Cloning Around – Champion Trees

Secrets of the Sequence Video Series on the Life Sciences • Grades 9 – 12

Teaching materials developed by VCU Life Sciences.

Virginia Commonwealth University

Classroom Tested Lesson

Video Description

"Secrets of the Sequence," Show 109, Episode 3
"Cloning Around" – approximately 7 minutes viewing time

Dave Milarch doesn't know if the oldest and largest trees in America have a genetic leg up – but their robust longevity is reason enough for him. He is grafting/cloning the biggest and oldest trees from every species in America so that when someone finally gets interested, their genes will still be around.

Ward Television
Producer: Kip Prestholdt
Featuring: David Milarch, Founder and President of Champion Tree Project, Jared Milarch, Co-Founder of Champion Tree Project, Francis Gouin, Natural Resource Sciences and Landscape Architecture at University of Maryland
Lesson Author; Reviewers: Ryan Templeton; Marilyn Elder, Cathie Alder, Sue Kirk, Catherine Dahl, Dick Rezba, and Kieron Torres
Trial Testing Teachers: Kate Henson, Brooke Williams

National and State Science Standards of Learning

National Science Education Standards Connection

Content Standard C: Life Science

As a result of their activities in grades 9-12, all students should develop understanding of:

- molecular basis of heredity

Content Standard F: Science in Personal and Social Perspectives

As a result of activities in grades 9-12, all students should develop understanding of:

- natural resources and
- science and technology in local, national, and global challenges

Selected State Science Standards Connections

Use http://www.eduhound.com (click on "Standards by State") or a search engine to access additional state science standards.

Virginia

BIO.6 The student will investigate and understand common mechanisms of inheritance and protein synthesis. Key concepts include:

- cell growth and division
- exploration of the impact of DNA technologies

BIO.8 The student will investigate and understand how populations change through time. Key concepts include:

- how genetic variation, reproductive strategies, and environmental pressures impact the survival of populations.

Arizona

4SC-P2 Describe the molecular basis of heredity

- PO 4. Describe how genetic information is transmitted from parents to offspring.

Overview

This episode demonstrates how cloning can be used to preserve great champion trees of North America, such as the featured Sycamore. The Champion Tree Project seeks to obtain the complete genomes of approximately 860 champion trees in North America that have lived to be 400 to 500 years old. A champion tree is the largest known tree of its species as determined by combining measurements of its trunk circumference, its height, and its crown spread. The genomes are collected by cutting living buds from the highest parts of the trees that contain the newest growth. It is thought that these giant, long-lived trees possess genetic qualities that predispose them to be so successful at living through pestilence, changing climate, and pollution. Once branches are obtained, they are grafted onto the root stock of younger plants of the same species and allowed to grow, a cloning process that is itself over 2,000 years old. Grafting and other types of asexual plant propagation may be the best way to maintain some species. Clones, plants that are identical to the parent tree, can only be propagated asexually. The Bartlett pear (1770) and the Delicious apple (1870) are two examples of clones that have been asexually propagated for many years. The major methods of asexual propagation are cuttings, layering, division, and budding/grafting. Cuttings involve rooting a cut piece of the parent plant; layering involves rooting a part of the parent and then cutting it; and budding and grafting are joining two plant parts from different plants. The DNA from these cloned plants can be studied and analyzed as well as preserved through continuous cloning for many years.

Testing: A sample related multiple choice item from State Standardized Exams

Which of the following is an example of a genetically engineered organism?

a. a plant that received external DNA to produce natural insecticides *
b. a new plant variety created by cross-pollination
c. seedless fruits resulting from grafting of one plant onto another
d. a plant that naturally possesses medicinal properties

(Source: Virginia Standards of Learning Assessments, Spring 2001 Biology Released Test)

Before Viewing

Ask several of the questions below and record student responses on the board:

1. "What would a tree need to survive the following stressors?"
    - Floods? *extensive root system, soil that allows the roots to grow down, available nutrients, general health*
    - Severe weather storms, such as tornadoes and hurricanes? *extensive root system, soil that allows the roots to grow down, general health, surrounding trees for support*
    - Diseases? *natural resistance, favorable weather conditions, available nutrients*
    - Insect Damage? *mechanical defenses such as spiny or hairy leaves and stems, tough stem and leaf coverings, chemical defenses (such as toxins in plant parts)*
    - Drought? *deep root system to reach ground water, general health, available nutrients*
    - Pollution from smoke/volcanic eruptions? *winds, oxygen and carbon dioxide exchange*

- Pollution from humans (acid rain, industrial gases)? *available amounts of water to dilute the pollution effect, available nutrients, short exposure time, human enforcement of pollution control emissions*

2. In what parts of a tree would you expect to find cells containing DNA?
All cells that contain a nucleus contain DNA. These would be in the actively growing areas of the tree. Not all plant cells contain a nucleus. Those areas of the plant that have finished growing and have specialized into mature cells (such as some cambium, xylem and phloem cells) have lost their nuclei. These cells cannot go through the process of mitosis.

3. How can you tell if a tree is alive and healthy? *new and abundant growth*

4. Optional question -- How do you plant seeds from seedless grapes? (Best asked with a smile.) *Seedless grapes are a result of genetic engineering (selective breeding); selecting grapes with smaller and smaller seeds and then crossing those to ultimately make grape vines that produce seedless grapes.*

During Viewing

1. START the video.

2. PAUSE the video (1.49 into the video) after Dave Milarch says "we're looking for new growth".

   Remind the students of the process of mitosis and where rapid cellular reproduction takes place.

3. RESUME the video and play to the end.

After Viewing

1. Ask and discuss the following questions:

   - When the sycamore was young, 200-300 years ago, do you think there were other trees in the same area? Were they competing for the same space, nutrients, and sunlight? How could this tree outlive all its competition?"
     All the trees in that area were competing. Perhaps this tree was situated in a place that was protected, received more water and nutrients, grew faster, and remained healthy. All the other trees surrounding it eventually died out, either naturally or by human actions.
   - What kinds of traits does the Sycamore tree have that would be important to preserve? *Height; size; health; vigor; resistance to pestilence, harmful insects, environmental changes, and air pollutants; ability to produce a broader and deeper root ball.*
     (Note: You might want to relate this to humans by asking: What are some traits of the oldest people you know? *healthy diet, healthy lifestyle, good genes, healthy heart, mental clarity, competitiveness*, etc.
   - Why was it necessary to climb to the top of the tree for branches? *to obtain the vigorously growing branches where the tree has actively reproducing cells.*
   - What are the requirements for a tree to be classified as a 'champion tree"? *old and large with vigorous growth*.
   - Why did the scientists wish to clone the trees planted by George Washington? *Apparently Washington had a good eye for good genetic predisposition.*

2. Draw on the board or on a transparency a picture of the Champion Tree grafting process. Use different colors for the root stock and the champion tree graft to show how the grafting/cloning process works. Describe why the

new tree has only the Champion Tree DNA. Then describe the DNA in an organ transplant patient and lead a discussion on how these two processes are similar.
connecting the nutrient supply, arteries, veins

3. Conduct the Student Activity: Observing Young Branches.

Teaching Notes for the Student Activity: Observing Young Branches

In this activity, students will observe freshly cut, young tree branches in order to examine the features that make them suitable for cloning/grafting. By using histological diagrams and slides, students will investigate the anatomy of the cambium layer of cells, including bark cambium and wood cambium, as well as apical meristematic tissues and buds. To successfully graft two plants, the vascular cambium of both plants must align correctly. This activity helps explains why scientists seek these tissues for cloning/grafting, while allowing students to explore the living tissue of trees and other plants.

Before the activity:

1. Review or demonstrate how to prepare a wet mount slide.
2. Remind students of safety procedures and precautions when working with glass slides, scalpels or razor blades. As an alternative, you may wish to create a station where cutting will be done for students.
3. If possible, have prepared slides of stained cross sections of young branches or saplings that show xylem, phloem, and other plant tissues. These same sections will also contain cambium layers that stain differently than underlying tissue. Slides of apical meristem will show the rapid cellular divisions that occur in this region as well.
   Note: A field test teacher suggested telling students to draw these slides on a separate sheet of paper to help clarify what they are looking at. If slides are unavailable go to http://www.uri.edu/artsci/bio/plant_anatomy/images.html#lab_6 for images or use a search engine for each particular type of tissue such as wood cambium, bark cambium or apical meristematic tissue.

After the activity:

1. Lead a class discussion on the differences in the cells that were observed.
2. Describe the parts of the cell including any organelles that students may have observed.
   Cell wall
   Nucleus
   Chloroplasts
   Nucleolus
   Cytoplasm
   Plasma membrane (students can not see but should be able to point to its location next to the cell wall).

Extensions

- Extract DNA from branch meristematic tissue using a procedure adapted from an onion DNA extraction lab to demonstrate the abundance of living cells in these areas. Alternatively, extracting DNA from onion cells themselves would be appropriate here to show the abundance of DNA in plant tissue.
- Graft geranium branches from one plant to another to demonstrate the importance of connecting cambium tissues. Different species could also be grafted to investigate plant-specific nutritional requirements. For example, a salt-intolerant plant could be grafted onto a salt-tolerant plant and monitored over time. Students could subject multiple plants to varying salt concentrations and compare them to controls (non-grafted plants).
- Plan a field trip to a local champion tree to observe it first-hand. Alternatively, find a large tree on school grounds or nearby that has survived many years. Students could research its history and document what

past disasters or diseases the tree has survived. Surrounding trees can be documented to see if other same-species trees have also survived along with it.

- Take a trip to a local orchard or tree farm where grafts are performed. Students could watch a graft in progress and see the outcome of past grafts.

Answer Key to Questions on the Student Handout

3. Are the cells in the stem longer or shorter than those in the apical meristem bud?
   longer

4. Which part of the plant (stem, bud) is going through the process of cellular division most rapidly?
   bud

5. How is the DNA different from the stem and the bud?
   it is not different but the way in which it is controlling cell growth and functions is different

6. Complete the following sentence: The Milarch's used the buds (apical meristems) of the trees for the cloning/grafting of the Champion Trees because … _____

   …that is where the new growth is occurring and the DNA still has maximum ability for differential cell growth

Student Handout: Observing Young Branches

Name:_____ Date:_____

In this activity, you will observe freshly cut, young tree branches so you can examine the features that make them suitable for cloning/grafting. By using histological diagrams and slides, you will also investigate the anatomy of the cambium layer of cells, including bark cambium and wood cambium, as well as apical meristematic tissues and buds. This activity will help you understand why scientists seek these tissues for cloning/grafting. Keep a log of your observations.

Materials

- Freshly cut branches from a maple, oak, or other tree
- Prepared or fresh stained slides of meristematic tissues
  - Wood cambium
  - Bark cambium
  - Apical meristem
- Magnifying lens
- Microscopes (optical or digital)
- Slides and cover slips
- Plastic knives (for scraping bark)
- Scalpel, razor blades, or a station where cutting will be done for you
- Tweezers or forceps
- Food coloring or some other kind of dye

Safety

Follow proper safety procedures when handling glass slides, scalpels, and razor blades.

Procedure

1. Carefully observe the various structures of the branch. Make a sketch of the branch and label as many parts as you can without dissecting it.

2. Next, use a plastic knife to gently scrape the bark off a thin portion of the stem and expose the wet, green layer of the cambium. Using forceps, try to peel away a very thin portion of this inner tissue and make a wet mount slide. See the following Web site for information on making a wet mount. http://www.sidwell.edu/sidwell.resources/bio/VirtualLB/wetmount.html

3. Observe your wet mount slide under a microscope and sketch your observations on the next page. Add stain such as iodine, food coloring, or bromothymol blue as directed by your teacher to show organelles.

4. Now observe the apical bud structure using a stereomicroscope or magnifying lens. Use a scalpel, razor blade, or the cutting station to make a very thin cross section of apical bud to study its inner layers. Sketch your observations on the next page

5. Observe the prepared slides of stained sections of these same tissues that your teacher has made available. Sketch your observations on the next page

6. Answer the questions that follow.

7. Observations
   Sketch the branch. Label the following parts: bark, stem, leaf, bud, leaf scar, apical meristem

8. Wet mounts

Thin portion of the stem	Apical bud structure (apical meristem)
Observations	Observations

9. Prepared slides

Thin portion of the stem	Apical bud structure (apical meristem)
Observations	Observations

Questions

1. Are the cells in the stem longer or shorter than those in the apical meristem bud? _____
2. Which part of the plant (stem, bud) is going through the process of cellular division most rapidly?

_____

1. How is the DNA different from the stem and the bud?

_____

_____

2. Complete the following sentence: The Milarch's used the buds (apical meristems) of the trees for the cloning/grafting of the Champion Trees because

_____

_____

Additional Resources

Because Web sites frequently change, some of these resources may no longer be available. Use a search engine and related key words to locate new Web sites.

Books and Articles

Dodds, John H. and Lorin W. Roberts. (1995). Experiments in Plant Tissue Culture. Cambridge University Press, 3rd edition.

Kyte, Lydiane, John Kleyn, and John G. Kleyn. (1996). Plants from Test Tubes: An Introduction to Micropropagation. Timber Press, Incorporated, 3rd edition.

Trigiano, Robert N. and Dennis J. Gray. (1999). Plant Tissue Culture Concepts and Laboratory Exercises. CRC Press, 2nd edition.

Web Sites

http://www.actionvideo.freeserve.co.uk/grafting.htm
Grafting has long been used to join parts of one plant to another, in order to combine the relative strengths of both.

http://instruct1.cit.cornell.edu/courses/hort494/mg/
This Web site is a "how to" for tree grafting.

http://www.nal.usda.gov/speccoll/images1/wye.html
An American Champion: Maryland's Wye Oak. This Web site describes the life & death of one of the Champion Trees.

http://www.championtrees.org/
The Champion Tree Project was founded in 1996 in Michigan to preserve these biggest, best, tallest, strongest, and eldest representatives of Earth's largest living plants.

http://instruct1.cit.cornell.edu/courses/hort494/mg/
This is the Time magazine Web site that has collected all related articles on "Dolly", the first cloned animal.

http://www.vcbio.sci.kun.nl/eng/virtuallessons/cellcycle/mitomeio/
Mitosis vs. meiosis

http://www.encyclopedia.com/html/section/reproduc_asexualreproduction.asp
Rapid Cellular Reproduction - Asexual Reproduction

http://sres.anu.edu.au/associated/mensuration/density.htm
Trees and competition

Genomic Revolution

http://www.ornl.gov/sci/techresources/Human_Genome/education/education.shtml
The Web site to the government-funded Human Genome Project with links about genomics, the history of the project, and more.

Secrets of the Sequence Videos and Lessons

This video and 49 others with their accompanying lessons are available *at no charge* from www.vcu.edu/lifesci/sosq