Reconstituting (Dissolving) Oligonucleotides (From Invitrogen / Life Tech)

Oligonucleotides are synthesized, then shipped in lyophilized form (dry). Our lab prepares a stock of the oligo at 50 µM and a working sample, at 5 µM. However, if the primers will be used for qPCR or RT-PCR analysis, prepare 100 µM stocks, and no working sample is needed.

1. Remove the tube(s) from the packaging and centrifuge for 20 s at top speed to bring the dry oligo to the bottom of the tube.
2. On the top of the tube, label it with the oligo name (e.g. "OLA 225")
3. On the side of the tube, indicate "50 µM" or "100 µM" (it doesn't matter where).
4. Find the "Certificate of Analysis" sheet. Leave the other forms that came with the oligo(s) with Ben. (If you order more than three oligos, there will be more than one Certificate of Analysis sheet).
5. On that sheet, you'll find information for each primer, including the number of nmoles produced.
    - If you are preparing 50 µM stocks, find this number and multiply it by 20. The result is the number of µL of 1X TE needed to get 50 µM.
    - If you are preparing 100 µM stocks, find this number and multiply it by 10. The result is the number of µL of 1X TE needed to get to 100 µM.
6. Add that amount of 1X TE to the tube. Let it sit for ~3-5 min. Vortex vigorously. Spin down for 10 s to collect everything at the bottom of the tube.
7. For 50 µM stocks, prepare a working sample tube in a regular 1.5 mL microfuge tube, labeled with the oligo name and "5 µM" on top (e.g. OLA 225 / 5 µM).
    - To the working sample tube, add 450 µL 1X TE.
    - Add 50 µL of the 50 µM stock that you just prepared to the 450 µL in the working tube.
    - Vortex.
8. Place the stock and working sample in the appropriate oligo box in the -20˚C freezer. Oligos should be in numerical order in the boxes, with working samples placed adjacent to the corresponding stock tube.