Accuracy vs. Reference Spectrophotometer

Introduction

The concentration accuracy across the entire dynamic range of the Thermo Scientific NanoDrop 2000 Spectrophotometer was evaluated by comparing nucleic acid samples measured on a NanoDrop™ 2000 and an Agilent 8453 Spectrophotometer fitted with a 2 mm pathlength quartz micro-cuvette and a 0.2 mm Implen cell.

Method

All samples measured on the NanoDrop 2000 were measured without dilution.

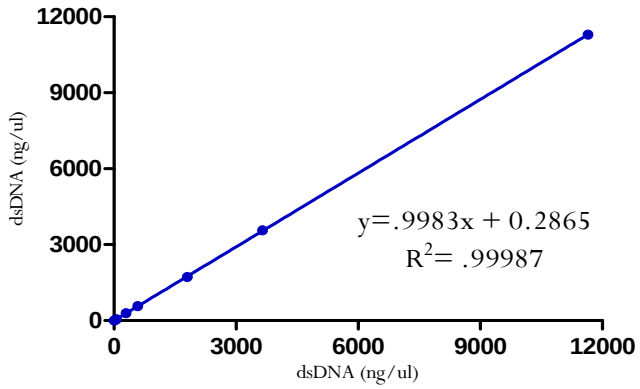
On the reference spectrophotometer a 2.0 mm quartz cell was used to measure the reference concentrations from 6 ng/uL to 295 ng/uL. A specialized cell capable of measuring samples at a 0.2 mm pathlength was used to measure the dsDNA concentrations from 580 ng/uL to 3650 ng/uL. The 11650 ng/uL sample was diluted 1:10 and measured with same specialized cell as above at a 0.2 mm pathlength.

DNA concentrations (ng/ul) for the Agilent 8453 were calculated by first multiplying the respective A260 nm value by 5, and then applying Beer's Law. This dilution factor is required to account for the 2 mm path. The 0.2 mm cell values were multiplied by 50.

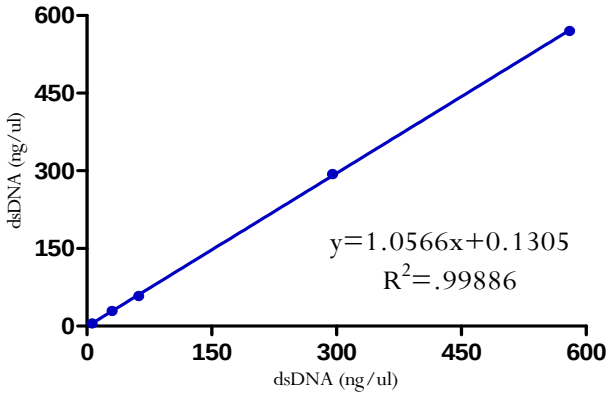
The average dsDNA concentration values obtained from the NanoDrop 2000 and the Agilent 8453 are listed in the below. All data shown for the NanoDrop 2000 Spectrophotometer was automatically normalized to a 10 mm pathlength .

Agilent 8453 dsDNA ng/ul	NanoDrop 2000 DNA ng/ul
6	5.3
30	29.3
62	58.4
295	293.6
580	570.1
1800	1720.8
3650	3565.7
11650	11299.2

NanoDrop 2000 vs Agilent 8453 Full Concentration Range



NanoDrop 2000 vs Agilent 8453 Low Concentration Range



The automatic path length capability of the NanoDrop 2000 spectrophotometer allowed for measurement of samples across the full concentration dynamic change using the instrument's standard method, with no dilutions and no special sample preparation required. By contrast, two different cells were necessary for the Agilent 8453 Spectrophotometer to measure samples across this broad concentration range. In addition, dilution of the sample was necessary to measure the highest concentration on the reference spectrophotometer.