

Measuring ssDNA Concentration Using the Quantus™ Fluorometer with the QuantiFluor® ssDNA System

Promega Corporation



Materials Required

- QuantiFluor® ssDNA System (Cat.# E3190)
- Quantus™ Fluorometer (Cat.# E6150)
- 0.5ml PCR Tubes (Cat.# E4941)
- shrimp DNase (USB Cat.# 78314)

Caution: We recommend the use of gloves, lab coats and eye protection when working with these or any chemical reagents.

Protocols: *Quantus™ Fluorometer Operating Manual* #TM396 and *QuantiFluor® ssDNA System Technical Manual* #TM376 are available at: www.promega.com/protocols/

Detecting and quantitating small amounts of single-stranded (ssDNA) is an important step in many molecular biology techniques, including DNA sequencing, site-directed mutagenesis, DNA amplification and gene expression. Traditional spectrophotometric assays cannot determine DNA concentrations below 2µg/ml; however, many isolated DNA samples have concentrations well below that level.

Using the Quantus™ Fluorometer (Cat.# E6150) with the QuantiFluor® ssDNA System (Cat.# E3190) provides a fast, easy and sensitive method for determining DNA concentration. Using this system, we have detected sample concentrations as low as at 200pg/µl when using 1µl of sample input per assay. It is possible to quantitate more dilute samples by adding more sample per assay. Up to 100µl of sample may be measured per 200µl assay.

The QuantiFluor® ssDNA System contains a fluorescent DNA-binding dye that enables sensitive and specific quantitation of small amounts of ssDNA in solution. For ssDNA samples that may contain contaminating double-stranded DNA (dsDNA), we recommend a brief shrimp DNase treatment to degrade any dsDNA present and ensure the most accurate ssDNA quantitation.

This Application Note describes the protocol for using the QuantiFluor® ssDNA Dye System with the Quantus™ Fluorometer. The Quantus™ Fluorometer measures sample volumes as little as 1µl in a 200µl assay volume without sacrificing instrument sensitivity.

For the complete protocol, see the *QuantiFluor® ssDNA System Technical Manual* #TM376.

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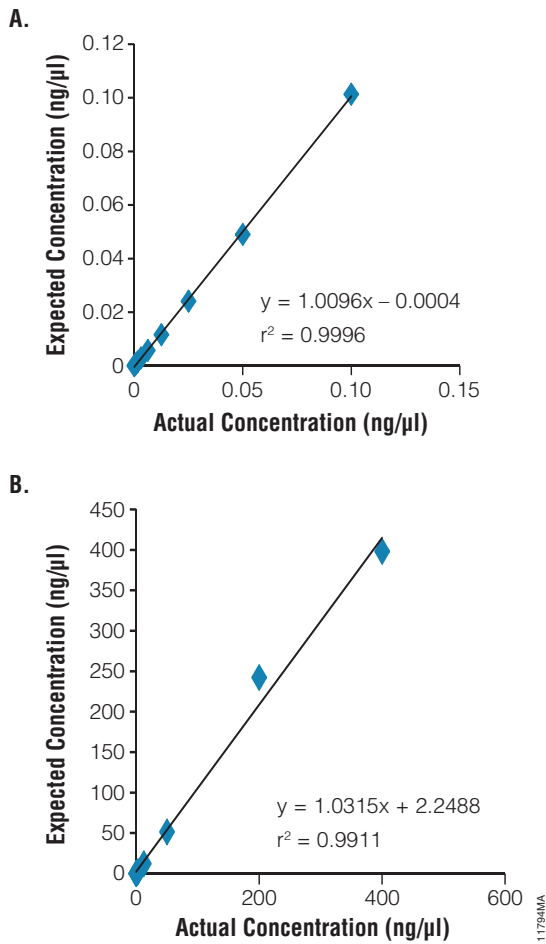


Figure 1. Measuring ssDNA concentration using the QuantiFluor® ssDNA System and the Quantus™ Fluorometer. Panel A. Assay linearity using the low-concentration standard curve. **Panel B.** Assay linearity using the high-concentration standard curve. The values presented correspond to assay tube concentration and not starting sample concentration.

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