We will use both FIJI and MATLAB in this lab. Unless specified, you can choose what you prefer for each task. FIJI often gives you a quicker and more visual way to apply filters, while MATLAB lets you be more involved in what you are doing.

Please put together a lab report in PDF format that is simple, well structured, well written and that shows that you have grasped all the tasks in the lab. Include necessary illustrations. You don't need to include screenshots of everything you do, just the relevant images and results. Take your time to understand the material. Let me know if you need extra help. If you are not done by the submission deadline, please submit what you did so far and you can complement later. (I will not take points off for this, the goal of the deadline is not to put stress and time pressure on you, just to make sure everyone follows along with the course.)

- **1 Object Recognition**

  Find an image of your choice in which you can try to find objects. If you don't have an image you want to work with, you can take the MATLAB rice grain image. You can use both FIJI and MATLAB to try to count the rice grains / your objects. Experiment with preprocessing the data to be able to find the grains, such as thresholding, blurring, spatial filtering with LoG or other edge detectors. See slides from Lecture 4 for support on this. Please also feel free to use the course textbook for how to work with this stuff in MATLAB.

- **2 Tracking Moving Objects**

  Now find a **movie** of your choice in which you can try to track objects. Start out with the movie of beads diffusing in 2D under Files using TrackMate. First step in tracking moving objects is to find the objects. You can either manually annotate or use some of the provided functions to detect the objects automatically. Adjust your parameters so that this works well. Step through the software and try out different ways to visualize and quantify your data.

  Extra task that will not be graded: If you are super interested in tracking, try out some other and more complex data sets, 2D or 3D. If you like, try other methods than trackmate.

- **3 Colocalization with color**

  Here you will try out some different ways to measure colocalization. Find an example image that has some colocalization of different channels in it. This can be data from your own research or found on the internet. If you cannot think of anything, you can use the example image provided under Files which is the same as we did in the Tuesday lecture. Tuesday's lecture slides should clearly illustrate the concept. If you need help, just let Sara know and we will go through the stuff together!.

  - Logical Operators.
    Try manually estimating the colocalization of the red and green proteins in the example image (or whatever the relevant channels are in the data you selected) by manually using logical operators on the red and green pixels ("and", "or" *etc.*) in either FIJI or MATLAB. Visualize your results. Try to quantify your results by e.g.

giving the percentage of green only, red only, and red and green pixels, in your image. Do the results make sense when you look at them?

- Hue Analysis.
  (Feel free to do task 4 first if the HSV format is not intuitive to you to get to explore it a bit before doing this stuff. Also see Lecture 4 slides for help. Or just ask Sara for help!)
  Try manually estimating the amount of yellow pixels in the red-green data set (or whatever the hue is of the mix of the two colors in your own data set) by converting the RGB to an HSV and finding the yellow hue. This format is very different from RGB. In RGB you have a red, green and blue matrix which each contain the intensity of each color. In HSV you have a hue, saturation and value matrix instead. Experiment with this and familiarize yourself with this color data format. (In MATLAB you can do this using the function rgb2hsv(). Try this and try plotting the three channels of your image; Hue, Saturation and Value). Now try to find the yellow (or other color) pixels. Does this give you a good result, or do you also need to include saturation and/or value? Try to quantify your result like you did with the previous attempt. Do the results make sense when you look at them?

- Extra task that will not be graded:
  If you are super interested in this stuff and will apply it in your own research, or if you just want to learn even more and have some time on your hands, I recommend some pedagogic youtube videos by Dr Craig Daly at the University of Glasgow. Nice tutorials on doing colocalization in ImageJ with different plugins and softwares. https://www.youtube.com/watch?v=rRyJnFo57xU Try some "already made" functions for analyzing colocalization. For example, Colocalization finder and Scatter Plot, Manders and Pearson coefficients, etc

- **4 Hue analysis of a color image / photograph**

  Find a color photograph that you like and try out converting it to HSV to do some hue analysis. Find some hue in the image and display where in the image you find it. Try something fun like changing the hue to another color, or try some operations on the Value and Saturation.

- **5 Please tell me how this lab was for you!**

  - Did you learn anything new or did you already know all of this?
  - Was this lab super hard, super easy, or an appropriate amount of work and difficulty?
  - How do you feel about going forward in this class? If you have any concerns please do let me know. You can send me a personal email or write it in your lab report.