Supporting Information

Systematic Study of Fluorescein-Functionalized Macrophotoinitiators for Colorimetric Bioassays

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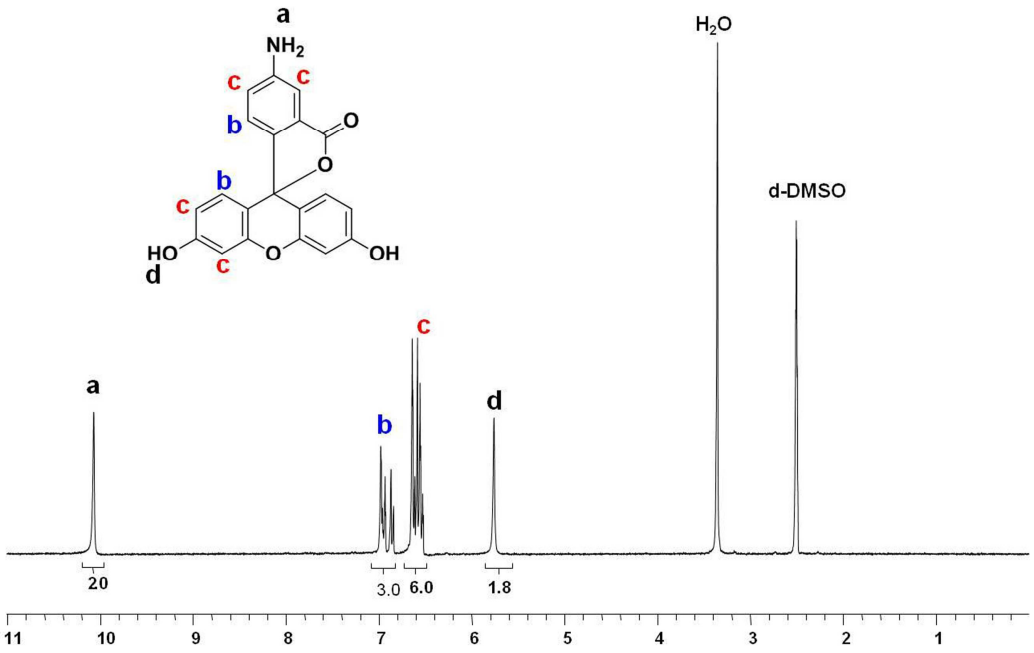
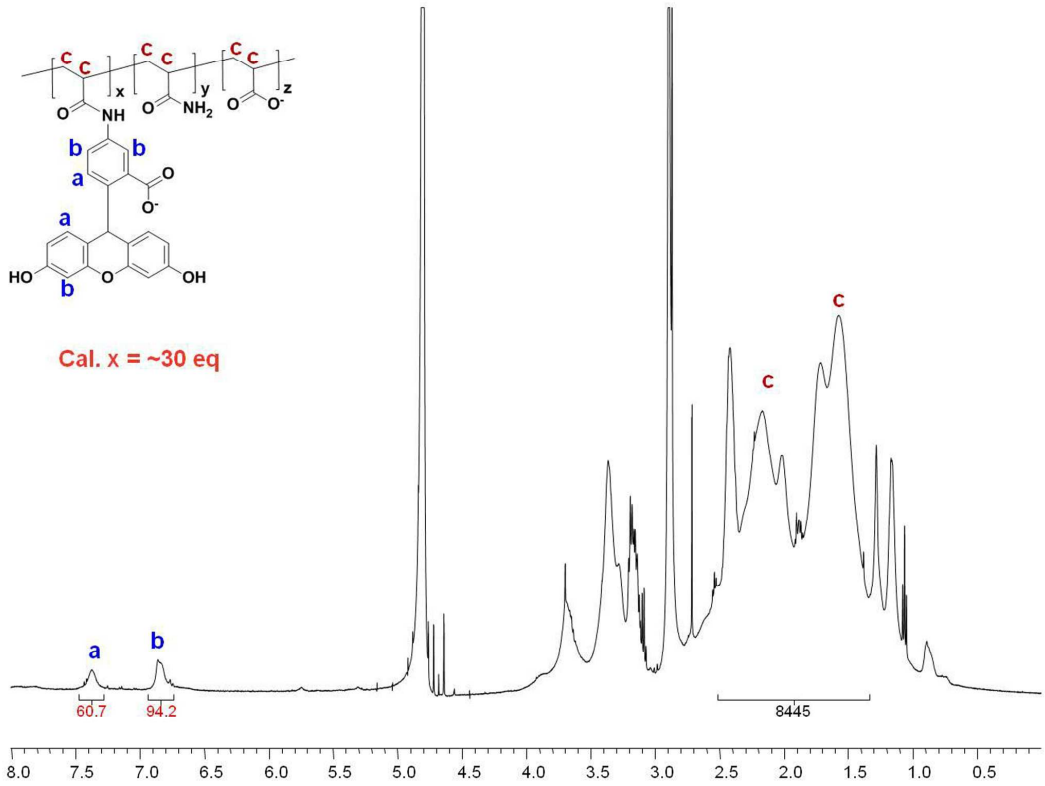
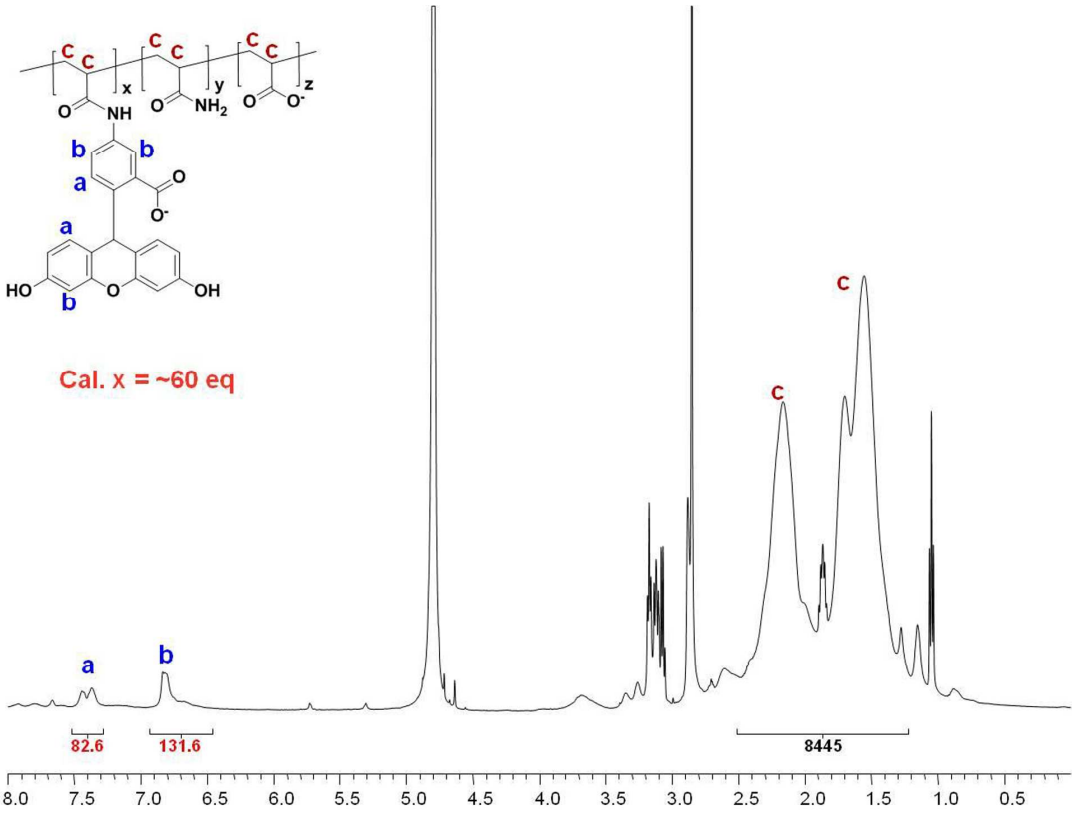
List of Supporting Figures.

Figure S1. [1]H NMR spectra and peak assignment of fluoresceinamine and compound **2-6**.[a]



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a
b
c
82.6
131.6
8445
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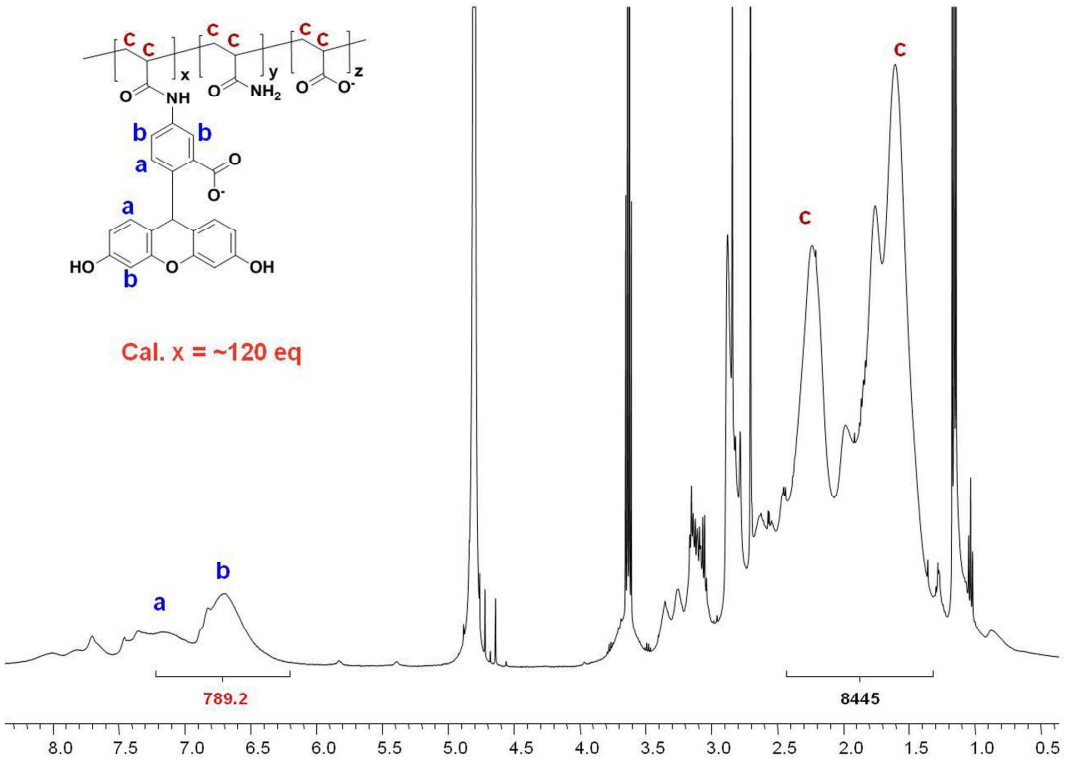
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b
c
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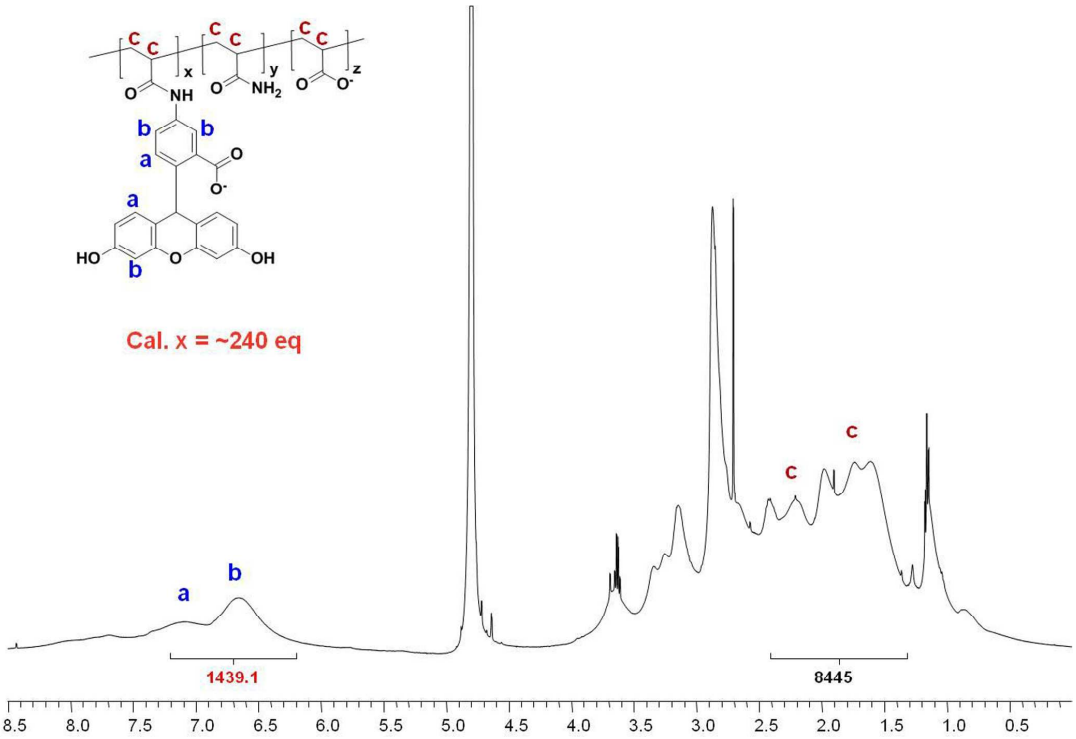
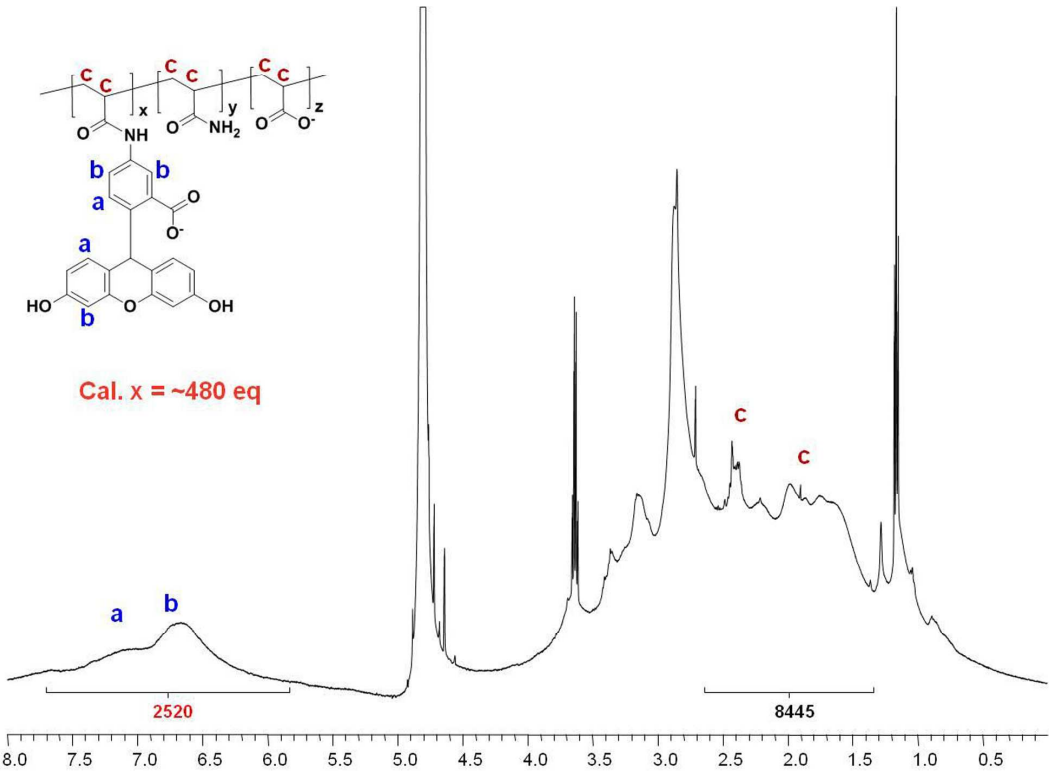
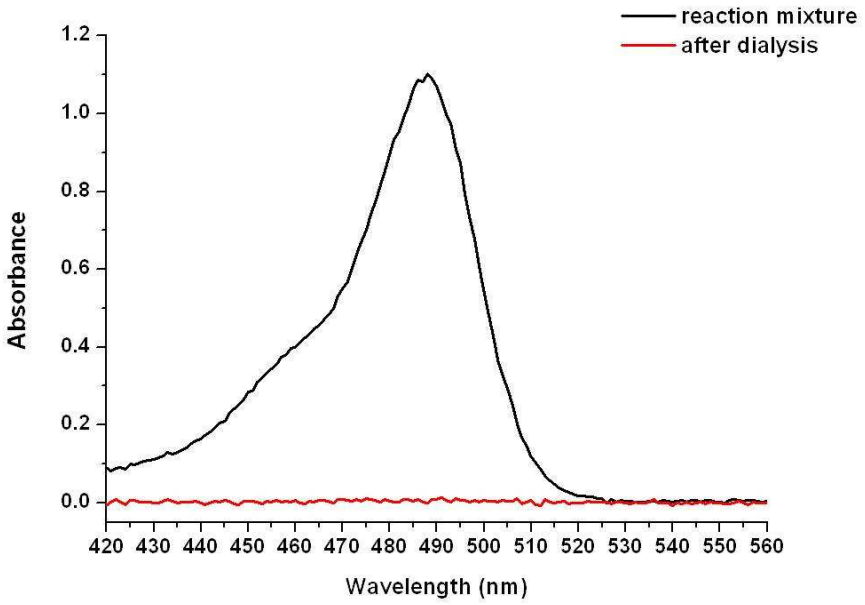
8445
8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5





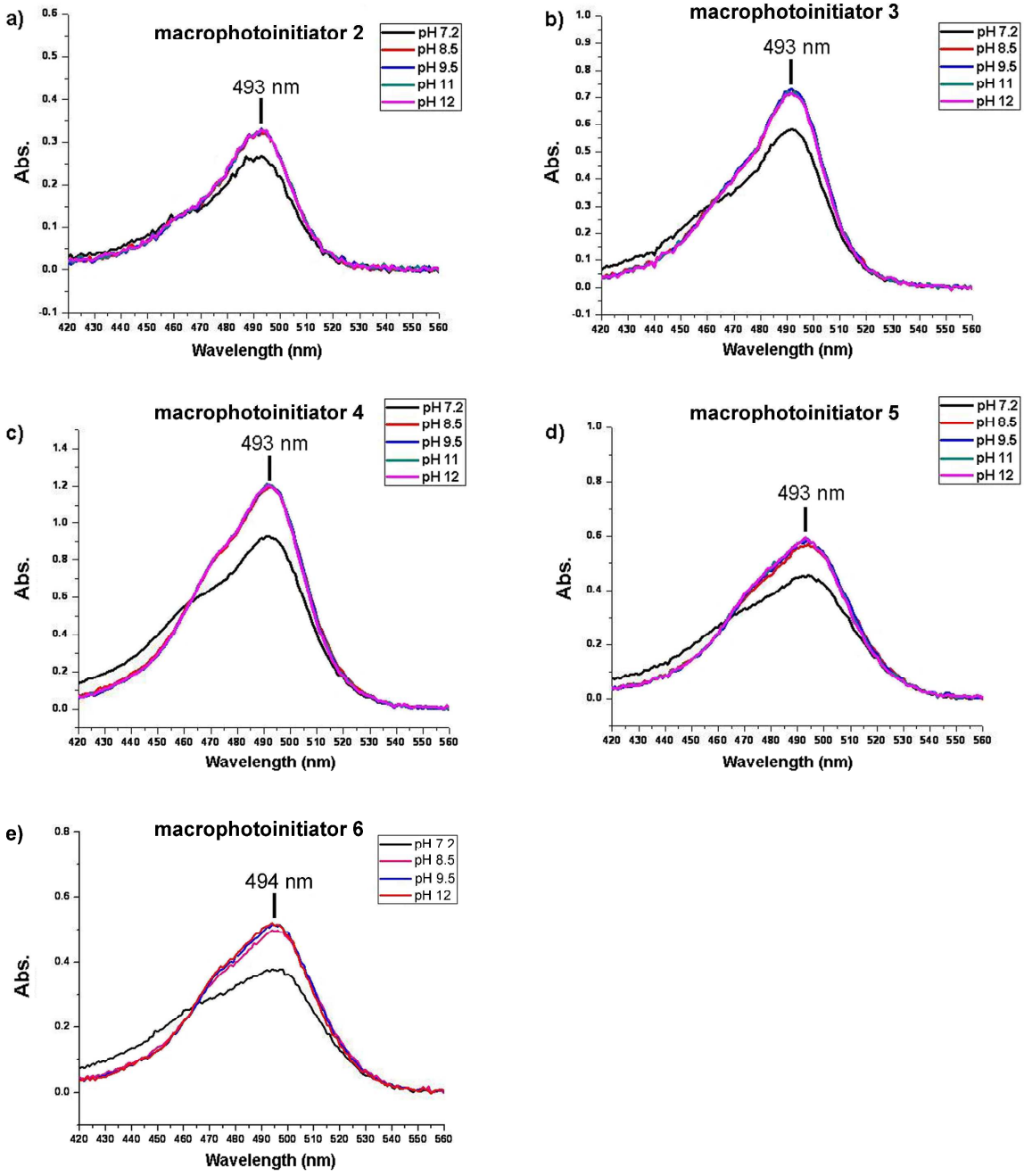
[a] Due to the solubility, fluoresceinamine and macrophotoinitiators **2-6** were measured with DMSO-d6 and D_2O, respectively.

Figure S2. UV/Vis absorption spectra of a physical blend of fluoresceinamine and copolymer **1** before and after dialysis.[a]



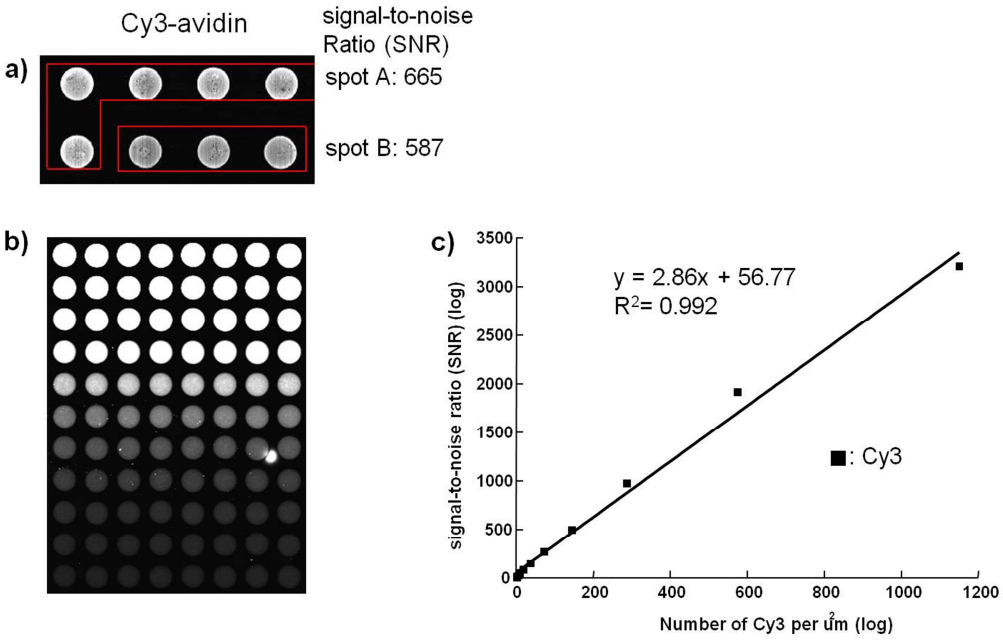
[a]Physical blend of copolymer (50 nmol, 10 mg) and fluoresceinamine (24 μmol, 8.34 mg) was dissolved in a mixture of DMSO (0.5 mL) and pH 8.0 buffer solution (1.5 mL), and then was stirred at 35°C for 2 days. The dialysis was carried out as described in the Methods section. Before and after dialysis, the same volume concentration of the resulting mixtures was used to measure UV/Vis absorbance.

Figure S3. UV/Vis absorbance spectra of macrophotoinitiator **2-6** under varying pH conditions.[a]



[a]As pH was increased, macrophotoinitiators **2-7** exhibited a hyperchromatic effect.

Figure S4. Determination of the density of biotin-DNA molecules on test surfaces by fluorescence intensity.



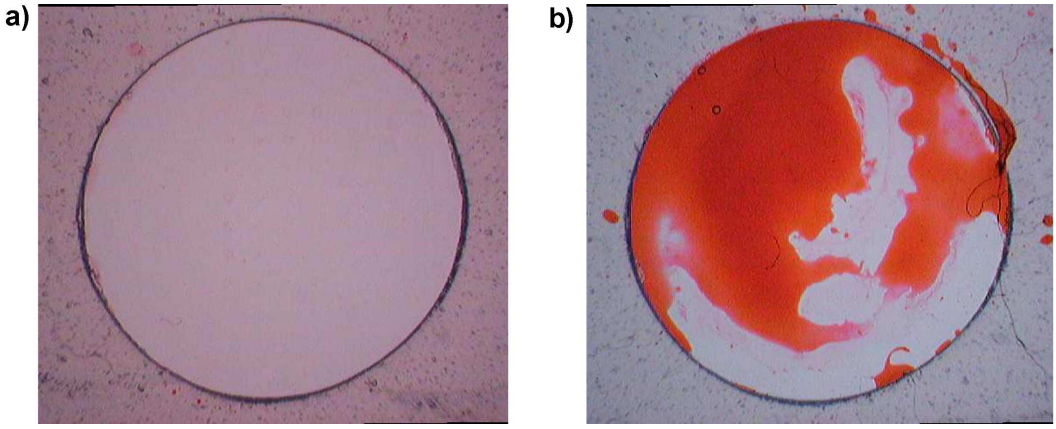
a) Fluorescence image of streptavidin-$(Cy3)_2$ bound on the test surfaces and average fluorescence intensity of each spot. b) Florescence image of a Cy3 calibration chip (Full Moon Biosystems), and c) its standard curve; black squares indicated the signal-to-noise ratio against 11 different numbers of Cy3 per square micrometer. The number of Cy3 on streptavidin was determined by following equation: $n_{cy3}/n_{sa} = (Abs_{cy3,552}/ \varepsilon_{cy3,552})/[(Abs_{sa,280}-Abs_{cy3,280}-Abs_{1\%BSA,280})/\varepsilon_{sa,280}]$, and $\varepsilon_{cy3,552}$ = 150,000 $M^{-1}cm^{-1}$, $\varepsilon_{cy3,280}$ = 12,000 $M^{-1}cm^{-1}$, $\varepsilon_{BSA,280}$ = 43824 $M^{-1}cm^{-1}$, $\varepsilon_{sa,280}$ = 173,000 $M^{-1}cm^{-1}$.[1~3] SNR= [(Average of signal intensity)- (Average of background intensity)]/ (standard deviation of background intensity).

1. http://www.nlv.ch/Molbiology/sites/Fluorescence1.htm

2. http://www.piercenet.com/files/TR0006-Extinction-coefficients.pdf

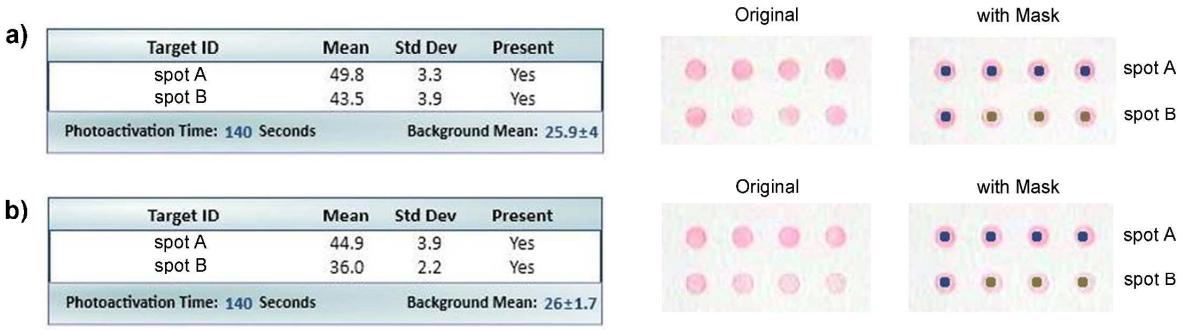
3. http://medicine.yale.edu/labs/henegariu/www/tavi/FISHdyes.html

Figure S5. Stained chip images after the photopolymerization using neutravidin/macrophotoinitiator **11** with varying irradiation time.[a]



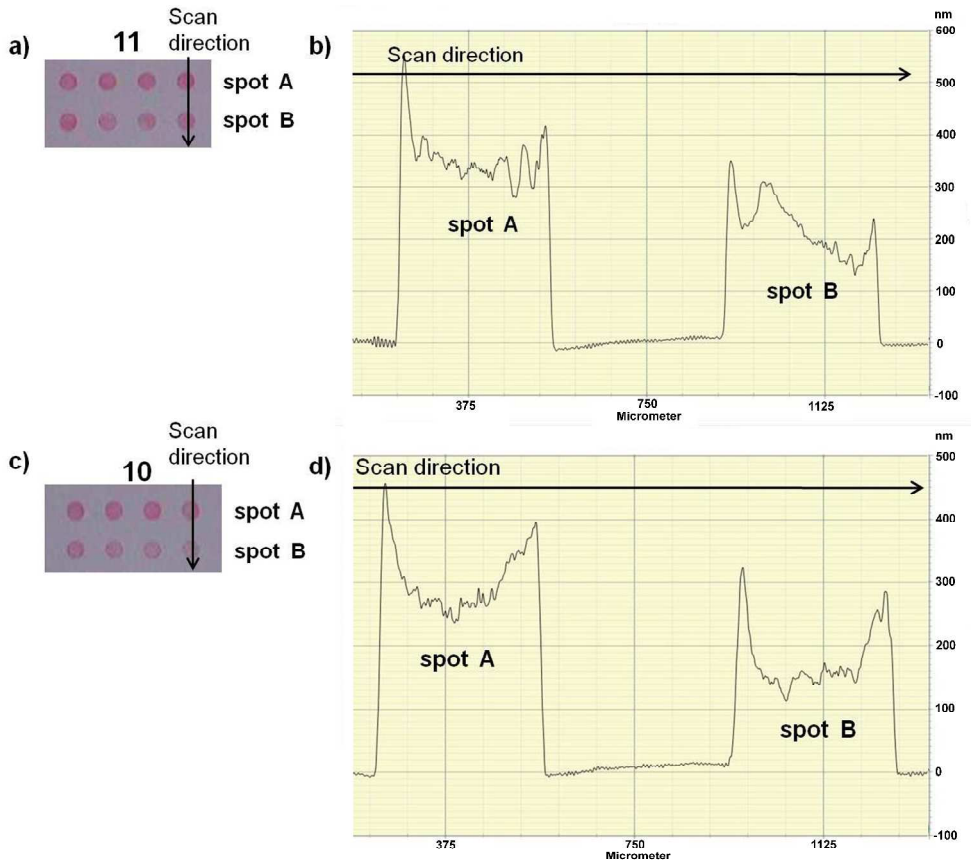
[a]The raw images of (a) 100 sec of irradiation; no polymerization detected, and (b) >190 sec of irradiation; hydrogel was excessively grown on the whole areas of the chip (bulk polymerization).

Figure S6. Analysis of color intensity after the photopolymerization of macrophotoinitiator **10** and **11** at 140 sec of irradiation, followed by the staining process.[a]



[a] Mean intensity for each row was obtained by the original image with mask, where the software calculated the mean intensity from the spots on each row; a) macrophotoinitiator **11** and b) macrophotoinitiator **10** was measured by three different areas on each row..

Figure S7. The images of surface profile after the photopolymerization using neutravidin/macrophotoinitiator **10** and **11** at 140 sec of irradiation.[a]



[a] The thickness of resulting polymer films were measured by Dektak 150 profilometer; a) image of a stained chip after photopolymerization with **11**, and b) the image of surface profile of a), c) image of a stained chip after photopolymerization with **10**, and d) the image of surface profile of c). The black arrow indicated a direction of the scanning target spots.