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Synthesis and pharmacological activity of Nickel Oxide nanoparticles from *Tadehagi Triquetrum*

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Abstract
Over the past few decades, nanotechnology has become a prominent field of study in modern material science. Green synthesized nanoparticles have garnered wide interest due to its inherent features like rapidity, eco-friendly and cost effectiveness. The aim of the study indicates the synthesis of nickel oxide nanoparticles utilizing nickel chloride hexahydrate ($NiCl_2 .6H_2O$) as a precursor and *Tadehagi triquetrum* (TT) leaf extract as a reducing agent and its anti-bacterial activity. The biosynthesized nickel oxide nanoparticles from *Tadehagi triquetrum* leaf extract were characterized by UV-Vis spectroscopy, Fourier Transform Infrared Spectroscopy (FTIR), X-Ray Diffraction (XRD) analysis, Scanning Electron Microscopy (SEM) and Energy Dispersive X-ray analysis (EDX). The UV -Visible spectroscopy of nickel oxide nanoparticles showed absorbance at 290 nm. FTIR spectroscopy was used to analyze the specific functional groups responsible for reduction, stabilization and capping agents present in the sample. The XRD results show that the nickel oxide nanoparticle is in amorphous state. The SEM analysis is to characterize the morphology of the nickel oxide nanoparticles. The nickel oxide nanoparticles showed effective antibacterial activity against *Staphylococcus aureus*, and *E. coli*. Thus, this study proves that nickel oxide nanoparticles would contain natural anti-bacterial agents through green synthesis.

Keywords: NiO nanoparticles, green synthesis, *Tadehagi Triquetrum* leaves

Introduction

Nano-science breakthroughs in almost every field of science and nanotechnologies make life easier in this era. It is revolutionizing many industrial and technological field due to the fact that it is possible with nanotech to orient material structures at extremely small scales, thereby extending science tool kit [1]. The earliest, widespread description of nanotechnology referred to the particular technological goal of precisely manipulating atoms and molecules for fabrication of macroscale products, also now referred to as molecular nanotechnology [2]. The study, exploitation, and use of materials and technologies at the nanoscale level is referred to as nanotechnology. The Greek word "nannos," which means dwarf or incredibly little, is where the word "nano" originates [3]. Nano-science is the study of science that deals with objects or matter at the nanoscale, and nanotechnology is the technology built upon this science. It entails working with nanoparticles, which are minuscule particles that usually range in size from one to 100 nanometers. A billionth of a meter is called a nanometer [4]. Nanoparticles are the fundamental building blocks in nanotechnology. These are the particles with dimensions at the nanoscale or a class of materials with properties distinctively different from their bulk and molecular counterparts [5].

The synthesis of nanoparticles involves the creation of these tiny structures from different starting materials using various methods and techniques. These methods, which include chemical, physical, and biological approaches, offer precise control over nanoparticle size, shape, composition, and properties. The nanoma terials are synthesized by various methods that are categorized in to bottom up or top-down method [6]. The two distinct methods are employed in the production of nanomaterials, including nanoparticles: top-down and bottom-up. These words describe the process by which the material is created or synthesized: top-down, where the material is created from bulk and reduced to nanoscale dimensions, or bottom-up, when the material is created from individual atoms or molecules to form nanoparticles [7].

Bio-technology and nanotechnology are connected through the green chemistry approach of biological synthesis of nanoparticles. The pace of synthesis is sluggish and the biological nanoparticles are not monodispersed despite their stability [8]. Synthesis of nanoparticle by plants is a green chemistry approach. Plant extract are used for the metal ions bio reduction to form nanoparticles [9]. The term "green synthesis" describes a sustainable technique of creating nanoparticles with ingredients and processes that are kind to the environment [10]. Green synthesis brings a clean, safe, cost effective, sustainable and environment friendly method to synthesize nanoparticles [11]. Green method of synthesis includes replacing hazardous toxic chemicals while developing new strategies to minimize or eradicate harmful chemicals and byproducts hazardous to health and environment [12].

Materials and methods

1. Materials

- *Tadehagi triquetrum* leaves
- Nickel chloride hexahydrate
- Distilled water
- Ethanol

2. Methods

2.1 Preparation of crude sample

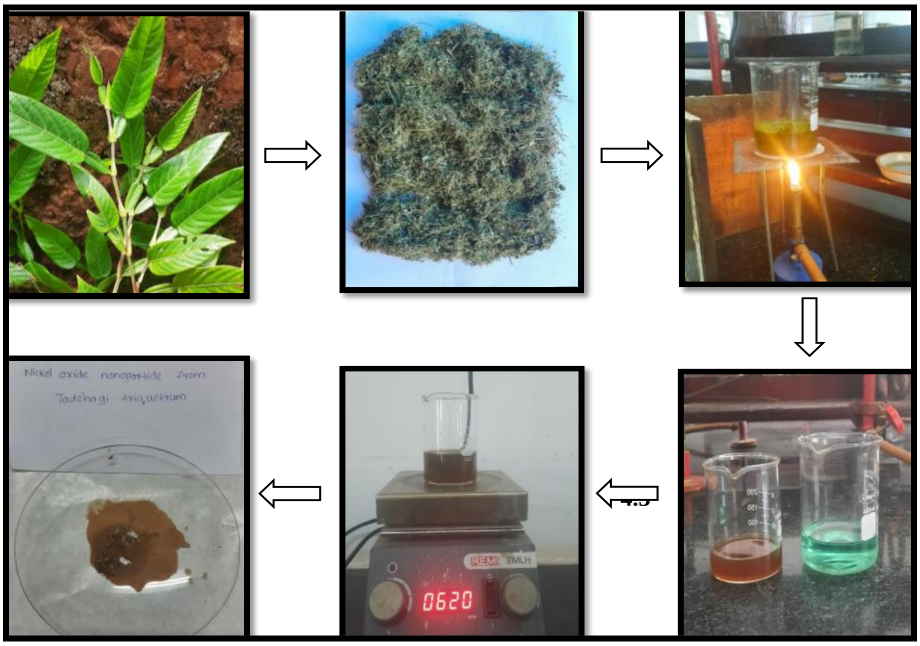
Tadehagi triquetrum is a traditional ethnic herb that is collected from Chemmarathur, Kozhikode, Kerala, were washed with distilled water and dried under shade. After shade drying, the leaves were made into fine powder using mechanical blender and transferred into air tight container.

2.2 Preparation of *Tadehagi triquetrum* leaf extract

Accurately 5g of powder of *Tadehagi triquetrum* leaves were weighed out and was added to 100 ml distilled water in a 400 ml beaker and heated for 40 minutes with constant stirring until the solution get brown colour. Then the solution kept aside for settle and filter the solution.

Green synthesis of nickel oxide nanoparticles

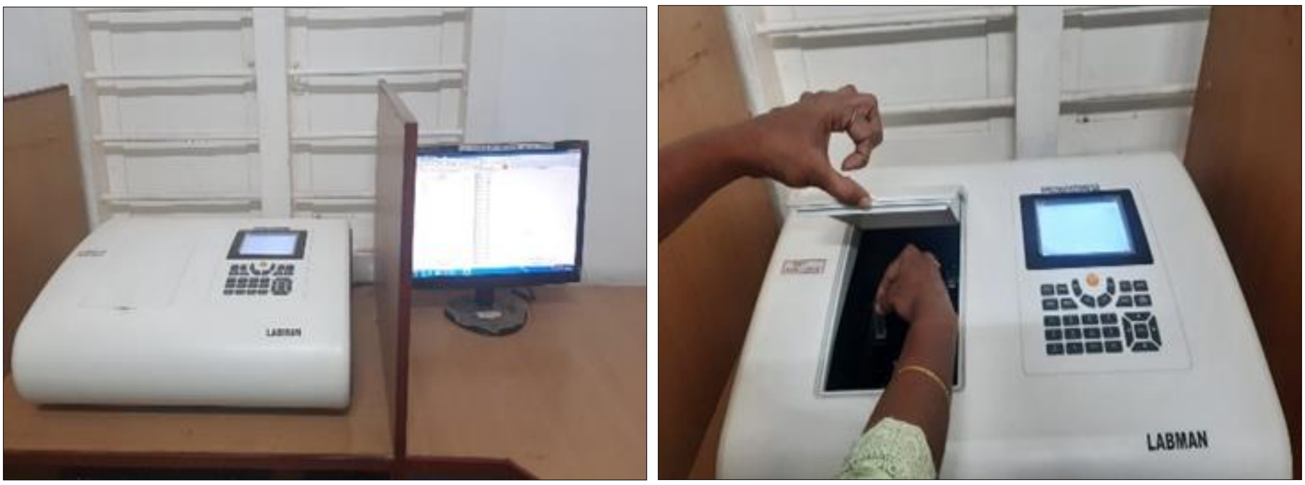
2.4g of nickel chloride hexahydrate ($NiCl_2$.$6H_2O$) was dissolved in 100 ml of distilled water. Shaken well. 50 ml of *Tadehagi triquetrum* leaf extract was added to 50 ml of $NiCl_2$.$6H_2O$ solution in 1:1 ratio with constant stirring. Then the mixture was placed at magnetic stirrer at ambient temperature for one hour, until a precipitate is formed. The formation of precipitate indicates the formation of nickel oxide nanoparticles. After stirring, the solution was centrifuged and the precipitate washed with alcohol. The precipitate was dried using oven and used for characterization of nickel oxide nanoparticle



Characterization of metal oxide nanoparticle

1. UV-Visible spectroscopy

The most important and simple technique for conforming the formation of nanoparticles is ultraviolet spectroscopy.



UV-Visible spectrometer

Ultraviolet-visible spectroscopy or visible spectroscopy or ultraviolet spectrophotometer involves the spectroscopy of photons in the UV-Visible region. The pure Nickel oxide nanoparticles were monitored by measuring the UV-Visible spectrometer by diluting a small aliquot of the sample into ethanol. UV-Vis spectroscopic analysis of nickel oxide nanoparticle synthesized from *Tadehagi triquetrum* was performed at Nirmala College for Women, Coimbatore

2. FT-IR Analysis

The functional group present in the sample was recorded using Fourier transform infrared spectroscopy. In FTIR Spectroscopy, the light is directed onto the sample of interest, and the intensity is measured using an infrared detector. FTIR analysis of green synthesized nickel oxide nanoparticle was performed at Avinashilingam Institute of Home Science and Higher Education for Women (Bharat Ratna prof.CNR Rao research Centre), Coimbatore.

3. X Ray diffraction analysis

The crystalline nature of the synthesized nickel oxide nanoparticle was analysed using X-ray diffraction. X-ray diffraction, a phenomenon in which the atoms of a crystal, by virtue of their uniform spacing, cause an interference pattern of the waves present in an incident beam of X-rays. The atomic planes of the crystal act on the X-rays in exactly the same manner as does a uniformly ruled diffraction grating on a beam of light. The average crystalline equation size of nickel oxide nanoparticles was calculated using the Debye Scherrer's equation D=KA/ B cos θ

Where D is the size of the particle, K is the Debye Scherer's constant which is equal to 904, Ais the Wavelength value equal to 0.15406, f is the full width half maximum of the diffracted peak and 8 is the Bragg's angle for the peak [13]. The XRD analysis of green synthesizes nickel oxide nanoparticle was performed at Avinashilingam Institute of Home Science and Higher Education for Women (Bharat Ratna prof.CNR Rao research Centre), Coimbatore.

4. Scanning Electron microscope (SEM) Study

The sample's surface topography and composition were recorded for the nickel oxide nanoparticles by using Scanning electron microscope (SEM). In most applications, data are collected over a selected area of the surface of the sample, and a 2-dimensional image is generated that displays spatial variations in these properties [14]. The dried sample was placed on a double conductive tap fixed on a sample holder at a normal temperature [14]. SEM analysis of nickel oxide nanoparticle synthesized by *Tadehagi triquetrum* plant extract was performed at Avinashilingam Institute of Home Science and Higher Education for Women (Bharat Ratna prof.CNR Rao research Centre), Coimbatore.

5. Energy Dispersive X-Ray (EDAX) Analysis

Energy Dispersive X-Ray Analysis, also referred to as EDS or EDAX, is an X-ray technique used to identify the elemental composition of materials. DAX systems are attachments to Electron Microscopy instruments (Scanning Electron Microscopy (SEM) or Transmission Electron Microscopy (TEM)) instruments where the imaging capability of the microscope identifies the specimen of interest [15]. The data generated by EDX analysis consist of spectra showing peaks corresponding to the elements making up the true composition of the sample being analyzed [15]. Elemental mapping of a sample and image analysis are also possible. The EDAX analysis along with SEM analysis of nickel oxide nanoparticle performed at Avinashilingam Institute of Home Science and Higher Education for Women (Bharat Ratna prof.CNR Rao research Centre), Coimbatore.

Biological activity

1. Anti-bacterial activity

The antibacterial activity of green synthesized NiO nanoparticles was analysed using Kirby-Bauer method. For that at least three to five well-isolated colonies of the same morphological type are selected from an agar plate culture. The top of each colony is touched with a loop, and the growth is transferred into a tube containing 4 to 5 ml of a suitable broth medium, such as Nutrient broth. The broth culture is incubated at 35°C until it achieves or exceeds the turbidity (usually 2 to 6 hours). The turbidity of the actively growing broth culture is adjusted with sterile saline or broth to obtain turbidity. This results in a suspension containing approximately 1 to 2 x 108 CFU/ml for *Staphylococcus aureus*, and *E. coli*. Optimally, within 15 minutes after adjusting the turbidity of the inoculum suspension, a sterile cotton swab is dipped into the adjusted suspension. The swab should be rotated several times and pressed firmly on the inside wall of the tube above the fluid level. This will remove excess inoculum from the swab. The dried surface of a Nutrient agar plate is inoculated by streaking the swab over the entire sterile agar surface. This procedure is repeated by streaking two more times, rotating the plate approximately 60° each time to ensure an even distribution of inoculum. As a final step, the rim of the agar is swabbed. The lid may be left ajar for 3 to 5 minutes, but no more than 15 minutes, to allow for any excess surface moisture to be absorbed before applying the drug impregnated disks. The media was punctured by making a well of 6 mm in diameter and filled with sample. Further the petri plates were placed inversely for complete diffusion and inhibition zones were examined by measuring the diameter (mm) formed around the well after 24 hrs incubation at 37°C. The zones were measured by using standard (Hi-Media) scale. The antio-bacterial activity of green synthesised nickel oxide nanoparticle performed at Avinashilingam Institute of Home Science and Higher Education for Women (Bharat Ratna prof.CNR Rao research Centre), Coimbatore.

Results and discussion

The nickel oxide nanoparticles were successfully synthesized in an easy ecofriendly way using *Tadehgi triquetrum* leaves extract as reducing agent. The current study involves the screening of leaf extract of *Tadehagi triquetrum* for the synthesis of nickel oxide nanoparticles which was characterized using UV-Visible spectroscopy, FTIR, XRD and FE-SEM. Elemental analysis was done by using EDAX analysis. The biological activity of nickel oxide nanoparticle was also investigated.

1. Characterization of Green Synthesized Nickel Oxide Nanoparticles

1.1 UV-Visible spectroscopy

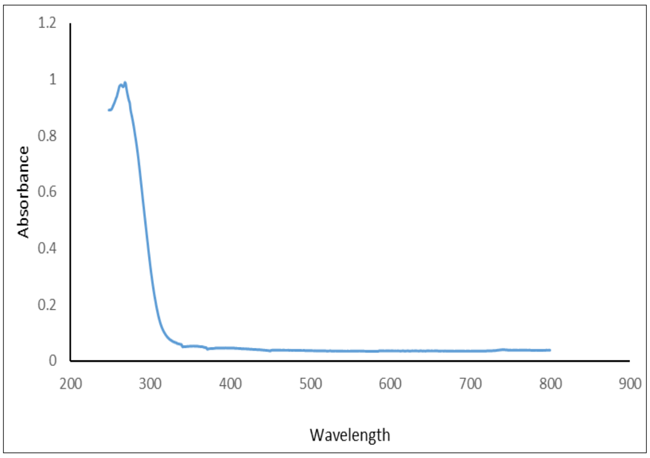


Fig 1: UV-Visible graph of nickel oxide nanoparticles

The UV-Visible spectrum of nickel oxide nanoparticle shown in the figure 1. The uv visible spectrum of nickel oxide nanoparticle synthesized from *Tadehaggi triquetr*um leaf extract shows absorbance at 268 nm. The characteristic absorbance peak of nickel oxide nanoparticles in the range of 200nm to 400 nm [16]. The UV-Visible spectroscopy confirms the formation of nickel oxide nanoparticles.

1.2 FT-IR Analysis

The FT-IR results of synthesized nickel oxide nanoparticles shown in the table 1. The peak at 2970.38 cm^{-1} shows the presence of C-H stretching. The peak at 2738.92 cm^{-1} shows the presence of O-H starching. The peak at 1597.06 cm^{-1} shows the presence of C=C stretching. The peak at 1280.73 cm^{-1} shows the presence of C=O stretching. The peak at 686.66 cm^{-1} shows the presence of C-H bending. The nickel-oxide stretching shows peak at 486.06 cm^{-1}. [17]

Table 1: FT-IR analysis of nickel oxide nanoparticles

Peak value	Possible groups
2970.38 cm^{-1}	C-H Stretching
2738.92 cm^{-1}	O-H Stretching
1597.06 cm^{-1}	C=C Stretching
1280.73 cm^{-1}	C=O Stretching
686.66 cm^{-1}	C-H Bending
486.06 cm^{-1}	Ni-O Stretching

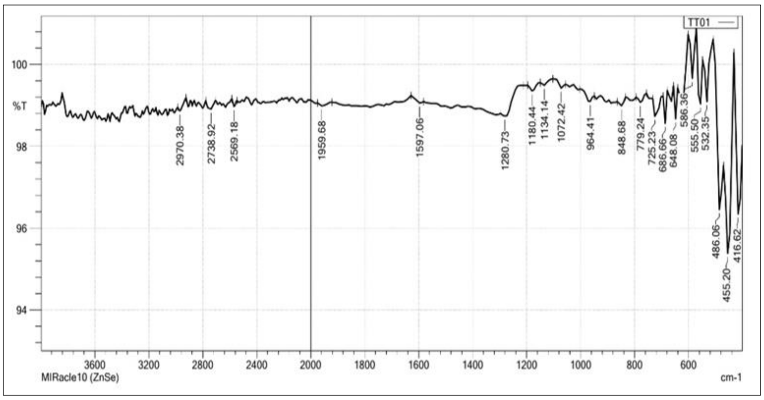


Fig 2: FT-IR spectrum of nickel oxide nanoparticles

1.3 XRD analysis

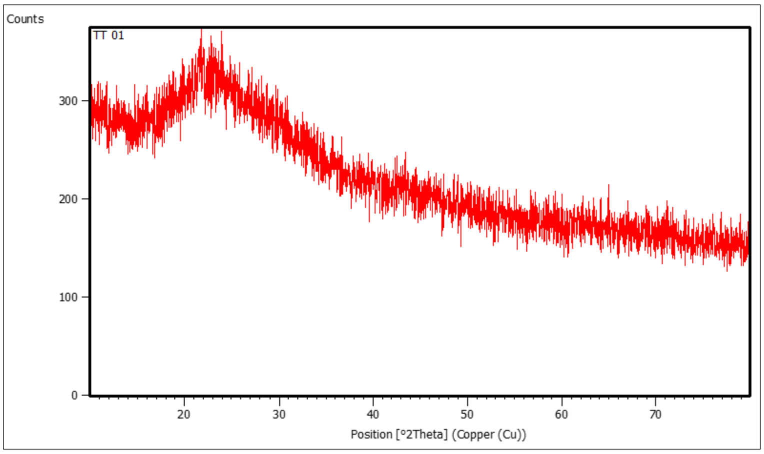
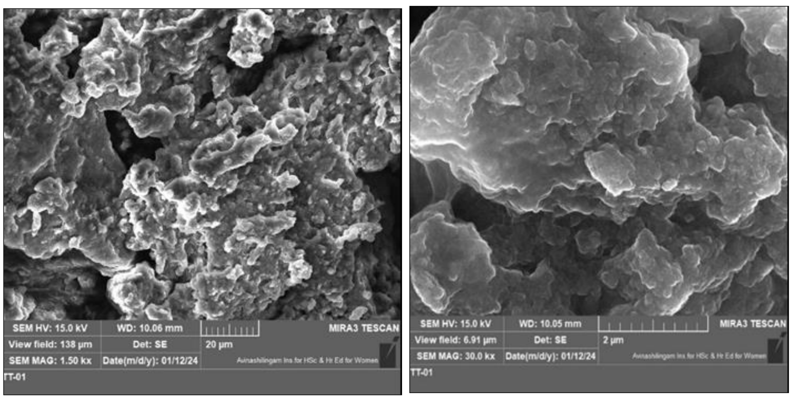


Fig 3: XRD pattern of nickel oxide nanoparticles

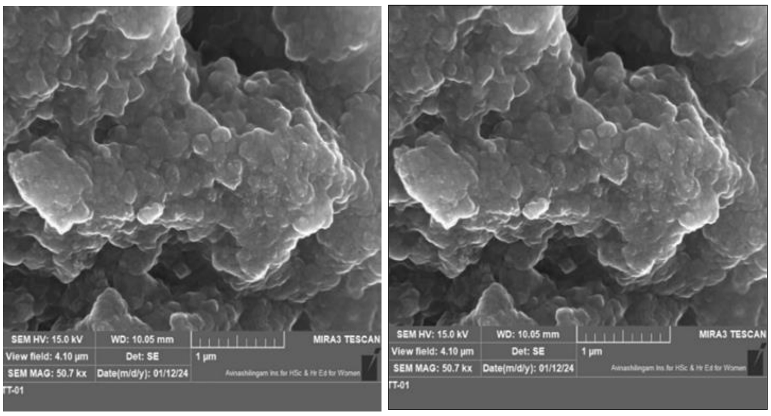
XRD pattern of synthesized nickel oxide nanoparticles using *Tadehagi triquetrum* extract shown in the figure 3. From the graph it can be seen that there are broad peaks are present. Henceforth the compound can be concluded as the synthesized nickel oxide nanoparticle is amorphous in nature.

1.4 SEM analysis

The structural features of the nickel nanoparticles that synthesized from *Tadehagi triquetrum* leaf extract were investigated through scanning electron microscopy. The SEM image in the figure 4 showed that the nickel oxide nanoparticles are floral in shape



I ii



iii iv

Fig 4: SEM images of nickel oxide nano particle (i, ii, iii, iv)

1.5 EDAX Analysis

The elemental composition of the nickel oxide nanoparticles that is synthesized from *Tadehagi triquetrum* was confirmed using EDAX spectrum. The EDAX spectrum revealed the presence nickel and oxygen along with carbon and chlorine. Nickel oxide nanoparticle sample constitute 6.98% of nickel and 31.9% of oxygen. So, this result confirms the formation of nickel oxide nanoparticles. Figure 5 shows the EDAX spectrum of nickel oxide nanoparticles

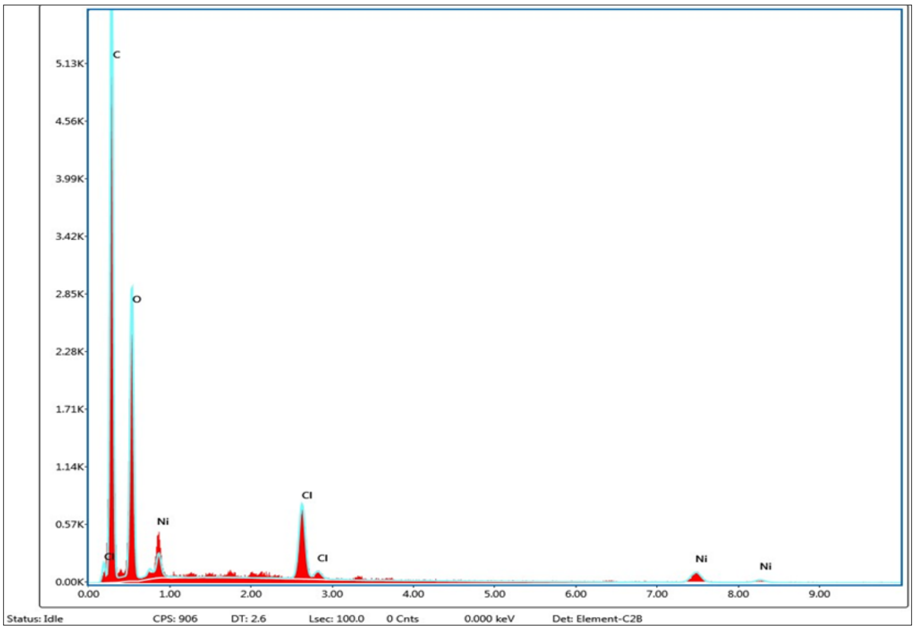


Fig 5: EDAX graph of nickel oxide nanoparticles

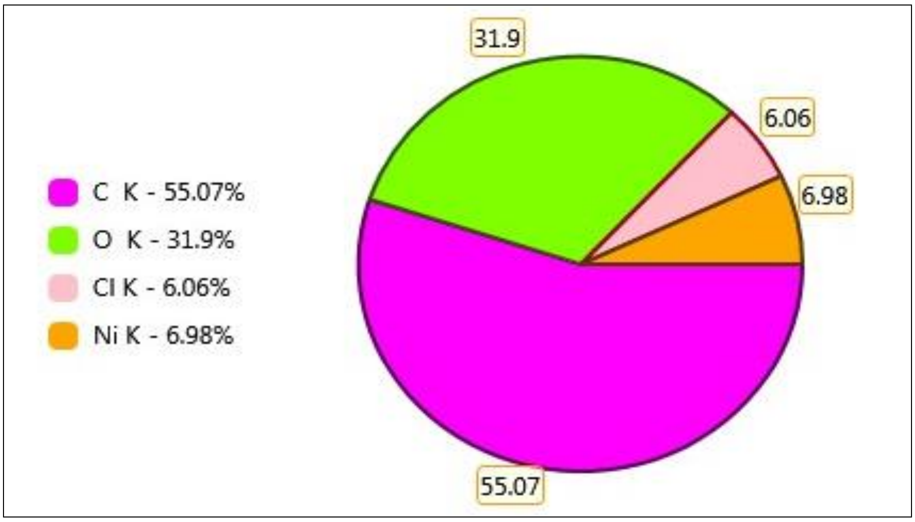


Fig 6: pie diagram EDAX analysis of nickel oxide nanoparticle

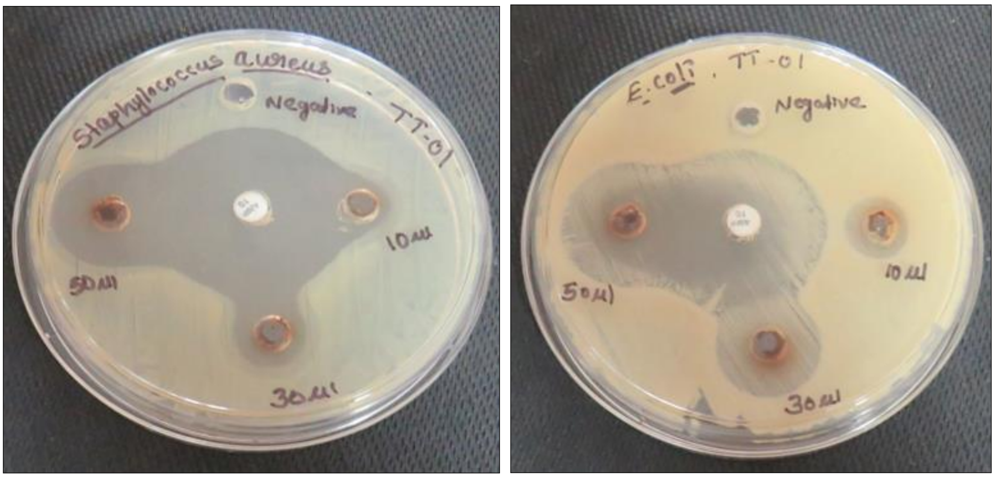
2. Biological Activity of Green Synthesized Nickel Oxide Nanoparticles

2.1 Anti-bacterial activity

The anti-bacterial activity of green synthesized nickel oxide nanoparticle was analyzed using Kirby-Bauer method. The nickel oxide nanoparticle was treated against with both gram-negative bacteria (*Escherichia coli*) and gram-positive bacteria (*Staphylococcus aures*). The green synthesized nickel oxide nanoparticles at 10 µl shows zone of inhibition against *staphylococcus aures* is 13 mm and *Escherichia coli* is 12 mm. The green synthesized nickel oxide nanoparticles at 30 µl shows zone of inhibition against *staphylococcus aures* is 14 mm and *Escherichia coli* is 20 mm. The green synthesized nickel oxide nanoparticles at 50 µl shows zone of inhibition against *staphylococcus aures* is 22 mm and *Escherichia coli* is 24 mm. The result shows that the nickel oxide nanoparticle synthesized using *Tadehagi triquetrum* leaf extract possess maximum zone of inhibition against *Staphylococcus aures* and *Escherichia coli*. The results of antibacterial activity are present in the table 2.

Table 2: Antibacterial activity of nickel oxide nanoparticles

Sample: TT-O1	Zone of Inhibition (mm)	
	Staphylococcus aureus	***Escherichia coli***
Control * (Ampicillin)	35mm	35mm
Negative control (Ethanol)	0mm	0mm
10µl	13mm	12mm
30µl	14mm	20mm
50µl	22mm	24mm



(i) **(ii)**

Fig 7: Antibacterial activity of green synthesized nickel oxide nanoparticle against *Staphylococcus aures* (i) and *Escherichia coli* (ii)

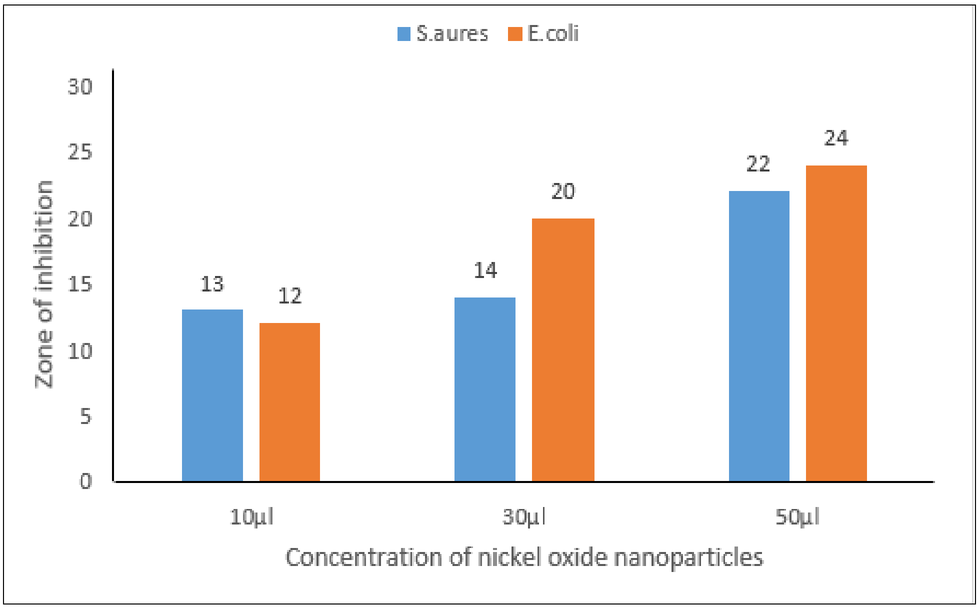


Fig 8: Bar chart for the antibacterial activity

The above results shows that the antibacterial activity of nickel oxide nanoparticles increases with increase in concentration for both gram-negative and gram-positive bacteria. From the result it can also shows that the green synthesized nanoparticles have higher activity against gram negative bacteria (*Escherichia coli)* than gram positive bacteria (*Staphylococcus aures)*

Conclusion

This study demonstrates the successful synthesis of nickel oxide nanoparticles utilizing nickel chloride hexahydrate ($NiCl_2 .6H_2O$) as a precursor and *Tadehagi Triquetrum* (TT) plant leaf extracts as a reducing agent. The biosynthesized Nickel oxide nanoparticles were characterized by UV-Vis spectrometer, Fourier transform infrared spectrometry

(FTIR), X-ray diffraction (XRD) and scanning electron microscope (SEM) coupled with energy dispersive microanalysis (EDAX). The UV -Vis spectrometer shows absorbance at 268 nm. FTIR spectroscopy was used to analyze the specific functional groups responsible for reduction, stabilization and capping agents present in the nanoparticles. The nickel-oxide stretching shows peak at 486.06 cm^{-1}. The XRD results show that the nickel oxide nanoparticle is in amorphous state. The morphology of the nickel oxide nanoparticles which was characterized by SEM shows that it is floral in shape. The EDAX confirms the nickel oxide nanoparticles composition. The ability of bacteria with synthesized NiO nanoparticles tested using Kirby-Bauer method were discussed. The nickel oxide nanoparticles showed effective antibacterial activity against *Staphylococcus aureus*, and *E. coli*. Thus, this study proves that nickel oxide nanoparticles would contain natural anti-bacterial agents through green synthesis.

References

1. Uddin S, Safdar LB, Anwar S, Iqbal J, Laila S, Abbasi BA, *et al*. Green synthesis of *nickel oxide nanoparticles* from Berberis balochistanica stem for investigating bioactivities. Molecules,2021:26(6):1548.
2. Bayda S, Adeel M, Tuccinardi T, Cordani M, Rizzolio F. The history of *nanoscience* and *nanotechnology*: from chemical–physical applications to *nanomedicine*. Molecules,2019:25(1):112.
3. Keiper A. The *nanotechnology* revolution. The New Atlantis,2003:1(2):17-34.
4. Biswas P, Wu CY. *Nanoparticles* and the *environment*. Journal of the air & waste management association,2005:55(6):708-746.
5. Bhaviripudi S, Mile E, Steiner SA, Zare AT, Dresselhaus MS, Belcher AM, *et al*. CVD synthesis of single-walled carbon *nanotubes* from gold *nanoparticle* catalysts. Journal of the American Chemical Society,2007:1296(6):1516-1517.
6. Arole VM, Munde SV. Fabrication of *nanomaterials* by top-down and bottom-up approaches-an overview. Journal of Material Science,2014:1:89-93.
7. Jamkhande PG, Ghule NW, Bamer AH, Kalaskar MG. Metal *nanoparticles* synthesis: An overview on methods of preparation, advantages and disadvantages, and applications. Journal of drug delivery science and technology,2019:1(53):101-174.
8. Kahn KB. Revisiting top-down versus bottom-up forecasting. The Journal of Business Forecasting. 1998:17(2):14.
9. Tripathy S, Rodrigues J, Shimpi NG. Top-down and Bottom-up Approaches for Synthesis of *Nanoparticles*. *Nanobiomaterials*: Perspectives for Medical Applications in the Diagnosis and Treatment of Diseases,2023:145:92-130.
10. Iravani S. Green synthesis of metal *nanoparticles* using plants. Green Chemistry,2011:13(10):2638-2650.
11. Parveen K, Banse V, Ledwani L. Green synthesis of *nanoparticles*: Their advantages and disadvantages. In AIP conference proceedings,2016:1724(1).
12. Hamouda RA, Hussein MH, Abo-Elmagd RA, Bawazir SS. Synthesis and biological characterization of silver *nanoparticles* derived from the cyanobacterium Oscillatoria limnetica. Scientific reports,2019:9(1):13071.
13. Akbari B, Tavandashti MP, Zandrahimi M. Particle size characterization of *nanoparticles*–a practical approach. Iranian Journal of Materials Science and Engineering,2011:8(2):48-56.
14. Srivastava R. Synthesis and Characterization Techniques of *Nanomaterials*. International Journal of Green *Nanotechnology*,2012:4(1):17-27.
15. Singh V, Shrivastava and Wahi N. Biosynthesis of silver *nanoparticles* by plants crude extracts and their characterization using UV, XRD, TEM and EDX. African Journal of Biotechnology,2015:14(33):2554-2567.
16. Iqbal J, Abbasi BA, Mahmood T, Hameed S, Munir A, Kanwal S. Green synthesis and characterizations of *Nickel oxide nanoparticles* using leaf extract of Rhamnus virgata and their potential biological applications. Applied Organometallic Chemistry,2019:33(8):4950.
17. Olajire AA, Mohammed AA. Green synthesis of *nickel oxide nanoparticles* and studies of their photocatalytic activity in degradation of polyethylene films. Advanced Powder Technology,2020:31(1):211-218.