

## HW 5

DUE: 10/20/2017 – 11:30 AM

MCDB 4312/5312

### Analysis Questions:

1. Consider a CFP-YFP FRET pair:
  - a. Draw the Jablonski diagram with energy levels labeled.
  - b. What is the Forster distance?
  - c. What is the FRET efficiency if the proteins are at a distance of 3 nm?
  - d. “ “ 8 nm?
  - e. What is the lifetime of the CFP when it's located 3 nm from the YFP as compared to when CFP is by itself?
  
2. TIRF
  - a. What is the critical angle for light traveling from glass to air?
  - b. “ “ glass to water?
  - c. What is the minimum numerical aperture needed to create a through the objective TIRF image for a live cell on a coverslip?
  
3. Assume you're imaging a live cell with a TIRF angle of  $78^\circ$ .
  - a. What is the penetration depth when using 561 nm light?
  - b. At what distance would you reduce the intensity by 90%?
  - c. At what distance would you reduce intensity by 90% moving from glass to air?
  
4. What is the Pearson's correlation coefficient of the following matrices:

2 4 3	4 1 3
1 3 3	2 4 3
1 4 1	2 2 1

### Matlab Questions:

1. Load in the red and green yeast image from the website.
  - a. Register the two images, and display the overlaid images.
  - b. Print out the 3x3 matrix of the transform to overlay each image.
  - c. Scatter plot the two images.
  - d. Scatter plot the two images of JUST the cells.
  - e. Calculate the PCC of the two images (whole and cells).