

# AccuGreen Broad Range dsDNA Quantitation Instructions

**ALL BROAD RANGE dsDNA COMPONENTS ARE MARKED WITH ORANGE LAB TAPE.**

1. The AccuGreen Broad Range dsDNA Quantification Solution and assay tubes are in the drawer labeled “Biotium”. The AccuGreen Standard 1 (0 ng/μl) and AccuGreen Standard 2 (100 ng/μl) are in the refrigerator. Both standards are labeled “BR” on the top and marked with orange tape.



2. Warm all components to room temperature before starting.
3. **The AccuGreen Broad Range dsDNA Quantification Solution (dark brown bottle) already contains the dye.** Aliquot enough Quantification Solution (dark brown bottle) for the number of samples and standards you have into the appropriate size tube (5 ml, 15 ml, or 50 ml). Include 10% overage.  
For calculations, assume 199 μl Quantification Solution is used per sample:  
 $[199 \mu\text{l} \times (\# \text{ samples} + 2 \text{ standards})] \times 1.10 = \text{volume of Quantification Solution needed}$
4. Label assay tubes. You will have one tube for each sample, one tube for standard 1, and one tube for standard 2.
5. For Standard 1 (0 ng/μl) and Standard 2 (100 ng/μl), add 190 μl of Quantification Solution to each tube. Then add 10 μl of the respective standard to each tube.
6. For each sample, add 199 μl of Quantification Solution to the assay tube. Then add 1 μl of sample.
7. Mix samples thoroughly by vortexing.
8. Incubate samples at room temperature for 2 minutes. Use timer that is available.
9. On the Qubit instrument, press **GO** to turn on. Use the arrows to navigate to **Quant-iT dsDNA, BR** then press **GO**.
10. Run a new calibration following the instructions on the screen.
11. Insert sample assay tube into Qubit and close the lid. Press **GO** to measure the sample. Arrow down to **Calculate sample concentration**, press **GO**, use the down arrow to select the volume of sample added to the assay tube (typically 1 μl), then press **GO**. The concentration of your sample will be displayed on the screen. Manually record the sample concentration.
12. Repeat step 11 until you have measured the concentration of all samples.
13. When finished, turn off the Qubit. (Press **HOME**, use the arrows to navigate to **Power OFF**, then press **GO**).
14. Record your Qubit usage on the Qubit log on the table by the door. Include the standards with the number of samples that you measured.