SUPPLEMENTARY INFORMATION:

Multiplexed Enrichment and Detection of Malarial Biomarkers using a Stimuli-Responsive Iron Oxide and Gold Nanoparticle Reagent System

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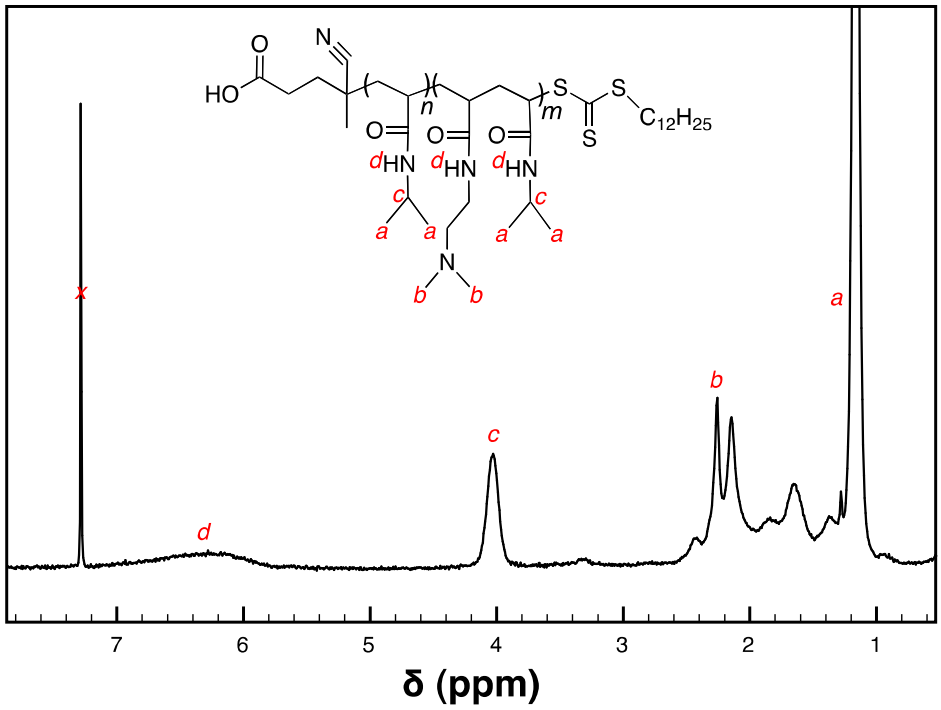
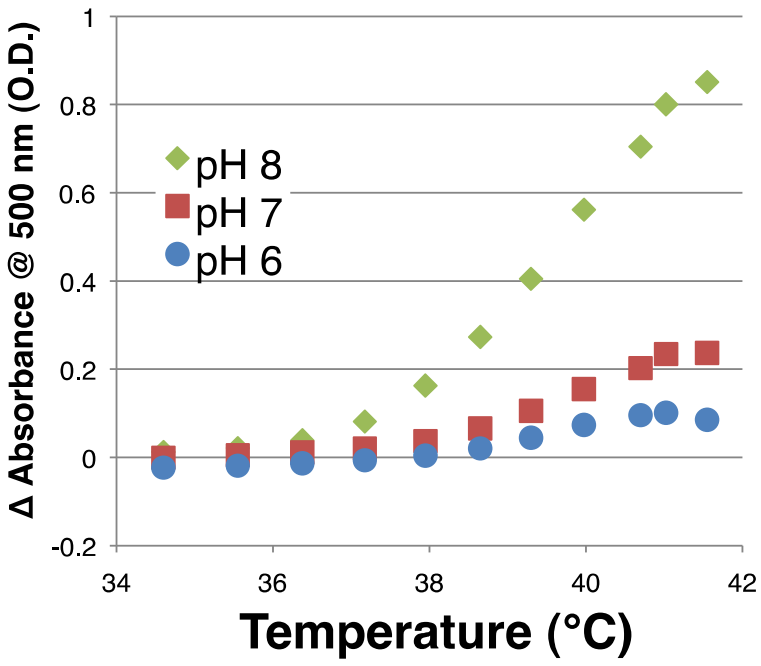
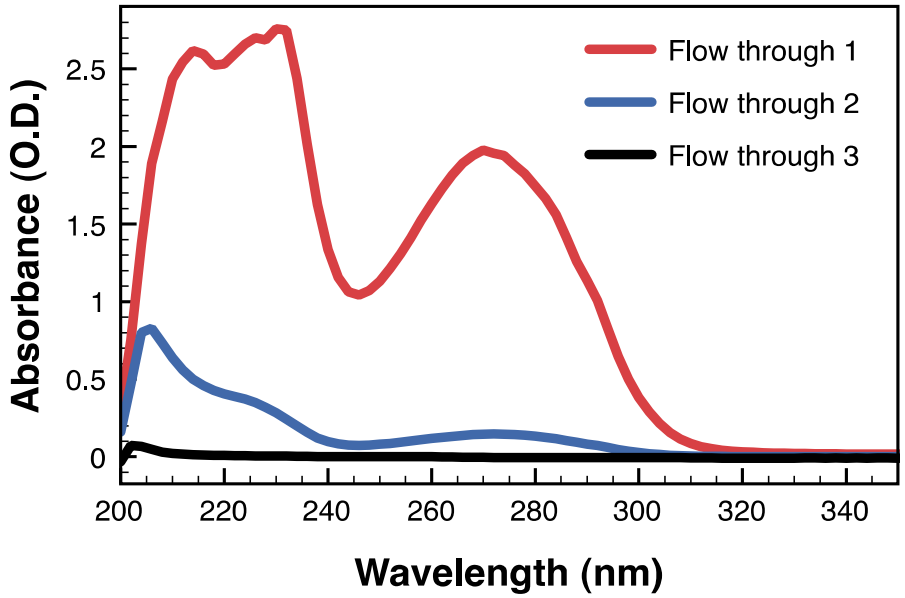


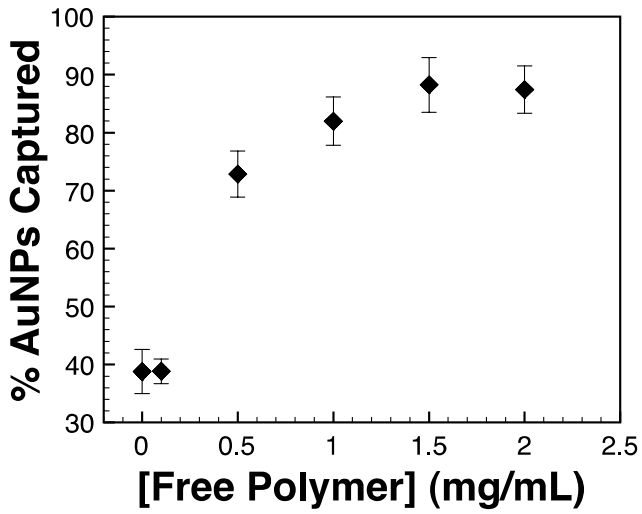
Supplementary Figure 1. Polymer NMR spectrum. The purified diblock copolymer in DI water was treated with 10 equivalents of NaOH to deprotonate the amine groups prior to lyophilization and analysis by ^{1}H-NMR (300 MHz., $CDCl_3$).



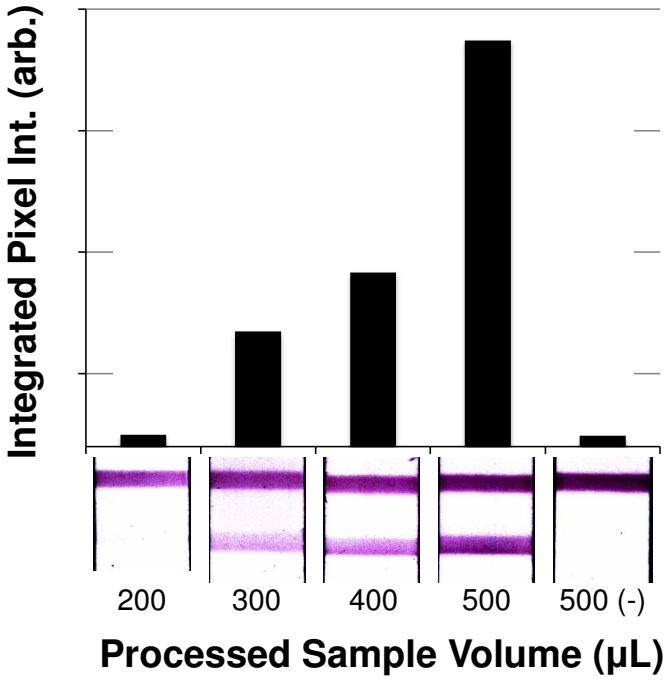
Supplementary Figure 2. Thermal aggregation profile of the diblock copolymer (2 mg/mL in PBS) exhibits pH-dependence due to amine groups. Light scattering (turbidity) was monitored as the sample temperature was ramped from room temperature to 45 ºC over six minutes. Protonation of the amine groups in the polymer at pH 6 and 7 resulted in electrostatic repulsions that prevented strong aggregation and light scattering, as was seen at pH 8.



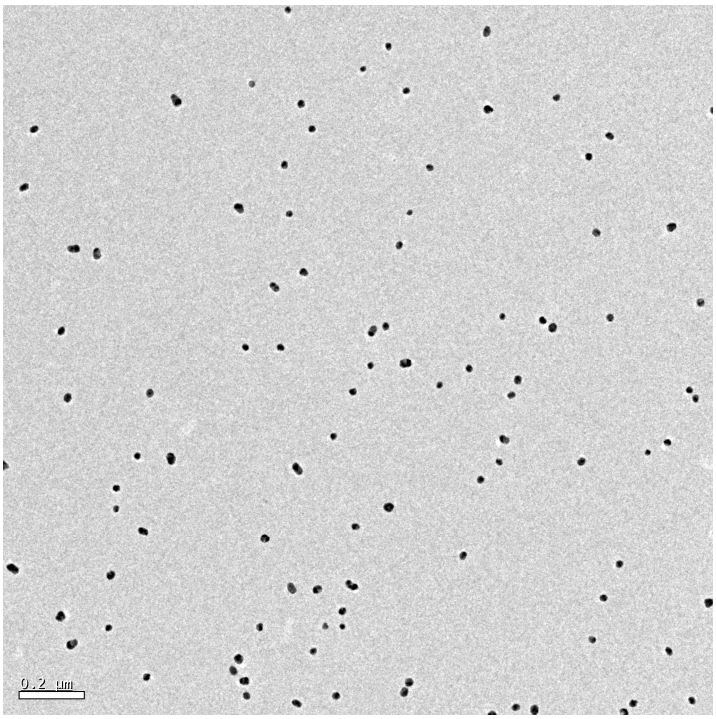
Supplementary Figure 3. Removal of non-conjugated streptavidin from SA-AuNPs. After conjugation of the polymer-modified AuNPs to streptavidin, the non-conjugated streptavidin was removed by membrane ultrafiltration. The UV absorbance at 280 nm was monitored during each round of ultrafiltration to confirm removal of the non-conjugated protein.



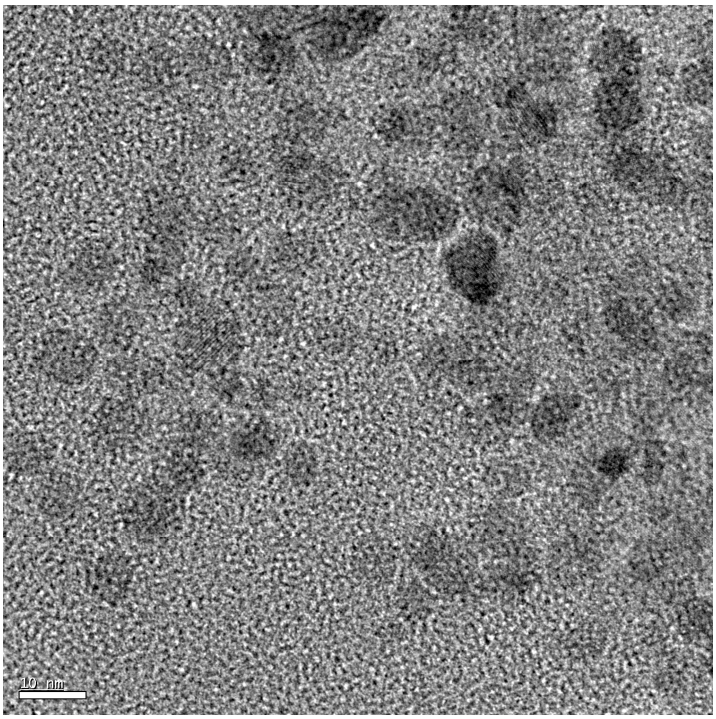
Supplementary Figure 4. Effect of 10 kDa homo-pNIPAm free polymer on AuNP capture efficiency (mean ± standard deviation, n=5). The optimum value of 1.5 mg/mL free polymer was used in further magnetic enrichment lateral flow immunoassay protocols.



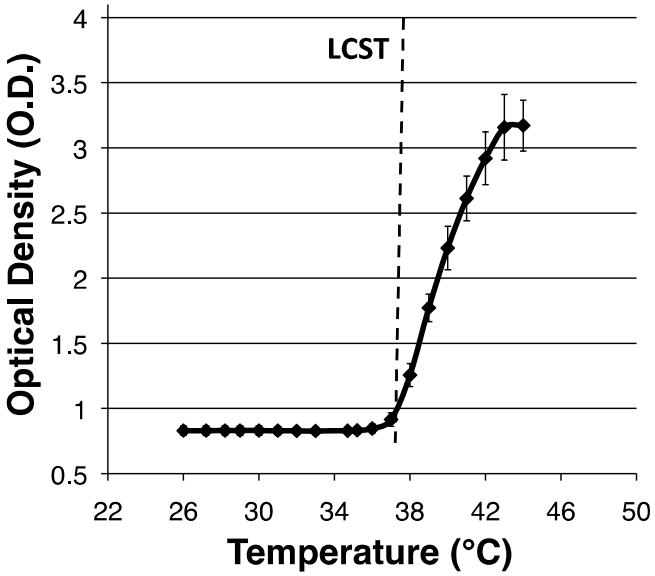
Supplementary Figure 5. (Top) Stepwise/incremental increases in signal for the clinical sample processed at increasing volumes from 200 to 500 µL. (Bottom) Corresponding flow strip images. A negative control at the highest volume processed (500 (-)) showed low background noise.



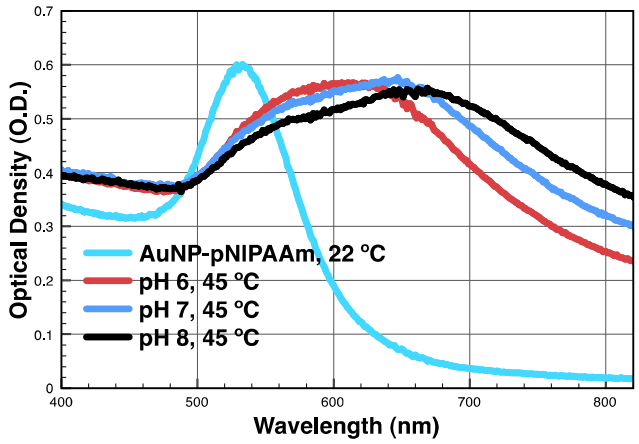
Supplementary Figure 6. Transmission electron micrograph (200 kV) of citrate-reduced AuNPs. The AuNPs were aerosolized onto carbon stabilized formvar coated copper grids (Ted Pella) prior to analysis. Scale bar=200 nm.



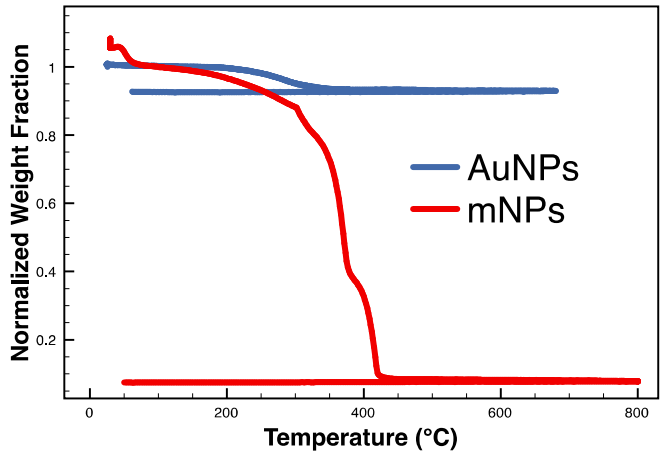
Supplementary Figure 7. Transmission electron micrograph (200 kv) of pNIPAm-mNPs. Scale bar = 10 nm.



Supplementary Figure 8. Phase separation of mNPs. The phase transition temperature of pNIPAm-mNPs dissolved at 2.5 mg/mL in PBS 7.4 was monitored using spectrophotometry. The solution temperature was raised at a rate of 1.2°/min (λ=600 nm).



Supplementary Figure 9. pH response of AuNPs above the transition temperature. The diblock-pNIPAm-modified AuNPs (PBS buffer pH 6, 7, or 8) were heated above the phase transition temperature of the polymer. Deprotonation of the tertiary amines upon raising the pH above the pKa (□8.0) resulted in increased nearfield plasmonic coupling at pH 8. Heated spectra were measured after a fixed heating time of 8.3 minutes at 45 ℃.



Supplementary Figure 10. Thermogravimetric analysis of the mNPs and AuNPs. The diblock modified AuNPs were found to be 7% organic content by weight. The mNPs were found to be 92.6% organic content by weight.