Quantitative Imaging and Analysis for Biologists (QIAB):

Fall, 2017 Lectures & Practical, Tuesdays / Thursdays 10AM – Noon; rm 104 Stellar Chance

Rationale for the course:
Students are making fundamental advances by capturing stunning, high-resolution digital images. Accurate and careful quantitative analysis of these can reveal important mechanistic insights into the underlying biology. However, most students are analyzing this data only qualitatively, with little guidance in techniques for quantitative imaging and its analysis. This course will provide an in-depth introduction to the analysis of images to engender confidence in applying quantitative imaging methods in one’s work.

Course Overview:
- How to acquire microscopy images suitable for quantitative analysis
- Knowledge of what comprises a digital image
- Steps necessary to extract information from 2-, 3-, and 4-D images, including:
  - Object identification / feature extraction
  - Object tracking over time
  - Co-localization
  - Quantitation
  - Advanced processing pipelines
  - Special applications such as Live cell & Tissue imaging
  - Visualizing single molecule dynamics
  - Responsible Conduct in Imaging Research, & Scientific Rigor

Class meetings: Combination of lecture with hands-on tutorials, over ~5 weeks
- Ten lectures (beginning Tuesday September 26th, carrying through the month of October)
- Hands-on image manipulation, both individually and in small group exercises during “practical portions” of class time
- Image manipulation homework

Course Application & Prerequisites: Open to BGS or SAS Biology graduate students. Enrollment will be capped under 20, with priority given to those with a demonstrated need and a preference to those past their qualifying exam. Fill out the Google Form at (https://goo.gl/forms/XdkxaWfMZb6HPELo1), and within that form explain your need for this course by:
A) succinctly describing the role played by light microscopy to date in your research,
B) the system(s) you have been using to gather data (Point scanning, Spinning disk, other?);
C) how you imagine that the analytical methods covered here might advance your studies.
This justification should take less than 1 page.

Instructors:
- Andrea Stout, Director of the CDB Microscopy Core
- Teaching Assistants: Terra Kuhn & Andy Moore to assist with exercises & office hours

Materials:
- Student-owned laptop, preloaded with the FIJI release of ImageJ

Grading & evaluation: attendance, plus a Quantitative Analysis Mini-Project
You must apply two of the image analysis methods from the class to your own image data set (e.g., images you’ve acquired for your thesis research)
- Execute measurements, extracting quantitative information
- Prepare a publication-quality Figure
- Present the Figure to the class, discussing its quantitative analysis
Course OUTLINE draft

Week #1 September, 26 / 28th
-- Tuesday, 26th: The basics of microscopy & image formation: Andrea Stout
  • How a microscope works
  • How images are formed
  • Choice of microscopy method; sample preparation

-- Thursday 28th: The basics of proper image acquisition: Andrea Stout
  • Optimization of imaging to collect data for quantitative analysis
    o Understanding lenses & optics to the degree necessary to set proper imaging parameters
    o Understanding Detectors (cameras and point-scanners) that have different noise characteristics & optimization strategies

Practical portion: Discuss graded assignment & its presentation, due October 31st.

Week #2: October 3rd / 5th
-- Tuesday, 3rd: Basics of image display, measurements and filters using Image J (FIJI): Andrea Stout
  • Channels; Bit depth; Resolution; Noise; File maintenance

-- Thursday 5th: Feature Extraction / Object identification Andrea Stout
While you “see” the object you care about, how can the computer learn to “see” and then report on that object?
  • Image segmentation
    o Preparing images for segmentation by filtering, smoothing, background subtraction, threshholding, etc.
    o Using Fiji “Analyze Particles” to understand segmentation
    o What to use if simple threshholding fails (other binary filters, watershed, etc)

Practical portion:
Exercises on image segmentation & threshholding.

Week #3: October 10th / 12th
-- Tuesday, 10th: Object tracking in 2-D / Motility / Kymographs: Instructor to be named
  • Movement dynamics; Working with object tracking algorithms; Kymographs

-- Thursday 12th Intro to Macros: Andrea Stout

Practical portion:
Exercises on tracking, kymographs, batch processing and macros
**QIAB course**
Stout, Granato, DiNardo.

**Week #4: October 17th / 19th**
--- **Tuesday, 17th**: Solving biological problems by applying modeling approaches using quantitative parameters from images as inputs  **Shawn Little**

Practical portion (potential):
Exercises on extracting data, and applying it to a biological problem

--- **Thursday 19th**: Advanced processing: **Melike Lakadamyali**
- *Beyond Image J – Challenges & Solutions to Single-molecule & Super-resolution imaging.*

**Week #5: October 24th / 26th**
--- **Tuesday, 24th**: Live Cell & Tissue imaging considerations:  **Sandra Maday**

Practical portion: Continue work on independent assignment for presentation

--- **Thursday 26th**: SRR / RCR – Scientific Rigor & Responsible Conduct in handling your Research Images:  **Michael Granato**
- What you **cannot** do, and why using case study round-table discussion

Practical session: Continue work on independent assignment for presentation

**Week #6: October 31st**
--- **Tuesday, 31st**: Student presentations & Pizza!!