

Resuspension of Primers

Overview

This protocol covers resuspension of dried (lyophilized) oligos that are to be used as primers. In this protocol both a stock (100 μ M) and working (10 μ M) solutions are made.

Materials

- Lyophilized Oligos (IDT)
 - o Each oligo should have a corresponding specification sheet, which should be stored in the specification sheet drawer.
- Labeled 1.7mL centrifuge tubes (1 per sample)
 - o Tubes should be labeled with primer name and 10 μ M, dates and initials should be included on the side
- Nuclease Free Water (NFW)ⁱ

Procedure

1. Briefly spin down oligo tubes.
 - o Use green benchtop centrifuge
2. Determine correct amount of NFW to add to each tube for a 100 μ M solution. This number can be found on the specification sheet. Alternatively, it can be determined by multiplying the number of nanomoles of DNA provided by 10 and then using that many μ l of NFW. Remember that each tube will have a different amount of NFW to add.
3. Add correct amount of NFW to each tube. Heat at 55C for 2 minutes. Vortex. Briefly spin down.
4. Prepare a 10 μ M working stock by adding a NFW and 100 μ M primer at a ratio of 9:1 (9 μ l NFW for every 1 μ l primer). Typically we make a 100 μ l worth of working solution.
5. Store primers in appropriate boxes in -20. Log their locations on google drive. Update the location column on appropriate primer excel sheet in the inventory folder on Dropbox.

ⁱ Other labs might use TE buffer, which is better for long term storage. As we've never had a problem with degradation, and switching to TE could conceivably cause minor problems downstream (from EDTA), we use NFW. Additionally, switching now would create an inconsistency.