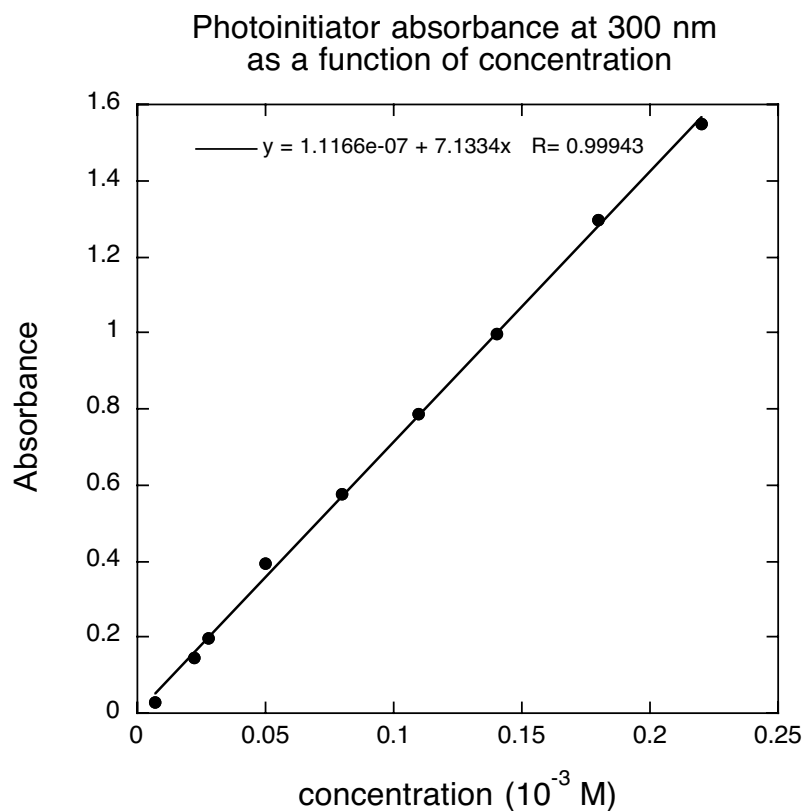


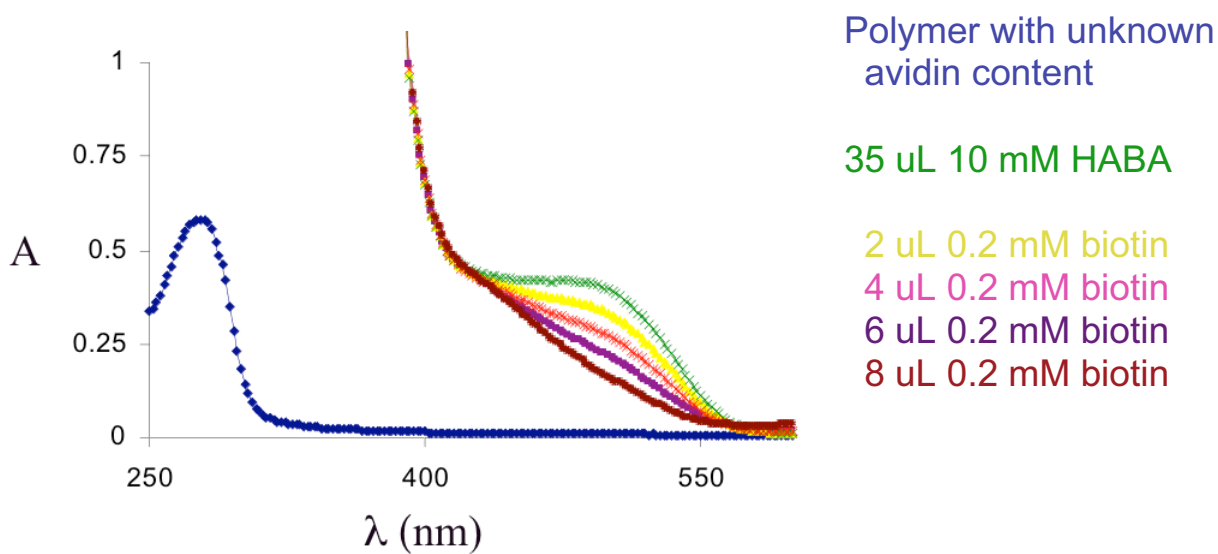
Supplementary Information

Macroinitiator characterization:

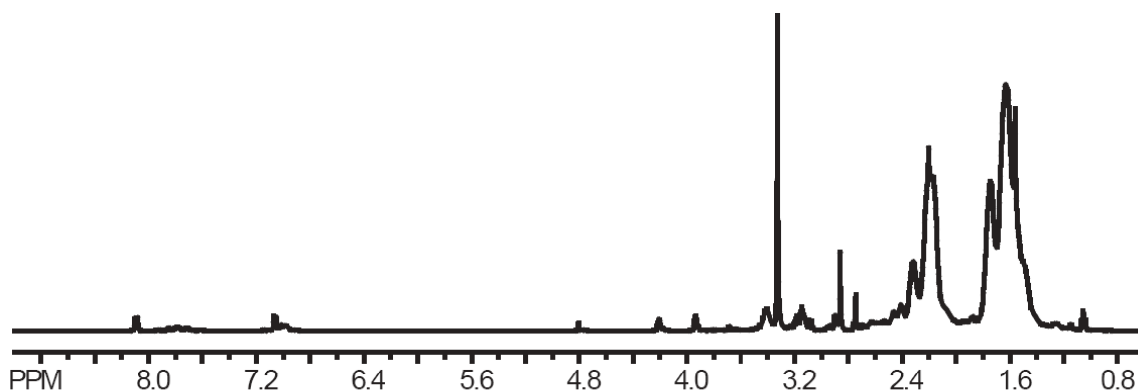
A standard curve of initiator absorbance as a function of concentration was prepared. The solvent was MES buffer at pH 6 (0.1 M MES, 0.5 M NaCl). Absorbance measurements of purified macroinitiator solutions (100 $\mu\text{g/ml}$ in MES buffer) were made, and the standard curve was used to determine the concentration of the initiator in the unknown product solution. The measured absorbance value was 0.7 for 0.5 ml of 100 $\mu\text{g/ml}$ of the macroinitiator.



HABA assays (described in Ref. 25) were used to determine the average number of Neutravidin substituents and verify retention of biotin-binding capability. HABA forms a complex with avidin that absorbs at 500 nm. Biotin displaces HABA due to the stronger affinity of the biotin-avidin interaction and as a result the absorbance at 500 nm decreases. The amount of biotin that had to be added to a solution of the purified macroinitiator (in MES buffer at pH 6) in order to make the shoulder in the absorbance spectrum disappear allowed calculation of the amount of avidin that was covalently attached to the macroinitiator.



^1H NMR spectra were collected using samples comprised of 20 mg of the macroinitiator dissolved in 0.7 ml of deuterium oxide. The water peak was pre-saturated for collection of the spectra.



NMR analysis requires a larger scale synthesis (60-fold compared with the description in the Methods section), and a methanol precipitation step was more practical in this case than purification with molecular weight cutoff (MWCO) filters. MWCO filters performed well for solutions of this product with concentrations of 1 mg/ml or less, but they did not provide good separation with higher product concentrations.

Note regarding macroinitiator design:

Commercial photoinitiators are commonly used as ~1% by weight additives in bulk polymerizations, a concentration many orders of magnitude higher than could be expected in this new application, particularly near the desired detection limit. Their UV extinction coefficients are sufficiently low that one-initiator-per-binding-event molecules were unlikely to result in large degrees of polymerization. In addition, the polymerization rate is proportional to the square root of the initiator concentration. As a result of these two considerations, we opted to synthesize macromolecules that contained a high ratio of photoinitiator substituents to recognition substituents.