Bi 1X, Spring 2011

Assignment 5: Single Molecule Restriction Digest (TPM)

Things to turn in:

- A write-up on what you did in week 4 (single molecule digest) in a lab report format. Follow the guidelines under the "Assignment" section in the <u>Grading Handout</u> on the Bi1x website. These include:
 - Introduction to single molecule digest: the tethered particle method (TPM)
 - o Summary that explain the protocols you used
 - Results, your conclusions and discussion, as well as any sources you cited
 - Figures and plots with their own captions such that you could independently understand it without having to refer to the text
 - Remember to use full sentences and walk the reader through each step. Do not write a list. In this case, your lab notebook is a reference for you to look up exactly what you did in the procedure.
 - Remember to attach your own Matlab code for full credit
- Be sure to include the following in your write-up:
 - Follow the Matlab tutorial for one frame of your choice taken during the single-molecule session. Create a gallery of some of the images generated in intermediate steps of the analysis: Im, ImThresh, ImMask, and ImOverlay. Make sure to show the before and after pictures if you're cropping the images.
 - Once you've worked out a strategy to find the beads, analyze your whole movie and generate a plot of the number of beads found as a function of time. Make sure to label the axes and title.
 - Print out some frames of the movie corresponding, for example, to the first, last and some middle frames (You might want to look into the Matlab command *montage* in order to plot many frames right next to each other). Make sure to add scale bars, time stamps, and captions on your images.
 - Estimate the rate of DNA cutting from your plot in units of number of beads per second. Be sure to explain how you estimated.