Flow Post-its
Kathy Daniels

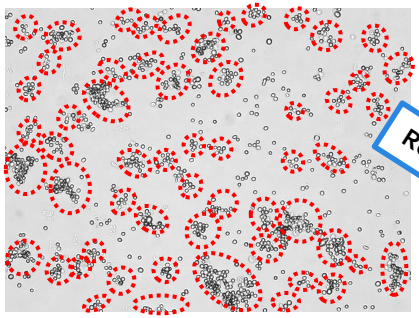
Cell Dissociation: One Size Does NOT Fit All

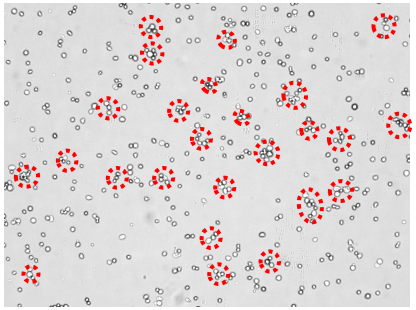
March 2021

Optimizing sample preparation is key for a successful flow cytometry experiment. For most applications, both analysis and sorting require a single cell suspension of cells. Due to the heterogeneity across different cell lines and tissue types, **the same dissociation methods will not work with all samples**. There are many variables that need to be assessed when optimizing preps.

Temperature

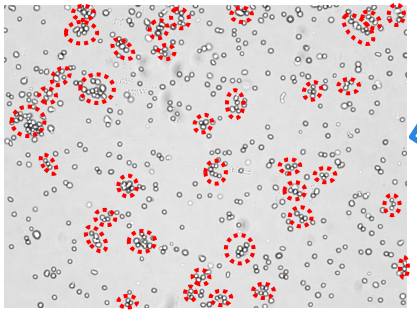
Temperature changes in storage and handling of cells can impact cellular metabolism and interactions. Assessments should be made to determine optimal temperatures.

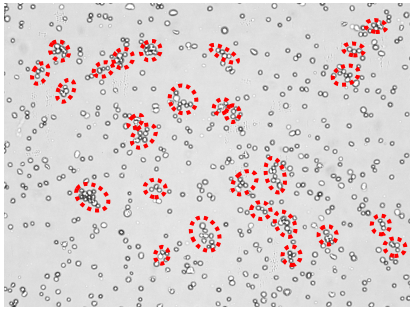




EDTA

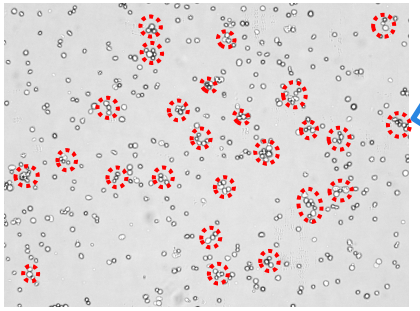
By chelating divalent cations necessary for cell to cell adhesion, the inclusion of EDTA can prevent reaggregation of cells. Suggested concentrations range between 2-5mM.

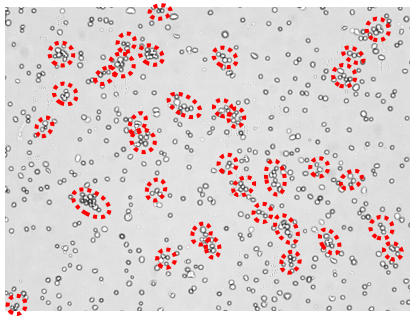




Dissociation Buffer

Various enzymatic digestion buffers are available to dissociate adherent cell lines or tissues. Comparisons should be made to determine which buffer results in the best single cell, viable suspension of cells that preserve surface epitopes of interest for subsequent staining.





After initial difficulties obtaining a single cell suspension for cell sorting, multiple dissociation conditions for a known difficult adherent cell line were tested. By looking at multiple conditions, it was observed that dissociation with Accutase, resuspension with 5mM EDTA and keeping the cells on ice prevented larger aggregates of cells.

The FCCF thanks Dr. Frances Weis-Garcia for preparing and providing the cells used in this Post-it

Flow Cytometry Core Facility
https://fccf.mskcc.org
Memorial Sloan Kettering Cancer Center