

Since, the nanoparticles possess a large surface area, the surface modification by a suitable adsorbate can produce different properties, hence FTIR spectroscopy was used for the detection of functional groups in pure compounds, mixtures and for comparison among compounds that correlated with the vibrational motion of atoms or molecules. Figure 4 depicted the FTIR of the AuNP synthesized from *S. cumini* leaf extract that shows the characteristic –

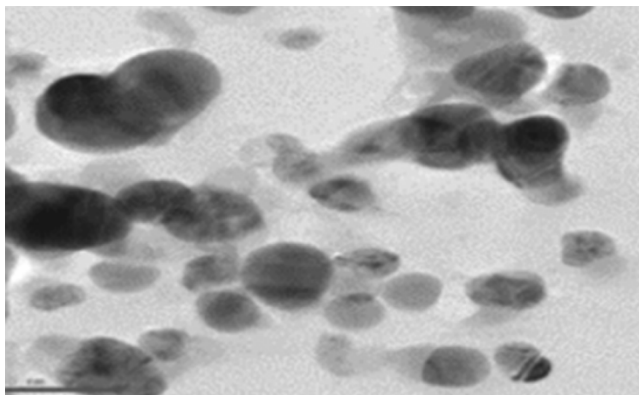


Fig. 3 — TEM images of gAuNPs

CHO peak around 2900 cm^{-1} , C-O stretch of alcohol group at 1100 cm^{-1} and have three peaks depicting amines groups *i.e.* C-N stretch at 1030 cm^{-1} , Ar-N stretch at 1320 cm^{-1} and NH₂ in plane bend at 1610 cm^{-1} (Fig. 4). FTIR analysis of *S. cumini* leaf indicated a broad and strong absorption band in a range of $685\text{--}638\text{ cm}^{-1}$. These absorptions are allocated to different stretching vibrations. The C-C, O-H stretching vibration appeared at 685 cm^{-1} , 1633 cm^{-1} and 3400 cm^{-1} , respectively. Further, the C=O stretching was observed at 1400 cm^{-1} .

Antimicrobial disc diffusion assay of the prepared AuNPs were done (Table 1 and Fig. 5) and antibacterial activity was observed against both Gram positive (*S. aureus*) or Gram negative (*E. coli*) bacteria. Figure 6A and B indicates the growth inhibition curves by AuNP at two different concentrations *i.e.* $10\text{ }\mu\text{g/mL}$ and $100\text{ }\mu\text{g/mL}$ on both *E. coli* and *S. aureus* bacteria Time dependent growth inhibitory activity was observed by AuNPs²¹.

Literature survey suggests that AuNPs were capable of adhering to the bacterial cell wall and

Fig. 4 — FTIR of gAuNPs Peaks at 2933 , 1633 , 1531 and 1415 cm^{-1} were attributed to the stretching vibrational frequencies of various functional groups such as H of methyl and methoxy groups, C=O of acid derivatives, C=C of aromatic rings and amide groups which come from the biomolecules of *Syzygium cumini*

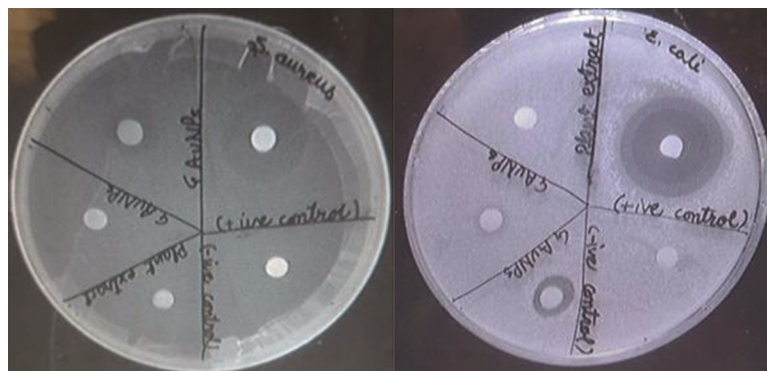


Fig. 5 — Antibacterial activity of test samples *i.e.* Leaf extract, gAuNPs in gram-positive (*S. aureus*) and gram-negative (*E. coli*) bacteria and its comparison with positive control (Gentamycin 5 mg/mL) & negative control

Table 1 — Antimicrobial disc diffusion assay

| Sample | Zone of Inhibition - ZOI (mM) | |
|---|-------------------------------|------------------|
| | <i>E. coli</i> | <i>S. aureus</i> |
| Positive Control (Gentamycin: 5 mg/mL) | 10±0.1 | 8±0.5 |
| Negative Control | 0±0 | 0±0 |
| Leaf extract | 0±0 | 0±0 |
| gAuNPs | 4±0.4 | 2±0.2 |
| gAuNPs | 2±0.2 | 1.5±0.2 |

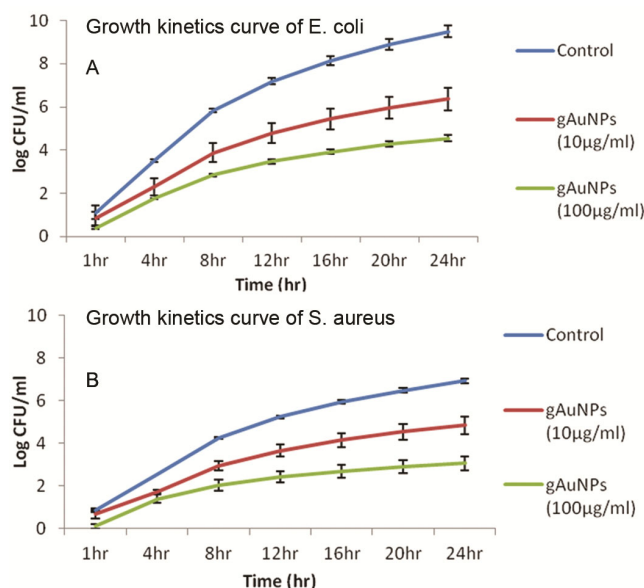


Fig. 6 —Growth kinetics curves of (A) *E. coli*; and (B) *S. aureus* when treated with gAuNP (10 µg/mL) and gAuNP at 100 µg/mL. Values are represented as mean ± standard deviation of three sets of identical experiments

penetrating it, causing conformational changes in the cell membrane ultimately leading to cell death²². Published evidences indicated strong interaction between AuNPs and the peptidoglycan (PGN). AuNPs interact with bacterial cell walls individually or *via* the released Au⁺ ions that generate “pits” in the cell walls owing to their nano size. This leads to the accumulation of AuNPs on the cell membrane as they begin to adhere strongly to the layers, thereby releasing more and more Au⁺ ions. This phenomenon strongly influences the destruction of Gram-positive bacteria as they have a thicker PGN layer, but Gram-negative bacteria are resistant to this phenomenon. According to Reidy *et al.* 2013, there are a series of mechanisms by which the AuNPs manifest their antibacterial property. As nanoparticles have an ultrasmall size and a large surface area, they are capable of making a strong contact with the bacterial surface. It has been reported earlier that Au-P III inhibited the division in both *S. aureus* and

E. coli. Secondly, the AuNPs penetrate the bacterial cell wall that leads to DNA damage. Thirdly, the dissolution of AuNPs releases Au⁺ ions that can act together with sulphur-containing proteins of bacteria to alter the structure and function. This phenomenon is an important mechanism of the antimicrobial activity of AuNP. The interaction of dissolved Au⁺ ions to extracellular as well as intracellular proteins highlights the binding of Au⁺ ions with the thiol group present in the vital enzymes that result in their inactivation or altered activity. Fourthly, Au⁺ ions may further interact with the phosphorous containing compounds like DNA which undoubtedly interferes with its replication process and inhibits the proliferation resulting in decreased growth over a period of time.

Conclusion

We report cost effective, eco-friendly, safe, simple, non-toxic, and single step synthesis protocol for AuNPs from leaf extracts of *Syzygium cumini*. The AuNPs made by leaf extract at 25°C was found to be more active in terms of antimicrobial activity. The efficacy of AuNPs was evident in Gram positive bacteria at nanogram concentrations as compared to Gram negative bacteria. These preliminary results warrant further experiments to elucidate the molecular mechanism both *in vitro* and *in vivo* models. The proposed cost-effective and eco-friendly approach for mass production of AuNPs has immense potential applications.

Conflict of interest

All authors declare no conflict of interest.

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