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# SOP16v1_TGD_Qubit_dsDNA_BR_Assays_for_DNAQuantification

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**Protocol status:** Working
**We use this protocol and it's working**

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## Abstract

**Guidelines**
Bench work should be completed on the **pre-PCR/DNA bench.**
**Blue DNA-only** labeled materials should be used for this protocol to avoid cross-contamination.

Lab attire: gloves, lab coat, and safety glasses/goggles.

**Before Start**
Grab ice.
Use this spreadsheet template to calculate reagent + buffer mix.
Qubit Prep Template - dsDNA BR As…

## Guidelines

Bench work should be completed on the **pre-PCR/DNA bench.**
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## Materials

| | A | B | C |
|---|---|---|---|
| | Material | Catalog | Supplier |
| | Qubit Assay Tubes | Q32856 | Life Technologies |
| | Qubit dsDNA BR Assay Reagent Kit | Q32853 | Life Technologies |
| | Qubit Fluorometer | (Snyder Lab, Rm 2265) | Life Technologies |

## Safety warnings

Waste Disposal: all components can be thrown in the normal trash.

## Before start

Grab ice.
Use this spreadsheet template to calculate reagent + buffer mix.
Qubit Prep Template - dsDNA BR As…

1 Prepare the correct amount of mastermix of Qubit reagent in buffer for your samples and standards.

**Note:** A calibration is required for every usage of the Qubit, so always include standards in the calculations.
Reagent and buffer mastermix is light sensitive. Wrap the tube in foil to protect from light.

1.1 Reagent must be diluted 1:200 in buffer.
Each standard requires 190 uL of mastermix.
Each sample requires 190-199 uL of mastermix. (You can put anywhere from 1 to 10 uL of sample in each qubit assay).

2 Select the reagent calculator on the Qubit Fluorometer main menu.
Ensure that your calculations match up with the numbers calculated by the Qubit for mastermix preparation.

3 Prepare samples: Aliquot the appropriate amount of mastermix (190-199 uL) into each 0.5 mL qubit assay tube. Add the appropriate amount of sample (1-10 uL) into each tube, and let sit for 2 minutes to incubate.

After incubation, the sample fluorescence is stable for 3 hours.

4 Prepare Standards 1 and 2. Each requires 10 uL of standard and 190 uL of mastermix. Keep the standards on ice.

5 Go back to home screen and select the dsDNA assay: Broad Range. Select "Read Standards".

6 Input Standard 1, then read. Then input Standard 2. Generate calibration curve.

7 Enter what sample volume you added to each tube.

8 After reading one tube, just take out the sample and put in the next tube. This way all samples will be saved under the same assay data set.

9 When finished, upload the data onto a USB.