Part 1B Plant & Microbial Sciences Practical : self-assessment worksheet

Reporter genes in plants

Gene fusions, GUS, GFP and microscopy.

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Web resources at: http://www.plantsci.cam.ac.uk/Haseloff/teaching/Index_teaching.htm

1. **Draw a diagram of a typical protein-coding plant gene. The annotation should include the following listed elements, and indicate their typical positions within the gene. Briefly describe the properties of the elements.**
promoter
upstream regulatory sequences
enhancer
silencer
RNA polymerase initiation site
intron
exons
UTRs
START codon
STOP codon
polyadenylation site

2. **What are transcription factors, and how do they interact with genes?**

3. **What is a reporter gene, and how is it detected?**

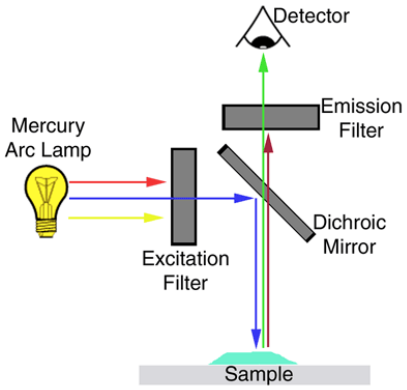
4. **Draw a schematic view of the differences between plant transformation vectors that might be used to produce protein fusions, transcriptional fusions and for enhancer detection in plants.**

5. **Why are these different gene fusions useful?**

6. **Both green fluorescent protein (GFP) and ß-glucuronidase (GUS) are widely used as reporter genes in plants. Describe major advantages of GFP over GUS, and vice versa.**

7. **A s a keen plant biologist, you wish to construct your own fluorescent microscope for work with a variant of green fluorescent protein. The excitation and emission spectra of the protein are shown below. You have access to a box of filters that transmit light in the following bands:**
   Bandpass Filter 1: 350-460nm; Filter 2: 450-490nm; Filter 3: 515-560nm, Filter 4: 550-570nm
   Beamsplitter Mirror 1: 560nm; Mirror 2: 505nm; Mirror 3: 580nm; Mirror 4: 595nm
   Longpass Filter 1: >470nm; Filter 2: >520nm; Filter 3: >580nm; Filter 4: >635nm

   You need to construct a suitable filter block for imaging GFP. What filters will you choose for the (i) excitation filter, (ii) beamsplitter and (iii) emission filter. Why choose this combination?



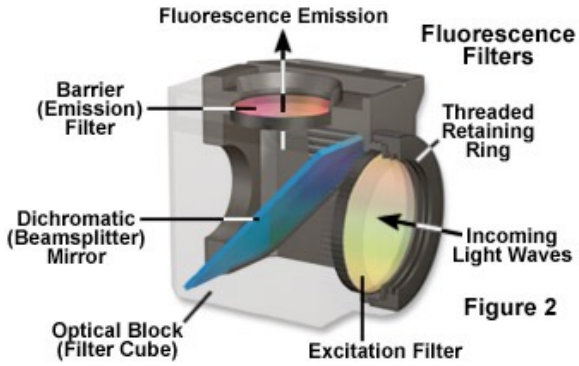
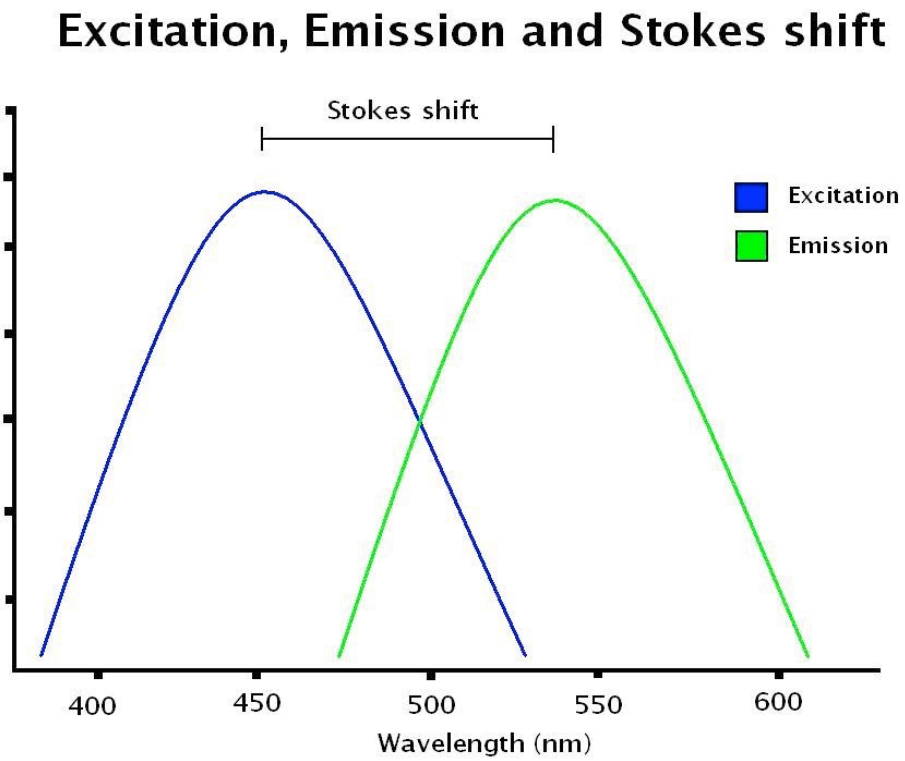


Figure 2



Microscopy weblinks

Molecular Expressions website featuring our acclaimed photo galleries that explore the fascinating world of optical microscopy. One of the Web's largest collections of photo-micrographs, and a lot of tutorials including diagrams and interactive Java applets:
http://micro.magnet.fsu.edu/index.html

Laser Scanning Confocal Microscopy
http://micro.magnet.fsu.edu/primer/techniques/confocal/index.html

Comparing wide-field and confocal microscopy techniques. Interactive Java applet.
http://micro.magnet.fsu.edu/primer/virtual/confocal/index.html
http://www.olympusconfocal.com/java/confocalvswidefield/index.html

Olympus and Nikon educational sites for confocal Microscopy
http://www.olympusconfocal.com
http://www.microscopyu.com/

Microscopy Primer: Physical and geometric optics of the microscope and their implications for practice. http://www.microscopy-uk.org.uk/full_menu.html

Including the Micropolitan Museum - http://www.microscopy-uk.org.uk/micropolitan/index.html

Fluorescent proteins

2008 Nobel prize for Chemistry - Fluorescent proteins
http://nobelprize.org/nobel_prizes/chemistry/laureates/2008/press.html

Fluorescent protein variants
http://www.olympusconfocal.com/applications/fpcolorpalette.html

Chromophore maturation in fluorescent proteins
http://www.olympusconfocal.com/java/fpfluorophores/index.html

Plant anatomy

The Photographic Atlas of Plant Anatomy contains photographs of plant structures.
http://botweb.uwsp.edu/anatomy/

The The Botanical Society of America Online Image Collection includes many photographs of plants and their anatomy. (http://www.botany.org/plantimages/)

An Introduction to Plant Tissues: McGraw-Hill
http://www.mhhe.com/biosci/pae/botany/histology/html/ptmodov.htm

Plant Study Resources: introduction to plant anatomy
http://biology.nebrwesleyan.edu/benham/plants/index.html

The Revision Modules in Plant Anatomy is an interactive site for reviewing plant structure.
http://bugs.bio.usyd.edu.au/2003A+Pmodules/home.html

The Plant Anatomy Laboratory contains micrographs and explanatory text from the textbook "Plant Anatomy", by James Mauseth. (http://www.sbs.utexas.edu/mauseth/weblab/)