

Alexa 555 Assay Linearity, Reproducibility and Sensitivity

Introduction

The family of Alexa fluors and Cy dyes are some of the most popular fluorophores used for microarray and fluorescence *in-situ* hybridization (FISH) experiments. The Thermo Scientific NanoDrop™ 3300 Fluorospectrometer can measure as little as 1 ul of sample, thereby significantly scaling-down the reaction volumes commonly needed for conventional cuvette-based fluorometers. This micro-volume capability of the NanoDrop 3300 allows the researcher to run quality control checks for a variety of reagents labeled with Alexa and Cy dyes. In addition, the versatility of the white LED allows for the simultaneous measurement of multiple fluorophores.

Method

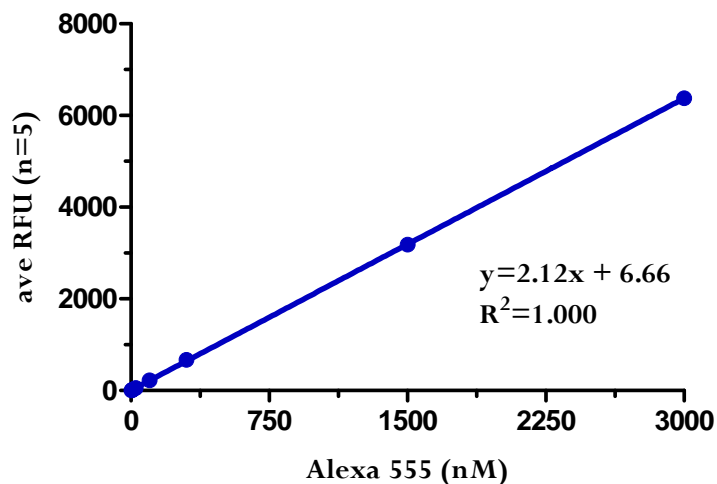
Serially diluted Alexa 555 was measured using the white LED excitation source (500-680nm) with the emission wavelength monitored at 565nm.

Results

Femtomoles Alexa 555 per assay	Alexa 555 (nM)	Ave RFU (n=5)	Stdev	%CV
5	2.5	5	0.6	11.5
10	5	12	1.1	9.2
16	8	19	1.2	6.4
20	10	23	0.8	3.3
25	13	31	2.5	8.3
50	25	58	1.4	2.5
200	100	226	1.6	0.7
600	300	668	7.3	1.1
3000	1500	3186	90.1	2.8
6000	3000	6374	82.7	1.3

Sensitivity is ~ 5 femtomoles or 2.5nM in this data set.
Linearity demonstrated between 2.5 nM to 3000 nM.

Alexa 555 Linearity on the
NanoDrop 3300



Alexa 555 Linearity on the
NanoDrop 3300
Low Concentrations

