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 Qubit

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Protocol status: Working

We use this protocol and it's working

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Abstract

Our protocols are constantly evolving and old versions will be deleted.
The documents here are not intended to be cited in publications



Scale Protocol

- 1 Double check that you have appropriately scaled this protocol. After clicking "run" above, you will need to enter your number of samples + 2 standards + 1 extra for pipette error (e.g. if you are running 6 samples, scale this protocol for 9).

Setup

- 2 Take out standards (1 and 2) and Qubit buffer from cabinet above NGS bench. Also get the Qubit reagent from the labeled blue box in the drawer under the NGS bench. Retrieve DNA extractions and defrost (if they were frozen previously). Be sure to vortex and centrifuge samples.
- 3 Clean the NGS area well, including the Qubit machine, vortex, and small centrifuge (if it is not already there, bring it to the NGS side of the lab bench - the blue one with the plastic lid).
- 4 Get out a number of Qubit tubes (glass cabinet above the NGS workspace) equal to your number of samples + 2 for the standards (N tubes + 2). Label each with an alcohol resistant marker, and write down each sample in your lab notebook (if you are using an abbreviated label, note this as well).

Master Mix

- 5 Each time you use the Qubit, you will need to prepare enough of the master mix for all the samples. Prepare 199 uL Qubit buffer + 1 uL of Qubit reagent * (N samples + 2 standards + 1 extra). For example, if you have 6 samples, you will need 199 uL Qubit buffer + 1 uL of Qubit reagent * (9) for all the tubes. Get out an appropriate number of tubes to fit the master mix and label them. Combine the amount of buffer and reagent necessary, then vortex and spin the master mix.

Making Tubes

- 6 For Standard tubes, pipette: 190 uL of Master Mix + 10 uL of standard into each tube. Do this for each separate standard, switching pipette tips for each one. Vortex and spin once done.
- 7 For DNA tubes: pipette 198 uL of Master Mix + 2 uL of extraction sample. Repeat for each sample, changing pipette tips in between each sample. Vortex and spin tubes once done.

Qubit



- 8 Start up the Qubit machine.
Select the necessary path, which usually is: DNA > dsDNA broadrange. When told to run new standards, click **yes**
- 9 Enter standard 1 when prompted, then run. Enter standard 2 when prompted, then run. Repeat with DNA samples until done.