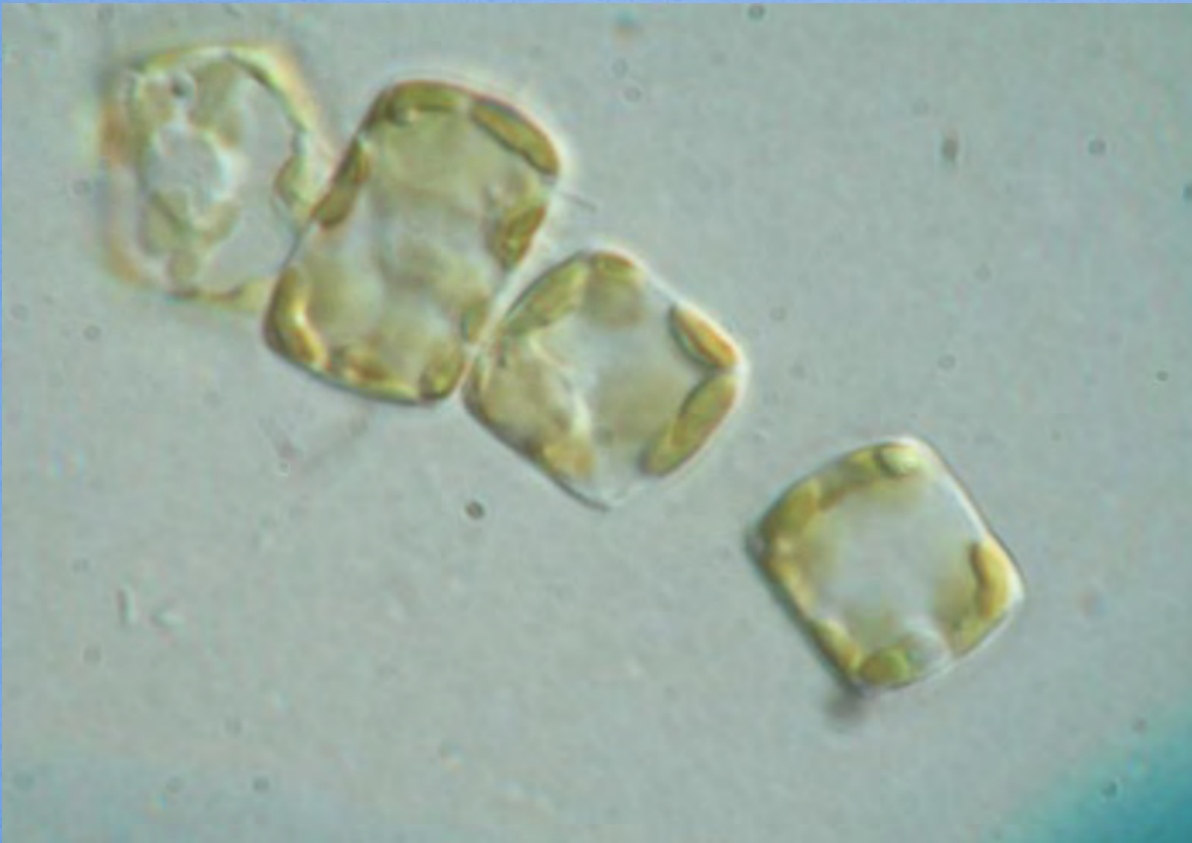


# RET 2013

A Comparative Investigation on the Formation of  
Marine Snow in a Hydrocarbon Contaminated  
Environment vs. an Uncontaminated Environment

Mentor Dr. Uta Passow  
Intern Catherine Borgard

Marine Snow are dead organic materials of various size (1.0 mm and larger) that are *suspended and then settle* to the ocean floor.

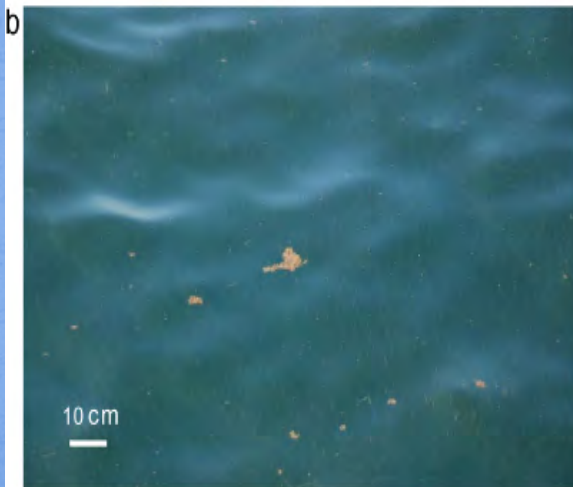
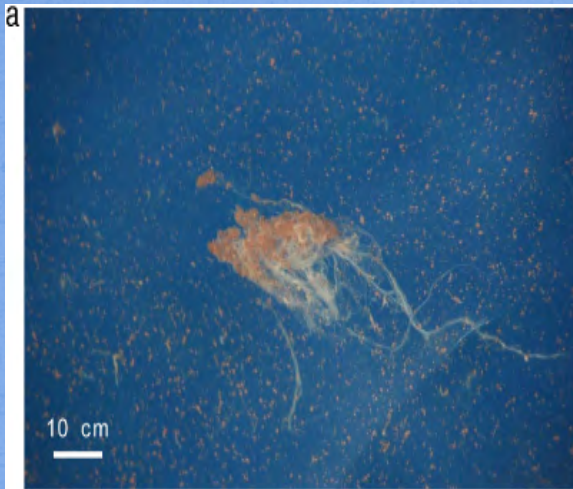


Thalassiosira weissflogii phytoplankton were used in this study as they grow well in culture and they are found in the Gulf of Mexico (GoM).

Image of *Thalassiosira weissflogii* (Grunow) G.A. Fryxell & Hasle  
Nordic Microalgae and aquatic protzoa



# Examples of Aggregates Observed in the GoM



Aggregates sink down and become Marine Snow. This example was found in the GoM after the Deepwater Horizon contamination.

[iopscience.iop.org](http://iopscience.iop.org)



# First Steps in the Quest for Marine Snow



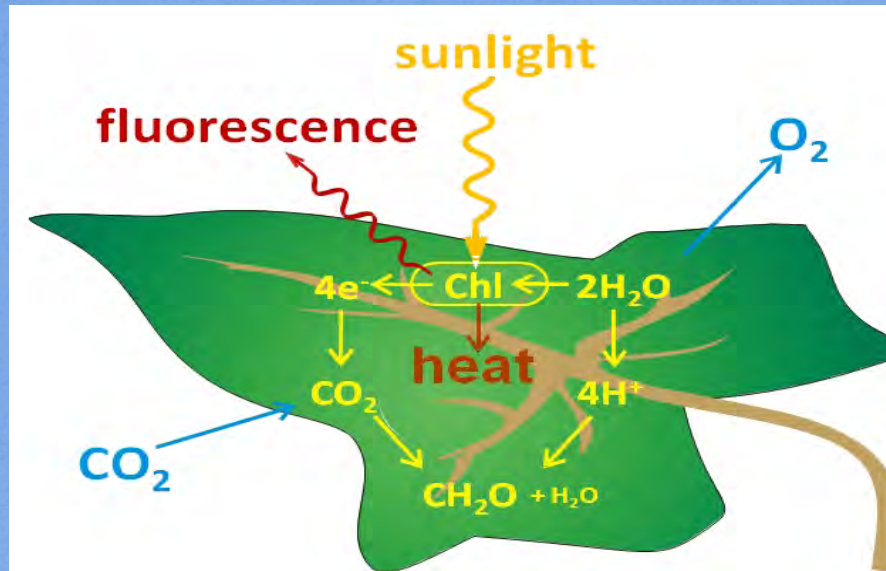
Create nutrient rich artificial seawater to “grow” a sufficient quantity of phytoplankton.

Wait.

Check every day to see if the inoculated seawater is “growing”.



# Fluorescence is the first standard.



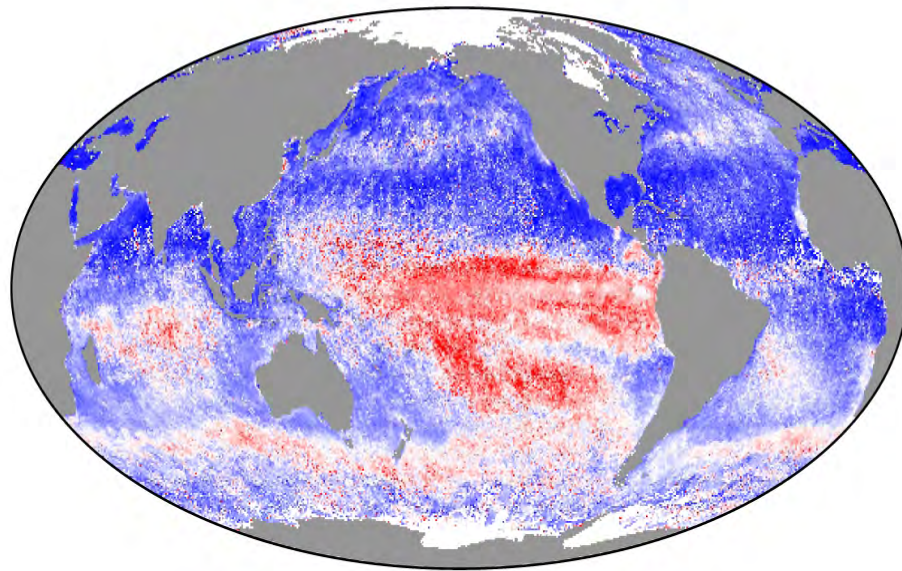
Using the AquaPen, fluorescence is determined. The higher the rate of fluorescence, the higher the rate of photosynthesis. This gives a quick idea of population growth.

<http://www.phenovation.com>





# Practical Applications of Fluorescence



NASA uses fluorescence to monitor how much CO<sub>2</sub> is removed from the atmosphere by phytoplankton by the photosynthetic process.

[Science1.nasa.gov](http://Science1.nasa.gov)



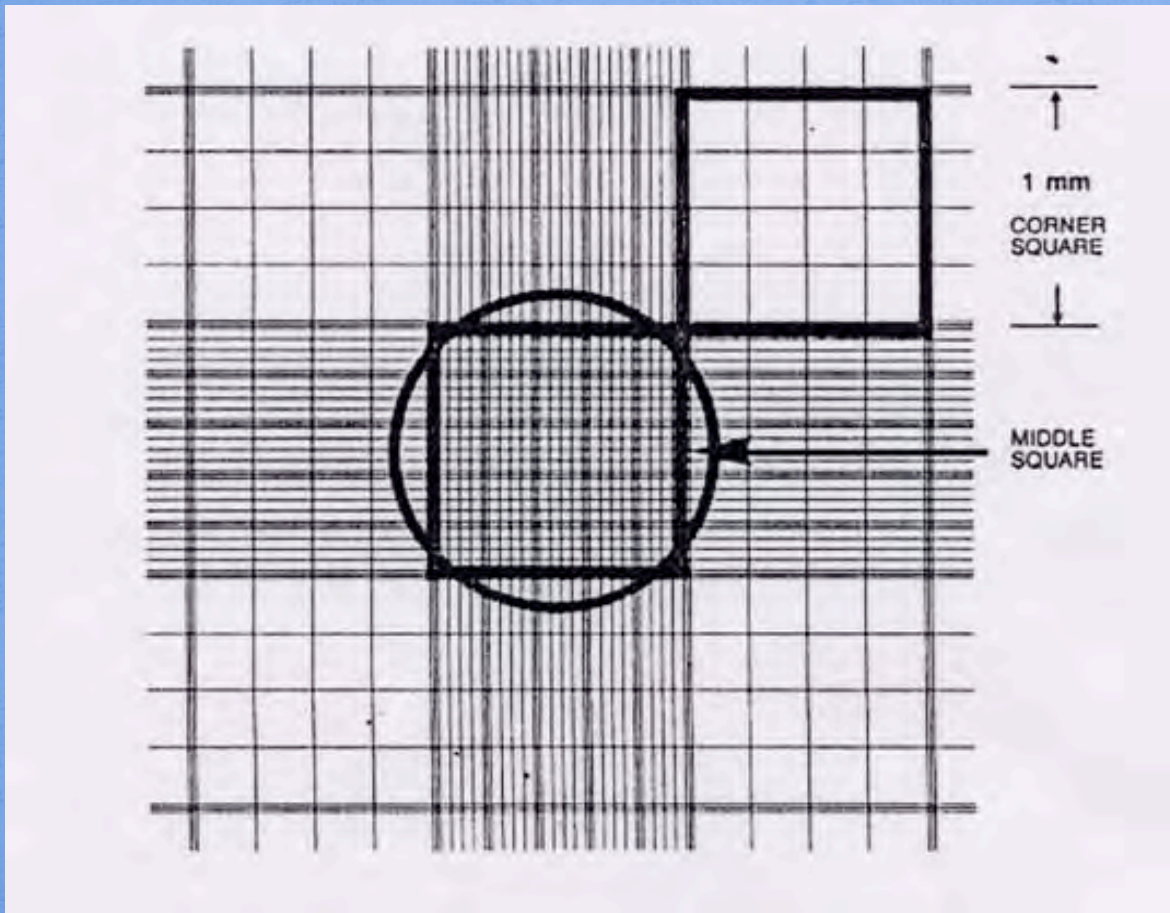
# Next Step is the Physical Counting



Magnification is at 800x the actual size of the organism. Phase contrasting was also used as this helped to visually “pop” the image of the phytoplankton (*Thalassiosira weissflogii*) into view.



# Hemocytometer Slide for Accuracy

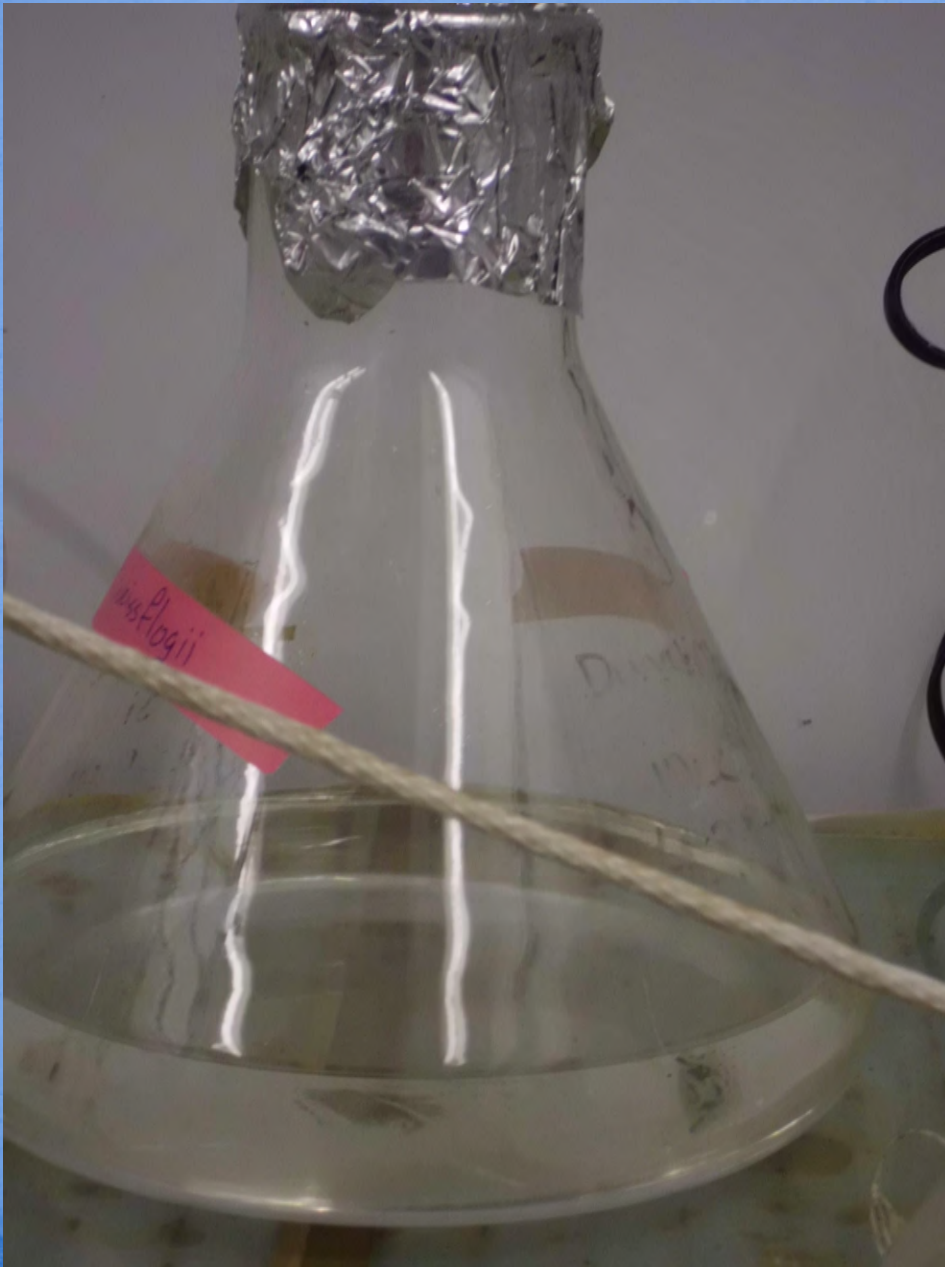


A hemocytometer slide allows one to calculate the population of cells per Liter of fluid. A count of 200 cells gives an approximate 20% percentage of error.



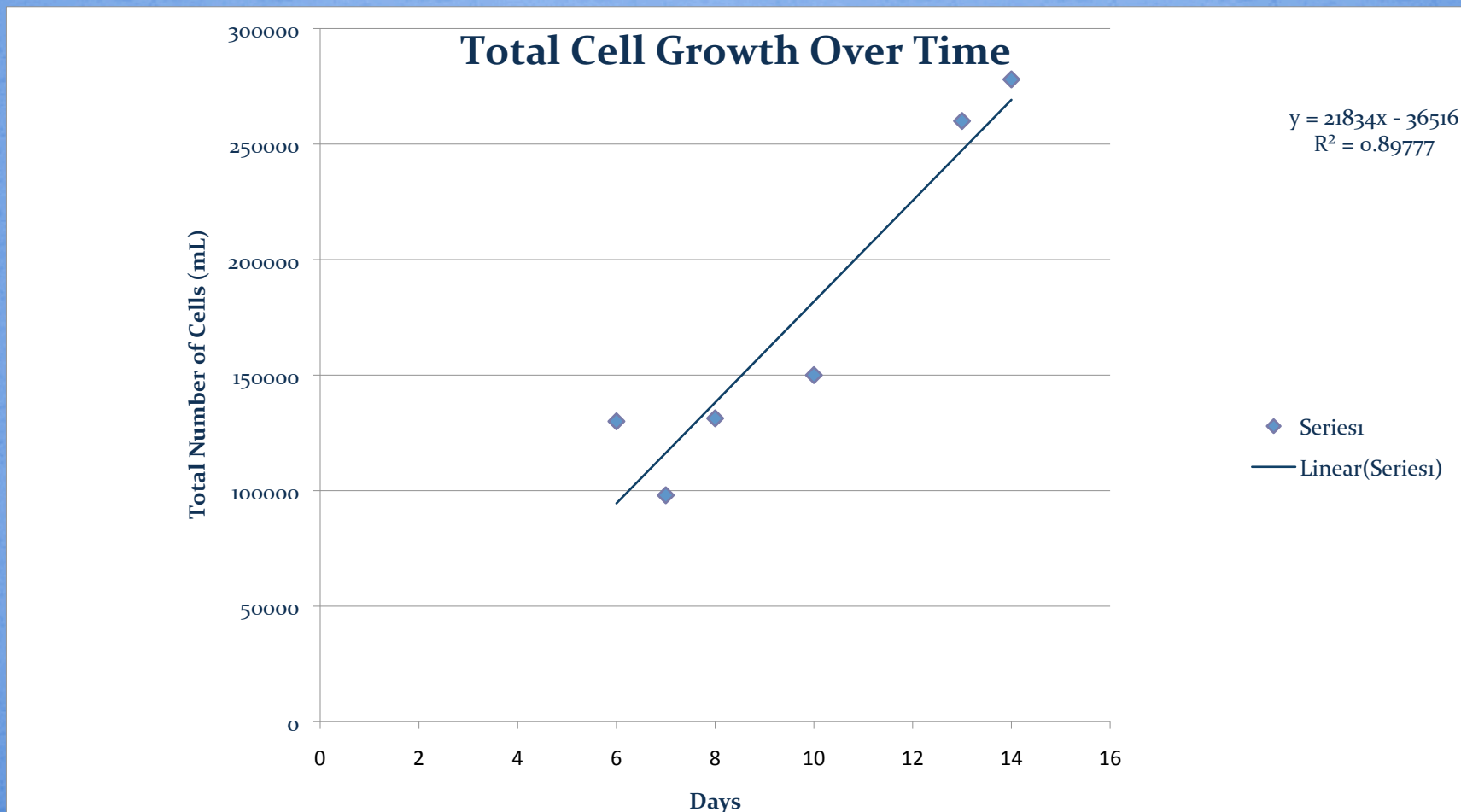
# First Results

12 Days have passed and the phytoplankton are not growing as expected. The clarity of the artificial seawater is one indicator that growth is not occurring.



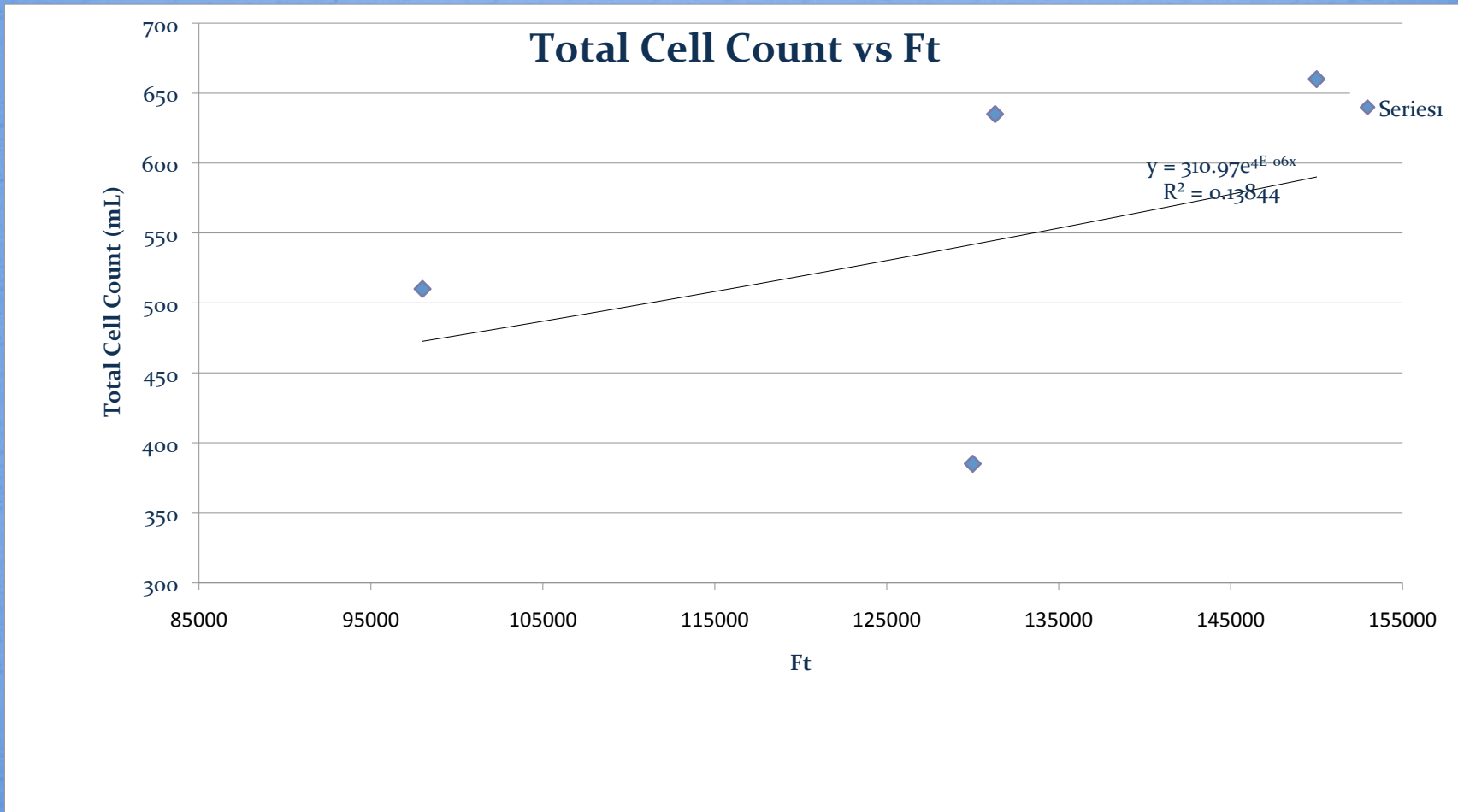


Initial growth was not what I had hoped for. The graph demonstrates linear growth, not exponential.





The linear trend is seen again with the quantity of fluorescence (Ft).





# Photosynthesis Assistance

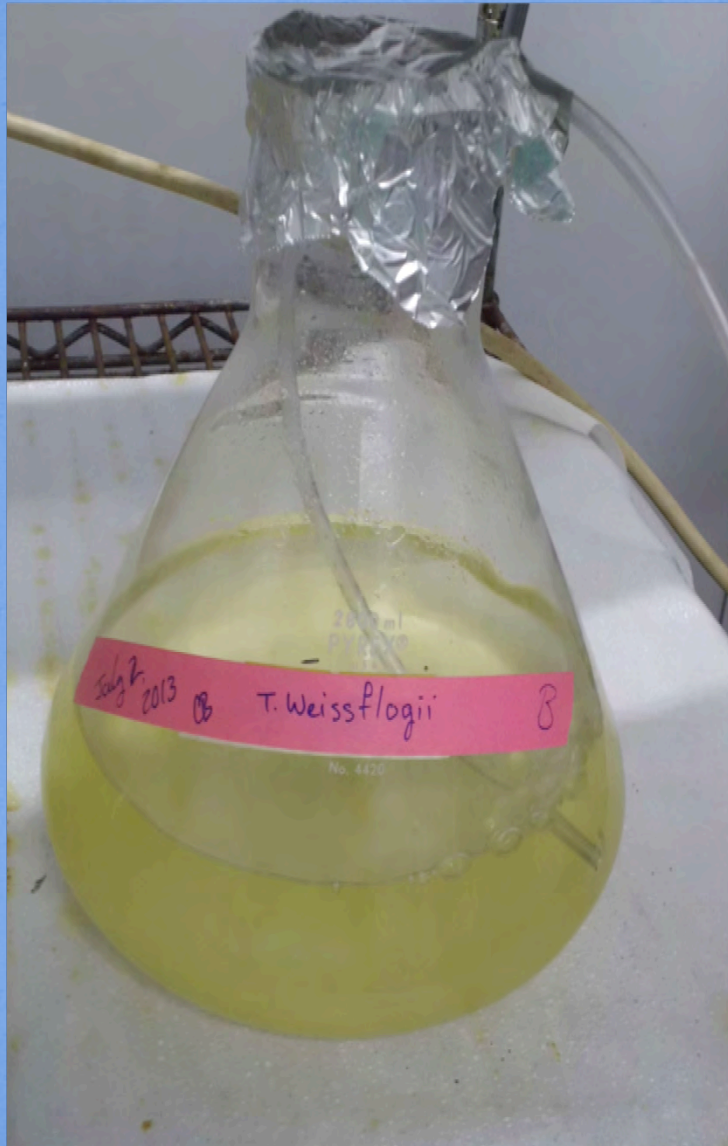


To assist the growth of the phytoplankton, CO<sub>2</sub> was bubbled through the media. A higher intensity of light was also used, but not the quantity of light. That remained at 12 hours on and 12 hours off.

Carbon Dioxide Bubbling Device



# The New Results

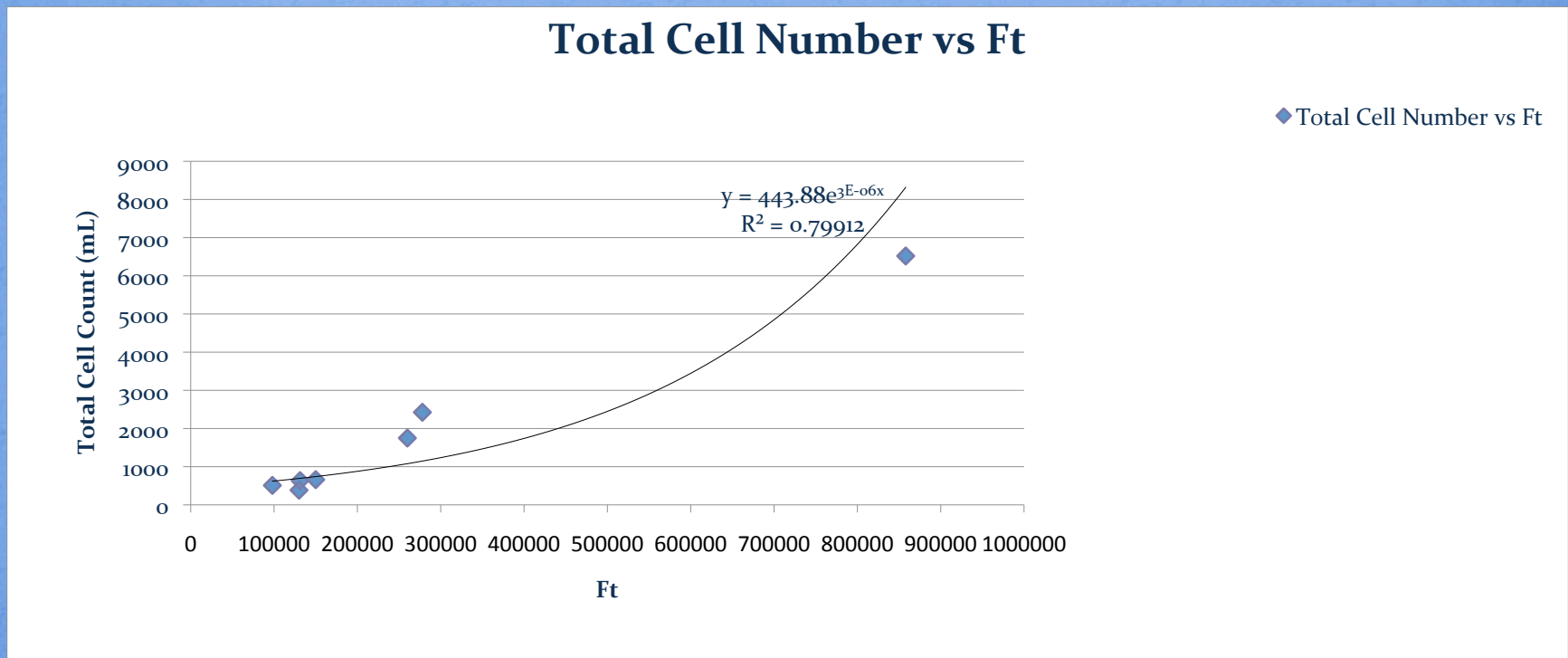


The results were immediately visible. The phytoplankton population grew to a density where the media changed to a green color. Note the tube in the flask delivering CO<sub>2</sub>



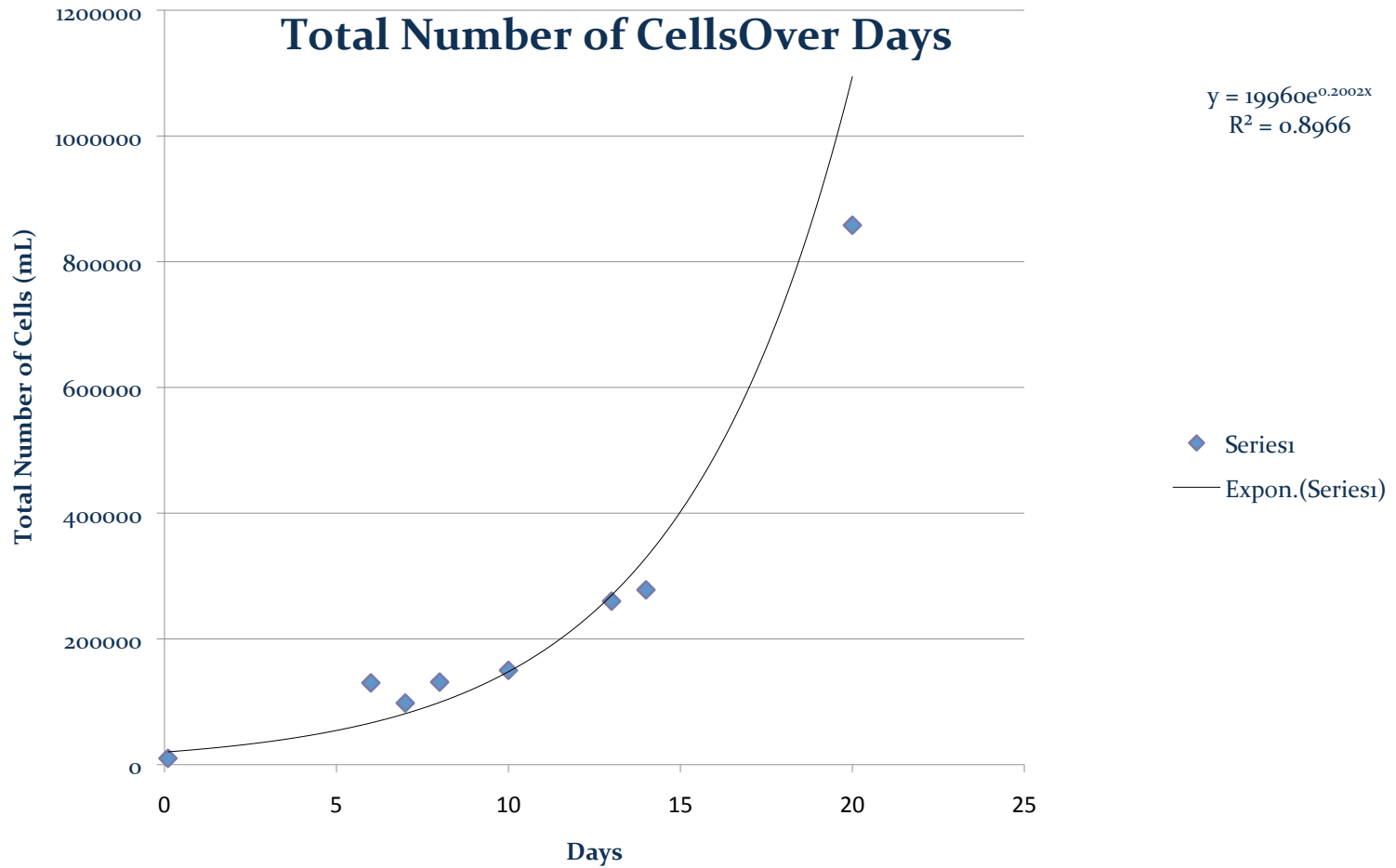
# Approximate Population

Ft measures fluorescence which indicates population size. Population jumped over the July 4<sup>th</sup> weekend (4 days).

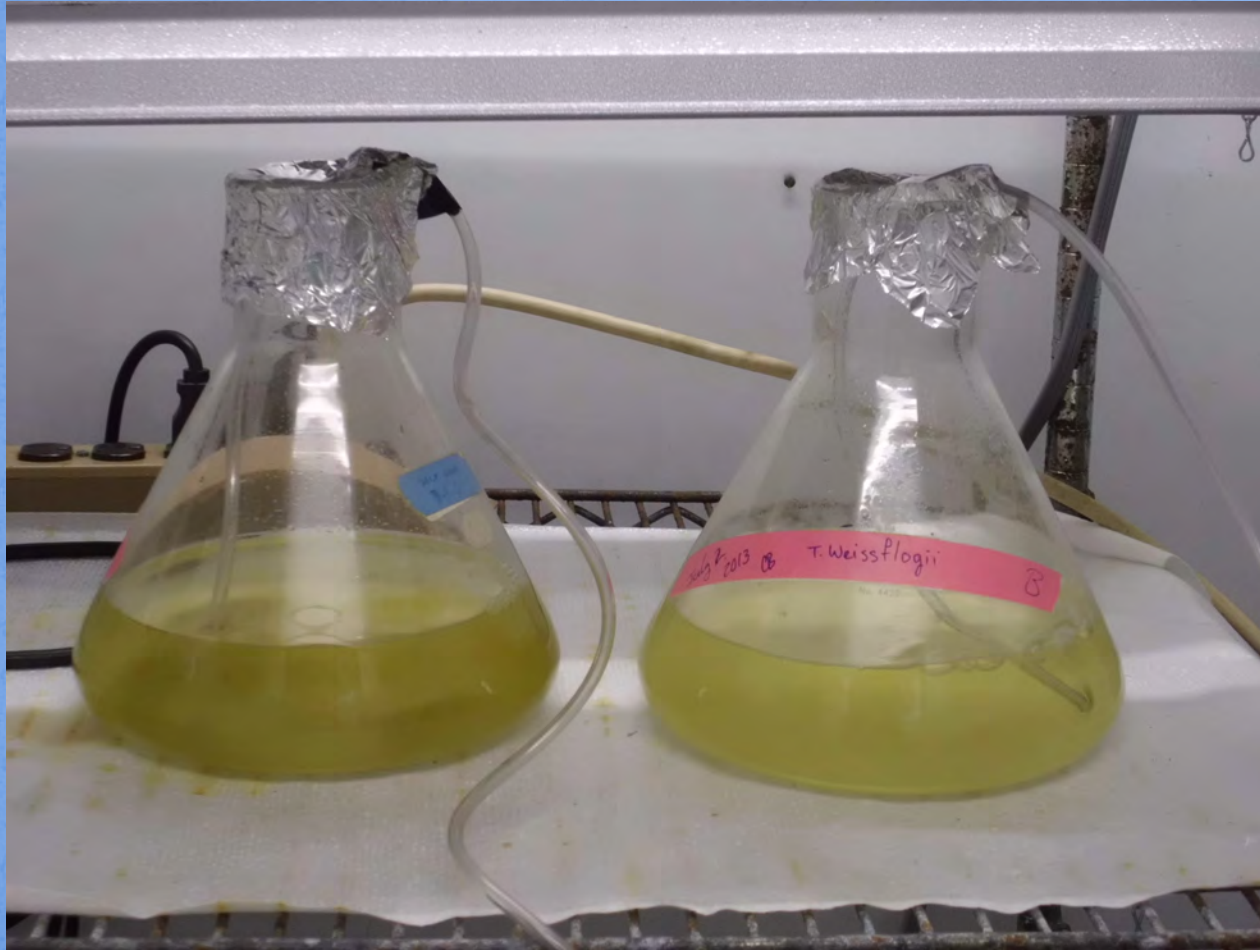




# Population Growth Over Time



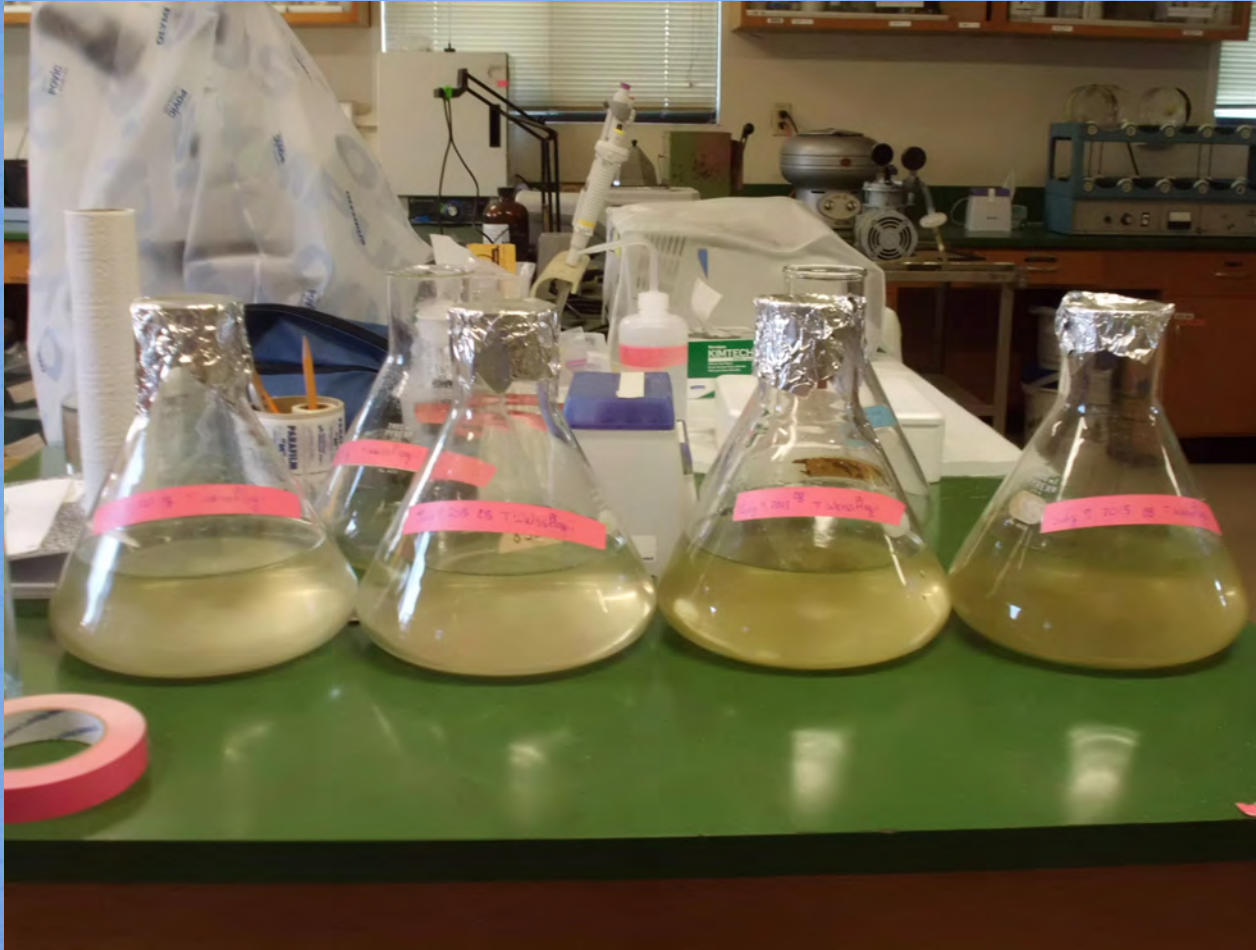
# The Quest for Marine Snow Cont.



As the phytoplankton grow, the culture is divided and added to new media. With  $\text{CO}_2$  and light the phytoplankton continues to grow.



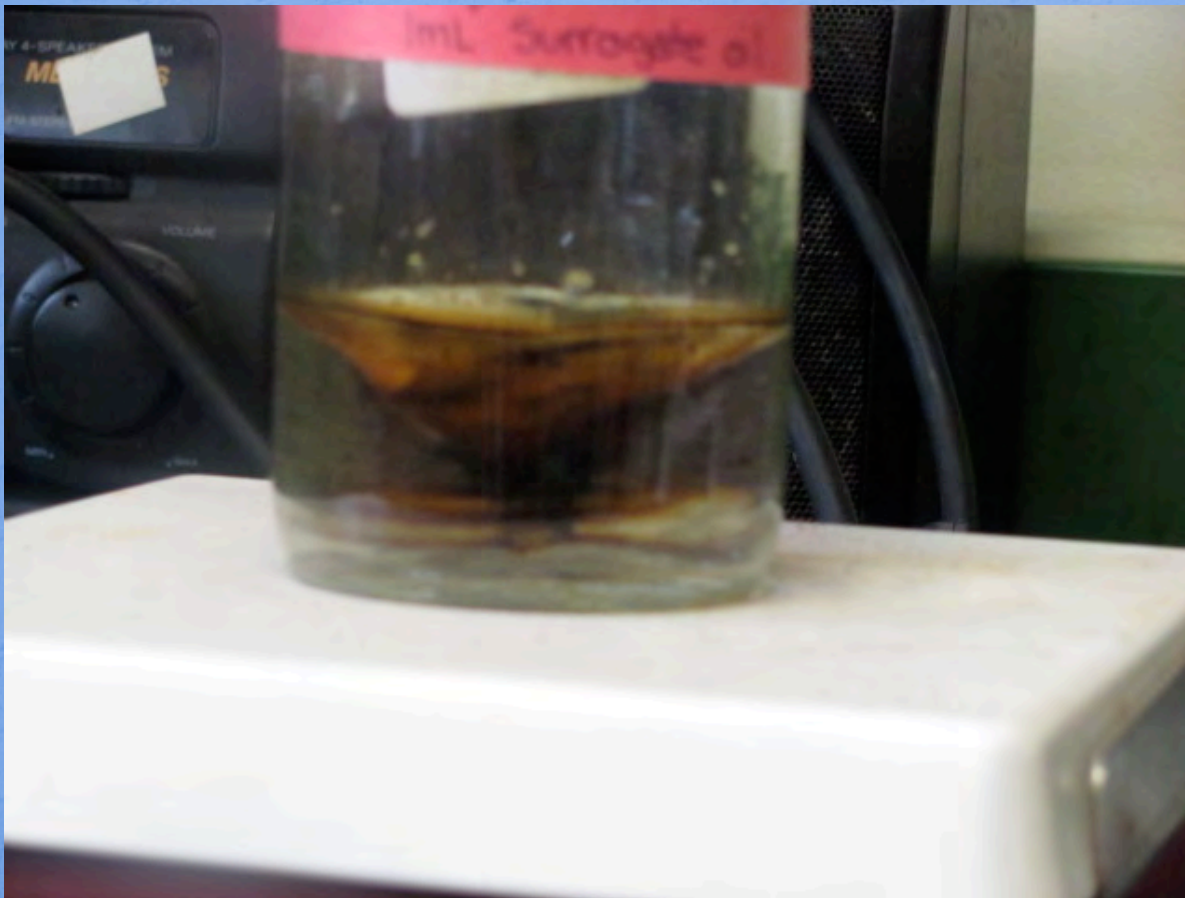
# More Phytoplankton is Needed



The process is repeated. This creates just over 6 L of phytoplankton filled media.



# Creating GoM Oil/Water Suspension Known as WAF (Water Accommodated Fraction)



Three experimental groups are created. One has Hydrocarbon Contaminated seawater forced into a suspension (WAF).

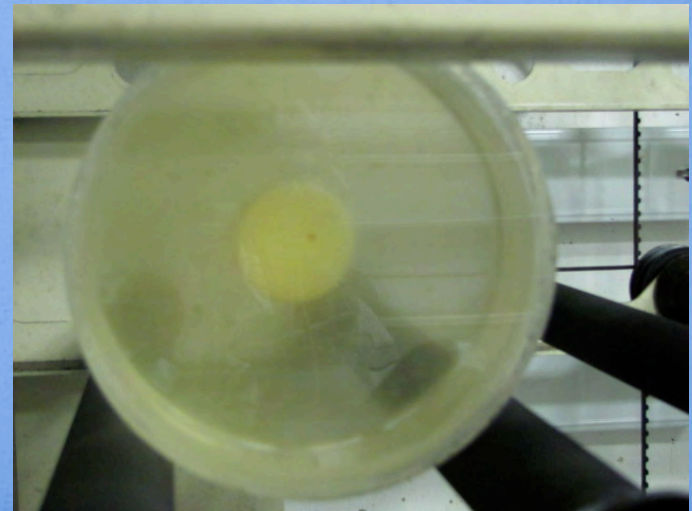


# No Bubbles Allowed!



Great care is taken to ensure No bubbles are in the tanks. The rolling simulates movement in the deep sea where there are no surfaces.

The six tanks are filled with an equal amount of phytoplankton media that had all been mixed (the 4 flasks) and topped with artificial seawater.





# Control Tanks and WAF Tanks

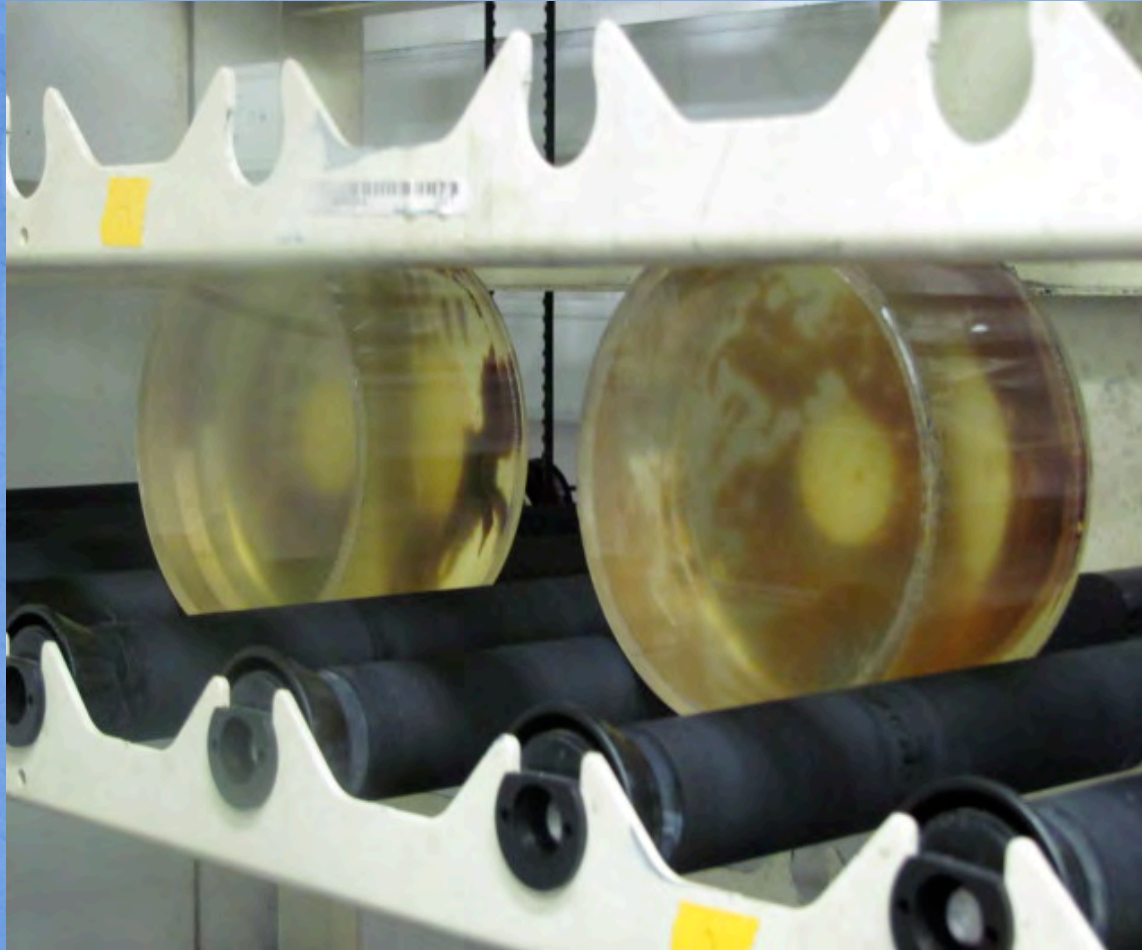


The first two tanks are controls with NO contamination from hydrocarbons.

The back two are contaminated with hydrocarbons via WAF.



# Direct Application of GoM Oil



1 mL of Oil from depth of the Deepwater Horizon was added to each of these two tanks.

# Aggregate Formation = Marine Snow!



Aggregates slowly form over days of undisturbed rolling. The tanks are kept at 13.5°C and in the dark to simulate the deep ocean. Average size of aggregate 1mm x 1mm.



# Collecting Aggregates: Quest Cont.

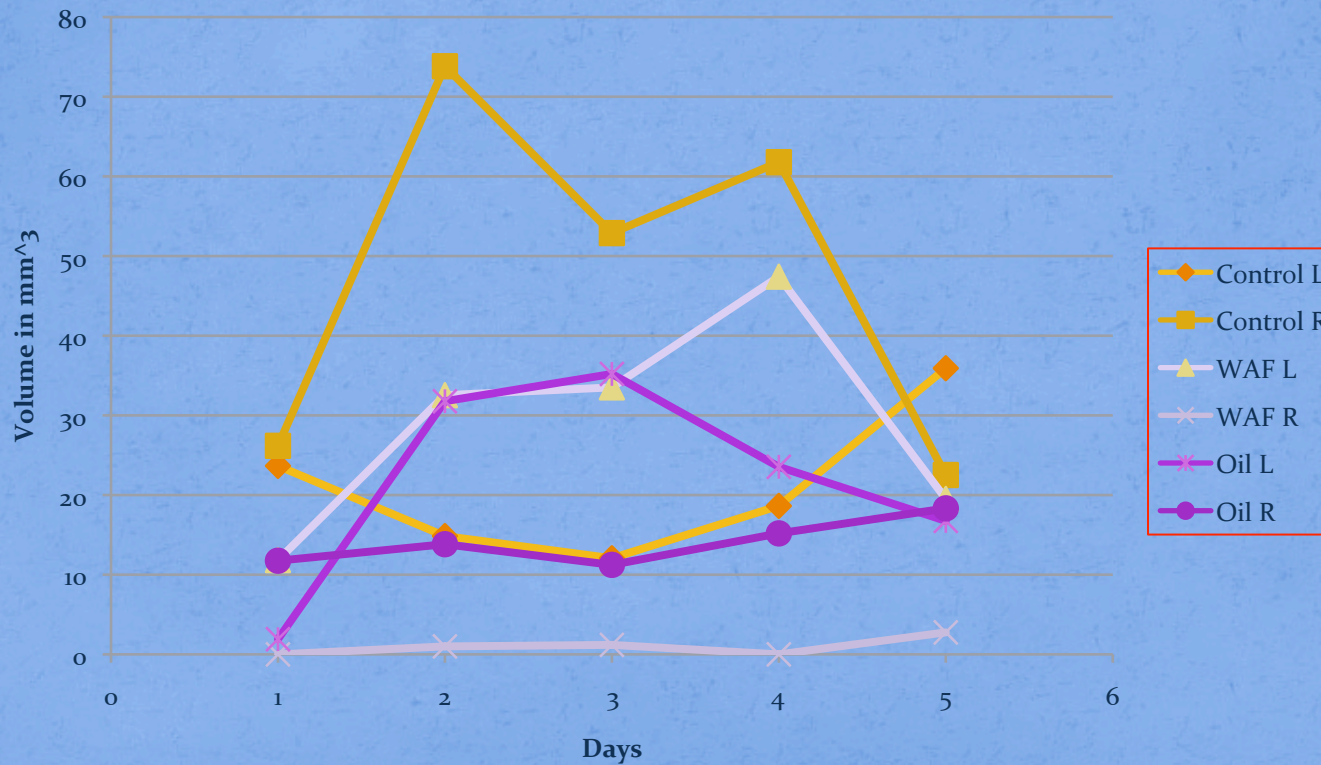


Aggregates are carefully removed. Duplicate dry weights are calculated after filtration. Samples of phytoplankton cells were also collected.



# Volume of Aggregates Measured

## Volume of Aggregates Over Time





# The Quest vs. Reality Check



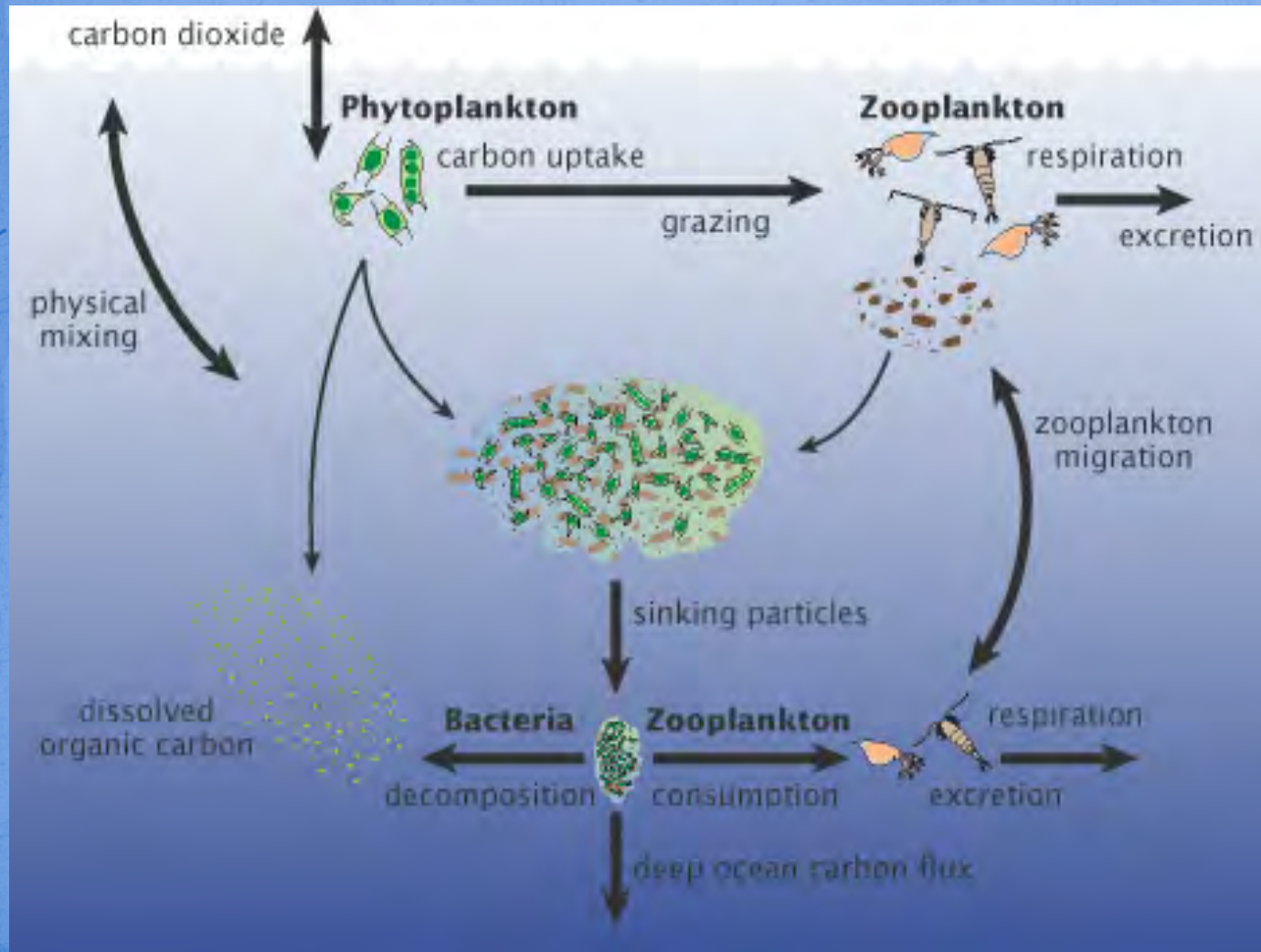
In my Quest there are two replicates of each: control, WAF, and oil. In reality, there can be more replicates but sometimes not. Plus, the tanks are left in situ far longer than in my Quest.

# Comparisons by Dr. Passow

Marine Snow Type	Type of Oil	$\delta^{13}\text{C}$ determining association oil with marine snow (%)
<i>D. fragilissimus</i> agg.	Spill oil from oil carpet	16% Marine Snow associated with oil
<i>D. fragilissimus</i> agg.	Macando Oil	65% Marine Snow associated with oil
<i>T. weissflogii</i> agg	Cold seep oil	91% Marine Snow associated with oil



# Carbon Pump: The Big Picture

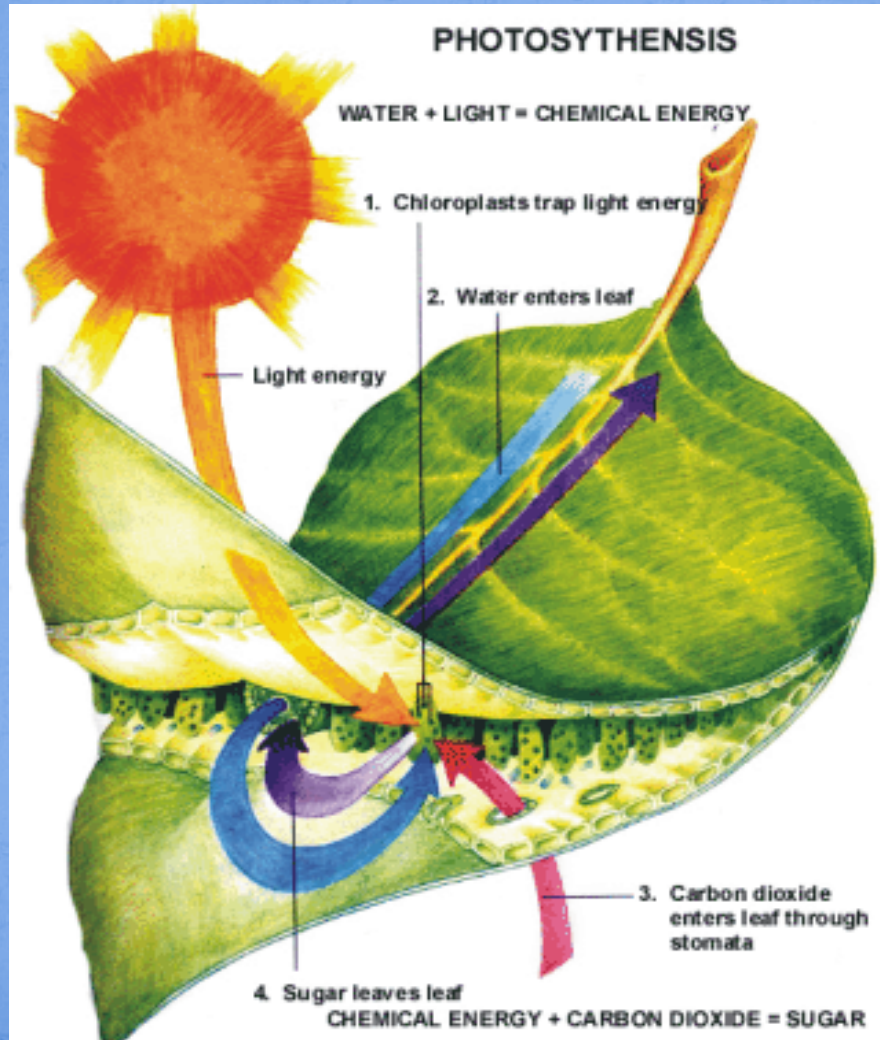


How does all this scientific information apply to the classroom?

Conceptual Education: Everything Interacts with Everything

<http://www.skepticalscience.com/pics/Biologicalcarboncycle.jpg>





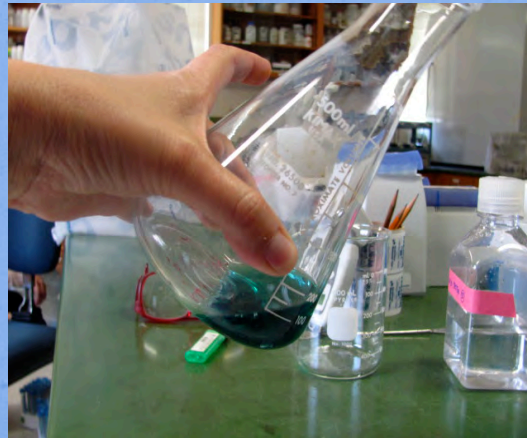
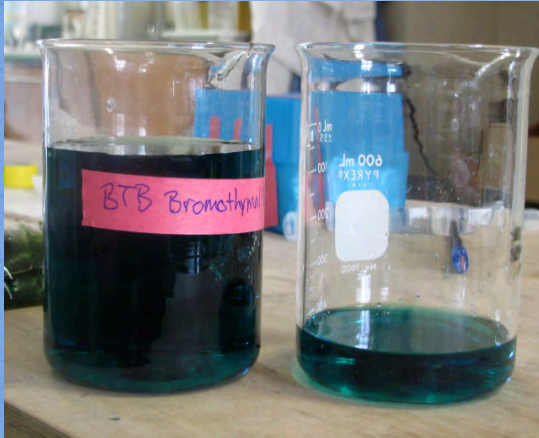
Photosynthesis is a basic concept and yet my students fail to grasp it as a “fact”.

As a concept, I hope to apply the role of Carbon in the students lives and then expand upon it.

[http://  
courtneystanifer.edublogs.org/  
files/2010/05/photosynthesis](http://courtneystanifer.edublogs.org/files/2010/05/photosynthesis)



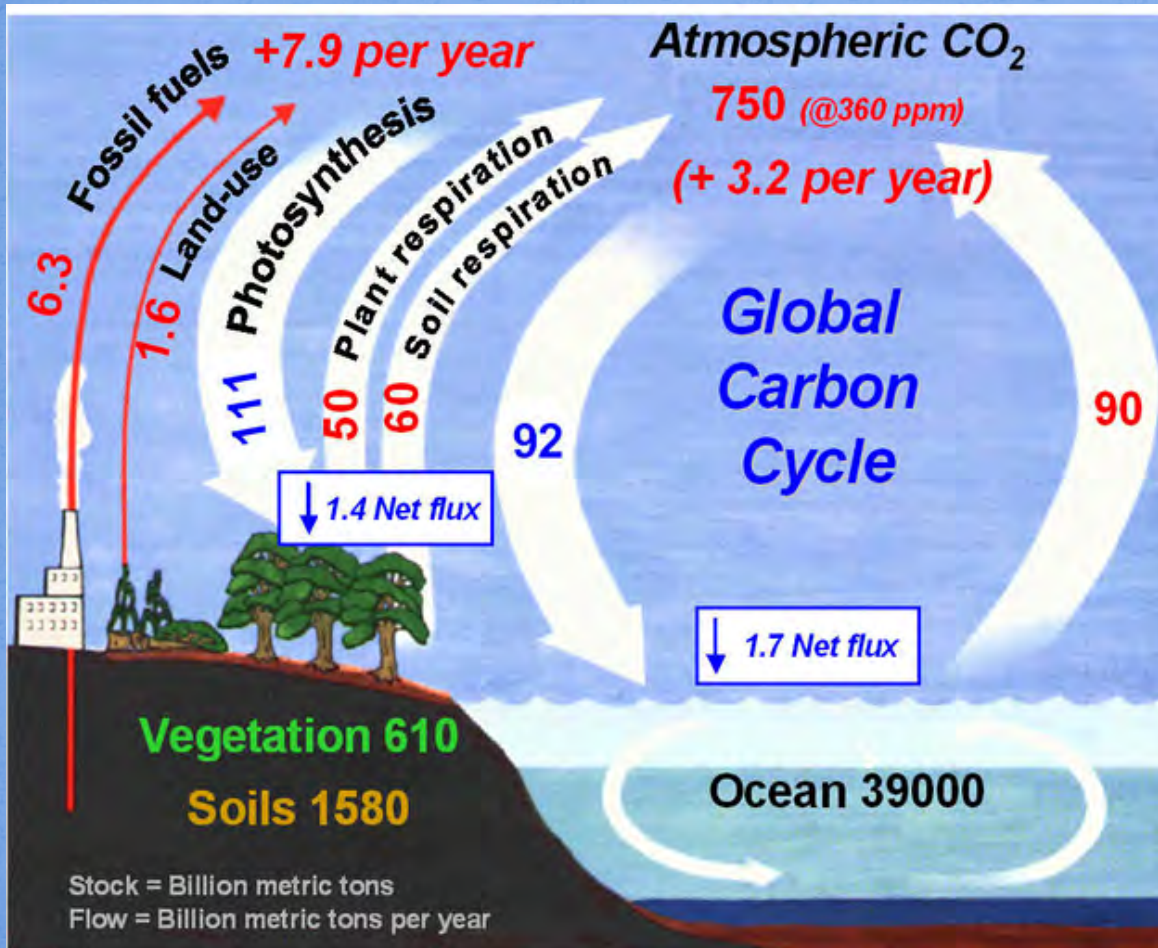
# Student Hands-On Applications



This lab demonstrates the release of CO<sub>2</sub> from burning organic matter and exhalation, plus the release of O<sub>2</sub> from plants.



*The frog does not drink up the pond in which it lives – Indian Proverb*



. . . And as one concept is grasped it will lead to a larger concept. The ultimate goal will be the understanding that all things are interconnected and we are connected too. [http://carbon\\_cycle/carbon\\_cycle.jpg](http://carbon_cycle/carbon_cycle.jpg)



# Thank You for a Great Summer

RET 2013

Thank you so much  
for this opportunity.



Special thanks to:  
Dr. Frank Kinnaman  
And  
Mary McGuan for  
encouraging me to  
participate.



Julia Sweet – Wealth of  
Knowledge

Mentor  
Dr. Uta Passow

One of the Best  
Instructors I have  
ever had – thank  
you.



Julie Gonzalez – Master of  
Excel

And a thank you to Philip Becker who just happened to be walking by at the right time – Mr. Bubble