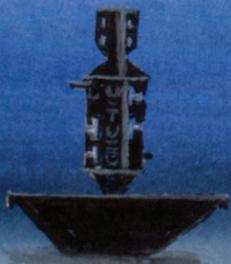
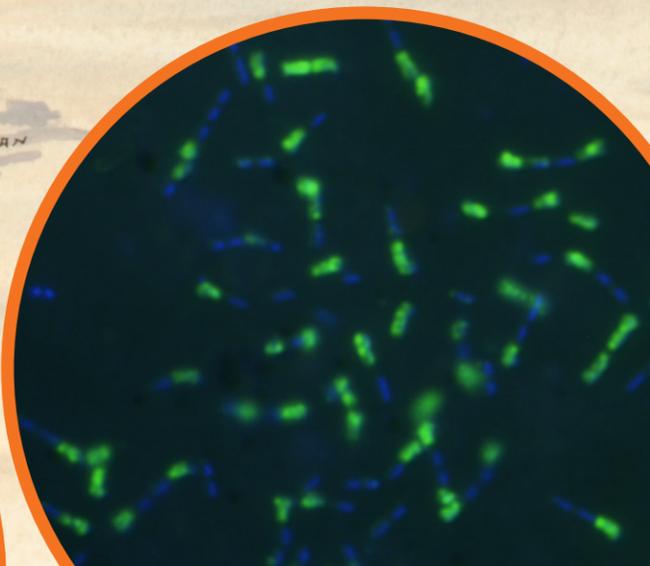
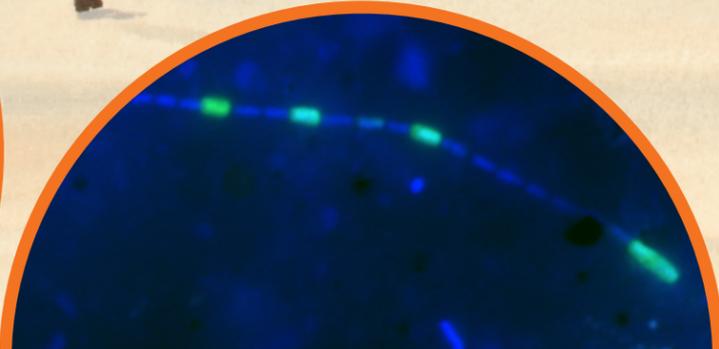
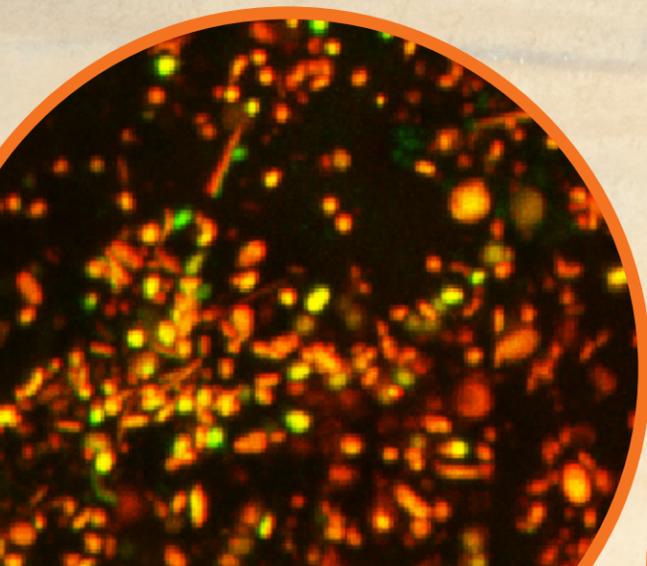


Searching for the Intra-terrestrials:

Microbiology beneath the Seafloor



DINAH BOWMAN



Tagging A Microbe - Teacher Guide

Overview

In this activity, students use Lego blocks to learn one method microbiologists use, called Fluorescence *In-Situ* Hybridization (FISH), to “tag,” identify, and study microbial diversity found deep below the sea floor. Students use the Lego pieces to build genetic strands of different bacteria and the probes that will tag them. This activity can be used with very little student background knowledge if pre-activity explanation is simple and vocabulary is kept to a minimum. It can also be used concurrently or after material on RNA base pairing is taught.

Objectives

1. To understand how RNA base pairing allows microbiologists to design fluorescent probes that attach to unknown microbes in a deep sea environment.
2. To learn the role the *JOIDES Resolution* plays in exploring the deep biosphere.

National Science Education Standards

A - Science as Inquiry

C – Life Science

E – Science and Technology

Ocean Literacy Principles

5. The ocean supports a great diversity of life and ecosystems.
7. The ocean is largely unexplored.

Background

In 1998, Whitman, et al. proposed the idea that 72% of the Earth’s prokaryotes (bacteria and archaea) live in the deep ocean sediments. Whitman did not include rocks at and below the ocean floor, but we now know from drilling expeditions on the *JOIDES Resolution* (*JR*) that there is a substantial number of bacteria living in the subsurface environment. By some calculations, it is estimated that 1/3 of the Earth’s entire biomass lies below the ocean floor. This concept is so compelling that in 2003, the Integrated Ocean Drilling Program (IODP) made studying sub-seafloor microbial diversity one of the main focuses for drilling expeditions.

Microbiologists working on the *JOIDES Resolution* are asking these questions:

1. What kind of bacteria are living in the rocks and sediments below the seafloor and how many are there?
2. How are the different microbes living and interacting with biotic and abiotic factors below the sea floor?
3. How do these bacteria survive in such extreme environments?
4. How are they affecting local and global ocean chemistry and biomass?

Because of the unique technological capabilities of the *JR*, scientists have been able to install CORKs (subseafloor observatories) in the ocean floor. The CORKs were originally designed to measure physical properties, but are now being adapted to collect and



measure microbes to learn more about the deep biosphere. One technique being used is Fluorescence In-Situ Hybridization or FISH. During FISH, the membranes of collected bacteria are made permeable to allow genetic “probes” to pass through and come into contact with their rRNA. Each type of probe is a small strand of base pairs attached to a backbone that has been designed as the compliment to a section of rRNA for a specific bacteria. The effectiveness of FISH relies on the rules of base-pairing; Adenine (A) binds to Uracil (U); Guanine (G) binds to Cytosine (C). Therefore a probe with the sequence AGCGUUGCA will bind to a cell that has the compliment (UCGCAACCGU). Once the rRNA of a cell is tagged with the probe, a fluorochrome is attached to the probe that makes the bacteria light up in an epifluorescence microscope so it can be identified and counted. This same technique is used medically, for example, to identify cancer cells.

Materials

- For each group:
 - (4) 8X1 Lego bricks – any color
 - (4) 4X1 Lego bricks – any color
 - (17) red 1X1 Lego bricks
 - (14) blue 1X1 Lego bricks
 - (4) black 1X1 Lego bricks
 - (9) yellow 1X1 Lego bricks
 - (4) green 1X1 Lego bricks with four different glow in the dark stickers
- Class set of bags to hold Legos
- 1 viewing box
- Marker and athletic tape to label bacteria probe backbones



Advance preparation:

1. Assemble bag kits for student groups, and expose the green bricks to light so they will glow.
2. Make a viewing box using a shoebox with a 4” cardboard roll inserted in the top.
3. Print student page.

Alternative: To make larger strands, tape two or more together to make 16X1 bricks and replace the 1X1 bricks listed below with 1X2.

Reference materials:

- The *JOIDES Resolution*: www.joidesresolution.org
- *JR* – Expedition 327 Juan de Fuca: joidesresolution.org/node/1154
- *JR* – Expedition 330 Louisville Seamount: joidesresolution.org/node/1622
- Thinkquest Cellupedia: library.thinkquest.org/C004535/
- Advanced Readings-deep biosphere research: earth.usc.edu/~kje/pubs.html

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To learn more, visit:

www.darkenergybiosphere.org and www.joidesresolution.org

