

## Alexa 647 Assay Linearity, Reproducibility and Sensitivity

### Introduction

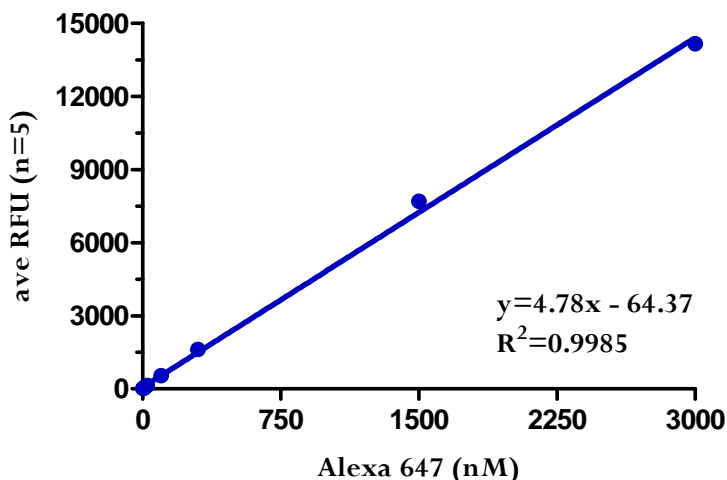
The family of Alexa fluors and Cy dyes are some of the most popular fluorophores used for microarray and fluorescence *in-situ* hybridization (FISH) experiments. The NanoDrop 3300 Fluorospectrometer can measure as little as 1 ul of sample, thereby significantly scaling-down the reaction volumes commonly needed for conventional cuvette-based fluorometers. This micro-volume capability of the Thermo Scientific NanoDrop™ 3300 Fluorospectrometer 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

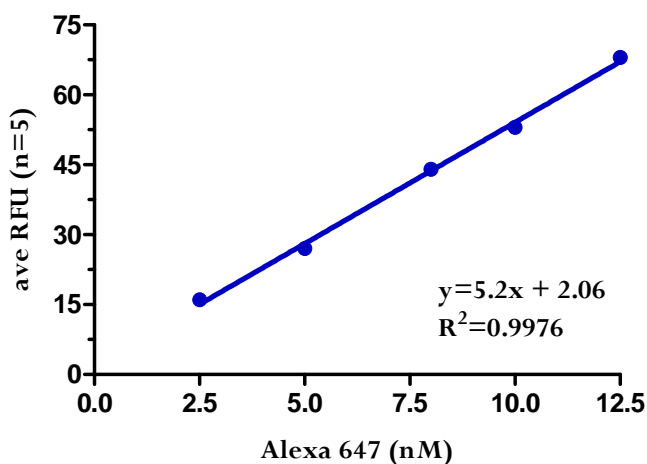
Serially diluted Alexa 647 was measured using the white LED excitation source (500-680nm) with the emission wavelength monitored at 680 nm.

Femtomoles Alexa 647 per assay	Alexa 647 (nM)	Ave RFU (n=5)	Stdev	%CV
5	2.5	16	0.6	3.9
10	5	27	0.8	2.9
16	8	44	0.9	2.2
20	10	53	2.1	4.0
25	12.5	68	0.6	0.8
50	25	141	5.7	4.0
200	100	540	9.6	1.8
600	300	1619	18.7	1.2
3000	1500	7696	154.4	2.0
6000	3000	14162	103.6	0.7

Alexa 647 Linearity on the  
NanoDrop 3300



Alexa 647 Linearity on the  
NanoDrop 3300  
Low Concentration Range



Sensitivity is ~ 5 femtomoles or 2.5nM in this data set.