



NSF POTATO GENOME

CENTER FOR PLANT GENOMICS
TRAINING AND EDUCATION

Summer Genomics Workshop

BAKER • BROWN • TIGR • UCBG

JUNE - JULY 2004

Workbook



Table of Contents

I. Presentation Worksheets	3
Potato Genome Project Overview (Barbara Baker)	4
Summer Genomics Workshop (Meghan Flanagan)	7
PCR/ Restriction Endonucleases (Amy deHart)	10
Bioethics of GMOs (Anthony Chien and Jack Sun)	13
Microsatellites (Wai Shum)	16
Microarrays (Miki Yamamoto)	19
Silencing (Rumi Asano)	22
II. Background and Review Worksheets	25
Solanaceae and Solanum	26
Molecular Biology	30
Genomics and Bioinformatics	34
Protocols	36
Microsatellites	42
cpDNA	45

I



Presentation Worksheets



Potato Genome Project Presentation Notes



Barbara Baker



Potato Genome Project



Barbara Baker

Exercises for Understanding

1. What does the term genomics mean?
2. What are the names of sub-topics or disciplines in genomics?
3. Name or describe one way plants detect and defend against pathogens.

Presentation Review



Summer Genomics Workshop Presentation Notes



Meaghan Flanagan



Summer Genomics Workshop



Meaghan Flanagan

Exercises for Understanding

1. What is the purpose of our work this summer?
2. What do we hope to accomplish by the end of summer?
3. List the techniques a researcher can use to classify a potato.

Presentation Review



PCR / Restriction Endonuclease Presentation Notes



Amy DeHart



PCR and Restriction Endonucleases



Amy DeHart

Exercises for Understanding

1. What is the function of a DNA polymerase?
2. What is special about the DNA polymerase Taq?
3. What are the three steps in PCR?

4. Which of the following DNA sequences is most likely to be recognized by a type II restriction endonuclease?

- a) 5' -CTCCCG-3' b) 5' -GAGAGA-3'
 3' -GAGGGC-5' 3' -CTCTCT-5'
- c) 5' -AGACTC-3' d) 5' -AGATCT-3'
 3' -TCTGAG-5' 3' -TCTAGA-5'

5. What technique can be used to differentiate between individuals in a population?



Bioethics of GMOs Presentation Notes



Anthony Chien and Jack Sun

Presentation Review



Bioethics of Genetically Modified Organisms



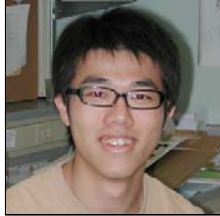
Anthony Chien and Jack Sun

Exercises for Understanding

1. What role do restriction enzymes and DNA ligases play in recombinant DNA?
2. How is genetic material introduced into a plant?
3. What are the three major categories of concern regarding the use of genetically modified organisms?



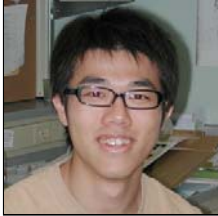
Microsatellite Presentation Notes



Wai Shum



Microsatellites



Wai Shum

Exercises for Understanding

1. Give examples of mono-, di-, trinucleotide simple sequence repeats.
2. Where are microsatellites found?
3. How do microsatellites become different lengths?

4. Give specific example of a microsatellite application.

5. Give examples of the three models of microsatellite mutation.



Microarray Presentation Notes



Miki Yamamoto



Microarrays



Miki Yamamoto

Exercises for Understanding

1. What does a microarray slide or have spotted on it?
2. What is hybridized onto a microarray?
3. What can be measured with microarrays?

4. How is the microarray technique more advantageous over the traditional way of measuring expression level using Northern Blots?

5. When analyzing the data, what does it mean when a spot is yellow?

6. Give 2 examples how microarrays can be applied in medical purposes or for basic research.

7. How does having a genome of an organism sequenced add to the utility of using microarrays?



Silencing Presentation Notes



Rumi Asano

Presentation Review



Silencing



Rumi Asano

Exercises for Understanding

1. What is another term for silencing?
2. At what level does silencing interfere with cellular processes (DNA, RNA or Protein)?
3. What are some applications for this technique?

4. Name a type of silencing technique.

5. Name a gene that has been silenced.

II



Background and Review Worksheets



Solanaceae and Solanum

Exercises for Understanding

1. Name the family and genus in which the common cultivated potato belongs.
2. What is inflorescence, and how does it relate to potato?
3. Do all potatoes have tubers?
4. What is a tuber, and what is its function?

10. Briefly describe the history of the potato.

11. Where is the ancestor of today's cultivated potatoes located?

12. What is potato Late Blight disease?

13. Please label the parts of the potato plant below.





Exercises for Understanding

1. What is the central dogma of biology?
2. What is a gene, and what do genes encode?
3. Draw one nucleotide of DNA with an adenine (A) residue, and its complementary nucleotide. (Include base pairing hydrogen bond rules.)
4. Draw one nucleotide of DNA with a guanine (G) residue, and its complementary nucleotide. (Include base pairing hydrogen bond rules.)

5. Fill in the base pair sequence complementary to the one shown below.



6. If the sequence shown in question 3 was repeated four times, what would be the number of As and Gs?

7. According to the number of As and Gs in the 4x repeated strand (question 4), how many Ts and Cs will be in the 'complementary strand' and why?

8. At the end of one round of DNA replication, how many strands are present?

9. Of the strands present after one round of DNA replication, what is their composition? (For example, 2 strands with parent DNA for one, daughter DNA for the other.) Draw a sketch if this helps.

10. What is necessary for DNA replication? (If you were going to set-up an in vitro replication in the lab, what component would you need?)

11. What is the component most active during transcription?

12. Transcription results in the production of _____?

13. What are the three main types of pre-mRNA processing?

14. Draw a picture of a pre-mRNA molecule and a picture of a processed mRNA molecule. (Include exons and introns).

15. What is the function of tRNA?

16. What is a codon?

17. What is the 'genetic code'?

4. How many genes are in a genome? (This doesn't have an exact answer, and is more like a trick question.)



Exercises for Understanding

1. What is a cationic detergent?
2. Give an example of a cationic detergent other than CTAB.
3. What does it mean when we say that CTAB "solubilizes" membranes?
4. In order to extract only genomic DNA, what major intracellular components must be eliminated from the extract?

5. What is used to separate these components from the DNA?

6. List four ways that agarose gel can affect the rate of DNA migration, and describe with one sentence.

7. Why will we use polyacrylamide gels for our SSR project, rather than agarose gels?

8. The process of silver staining for polyacrylamide gels requires a "reductant." What is a reductant, and what purpose does it serve in polyacrylamide gel staining?

9. In the diagram below, which fraction is the supernatant (green or white)?



10. How are the bands of agarose gels visualized?

11. What compound allows researchers to visualize the bands in an agarose gel?

12. If the percentage of agarose: TAE is increased from 1% to 4%, how will this affect DNA migration?

13. What basic cellular activity is the polymerase chain reaction based upon?

14. What are primers and how do they work in PCR?

15. Why do scientists use PCR?

16. What happens at each temperature in the PCR cycle?

17. When will we use PCR this summer (for what experiments)?

18. Draw a diagram of the products of the first three cycles of a PCR reaction.

19. Using two primers and one original DNA template, how many copies of DNA are present after seven PCR cycles?



Exercises for Understanding

1. Give examples of mono-, di-, trinucleotide simple sequence repeats.
2. Circle the examples that are considered microsatellites.
3. Give specific example of a microsatellite application

4. Provide real-world examples of mutations that cause differential length microsatellites (Search terms: human disease, microsatellites, SSRs)

5. Give an example of polymorphic microsatellites.

6. Give examples of the three models of microsatellite mutation.



Exercises for Understanding

1. What set of experiments in 1962 proved that chloroplasts contain DNA?
2. Name two organelles other than chloroplasts that contain DNA separate from genomic DNA.
3. How can we define T type potato cpDNA?

7. What molecular biology techniques will we use for our cpDNA analysis?