Supplementary

A facile assay for rapid detection of COVID-19 antibodies

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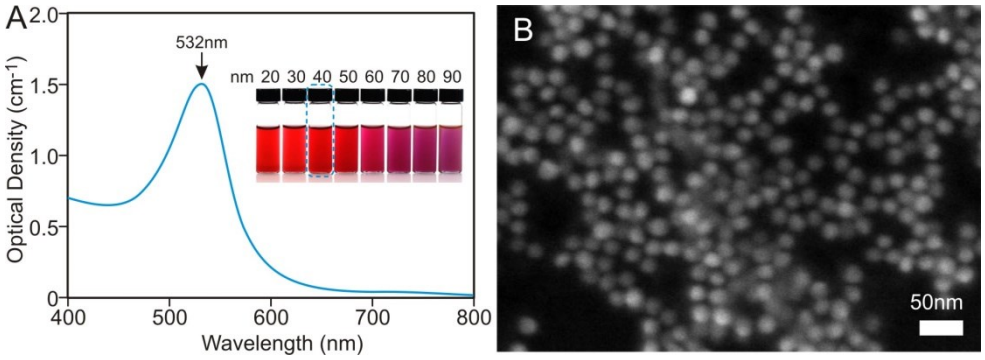

Fig S1. (A) The UV-Vis spectrum and the appearance (inset [1]) of synthesized AuNPs. (B) SEM image of the synthesized AuNPs.

Supplementary, S1

The optimization processes include 2 parts as follows:

1st part: Optimization of the antigen Ag8 and AuNPs coupling rate:

According to the method in the manuscript, 5μg, 10μg, 15μg and 20μg of Ag8 were added to 1ml

of AuNPs solution (pH 8.5) and bound for 30min, after that the reaction mixture was centrifuged at 9000 rpm and 4°C for 30 min. Nanodrop-2000 and BSA were used to plot the standard curve of protein (OD280), and the protein in the supernatant was examined (5 times for each sample) to calculate the coupling rate of AuNP-Ag8. The results showed that in 1 ml AuNPs solution, for 5μg, 10μg, 15μg and 20μg of Ag8, the antigen coupling rates were 99.72%, 97.75%, 95.33% and 93.29%, respectively. Furthermore, the precipitate was also processed as the methods in the manuscript, and 4 different AuNP-Ag8 conjugates were prepared respectively. After the strip preparation, the serial diluted serum of a COVID-19 patient * was used to test the performance of IgM/IgG examination. The results showed that 10μg/ml, 15μg/ml Ag8 conjugation could detect the IgM/IgG precisely, while the 20μg/ml Ag8 conjugation made the results oversaturated.

*: The chemiluminescence microparticle immunoassay (CMIA) was used to quantitative detection of the COVID-19 specific IgM/IgG contents of the confirmed patient. The cut-off values were set as 10.0 AU/ml for IgM and IgG, and the result higher than 10.0 AU/ml was judged as positive. The value of IgM and IgG were 252.53AU/ml and 289.87AU/ml respectively of the patient's serum used in this confirmatory test.

2nd part: Coating optimization of AuNP-Ag8 in the conjugation pad:

The AuNP-Ag8 and the AuNPs-rabbit IgG conjugate were mixed evenly at 2:1 (v/v), and then sprayed on the conjugation pad at volume of 6 μl/cm, 8 μl/cm, 10 μl/cm, 15 μl/cm and 20 μl/cm. After the strip preparation, the strips were verified with the patient's sample as mentioned above. The results manifested that the strips coated with 15 μl/cm of AuNPs conjugation could accurately detect the serial diluted IgM and IgG, and obtained the visible band for naked eyes.

Supplementary S2: The RT-PCR test.

The RT-PCR reagents were purchased from Shanghai BioGerm Medical Biotechnology Co.,Ltd (Registration Certificate for Medical Device of PRC, NO. 20203400065). The reagent uses real-time fluorescent PCR technology, the specific primers and TaqMan probes were designed with the ORF1ab and N genes of COVID-19. By amplification in fluorescence quantitative PCR instrument (ABI-7500) to detect the nucleic acid of COVID-19. The specimen collection and nucleic acid extraction were carried out in the designated conditions with PPE mentioned in the

manuscript. After the nucleic acid was extracted (ZTLYB-Y64 Nucleic Acid Extraction Kit, Tianlong Technology, and GeneRotex96 Extractor) from the clinical sample, the one-step RT-PCR amplification was performed and the fluorescence signal was detected, and the real-time amplification curve was plotted automatically by the 7500 Software (v2.3) system. The internal reference control in the kit was used to monitor the process of RT-PCR to avoid the false negative results, partial amplification curves were listed in **Fig S2**.

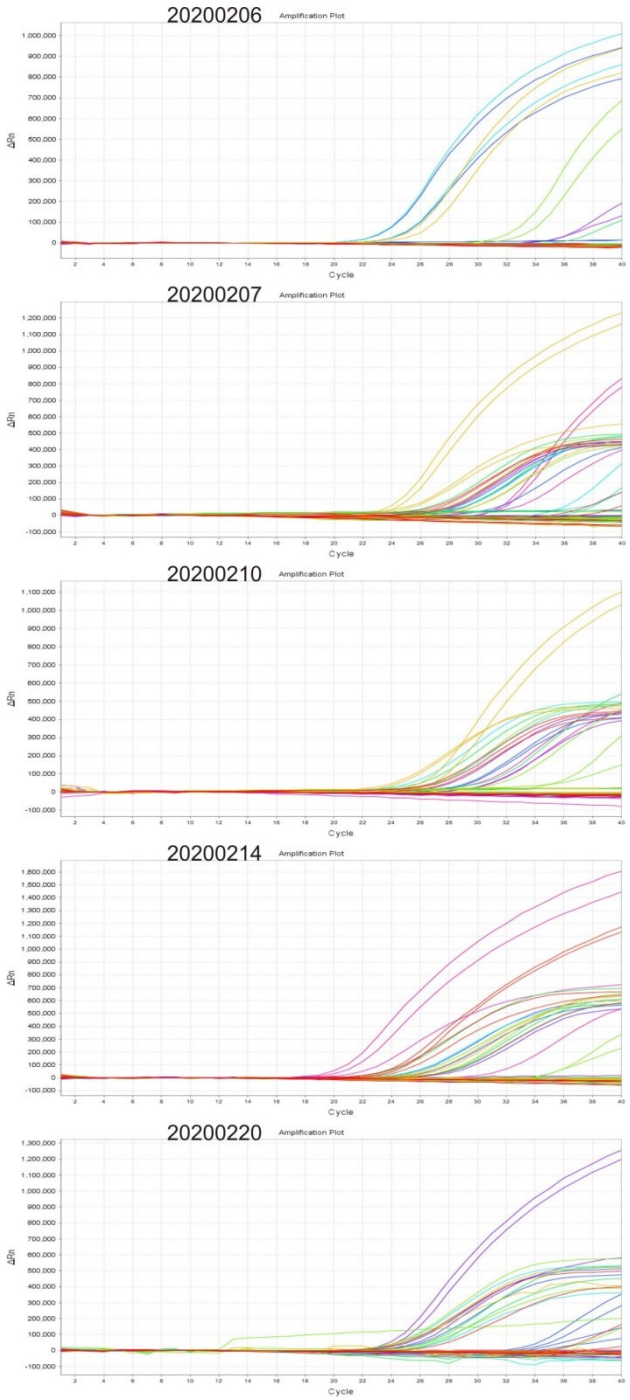

Fig S2. Partial amplification curves of RT-PCR for COVID-19 detection.

Reference

1. https://nanocomposix.eu/pages/gold-colloid (Accessed June, 30. 2020).