

Ethanol precipitation for DNA concentration (protocol provided by Cindy Converso, Luk Lab)

Protocol:

1. Add 0.1x volume of 3 M NaOAc. Add 2.5x volume of 100% EtOH. Vortex.
2. Spin: 4 C, 15k rpm, 15 min. Remove supernatant by pipetting.
3. Wash pellet two times with 500 ul 100% EtOH.
4. Wash with 70% EtOH. Remove all liquid.
5. Allow pellet to air dry for at least 30 minutes (until completely dry).
6. Redissolve in 5 ul of molecular H₂O (or in greater amounts of water depending on the amount and concentration goal). Usually there is a loss of about 30% of the product which has to be considered.

Changes to protocol:

- We could not see the pellet, so we had to work under the assumption of its placement. We made sure to always centrifuge with the eppendorf in the same position relative to the center of the centrifuge and then assumed the pellet is on the opposite side of the tube.
- After each wash and before discarding the wash fluid, we centrifuged the eppendorf for about 1 minute to make sure that the pellet went back to its position.