

# UNCC Biotechnology and Bioinformatics Camp

Dr. Jennifer Weller

Summer 2010

# Part 2 - Agenda

- The Scientific Method
- Record Keeping
- The American Chestnut

# Focus Areas

- Scientific inquiry : how and why things happen
  - In order to understand and control mechanisms
- The scientific method
  - Posing answerable questions
  - Making measurements
- Record keeping
- Biology: study of living organism(s)
  - The American chestnut
    - Development, lifespan, nutritional needs, etc.
    - Range and co-occurring species
    - Diversity
    - Pathogens

# Scientific Inquiry

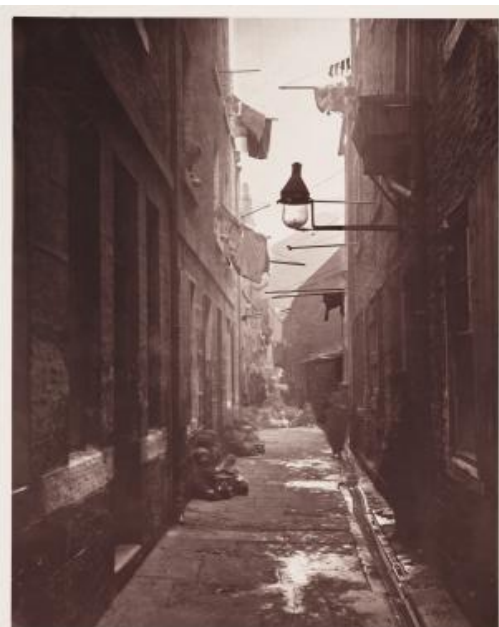
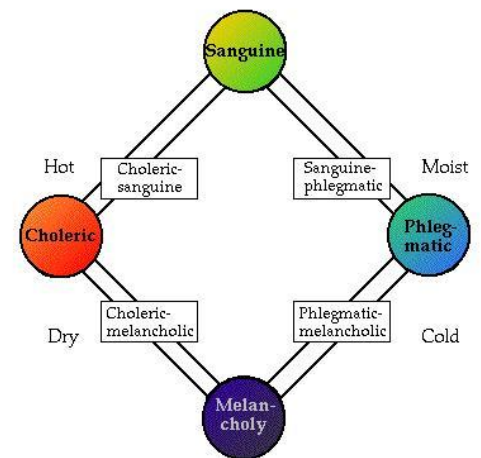
- Starts with a why of how question “Why does an organism get sick?”
  - Early models: external: ‘Miasma’ vs. internal: ‘humors’
  - Observations leading to these hypotheses?
- Humors → nutrition
- Miasma’ → something passed in the air or water

‘More questions: how does the body use the nutrients? How does the bacterium invade the body?’

Good experiments lead to more questions than they answer.

Theories are generalizations that explain many experiments.

**The Four Humours**



# Scientific Method



- Observation of a phenomenon
  - Apply experience to see if you can explain it; see what others may have done.



Form a conjecture about what might be going on.

- If your conjecture is right, what prediction could you make about other consequences
- Develop an experiment to test the prediction and its counter-result

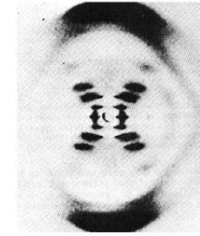


# Experimentation

- Carry out the experiment, keeping track of all the details about instruments and measurements
- Analyze the data and reflect on how it matches the predicted results
- Publish, → repeat



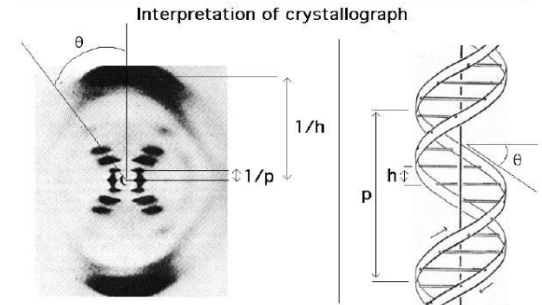
# DNA characterization



X-ray diffraction pattern from B form of DNA

- Observations:

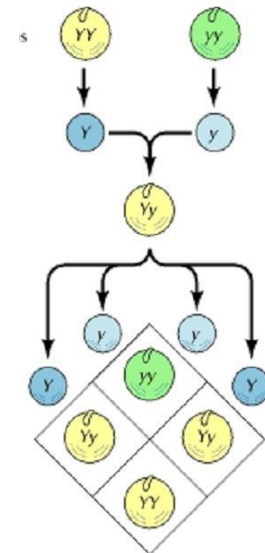
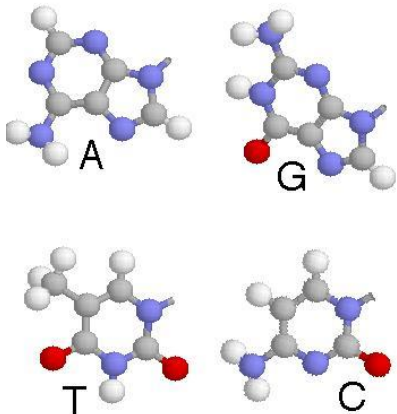
- DNA has only 4 basic subunits  
A=T and G=C.
- DNA has a regular structure, which allows it to form crystals  
- thus X-ray crystallography could be used.
- The bond lengths and chemical properties of the subunits and backbone were well known.
- DNA occurs in every living cell, and offspring inherit half their traits from each parent , copies are 'semi-conservative'.



$\theta$  - tilt of helix (angle from perpendicular to long axis)

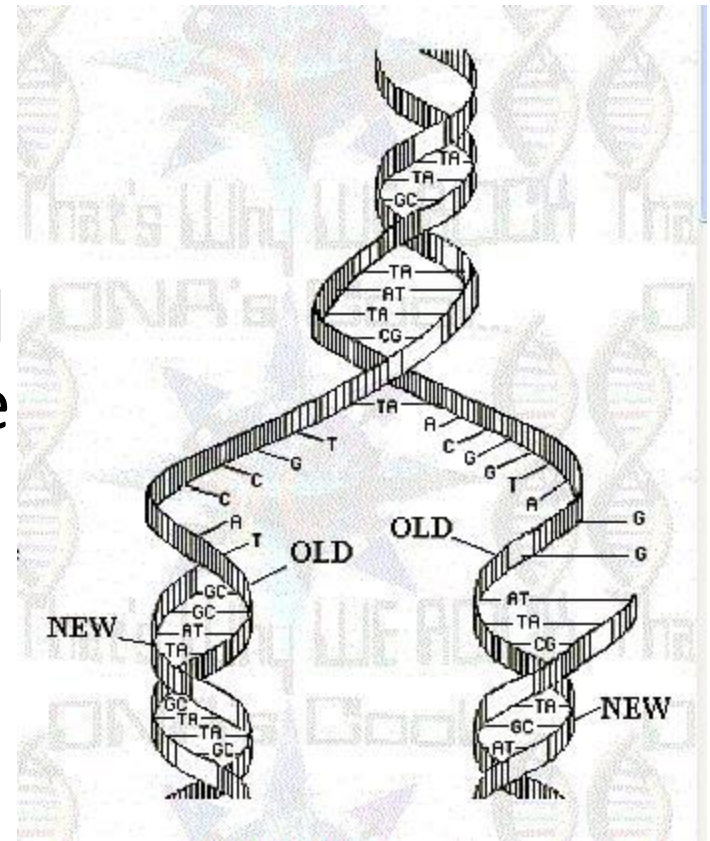
$h = 3.4 \text{ \AA}$  (Distance between bases)

$p = 34 \text{ \AA}$  (Distance for one complete turn of helix; Repeat unit of the helix)



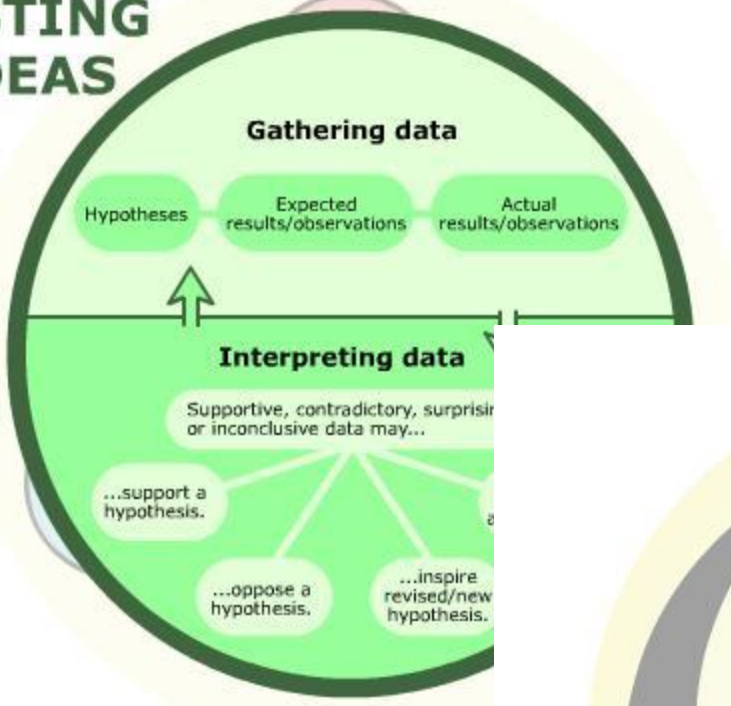
# DNA hypothesis

- The structure is some form of helix
- Predictions: The x-ray crystallography pattern would be typical of a helix. Given the double helix and H-bonds across the core, DNA replication would involve an enzyme that made a complementary copy of one strand.



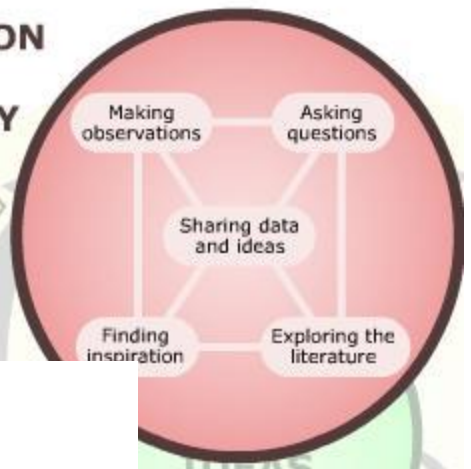


# TESTING IDEAS

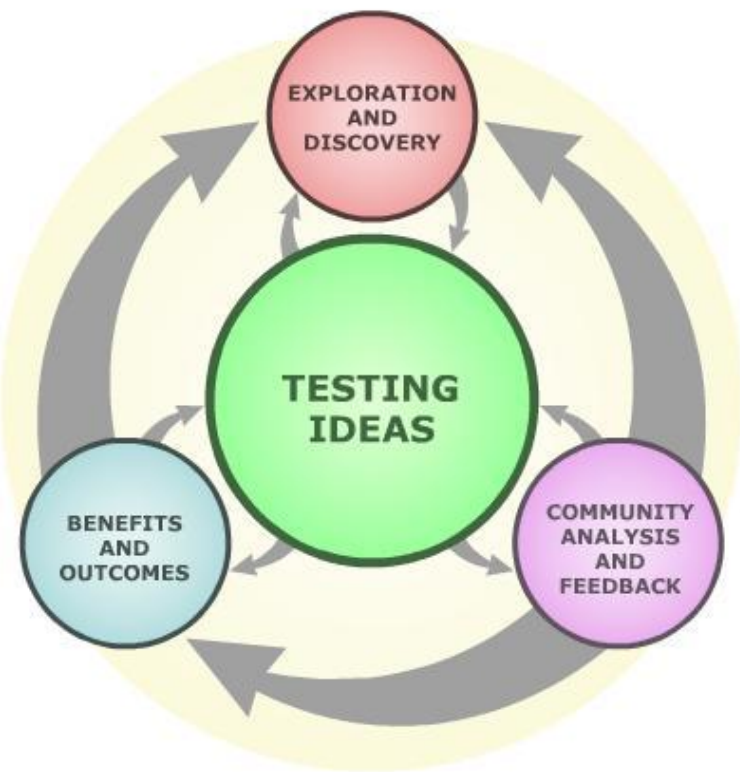
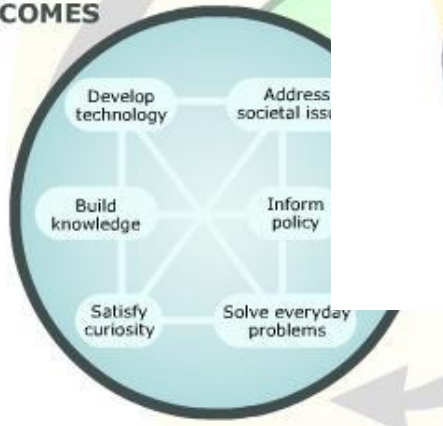


# EXPLORATION AND DISCOVERY

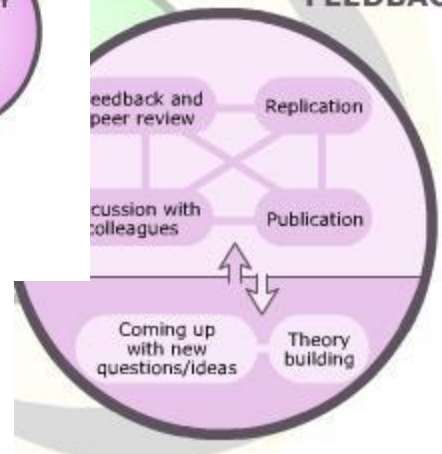
New technology  
Practical problem  
Curiosity



# BENEFITS AND OUTCOMES



# COMMUNITY ANALYSIS AND FEEDBACK



# Reporting Experiments

- A scientific paper with aspects of the scientific method expanded upon can be found here:
- <http://www.sciencemag.org/feature/data/scope/keystone1/>
- Some bioinformaticians compared gene sequences between humans and bacteria, to see if bacterial genes had got spliced into the human genome. Lateral Gene Transfer.
- Notebooks vs journals vs published reports

# Record Keeping

- You must record and report on the experimental design and conditions and results fully.
  - Others can judge if your experiment was effective and the integrity of your procedures .
  - Others can replicate your experiment
  - Journals are good for for notes, ideas, rough drafts of experimental designs
  - Data Notebook is for recording experiment set-up, measurements, immediate observations

# Data Notebooks

- Use a bound notebook
- Set up the notebook (always use ink):
  - Title page with name, date, purpose
  - Reserve 2-3 pages for a Table of Contents
  - Print page numbers
- Entering Data
  - Give a title to the experiment at the top of a page
  - Briefly summarize the question to be answered
  - Write down the experimental design, and equipment and reagents needed.
  - Put in a copy of the protocol (Methods)
  - As you carry out the protocol note actual values used, step-by-step as in the protocol
  - Record all observations and measurements , put in print-outs or copies of digitally recorded values. If instruments need to be calibrated record that information also.
  - Leave several pages to perform analyses, or the details and outcomes if a computer is used
  - Summarize the conclusions and your reasons for making them. Restate any errors made that you think might affect the outcome.
  - Leave room to note any paper citation information that uses the data.
- Completing the record
  - When done, fill in the Table of Contents and sign the bottom of each page.
  - Never tear pages out of the Data notebooks.

# Journals

- Use either a 3-ring binder or a bound notebook
- Put your name and the data on the front or title page
- During lectures and seminars, note the date, place, speaker, topic. Leave 1 page to summarize
- When you read source material: note the question that lead you to the article, the reference citation information. If you take notes directly, leave a page to summarize.
- If this is used in tandem with a data notebook you can note the pages that are relevant in the other notebook.

# Electronic lab books

- There are electronic lab notebooks, software that runs on tablets.
- You have to carry the tablet around with you (and many of them are commercial).
- GoogleDocs, with sharing, is another way to make your data available – I would like to use it if everyone has access.

File Edit View Insert Format Table Tools Help

Emily Galloway  
 BINF 6010 - Spring 2010  
 Lab Assignment Record

January 12, 2010

**Transfer using micropipettes:**

- ddH<sub>2</sub>O

Volume	Weight 1	Weight 2	Weight 3
7.500uL	0.007g	0.007g	0.007g
14.50uL	0.013g	0.014g	0.015g
76.20uL	0.076g	0.076g	0.077g
125.3uL	0.125g	0.124g	0.124g
450.4uL	0.448g	0.449g	0.448g

- 25% Glycerol

Volume	Weight 1	Weight 2	Weight 3
7.500uL	0.007g	0.008g	0.008g
14.50uL	0.016g	0.015g	0.015g
76.20uL	0.080g	0.081g	0.079g
125.3uL	0.131g	0.130g	0.131g
450.4uL	0.458g	0.458g	0.458g

**Practice dilutions: Fluorescent labeled and non-labeled oligomers**

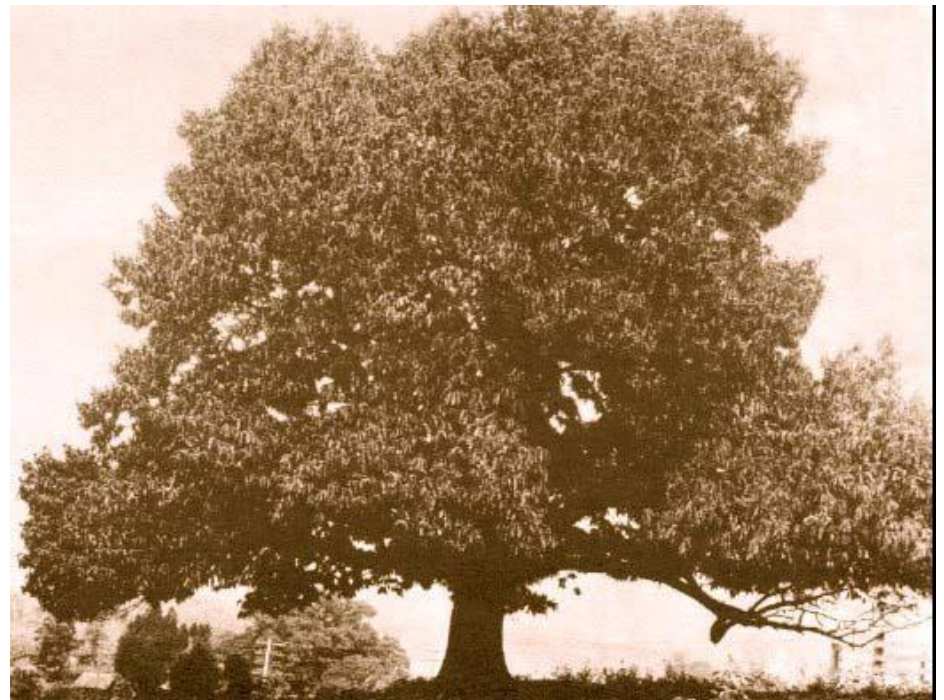
- Make 10mL of 3X SSC working stock from 20X SSC stock:

$$1.5\text{mL } 20\text{X SSC} + 8.5\text{mL ddH}_2\text{O} = 10.0\text{mL } 3\text{X SSC}$$

- 180 mL of 3X SSC was added to each of 24 microcentrifuge tubes:

Tube #	Addition
OL 1:1 A/B	20uL 100uM Oligo-1 stock
OL 1:2 A/B	20uL OL 1:1
OL 1:3 A/B	20uL OL 1:2
OL 1:4 A/B	20uL OL 1:3
OL 1:5 A/B	20uL OL 1:4
OL 1:6 A/B	20uL OL 1:5
OL 2:1 A/B	20uL 100uM Oligo-2 stock
OL 2:2 A/B	20uL OL 2:1

# The American Chestnut



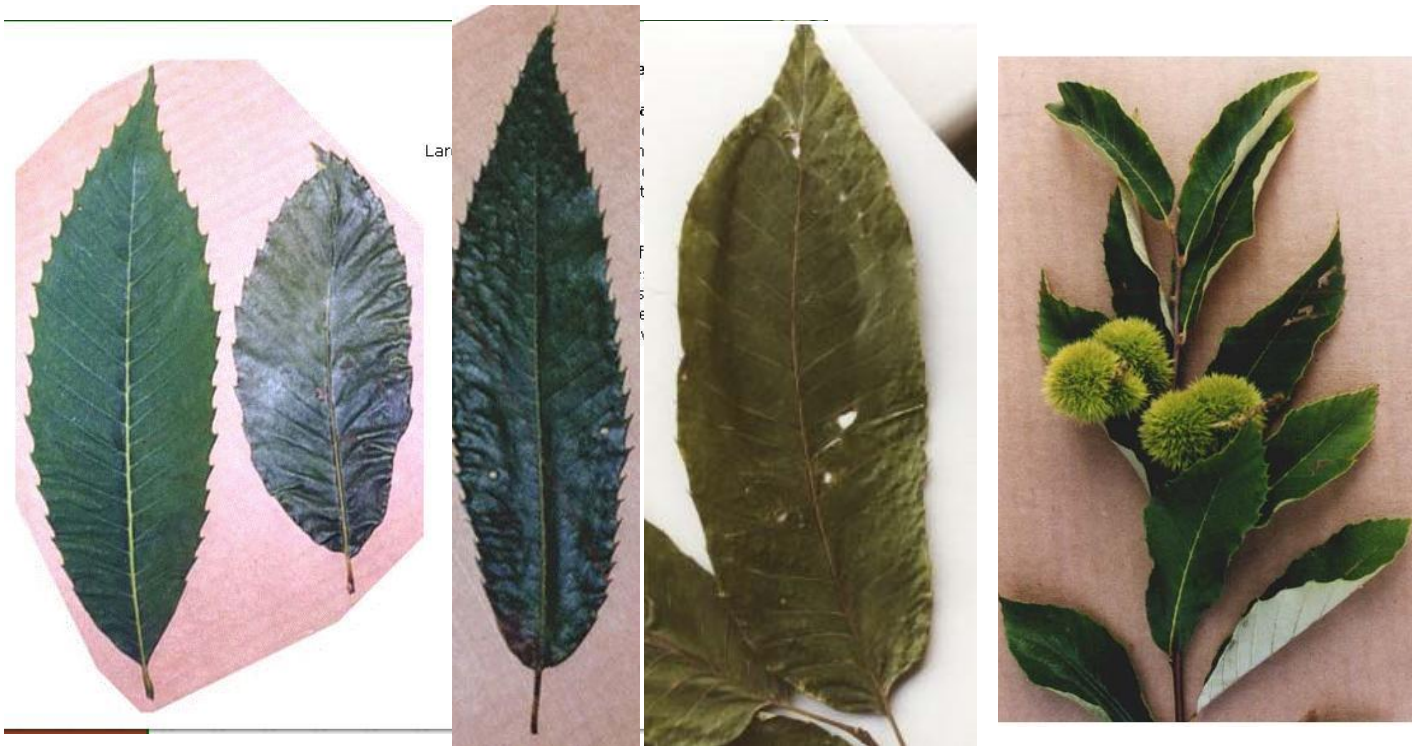


# Castanea dentata



# Field Notes

- American chestnut: Leaves are long compared to width, teeth on edges curve inwards, stems are reddish,
- Chinese chestnut: Oval leaves, thicker. If sunlit, the leaf backs are whitish, stems are grey with white bumps
- European chestnut leaves look like American chestnut leaves, teeth are triangular
- Japanese chestnut: leaves are a dark shiny green on top, sides are parallel
- Chinquapin



ULUS

## Chinkapin Leaves

Usually smaller than American chestnut leaves, but highly variable

Chinkapin leaves exposed to the sun are whitish underneath because of numerous leaf hairs

American chestnut leaves, in contrast, have few hairs and are light green underneath



**American Bur ----- Chinese Bur**

**American Chestnut Burs:**  
 A dense mass of long, slender spines  
 Spines are 2 to 3 cm long, 0.5 mm thick

**Chinese Chestnut Burs:**  
 A sparse mass of short, thick spines  
 Spines are 1 to 2 cm long, 1 mm thick



**American - Chinese - Japanese - European**



**American - Chinese - Japanese - European**



**American Bur ----- Japanese Bur**

**American Chestnut Burs:**  
 A dense mass of long, slender spines  
 Spines are 2 to 3 cm long, 0.5 mm thick  
 Up to 3 nuts per bur

**Japanese Chestnut Burs:**  
 A mass of spines that interlock in a  
 thatched pattern  
 Up to 3 nuts per bur

# Images of bagging trees (ACF)

- <http://www.youtube.com/watch?v=O098JFHSmmo>
- [www2.volstate.edu/jschibig/Pollination2006i.htm](http://www2.volstate.edu/jschibig/Pollination2006i.htm)