



Green synthesis of silver nanoparticles using Citrus limon peel extract and evaluation of their antibacterial properties and cytotoxic activities

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Introduction:

- Nanotechnology nowadays has received considerable attention due to their unique properties and potential applications.
- As a results of their properties, nanoparticles can be employed in several felids, for example, the felid of medicine.
- There's an urgent need to find alternatives to drugs used nowadays, one of the alternatives is the use of nanoparticles.







Why is it important to green synthesize silver nanoparticles ?

• Development of <u>eco-friendly</u>, <u>economic</u> and <u>nontoxic</u> process for the synthesis of metallic nanoparticles is an important step in the field of application of nanotechnology.







Objectives:

Objective 1:

To green synthesize silver nanoparticles using Lemon peels extract and characterize them using: UV spectrophotometer, DLS, TEM, EDX and FTIR.

Objective 2:

To evaluate the antibacterial activity against various bacteria, and combination effect of the silver nanoparticles with different antibiotics.

Objective 3:

To asses the cytotoxic activity of the green synthesized silver nanoparticles on the cell lines of MCF-7 & HCT-116.





Review of literature:

• Reena & Menon (2017), have examined the green synthesis of AgNPs using citrus fruit peel extract under two different conditions, sun dried and oven dried form. The biosynthesized AgNPs from the sun dried *Citrus sinensis* and mixed fruit peel extract resulted best antibacterial activity against *Pseudomonas aeruginosa*, *Escherichia coli* and *Salmonella typhimurium*.







Methodology:



Collection of the peels and preparation of the extract.



70 grams of peels were transferred into 50ml of boiled distilled water and left for 10 minutes, then the extract was filtered.



Green synthesis of AgNPs Involved the addition of 0.008g (1mM) silver nitrate (AgNO₃) in 50 ml distilled water, then 5ml of the LPE has been added.



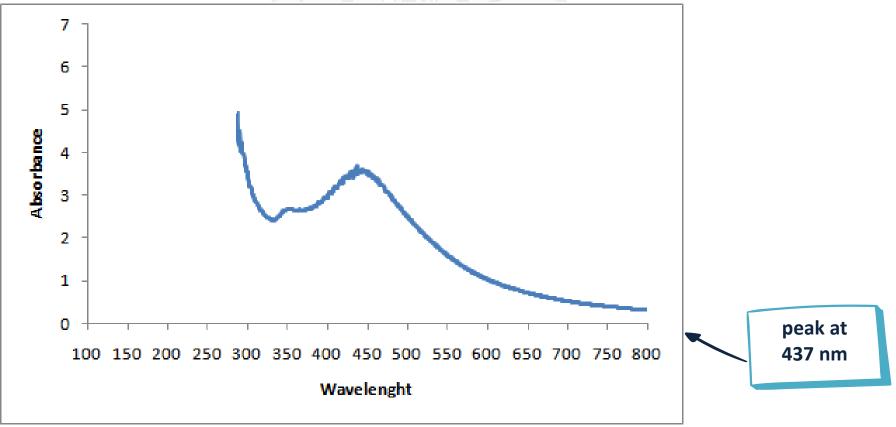
The color change was observed after 15 minutes.





Results

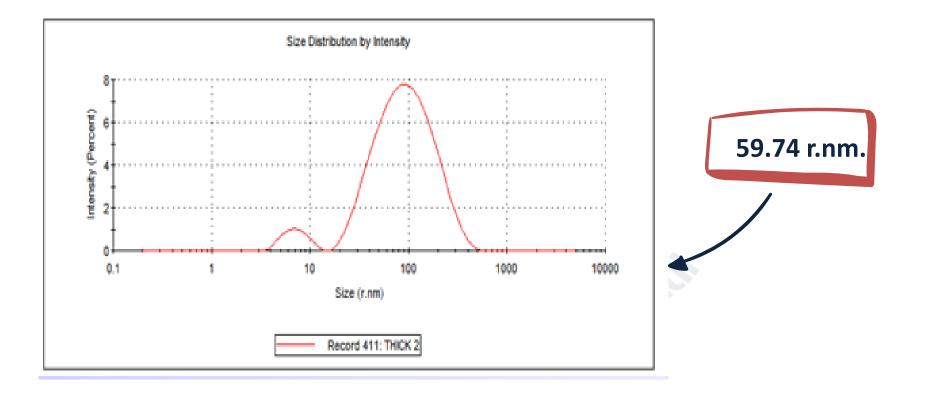
• UV-visible spectrophotometer:







• Dynamic Light Scattering evaluation:

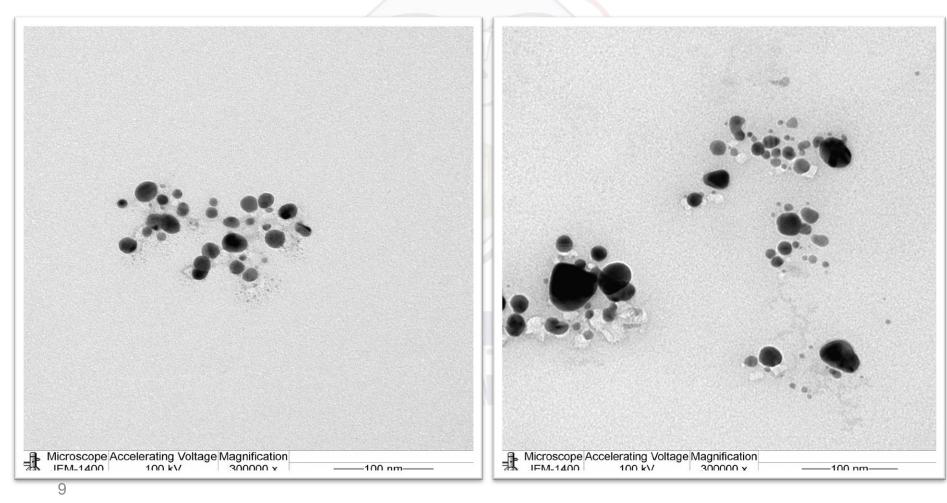






• Transmission electron microscope:

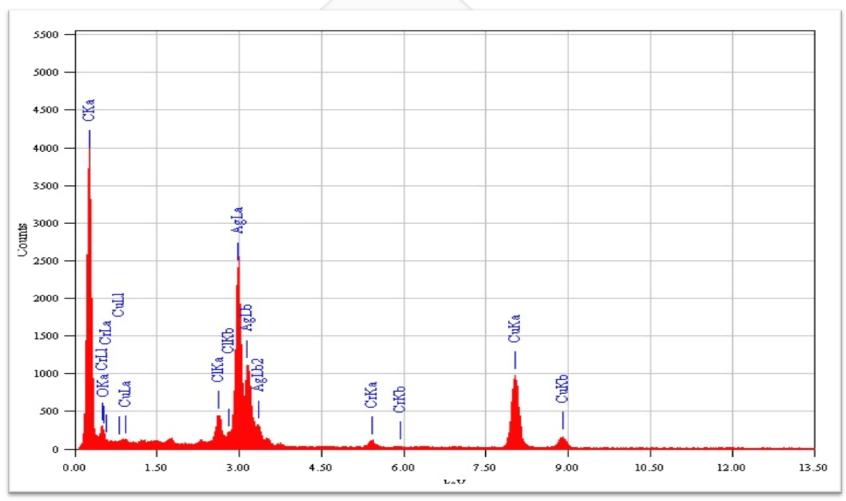
• AgNPs are agglomerated and spherical in shape.







• Energy dispersive x-ray analysis:

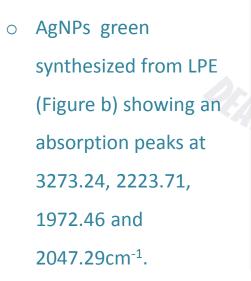


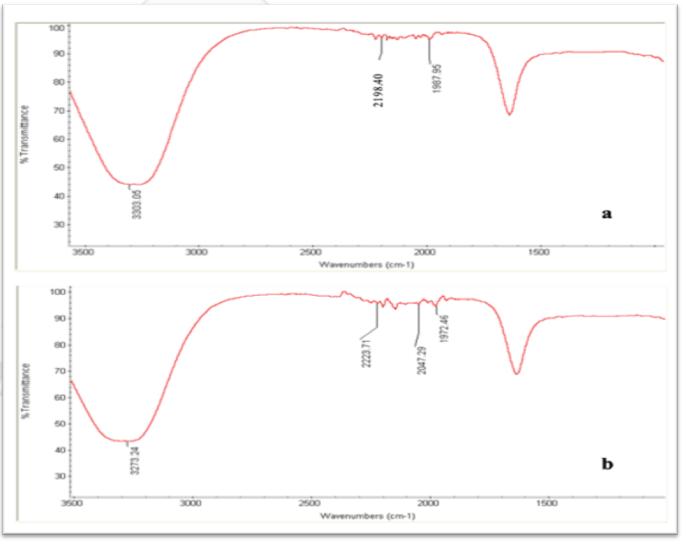




• Fourier-transform infrared spectroscopy :

 LPE (Figure a) showing an absorption peaks at 3303.05, 2198.40 and 1987.95 cm⁻¹.





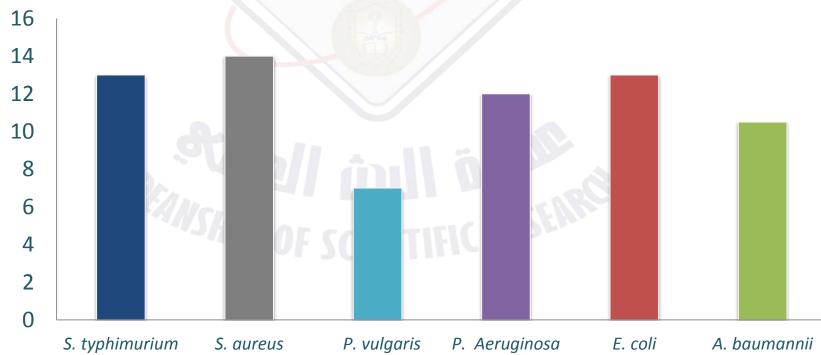


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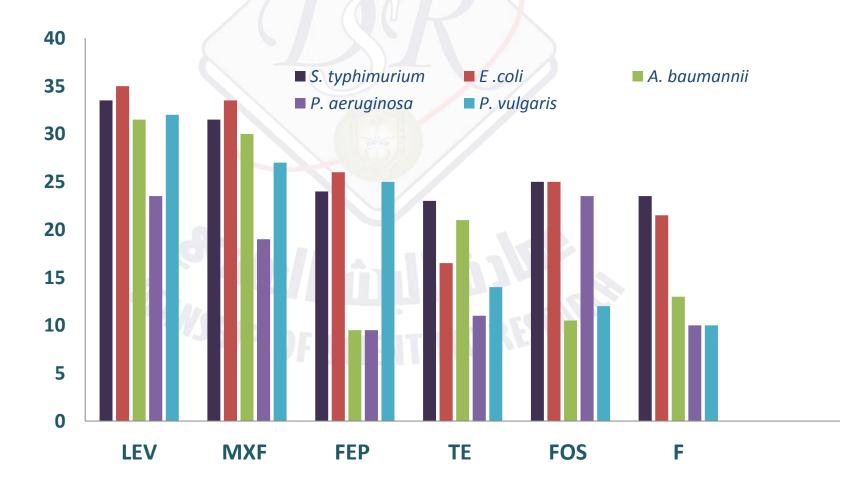
- Determination of the antibacterial activity of the green
 synthesized silver nanoparticles:
 - S. typhimurium
 S. aureus
 P. vulgaris
 P. Aeruginosa
 E. coli
 A. baumannii







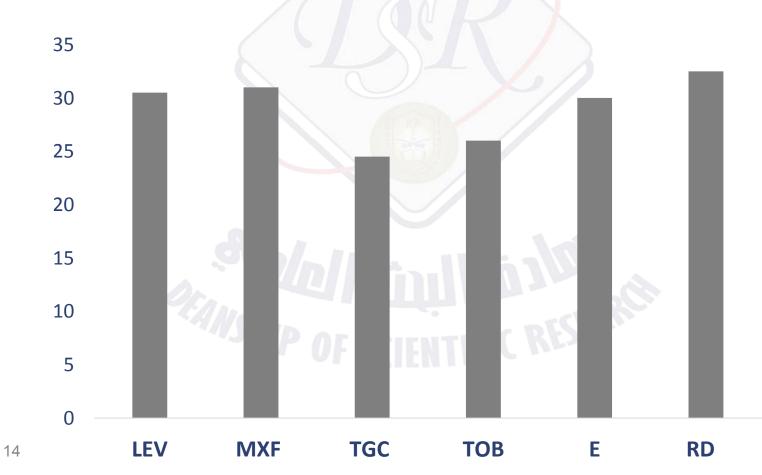
 Combination effect of the green synthesized silver nanoparticles with Antibiotics on gram negative bacteria:







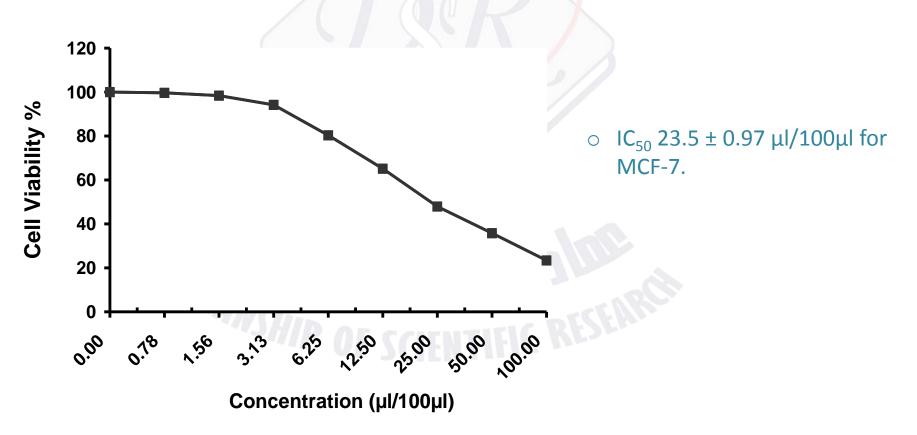
 Combination effect of the green synthesized silver nanoparticles with Antibiotics on gram positive bacteria *S. aureus*:





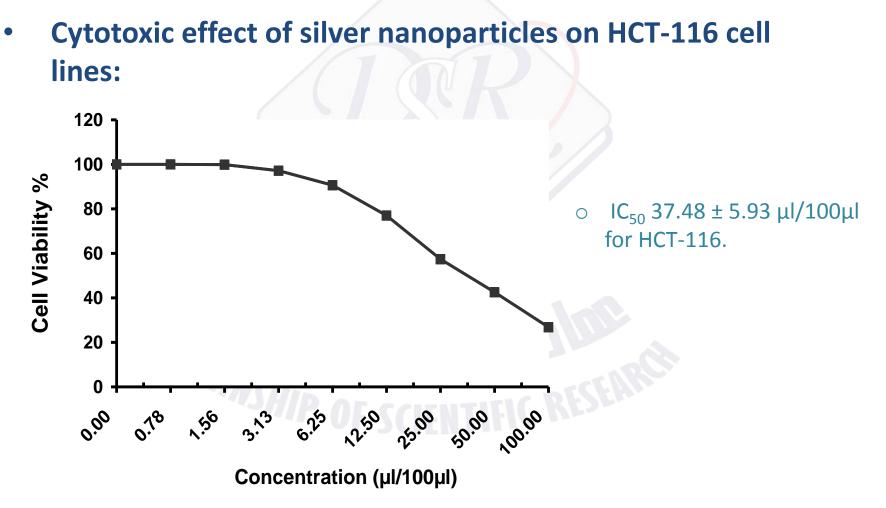


• Cytotoxic effect of silver nanoparticles on MCF-7 cell lines:









Discussion:

UV- visible spectrophotometer & DLS

UV peak was attributed to the excitation of the surface Plasmon resonance. DLS evaluation, the semi-broad peak is due to the variation in size, or an aggregated structure of AgNPs.

Transmission Electron Microscopy

The aggregation is due to the layer covering the NPs, this layer causes the NPs to be attached to each other resulting in a decreased space.

Energy Dispersive X-ray

The absorption peak observed at 3 KeV indicates the presence of the element silver (Velmurugan et al., 2012).

Fourier-transform infrared spectroscopy

From the comparison between the spectrums, the shifts in the peaks positions indicates the presence of the functional groups that reduced the silver ions to silver nanoparticles.







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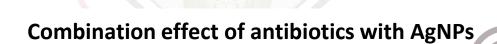


Discussion:



The antibacterial effect

Green synthesized AgNPs showed good antibacterial effect against tested bacteria, particularity on *S. aureus*.



best effect was on *A*. *baumannii*, zone of inhibition size was 10.5 mm, with Moxifloxacin, the zone of inhibition enhanced to 30 mm.



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Cytotoxic activity

Synthesized AgNPs displayed the best effect on MCF-7. Effect might be due to the ability to stimulate the reactive oxygen species on the cellular components, resulting in cell death (Venuegopal *et al.,* 2017).







Conclusion

To conclude, AgNPs synthesized from *Citrus limon* peel extract showed the best antibacterial effect on *Staphylococcus aureus*. Yet when combining the AgNPs with the antibiotics, an enhanced and decreased inhibition zone size was observed. Moreover, the cytotoxic effect of the AgNPs showed the best effect against MCF-7 cell lines.

Recommendation

In the future, we recommend optimizing the conditions when synthesizing the AgNPs, as well as understanding the mode of action and determining the toxic effects on animals, in order to employ the AgNPs in the field of medicine as a possible therapeutic agents.





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References:

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- Venugopal, K., Ahmad, H., Manikandan, E., Thanigai Arul, K., Kavitha, K., Moodley, M., Rajagopal, K., Balabhaskar, R. and Bhaskar, M. (2017). The impact of anticancer activity upon Beta vulgaris extract mediated biosynthesized silver nanoparticles (ag-Nps) against human breast (MCF-7), lung (A549) and pharynx (Hep-2) cancer cell lines. *Journal of Photochemistry and Photobiology B: Biology*, 173, pp.99-107.

