PLB 316: Cell & Molecular Section

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http://farrelab.openwetware.org teaching PLB316

Final, Room 255

Fri May 6th 7:45-9:45 am

OR

10 am-Noon?

Grading	
Protocols and Lab Notebook	10%
Summaries and Questions	10%
Quizzes	10%
Worksheets	10%
Photosynthesis Report	15%
GA5-1 Report	15%
Alpha-Amylase Worksheet	15%
Final Exam	15%
Total M and C	100%

For each class you need to prepare:

To hand in at the beginning of class:

- Summaries
- Questions

In your lab notebook (no scrap paper allowed!):
Protocol

For our next class:

- Read GA5-Complementation lab manual pages 1-6 (until step 2, PCR).
- Write the summary of what you will do .
- •Answer questions 1-4 on the lab manual.

OTHER POINTS:

There is NO text book but you are expected to know everything mentioned in class and included in the Lab Manual.

Some reference books are available in the class.

Background information can be found in any plant physiology and molecular biology/genetics book.

I expect you to think. Think first and ask second.

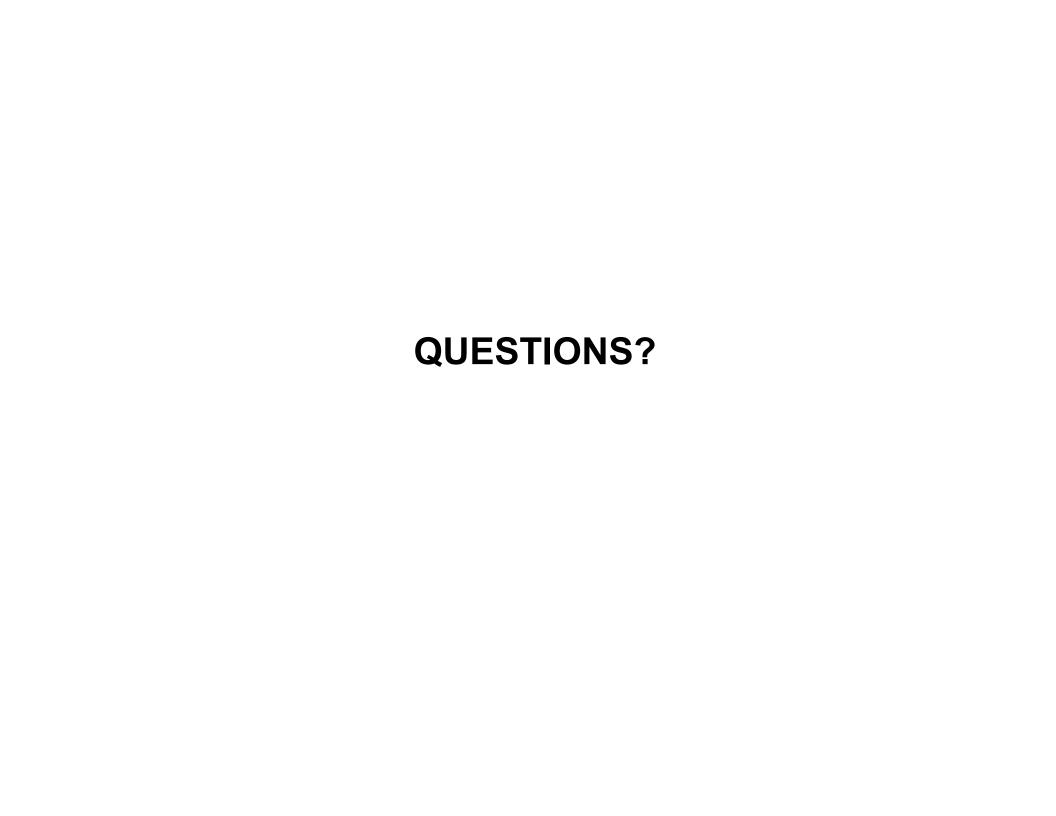
Things to bring to class:

Notebook

Scientific calculator

Pen

(laptop if available)



You will learn how to answer the following questions:

- 1. Is the gene "X" responsible for a particular mutant phenotype?
- 2. Is gene/protein "Y" induced under particular circumstances?

As a test case we will work on the plant hormone GIBERRELLIN

WHAT ARE HORMONES?

GENERIC DEFINITION

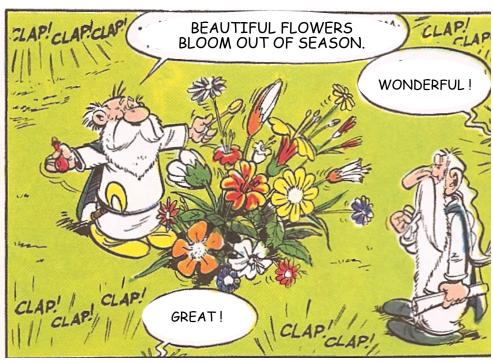
- •Are present in very low concentrations.
- •Are produced in one organ/tissue and regulates the function of a different one.

SPECIFIC CHARACTERISTICS OF PLANT HORMONES

•Mediate developmental processes often in response to the environment.

The power of plant hormones





GAs affect many aspects of plant growth and development







Gibberellins







http://www.hort.purdue.edu/newcrop/tropical/lecture_20/64m.jpg

A bit of history

Figure 1.
Elongation
symptoms of
bakanae disease on mature
plants at heading.



http://www.plantsciences.ucdavis.edu/uccerice/NEWS/RiceNL_mar2003.pdf

Gibberella fujikuroi



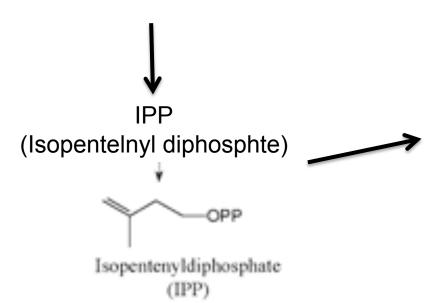
Fig. 1. Semi-dwarf rice cultivars and their tall isogenic lines. From left to right: Deegeo-woo-gen (dwarf *indica* cultivar), woo-gen (tall equivalent), Calrose 76 (dwarf *japonica* cultivar), Calrose (tall equivalent).

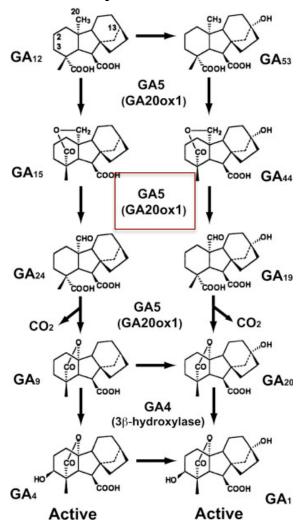
Hedden (2003)

"The Green Revolution"

Biosynthesis of GAs in plants

pyruvate + glycerol-3-phosphate





See: http://4e.plantphys.net/index.php

How do you find the genes involved in a particular process?

- a. Forward genetic approach; you start with a mutant phenotype
- b. Reverse genetic approach you start with a candidate gene

Our test case:

Xu et al., (1995):

ga5 semidwarf mutant contains a mutation in a GA 20-oxidase.

Is it possible that the mutation in the GA20 oxidase might NOT be the cause of the phenotype?

To confirm that this particular mutation is the cause of the phenotype we will carry out a complementation experiment.

Complementation Experiment

- 1. Clone the gene
- 2. Transform the mutant
- 3. Check the phenotype

Today

- 1. Practice some pipetting and check our pipettes.
- 2. Establish our cloning strategy for GA20OX.