Supporting Information

Dynamic Light Scattering

Changes in the size of the aptamer-conjugated AuNPs upon binding to the viruses were observed by dynamic light scattering (DLS). Changes in the size of scattering particles using DLS when the AuNP.virus complexes are formed has previously been reported by Driskell *et al.* using antibodies conjugated to 30, 60, 80nm AuNPs.[1] For a direct comparison with their work we used AuNPs of similar diameters (20nm, 80nm) conjugated with the aptamers. Moreover, using larger AuNPs for DLS experiments, as compared to the 5nm NPs depicted in Figure 1, gives a clearer distinction between the AuNPs.virus and the unconjugated virus (100nm). Calculations based on close packing of these NP's suggest that each virus particle could accommodate 200 NPs of 20nm diameter or 12 NPs of 80nm diameter on their surface.



Figure S1. DLS measurements. (a) aptamer-conjugated AuNPs without (red) and with (light green) *J1999V*. (b) aptamer-conjugated AuNPs without (red) and with *P2007V* (light green). Replacing by a random RNA sequence of the same length with the aptamers, no significant change in AuNP's light scattering size was seen in the absence of the specific aptamer conjugation.

DLS measurements revealed that the aptamer-AuNP complex has a mean diameter of 53nm. This is consistent with a 20nm AuNP core surrounded by the biomolecular recognition layer (streptavidin/oligonucleotide linker/aptamer). When J1999V was added, the mean particle diameter increased to 241nm consistent with the aptamer-conjugated AuNPs forming a shell around the viral envelope. Similarly, a corresponding change was observed when P2007V was added to the relevant aptamer conjugated with 80nm AuNPs and these values are consistent with the DLS data obtained from the antibody conjugated AuNPs [1]. DLS was also used to assess the aptamer cross-reactivity. Addition of the J1999 aptamer to the P2007 virus showed a small degree of binding; however the P2007 aptamer showed no evidence of binding to the J1999 virus. Although a very sensitive technique, DLS is not suitable for point-of-care tests.

ELONAs

As the method described in this paper involved using 5nm AuNPs as the detection moiety, affinities of the AuNP-conjugated aptamers for the virus were also investigated to assess the effect of attachment of the AuNPs on their affinity. In order to assess binding affinities of the aptamers conjugated to the AuNPs, we employed a modified ELONA protocol. In this modified assay format (competitive inhibition ELONA), we establish a competitive binding reaction between the unconjugated and conjugated forms. The aptamers were labelled with biotin at the 3'-ends through DNA linkers. In the unconjugated form, the biotin is free to couple with streptavidin-HRP for subsequent enzymatic reaction once TMB is added. In the conjugated form, the biotin is bound to the streptavidin-coating on the AuNP, preventing the aptamers from coupling with streptavidin–HRP.

1 J. D. Driskell, C. A. Jones, S. M. Tompkins and R. A. Tripp, *Analyst*, 2011, **136**, 3083-3090.