

Freezing adherent mammalian cells

1. Trypsinize cells.
2. Make freezing media (40% FBS and 20% DMSO in media without supplements).
3. Label cryogenic vials with date, initials, type of cell and type of protein(s) expressed, if any. It would help tremendously if you also fill out a “Bjorkman Lab Cell Collection” sheet every time you generate a new cell line because a cryovial can only store so much information.
4. Once cells have been trypsinized and pelleted, resuspend in 1:1 mixture of 40% FBS and 20% DMSO to end up with final concentrations of 20% FBS and 10% DMSO.
5. Store cells in cryogenic vials at -80°C for at least 24 hours before transferring to liquid N_2 Dewar.
6. Transfer the cryovials to our liquid N_2 Dewar.

Note: I usually freeze 3 cryovials of cells (1mL each) per 10cm^2 dish and 5 cryovials of cells (1mL each) per 15cm^2 dish.