Case It: Case-based Learning in Molecular Biology

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Overview

- Introduction to *Case It!* project
- New cases on honey bee biology
  - Virus detection by PCR
  - Bee virus bioinformatics
- Case It Mobile prototype
- Open-ended research applications
  (if time permits)
  - HHMI SEA-PHAGES project

Case It! Project

URL for Case It! Home Page:

http://www.caseitproject.org

- Includes tutorials and download links
- Access to case descriptions

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Case It! Project

Electronic framework for analyzing and discussing case studies in molecular biology

- Genetic and infectious diseases and associated ethical issues
- Students gather background information on cases
- Analyze DNA and/or protein sequences using Case It! simulation
- Online poster sessions
- Role-playing

Techniques for DNA and protein analysis Case It! simulation

Features of Case It! simulation

- DNA analysis
  - PCR, Restriction enzyme digestion, electrophoresis, Southern blot, Dot blot
- Protein Analysis
  - ELISA, Western blot, protein electrophoresis
- Microarrays (SNP and Expression)
- Bioinformatics tools
  - Connection to BLAST and other NCBI tools
  - Integration with MEGA software - Alignments, Tree building

➢ Case studies in genetic and infectious diseases and other biology topics

Honey bee cases

- Collaborators:
  - Kim Mogen and Brad Mogen, UWRF Biology
  - Marla Spivak, UM Bee Lab

- Incorporating research on honey bee health and colony collapse disorder in first-year biology classes
  - Virus detection by RT-PCR
  - Effects of pesticide exposure, mite levels, etc.

Case scenario

Honey bees are commonly exposed to pesticides as they forage for pollen and nectar. Some pesticides are known to disorient bees and thus affect their behavior. Sub-lethal exposures of some pesticides are considered possible contributing factors to Colony Collapse Disorder (CCD). Dr. Muskiver was curious if pesticide exposure was linked to virus infection, another possible contributing factor to CCD.

To test this question, Dr. Muskiver set up test colonies, and fed the honey bees either with untreated pollen or pollen treated with sub-lethal doses of pesticides. She then tested the bees for the presence of several viruses using multiplex PCR.

PCR Result

1.0 % agarose runtime = 60 100 volts	1. 100 bp ladder 2. neg control 3. pos control all viruses 4. Hive 1 pesticide-treated 5. Hive 2 pesticide-treated 6. Hive 3 no pesticide 7. Hive 4 no pesticide

Case scenario - bioinformatics

Recent declines in honey bee populations have given rise to the syndrome named Colony Collapse Disorder (CCD). Several potential stressors have been identified. A team of research scientists, funded by the North American Honey Bee Council, decide to survey colonies from around North America for two of the notable stressors – Deformed Wing Virus (DWV), a virus that causes wing deformation, and *Varroa destructor*, a parasitic mite that feeds on the bee.

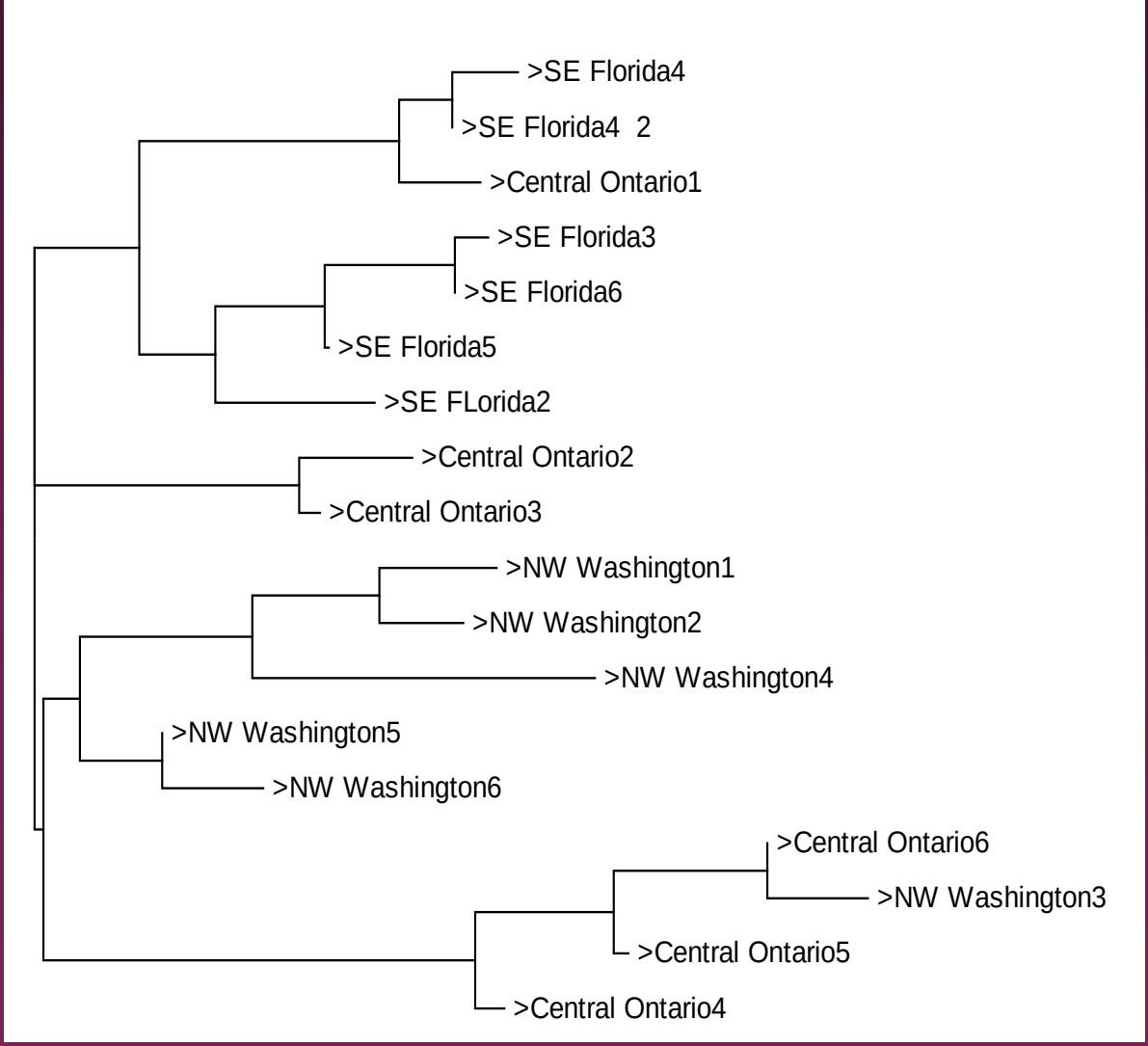
It has recently been reported that there are *V. destructor* transmit certain strains of DWV more effectively, and that long-term mite infection reduces virus diversity and leads to the prevalence of more pathogenic viruses. The scientists are interested in testing the relationship between DWV strains and the *Varroa* mite in North America.

Case scenario - bioinformatics

Bees tested from:

- Central Ontario - low mite levels
- Northwestern Washington - low mite levels
- Southeast Florida - high mite levels
- Oahu, Hawaii - high mite levels
- Northern Arizona - moderate mite levels
- Southern British Columbia - moderate mite levels

DNA sequence analysis



Case It! mobile

- Access to case scenarios and lab results from tablets, smart phones, and Macs

- See prototypes at www.caseitproject.org/mobile

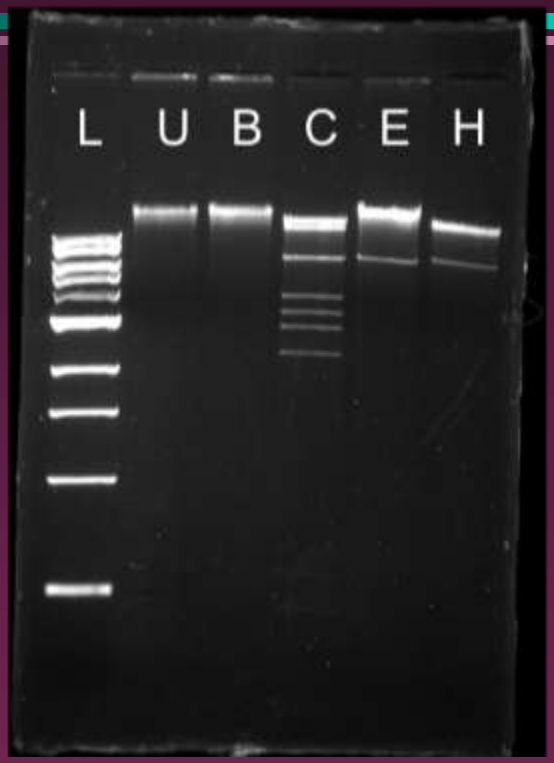
Open-ended research

- HHMI SEA-PHAGES project in General Biology course for freshmen
- Lab sequence replaced by phage research
  - Isolate mycobacteriophages from soil
  - Isolate phage DNA and analyze by restriction enzyme digestion
  - Select one phage to send for sequencing

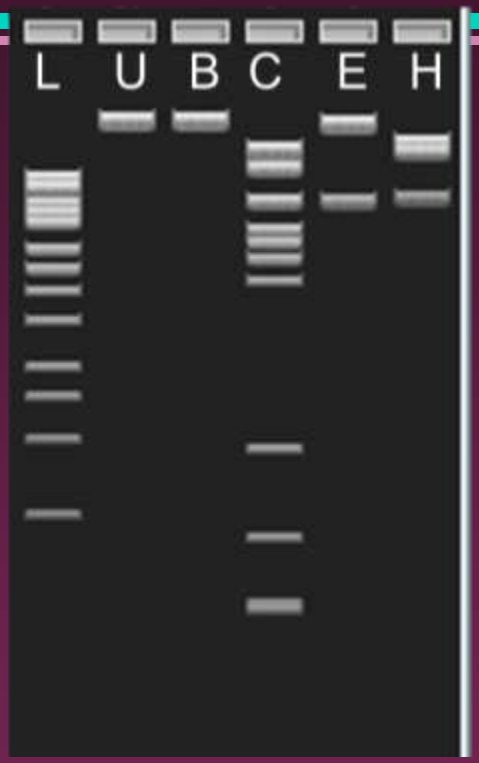
Open-ended research

- Spring semester – phage genomics
- Retrieve complete phage genome sequence
  - Annotate genes
  - Comparative genomics
  - Research projects on phage biology
- www.phagesdb.org

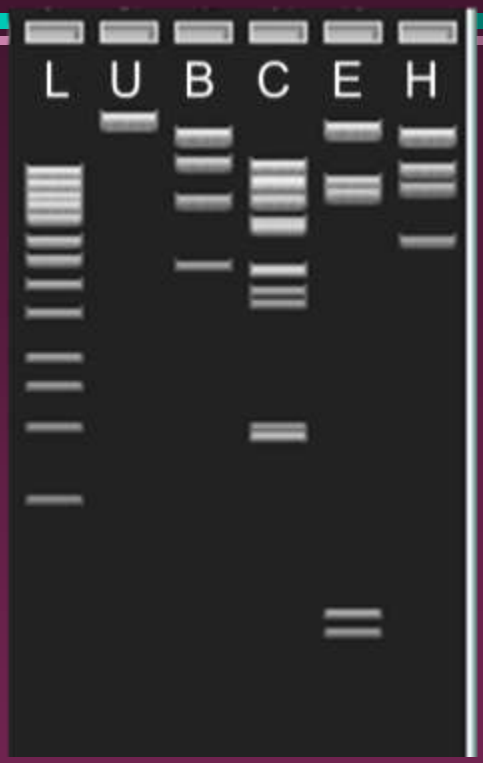
Abrogate lab gel



Abrogate virtual gel

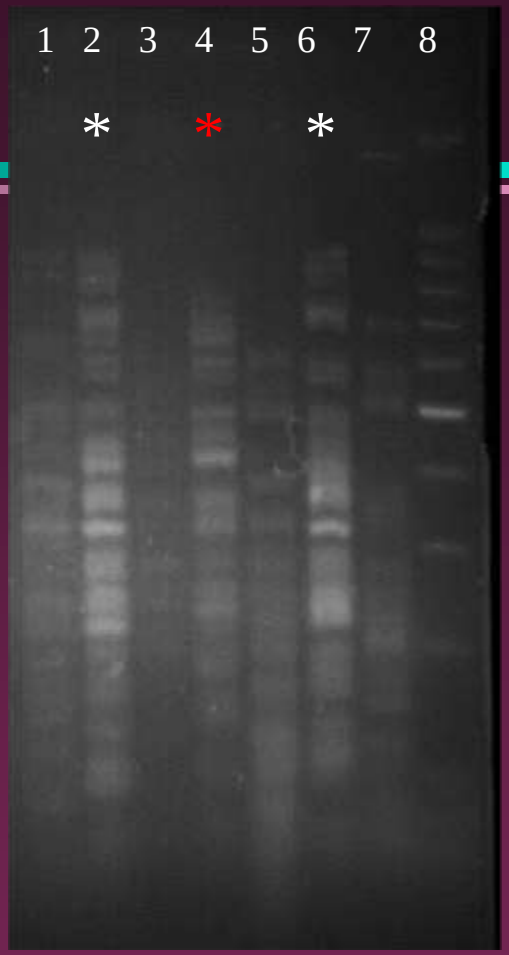


Bxb1 virtual gel

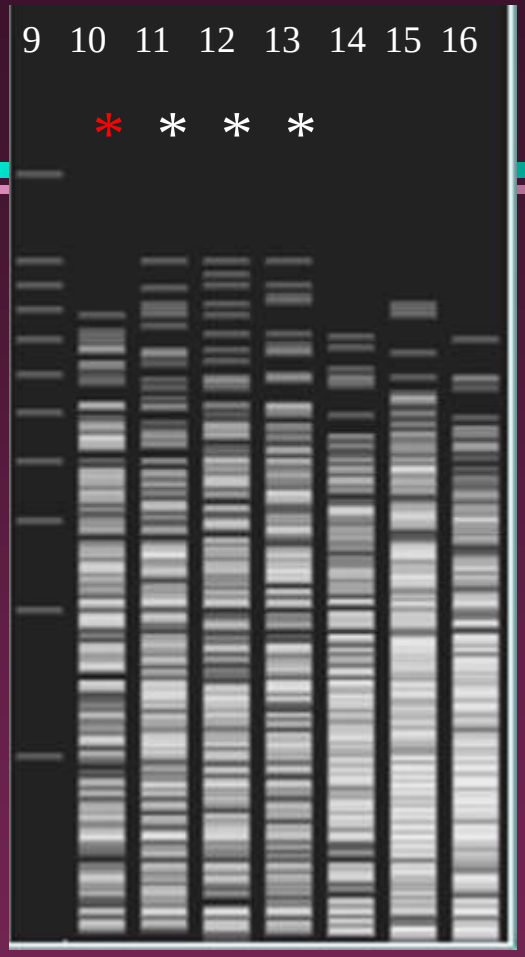


L=1 kb ladder; U=undigested; B=BamHI; C=ClaI; E=EcoRI
H=HindIII

HaeIII Lab Gels



HaeIII Virtual Gel



- Ran separate in 2% agarose gel
- * Indicates A1 phages
- * Indicates Abrogate
- Abrogate significantly different from A1 phages

Huntington's disease case

Case scenario - from Case It web site

Restriction enzyme digestion and Southern blot
or
PCR and gel electrophoresis

Sequence analysis - detect triplet base repeat, sequence alignment and BLAST to identify gene

Sample case: Huntington's Disease

Susan is a 23-year-old whose father, age 55, and paternal aunt, age 61, have been diagnosed with Huntington's chorea. A paternal uncle, age 66, appears to be unaffected by the disease. Susan wants to know if she inherited the mutated gene from her father so that she can prepare for that future if necessary. She arranges to undergo DNA testing for Huntington's. Her 17-year old brother, John, also decides to be tested after talking with Susan.

Role playing

- Students present the results of their case analysis as a web poster
  - Includes a statement to the "family"
  - Wiki system provides group web posters with associated discussions

    caseitconferencing.wikispaces.com
- Visit another group's web poster and post questions in the role of a person in the case
- Authors respond to questions in the role of a genetic or health counselor

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- New – microarray cases (SNP and expression)
  - Breast cancer expression microarray
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  - HIV cases with introduction to bioinformatics
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Breast cancer case

- Woman diagnosed with breast cancer, without prior risk factors
- Microarray analysis used to determine potential for aggressive growth and invasiveness of tumor

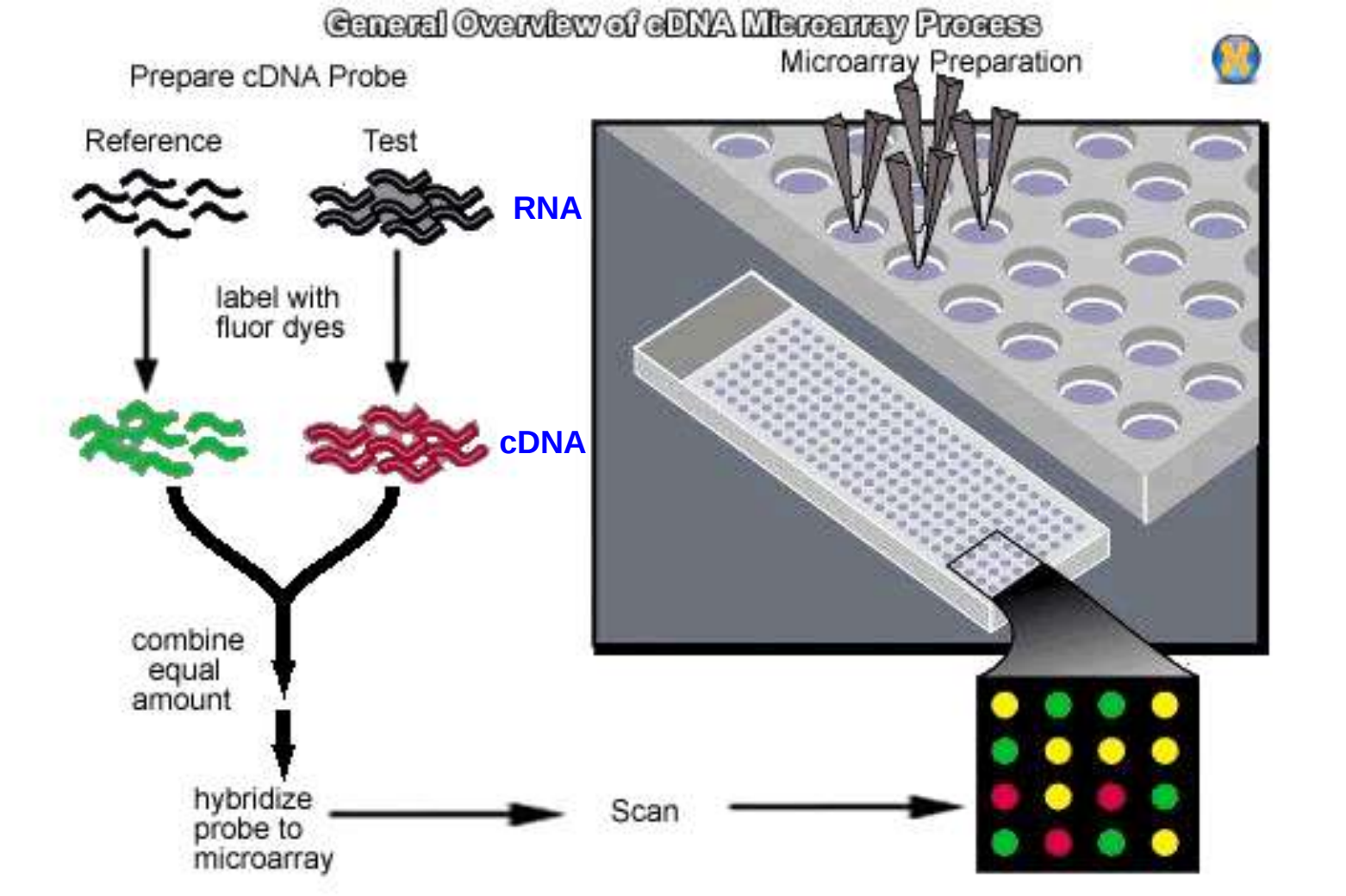
Breast cancer microarray case

Sarah was devastated when she received a diagnosis of breast cancer. It did not seem to run in her family, so she assumed she did not have to worry about it. She is grateful for the support of her friends, especially Molly, who is a clinical lab pathologist. Molly is helping her think about the difficult decisions regarding how aggressive her treatment should be, in terms of surgery, chemotherapy, etc.

Breast cancer microarray case

Molly explains that the oncologist recommended running a lab test that uses a microarray to measure the expression of specific genes. The pattern of gene expression can predict how quickly the tumor cells will grow and whether they will respond to various treatments. Sarah is meeting with the oncologist to review the results, and she has asked Molly to go with her.

Microarray method



Breast cancer microarray

Genes associated with increased cell proliferation (or rapid growth) in breast cancer tumors:

- Ki-67, STK15, Survivin, Cyclin B1, MYLB2

Genes associated with increased tumor cell invasion:

- Stomelysin 3, Cathepsin L2

Genes associated with proliferation in response to estrogen:

- SCUBE2, PGR, ERBB2

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HIV Case studies

Case scenario - video and text (Anna case), from Case It web site

ELISA test - initial screening (new autoload feature)

Western blot to follow up ELISA results

PCR to amplify HIV DNA for viral load or sequence analysis

Sequence analysis to determine source of HIV infection

Discussion

- What kinds of case studies would be useful in your classes involving the analysis of DNA or protein sequences?
- How could the Case It simulation be useful for the analysis of open-ended research questions?
- How do these approaches address the Vision and Change initiative?