Fluorescence-based and High Throughput Technologies

- You are a researcher in tissue engineering lab (perhaps this is already true!) and your boss has asked you to design an experiment to measure the expression of protein X in your cultured cells in response to an array of doses of drug Y. You initially proposed a high-throughput experiment but your boss thinks it is too expensive. Justify your reasoning for this choice. How would you convince your boss otherwise?
- Respond to at least two of your classmates.

To measure expression of protein X interaction with drug molecule Y, if you don't use high-throughput technologies you will then use FRET or maybe more sensitive techniques like FCS or two-photon dual-color FCS. For FRET you will then use either PCR or cloning vectors with for example Cy3 and Cy5 fluorophore pairs, and insert them into X and Y proteins for expression. Then engineered cells will need to grow in culture and experiments will be performed. Data analysis will require a fluorescence imaging system with image capture, and image processing and it will also require a microscope that uses a two or more-photon excitation laser.

One high-throughput technic which could be used for this task; is to create an interaction map which starts with fluorescent labeling similar to the experiment just described above. Then fluorescent protein microarray of some protein complex built with protein X and molecule Y are scanned using a molecular scanner device, these devices are popular in research and usually already available in your team. The alternative is to send the arrays to specialized companies which will process them and send back protein expression data. In possession of these data files, you can use a variety of free software packages to run the analysis and query very exhaustive and freely available online databases like the Human metabolome Project (HMP) to help you to analyze the metabolites produced and quantified the results. For the FRET study the major cost is the purchase of a fluorescence imaging system which could be significant compared to the purchase of a protein microarray scanner. To avoid the purchase of a scanner; partnerships with scanner vendors could be considered based on shared credits for future publications.