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Two-way gold nanoparticle label-free sensing of specific sequence and small molecule targets using switchable concatemers

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A two-way colorimetric biosensor based on unmodified gold nanoparticles (GNPs) and a switchable double-stranded DNA (dsDNA) concatemer have been demonstrated. Two hairpin probes (H1 and H2) were first designed that provided the fuels to assemble the dsDNA concatemers via hybridization chain reaction (HCR). A functional hairpin (FH) was rationally designed to recognize the target sequences. All the hairpins contained a single stranded DNA (ssDNA) loop and sticky end to prevent GNPs from salt-induced aggregation. In the presence of target sequence, the capture probe blocked in the FH recognizes the target to form a duplex DNA, which causes the release of the initiator probe by FH conformational change. This process then starts the alternate-opening of H1 and H2 through HCR, and dsDNA concatemers grow from the target sequence. As a result, unmodified GNPs undergo salt-induced aggregation because the formed dsDNA concatemers are stier and provide less stabilization. A light purple-to-blue color variation was observed in the bulk solution, termed the light-o sensing way. Furthermore, H1 ingeniously inserted an aptamer sequence to generate dsDNA concatemers with multiple small molecule binding sites. In the presence of small molecule targets, concatemers can be disassembled into mixtures with ssDNA sticky ends. A blue-to-purple reverse color variation was observed due to the regeneration of the ssDNA, termed the light-on way. The two-way biosensor can detect both nucleic acids and small molecule targets with one sensing device. This switchable sensing element is label-free, enzyme-free and sophisticated-instrumentation-free. The detection limits of both targets were below nanomolar.

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