Title: Dengue: Using Breakbone Fever to Introduce Proteomics

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Abstract:

High school students often have little knowledge about the workings of biotechnology and are intimidated to try and understand molecular biology. Dengue fever, while sounding exotic, is an emerging pathogen that could affect Florida students today. The study of this disease will be used as a hook to get students to utilize biotechnology tools and protocols to understand proteomics. Mosquito egg clusters will be reared in the lab, with specimens being removed at the larval, pupal and adult stages. After DNA is extracted from each stage, it will be run through PCR and sent to an outside source for sequencing to confirm identical DNA at all stages. Protein will be extracted from each stage as well, and gels will be compared to find differences in protein expression. Samples will be run through the MALDI mass spectrometer at the University of Florida with the results being sent back to students at a later date.

Rationale:

Understanding biology at the molecular level is critical in any of the life science fields students may pursue at the secondary level. Biology teachers at my school do not conduct lab activities associated with biotechnology, leaving students with a high level of ignorance and intimidation concerning molecular biology. This is reflected by the number of students taking Advanced Placement Biology and Genetics Honors. With a student population of about 3,000 students, Palm Beach Central currently has ten sections of Advanced Placement Environmental Science which shows an interest in higher level science. However, there are only two sections of Advanced Placement Biology and one section of Genetics Honors which indicates a significant drop in students advancing to the more difficult science courses. This unit will expose students to many of the concepts of these courses, and an increase in enrollment of these courses is expected. In addition, many Research Honors students need to incorporate biotechnology into their individual projects but lack the knowledge of how to do this. The practical experience of this unit will assist them in following protocols for several common biotechnology tasks.

Description of Teaching Unit:

The following unit will take approximately five weeks, which will be followed by extension activities for the following semester. Since students are in grades 9-12, with some not having taken biology yet and others having completed AP Biology, the Viral Quest curriculum will be utilized to bring all students up to a baseline level of molecular biology.
Day One: Pre Test on emerging pathogens and biotechnology
Day Two: Viral Quest
Day Three: Viral Quest
Day Four: Viral Quest
Day Five: Viral Quest
Day Six: Viral Quest
Day Seven: Viral Quest
Day Eight: Viral Quest
Day Nine: Viral Quest
Day Ten: Viral Quest
Day Eleven: Viral Quest
Day Twelve: Lecture on insect taxonomy and anatomy
Assessment: Mosquito life cycle diagram
Day Thirteen: Grasshopper dissection
Assessment: Lab Packet
Day Fourteen: Mosquito IPM DVD
Assessment: Video worksheet
Day Fifteen: Dengue Fever WebQuest
Assessment: WebQuest packet
Day Sixteen: HHMI PCR Virtual Lab
Assessment: Lab worksheet
Day Seventeen: Micropipette activity
Day Eighteen: DNA extraction and PCR
Assessment: Lab report
Day Nineteen: Run PCR product on gels, send out for sequencing
Assessment: Lab report
Day Twenty: Protein extraction
Assessment: Lab report
Day Twenty One: Protein gels and digestion
Assessment: Lab report
Day Twenty Two: Lecture on mass spectrometry
Day Twenty Three: Post Test on emerging pathogens and biotechnology
Samples sent to Dr. Chen’s lab for mass spec.

Extensions: Classroom visit by person who had Dengue
- the human side of disease
Classroom visit by emergency room physician (former student)
- how doctors detect uncommon pathogens
- academic pathways to a medical career
Classroom visit by Scripps research scientist
- how researchers up/down regulate protein expression
Classroom visit by UF entomologist
Student insect collection
Group presentation on an emerging pathogen
Confirmation of mass spec by using BLAST
Data Collection:

In addition to completing a pre test and post test, nine assignments will be collected and graded along with the Viral Quest materials. The number of students who incorporated biotechnology in their individual projects will be recorded this year and compared to the number incorporating them last year. Enrollment numbers for AP Biology and Genetics Honors will be monitored to determine any increase.

Equipment Usage:

The equipment locker will be needed during the week of DNA and protein extraction to use the micropipettes and gel chambers. Samples will be run on the MALDI in Dr. Chen’s lab at UF.

ICORE Connections:

Knowledge of Dengue was obtained from a lecture by Dr. Connelly during ICORE. The labs for DNA extraction, PCR, protein extraction and mass spec were conducted during ICORE.

Budget:

<table>
<thead>
<tr>
<th>Item</th>
<th>Cost</th>
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<tr>
<td>Mosquito Breeding Chamber</td>
<td>$20</td>
</tr>
<tr>
<td><em>Culex</em> egg cluster</td>
<td>$20</td>
</tr>
<tr>
<td>Primer</td>
<td>$10</td>
</tr>
<tr>
<td>DNA sequencing</td>
<td>($8x9) $72</td>
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</tbody>
</table>
Title: Dengue: Using Breakbone Fever to Introduce Proteomics

Key Questions: What is Dengue Fever and why should I care about it?

Science Subject: Research Honors, Advanced Placement Environmental Science

Grade and Ability Level: Honors (9-12), Advanced Placement (10-12)

Science Concepts: How are vector borne diseases transmitted?

Overall Time Estimate: Unit will take 25 days, lesson will take 50 minutes.

Learning Styles: Students will be viewing video clips and reading text from various web sites.

Vocabulary: symptom, vector, endemic, pandemic, epidemic, serotype, antigen, antibody, vaccine

Lesson Summary: Students will review several web sites to understand what Dengue Fever is, where it is found, how it is transmitted along with current research.

Student Learning Objectives with Standards:
1. Student will describe the symptoms of Dengue Fever.
2. Student will describe the transmission of Dengue Fever.
3. Student will construct a graph of Dengue Fever cases and calculate percent increase over time.
4. Student will describe current research relating to the development of a Dengue Fever vaccine.

Florida Standard: SC.912.2.14.6 Explain the significance of genetic factors, environmental factors, and pathogen agents to health from the perspective of both individual and public health.

Materials: Dengue Fever packet, computer with internet access

Background Information: Dengue Fever is a viral disease most commonly found in tropical climates. However, it has been increasing in both number of cases worldwide and in the number of geographic areas affected. Although Dengue Fever is not endemic to Florida, it has been found occasionally in travelers returning from an overseas trip and has been transmitted locally. The virus that causes Dengue Fever is transmitted by mosquitoes in the Aedes genus which limits the virus to areas where the mosquito lives. A mosquito bites an infected person and picks up the virus, and 8-10 days later it can transmit the virus through its salivary secretions when it bites another human. Typical symptoms of Dengue Fever include headache, fever, rash, and severe joint pain which last about a week. There are four different strains of viruses that cause Dengue Fever, so the body does not build up an immunity to the other strains after being infected with one variety. Some patients develop a more serious form of Dengue called Dengue Hemorrhagic Fever which causes a loss of blood and can be fatal. To prevent Dengue Fever, communities need to monitor mosquito populations and remove breeding areas that Aedes
prefers, like small containers that collect water. Scientists are currently working on creating a vaccine for Dengue Fever, but have had difficulty because there are four serotypes.

- **Symptom** – sign of abnormality of function in a patient
- **Vector** – organism that transmits a pathogen
- **Endemic** – disease is normally found in a population
- **Pandemic** – when a disease spreads across continents
- **Epidemic** – new cases of a disease increase rapidly
- **Serotype** – variety of virus based on its antigens
- **Antigen** – molecule that causes the body to produce antibodies
- **Antibody** – protein produced by the body to identify foreign particles
- **Vaccine** – weakened/dead pathogen introduced so the body produces antibodies

**Advance Preparation:** It is best to run through the Web Quest to make sure all links are still operating.

**Procedure and Discussion Questions with Time Estimates:** To introduce the lesson, I will ask if anyone has heard of Dengue Fever before, and if so, what they think they know about the disease. It is expected that no student will have background knowledge about Dengue, and this should only take 2 minutes. Students will then move quickly to complete the Dengue web quest, which will take 40 minutes. If it takes longer for the students to complete the web quest, the follow-up discussion will take place the following class period. Discussion questions will include:

1. **What are the symptoms of Dengue Fever?**
   *Fever, headache, joint pain.*

2. **Where is Dengue usually found? (endemic)**
   *Tropical Asia, Central America, South America, Puerto Rico.*

3. **How could someone in Florida get Dengue?**
   *By traveling to an area that has Dengue. There is a lag time of several days from being bitten to showing symptoms.*

4. **What limits the spread of Dengue?**
   *Only Aedes mosquitoes transmit Dengue. There needs to be mosquitoes and the virus present to start an epidemic.*

5. **So what actually causes Dengue?**
   *Dengue virus.*

6. **Why are the female mosquitoes important?**
   *They are the vectors, transmitting the virus from one infected person to another.*

7. **What government agencies track diseases like Dengue?**
   *CDC in the U.S. and WHO worldwide*

8. **What do we see worldwide as far as the number of Dengue cases?**
   *Large increase (200%) over the last two decades.*
What is the most serious form of Dengue?
*Dengue Hemorrhagic Fever, which can be fatal and more often acquired after having Dengue multiple times.*

How many different types of Dengue virus are there?
*There are four serotypes.*

Why is creating a vaccine so difficult for Dengue?
*The difficulty has been getting one vaccine that is good for all four serotypes. Trials are currently underway in Thailand.*

**Assessment Suggestions:** In addition to the web quest packet, questions will be on the pre/post test.

1. The agent that causes Dengue Fever is a
   a. Virus
   b. Bacterium
   c. Protozoan
   d. Mosquito
   e. None of the above

2. Dengue gets introduced into the U.S. by
   a. Infected food products
   b. Migratory birds
   c. International travelers
   d. Tainted medical supplies
   e. All of the above

3. The vector for Dengue is
   a. Ticks
   b. Mosquitoes
   c. Lice
   d. Bed bugs
   e. All of the above

4. How many varieties of DEN are there?
   a. One
   b. Two
   c. Three
   d. Four
   e. Five

5. In the U.S. diseases like Dengue are tracked by the
   a. USDA
   b. USFWS
   c. USPS
   d. CDC
   e. EPA

6. The most severe form of Dengue is
   a. Dengue Hemorrhagic Fever
   b. Dengue Encephalitis
c. Dengue Dysentery
d. Dengue Sleeping Syndrome
e. There is only one form of Dengue.

7. Why has the development of a Dengue vaccine taken so long?
   a. There are so few cases of Dengue worldwide, it isn’t cost effective to produce.
   b. The vaccine must work for all Dengue varieties.
   c. Birds are resistant to most antibiotics.
   d. Dengue cases have been declining steadily worldwide.
   e. A different vaccine must be produced for each human ethnic group.

8. In which stage of the vector life cycle is Dengue transmitted?
   a. Egg
   b. Larva
   c. Pupa
   d. Adult
   e. All stages

9. Symptoms of Dengue include
   a. Fever
   b. Rash
   c. Headache
   d. Joint pain
   e. All of the above

**Extensions:** This is part of a unit, which will include using ViralQuest, HHMI virtual PCR lab, UF IPM for Mosquito Control DVD and print resources, rearing of mosquitoes, DNA extraction, and protein extraction.

**Articles:**

Linda Marsa, *Discover*, The Hot Zone, December 2010

Peter Arensburger et al, *Science* 330, Sequencing of *Culex quinquefasciatus* Establishes a Platform for Mosquito Comparative Genomics, October 2010

**Resources:** Several web sites are utilized in this activity.

BBC News  [http://news.bbc.co.uk/2/hi/health/8237529](http://news.bbc.co.uk/2/hi/health/8237529)


U.S. Centers for Disease Control  [www.cdc.gov/dengue](http://www.cdc.gov/dengue)

Wikipedia  [www.wikipedia.org](http://www.wikipedia.org)
Thai News  www.dailymotion.com/video/xhqy06_dengue-fever-vaccine-developed-in-thailand_news
Dengue: Using Breakbone Fever to Introduce Proteomics

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Abstract:

High school students often have little knowledge about the workings of biotechnology and are intimidated to try and understand molecular biology. Dengue fever, while sounding exotic, is an emerging pathogen that could affect American students today. The study of this disease will be used as a hook to get students to utilize biotechnology tools and protocols to understand proteomics. Mosquito egg clusters are to be reared in the lab, with specimens being removed at the larval, pupal and adult stages. After DNA is extracted from each stage, it will be run through PCR and then gel electrophoresis to examine banding. Protein will be extracted from each stage as well, and banding can be compared to find differences in protein expression. Students will find that although the mosquitoes are the same species with the same DNA, different proteins will be expressed at different stages of the life cycle.

Keywords:

proteomics     gel electrophoresis     PCR    pathogen    epidemiology    vector

Dengue fever has been a scourge of tropical regions for decades, spreading quickly after World War II. The nickname “breakbone fever” originates from the intense joint pain associated with the disease. However, multiple infections can result in Dengue hemorrhagic fever which can be fatal. There are four varieties of Dengue virus which cause Dengue fever, each with unique antigens that prevent immunity from a single infection. The vector of Dengue is the *Aedes aegypti* mosquito, which is found throughout the tropics including the southern United States. Americans who contract Dengue typically acquire the disease while travelling, although several high profile cases of locally transmitted Dengue have occurred in Florida and Texas over the last few years. If climate change increases global temperatures, it is predicted that *Aedes aegypti* can expand its range and spread Dengue beyond the tropics. Current research to develop a vaccine is hindered by the four distinct serotypes of the Dengue virus.

Students are taught in biology class that DNA will code for proteins, but this is an abstract concept for most students. A common misconception is that if the DNA code is present, the protein will be produced. Scientists are learning a great deal about the complexity of when a cell produces a protein or silences a gene. A mosquito has a complete life cycle consisting of the egg, larva, pupa and adult stages. While the DNA is the same for all stages, the stages look very different as different proteins are being expressed.
Objectives

At the end of this unit students will be able to:
1. describe the mosquito life cycle
2. describe how the Dengue virus is transmitted
3. describe how Dengue affects the body
4. explain the steps to extract DNA
5. explain the steps of PCR
6. explain the steps of protein extraction
7. predict how environmental change can affect the range of Dengue

Materials

- mosquito breeding chamber (available from BioQuip)
- *Culex* egg clusters (available from Carolina Biological)
- PCR Informatics Kit #166-2100EDU from Bio-Rad
- micropipets (200 microliters, 2-20 microliters)
- centrifuge
- water bath
- thermal cycler
- gel electrophoresis chamber with gels
- Mini-PROTEAN Tetra gel box from Bio-Rad
- Mini-PROTEAN gels (not included in kit)
- Comparative Proteomics Kit I #166-2700EDU from Bio-Rad

Procedure

Order mosquito egg clusters two weeks prior to the unit. Eggs typically are starting to hatch upon arrival, and take 8-9 days to achieve the adult stage. Remove and store in a freezer several larvae, pupae and adults for later useage.

To introduce Dengue, use the attached Dengue Web Quest. This could be completed in a 50 minute class period, but might require time outside the classroom to finish.

Students can view Integrated Pest Management for Mosquito Control: The Basics DVD from the University of Florida Institute of Food and Agricultural Sciences.

How is Dengue affecting the U.S.? Have students complete the Dengue articles worksheet by accessing the following articles:

Palm Beach Post

1. Dengue virus found in 5 percent of Key West residents (7/15/2010)
   http://www.palmbeachpost.com/health/dengue-virus-found-in-5-percent-of-key-800696.html?showComments=true
2. First case of dengue fever reported in Palm Beach County (10/13/2011)
http://www.palmbeachpost.com/news/first-case-of-dengue-fever-reported-in-palm-
1911593.html

3. Second case of dengue fever reported in Palm Beach County (10/18/2011)
http://www.palmbeachpost.com/news/second-case-of-dengue-fever-reported-in-palm-
1919238.html

4. Keys mosquito-suppression plan raises questions
http://www.winknews.com/Local-Florida/2011-12-26/Keys-mosquito-suppression-plan-
raises-questions

Science

1. GM Mosquito Trial Alarms Opponents, Strains Ties in Gates-Funded Project
(11/19/2010) Volume 330 pp. 1030-1031

Follow protocol for DNA extraction and PCR from PCR Informatics Kit #166-2100EDU
from Bio-Rad. One 50 minute class period should be devoted to the DNA extraction and
setting up the PCR. The PCR can run overnight and set on cold soak. Another 50 minute
period is needed to run the gel electrophoresis followed by staining.

Follow the protocol for protein extraction from Comparative Proteomics Kit I #166-
2700EDU from Bio-Rad. It will take one 50 minute period to extract the protein
followed by running the gel and staining.

Discussion

Materials can be found in the Bio-Rad kits to explain procedures and provide assessments
in the form of worksheets which can accompany a formal lab report. Students should see
similar DNA banding in all life cycle stages. Protein gels should show different banding
patterns at the different life cycle stages. Depending on the level of student, an
introduction to epigenetics might be appropriate.

Extensions

The movies Contagion and Ebola: The Plague Fighters give a view of how CDC
epidemiologists work. A writing assignment might compare the actual work of the
epidemiologist with that portrayed in the Hollywood movie. Students can also break into
small groups, and come up with a list of supplies needed to last an epidemic lasting 3-6
months and present their findings to the class.

Since insects are vectors for many pathogens, have students put together an insect
collection to show the biodiversity of your local area.
Students can pick an emerging pathogen and present a report on it. One way of doing this is through a Glog. Glog are like an online science display created by the students that allow for video clips, graphics and text. The editing is in non-technical jargon so all students can learn to produce the Glog with relative ease. Check out www.edu.glogster.com for more information.

The Howard Hughes Medical Institute has resources available to teachers for free. The Bacterial Identification Virtual Lab CD-ROM allows students to practice PCR and other procedures on the computer through simulations. The 2010 Holiday Lecture Series DVD features researchers who are currently working on Dengue (only use with high level students). http://www.hhmi.org/biointeractive/disease/lectures.html

Conclusion

Proteomics is higher level biology, and this unit will give students a real life application to introduce the concepts of selective protein expression. Practical experience using equipment and procedures will give students confidence to tackle biotechnology and embrace higher level biology courses.
For my unit on Dengue, 21 Research Honors students were chosen to participate. The mean score of 42% was attained on the Dengue pre-test, with a mean score of 96% on the post-test. The Viral Quest curriculum was used as an introduction on how viruses function. The student produced Glogs were excellent and students found the format easy to work with. Students now have a better understanding of viral diseases, DNA, PCR and protein extraction after completing the labs. Several students utilized these procedures as part of their individual science fair projects. I am still planning to have Dr. Darwiche visit my classroom next month to complete the protein extraction portion. During department meetings, I explained the ICORE program and what my unit of study is about. One teacher from my school will be participating in ICORE this summer. I feel that I gained a great deal of knowledge about emerging pathogens from participating in ICORE and developing this unit.
Contagion

1. What organization tracks diseases globally?

2. What U.S. government agency tracks diseases here in the U.S. (and monitors world diseases as well)?

3. What is a quarantine?

4. How many times does a person touch their face each day?

5. What is an RO (R nought)?

6. Describe the safety precautions taken in a BSL 4 lab.

7. The virus in the movie comes from what animals?

8. What does it mean if a virus is novel?

9. What is an index patient?

10. What was the worst global flu pandemic?
11. What is a homeopathic treatment?

12. How did the viral mutation affect its RO?

13. How did AIMM contribute to the initial spread of the virus?

14. What factors make a country like China the primary ‘breeding ground’ for viruses?

15. How does the outcome of the H1N1 threat affect people’s view of warnings from the CDC?
Contagion Part II

Bioterrorists in Switzerland have read the *Science* article about making bird flu airborne and figured out how to do it themselves. They have released the flu into Orlando, and it is spreading. Approximately 30% of those infected die and it has an RO of 6. People are panicking and the National Guard has placed a quarantine around Orlando. Travel in and out of Florida is prohibited. There is already a run on food supplies at the grocery stores and little is left on the shelves. If the flu spreads, it is expected that basic services like water and electric will not be in service.

With your group, determine how you will survive this scenario that is expected to last 6 months. Determine where you will go. Create a list of essential items you will need to take with you. The only items you can take must be carried by the people in the group. Describe how you will get water, food, and shelter.

Create a powerpoint presentation that is consists of the following slides: location, shelter, water, food, defenses, item list with justification for each item.
Dengue Fever
I. What is Dengue Fever?
A. Disease common in tropical areas worldwide
   - endemic in Puerto Rico
B. Sometimes called Breakbone Fever due to the pain it causes
C. 100,000,000 people contract it annually, with 25,000 deaths
D. There is currently no vaccine
II. Symptoms of Dengue

A. Many people show no symptoms

B. Symptoms start 3-5 days after being infected

C. Headache, fever, joint pain and rash are common
D. Some develop Dengue Hemorrhagic Fever

1. marrow is infected, reduces platelets
2. bleeding common, can be fatal
3. more common after multiple infections
III. Cause of Dengue

A. Caused by DENV virus

B. There are four different types (serotypes)
   - DENV-1, DENV-2
   - DENV-3, DENV-4

C. Body makes antibodies after infection, but are only specific to one type of DENV

D. 11,000 base pairs in DENV
IV. How Dengue spreads

A. Vector is the mosquito *Aedes aegypti*
   1. common in tropics, including Florida
   2. day fliers
   3. common inside homes
B. Female mosquito bites an infected person
   1. virus reproduces in the gut
   2. spread to the salivary glands after 8-10 days
C. Female mosquito bites another person, viruses enter in the mosquito saliva
D. Viruses infect human white blood cells
V. Preventing Dengue

A. Use insect repellant with DEET

B. Remove standing water from yards to stop mosquito life cycle

C. GMO mosquitoes

D. Current work on a vaccine in Thailand
Prevention in the Philippines
Prevention in Singapore

IF THEY BREED, YOU WILL BLEED.
STOP DENGUE. ACT NOW!
Dengue in Florida

1. What specific type of mosquito spreads Dengue?

2. Where in Palm Beach County was the case of Dengue?

3. What are the symptoms of Dengue?

4. What does locally transmitted mean?

5. How can you protect your home from Dengue?

6. How did epidemiologists determine that 5% of Key West households have Dengue?

7. Why don’t doctors recognize Dengue more often?

8. How do you treat Dengue?
9. What is the plan to stop Dengue in the Keys?

10. How much will the program cost each year?

11. What is a GMO?

12. Who helped fund studies on tropical disease like Dengue and malaria?

13. How does Oxitec plan to use mosquitoes to stop Dengue?

14. What did Oxitec claim happened in their experiment?

15. How was the experiment in the Cayman Islands different from the experiment in Mexico in terms of public input?

16. Should Oxitec be allowed to execute its plans in the Keys?
Dengue Fever is a condition you are probably not familiar with. To find out more, watch the video clip at http://news.bbc.co.uk/2/hi/health/8237529.stm.

1. Why are cities like Manila a haven for Dengue? ______________________________________

2. How many countries now have to deal with Dengue? ____________

You don’t live in the Philippines, so you don’t need to worry about Dengue, right? Watch the video clip at www.abcnews.go.com/Health/video/dengue-fever-outbreak-risk-mosquito-10730699.

3. What are the symptoms of Dengue? ______________________________________

4. What is the nickname given to Dengue? ______________________________________

5. Is Dengue common in the U.S.? ______ Where in Florida has Dengue been a problem? ___________________________________________________________________

A vector is an organism (usually a biting insect) that doesn’t cause the disease, but carries the agent that does cause the disease and transmits it to a host. Go to Wikipedia and search Dengue Fever to answer the following questions.

6. Can you get Dengue from any kind of mosquito? _____ What is the scientific name of the mosquito that transmits Dengue? ______________________________________
7. Describe what this mosquito looks like.

8. After the mosquito acquires the Dengue virus by biting an infected person, how many days does it take before the mosquito can transmit the virus to another person? ________

In the U.S., the Centers for Disease Control (CDC) tracks many diseases that affect human health. Their web site contains a great deal of information about these diseases. Go to www.cdc.gov/dengue and find the link titled Entomology to answer the following questions.

9. What are the four stages of the mosquito life cycle? ____________________________

10. How many days does it take to reach the adult stage? __________

Go back to the Dengue home page at the CDC. Click on the link for the Dengue map. Color in the areas on the map where Dengue is regularly found.

11. Based on the map, what environmental factor(s) are necessary for Dengue? ________

12. Define the following terms:
   - Endemic ____________________________
   - Pandemic ____________________________
   - Epidemic ____________________________

13. In what part of the U.S. is Dengue endemic? ____________________________

The World Health Organization (WHO) monitors health conditions, include disease, on a worldwide scale. Use the data from WHO to see how Dengue infection rates have changed over the decades.
Average Annual Dengue Rates

<table>
<thead>
<tr>
<th>Decade</th>
<th>Average Annual DF Cases</th>
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<tbody>
<tr>
<td>1950s</td>
<td>908</td>
</tr>
<tr>
<td>1960s</td>
<td>15,497</td>
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<td>1970s</td>
<td>122,174</td>
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<td>1980s</td>
<td>295,554</td>
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<td>1990s</td>
<td>479,848</td>
</tr>
<tr>
<td>2000s</td>
<td>968,561</td>
</tr>
</tbody>
</table>

14. Calculate the percent increase in Dengue cases from 1980 to 2000. _____________

Go back to Wikipedia to find out more about the virus that causes Dengue.

15. What is the family of the virus that causes Dengue? ___________________________

16. How many different types (serotypes) of Dengue virus are there? ________

17. If you get Dengue, your body will make antibodies to recognize the virus if it enters again. After having Dengue, are you protected from the other types of Dengue? ________
Go back to the CDC page and look at the FAQ link.

18. What is the most serious version of Dengue that affects humans? ________________
________________________________________________________________________

19. How does someone get this type of Dengue? ________________________________
________________________________________________________________________

20. Why should you not take aspirin or ibuprofen if you think you have Dengue? ______
________________________________________________________________________

Since Dengue is such a serious problem, what are scientists doing to try and contain it?
Go to www.dailymotion.com/video/xhq06_dengue-fever-vaccine-developed-in-
thailand_news to answer the following questions.

21. Where are scientists working on a Dengue vaccine? ___________________________

22. Why is it so difficult to make a working vaccine? ______________________________
________________________________________________________________________

23. If human trials are successful, how long will it be until a Dengue vaccine is available?
________________________________________________________________________

Now go to www.bbc.co.uk/2/hi/science/nature/7814539.stm.

24. How are scientists trying to stop mosquitoes from breeding? ________________
________________________________________________________________________
Mosquitoes!

1. What do mosquito larvae feed on?
2. Why do only female mosquitoes need blood?
3. How many wings does a mosquito have?
4. What is a vector?
5. Name three modern mosquito borne diseases found in Florida.
6. For IPM, which stage of the mosquito life cycle is most difficult to kill?
7. What is source reduction?
8. Why is an adulticide not very effective?
9. What order are all insects in?
10. What are some ways to determine what genus a mosquito is in?
11. What is a container larval habitat?
12. In which aquatic habitats will you not find mosquito larvae?
13. What is different about the feeding of Aedes egyptii?
14. How do mosquitoes recognize species for mating?
15. What do mosquitoes key on to find their victim?
16. How long does it take a female to digest a blood meal?

17. What is synchronous emergence?

18. T/F It is best to treat all potential larval water sources to make sure they are killed.

19. What is the dipper method?

20. Name three aquatic plants that some mosquito larvae live in.

21. How is a landing rate conducted?
## Glog Pathogen Presentation

Now that you have experience with the Glog format, your next challenge is to focus on an emerging/re-emerging pathogen.

<table>
<thead>
<tr>
<th>Component</th>
<th>Requirements</th>
</tr>
</thead>
<tbody>
<tr>
<td>Photo Gallery</td>
<td>At least 5 pictures which can be from your pathogen 10 pts.</td>
</tr>
<tr>
<td>Video Clips</td>
<td>At least 2 video clips about your pathogen. 20 pts.</td>
</tr>
<tr>
<td>Cause</td>
<td>What actually causes the disease? 5 pts.</td>
</tr>
<tr>
<td>Symptoms</td>
<td>What are the symptoms of the disease? 5 pts.</td>
</tr>
<tr>
<td>Mechanism</td>
<td>How does the disease affect biology? 10 pts.</td>
</tr>
<tr>
<td>Treatments</td>
<td>How is the disease treated? 5 pts.</td>
</tr>
<tr>
<td>Vectors</td>
<td>Are there any vectors? 5 pts.</td>
</tr>
<tr>
<td>Prevention</td>
<td>How can disease transmission be prevented? 5 pts.</td>
</tr>
<tr>
<td>Data</td>
<td>U.S. and Worldwide infection rates for the last 20 years. A data table is acceptable. 5 pts.</td>
</tr>
<tr>
<td>Outbreaks</td>
<td>Map with U.S. or global outbreaks. 10 pts.</td>
</tr>
<tr>
<td>Bibliography</td>
<td>At least 10 sources in proper format. 10 pts.</td>
</tr>
<tr>
<td>Aesthetics</td>
<td>Glog must be attractive with working links and error free. 20 pts.</td>
</tr>
</tbody>
</table>

The Glog counts as a test grade for the fourth quarter. It is due by April 6th, which means it must be uploaded before 11:59 PM on April 6th. It is irresponsible to attempt to complete the Glog at the last minute and then use technical issues as an excuse for not having the project completed. Any technical problems must be brought to my attention in writing at least 72 hours before the due date.