

Title:**May the Odds Be Ever in Our Favor!
Facts about Genetically Modified Organisms**

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ABSTRACT:

This four-day study is designed to combine the study of genetic modification in the agricultural industry with acquisition of laboratory skills and understanding emerging pathogens.

Background is presented, followed by a case study of papaya ringspot virus. The students are introduced to the disease, its causative pathogen, papaya ringspot virus (PRSV), and vector (aphids). Other GMO cases, including citrus greening (HLB), are briefly considered and further research is encouraged.

The students learn proper pipetting techniques and the mechanism of GMO detection using enzyme-linked immunosorbent assay (ELISA) simulation. Using a puzzle (sequencing activity) that illustrates the steps of the antigen-antibody reaction, they learn the basis of the test and use it to “test” several foods for the presence of GMO.

Using background knowledge, class laboratory results, and additional research, each student will individually prepare a report, in the form of a letter, that will detail the student’s fully supported recommendations concerning GMO, its value, safety, labeling, ethics, and other valid concerns.

RATIONALE:

Human beings have a long history of modifying the environment to meet their needs – with mixed results. If we have learned nothing else from history, we have learned that every thing we do to modify nature has consequences. In this lesson, the student is introduced to the relatively recent practice of genetic modification and the process of producing the genetically modified organism (GMO). The lesson is based on the study of the papaya crop in Hawaii and its decimation by the papaya ringspot virus (PRSV). The lesson uses the production of the genetically modified “Rainbow” papaya as an example of both the successful solution to a biological problem and the source of a massive local, national, and worldwide debate that addresses the need for more comprehensive testing for human and environmental safety, genetic copywriting, truth in labeling, and ethical imperatives.

The student learns the basic genetic modification process and the scientific principles upon which it is based. He/she is also introduced to the antibody-antigen mechanism by which the ELISA procedure detects the presence of GMO. Previously acquired skills are reinforced, including laboratory set up, multiple trials, and the use of positive and negative controls. New laboratory skills are introduced including accurate and sterile pipetting skills and interpretation of color change results.

TIMELINE OVERVIEW:

The study will be completed in 4 days. Class periods average 50 minutes. A posttest will be given later to allow students time to complete their further research, write their letters, and study their notes and results.

Day 1 – Introduction to GMO (power point – 20 minutes)
Video with handout (10 minutes)
Background and Introduction to the papaya problem (20 minutes)

Day 2 – Pipetting skills – *Pipetting by Design* activity
Introduction and instruction (10 minutes)
Lab Activity (30 minutes)
Introduction to the ELISA
Sequencing activity (10 minutes)

Day 3 – ELISA simulation test for GMO in common food items (45 minutes)

Day 4 – ELISA Discussion and Letter writing assignment (50 minutes)

STUDENT OUTCOMES:

The student will:

- Define and describe the interactions between a pathogen, a vector, and a host. (background)
- Discuss ways to interrupt the pathogen-vector-host relationship. (background)
- Describe the structure and function of DNA and relate that structure and function to the process of producing GMO. (content)
- Outline the basic process of GMO production. (content)
- Accurately pipette specified measured amounts into appropriate wells on a well plate and micro tube. (laboratory skill)
- Successfully interpret the presence or absence of GMO genes in a food source using ELISA results. (laboratory skill)
- Relate the antigen-antibody basis of the ELISA procedure. (background)
- Describe the outcomes of GMO and their effect on agriculture. (application)
- Discover the positive and negative outcomes of GMO that are concerns to society. (extension)
- Articulate a fully supported position on the issue of GMO. (extension)

STANDARDS*:

- GSI.2. Perform systematic observations.
- GSI.5. Use tools to gather, analyze, and interpret data.
- GSI.7. Use appropriate evidence and reasoning to justify explanations to others.
- GSI.8. Communicate the results of scientific investigations.
- GSI.9. Evaluate the merits of explanations produced by others.
- GSI.12. Recognize that the strength or usefulness of a scientific claim is established through logical argumentation and includes active consideration of alternative scientific explanations.
- GSS.1. Identify ways in which science influences society and is influenced by society.
- GSS.2. Identify sources of information and assess their reliability.
- GSS.3. Weigh the merits of alternative strategies by comparing a number different costs and benefits (human, economic, environmental).
- GSS.5. Discuss the relationship between faith, science, and reason and explain how the principles and patterns of nature can be indicative of a higher power with purpose.
- LS.4. Compare and contrast the general structures, and the functions, found in prokaryotic and eukaryotic cells.
- LS.14. Describe the structure of DNA and why that structure is vital to its function.
- LS.16. Explain the processes involved in genetic engineering and discuss its impact on human life and society.
- LS.22. Characterize the life cycles, reproductive methods, structure, and reproductive requirements of bacteria, protists, fungi, and viruses.

- LS.23. Characterize the life cycles, reproductive methods, structure, and reproductive requirements of members of the plant and animal kingdoms.
- LS.26. Recognize the positive and negative anthropogenic influences on the biosphere.

*These standards refer to Jesuit High School, Tampa Florida, Science Standards

DATA COLLECTION:

- Pretest assessment of prior knowledge
- Video worksheet
- Lab Assignments: Designer plates and ELISA simulation
Participation
Reports (responses)
- Antigen-antibody activity results
- Individual student-produced position paper in the form of a letter to a legislator
- Posttest assessment of knowledge and skills gained

USE OF UF ICORE EQUIPMENT LOCKERS:

Pipetting Stations
Designer plates activity
(ELISA simulation kits created during ICORE 2013)

ICORE SUMMER INSTITUTE CONNECTIONS:

- Morris, J. Glenn. *Emerging Pathogens and Pandemics: Things that go bump in the Night*. Presentation to UF HHMI ICORE Summer Institute, June 9, 2013.
- Gabriel, Dr. Dean. *Transgenic Citrus with Immunity to Citrus Greening Disease*, Presentation to UF HHMI ICORE Summer Institute, June 10, 2013.
- Lab Activity: *Designer Plates*. UF HHMI ICORE Summer Institute, June 10, 2013.
- Green, Linda. *ELISA Technique*. Presentation to UF HHMI ICORE Summer Institute, June 10, 2013
- Bokor, Julie. *Dengue curriculum (ELISA simulation)*. Presentation to UF HHMI ICORE Summer Institute, June 19, 2013
- Bokor, Julie. *Dengue curriculum (Antigen-Antibody Activity)*. Presentation to UF HHMI ICORE Summer Institute, June 19, 2013

IMPROVEMENT ON TRADITIONAL TEACHING TECHNIQUES:

Genetic modification is a topic that was previously only mentioned in a list of biotechnologies when discussing the practical application of science. This topic usually only comes up in that chapter one discussion of “what is science?” The students have none of the prerequisite knowledge to even begin to discuss genetic modification at this time - so I never have.

Up to the minute information, and specific examples of the uses of genetic modification, give me the confidence to expand on my classroom instruction in this area. The importance of this process to our food supply (citrus greening, papaya modification, anti-herbicide gene) and the widespread opposition to its utilization, make GMO an important topic for students to consider as informed citizens and eventual voters.

The GMO simulation activity models for students the concept that many of the foods they actually eat are modified and that it isn't noticeable or particularly scary. The background information also shows them that not just particular food products, but the nutrition and livelihood of many people depend on the preservation of food industries. The moral imperative of this technology in the feeding of the world's poor is also explored.

Presentation of opposing viewpoints is also a vital part of this unit. Any one-sided presentation is automatically suspect – as it should be. The students should understand that individuals, groups and entire nations have concerns about the safety and ethics of this process.

The more complete coverage of this topic is certainly an improvement over practically ignoring such an important biological, social, and political current topic

BUDGET AND BUDGET JUSTIFICATION:

The following items are the equipment and consumables needed to complete this series of activities.

UF ICORE EQUIPMENT LOCKERS: (no cost to teacher or school)

Pipetting Stations – contains classroom set of micropipettes and pipette tips
Designer plate activity – contains classroom set of 96-well plates

ICORE ELISA simulation kit – contains teacher made fluorescent plates (8), required test tubes. UV light

BIBLIOGRAPHY

Video:

Green, H. (Performer) (2013). *The science of genetically modified food* [Web]. Retrieved from <http://www.youtube.com/watch?v=vUzVm-zpyR8>

Fictional reference:

Collins, S. (2008). *The hunger games*. New York: Scholastic, Inc.

Citrus Greening references:

Florida Department of Citrus, (2010). *Florida grapefruit*. Retrieved from website: <http://www.gofloridagraperfruit.com/about-fdoc/>

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Kamiya, K. (2012, May 31). Hawaii's biotech papayas hold a lesson for America. *Truth About Trade Technology*, Retrieved from <http://www.truthabouttrade.org/2012/05/31/hawaiis-biotech-papayas-hold-a-lesson-for-america/>

Vatican Reference:

(2010). Vatican scientists see "moral imperative" in GMO. *GMO compass*, Retrieved from http://www.gmoccompass.org/eng/news/547.vatican_scientists_see

[_moral_imperative_gmo.html](#)

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Title:

**May the Odds Be Ever in Our Favor!
What Are Genetically Modified Organisms?**

**LESSON PLAN – DAY 1
INTRODUCTION TO GMO AND CASE STUDY**



TARGET GRADE LEVEL: First year biology students (grades 9 or 10)

KEY QUESTIONS:

1. What key historical decisions may be considered when deciding on a position concerning GMOs?
2. What organism and vector are responsible for the papaya ringspot virus (PRSV)?
3. What were the effects of PRSV on the papaya industry in Hawaii?
4. How did GMO “rainbow” papaya affect the papaya industry in Hawaii?
5. What are some of the objections to GMO?
6. By what processes are GMO organisms produced?
7. Why is there so little independent testing of the effects of GMO foods?

OVERALL TIME ESTIMATE: 50 minutes

LEARNING STYLES: Visual and auditory

VOCABULARY:

Pathogen	Transgenics	Glyphosate (Round Up)
Hybrid	Psyllid	Gene gun
GMO	moral imperative	Agrobacterium
Aphid	HLB	Backcross breeding
PRSV	commodity crops	Resistance

LESSON SUMMARY: Using an introductory powerpoint, video with handout, and a case study powerpoint, the student is introduced to the topic of production of genetically modified foods and their impact on agriculture in the United States.

STUDENT LEARNING OBJECTIVES:

The student will:

- Define and describe the interactions between a pathogen, a vector, and a host. (background)
- Discuss ways to interrupt the pathogen-vector-host relationship. (background)
- Describe the structure and function of DNA and relate that structure and function to the process of producing GMO. (content)
- Outline the basic process of GMO production. (content)

STANDARDS*

- GSI.7. Use appropriate evidence and reasoning to justify explanations to others.
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- GSI.9. Evaluate the merits of explanations produced by others.
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- LS.26. Recognize the positive and negative anthropogenic influences on the biosphere.

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MATERIALS:

Powerpoint and notes – **May the Odds Be Ever in Our Favor –Introduction**
 Access to video (youtube) <http://www.youtube.com/watch?v=vUzVm-zpyR8>
 Video questions - **May the Odds Be Ever in Our Favor – Video Questions**

Powerpoint and notes - **May the Odds Be Ever in Our Favor – Case Study**

BACKGROUND INFORMATION FOR TEACHER:

INTRODUCTION:

In the popular Hunger Games trilogy, a (fictional) bird, called the mockingjay is produced when the Capitol (the government) releases jabberjays into the wild, expecting them to die out. Jabberjays, all males, were genetically enhanced birds that the Capitol used to spy on the people. Instead of dying as expected, the jabberjays bred with mockingbirds and the offspring were hybrids called mockingjays and their hybrid traits were eventually used against the Capitol. This bird came to symbolize the revolution that occurs in that series because of its resistance, resilience, and persistence.

From the beginning of time, man has altered his environment for his own benefit. Beginning with early agricultural practices, the land produced and mankind benefited. When man got greedy, or careless, the land responded by no longer producing (think dust bowl). As man continues to overpopulate, we see the reemergence of pathogens that were once under better control. (cholera) As food-handling mega-corporations handle tons of products, mass contamination events are on the rise. (*e. coli*) While we continue to deforest and incur into formerly uninhabited areas, previously unknown pathogens emerge. (ebola)

What lessons can be learned about genetically engineering organisms from history and from the story of the mockingjay? What are the risks and benefits of research and development of GMOs – genetically modified organisms? Could GMOs created to increase crop yields or improve an organism's resistance to disease eventually hybridize with a wild animal or plant? Can this happen in the real world? Do we know for sure?

What we do know for sure is that GMOs are already here. And they got here using a relatively simple process.

The following video will explain the process:

THE SCIENCE OF GENETICALLY MODIFIED FOOD

<http://www.youtube.com/watch?v=vUzVm-zpyR8> (10 minutes)

References Cited:

1. "Papayas." GMO Compass (database). Accessed 13 June 2013.
http://www.gmo-compass.org/eng/grocery_shopping/fruit_vegetables/14.genetically_modified_pa

payas_virus_resistance.html

2. "Japan Approved GM Papaya." The Gain Report of the USDA Foreign Agricultural Service. 2011, 19 December.
http://gain.fas.usda.gov/Recent%20GAIN%20Publications/Japan%20approved%20GM%20papaya_Tokyo_Japan_12-19-2011.pdf
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<http://www.truthabouttrade.org/2012/05/31/hawaiis-biotech-papayas-hold-a-lesson-for-america/>

CASE STUDY:

Now that you know a little bit about the process used to make GMOs, consider the following problem:

Papaya is a major crop of the state of Hawaii. A specific virus, the papaya ringspot virus (PRSV) infects papaya and some members of the melon family. It is transmitted between plants by pruning, but mostly by an insect vector of one of numerous aphid species.

Around 1950 the PRSV was introduced into Hawaii and within a decade, production of papaya had dropped over 90%. Symptoms are typical of viral diseases. Papaya exhibits yellowing, leaf distortion, and severe mosaic. The fruit will exhibit bumps and the classic "ringspot". By 1995, despite efforts to contain the virus, commercial production was impossible on some islands and severely limited on the others.

In the late 1980s, the University of Hawaii developed a papaya resistant to PRSV [by genetically modifying papayas](#). Certain viral genes were transferred into the papaya genome, creating an "immune-like response" from the papaya plant. These new plants are no longer susceptible to PRSV. (1)

The first virus resistant or "Rainbow" papayas were grown in Hawaii in 1999. Transgenic papayas now make up 75% of the papaya crop in Hawaii. These papayas are approved for consumption in the US and Canada, and were very recently approved for shipment into Japan. They are the first genetically modified (GM) food approved in Japan. (2)

This is a story of how cutting edge agriculture saved a major Hawaiian crop industry and many livelihoods in that state. Papaya is a local industry worth \$11 million annually. (3)

Not everyone in Hawaii is happy about the transgenic papayas. Concern about safety has created a significant backlash. Terrorist opponents have destroyed

papaya plantations under the cover of darkness. Farmers have lost tens of thousands of dollars worth of trees in these attacks.

The Vatican has even weighed in on the topic. In a statement released at the end of November 2010, forty international scientists including seven Vatican advisors have called for the relaxation of “excessive, unscientific regulations” applied to genetically modified crops. The scientists cited the “magnitude of challenges facing the world’s poor and undernourished” as a “matter of urgency” and the making of the benefits of GE available to poor and vulnerable populations a “moral imperative”. (4)

What if you don’t even like papaya and no one you know is economically affected by the papaya industry. So what? Well, how do you feel about orange juice and whether you know it or not, all of the residents of Florida are economically impacted by the citrus industry. What if something like that happened to citrus? Well it has. It is called citrus greening (Huanglongbing or HLB), and it is working on wiping out the citrus industry in Florida.

Like PRSV, HLB is a vector borne pathogen that was first noticed in Florida in 2005. Eight years later, it can be found in every citrus-producing county in Florida. The fruit from infected trees has an unacceptable flavor, and the virus is eventually fatal to the tree. The vector is an Asian psyllid that can easily move



from tree to tree and from grove to grove.

According to the Florida Department of Citrus, the industry employs approximately 76,000 workers and has an annual economic impact of 9 billion dollars. (5) The United States leads the world in grapefruit production supplying the world with over 30% of its grapefruit, and is the third largest overall citrus producer in the world. The majority of the citrus grown in the United States comes from Florida. In fact Florida produces three times as many tons of oranges and four times as many tons of grapefruit as its closest competitor, California. (6) With over 1/2 million acres of citrus groves and 74 million trees, Florida is second only to Brazil in orange juice production and supplies approximately 80% of the orange juice in the United States during any given growing season. (7)

You will probably not be surprised to learn that the scientists at the University of Florida are following the example of those at the University of Hawaii and are working on the creation of HLB resistant citrus.

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1. "Papayas." GMO Compass (database). Accessed 13 June 2013.
http://www.gmo-compass.org/eng/grocery_shopping/fruit_vegetables/14.genetically_modified_papayas_virus_resistance.html
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http://www.gmocompass.org/eng/news/547.vatican_scientists_see_moral_imperative_gmo.html

ADVANCE PREPARATION FOR TEACHER:

Prepare copies of the notes from each powerpoint and the video study questions for each student. These pages begin on the next page.

May the Odds Be Ever in Our Favor!

LESSON ONE - INTRODUCTION



In the popular Hunger Games trilogy, a (fictional) bird, called the mockingjay is produced when the Capitol (the government) releases jabberjays into the wild, expecting them to die out. Jabberjays, all males, were genetically enhanced birds that the Capitol used to spy on people. Instead of dying as expected, the jabberjays bred with mockingbirds and the offspring were hybrids called mockingjays and their hybrid traits were eventually used against the Capitol. This bird came to symbolize the revolution that occurs in that series because of its resistance, resilience, and persistence.

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What lessons can be learned about genetically engineering organisms from history and from the story of the mockingjay? What are the risks and benefits of research and development of GMOs – genetically modified organisms? Could GMOs created to increase crop yields or improve an organism's resistance to disease eventually hybridize with a wild animal or plant? Can this happen in the real world? Do we know for sure?

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The following video will explain the process: THE SCIENCE OF

GENETICALLY MODIFIED FOOD

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May the Odds Be Ever in Our Favor!

VIDEO: THE SCIENCE OF GENETICALLY MODIFIED FOOD STUDY QUESTIONS



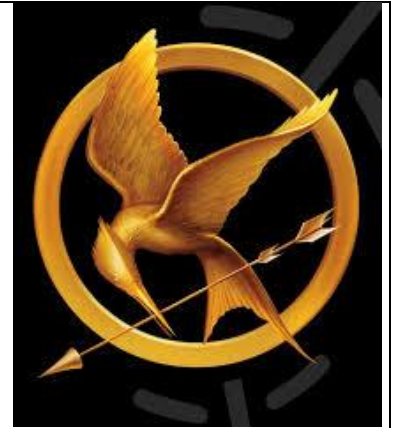
<http://www.youtube.com/watch?v=vUzVm-zpyR8> (10 minutes)

1. What types of genetically modified (GM) foods might you be eating every day?
2. What was the first GM animal approved by the USDA for human consumption?
3. What traits are being engineered in crops like soy, corn, and sugar beets?
4. Why have some countries like Russia and Peru banned the use and import of GM foods?
5. What should GMO foods more accurately be called?
6. In what ways have we been genetically modifying plants and animals for thousands of years?
7. What is transgenics?
8. How did scientists increase genetic diversity in plants in the 1920s?
9. What tactic did scientist use to modify plants in 1983?
10. What GM product was approved by the USDA in 1994?
11. What type of crop is most of the GM foods produced today?
12. What are Round-Up ready crops?
13. How are crops engineered to be Round-Up ready?
14. What is a gene gun?
15. What is an agrobacterium?
16. What is the use of the agrobacteria?
17. What is backcross breeding and how long does it take?
18. Why is there so little independent research on the possible environmental and health impacts of GM foods in the United States?
19. Describe some of the results of independent testing done in European countries.

20. What is definitely working against GM crops?

May the Odds Be Ever in Our Favor!

VIDEO: THE SCIENCE OF GENETICALLY MODIFIED FOOD STUDY QUESTIONS WITH ANSWERS



1. What types of genetically modified (GM) foods might you be eating every day?

Sugar beets (sugary products), soybeans (mayonnaise), hamburger (cows eat GM feed)

2. What was the first GM animal approved by the USDA for human consumption?

Arctic Salmon made from salmon with genes from the Pacific pout.

3. What traits are being engineered in crops like soy, corn, and sugar beets?

Herbicide and pesticide resistance

4. Why have some countries like Russia and Peru banned the use and import of GM foods?

GM foods are expensive to produce and their impacts on the environment and human health are yet unknown

5. What should GMO foods more accurately be called?

Genetically engineered organisms

6. In what way have we been genetically modifying plants and animals for thousands of years?

Artificial selection (selective breeding)

7. What is transgenics?

Transgenics is a process in which genes from one species are extracted and inserted into the genes of another species

8. How did scientists increase genetic diversity in plants in the 1920s?

Scientists caused mutations in plants by exposing them to x-rays, gamma rays and chemicals

9. What tactic did scientist use to modify plants in 1983?

Scientists took the gene that caused a particular bacterium to be resistant to antibiotics and inserted it into the DNA of a tobacco plant. They created an antibiotic resistant plant that was not really good for anything.

10. What GM product was approved by the USDA in 1994?

The flavor saver tomato – a fruit genetically engineered to ripen more slowly so it had a longer shelf life. Turns out it didn't taste so good.

11. What type of crop is most of the GM foods produced today?

Commodity crops – like feed corn and soybeans that are used in processed foods and as feed for animals

12. What are Round-Up ready crops?

Crops that are engineered to be resistant to glyphosate (Round-Up, a common herbicide made by Monsanto)

13. How are crops engineered to be Round-Up ready?

Glyphosate resistant bacteria produce an enzyme that causes them to be unaffected by the chemical. The small pieces of DNA for that enzyme are extracted and introduced into the crop plant.

14. What is a gene gun?

A gene gun is a device that is used to blast DNA into plant cells. Gold is coated with the desirable donor genes (called transgenes) and blasted into the plant cells. Once inside the nucleus of the plant cell, the gold dissolves. The transgenes are taken up by the chromosomes of the plant. The plant with the new transgenic DNA can then be bred into GM plants.

15. What is an agrobacterium?

An agrobacterium is a soil dwelling bacterium with an extra piece of DNA called a plasmid. The plasmid can move outside the bacterium and implant itself into a plant cell.

16. What is the use of the agrobacteria?

Agrobacteria are used as carriers for transgenes.

17. What is backcross breeding and how long does it take?

Backcross breeding is repeated crossing of the new transgenic plant with breeding stock until a new GM crop is produced. It can take up to 15 years to complete.

18. Why is there so little independent research on the possible environmental and health impacts of GM foods in the United States?

GMOs are patented and research related to GMOs is tightly controlled by their manufacturers.

19. Describe some of the results of independent testing done in European countries.

One study has seen increased cancer rates and organ damage in rodents. Another study found that mice fed Round-Up ready soybeans made fewer digestive enzymes and had modifications of their liver cells.

20. What is definitely working against GM crops?

Nature. Over time there has been a dilution of the effects of GMOs. There are now pest resistance to BT (an insect resistance modification) and “superweeds” resistant to Round-Up. GM seeds escape from fields and cross breed with “wild strains”.

RESOURCES/REFERENCES:

Video:

Green, H. (Performer) (2013). *The science of genetically modified food* [Web]. Retrieved from <http://www.youtube.com/watch?v=vUzVm-zpyR8>

Fictional reference:

Collins, S. (2008). *The hunger games*. New York: Scholastic, Inc.

May the Odds Be Ever in Our Favor!

CASE STUDY – HAWAIIAN PAPAYA



Now that you know a little bit about the process used to make GMOs, consider the following problem:

Papaya is a major crop of the state of Hawaii. A specific virus, the papaya ringspot virus (PRSV) infects papaya and some members of the melon family. It is transmitted between plants by pruning, but mostly by an insect vector of one of numerous aphid species.

Around 1950 the PRSV was introduced into Hawaii and within a decade, production of papaya had dropped over 90%. Symptoms are typical of viral diseases. Papaya exhibits yellowing, leaf distortion, and severe mosaic. The fruit will exhibit bumps and the classic "ringspot". By 1995, despite efforts to contain the virus, commercial production was impossible on some islands and severely limited on the others.



Papaya infected with PRSV

In the late 1980s, the University of Hawaii developed a papaya resistant to PRSV. Certain viral genes were transferred into the papaya genome, creating an "immune-like response" from the papaya plant. These new plants are no longer

susceptible to PRSV. (1)

The first virus resistant or “Rainbow” papayas were grown in Hawaii in 1999. Transgenic papayas now make up 75% of the papaya crop in Hawaii. These papayas are approved for consumption in the US and Canada, and were very recently approved for shipment into Japan. They are the first genetically modified (GM) food approved in Japan. (2)

This is a story of how cutting edge agriculture saved a major Hawaiian crop industry and many livelihoods in that state. Papaya is a local industry worth \$11 million annually. (3)

Not everyone in Hawaii is happy about the transgenic papayas. Concern about safety has created a significant backlash. Terrorist opponents have destroyed papaya plantations under the cover of darkness. Farmers have lost tens of thousands of dollars worth of trees in these attacks.

The Vatican has even weighed in on the topic. In a statement released at the end of November 2010, forty international scientists including seven Vatican advisors have called for the relaxation of “excessive, unscientific regulations” applied to genetically modified crops. The scientists cited the “magnitude of challenges facing the world’s poor and undernourished” as a “matter of urgency” and the making of the benefits of GE available to poor and vulnerable populations a “moral imperative”. (4)

What if you don’t even like papaya and no one you know is economically affected by the papaya industry. So what? Well, how do you feel about orange juice and whether you know it or not, all of the residents of Florida are economically impacted by the citrus industry. What if something like that happened to citrus? Well it has. It is called citrus greening (Huanglongbing or HLB), and it is working on wiping out the citrus industry in Florida.

Like PRSV, HLB is a vector borne pathogen that was first noticed in Florida in 2005. Eight years later, it can be found in every citrus-producing county in Florida. The fruit from infected trees has an unacceptable flavor, and the virus is eventually fatal to the tree. The vector is an Asian psyllid that can easily move



from tree to tree and from grove to grove.

According to the Florida Department of Citrus, the industry employs approximately 76,000 workers and has an annual economic impact of 9 billion dollars. (5) The United States leads the world in grapefruit production supplying the world with over 30% of its grapefruit, and is the third largest overall citrus producer in the world. The majority of the citrus grown in the United States comes from Florida. In fact Florida produces three times as many tons of oranges and four times as many tons of grapefruit as its closest competitor, California. (6) With over 1/2 million acres of citrus groves and 74 million trees, Florida is second only to Brazil in orange juice production and supplies approximately 80% of the orange juice in the United States during any given growing season. (7)

You will probably not be surprised to learn that the scientists at the University of Florida are following the example of those at the University of Hawaii and are working on the creation of HLB resistant citrus.

References Cited:

1. "Papayas." GMO Compass (database). Accessed 13 June 2013.
http://www.gmo-compass.org/eng/grocery_shopping/fruit_vegetables/14.genetically_modified_papayas_virus_resistance.html
2. "Japan Approved GM Papaya." The Gain Report of the USDA Foreign Agricultural Service. 2011, 19 December.
http://gain.fas.usda.gov/Recent%20GAIN%20Publications/Japan%20approved%20GM%20papaya_Tokyo_Japan_12-19-2011.pdf
3. Kamiya, Ken. "Hawaii's Biotech Papayas Hold a Lesson for America." Truth About Trade Technology. 2012, 31 May.
<http://www.truthabouttrade.org/2012/05/31/hawaiis-biotech-papayas-hold-a-lesson-for-america/>
4. "Vatican scientists see "moral imperative" in GMO." GMO Compass. 3 December 2010.
http://www.gmocompass.org/eng/news/547.vatican_scientists_see_moral_imperative_gmo.html
5. "Florida Grapefruit." Florida Department of Citrus. 2010. 30 Aug. 2011.
<http://www.gofloridagrapefruit.com/about-fdoc/>
6. "Background Statistics: U.S. Citrus Market ." United States Department of Agriculture. 22 Jan. 2007. 28 Aug. 2011.

<http://www.ers.usda.gov/News/citruscoverage.htm>

7. "Citrus Facts." Florida Citrus. 2008, 30 August 2011.
<http://www.floridajuice.com/juice.php>.

Citrus Greening references:

Florida Department of Citrus, (2010). *Florida grapefruit*. Retrieved from website:
<http://www.gofloridagraperfruit.com/about-fdoc/>

United States Department of Agriculture, (2007). *Background statistics: U.S. citrus market*. Retrieved from website:
<http://www.ers.usda.gov/News/citruscoverage.htm>

Florida citrus, (2008). *Citrus facts*. Retrieved from website:
<http://www.floridajuice.com/juice.php>.

Papaya Ringspot Virus references:

Papayas. *GMO compass-online database*, Retrieved from http://www.gmo-compass.org/eng/grocery_shopping/fruit_vegetables/14.genetically_modified_papayas_virus_resistance.html

USDA foreign agriculture service, Gain report. (2011). *Japan approved gm papaya (JA1048)*. Retrieved from website:
http://gain.fas.usda.gov/Recent%20GAIN%20Publications/Japan%20approved%20GM%20papaya_Tokyo_Japan_12-19-2011.pdf

Kamiya, K. (2012, May 31). Hawaii's biotech papayas hold a lesson for America. *Truth About Trade Technology*, Retrieved from
<http://www.truthabouttrade.org/2012/05/31/hawaiis-biotech-papayas-hold-a-lesson-for-america/>

Vatican Reference:

(2010). Vatican scientists see "moral imperative" in GMO. *GMO compass*, Retrieved from
http://www.gmocompass.org/eng/news/547.vatican_scientists_see_moral_imperative_gmo.html

Additional References and Resources:

University of Florida ICORE, June, 2013.

SUGGESTED ANSWERS TO VOCABULARY:

Pathogen - a disease-causing organism

Hybrid – an offspring resulting from crossbreeding- in this case, a GMO with a nonGMO.

GMO – genetically modified organisms

Aphid – a small insect known to act as a vector for PRSV

PRSV – papaya ringspot virus – a disease fatal to papaya and other melons.

Psyllid – a small insect known to act as a vector for HLB

Transgenics - a process in which genes from one species are extracted and inserted into the genes of another species

Moral imperative – something that must occur because it is the right or virtuous thing to do.

Agrobacterium - a soil dwelling bacterium with an extra piece of DNA called a plasmid. The plasmid can move outside the bacterium and implant itself into a plant cell.

HLB - a deadly bacterial disease of citrus called Huanglongbing, or citrus greening disease.

Gene gun - a device that is used to blast DNA into plant cells.

Glyphosate (Round Up) - a common herbicide made by Monsanto.

Commodity crops - crops like feed corn and soybeans that are used in processed foods and as feed for animals

Backcross breeding - repeated crossing of the new transgenic plant with breeding stock until a new GM crop is produced.

Resistance – a term used to describe an organism that is no longer affected by a drug or chemical as in weeds no longer affected by Round Up.

ASSESSMENT SUGGESTIONS:

To assess preparation and readiness

Vocabulary assessment

To assess understanding and communication of relevant concepts

Questioning and contribution to discussion

To assess overall understanding and application to the larger topic.

Questioning and contribution to discussion
Evaluate answer to video questions

Title:

**May the Odds Be Ever in Our Favor!
What Are Genetically Modified Organisms?**

**LESSON PLAN – DAY 2
PIPETTING SKILLS – PIPETTING BY DESIGN,
ELISA ANTIGEN-ANTIBODY BACKGROUND
INTRODUCTION**



TARGET GRADE LEVEL: First year biology students (grades 9 or 10)

KEY QUESTIONS:

1. How do you properly use a micropipette?
2. What is an antigen-antibody reaction?
3. What are the steps of an antigen-antibody reaction?

OVERALL TIME ESTIMATE: 50 minutes

LEARNING STYLES: Visual, auditory, kinesthetic

VOCABULARY:

Micropipette Substrate
Microliter
Antigen
Antibody

LESSON SUMMARY: Using the designer plate procedure, the student will develop micropipetting skills. When finished with the pipetting activity, the student will complete a puzzle-like antigen-antibody reaction sequencing activity as an introduction to the mechanism of the ELISA procedure.

STUDENT LEARNING OBJECTIVES:

The student will ...

- Accurately pipette specified measured amounts into appropriate wells on a well plate and micro tube. (laboratory skill)
- Relate the antigen-antibody basis of the ELISA procedure. (background)

STANDARDS*

- GSI.5. Use tools to gather, analyze, and interpret data.

- GSS.1. Identify ways in which science influences society and is influenced by society.
- LS.4. Compare and contrast the general structures, and the functions, found in prokaryotic and eukaryotic cells.
- LS.23. Characterize the life cycles, reproductive methods, structure, and reproductive requirements of members of the plant and animal kingdoms.

*These standards refer to Jesuit High School, Tampa Florida, Science Standards

MATERIALS:

Pipetting by Design

Requires the use of UF ICORE equipment locker (micropipettes) and the pipetting by design kit

Contents (per group):

- 1 96 well plate
- Colored water
- 1 P20 micropipette
- 1 P200 micropipette
- electronic scale

Protocols for the designs produced can be found in the **ADVANCED PREPARATION** section

Sequencing activity materials:

- Per student or group:
- 1 set of sequencing cards
- 1 student worksheet

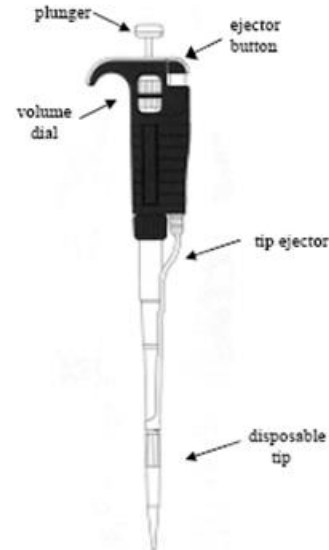
Instructions for the sequencing cards can be found in the **ADVANCED PREPARATION** section

BACKGROUND INFORMATION FOR TEACHER:

Pipetting by Design:

Micropipettes are precise instruments used to accurately measure very small quantities of liquids in science laboratories. Image 1 shows a micropipette and the main components of the instrument. They are available in a variety of sizes to best match your measurement needs. The size of the micropipette is indicated directly on the instrument. The most commonly used micropipettes are the P10, P20, P200, and P1000. The number following the “P” refers to the maximum volume in microliters (μl) that can be measured using the instrument.

In this activity, P20 and P200 micropipettes will be used. The proper method for reading the volume indicator and directions on how to use the P20 and P200 micropipettes are listed below:



Reading the volume on the micropipette:

- **P20 Micropipettes:** The volume indicator consists of three number dials and is read from top to bottom. Black digits indicate tens of microliters and microliters; red digits indicate tenths of microliters. A P20 is used to measure volumes up to 20 μ l. **NOTE: Do not dial past 20 μ l.**

0
7
3

7.3 μ l

2
0
0

20.0 μ l

- **P200 Micropipettes:** The volume indicator consists of three number dials and is read from top to bottom. Black digits indicate hundreds and tens of microliters; red digits indicate microliters. A P200 is used to measure volumes between 20 μ l and 200 μ l. **NOTE: Do not dial past 200 μ l.**

0
7
3

73 μ l

2
0
0

200.0 μ l

Directions on how to use a micropipette:

- Hold micropipette in one hand. With the other hand turn the black volume adjustment dial 1/3 of a revolution above the desired setting then slowly down until the required volume shows on the digital indicator. *This prevents mechanical backlash from affecting accuracy.*
- Press disposable tips firmly onto the shaft to ensure an airtight seal. Do this by tapping the micropipette in the tip (tapping the tip on).
- Depress plunger to the **first stop**. Holding the micropipette vertically, immerse the tip approximately two mm into the sample liquid. **Allow the pushbutton to return slowly to the up position!**
- Withdraw the tip from the liquid. Touch the tip end against the side wall of the receiving vessel and depress the plunger slowly to the first stop.
- Wait one second then press the plunger to the second stop, expelling any residual liquid in the tip.
- With the plunger fully depressed, withdraw micropipette and allow the plunger to slowly return to the up position.
- Discard the tip by depressing the ejector button. **Use a fresh tip for the next sample to avoid contamination.**

ELISA Sequencing Activity:

In ELISA, an unknown amount of antigen is affixed to a surface, and then a specific antibody is applied over the surface so that it can bind to the antigen. This antibody is linked to an enzyme, and, in the final step, a substance

containing the enzyme's substrate is added. The subsequent reaction produces a detectable signal, most commonly a color change in the substrate.

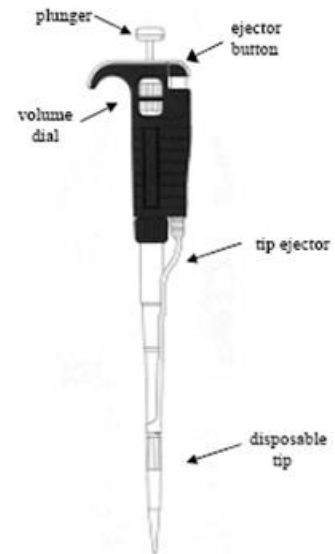
Performing an ELISA involves at least one antibody with specificity for a particular antigen. The sample with an unknown amount of antigen is immobilized on a solid support (usually a polystyrene microtiter plate) either non-specifically (via adsorption to the surface) or specifically (via capture by another antibody specific to the same antigen, in a "sandwich" ELISA). After the antigen is immobilized, the detection antibody is added, forming a complex with the antigen. The detection antibody can be covalently linked to an enzyme, or can itself be detected by a secondary antibody that is linked to an enzyme through bioconjugation. Between each step, the plate is typically washed with a mild detergent solution to remove any proteins or antibodies that are not specifically bound. After the final wash step, the plate is developed by adding an enzymatic substrate to produce a visible signal, which indicates the quantity of antigen in the sample.

May the Odds Be Ever in Our Favor! What Are Genetically Modified Organisms?

STUDENT LABORATORY SKILLS – PIPETTING BY DESIGN



Micropipettes are precise instruments used to accurately measure very small quantities of liquids in science laboratories. Image 1 shows a micropipette and the main components of the instrument. They are available in a variety of sizes to best match your measurement needs. The size of the micropipette is indicated directly on the instrument. The most commonly used micropipettes are the P10, P20, P200, and P1000. The number following the “P” refers to the maximum volume in microliters (μl) that can be measured using the instrument.



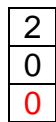
In this activity, P20 and P200 micropipettes will be used. The proper method for reading the volume indicator and directions on how to use the P20 and P200 micropipettes are listed below:

Reading the volume on the micropipette:

- **P20 Micropipettes:** The volume indicator consists of three number dials and is read from top to bottom. Black digits indicate tens of microliters and microliters; red digits indicate tenths of microliters. A P20 is used to measure volumes up to 20 μl . **NOTE: Do not dial past 20 μl .**



7.3 μl

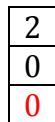


20.0 μl

- **P200 Micropipettes:** The volume indicator consists of three number dials and is read from top to bottom. Black digits indicate hundreds and tens of microliters; red digits indicate microliters. A P200 is used to measure volumes between 20 μl and 200 μl . **NOTE: Do not dial past 200 μl .**



73 μl



200.0 μl

Directions on how to use a micropipette:

- Hold micropipette in one hand. With the other hand turn the black volume adjustment dial 1/3 of a revolution above the desired setting then slowly down until the required volume shows on the digital indicator. *This prevents mechanical backlash from affecting accuracy.*
- Press disposable tips firmly onto the shaft to ensure an airtight seal. Do this by tapping the micropipette in the tip (tapping the tip on).
- Depress plunger to the **first stop**. Holding the micropipette vertically, immerse the tip approximately two mm into the sample liquid. **Allow the pushbutton to return slowly to the up position!**
- Withdraw the tip from the liquid. Touch the tip end against the side wall of the receiving vessel and depress the plunger slowly to the first stop.
- Wait one second then press the plunger to the second stop, expelling any residual liquid in the tip.
- With the plunger fully depressed, withdraw micropipette and allow the plunger to slowly return to the up position.
- Discard the tip by depressing the ejector button. **Use a fresh tip for the next sample to avoid contamination.**

Materials: 1 96 well plate
Beakers or tubes of colored water
1 P20 micropipette
1 P200 micropipette
electronic scale

Procedure:

1. Determine the mass of your 96 well plate. Be sure it is clean and dry. Record.
2. Follow the protocol assigned to your group precisely.
3. When you are finished, mass your 96 well plate with the liquid you have pipetted into it. Record.
4. Determine the mass of the liquid pipetted into the plate.

Data:

	Mass (g)
Clean, dry plate	
Plate after pipetting	
Liquid pipetted into plate	

Questions:

1. Which protocol did you follow? (the letter on the instruction sheet)
2. What design did you create in this activity?
3. What was the mass of the liquid you pipetted into the plate?

RESOURCES/REFERENCES:

Background Information Modified From: “Biotechnology Laboratory: Micropipet Technique.” Biotechnology In The Classroom- University of California Davis. N.p., 2002. Web. <<http://ceprap.ucdavis.edu>>.

Procedures and Protocols Modified From: “Pipetting by Design” Lesson Plan. ICORE 2013 Resources. University of Florida. June, 3013.

ELISA SEQUENCING ACTIVITY – STUDENT WORKSHEET

A common test used to detect if a patient has been exposed to a virus such as HIV, Dengue, or West Nile is called an ELISA (**E**nzyme **L**inked **I**mmuno**S**orbant **A**ssay). This test takes advantage of the interactions between antigens and antibodies. Often compared to a lock and key, an antigen/antibody interaction is very specific. ELISA tests usually take place in plastic plates containing wells, or depressions.

Match the statements and images below to sequence the steps of an ELISA test.

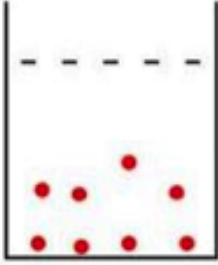
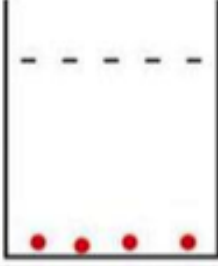
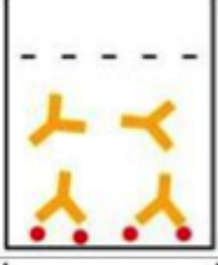
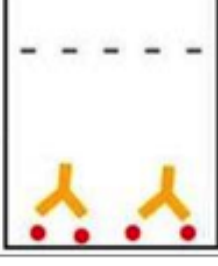
	<p>Virus proteins (antigens) are added to wells of a 96-well plate.</p>
	<p>The antigens bind to the plastic, coating the bottom of the wells.</p>
	<p>The primary antibody is added to the well. In the case of the dengue ELISA, the primary antibodies (IgM) are from the patient's serum sample.</p>
	<p>Excess antibody is washed away, leaving only antibodies bound to the antigens behind. This wash removes excess antibodies that are unbound and prevents non-specific binding.</p>

	<p>A secondary antibody is added to the wells. This antibody recognizes the patient IgM antibodies, bound to the antigens. The secondary antibody also has a colorimetric tag attached.</p>
	<p>Excess secondary antibody is washed away, leaving only secondary antibodies, bound to the patient IgM antibodies. This wash removes excess antibodies that are unbound.</p>
	<p>A substrate is added to the wells.</p>
	<p>Bound secondary antibody containing a colorimetric tag will cause a color change when exposed to the substrate. A color change indicates a positive reaction.</p>

ELISA SEQUENCING ACTIVITY - TEACHER ANSWER SHEET

A common test used to detect if a patient has been exposed to dengue virus is called an ELISA (Enzyme Linked ImmunoSorbant Assay). This test takes advantage of the interactions between antigens and antibodies. Often compared to a lock and key, an antigen/antibody interaction is very specific. ELISA tests usually take place in plastic plates containing wells, or depressions.

Match the statements and images below to sequence the steps of an ELISA test.

	Virus proteins (antigens) are added to wells of a 96-well plate.
	The antigens bind to the plastic, coating the bottom of the wells.
	The primary antibody is added to the well. In the case of the dengue ELISA, the primary antibodies (IgM) are from the patient's serum sample.
	Excess antibody is washed away, leaving only antibodies bound to the antigens behind. This wash removes excess antibodies that are unbound and prevents non-specific binding.

	<p>A secondary antibody is added to the wells. This antibody recognizes the patient IgM antibodies, bound to the antigens. The secondary antibody also has a colorimetric tag attached.</p>
	<p>Excess secondary antibody is washed away, leaving only secondary antibodies, bound to the patient IgM antibodies. This wash removes excess antibodies that are unbound.</p>
	<p>A substrate is added to the wells.</p>
	<p>Bound secondary antibody containing a colorimetric tag will cause a color change. A color change indicates a positive reaction.</p>

ADVANCE PREPARATION FOR TEACHER:

Pipetting by Design –

1. Reproduce the protocols – one for each student or group.
2. Prepare tubes or beakers of colored water appropriate for the protocols you will be using:

Protocol A: 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,

- 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: 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 μL : D1, D4, D8
88 μL : E2, E3, E4, E8
129 μL : A2, A3, A4, A6, A7, A8, A9, A10
107 μL : C1, C3, C4, C8

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

Using the **GREEN** dye:

20 μL : A1, A9, A10, A11
19.5 μL : B2, B3, B8, B10, B11, B12
18.2 μL : D9, D10, D11, D12
17.7 μL : D4, D5, D6, D7, D8
89 μL : D10, D11, D12, B2, B3
95 μL : B8, B10, B11, B12
100 μL : H3, H4, H10, H11
111 μL : A1, A9, A10, A11, C3, C4, C11, C12
120 μL : C3, C4, C11, C12
135 μL : D4, D5, D6, D7, D8, D9
15.7 μL : H3, H4, H10, H11
13 μL : G5, G11
12 μL : F5, F6, F7, F9, F10, F11, A2, C5
11 μL : G5, G11
140 μL : D4, D5, D6, D7, D8, D9, D10, D11, D12
160 μL : H3, H4, H10, H11,
180 μL : G5, G11
177 μL : F5, F6, F7, F9, F10, F11
188 μL : E1, E2, E3, E5, E6
190 μL : E7, E8, E9, E10, E11

Using the **BROWN** dye:

200 μL Brown: A2, C5

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

Using the **ORANGE** dye:

20 μL : B2, C2
19 μL : B6, C6
18.6 μL : B8, C8
17.3 μL : F2, F6, F8
15.9 μL : G3, G4, G5, G8

Using the **BLUE** dye:

13 μL : D2, E2
11.5 μL : D6, E6, E8
10 μL : D8, D9, D10

Using the **ORANGE** dye:

179 μL : B9, B10, B11
164 μL : B8, C8
159 μL : B6, C6
143 μL : B2, C2
133 μL : F2, F6, F8

127 μ L: G3, G4, G5, G8

Using the **BLUE** dye:

111 μ L: D2, E2

100 μ L: D6, E6, E8

120 μ L: D8, D9, D10

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

Using the **RED** dye,

20 μ L: A12

16 μ L: C3, C4

17 μ L: D2, D3, D4, D5, D12

18 μ L: E3, E4

19 μ L: G12

Using the **PURPLE** dye,

8 μ L: A10, A11, B3, B4, B9

6 μ L: D1, D6, D7, D8, D9, D10, D11

8 μ L: E2, E5, E9

9 μ L: F3, F4, F9

7 μ L: C2, C5, C9, G10, G11

Using the **RED** dye,

70 μ L: A12

116 μ L: C3, C4

110 μ L: D2, D3, D4, D5, D12

93 μ L: E3, E4

85 μ L: G12

Using the **PURPLE** dye,

118 μ L: A10, A11, B3, B4, B9

133 μ L: D1, D6, D7, D8, D9, D10, D11

122 μ L: E2, E5, E9

129 μ L: F3, F4, F9

141 μ L: C2, C5, C9, G10, G11

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

Using the **RED** dye,

20 μ L: A2, A3, E4

14 μ L: B2, B4, C2, C5, D3

15 μ L: G9, F8, H7, H8

Using the **ORANGE** dye,

20 μ L: A8, A9, A10

18 μ L: B7, B10, C10, D4, D9

17 μ L: E3, E8, F3, F7

19 μ L: G3, G6, H4, H5

Using the **BLUE** dye,

18 μ L: C6, D5, D6, D7

19 μ L: 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 G: Micropipette indicated amounts into designated wells on the 96 well plate!

Using the **BLUE** dye,

17 μL : A7, A8, A9, C5, C9

19 μL : B6, B9, E2, E9

21 μL : D2, D3, D4, D9

18 μL : F2, F3, F4, F9, G5, G9

15 μL : H6, H7, H8, H9

Using the **ORANGE** dye, 1

8 μL : B7, B8, D7, D8

14 μL : C7, C8, E7, E8

16 μL : F7, F8, G7, G8

Using the **RED** dye,

19 μL : C11, D10, D11, D12

20 μL : E10, E11, E12, F11

Using the **BLUE** dye,

89 μL : A7, A8, A9, C5, C9

72 μL : B6, B9, E2, E9

84 μL : D2, D3, D4, D9

91 μL : F2, F3, F4, F9, G5, G9

105 μL : H6, H7, H8, H9

Using the **ORANGE** dye,

108 μL : B7, B8, D7, D8

104 μL : C7, C8, E7, E8

79 μL : F7, F8, G7, G8

Using the **RED** dye,

95 μL : C11, D10, D11, D12

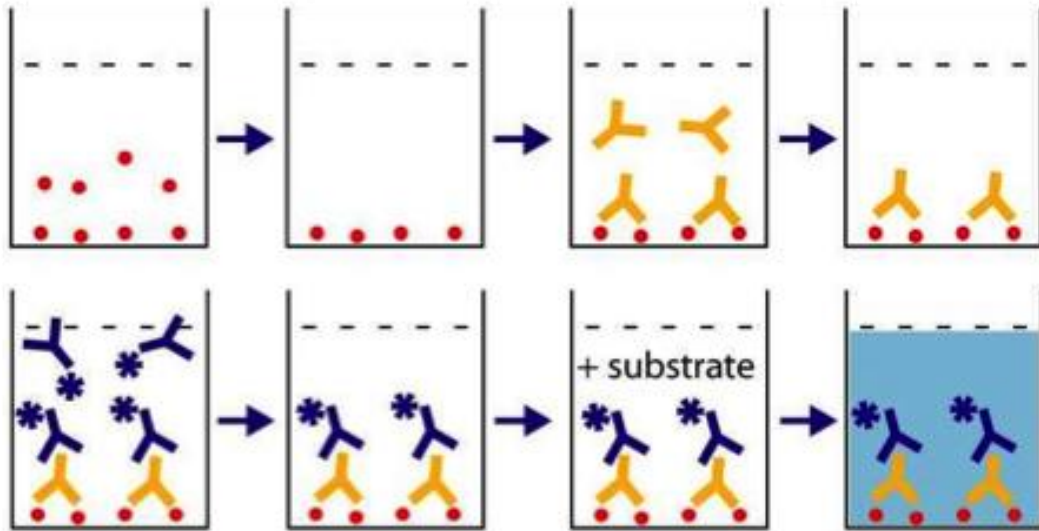
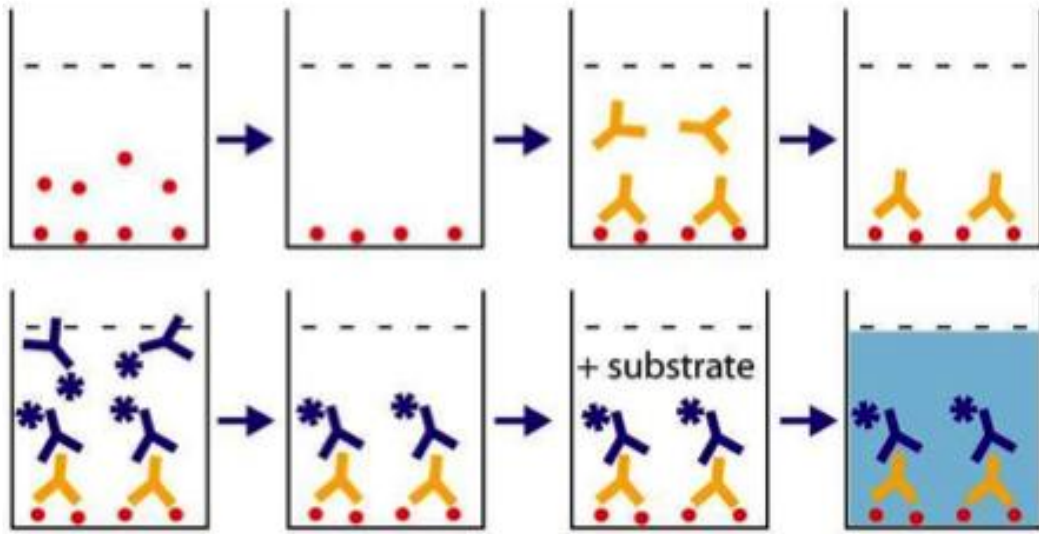
100 μL : E10, E11, E12, F11

ELISA Sequencing Activity:

1. Prepare sequencing activity by cutting out and laminating (optional) the various pieces on

the following page.

Make one set of eight cards for each group. Cut on both sides of arrows and between rows to separate into eight cards, each representing a step in the ELISA reaction.



Additional References and Resources:

University of Florida ICORE, June, 2013.

Bokor, Julie. *Detecting dengue in the lab and field.*

SUGGESTED ANSWERS TO VOCABULARY:

Micropipette - a very slender pipette for transferring or measuring minute amounts of fluid, microorganisms.

Substrate - a molecule upon which an enzyme acts.

Microliter - a unit of volume equal to one millionth of a liter

Antigen - A substance that when introduced into the body stimulates the production of an antibody.

Antibody - A Y-shaped protein that is secreted into the blood in response to an antigen, and neutralizes the antigen by binding specifically to it.

ASSESSMENT SUGGESTIONS:

To assess preparation and readiness	Pre laboratory questioning
To assess skills acquisition	Laboratory participation and success
To assess understanding and communication of relevant concepts	Successful completion of sequencing activity
To assess overall understanding and application to the larger topic	Questioning and contribution to discussion Evaluate answer to laboratory questions

Title:

May the Odds Be Ever in Our Favor!

**LESSON PLAN DAY 3
DETECTION OF GENETICALLY MODIFIED
FOOD LAB**



TARGET GRADE LEVEL: First year biology students (grades 9 or 10)

KEY QUESTIONS: Can you determine if food product contains GMO?
Which varieties of papaya contain GMO?

OVERALL TIME ESTIMATE: 45 minutes

LEARNING STYLES: Visual and kinesthetic

VOCABULARY:

ELISA
Antigen
Antibody
Substrate
Primary antibody
Secondary antibody
T ransgene

LESSON SUMMARY: Using an ELISA simulation kit, students will test papaya samples from three sources -US (Hawaii grown –Rainbow), US (Hawaiian grown – organic), Non American grown, for the presence of GMO.

STUDENT LEARNING OBJECTIVES:

The student will:

- Perform an ELISA simulation test
- Know (Recognize) that an ELISA is an antibody-based test
- Explain the steps of an ELISA
- Consider (Propose) other uses of an ELISA

OUTCOMES AND STANDARDS:

STUDENT OUTCOMES:

The student will be able to...

- Outline the basic process of GMO production. (LS.16)*
- Accurately pipette specified measured amounts into appropriate wells on a well plate and micro tube. (GSI.5)*
- Successfully interpret the presence or absence of GMO genes in a food source using ELISA results. (GSI.2, GSI.7)*
- Relate the antigen-antibody basis of the ELISA procedure. (GSI.8, LS.16)*

STANDARDS*:

- GSI.2. Perform systematic observations.
- GSI.5. Use tools to gather, analyze, and interpret data.
- GSI.7. Use appropriate evidence and reasoning to justify explanations to others.
- GSI.8. Communicate the results of scientific investigations..
- LS.16. Explain the processes involved in genetic engineering and discuss its impact on human life and society.

*These standards refer to Jesuit high School Science Standards

MATERIALS:

Fluorescent ink pen
96-well plate
Assorted 1.5 or 2.0ml microfuge tubes
Microfuge racks
Disposable transfer pipettes
P200 micropipette
Disposable tips, 20-200ul
Clear or white unscented soap
Food coloring
Small beakers
UV lights

BACKGROUND INFORMATION FOR TEACHERS:

Several companies in the United States and Europe test foods for genetic modification. Methods vary among the different companies and different countries. One method tests food for the presence of the product of the transgene, a protein. Proteins are assayed (determine the biochemical or immunological activity) using an ELISA (Enzyme Linked ImmunoSorbent Assay). In the ELISA test, an enzyme is linked to an antibody bound to

the transgene-produced protein, which then reacts with a colored substrate enabling detection of the specific protein. ELISA tests are usually relatively low cost, offer quick results, and can sometimes be done on site. The big drawback is that ELISA tests do not work well on processed foods since heating during processing may destroy the protein.

In this lab we will use an ELISA simulation to determine whether the foods we are testing contain the protein produced by the transgene. Remember, the transgene expresses a particular protein (antigen). We will first coat the wells of the plate with a specific antibody that will react with that antigen. We will then add a secondary antibody that is tagged with a gene that expresses fluorescence. After these antibodies have bound to their targets, an enzyme substrate is added. This substrate reacts with the enzyme producing a color change. Using a UV light, we will see fluorescence in the samples that contain the target (GMO) protein. (Remember we are simulating each of these reactions)

ADVANCE PREPARATION:

1. Prepare the ELISA plates. If using 12-well microplate strips, use a Sharpie or other permanent marker to number the wells at the top 1-12. If using 96-well plates, they should come with columns and rows marked.
2. Using a fluorescent ink pen, “paint” the bottom of wells 1-3 (positive serum) and wells 8 and 9 (contain GMO) on rows 1-3. Allow the ink to dry prior to use.
3. Prepare student station reagents using the chart below. Note: This provides quantities for 8 student workstations, each with 2-4 students.

Tubes (number needed)	Description	Label	Contents (Each Tube)
Violet tubes, 8	Positive controls	+	0.5ml water
Blue tubes, 8	Negative controls	-	0.5ml water
Green tubes, 8	Primary antibody	PA	1.5ml water
Orange tubes, 8	Secondary antibody	SA	1.5ml water
Brown tubes, 8	Enzyme substrate	SUB	1.5ml water
Clear tubes, 8	Papaya antigen US grown	PUS	0.25ml water
Clear tubes, 8	Papaya antigen US organically grown	PUSO	0.25ml water
Clear tubes 8	Papaya antigen non US grown	NPUS	0.25 ml water

4. Prepare wash buffer
 - Add 5ml clear or white unscented dish soap to 1000ml water. Mix well.
 - Allow 50ml wash buffer per student group.
5. Assemble student workstations, or have students collect the items below from a common station.

Item (Label)	Contents	Number per station
Yellow tube (PUS)	(0.25ml) Papaya US grown – antigen	1
Yellow tube (PUSO)	(0.25ml) Papaya US grown organic – antigen	1
Yellow tube (NPUS)	(0.25ml) Papaya non US grown – antigen	1
Violet tube (+)	Positive control (0.5ml)	1
Blue tube (-)	Negative control (0.5ml)	1
Green tube (PA)	Primary antibody (1.5ml)	1
Orange tube (SA)	Secondary antibody (1.5ml)	1
Brown tube (SUB)	Enzyme substrate (1.5ml)	1
Beaker of wash buffer		1
96-well microplate		1
Disposable transfer pipette		1
20-200ul micropipette		1
20-200ul tips		1 box
Stack of paper towels		1

SUGGESTED ANSWERS TO PRELAB VOCABULARY:

Assay - a process used to determine biochemical or immunological activity

ELISA - Enzyme Linked ImmunoSorbent Assay

Antigen - protein that stimulates the production of an antibody

Antibody- protein that is secreted in response to a foreign protein antigen and neutralizes it by binding specifically to it)

Substrate - a molecule upon which an enzyme acts

Primary antibody - antibody against an antigen target of interest

Secondary antibody - antibody that binds to primary antibodies.)

Transgene - an organism that has genes from another organism put into its DNA

UV light - light that emits radiation lying in the ultraviolet range

Florescence – glow that occurs when exposed to a UV light

ANSWERS TO THE POST LAB QUESTIONS:

1. Our testing goal was to determine whether each food sample contained GMO.
2. The ELISA test uses the antigen-antibody reaction to test for the presence (in this case) of a protein antigen produced by the GMO gene.
3. Yes the test showed clear results that the US grown papaya (PUS) contained GMO while the organic US grown papaya (PUSO) and the non US grown papaya (NPUS) did not.
4. The positive control and the US grown papaya (PUS) fluoresced, while the organic US grown papaya (PUSO) and the non US grown papaya (NPUS) did not.
5. ELISA can be used to test for anything that enters into an antigen-antibody reaction. Some possibilities are pathogens that cause dengue, HIV, Hepatitis B or C, toxoplasmosis, Lyme disease, mumps, rubella as well as for pregnancy tests, food allergies, Lupus, rheumatoid arthritis, and drug tests for cocaine and methamphetamines, to name some.
6. Yes, I expected Hawaiian (US grown papaya) to contain GMO since a large percentage of the Hawaiian papaya are GMO and I expected the organic and non US grown papaya would not contain GMO since organic foods are not supposed to contain GMO and most other countries are not using GMO technology as widely as the US is using it.

ASSESSMENT SUGGESTIONS:

To assess preparation and readiness

The student pre-laboratory activities must be completed BEFORE the student is allowed to participate in the lab. This work is collected on the day the procedure is to begin.

To assess skills and procedures

Teacher observations are made and recorded during laboratory activities

To assess understanding and communication of results

Laboratory questions and/or lab reports are prepared and collected.

To assess overall understanding and application to the larger topic.

Laboratory questions are included on unit test.

REFERENCES/RESOURCES:

University of Florida ICORE. June, 2013.

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Title:

**May the Odds Be Ever in Our Favor!
Detection of Genetically Modified Foods Lab**

STUDENT LABORATORY INSTRUCTIONS



Background Information

Several companies in the United States and Europe test foods for genetic modification. Methods vary among the different companies and countries. One method tests food for the product of the **transgene** (an organism that has genes from another organism put into its DNA), a protein. Proteins are **assayed** (a process used to determine the biochemical or immunological activity) using an **ELISA** (Enzyme Linked ImmunoSorbent Assay). In the test an enzyme is linked to an **antibody** (protein that is secreted in response to an foreign protein antigen and neutralizes it by binding specifically to it) bound to the protein **antigen** - (protein that stimulates the production of an antibody), which then reacts with a substrate enabling detection of the specific protein (antigen). ELISA tests are usually relatively low cost, offer quick results, and can sometimes be done on site. The big drawback is that ELISA tests do not work well on processed foods because heating during processing may destroy the protein.

In this lab we will use an ELISA procedure to determine whether the foods we are testing contain the protein produced by the transgene. Remember, the transgene expresses a particular protein (antigen). We will first coat the wells of the plate with a specific **primary antibody** (antibodies against an antigen target of interest) that will react with that antigen. We will then add a **secondary antibody** (antibody that binds to primary antibodies.) that is tagged with a gene that expresses florescence (glow). After these antibodies have bound to their targets, an enzyme **substrate** (a molecule upon which an enzyme acts) is added. This substrate reacts with the enzyme producing a color change. Using a **UV light** (light that emits radiation lying in the ultraviolet range) look for **fluorescence** (glowing) in the samples that contain the target (GMO) protein.

Pre-laboratory Preparation

1. Read the background information,
2. After reading the introduction material, define each of the following vocabulary from the context of the reading.

Assay

ELISA

Antigen

Antibody

Substrate

Primary antibody

Secondary antibody

Transgene

UV light

Florescence

3. Read the procedure carefully and be prepared to perform it flawlessly.

Laboratory activity

Purpose

Using the ELISA procedure, answer the following questions:

1. Can you determine if food product contains GMO?
2. Which varieties of papaya contain GMO?

Materials

Item (Label)	Contents	Number per station
Yellow tube (PUS)	(0.25ml) Papaya US grown – antigen	1
Yellow tube (PUSO)	(0.25ml) Papaya US grown organic – antigen	1
Yellow tube (NPUS)	(0.25ml) Papaya non US grown – antigen	1
Violet tube (+)	Positive control (0.5ml)	1

Blue tube (-)	Negative control (0.5ml)	1
Green tube (AG)	Primary antibody (1.5ml)	1
Orange tube (SA)	Secondary antibody (1.5ml)	1
Brown tube (SUB)	Enzyme substrate (1.5ml)	1
Beaker of wash buffer		1
96-well microplate		1
Disposable transfer pipette		1
20-200ul micropipette		1
20-200ul tips		1 box
Stack of paper towels		1

Procedure

1. Orient your plate like the diagram below:

	1	2	3	4	5	6	7	8	9	10	11	12
A												
B												
C												
D												
E												
F												
G												
H												

Now label the plate as follows (Label rows A,B, and C):

Positions A1, A2, and A3	Label with a +
B1, B2, and B3	Label with a +
C1, C2, and C3	Label with a +
Positions A4, A5, and A6	Label with a -
B4, B5, and B6	Label with a -
C4, C5, and C6	Label with a -
Positions A7, A8, and A9	Label with a PUS
B7, B8, and B9	Label with a PUSO
C7, C8, and C9	Label with a NPUS
Leave ALL other positions	UNLABELED

Does your plate look like this?

	1	2	3	4	5	6	7	8	9	10	11	12
A	+	+	+	-	-	-	PUS	PUS	PUS			
B	+	+	+	-	-	-	PUSO	PUSO	PUSO			
C	+	+	+	-	-	-	NPUS	NPUS	NPUS			
D												

2. With a fresh pipette tip, pipette 50ul of the primary antibody (PA) into wells 1 – 9 of row A.
3. Wait 5 minutes for the primary antibody to bind to the plastic wells.
4. Tip the plate upside down onto a paper towel and tap the plate a couple of times. Avoid splashing the liquid into neighboring wells. Discard the paper towel.
5. Wash:
 - a. Use a plastic pipette to fill each well (1-9 in rows A, B, and C) with wash buffer. Be careful not to overflow into neighboring wells. Save that pipette for use in each washing step.
 - b. Tip the plate upside down onto a paper towel and tap the strip a couple of times. Avoid splashing the liquid into neighboring wells.
 - c. Discard paper towel
6. Repeat wash step 5. (That means you are washing **TWO TIMES**)
7. Use a fresh pipette tip to transfer 50ul of the positive control (+) into wells 1-3 in rows A, B, and C.
8. Use a fresh pipette tip to transfer 50ul of the negative control (-) into wells 4-6 in rows A, B, and C.
9. Use a fresh pipette tip to transfer 50ul of the papaya antigen (PUS) into well 7, 8, and 9 in row A.
10. Use a fresh pipette tip to transfer 50ul of the papaya antigen (PUS0) into well 7, 8, and 9 in row B.
11. Use a fresh pipette tip to transfer 50ul of the papaya antigen (NPUS) into well 7, 8, and 9 in row C.
12. Leave wells 10-12 empty in all rows.
13. Wait 5 minutes for the antigens to bind to the antibodies.

14. Wash the unbound antigens out of the wells by repeating all of wash step 5 two times. (Wash **TWO TIMES**)

15. Use a fresh pipette tip to transfer 50ul of secondary antibody (SA) into wells 1-9, in rows A, B, and C, of the plate.

16. Wait 5 minutes for the antibodies to bind to their targets.

17. Wash the unbound secondary antibody out of the wells by repeating wash step 5 three times. (Wash **THREE TIMES**)

18. Use a fresh pipette tip to transfer 50ul of enzyme substrate (SUB) into wells 1-9 in rows A, B, and C, of the plate.

19. Wait 5 minutes to allow the enzyme-substrate reaction to occur.

20. Using a UV light look for florescence in each of the wells. Record your results in the data table.

Data Table

WELL NUMBER	1	2	3	4	5	6	7	8	9
RESULTS - ROW A Presence or absence of florescence (+ or -)									
RESULTS - ROW B Presence or absence of florescence (+ or -)									
RESULTS - ROW C Presence or absence of florescence (+ or -)									

Questions

1. What were our testing goals the ELISA procedure?
2. What does the ELISA test actually test for?

3. Were the results of your ELISA test adequate to answer your two key questions?
4. How do you know whether the ELISA test was adequate to answer your two key questions?
5. For what other things do you think you could test for using an ELISA procedure?
6. Did your results turn out as you expected? Explain fully.

Title:

**May the Odds Be Ever in Our Favor!
What do you think and why do you think it?**

LESSON PLAN DAY 4



TARGET GRADE LEVEL: First year biology students (grades 9 or 10)

KEY QUESTIONS:

1. What is your fully supported opinion about GMO?

OVERALL TIME ESTIMATE: 45 minutes

LEARNING STYLES: Visual

VOCABULARY: no new vocabulary

LESSON SUMMARY: Using the information they have received in class, in the laboratory and in outside reading, each student will develop a fully supported opinion. This will be submitted in the form of a persuasive letter to a newspaper editorial page or a letter to a senator or congressperson.

STUDENT LEARNING OBJECTIVES:

The student will:

Develop and communicate a fully supported opinion concerning the various aspects of GMO. This will be submitted in the form of a persuasive letter to a newspaper editorial page or a letter to a senator or congressperson. All letters will be written independently.

STUDENT OUTCOMES: STUDENT OUTCOMES:

The student will:

- Describe the structure and function of DNA and relate that structure and function to the process of producing GMO. (content)
- Outline the basic process of GMO production. (content)
- Relate the antigen-antibody basis of the ELISA procedure. (background)
- Describe the outcomes of GMO and their effect on agriculture. (application)
- Discover the positive and negative outcomes of GMO that are concerns to society. (extension)

- Articulate a fully supported position on the issue of GMO. (extension)

STANDARDS*:

- GSI.7. Use appropriate evidence and reasoning to justify explanations to others.
- GSI.8. Communicate the results of scientific investigations.
- GSI.9. Evaluate the merits of explanations produced by others.
- GSI.12. Recognize that the strength or usefulness of a scientific claim is established through logical argumentation and includes active consideration of alternative scientific explanations.
- GSS.1. Identify ways in which science influences society and is influenced by society.
- GSS.2. Identify sources of information and assess their reliability.
- GSS.3. Weigh the merits of alternative strategies by comparing a number different costs and benefits (human, economic, environmental).
- GSS.5. Discuss the relationship between faith, science, and reason and explain how the principles and patterns of nature can be indicative of a higher power with purpose.
- LS.16. Explain the processes involved in genetic engineering and discuss its impact on human life and society.
- LS.26. Recognize the positive and negative anthropogenic influences on the biosphere.

*These standards refer to Jesuit High School, Tampa Florida, Science Standards

MATERIALS:

Paper and pencil

Computer with word processing software (optional)

BACKGROUND INFORMATION FOR TEACHERS:

1. Spend the first few minutes of class leading a discussion that helps the student recall the facts previously discussed.
2. Describe the assignment (handout)
3. Allow students to complete in class.

ADVANCE PREPARATION:

1. Prepare talking points appropriate for each individual class.
2. Produce handout – one for each student.
3. Students should not collaborate.

Title:

May the Odds Be Ever in Our Favor!

**What do you think and why do you think it?
STUDENT HANDOUT**



Now it is time for you to tell me what you think about GMO. Your assignment for today is to write a fully supported persuasion letter to the newspaper editorial page or to one of your senators or representatives. This letter should include AT LEAST the following:

1. An introduction that illustrates to the reader that you know something about what you are writing about.
2. Some reference to the historical perspective that applies to this topic. Use examples.
3. Acknowledgement of the positive aspects of GMO, whether or not you are in favor of its use. Use examples.
4. Acknowledgement of the negative aspects of GMO, whether or not you are in favor of its use. Use examples
5. Your opinion concerning
 - a. Its use at all – why or why not?
 - b. Safety issues– why or why not?
 - c. Copywriting issues– why or why not?
 - d. Labeling issues– why or why not?
 - e. Ethical/religious/moral issues.

Get busy – this is due at the end of class.

ASSESSMENT SUGGESTIONS:

To assess understanding and communication of results and to assess overall understanding-

Carefully read and comment on each letter.

REFERENCES/RESOURCES:

University of Florida ICORE. June, 2013.

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**May the Odds Be Ever in
Our Favor!**



**What about Genetically Modified
Organisms?**

In the popular *Hunger Games* trilogy, a (fictional) bird, called the mockingjay is produced when the Capitol (the government) releases all male jabberjays into the wild, expecting them to die out.

Jabberjays were genetically enhanced birds that the Capitol used to spy on the people. Instead of dying out as expected, the jabberjays bred with mockingbirds and the offspring were hybrids called mockingjays. Their hybrid traits were eventually used against the Capitol.

This bird came to symbolize the revolution that occurs in that series of books because of its resistance, resilience, and persistence.

From the beginning of time, man has altered his environment for his own benefit.



Beginning with early agricultural practices, the land produced and mankind benefited.



When man got greedy, or careless, the
land responded by no longer
producing.



Image – United States “dust bowl” in the 1930s

As man continues to overpopulate, we see the reemergence of pathogens that were once under better control.



Image – cholera in Haiti



As food- handling mega-corporations handle tons of products, mass contamination events are on the rise.

image – industrial ground beef is often the source of widespread e.coli contamination

While we continue to deforest and incur into formerly uninhabited areas, previously unknown pathogens emerge.

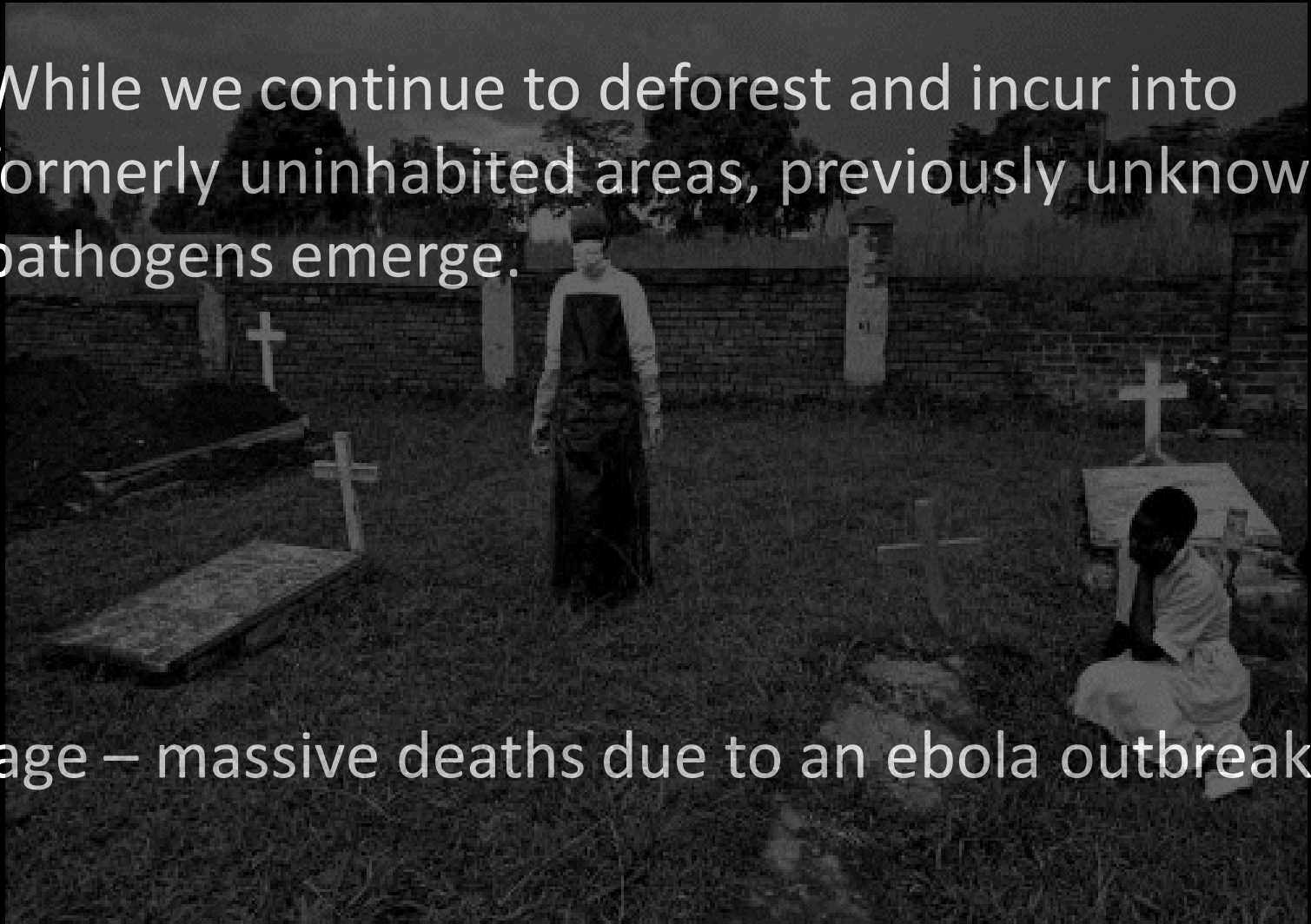


Image – massive deaths due to an ebola outbreak

What lessons can be learned about genetically engineering organisms from history and from the story of the mockingjay?



What are the risks and benefits of research and development of GMOs – genetically modified organisms?

Could GMOs created to increase crop yields or improve an organism's resistance to disease eventually hybridize with a wild animal or plant?



Can this happen in the real world?



Do we know for sure?



What we do know for sure is that GMOs are already here. And they got here using a relatively simple process.



The following video will explain the process:

THE SCIENCE OF GENETICALLY MODIFIED FOOD

<http://www.youtube.com/watch?v=vUzVmzpyR8>

**May the Odds Be Ever in
Our Favor!**



**Case Study : Hawaiian Papaya
Industry**

Now that you know a little bit about the process used to make GMOs, consider the following problem:

Papaya is a major crop of the state of Hawaii.



A specific virus, the papaya ringspot virus (PRSV), infects papaya as well as some members of the melon family.



It is transmitted between plants by pruning, but mostly by an insect vector of one of numerous aphid species.



Around 1950, the PRSV was introduced into Hawaii and within a decade, production of papaya had dropped over 90%.

Symptoms are typical of viral diseases. Papaya exhibits yellowing, leaf distortion, and severe mosaic.



The fruit will exhibit bumps and the classic "ringspot".



By 1995, despite efforts to contain the virus, commercial production was impossible on some islands and severely limited on the others.

In the late 1980s, the University of Hawaii developed a papaya resistant to PRSV.



Certain viral genes were transferred into the papaya genome, creating an “immune-like response” from the papaya plant.



These new plants are no longer susceptible to PRSV.

The first virus resistant or “Rainbow” papayas were grown in Hawaii in 1999.



Transgenic papayas now make up 75% of the papaya crop in Hawaii.

These papayas are approved for consumption in the US and Canada, and were very recently approved for shipment into Japan.



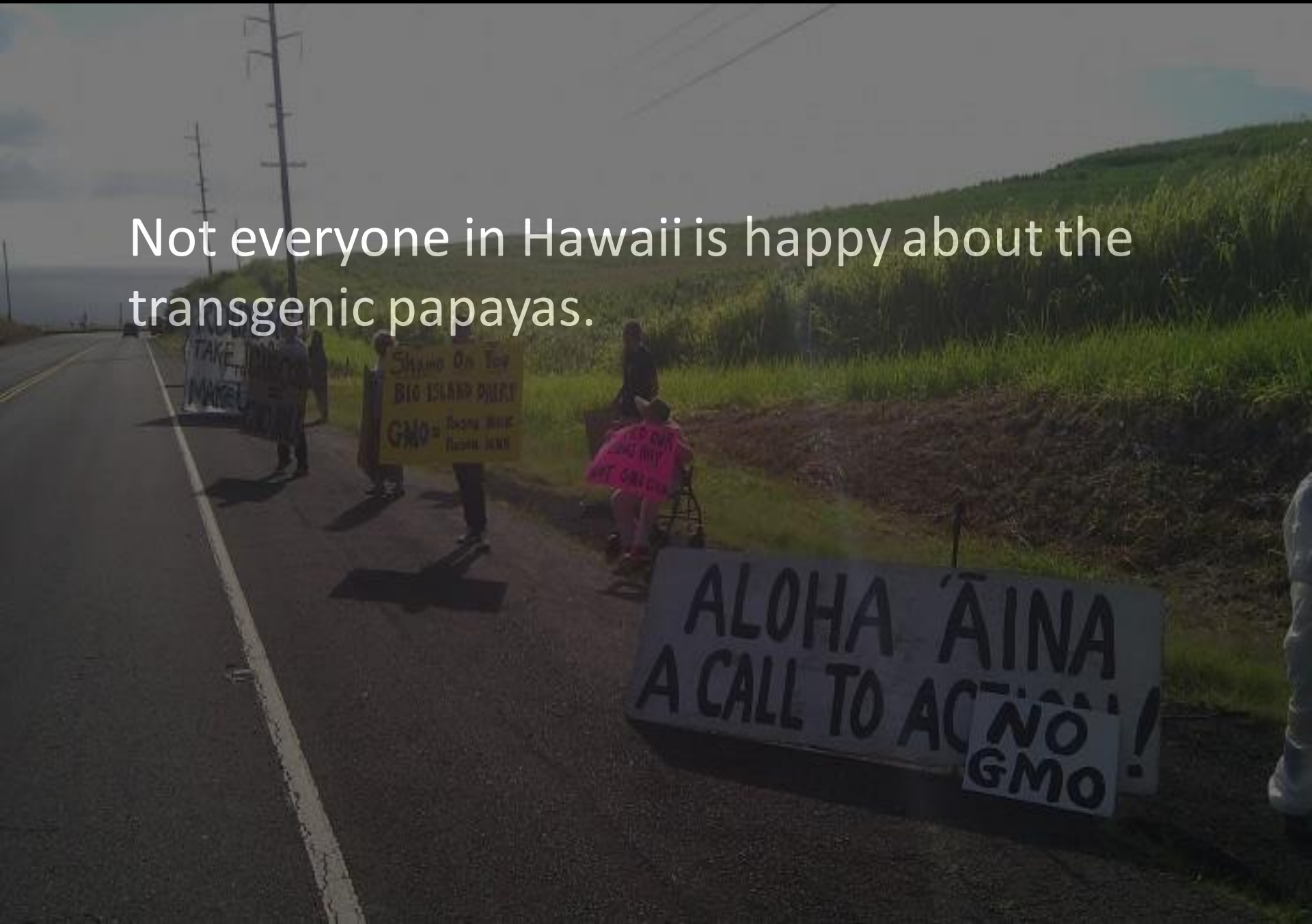
They are the first genetically modified (GM) food approved in Japan.

This is a story of how cutting edge agriculture saved a major Hawaiian crop industry and many livelihoods in that state.



Papaya is a local industry worth \$11 million annually.

Not everyone in Hawaii is happy about the transgenic papayas.



Concern about safety has created a significant backlash.




Terrorist opponents have destroyed papaya plantations under the cover of darkness.

Farmers have lost tens of thousands of dollars worth of trees in these attacks.



The Vatican has even weighed in on the topic of GMOs.



A man in a dark suit is seated at a table, gesturing with his right hand. The background is slightly blurred, showing a blue flag with yellow stars and a white wall. The text is overlaid on the image in white font.

In a statement released at the end of November 2010, forty international scientists including seven Vatican advisors have called for the relaxation of “excessive, unscientific regulations” applied to genetically modified crops.

The scientists cited the “magnitude of challenges facing the world’s poor and undernourished” as a “matter of urgency” and the making of the benefits of GE available to poor and vulnerable populations a “moral imperative”.



What if you don't even like papaya and no one you know is economically affected by the papaya industry? So what?



Well, how do you feel about orange juice?



Whether you know it or not, all of the residents of Florida are economically impacted by the citrus industry.



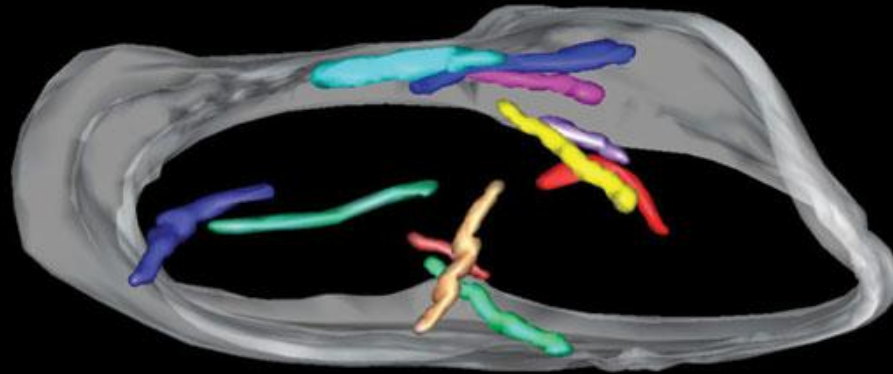
What if something like that happened to citrus?

A photograph of an elderly man with a white beard and a camouflage cap, wearing a blue and white checkered shirt. He is holding a citrus fruit in his hands. The background is a dense orchard of citrus trees with green leaves and some yellow fruit. The image has a dark, semi-transparent overlay.

Well it has.

It is called citrus greening (Huanglongbing or HLB), and it is working on wiping out the citrus industry in Florida.

Like PRSV, HLB is a vector borne pathogen (bacterial instead of viral) that was first noticed in Florida in 2005.




Eight years later, it can be found in every citrus-producing county in Florida.

The fruit from infected trees has an unacceptable flavor, and the virus is eventually fatal to the tree.




The vector is an Asian psyllid that can easily move from tree to tree and from grove to grove.





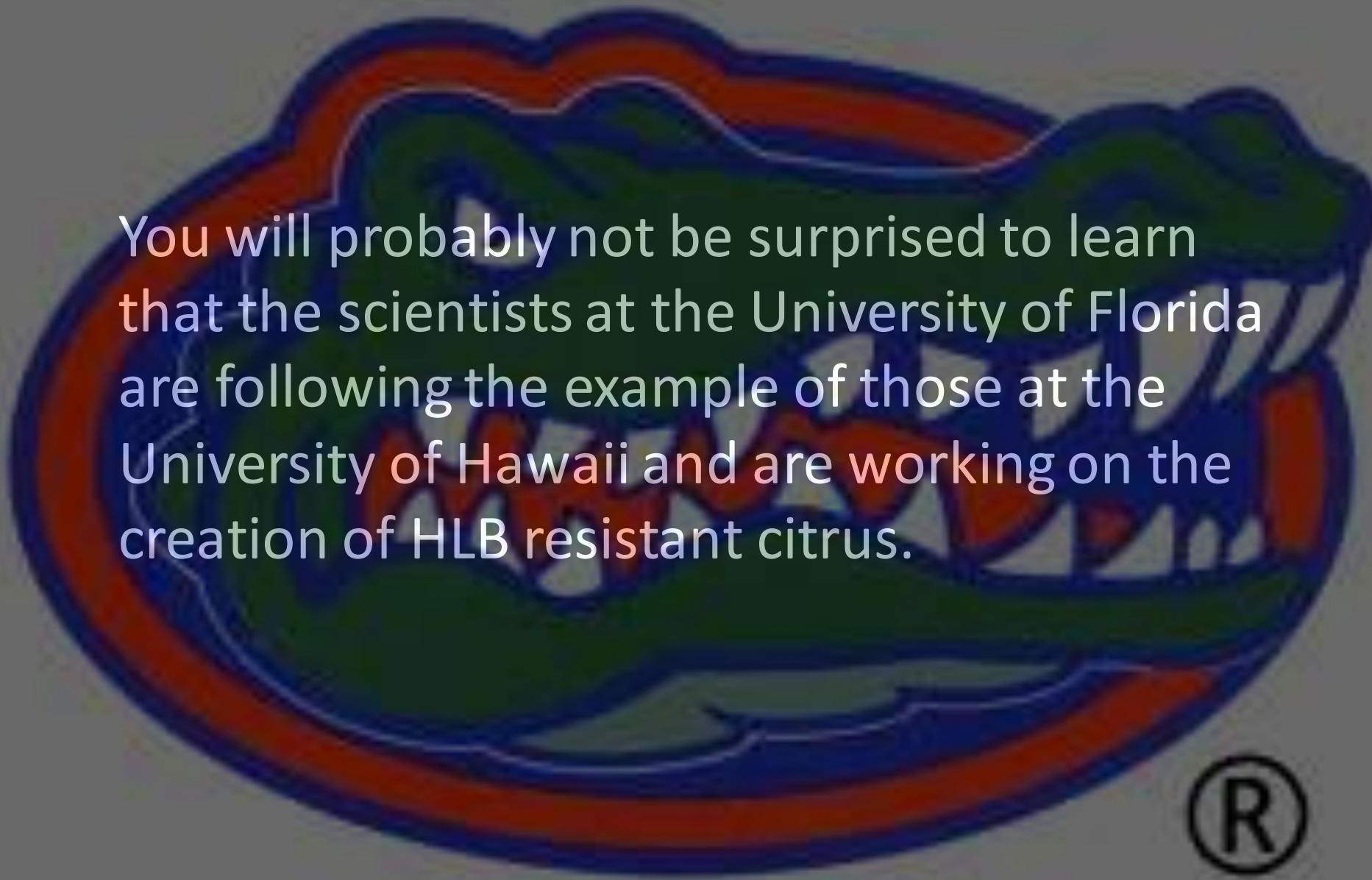
According to the Florida Department of Citrus, the industry employs approximately 76,000 workers and has an annual economic impact of 9 billion dollars.



The United States leads the world in grapefruit production supplying the world with over 30% of its grapefruit, and is the third largest overall citrus producer in the world.

The background of the slide features a close-up, slightly blurred image of several oranges and orange slices. The oranges are a vibrant orange color, and the slices show the internal segments and white pith. The lighting is warm, creating a soft glow around the fruit. The text is overlaid on this background in a light yellow, sans-serif font.

In fact, Florida produces three times as many tons of oranges and four times as many tons of grapefruit as its closest competitor, California. With over 1/2 million acres of citrus groves and 74 million trees, Florida is second only to Brazil in orange juice production and supplies approximately 80% of the orange juice in the United States during any given growing season.

The image features the University of Florida Gator logo, which is a stylized orange and blue outline of a gator's head. The gator's mouth is open, showing its teeth and tongue. The logo is set against a dark gray background. A registered trademark symbol (®) is located in the bottom right corner of the logo.

You will probably not be surprised to learn that the scientists at the University of Florida are following the example of those at the University of Hawaii and are working on the creation of HLB resistant citrus.

What do you
think?