

TITLE: Incorporating Biotechnology in the High School Classroom: Professional Development for Biology Teachers.

PRINCIPLE INVESTIGATOR:

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ABSTRACT: The purpose of this project is to develop 3-4 professional development sessions aimed towards life science teachers. Using the emerging pathogens as the theme, biotechnology skills and equipment will be employed to educate teachers on how these skills and equipment can be incorporated into their current curriculum.

RATIONALE: Making changes can be difficult and this difficulty in making change is evident in many science classrooms. Many science teachers come to the classroom lacking laboratory skills necessary to be able to incorporate current technologies into their curriculum. Add to this administrative and district mandates such as curriculum pacing guides and pressure due to high stakes testing, it is understandable that teachers will teach as they have been taught and within their comfort zone. This scenario is evident in my current school. It is hoped that by including the life science teachers at my school in several interactive professional development sessions utilizing some of the current technologies available, these teachers will develop knowledge that will allow them to introduce new ideas and activities using technology into their curriculums.

DESCRIPTION OF TEACHING UNIT OR MODULE, INCLUDING EXPECTED OUTCOMES: This unit will require three or four hours of professional development spread throughout the first semester of the 2011-2012 school year. A pre-assessment survey will be given to participating teachers to determine their knowledge of and comfort level in using technologies that will be taught. A post-assessment survey will be given at the end of the professional development.

Module 1 – The first module will consist of introducing the teachers to pipetting skills. This is a skill that has not been seen in my school. While we have a few pipettes, they have not been used extensively. The pipetting locker will be obtained from ICORE at the University of Florida and used to introduce teachers to pipetting. Using money from the ASBMB and ICORE, pipettes will be purchased and an equipment locker for Celebration High School will be started.

Module 2- This module will introduce teachers to antigen/antibody testing and Microarrays through the simulation of A Medical Mystery of Epidemic Proportions from Science Take Out. Using a cholera epidemic as the basis, participants will attempt to discover why people who have been vaccinated against cholera or have had cholera in the past are becoming sick with cholera. Other simulation activities will be developed and incorporated into an equipment locker for future use.

Module 3 – Building on the previous two modules, the third module will use restriction digestion and gel electrophoresis to examine bacterial DNA. This activity will use the bacterial kit from Carolina Biological, Outbreak, to teach these skills. Our school has a few of the gel electrophoresis kits where agarose gels must be prepared ahead of time. Teachers will be taught how to do this and how to run the gels with this equipment but E-Gels from Bio Rad will be purchased and placed into an equipment locker for use

by teachers of Celebration High School. Preparation of the agarose gels and staining of those gels is time consuming. Celebration High School has 52 minute periods so using E-Gels, preparation time can be minimized.

Module 4 – En Elisa Antibody Test will also be one to continue with the cholera theme, building on the three previous modules.

The expected outcomes are that participating teachers will develop a level of knowledge and skills that will allow them to incorporate biotechnology into their classrooms. Skills learned:

- Pipetting
- Gel Electrophoresis
- Use of simulations such as ELISA Testing and DNA analysis
- How to prepare agarose gels
- Writing curriculum modules to use with the equipment lockers

Many of the teachers at our school are not prepared to incorporate the activities described above into their curriculum. They would be extremely happy to participate in these workshops. My goal for them would be to raise their level of confidence and knowledge so that incorporation of these activities, with support from other teachers, would occur.

DATA COLLECTION/ASSESSMENT

Data collected will consist of a survey of participating teachers assessing the knowledge, experience and comfort using the techniques included in the modules. The survey will be given pre in-service as well as post in-service. Besides surveys, the use of the equipment lockers developed for Celebration High School will be monitored.

EQUIPMENT LOCKERS FROM UNIVERSITY OF FLORIDA

Pipettes and tips	from UF Equipment Locker
E-Gels	from UF Equipment Locker
Simulation Kits	from UF Equipment Locker

ICORE ELEMENTS INCLUDED

The ICORE Summer Institute used the theme of emerging pathogens to engage participants in biotechnology. The modules developed for Celebration High School also use emerging pathogens, specifically cholera, to engage and instruct participating life science teachers.

LITERATURE

Cholera Online: A Modern Pandemic in Texts and Images. Retrieved August 12, 2011 from www.nlm.nih.gov/exhibition/cholera/.

Lyn, T. E. (2011) Cholera outbreaks closely follow temperature rise, rainfall. Retrieved August 10, 2011 from www.nlm.nih.gov/medlineplus/news/fullstory_112671.html.

Science Take Out – A Medical Mystery of Epidemic Proportions. (2009). University of Rochester. www.sciencetakeout.com

BUDGET/EQUIPMENT NEEDED (For 7 teachers)

Invitrogen	6 E-Gel 1.2% with SYBER Safe Starter Kit	\$114 each	\$684
Invitrogen	1 E-Gel 1.2% with SYBER Safe 18 Pak	\$177 each	\$177
Carolina Biological	Fixed Micropipettes 2- 5 µl -214300 5- 10 µl - 214301 5 - 20 µl - 214302 5 – 100 µl -214304 3 - 200 µl - 214308	\$34 each	\$680
Carolina Biological	Pipette Tips 1-200 µl 215051	5 -200 (case of 1000)	\$72.50
Bio Rad	ELISA Immuno Explorer Kit 166-2400EDU	\$155	\$155
Carolina Biological	OUTBREAK 211208	\$89	\$89
Science Take Out	Medical Mystery of Epidemic Proportion STO-116	25 @ \$11.20 each	\$280
Total			\$2137.50

T²: Teachers and bioTechnology

KEY QUESTION: There are four in-service sessions with the goal of increasing the use of biotechnology in the science classrooms at Celebration High School. Many of the teachers are not familiar with biotechnology and do not feel comfortable using it with their students. It is hoped that by exposing teachers to aspects of biotechnology in a learning community of other teachers, the use of biotechnology will increase in the classroom. The question then becomes, “Does science teacher professional development increase the use of biotechnology in the science classroom?”

SCIENCE SUBJECT: The professional development will be aimed towards high school life science classes but open to all science teachers at Celebration High School.

GRADE LEVEL AND ABILITY LEVEL: While the professional development is for teachers only, students of all grade levels will ultimately be impacted as the participating teachers are involved in all levels of high school science.

SCIENCE CONCEPTS: The broad science concepts taught to science teachers will include pipetting, gel electrophoresis, ELISA testing and DNA analysis but the goals of the professional development are as follows:

- 1). Develop teacher understanding of current concepts/skills in biotechnology.
- 2) Develop pedagogy appropriate for life science students.
- 3) Provide ongoing support to participating teachers for implementation and use of biotechnology in their science classrooms.
- 4) Encourage collaboration of ideas and sharing of materials between all science teachers at Celebration High School.

TIME ESTIMATE: It is anticipated that the professional development will consist of 4-5 one hour sessions.

LEARNING STYLES: All learning styles will be incorporated.

VOCABULARY:

Agarose- a polymer made of repeating monomers of agarobiose – a disaccharide.

Antibody- antibodies are produced by the immune system in response to harmful substances called antigens.

Antigen- the harmful substances to which the body produces antibodies.

Biotechnology- technology based on biology in which cellular and biomolecular processes are developed and uses to improve lives and health of the planet. It includes but is not limited to, genetic engineering, bioengineering, cloning, forensics and many others.

Cholera- an acute and usually fatal disease caused by the bacterium, *Vibrio cholera*.

Produces severe gastrointestinal symptoms. The cholera bacterium is found in contaminated food and water.

ELISA- Enzyme linked immunosorbent assay – used to detect the presence of an antibody or antigen in a sample.

Gel-electrophoresis- used to separate molecules such as proteins, DNA or RNA fragments by charge or size. An electric field is applied and negatively charged particles are moved through the agarose mixture. Smaller, shorter molecules move faster and migrate farther while the longer molecules do not. The agarose gel has pores to allow movement of these molecules.

LESSON SUMMARY:

Module 1 – The first module will consist of introducing the teachers to pipetting skills. This is a skill that has not been seen in my school. While we have a few pipettes, they have not been used

extensively. The pipetting locker will be obtained from ICORE at the University of Florida and used to introduce teachers to pipetting. Using money from the ASBMB and ICORE, pipettes will be purchased and an equipment locker for Celebration High School will be started.

Module 2- This module will introduce teachers to antigen/antibody testing and Microarrays through the simulation of A Medical Mystery of Epidemic Proportions from Science Take Out. Using a cholera epidemic as the basis, participants will attempt to discover why people who have been vaccinated against cholera or have had cholera in the past are becoming sick with cholera. Other simulation activities will be developed and incorporated into an equipment locker for future use.

Module 3 – Building on the previous two modules, the third module will use restriction digestion and gel electrophoresis to examine bacterial DNA. This activity will use the bacterial kit from Carolina Biological, Outbreak, to teach these skills. Our school has a few of the gel electrophoresis kits where agarose gels must be prepared ahead of time. Teachers will be taught how to do this and how to run the gels with this equipment but E-Gels from Bio Rad will be purchased and placed into an equipment locker for use by teachers of Celebration High School. Preparation of the agarose gels and staining of those gels is time consuming. Celebration High School has 52 minute periods so using E-Gels, preparation time can be minimized.

Module 4 – En Elisa Antibody Test will also be one to continue with the cholera theme, building on the three previous modules.

STUDENT LEARNING OBJECTIVES WITH STANDARDS:

The expected outcomes are that participating teachers will develop a level of knowledge and skills that will allow them to incorporate biotechnology into their classrooms. Skills learned:

- Pipetting
- Gel Electrophoresis

- Use of simulations such as ELISA Testing and DNA analysis
- How to prepare agarose gels
- Writing curriculum modules to use with the equipment lockers

1. SC.912.L.16.7 – Describe how viruses and bacteria transfer genetic material between cells and the role of this process in biotechnology.

2. SC.912.L.16.10 – Evaluate the impact of biotechnology on the individual, society and environment, including medical and ethical issues.

3. SC.912.L.16.11 – Discuss the technologies associated with forensic medicine and DNA identification, including restriction fragment length polymorphism (RFLP) analysis.

4. SC.912.L.16.12 – Describe how basic DNA technology (restriction digestion by endonucleases, gel electrophoresis, polymerase chain reaction, ligation and transformation) is used to construct recombinant DNA molecules (DNA cloning).

MATERIALS:

Essential: 6 E-Gels, 18 prepared agarose gels, fixed micropipettes (2-5, 5-10, 5-20, 5-100 and 3-200 µl), one case (1000) pipette tips, 1 ELISA Immuno Explorer Kit, 1 OUTBREAK simulation, 25 Medical Mystery of Epidemic Proportion Simulation Kit,

Supplemental: 4 Plastic storage bins for equipment lockers, agarose, Carolina Blue dye, microwave, pre and post surveys,

BACKGROUND INFORMATION:

National goals regarding science education emphasize an inquiry approach and placement of science concepts into a context relevant to students. There is concern that implementation of reform-based ideas are not wide spread in science classrooms of today (Bybee, 1997). There is a disconnect between what educational researchers view as goals and the practices seen in the classroom (Hurd,

1991). As stated by Courson and Zembel-Saul (2002), "Sometimes teachers are not even aware of new reform initiatives and national policy documents (pg. 1). It is the teacher who is the critical in building the bridge between curriculum and implementation (Roberts, 1982).

Concepts of biotechnology are currently not found in the educational programs of secondary science educators (Courson & Zembel-Saul, 2002) but with the rapid rise of biotechnology topics seen in the news as well as new products of biotechnology and biotechnology jobs, students must be knowledgeable with regards to biotechnology (Courson & Zembel-Saul). According to Dawson and Schibeci (2003), many students do not understand current biotechnology concepts or the science behind social issues such as genetically modified organisms and forensic science. This lack of knowledge of biotechnology by high school students was noted by Murat (2009) in his study of high school students' knowledge and attitudes regarding biotechnology. Scientific literacy is a priority for all members of society. Schools have the job of preparing students for their role as citizen.

Dawson (2003) states, "Not all science teachers will have an understanding of modern biotechnology. More importantly, they may not feel comfortable dealing with science knowledge that has a controversial aspect. Clearly professional development for science teachers is important here" (pg. 66). Thus the reason for development of professional development modules dealing with biotechnology for life science teachers at Celebration High School. It is hoped that by involving teachers in inquiry and relevant biotechnology activities, these teachers will take this knowledge and skills back to their classrooms, therefore, impacting all students at Celebration High School.

ADVANCED PREPARATION:

Module 1: Prepare colored water necessary for participants to pipette.

Module 2: The simulation comes with all necessary materials.

Module 3: The simulation comes with all necessary materials

Module 4: Obtain kit from UF ICORE program

PROCEDURE AND DISCUSSION QUESTIONS WITH TIME ESTIMATES.:

All procedures from ICORE summer institute- University of Florida.

Module 1 – Pipetting

Participants will follow directions regarding pipetting to make a “picture” in a well plate using pipettes and the appropriate amount of colored water.

Protocol A – DNA

Micropipette indicated amounts into designated wells on the 96 well plate!

Using the RED dye,

20 μ L: B1, B2, B3, B11,
16 μ L: D1, D3, D10, D11, D12
17 μ L: E1, E3, E10, E12
18 μ L: F1, F2, F10, F12
19 μ L: C1, C3, C10, C12

Using the BLUE dye,

8 μ L: B5, B8
6 μ L: D5, D7, D8
8 μ L: E5, E7, E8
9 μ L: F5, F8
7 μ L: C5, C6, C8

Using the RED dye,

70 μ L: B1, B2, B3, B11,
116 μ L: D1, D3, D10, D11, D12
110 μ L: E1, E3, E10, E12
85 μ L: F1, F2, F10, F12
93 μ L: C1, C3, C10, C12

Using the BLUE dye,

118 μ L: B5, B8
96 μ L: D5, D7, D8
88 μ L: E5, E7, E8
129 μ L: F5, F8
107 μ L: C5, C6, C8

Protocol B- GATC

Micropipette indicated amounts into designated wells on the 96 well plate!

Using the GREEN dye,

20 μ L: E6, E10, E11, E12

16 μ L: G5, G6, G7, G9

17 μ L: F5, F7, F9

18 μ L: H5, H7, H10, H11, H12

Using the BLUE dye,

8 μ L: B1, B8

6 μ L: D1, D4, D8

8 μ L: E2, E3, E4, E8

9 μ L: A2, A3, A4, A6, A7, A8, A9, A10

7 μ L: C1, C3, C4, C8

Using the GREEN dye,

70 μ L: E6, E10, E11, E12

116 μ L: G5, G6, G7, G9

110 μ L: F5, F7, F9

93 μ L: H5, H7, H10, H11, H12

Using the BLUE dye,

118 μ L: B1, B8

96 μ L: D1, D4, D8

88 μ L: E2, E3, E4, E8

129 μ L: A2, A3, A4, A6, A7, A8, A9, A10

107 μ L: C1, C3, C4, C8

Protocol C- DNA Strand

Micropipette indicated amounts into designated wells on the 96 well plate!

Using the BLUE dye,

17 μ L: C1, C10, C11, E8

19 μ L: D2, D9, D12

18 μ L: F4, F7, G5, G6

Using the RED dye,

16 μ L: G1, G10, G11

19 μ L: F2, F9, F12, E3

22 μ L: D4, D7

14 μ L: C5, C6

Using the BLUE dye,

101 μL : C1, C10, C11, E8

74 μL : D2, D9, D12

96 μL : F4, F7, G5, G6

Using the RED dye,

72 μL : G1, G10, G11

108 μL : F2, F9, F12, E3

105 μL : D4, D7

102 μL : C5, C6

Protocol D – Atom

Micropipette indicated amounts into designated wells on the 96 well plate!

Using the RED dye,

20 μL : A2, A3, E4

14 μL : B2, B4, C2, C5, D3

15 μL : G9, F8, H7, H8

Using the ORANGE dye,

20 μL : A8, A9, A10

18 μL : B7, B10, C10, D4, D9

17 μL : E3, E8, F3, F7

19 μL : G3, G6, H4, H5

Using the BLUE dye,

18 μL : C6, D5, D6, D7

19 μL : E5, E6, E7, F6

Using the RED dye,

85 μL : A2, A3, E4

90 μL : B2, B4, C2, C5, D3

70 μL : G9, F8, H7, H8

Using the ORANGE dye,

90 μL : A8, A9, A10

85 μL : B7, B10, C10, D4, D9

75 μL : E3, E8, F3, F7

102 μL : G3, G6, H4, H5

Using the BLUE dye,

93 μL : C6, D5, D6, D7

97 μL : E5, E6, E7, F6

Protocol E – Insect

Micropipette indicated amounts into designated wells on the 96 well plate!

Using the GREEN dye,

9 μ L: A5, A7, C4, C6, C8

12 μ L: B3, B4, B6, B8

8 μ L: D4, D5, D6, D7, D8, D9

14 μ L: E4, E5, E6, E7, E8, E9

16 μ L: F4, F6, F8, H5, H7

13 μ L: G3, G4, G6, G8

Using the BLUE dye,

14 μ L: A11, B11, G11

12 μ L: C10, F10, H10

Using the GREEN dye,

99 μ L: A5, A7, C4, C6, C8

102 μ L: B3, B4, B6, B8

85 μ L: D4, D5, D6, D7, D8, D9

114 μ L: E4, E5, E6, E7, E8, E9

106 μ L: F4, F6, F8, H5, H7

131 μ L: G3, G4, G6, G8

Using the BLUE dye,

104 μ L: A11, B11, G11

112 μ L: C10, F10, H10

Module 2 – A Medical Mystery of Epidemic Proportion

Medical Mystery of Epidemic Proportions

This activity was adapted from the Science Take Out/University of Rochester Medical School activity, “Medical Mystery of Epidemic Proportions”, to better reflect real-world scenarios in Haiti, as well as use more inquiry-based techniques

Part 1:

Materials:

- Strips of 4 pad pH indicator testing as the cholera dipsticks
- Tubes of “positive” for cholera samples (water and diarrhea – can add small amounts of color for more realistic sample) using a pH 10 buffer, and “negative” for cholera using an acidic buffer.

- Print out 1 copy of the “Camps, water sources” document, cutting out a camp name to be assigned to each student group

Assign each student group a camp and water source from the following list*:

*Based on WFP Emergency Preparedness and Response Branch (ODEP)’s map of Port-au-Prince IDP (Internally displaced people) Camps and Main Distribution Sites – *available on accompanying PowerPoint*

**Images of the different water sources (and what they actually look like in Haiti, specifically), would likely help students imagine why different sources are more prone to contamination than others – *available on accompanying PowerPoint*

- Camp 19: Camp Automeca Hyndai; 11,000 ppl; river (washing), Water tank
- Camp 21: Canape Vert (Poste de Polie et terrain basquet); 12,000 ppl; River (washing), water tank
- Camp 38: Cite Soleil; 4,000 ppl; River
- Camp 43: CX Mission (eglise); 2,000 ppl; well
- Camp 57: Delmas 89 A impasse Oseille; 7,500 ppl; Water tank
- Camp 70: Henfrasa; 3750 ppl; water bladder
- Camp 110: Martissant – Cite la Joie; 10,000 ppl; River (washing), water bladder
- Camp 116: Parc de la Fe/Delmas 2; 16,305 ppl; Stream (washing), water tank
- Camp 123: Place de la Paix, Rue St Martin/Del Mar 2; 2,000 ppl; stream
- Camp 125: Presbytere Notre Dame de Lourdes; 3,000 ppl; stream (washing), water bladder
- Camp 140: Site Kawas village Afka Route Patrick; 1,000 ppl; Stream
- Camp 147: Terrain Bayejou/Parc Flamboyant; 2,500 ppl; Stream (washing), water bladder
- Camp 148: Terrain de foot pre du Capitol; 1,000 ppl; stream (washing), water bladder
- Camp 151: Terrain Miron; 899 ppl; Stream
- Camp 153: Terrain Pere Solina; 7,000 ppl; stream (washing), Water bladder

The following camps should test POSITIVE for cholera in one water source:

- Camp 19: Camp Automeca Hyndai; 11,000 ppl; river (washing – positive), Water tank (negative)
- Camp 21: Canape Vert (Poste de Polie et terrain basquet); 12,000 ppl; River (washing - positive), water tank (negative)
- Camp 38: Cite Soleil; 4,000 ppl; River
- Camp 57: Delmas 89 A impasse Oseille; 7,500 ppl; Water tank
- Camp 110: Martissant – Cite la Joie; 10,000 ppl; River (washing - positive), water bladder (negative)
- Camp 116: Parc de la Fe/Delmas 2; 16,305 ppl; Stream (washing - positive), water tank (negative)

- Camp 123: Place de la Paix, Rue St Martin/Del Mar 2; 2,000 ppl; stream
- Camp 125: Presbytere Notre Dame de Lourdes; 3,000 ppl; stream (washing), water bladder (negative)
- Camp 140: Site Kawas village Afka Route Patrick; 1,000 ppl; Stream
- Camp 147: Terrain Bayejou/Parc Flamboyant; 2,500 ppl; Stream (washing - positive), water tank (negative)
- Camp 148: Terrain de foot pre du Capitol; 1,000 ppl; stream (washing – positive), water bladder (negative)
- Camp 151: Terrain Miron; 899 ppl; Stream
- Camp 153: Terrain Pere Solina; 7,000 ppl; Water bladder

The following camps should test NEGATIVE for cholera in the water:

- Camp 43: CX Mission (eglise); 2,000 ppl; Well
- Camp 70: Henfrasa; 3750 ppl; water bladder

The following camps should test POSITIVE for cholera in patients:

- Camp 19: Camp Automeca Hyndai; 11,000 ppl; river (washing), Water tank
- Camp 21: Canape Vert (Poste de Polie et terrain basquet); 12,000 ppl; River (washing), water tank
- Camp 38: Cite Soleil; 4,000 ppl; River
- Camp 57: Delmas 89 A impasse Oseille; 7,500 ppl; Water tank
- Camp 110: Martissant – Cite la Joie; 10,000 ppl; River (washing), water bladder
- Camp 116: Parc de la Fe/Delmas 2; 16,305 ppl; Stream (washing), water tank
- Camp 123: Place de la Paix, Rue St Martin/Del Mar 2; 2,000 ppl; stream
- Camp 125: Presbytere Notre Dame de Lourdes; 3,000 ppl; stream (washing), water bladder
- Camp 140: Site Kawas village Afka Route Patrick; 1,000 ppl; Stream
- Camp 147: Terrain Bayejou/Parc Flamboyant; 2,500 ppl; Stream (washing), water bladder
- Camp 151: Terrain Miron; 899 ppl; Stream
- Camp 153: Terrain Pere Solina; 7,000 ppl; stream (washing), Water bladder

The following camps should test NEGATIVE for cholera in patients:

- Camp 43: CX Mission (eglise); 2,000 ppl; Well
- Camp 70: Henfrasa; 3750 ppl; water bladder
- Camp 148: Terrain de foot pre du Capitol; 1,000 ppl; stream (washing), water bladder

Port-au-Prince map:

Print a large copy of the map (available on accompanying PowerPoint), and put up on a corkboard (for using pushpins), or a wall (for using sticky tabs). Alternatively, you can project this map onto a

whiteboard, and have students mark the locations with whiteboard markers or sticky tabs, although the resolution may be too poor when projected to read the camp names.

Water source table:

Water source	Cholera present	Cholera absent
Wells	0	1
Rivers/streams	11	0
Water bladders	1	5
Water tanks	0	4

Part 2:

Materials:

- Print out antibody-antigen test circles on overhead sheets, 1 per group/camp
- Use 2 colorless chemicals (saturated sodium bicarbonate solution – baking soda, and calcium chloride are two such chemicals) that will create a precipitate when combined (just a few drops), place one (that won't react with water) in tubes labeled "O1 antibody", and the other in a tube labeled "positive control". In tubes labeled with patients A-C, put either water or the precipitating solution, depending on whether it should test positive for the O1 antigen (with precipitate) or negative (water).

Cholera Type:

The following camps should test POSITIVE for type O1 cholera

- Camp 19: Camp Automeca Hyndai; 11,000 ppl; 1-2 PATIENTS
- Camp 21: Canape Vert (Poste de Polie et terrain basquet); 12,000 ppl; 0-1 PATIENTS
- Camp 38: Cite Soleil; 4,000 ppl; 1-2 PATIENTS
- Camp 57: Delmas 89 A impasse Oseille; 7,500 ppl; ALL PATIENTS
- Camp 110: Martissant – Cite la Joie; 10,000 ppl; 1-2 PATIENTS
- Camp 116: Parc de la Fe/Delmas 2; 16,305 ppl; 0-1 PATIENTS
- Camp 123: Place de la Paix, Rue St Martin/Del Mar 2; 2,000 ppl; ALL PATIENTS
- Camp 125: Presbytere Notre Dame de Lourdes; 3,000 ppl; ALL PATIENTS
- Camp 140: Site Kawas village Afka Route Patrick; 1,000 ppl; ALL PATIENTS
- Camp 147: Terrain Bayejou/Parc Flamboyant; 2,500 ppl; ALL PATIENTS
- Camp 151: Terrain Miron; 899 ppl; ALL PATIENTS

- Camp 153: Terrain Pere Solina; 7,000 ppl; 1-2 PATIENTS

The following camps should also test NEGATIVE for type O1 cholera

- Camp 19: Camp Automeca Hyndai; 11,000 ppl; 1 PATIENT
- Camp 21: Canape Vert (Poste de Polie et terrain basquet); 12,000 ppl; 2 PATIENTS
- Camp 38: Cite Soleil; 4,000 ppl; 1 PATIENT
- Camp 110: Martissant – Cite la Joie; 10,000 ppl; 1 PATIENT
- Camp 116: Parc de la Fe/Delmas 2; 16,305 ppl; 2 PATIENTS
- Camp 153: Terrain Pere Solina; 7,000 ppl; 1 PATIENT

Part 3:

Materials:

- Create a simulated DNA microarray by using either 96 well plates (or similar), or using the template printed on cardstock. In the circles/wells for the following genes, place 1-2 drops of a 2% phenolphthalein solution:
 - Cholera Toxin
 - Catalase enzyme
 - DNA polymerase
 - Flagella
 - Pilus
- The O1 antigen gene circle/well should receive either water, or nothing at all
- In a tube labeled “new cholera DNA”, put a pH 10 buffer, which should turn pink when combined with the phenolphthalein solution (each circle but the O1 antigen circle should turn pink)

For more information on DNA microarray technology, go to: <http://www.genome.gov/10000533>, or the virtual lab <http://learn.genetics.utah.edu/content/labs/microarray/>

Evolution Hypotheses:

In general, there are 2 types of hypotheses for this part of the lab, although some students might think of alternatives, as well. One hypothesis is that the new strain of cholera was a harmless *Vibrio cholerae* that picked up/evolved the cholera toxin independently. The second hypothesis is that a pathogenic *Vibrio cholerae* bacterium evolved from a type O1, but mutated in a way that it lost the gene to code for the O1 antigen. The second hypothesis is the one supported by the results of the microarray.

Extensions:

After completion of this activity, many students will likely wonder how cholera can be prevented in the first place. The difficulty is that countries vulnerable to cholera epidemics have poor water and sanitation, and public health systems are lacking. To show students the extent of this problem, you can have them complete individual or group research projects on the nature of cholera epidemics. Possible topics include:

- Map out cholera outbreaks in the past 50-100 years, globally. Indicate the per capita income in each country, as well as the status of their sanitation and public health systems.
- Artistic representations of the sanitation and public health systems in countries that have recently experienced cholera outbreaks. This includes drawings, paintings, audio, and video (ex. public service announcements).
- Research CDC and/or WHO data on cholera outbreaks, particularly the recent outbreak in Haiti. Projects could include a public service announcement on preventative measures to help prevent the spread of cholera.
- *Any of these projects should ask students to consider how the preventative measures might be difficult for citizens to implement due to cost, i.e. why adding bleach in water or boiling water might be too expensive for many Haitians*
- Antibody Test: print 1 per group/camp on overhead sheets for Part 2

O1 Antibody



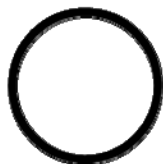
Positive control

O1 Antibody



Vibrio cholerae
sample from
patient A

O1 Antibody



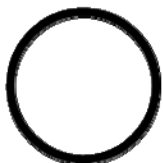
Vibrio cholerae
sample from
patient B

O1 Antibody



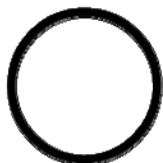
Vibrio cholerae
sample from
patient C

O1 Antibody



Positive control

O1 Antibody



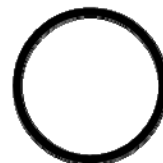
Vibrio cholerae
sample from
patient A

O1 Antibody



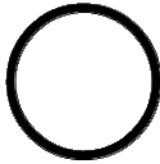
Vibrio cholerae
sample from
patient B

O1 Antibody



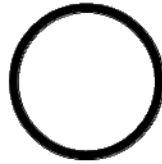
Vibrio cholerae
sample from
patient C

O1 Antibody



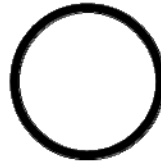
Positive control

O1 Antibody



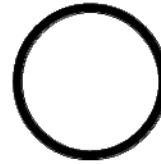
Vibrio cholerae
sample from
patient A

O1 Antibody



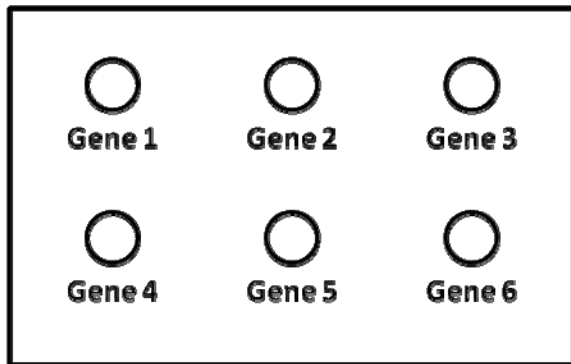
Vibrio cholerae
sample from
patient B

O1 Antibody



Vibrio cholerae
sample from
patient C

DNA Microarray Test



O1 *Vibrio* genes

spotted on microarray

Gene 1: Cholera toxin gene

Gene 2: Catalase enzyme gene

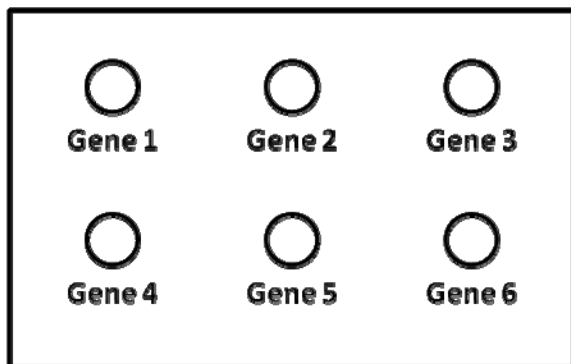
Gene 3: DNA polymerase gene

Gene 4: Flagella gene

Gene 5: O1 Antigen gene

Gene 6: Pilus gene (attachment
to intestine)

DNA Microarray Test



O1 *Vibrio* genes

spotted on microarray

Gene 1: Cholera toxin gene

Gene 2: Catalase enzyme gene

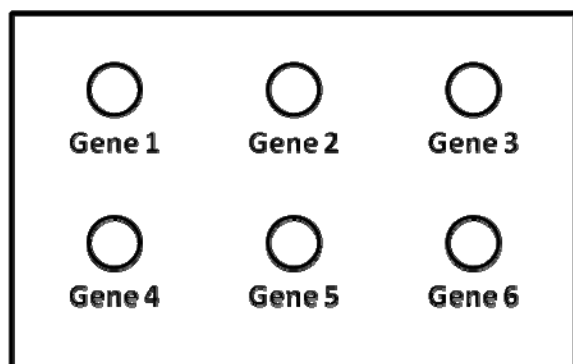
Gene 3: DNA polymerase gene

Gene 4: Flagella gene

Gene 5: O1 Antigen gene

Gene 6: Pilus gene (attachment
to intestine)

DNA Microarray Test



O1 Vibrio genes

spotted on microarray

Gene 1: Cholera toxin gene

Gene 2: Catalase enzyme gene

Gene 3: DNA polymerase gene

Gene 4: Flagella gene

Gene 5: O1 Antigen gene

Gene 6: Pilus gene (attachment to intestine)

Module 3 – Outbreak

After leaving your volunteer commitment in Haiti, you return home to Florida. After your first week of being back, your community is hit with a mysterious illness, which has symptoms similar to the cholera that you recently witnessed in Haiti. Although you're quick to assume that everyone has cholera, you realize that there are other bacterial diseases with the same general symptoms – including the enterohemorrhagic strains of *Escherichia coli*. You culture the bacteria from a patient, and discover that the disease is indeed *E. coli*. Since a tour group from your community just returned home from Europe, you worry that they may have contracted the new strain of *E. coli*, O104:H4, which has a much higher mortality.

You decide to use restriction digestion and gel electrophoresis to examine the bacterial DNA to determine which *E. coli* strain is causing the epidemic in your community. You have added enzymes to the bacterial DNA extracts from *E. coli* O157:H7 (the most common culprit of *E. coli* outbreaks in the United States), and *E. coli* O104:H4 (the new strain from Germany). The enzymes cut the DNA at specific locations in the bacterial genome, which are then electrophoresed to produce a bacterial fingerprint.

1. Why would the restriction fragments patterns from the different bacteria be different?

Procedure 1: Prepare gel

1. Plug PowerBase™ into an electrical outlet.
2. Remove gel cassette from package

3. Insert the gel (with comb in place) into the base right edge first. The Invitrogen logo should be located at the bottom of the base. Press firmly at the top and bottom to seat the gel cassette in the PowerBase™. A steady, red light will illuminate if the gel cassette is correctly inserted.
4. Pre-run the gel (with comb in place) by pressing and holding either the 15 minute or 30 minute button until the red light turns to a flashing green light indicating the start of the 2 minute pre-run. Release the button. At the end of the pre-run, the current will automatically shut off and the power base will display a flashing red light and beep rapidly. Press either button to stop the beeping.

Procedure 2: Load prepared samples

(If two groups are sharing a gel, Group A will load DNA samples in wells 1-4; Group B will load DNA

Well #	1	2	3	4	5	6	7	8	9	10	11	12
What to add to the well	10µl water + 8µl patient	10µl water + 8µl <i>E. coli</i> O157:H7	10µl water + 8µl <i>E. coli</i> O104:H4	20µl water	20µl water	20µl water	10µl water + 8µl patient	10µl water + 8µl <i>E. coli</i> O157:H7	10µl water + 8µl <i>E. coli</i> O104:H4	20µl water	20µl water	20µl water

samples in wells 7-10.)

1. Remove and discard comb from the E-Gel® cassette.

GROUP A

2. Add 10µl sterile distilled H₂O to wells 1-3.
3. Add 20µl sterile distilled H₂O to wells 4-6.
4. Add 8µl DNA samples to wells 1-3.

GROUP B

5. Add 10µl sterile distilled H₂O to wells 7-9.
6. Add 20µl sterile distilled H₂O to wells 10-12.
7. Add 8µl DNA samples to wells 7-9.

Procedure 3: Run gel

1. Press and release the 30 minute button on the E-Gel® PowerBase™ to begin electrophoresis.

2. At the end of the run, the current will automatically shut off and the power base will display a flashing red light and beep rapidly. Press either button to stop the beeping.
3. Remove the gel cassette and analyze your results by viewing on one of the transilluminators.

1. Compare the fingerprints from *E. coli* O157:H7 and *E. coli* O104:H4. What similarities and differences do you notice?

2. Which *E. coli* strain is causing the epidemic in your community?

Module 4 – ELISA

From BIO Rad – ELISA – see attached PDF

ASSESSMENT SUGGESTIONS: As this is professional development for teachers, assessment in the true sense of the word will not be done. Data will be collected through the use of a pre and post survey which examines past experiences, knowledge and comfort level of each participant. The survey is located on Google Docs and can be found at:

https://docs.google.com/spreadsheet/viewform?hl=en_US&formkey=dE43NllcmdjUF9nMzVKTDIWIY28xcIE6MQ#gid=0

EXTENSIONS: It is hoped that through collaboration with teachers in the professional development, other lesson plans using biotechnology will be written and other equipment lockers developed for use by current and future Celebration High School science teachers. It would be ideal to eventually institute this program at the district level for all high schools in Osceola County.

RESOURCES

Kits, Equipment and Procedures for Modules are from ICORE Summer Institute – University of Florida.

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