

Documentation No. 2

Statistics of fluorescence lifetime measurement with LF502 NanoScan FLT

Aim of the experiment:

- demonstration of the precision of fluorescence lifetime (FLT) measurements with LF502 NanoScan FLT
- comparison of fluorescence intensity read-out and fluorescence lifetime read-out



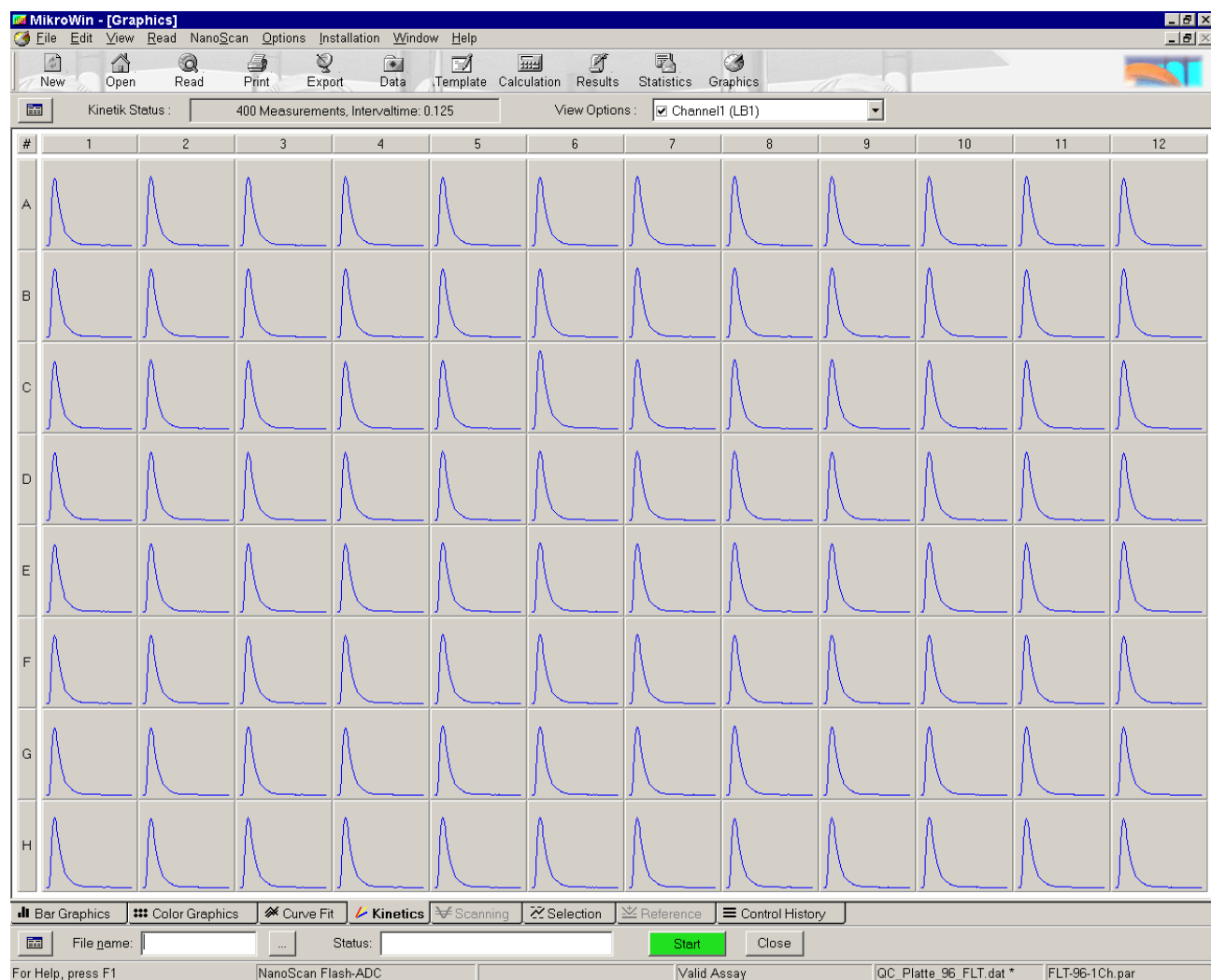
Measurement:

Measurement of a QC microplate (96-well-overlay) containing a homogeneous, light blue fluorescent standard material.

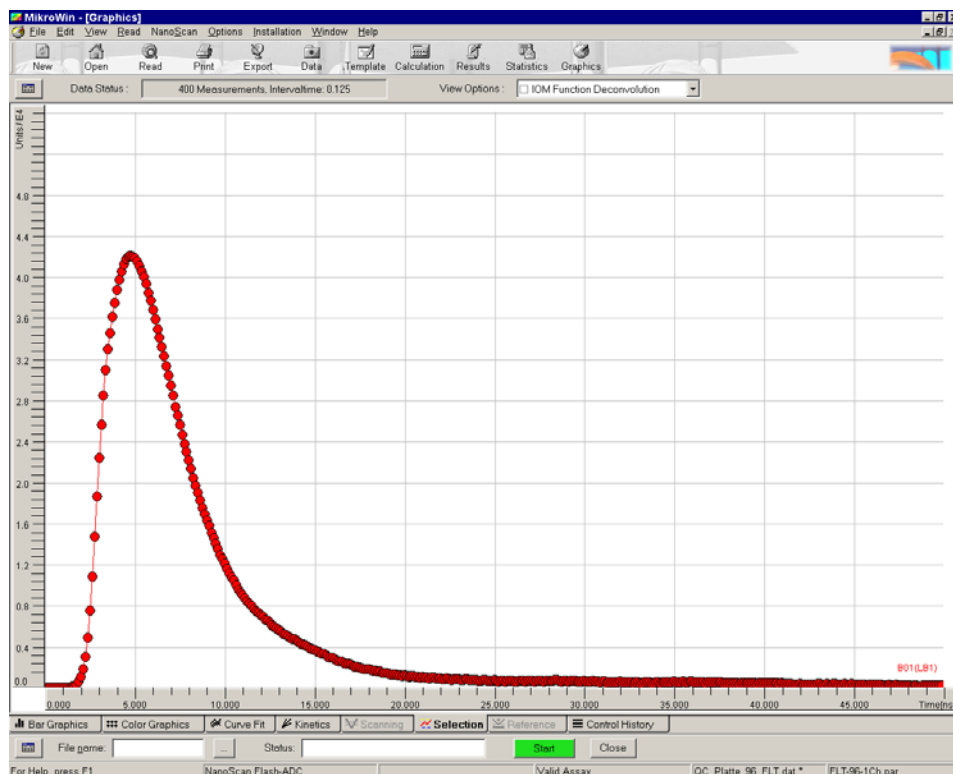
laser module: 488 nm
 detection wavelength: 520 nm
 laser frequency: 30 Hz
 laser energy: 15 %
 sensitivity: reduced gain
 averaging: 16 laser pulses
 detection window: 50 ns
 time resolution: 0,125 ns

Results:

overview of all 96 fluorescence decay curves:



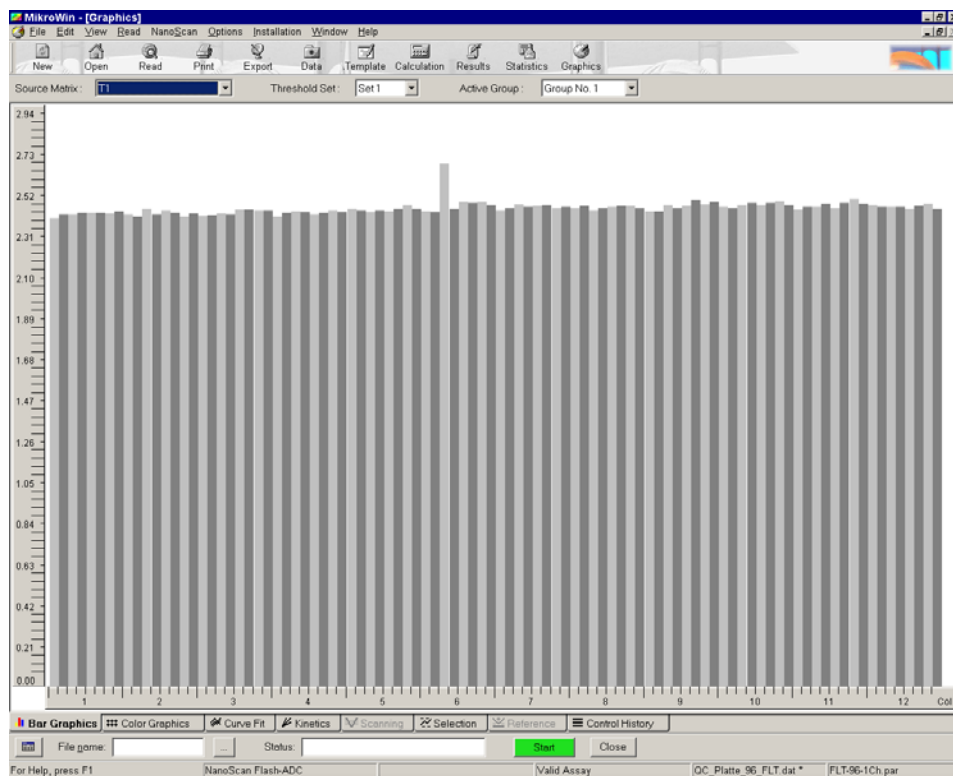
selection of one decay curve (B01) for enlarged display



data analysis

graphical display of single-exponential FLT analysis for all 96 wells

Fit Range: 2 .. 50 ns
 number of exponentials: 1
 determination of start values: automatically



As can be seen from the diagram, the result of one data point (C06) reproducibly differs from all others. The source of the error is presumably a piece of dust on the plate's surface.

Statistics

	fluorescence intensity [rfu]	fluorescence lifetime [ns]
average of all 96 wells	278.887	2,446
standard deviation of all 96 wells	6.957	0,031
cv of all 96 wells	2,49%	1,29%
average without well C06	278.278	2,444
standard deviation without well C06	3.642	0,021
cv without C06	1,31%	0,87%

Conclusion:

- The LF502 NanoScan FLT enables measurements of fluorescence lifetimes with a precision of as low as 20 .. 30 ps.
- The cv value for the FLT measurement is reproducibly below 1%.
- The measurement of fluorescence lifetimes is proven to be more precise than the measurement of fluorescence intensities.