# Sunlight- mediated synthesis of silver and gold nanoparticles using Active Manuka Honey 20+ UMF$^{R}$ against wound infection causing bacteria

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***Abstract-*** The noble metal gold and silver nanoparticles have been successfully prepared using a bio-derived product-Active Manuka honey 20+ UMFR. Manuka honey is well recognized for its beneficial properties over and above mixed flora honey for use in wound treatment. Having been found to offer effective antimicrobial, anti-inflammatory and debridement properties as a dressing, it is only the medical grade Active Manuka honey that should be used in open wounds. A high UMFR grading above 10+ is recommended for wounds. The formation of silver and gold nanoparticles has been confirmed using UV–vis, SEM and FTIR analysis and its antibacterial activity was evaluated against *Enterococcus faecalis*, *Staphylococcus aureus* and *Escherichia coli*. Silver and gold nanoparticles showed a clear well defined inhibition zone compared to control. The results suggest the stabilized and reduced molecules of silver and gold nanoparticles may act as an effective antibacterial agent for wound healing.

***Index Terms***- Manuka Honey; Nanoparticles; Antibacterial activity; Wound healing.

## I. INTRODUCTION

Manuka honey obtained from the *Leptospermum scoparium* (Manuka tree) in New Zealand. It is well-known for a pronounced antibacterial activity which cannot be found in any other honey. This honey demonstrates exceptional antimicrobial activity; however, studies have shown its antimicrobial process is independent of hydrogen peroxide, antimicrobial phenolics and flavonoids, all of which exert some degree of antimicrobial activity [1]. The antimicrobial activity of methylglyoxal forms a near perfect linear correlation with that of phenol, allowing its activity to be expressed as an equivalent phenol concentration commonly known as its unique manuka factor (UMF) [2]. The UMF rating given to manuka honeys increases with antimicrobial activity and ranges increases [3]. Manuka honey itself has long been in employment for clearing up infections, including abscesses, surgical wounds, traumatic wounds, burns and ulcers of different etiology [4]. Nanoparticles often show unique and considerably changed biological, physical and chemical properties compared to their macro scaled counterparts [5, 6]. Nanocrystalline silver and gold particles have been found tremendous application in the fields of high sensitivity biomolecular detection, antimicrobials, micro-electronics, diagnostics and therapeutics [7]. Silver and gold belong to a family of 'free' electron metal that have a filled valence shell but an unfilled conduction band. When nanoparticles of silver and gold are irradiated with incident light, the 'free' electrons move under the influence of the electromagnetic field and are displaced relative to their positive core, which creates an oscillating dipole. Oscillating dipole absorb maximum energy at their resonance frequency, which lies in the visible range of the electromagnetic spectrum for silver and gold nanoparticles. To the best of our knowledge, the use of Manuka honey for the green synthesis of silver and gold nanoparticles has not been reported. Hence the present study was carried out to synthesize and characterize the silver and gold nanoparticles using Active Manuka honey20+ UMFR and evaluate its antibacterial efficiency against wound infection causing bacteria *Enterococcus faecalis*, *Staphylococcus aureus* and *Escherichia coli*.

## II. MATERIALS AND METHODS

### A. Chemicals and materials

AgNO3 and AuCl4 were purchased from Sigma Aldrich. Manuka honey was purchased online from New Delhi.

### B. Synthesis of silver nanoparticles

For the reduction of silver ions, 1 ml of honey was added to 9 ml of 0.1M aqueous AgNO3. The reaction mixture was stirred properly and exposed to bright sunlight. The color of solution turned to yellowish- brown. The changed colour indicates the formation of silver nanoparticles.

### C. Synthesis of gold nanoparticles

Honey dilution was prepared by mixing 1 ml honey to 9 ml of distill water. For the reduction of gold ions, 5 ml of aqueous honey was added to 5ml of aqueous AuCl4. The reaction mixture was stirred properly and exposed to bright sunlight. The color of solution turned to brownish-black indicating the formation of gold nanoparticles.

### D. Characterization

UV-vis spectroscopic analysis of the yellowish-brown colored solution and brownish-black solution was carried out using UV-vis spectrophotometer. The reduction of silver ions was monitored from (350-650) nm. And the reduction of gold ions was monitored from (400-700) nm. For FTIR measurement

the dried powder was analyzed on a FTIR spectroscopy. The morphology of the silver and gold nanoparticles was analyzed using scanning electron microscope (SEM).

E. Antibacterial assay

The efficiency of silver and gold nanoparticles as an antibacterial compound was evaluated against *Enterococcus faecalis*, *Staphylococcus aureus* and *Escherichia coli*. The disc diffusion method was used to assess the antibacterial activity of the synthesized silver and gold nanoparticles.

## III. RESULTS AND DISCUSSION

A. UV-Vis Spectrophotometric analysis

UV-Visible spectroscopy is a significant technique to ascertain the formation and stability of metal nanoparticles in aqueous solution. Metallic nanoparticles display characteristic optical absorption spectra in UV-visible region called surface plasmon resonance (SPR). Metal nanoparticles have free electrons, which gives surface plasmon resonance absorption band, due to the combined vibration of electron of metal nanoparticles in resonance with light wave. The UV-Visible absorption of silver nanoparticles are exhibit maximum in range of 400-500 nm and gold nanoparticles are exhibit maximum in range of 500-600 due to this property[8]. Figure 1 shows the sharp band of silver nanoparticles was observed at 413 nm and gold nanoparticles band were observed at 534 nm.

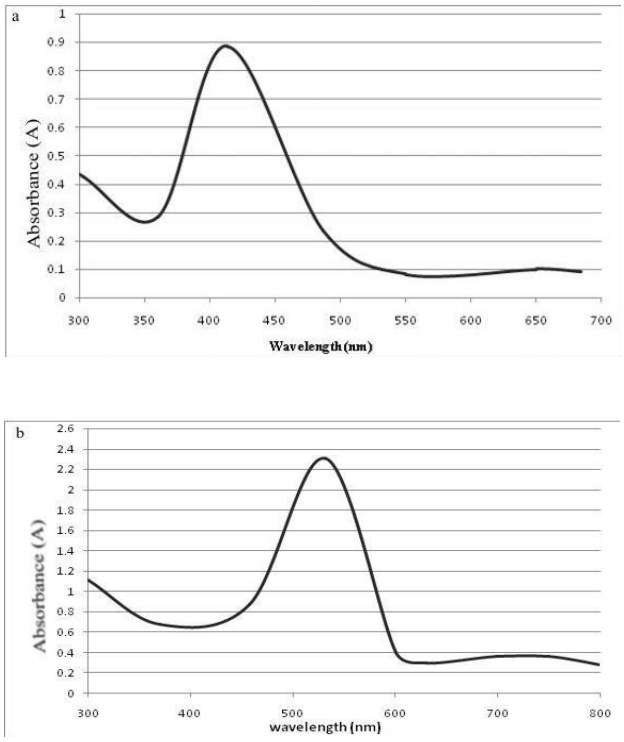


**Figure 1: UV-vis spectra of manuka honey containing (a): AgNPs and (b): AuNPs**

B. Scanning Electron Microscopy (SEM)

Morphology of silver and gold nanoparticles was confirmed using SEM analysis, which indicates that the metal nanoparticles presence in nano-size. Figure 2 shows aggregation of spherical shaped silver nanoparticles and spherical and square shaped gold nanoparticles was observed below 100 nm with the magnification of 20,000X

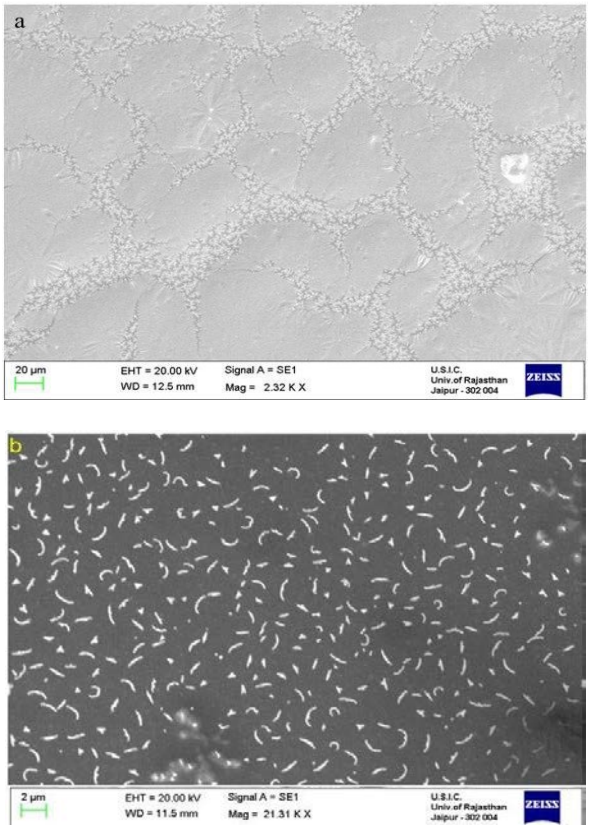


**Figure 2: SEM micrograph of (a) silver nanoparticles (b) gold nanoparticles**

C. Fourier transform infrared spectroscopy (FTIR)

FTIR measurement was carried out to identify the possible biomolecules of Manuka honey 20+ UMFR responsible for capping leading to efficient stabilization of the silver and gold nanoparticles. The FTIR spectrum obtained for Manuka honey 20+ UMFR displays a number of absorption peaks, reflecting its complex nature due to biomolecules. The IR spectrum of silver nanoparticles manifests prominent absorption bands at 3927, 3561, 3198, 3068, 2756, 2539, 2401, 1811, 1627, 1546, 1482, 1328, 1278, 1228, 1045, 854, 698, 588. The strong bands at 1627 cm-1 and 1546 cm-1 are due to the carboxyl stretch and N-H deformation vibrations in the amide linkages of protein. Ag nanoparticles can bind to protein through free carboxylate ion or amine group of the amino acid residue in it. The bond at 1045 cm-1 is due to the C-O-C symmetric bending and C-O-H bending vibrations of protein in the manuka honey. The IR spectrum of gold nanoparticles manifests prominent absorption bands at 3956, 3447, 3058, 2902, 2832, 2379, 1993, 1646, 1442, 1329, 1099, 847, 673, 589, 463 cm-1. The strong band at 3447 cm-1 is due to N-H stretching vibration of primary amines. The band at 1329 and

1099 cm-1 are may be due to C-N and –C-O-C stretching modes, respectively. The band at 673 cm-1 is due to the bending vibration of N-H groups in proteins. This result suggested that the opening of the glucose ring by abstraction of the alpha-proton of the sugar ring oxygen and the metal ions oxidize glucose to gluconic and sucrose and protein/enzyme play a role in reduction.

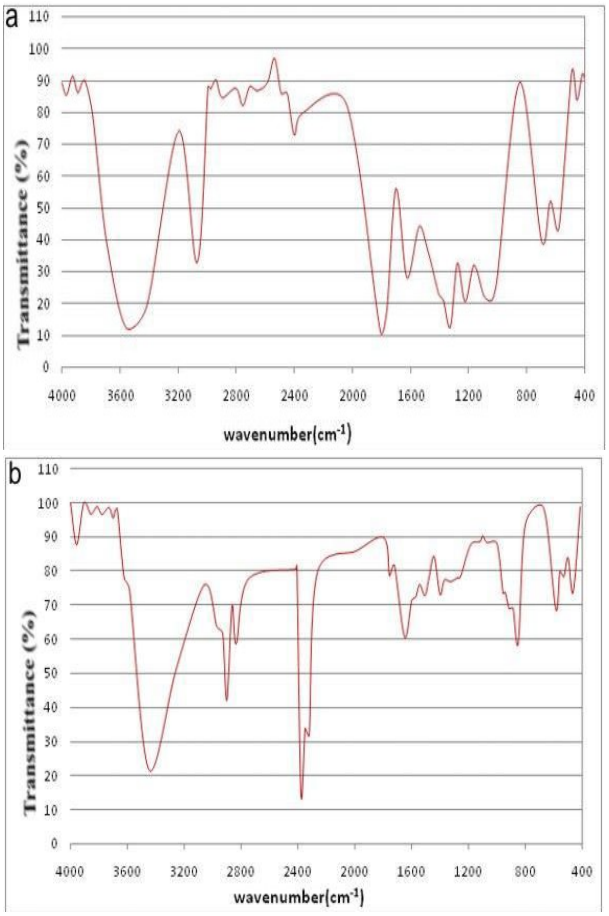


**Figure 3: FTIR spectrum of (a) silver nanoparticles (b) gold nanoparticles**

D. Antibacterial Screening

Silver and gold nanoparticles have been used for its antimicrobial properties. The more advances in generating silver and gold nanoparticles use as a powerful bactericide against various clinical pathogens.

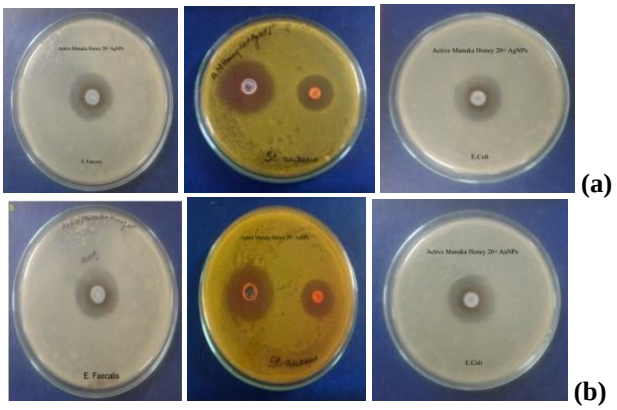


**Figure 4: (a)** Zone of inhibition of Silver nanoparticles against *Enterococcus faecalis*, *Staphylococcus aureus* and *Escherichia coli* and **(b)** Zone of inhibition of Gold nanoparticles against *Enterococcus faecalis*, *Staphylococcus aureus* and *Escherichia coli.*

In the present study, zone of inhibition of biologically synthesized silver and gold nanoparticles was evaluated against *Enterococcus faecalis*, *Staphylococcus aureus* and *Escherichia coli*. Figure: 4 depict synthesized silver and gold nanoparticles showed a clear well defined zone of inhibition against all the bacterial species. Thus, in the present investigation, the application of silver and gold nanoparticles was proved as an antibacterial agent by green route method.

## IV. Conclusion

For the first time, a completely eco-friendly and fast method for the synthesis of metallic nanoparticles using Active Manuka honey 20+ UMFR and sunlight is demonstrated. The use of natural honey, distilled water and practically nontoxic reagents can be produced silver and gold nanoparticles through “Sunlight- mediated”, methodology thus avoiding the presence of hazardous and toxic solvent and waste. The synthesised nanoparticles are used in sensitive areas such as biomedicine. Since the synthesised silver and gold nanoparticles using Active Manuka honey 20+ UMFR shows antibacterial activity against tested bacterial pathogens, it could be targeted for the promising potential to prepare metallic nanoparticles impregnated wound dressing material for healing wounds effectively in future.

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