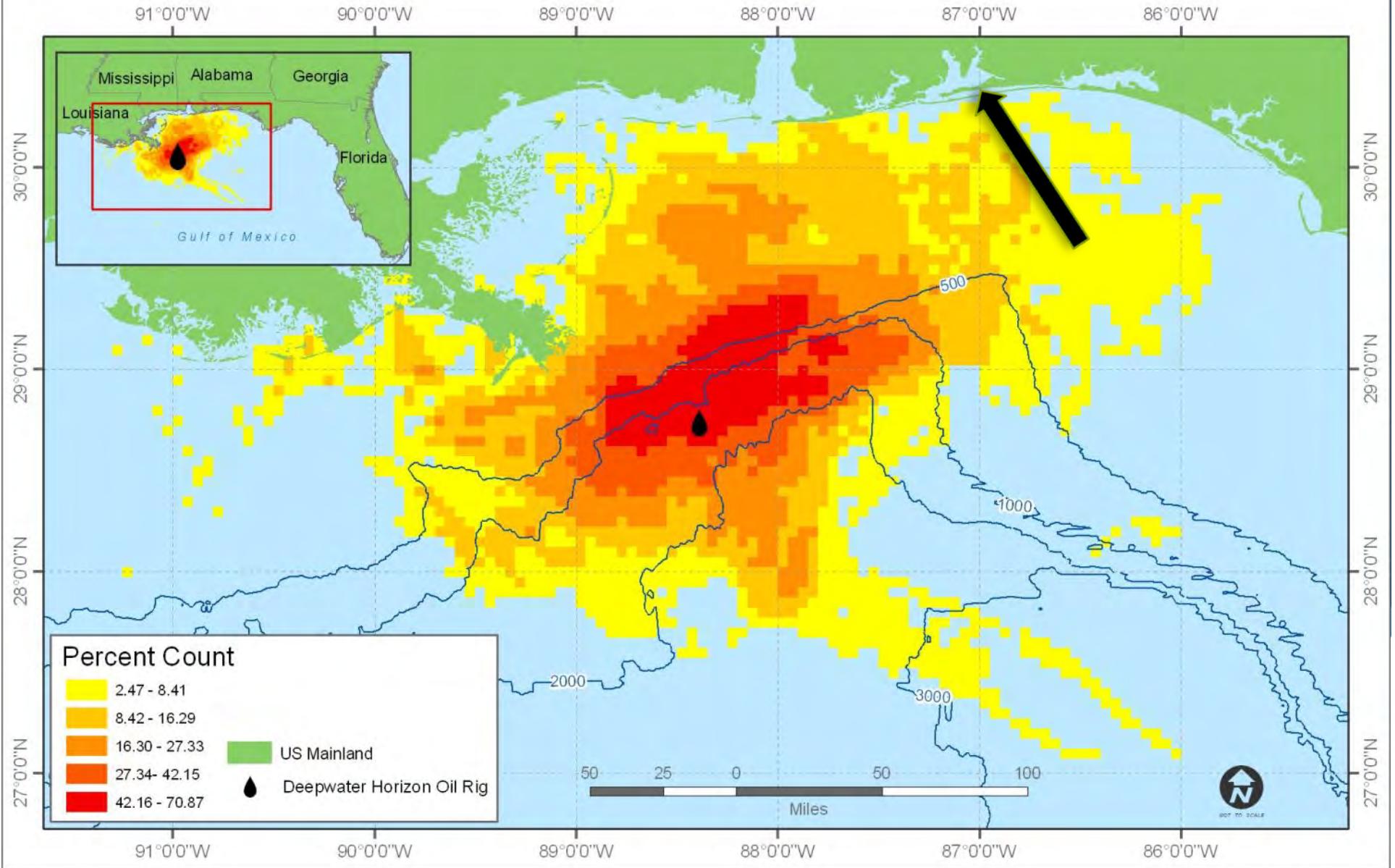


Photograph by Jonathan Blair, National Geographic

# BP's Oil Spill Analysis. Normalized Detection of Surface Oil.



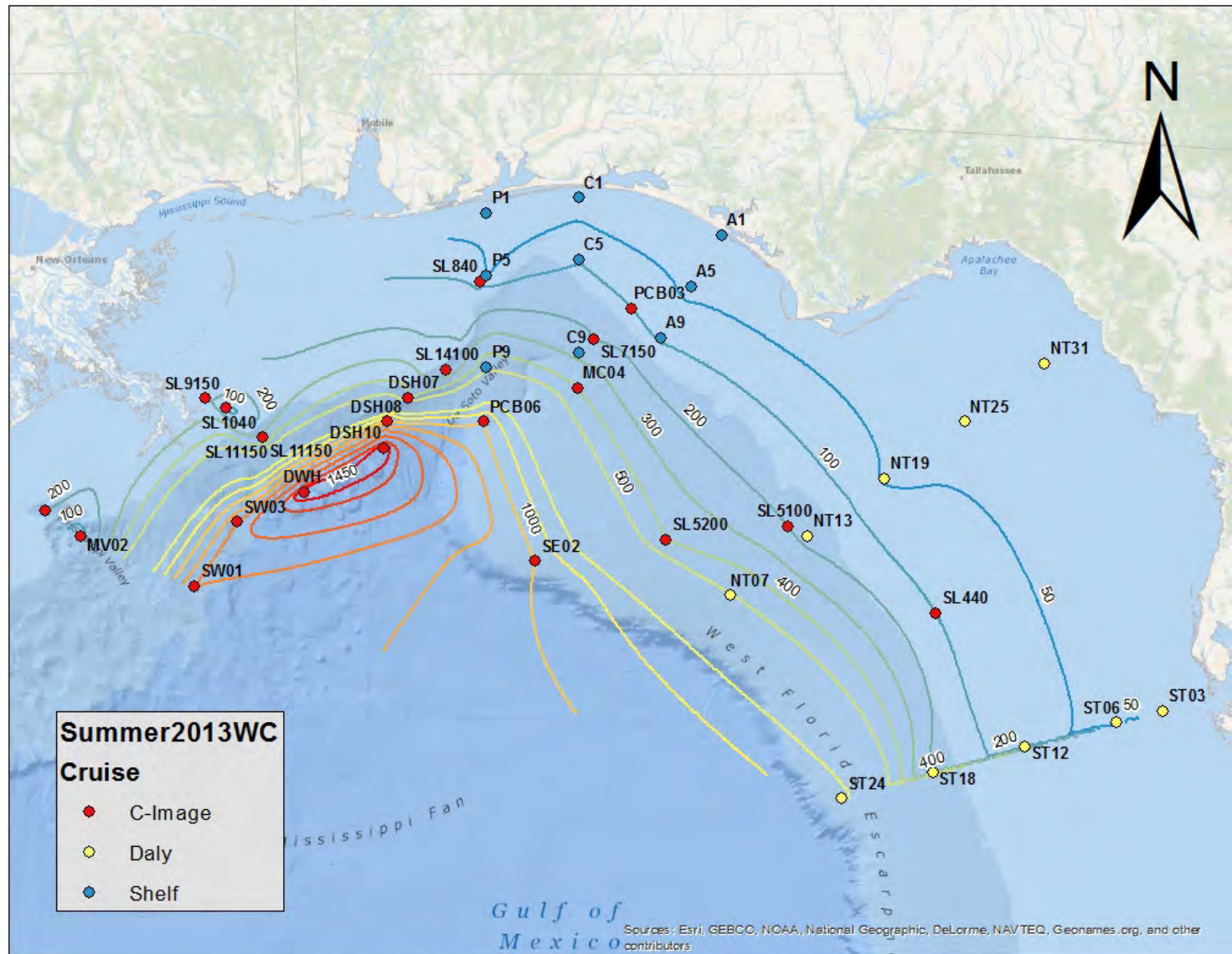
MacDonald Image Lab  
Dr. Oscar Garcia-Pineda  
EOAS Department



Projection: WGS\_1984  
Date: September 10, 2010

Source: TCNNA  
32 Envisat ASAR frames

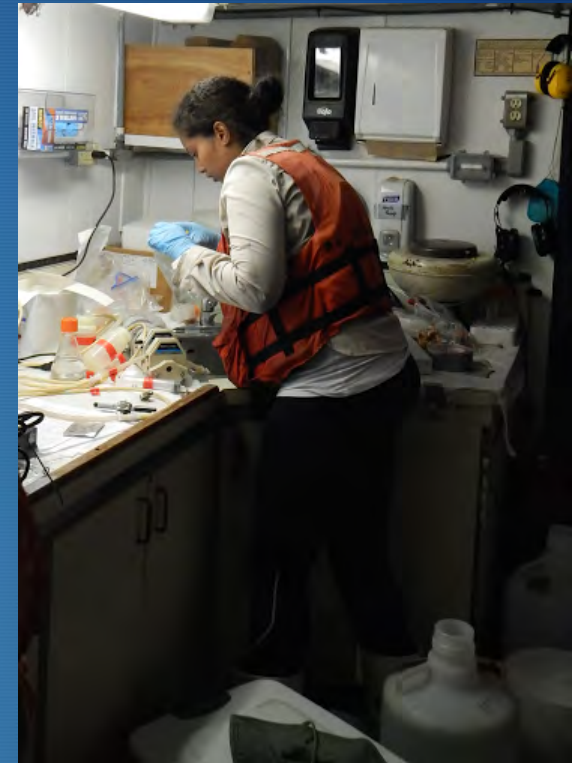
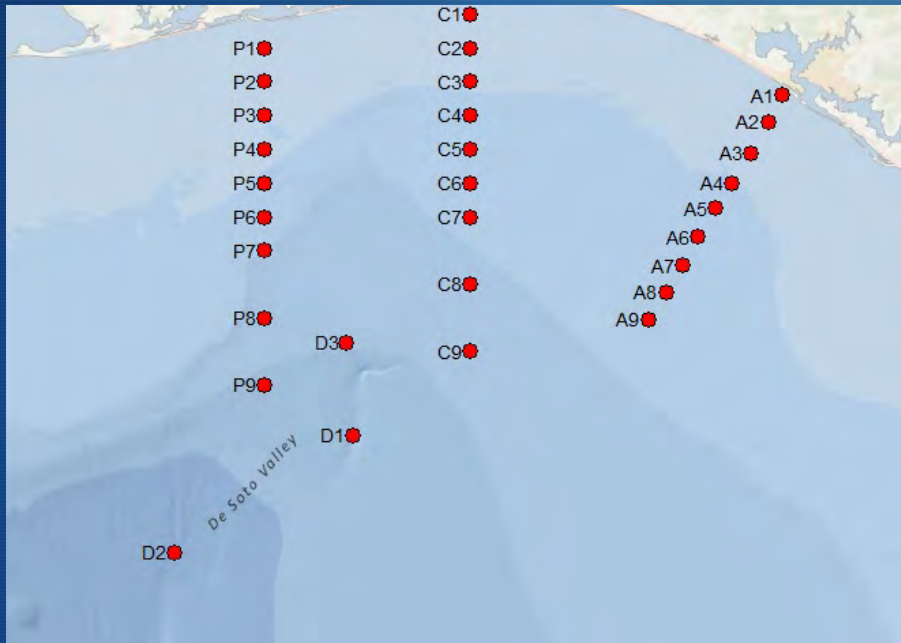
# Sampling Locations



# Water over sediment (core) collection



# Research Cruise

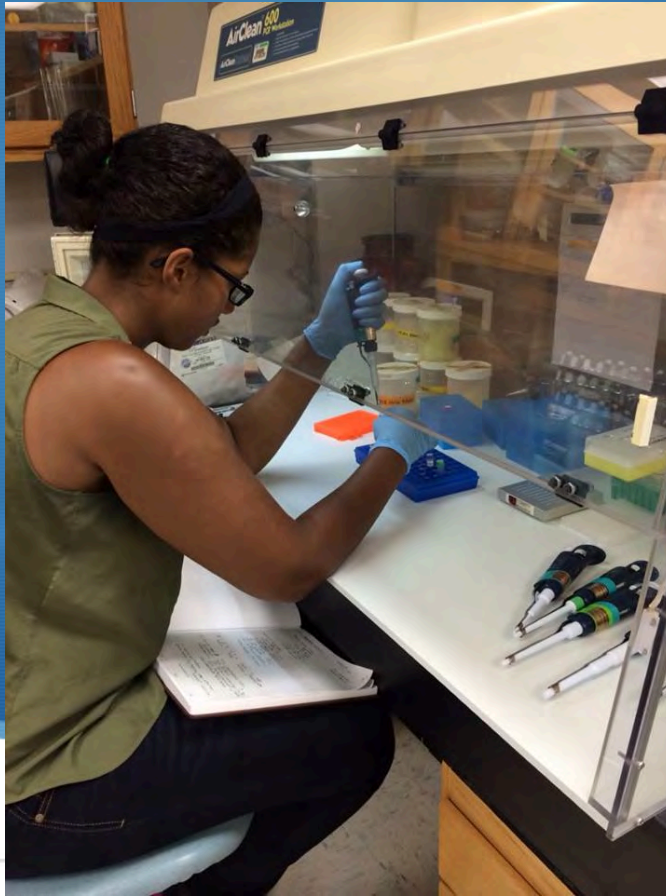




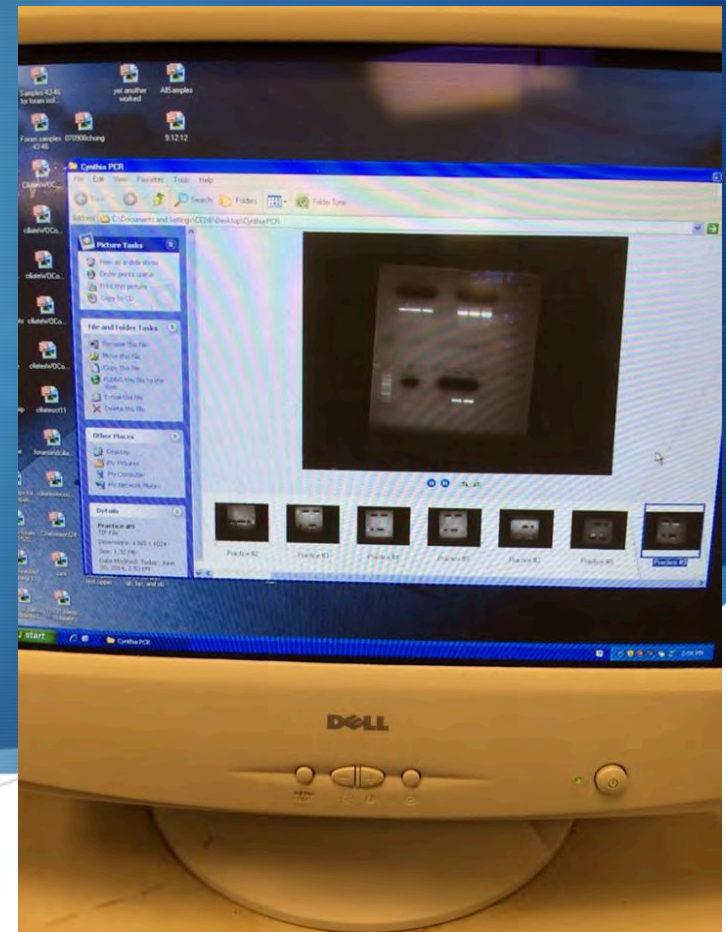
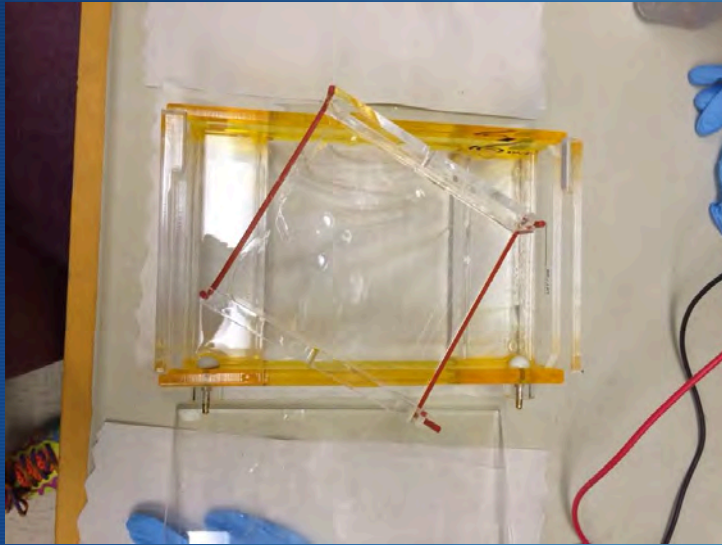
# My project

- ◆ Archaea
- ◆ Examining water over core samples for types of Archaea present
- ◆ Molecular techniques including PCR, gel electrophoresis, clone libraries, and DNA sequencing

# Polymerase Chain Reaction (PCR)



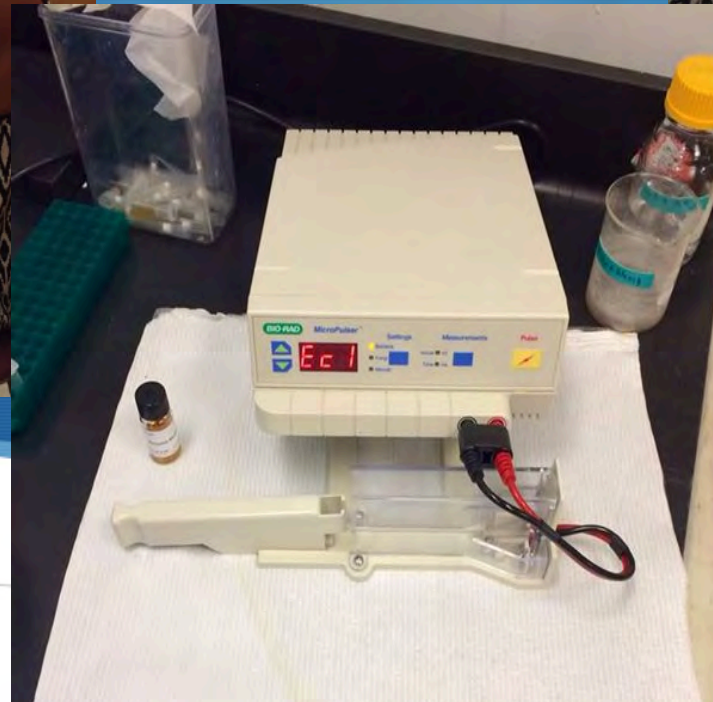
# Gel Electrophoresis



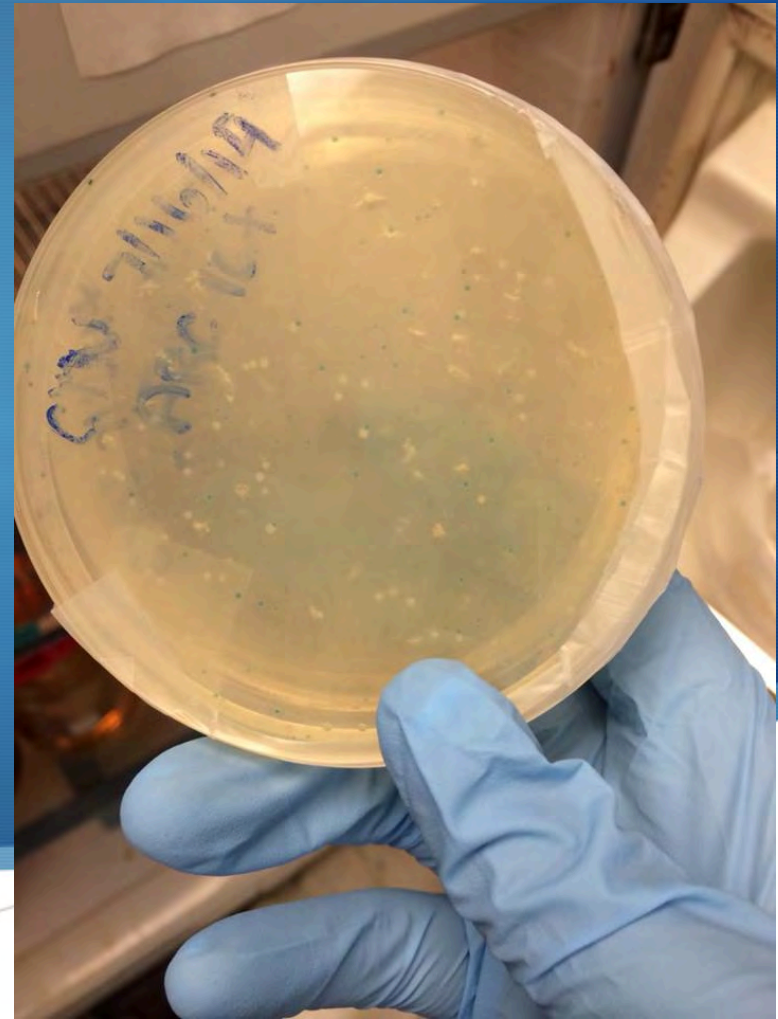
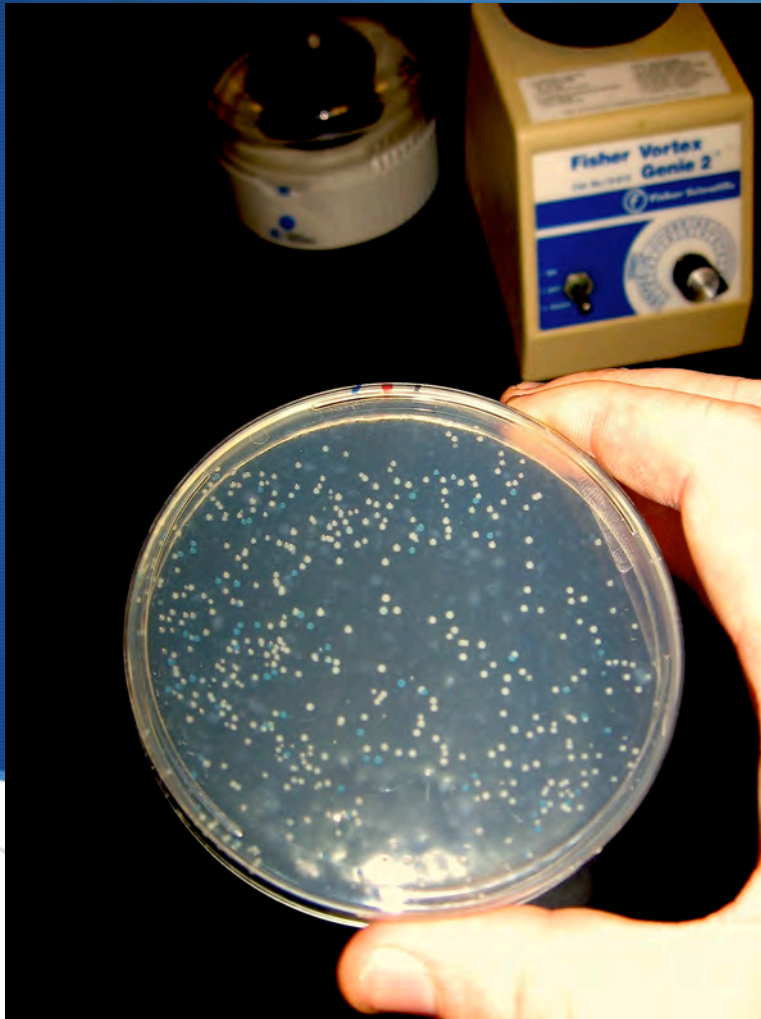
# DNA purification



# Cloning & Transformation Reactions



# Clone Library



# Final Incubation



# Sequence Analysis

The screenshot shows the NCBI BLAST website. The browser address bar displays `blast.ncbi.nlm.nih.gov/Blast.cgi`. The page header includes the BLAST logo, the text "Basic Local Alignment Search Tool", and navigation links for "Home", "Recent Results", "Saved Strategies", and "Help". On the right, there are links for "My NCBI", "[Sign In]", and "[Register]".

The main content area is titled "NCBI/BLAST Home" and contains a search box with the text "BLAST finds regions of similarity between biological sequences. [more...](#)". Below the search box is a "New" button and a link to "DELTA-BLAST, a more sensitive protein-protein search" with a "Go" button.

The "BLAST Assembled RefSeq Genomes" section prompts users to "Choose a species genome to search, or [list all genomic BLAST databases.](#)" and provides a grid of species links:

- Human
- Mouse
- Rat
- Cow
- Pig
- Dog
- Rabbit
- Chimp
- Guinea pig
- Sheep
- Fruit fly
- Honey bee
- Chicken
- Zebrafish
- Clawed frog
- Arabidopsis
- Rice
- Yeast
- Neurospora crassa
- Microbes

The "Basic BLAST" section prompts users to "Choose a BLAST program to run." and lists several options:

- nucleotide blast**: Search a **nucleotide** database using a **nucleotide** query. Algorithms: *blastn*, *megablast*, *discontiguous megablast*.
- protein blast**: Search **protein** database using a **protein** query. Algorithms: *blastp*, *psi-blast*, *phi-blast*, *delta-blast*.
- blastx**: Search **protein** database using a **translated nucleotide** query.
- tblastn**: Search **translated nucleotide** database using a **protein** query.
- tblastx**: Search **translated nucleotide** database using a **translated nucleotide** query.

On the right side, there are two sections: "Your Recent Results" with a link to "All Recent results..." and "News" with a link to "More BLAST news...". The news section features a headline "BLAST in the Cloud Webinar, July 30th, 3:00 PM" and a brief description of the webinar, dated "Mon, 07 Jul 2014 17:00:00 EST".

At the bottom right, there is a "Tip of the Day" section with a link to "More tips...".



# Results & Conclusions

- ◆ Ran 38 PCR reactions, extracted 21 samples, created 2 clone libraries
- ◆ Waiting for sequencing results to come back, continuing analysis of samples
- ◆ Included as author in paper when published

# Skills

- ◆ How to properly use a pipette (how to calibrate it, the correct way to adjust the volume, how much/how little each pipette could hold, etc.)
- ◆ How to use the Thermocycler (used for PCR), including how to program it
- ◆ How to purify DNA using a Qiagen kit
- ◆ How to prepare agar and pour gel plates for the clone libraries
- ◆ How to analyze the clones after incubation
- ◆ How to interpret the results from the DNA sequencing

# Concepts

- ◆ Properties of DNA (denaturation temperature, pH range, etc)
- ◆ The components of the Master Mix and their functions
- ◆ The purpose of using ethidium bromide in the gel for electrophoresis
- ◆ The mechanism behind the Qiagen kit buffers
- ◆ How/why of the cloning reaction (the vectors, shocking the cells, the LB media used)

# Challenges

- ◆ Learning how to use all the equipment/finding my way around the three different labs that were used
- ◆ Learning how to look critically at and not be discouraged by unsuccessful experiments

# Internship Experience

- ◆ Field experience (ship board operations, oceanographic sampling)
- ◆ Valuable lab skills and an understanding of the proper way to work in a lab (the do's and don'ts)
- ◆ First hand experience of the field of microbiology

# Acknowledgements

- ◆ NOAA-NGI Diversity Internship
- ◆ Deep-C Consortium
- ◆ Dr. Richard Snyder
- ◆ Joe Moss
- ◆ Dr. Wade Jeffrey