# Resuspending Primers

## Introduction

Primers arrive from IDT dehydrated; to use them, you need to resuspend them in TE to a known concentration. By convention, the stock concentration (in the blue-capped IDT tube) is 100 $\mu$M; the working concentration depends on the application:

- For PCR, the working concentration is usually 10 $\mu$M
- For sequencing, the working concentration is usually 5 $\mu$M

## Materials

- Nuclease-free TE
  - We resuspend in TE to help supress nuclease activity that would degrade the primers.
- Dehydrated primers from IDT
- One extra epi tube per primer
  - ...for the working stock.

## Procedure

### Resuspend the dried primer

1. Label the top of the oligo tubes. I recommend the group initials and a number.
2. In the little microfuge, spin the (dry) oligos briefly.

   Sometimes the freeze-dried primers flake off the bottom of the tube.
3. Determine how many nanomoles of primer are in the tube.

   This is the bottom line on the tube label. As an example, the BT-01 primer says
   7.3OD = 33.4 nmol = 0.23 mg. <-- the number of nanomoles is "33.4".
4. Add 10 $\mu$l of nuclease-free TE for each nanomole of primer.

   For the BT-01 primer, this volume is 334 $\mu$l.
5. Vortex briefly.
6. Check the bottom of the tube to see if the primer is fully resuspended. If not, vortex again.
7. Pulse spin the primers.

### Make a working stock

8. Label the top of the epi tube the same as the oligo.

9. Determine the working stock concentration that you want.  Label the side of the epi tube with the working concentration.

10. Make 100 $\mu$l of working stock.

    For example, if you're making a 10 $\mu$M working stock, dilute 10 $\mu$l of the concentrated stock into 90 $\mu$l of TE.

11. Vortex  briefly.

12. Freeze both the concentrated and working stocks in the primers box.