Supplementary Information

Plasmonic gold nanoparticle for detection of fungi and human cutaneous fungal infections

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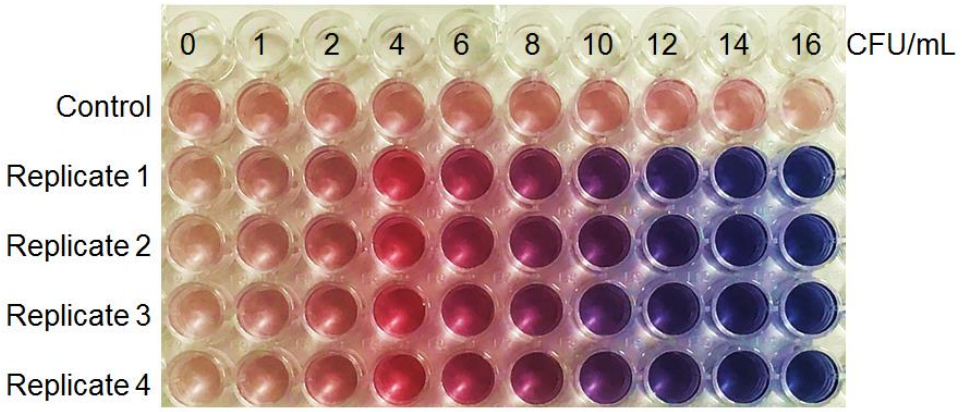
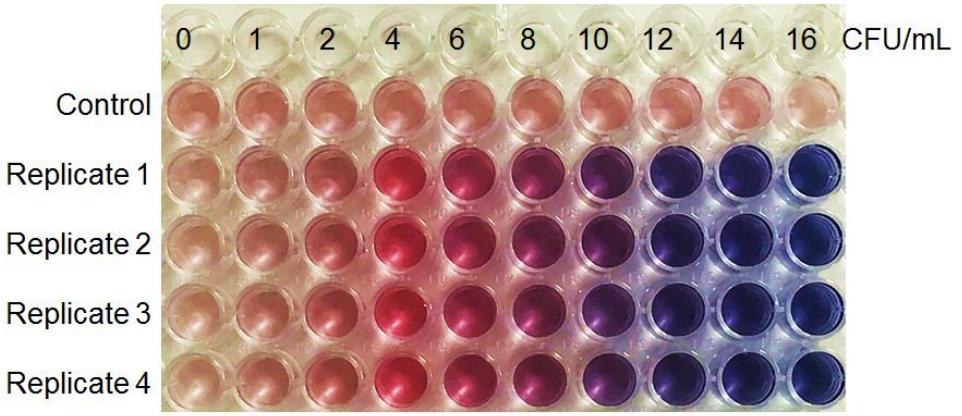
Layout of concentration of fungi and bacteria on 96 well plates

The mixture with 2 mM Chloroauric acid (10 µl) and 2 µl HEPES buffer in 88 µl distilled water was added to the 96 well plate. The wells of the first row were blank, containing 100 µl Ultrapure deionized water. The wells from the second to fifth row were filled with 100 µl fungi suspensions. The concentration of fungi were 0, 1, 2, 4, 6, 8, 10, 12, 14, 16 CFU/ml from left to right in each row. Four replicates have been measured for each data point. A plate layout of four fungi from top to bottom was comprised of *A. niger*, *A. oryzae*, *P. chrysogenum* and *M. hiemalis*. The wells from second to fifth row were replicas for each fungi 96-well plate (S1).

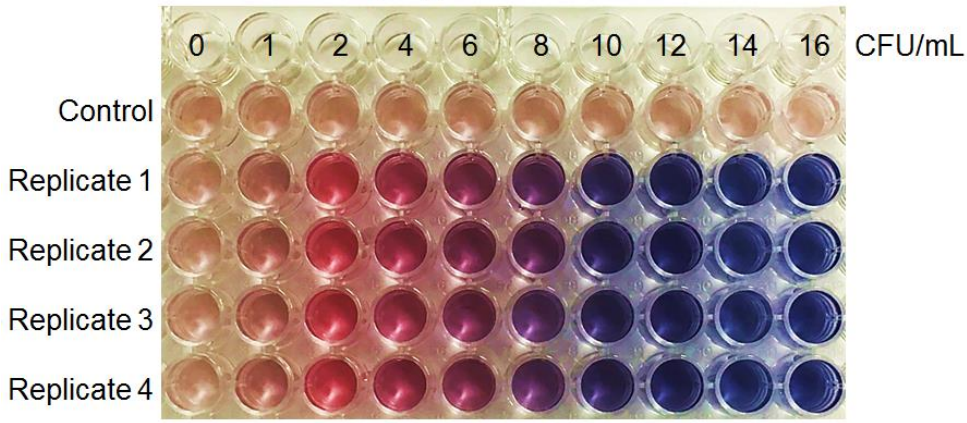
Aspergillus *niger*



Aspergillus *oryzae*



Penicillium *Chrysogenum*



Mucor *hiemalis*

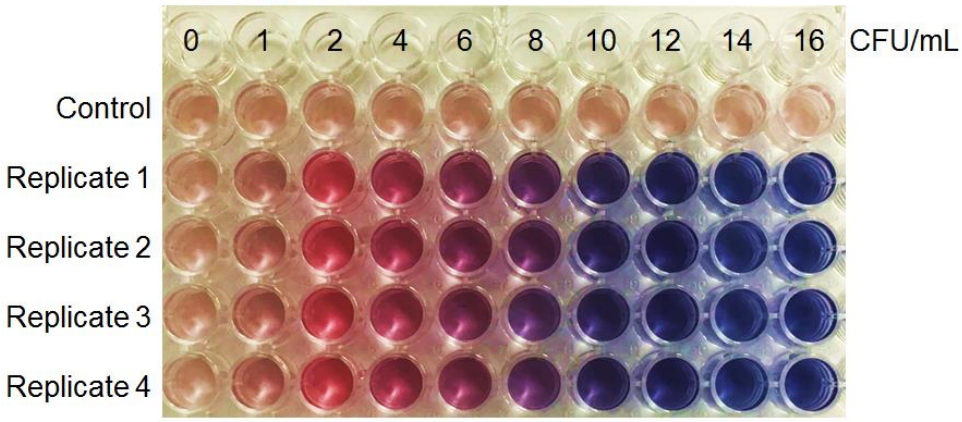


Figure S1. Colorimetric assay of Gold nanoparticle on four different fungi in 96 well plate. The concentration of fungi increases from left to right from 0 to 16 CFU/mL and the wells from 2nd row to 5th row are replicates. The colour changes from red to pink-purple and to blue indicated the increase in fungi concentration.

Layout of concentration of bacteria on 96 well plates

The mixture with 2 mM Chloroauric acid (10 µl) and 2 µl HEPES buffer in 88 µl distilled water was added to the 96 well plate. The wells of the first row were blank, containing 100 µl Ultrapure deionized water. The wells from the second to fifth row were filled with 100 µl bacteria suspensions. The plate layout of three bacterial samples starting from the second row to fourth row were *Escherichia coli*, *Bacillus cereus* and *Staphylococcus aureus* (S2).

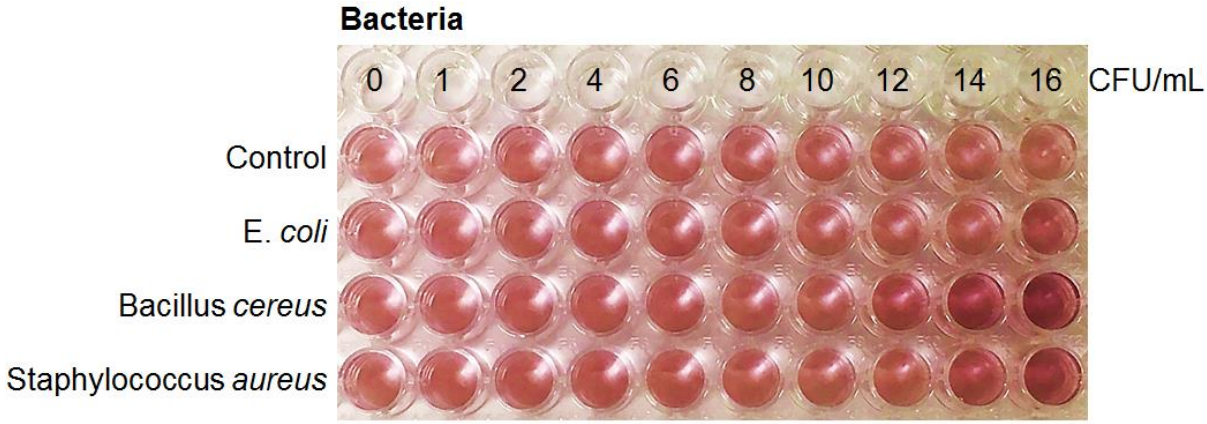


Figure S2. Colorimetric assay of Gold nanoparticle on three different bacteria in 96 well plate. The concentration of *bacteria* increases from left to right from 0 to 16 CFU/mL. *Escherichia coli*, *Bacillus cereus* and *Staphylococcus aureus* were loaded from top to bottom, respectively. The colour of the solution in bacteria samples changes from pale yellow to pink in 50 mins.

Concentration depended Surface Plasma Resonance of gold nanoparticles on *Escherichia coli*

50 minutes after the addition of bacteria with HEPES buffer and $HAuCl_4$, a slow colour change was observed. When the bacterial concentration is increased over the range 0-16 CFU/mL, there is a minor shift of the SPR to higher wavelengths, with a slow decrease in the absorbance ratio concentration 520nm / 620nm. There is a linear relationship between concentrations of *Escherichia coli* and the ratio from 0-16 CFU/mL. The range of ratio observed was 2.3-1.6 at bacterial concentrations 0-16 CFU/mL (S3).

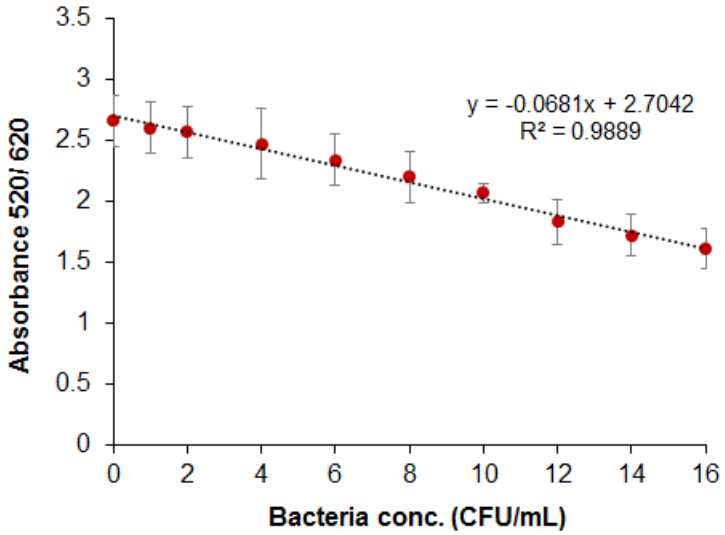


Figure S3. SPR of gold nanoparticle formation and morphological transformation incubated with increasing concentrations of *Escherichia coli* at pH=5.5. Absorbance ratio 520nm/620nm following incubation of gold nanoparticles with increasing concentrations of *Escherichia coli*, in HEPES buffer.

Time depended GNPs formation and morphological transformation on fungi and bacterial

With increasing time, a sharp and narrow SPR peak was observed at 530 nm within first 0.5 min of addition of fungi to $HAuCl_4$ with HEPES buffer in pH=5.5. Within 1 min, the absorption intensity of SPR peak was found to increase, and it was seen to red shift and broaden. This signifies polydispersity and increase in the size of synthesized GNPs within 2 mins. The ratio of absorbance 520 nm/620 nm was round to rapidly decrease from 4-0.8 in the time 0.5 min to 2 mins. At the time point of 2mins, the ratio stabilised at 0.8 (blue dots, S4), suggesting the completion of GNPS formation and morphological transformation.

In the case of bacteria, a modest and broad SPR peak was observed at 530 nm within the first 0.5 min of bacteria addition to $HAuCl_4$ with HEPES buffer. Within 2 min the absorption intensity of SPR peak was found to increase, at the same wavelength. The ratio of absorbance 520 nm/620 nm were increased slightly from 1.0-1.08 over the period from 0.5 min to 2 mins (red dots, S4). Bacteria can cause minor cause aggregation and red shifting of the SPR over a prolonged period of 50 mins (S3). This is a major drawback of procedures compared to fungi for rapid detection. Hence, the potential of this fungus towards such rapid GNP synthesis yielding a specific ratio of absorbance at 520 nm/620 nm at 0.8 presents a significant advantage in the context of a biosensor.

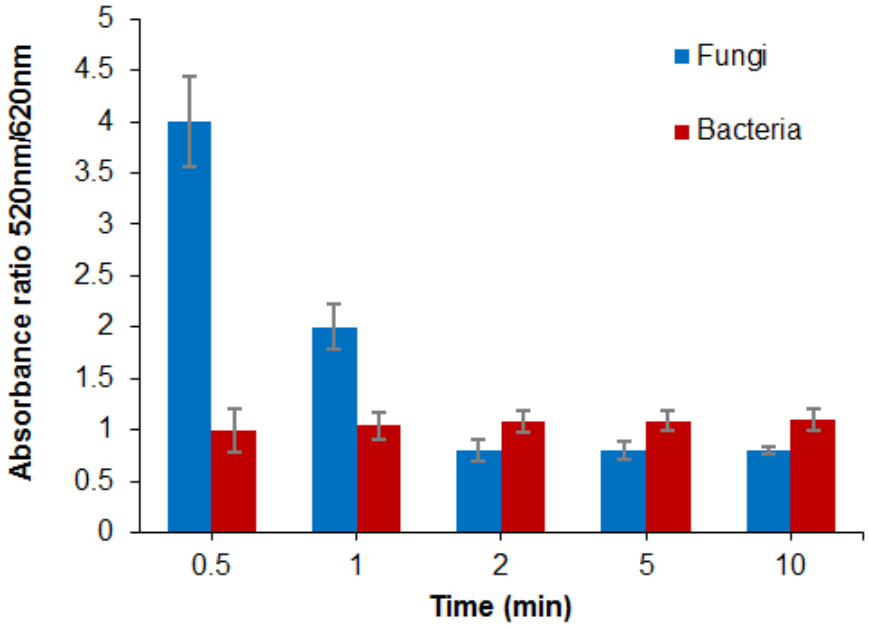


Figure S4. Time dependent gold nanoparticle formation and morphological transformation on Bacteria (Red) and Fungi (Blue). The formation of gold nanoparticles over time was measured by the ratio of absorbance at 520nm/620nm from 0.5 min to 10 mins.

Time depended Surface Plasma Resonance of GNPs on 200 mM of $NaNO_3$

In the presence of Na^+ alone, a different spectral evolution profile is observed, as shown in the typical time-course of GNP evolution in the presence of $NaNO_3$ (S5). The initial SPR resonance peak at ~530nm is slightly shifted and broadened over a period of ~50minutes and a broader resonance with maximum > 800nm evolves, associated with a colour change from pink to grey and transpired (S5).

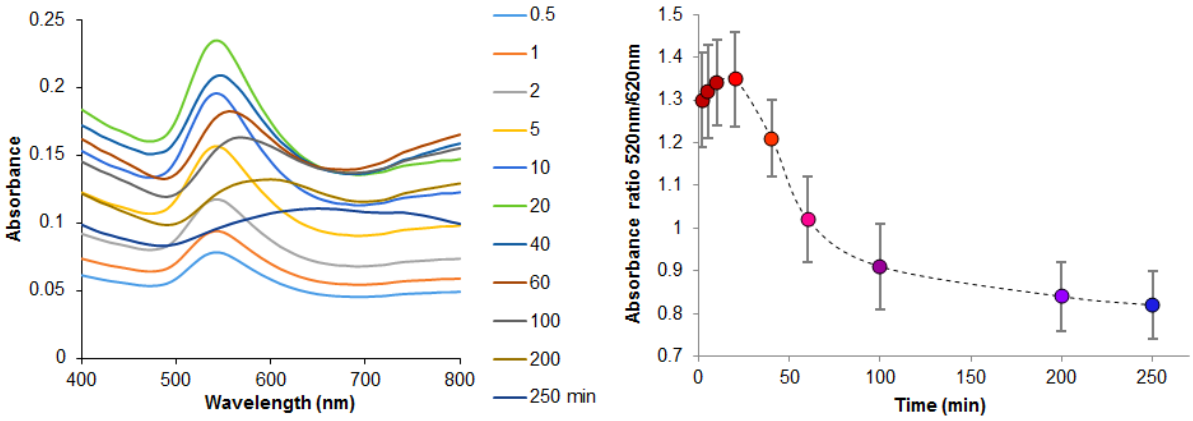


Figure S5. Time dependent surface plasmon resonance (SPR) of gold nanoparticles formation and morphological transformation in sodium nitrate ($NaNO_3$). (Left panel) Absorbance spectra of nanoparticles after mixture with 200 mM of $NaNO_3$ in HEPES buffer at pH=5.5. The dotted lines shown the spectra of GNPs in 20 mins. (Right panel) The absorbance ratio 520 nm/620 nm gradually increased from 0.5 min to 20 mins, and slowly decreased at 40 min. At the time point of 200 mins, the ratio 520 nm/620 nm reached 0.8 (panel right).