

Biosynthesis of gold nanoparticles from the seaweed for the antibacterial activities

Submitted in partial fulfilment of the requirements for the award of

Bachelor of Technology degree in Biotechnology

By

K . GOWSIGA (38230014)



DEPARTMENT OF BIOTECHNOLOGY

SCHOOL OF BIO AND CHEMICAL ENGINEERING

SATHYABAMA

INSTITUTE OF SCIENCE AND TECHNOLOGY (DEEMED TO BE UNIVERSITY)

Accredited with Grade “A” by NAAC

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APRIL 2022



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DEPARTMENT OF BIOTECHNOLOGY

BONAFIDE CERTIFICATE

This is to certify that this Project Report is the bonafide work of **K. GOWSIGA (48230014)** who carried out the project entitled **“Biosynthesis of GOLD nanoparticles from seaweed for the antibacterial activities”** under our supervision from January 2022 to April 2022.

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DECLARATION

I, **K.GOWSIGA (38230014)** hereby declare that the Project Report entitled “**Biosynthesis of GOLD nanoparticles from seaweed for the antibacterial activities**” done by us under the guidance of **Dr.R.THIYAGARJAN** is submitted in partial fulfillment of the requirements for the award of Bachelor of Technology degree in Biotechnology.

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ACKNOWLEDGEMENT

I am pleased to acknowledge my sincere thanks to Board of Management of SATHYABAMA for their kind encouragement in doing this project and for completing it successfully. I am grateful to them.

I convey my thanks to Dr. Ramesh Kumar. V, Head of the Department, Dept. of Biotechnology for providing me necessary support and details at the right time during the progressive reviews.

I would like to express my sincere and deep sense of gratitude to my Project Guide Dr.R.Thiyagarjan for his valuable guidance, suggestions and constant encouragement paved way for the successful completion of my project work.

I would like to express my sincere and deep sense of gratitude to my Project Guide Dr Rajasekar T for his valuable guidance, suggestions and constant encouragement paved way for the successful completion of my project work.

I wish to express my thanks to all Teaching and Non-teaching staff members of the Department of Biotechnology and Centre for the Drug Discovery and Development who were helpful in many ways for the completion of the project.

ABSTRACT

Green synthesis technology uses microbes or plants to make nanoparticles that are biologically safe, cost-effective, and environmentally benign. The ability of a biological system to use its own organic chemistry processes to modify inorganic metal ions into nanoparticles has opened up a hitherto unknown field of biochemical research. The potential of a biological system to use its inherent organic chemistry processes to modify inorganic metal ions into nanoparticles has opened up a hitherto unknown field of biochemical research. When nanotechnology is combined with biology, a new field called nanobiotechnology emerges, involving living organisms of both prokaryotic and eukaryotic origin. And in this study, synthesis of gold nanoparticles from marine microalgae also called seaweed and checking its characterizations. The use of nanotechnology for medication delivery and tissue engineering has made significant contributions to translational research on pharmaceutical products and their applications.

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LIST OF SYMBOLS AND ABBREVIATIONS

AUCL3	Gold chloride
GNPs	Gold nanoparticles
TEM	Transmission electron microscopy
SEM	Scanning electron microscopy
XRD	X-ray diffraction
FTIR	Fourier transform infrared spectroscopy
UV-VIS	Ultraviolet visible spectroscopy

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1.INTRODUCTION

Gold nanoparticles may be used in different domains, one of most important being in the biomedical field. They have suitable properties for controlled drug delivery, cancer treatment, biomedical imaging, diagnosis and many others, due to their excellent compatibility with the human organism, low toxicity and tunable stability, small dimensions, and possibility to interact with a variety of substance. Gold nanoparticles are used in resonance scattering dark field microscopy for the detection of microbial cells and their metabolites, the bio-imaging, the bio-imaging of tumor cells, and for the detection of receptors on tier surface and for the study of endocytosis. There is increasing optimism that nanotechnology applied to medicine and dentistry will bring significant advances in the diagnosis, treatment, and prevention of disease. Growing interests in the future medical application nanotechnology is leading to the emergence of a new field called nanomedicine. Particles are engineered so that they are attracted to diseased cells, which allows direct treatment of those cells. This technique reduces damage to healthy cells in the body and allows for earlier detection of disease. It is beyond doubt that modern medicine can benefit greatly from it; thus nanomedicine has become one of the main branches of nanotechnological research. Currently it focuses on developing new methods of preventing, diagnosing and treating various diseases.

This study using water extract from different bio-based plants product from an aqueous solution to check their potential to form metallic nanoparticles. For clinical diagnosis of folin-ciocalteus's reagent, for synthesis gallic acid 1-hydrate,99% is used for analysis sodium anhydrous(panreac Quimica, Barcelona, Spain).plant extract were prepared by a total of 1g of dry bio-product mixed with 100ml of deionized water. an only on the extract from brown algae *sargassum muticum* was studied balance data are plant extract for the synthesis of metal nanoparticles a volume of 50ml of a 0.1m metal salt(AgNO_3 , $\text{Fe}(\text{NO}_2)_3 \cdot 9\text{H}_2\text{O}$, $\text{Cu}(\text{NO}_2)_3$, $\text{Ni}(\text{NO}_2)_3 \cdot 6\text{H}_2\text{O}$ or $\text{Pb}(\text{NO}_2)_3$ mixed with 50 ml of plant extract. and synthesis at 25deg C. in the determination of DPPH radical scavenging capacity method analysis the highest antioxidant capacity was only from plant extract, seaweed extract only prevented the smallest amount of antioxidant like <1TE. The phenolic content checking is the same as DPPH. only leaf extract showed the highest value of above4GAE and algae extract is lowest phenolic concentration <2GAE antioxidant capacity of synthesized silver nanoparticle showed SPR band at 423nm which is constant with time. TEM image showed that the metal NPs were heterogeneous in shape with an average size range of

50nm. however, with the DLS method main size distribution of NPs were is in above 100nm but AgNPs were under 100nm themselves. (Maria Martinez-cabanas et al., 2021)

2. REVIEW OF LITERATURE

NANOBIOTECHNOLOGY

The term '[Nanobiotechnology](#)' refers to the application of biological entities like plants, animals, and microorganisms for the synthesis of nanoparticles (NPs) which have at least one dimension in the range of 1-100 nm. These methods are commonly known as 'Green synthesis', which are biologically safe, cost-effective, and environment friendly. There is a vast range of plants and plant-based materials reported for the synthesis of metal nanoparticles (MNPs) such as zinc, copper oxide, MnO, silver and gold NPs and their respective metal oxide NPs. The plant-based synthesis of NPs is known as 'Phytosynthesis'. In a similar way, the microorganisms are also used for the synthesis of MNPs like copper and copper oxide, zinc and zinc oxide, aluminum oxide, silver, gold, and many more. As we go through the literature in the recent past, it shows an enormous variety of microorganisms that are used for the synthesis of these NPs. The microorganisms range from bacteria, myxobacteria, actinomycetes, fungi, and yeasts, or sometimes the combination of any of them. Among this biological synthesis of silver and gold, NPs have gained great importance and have started a new era of biogenic NPs from the past few decades. The major group of microorganisms used for the synthesis of silver NPs includes bacteria and fungi.

DEVELOPMENT OF NANO WORLD

The concept of nanotechnology was introduced by an American physicist and Nobel prize laureate Richard Feynman in 1959, and also presented the lecture called "there's plenty of room at the bottom". And he is considered as the father of modern nanotechnology. After that a Japanese scientist norio taniguchi was first the use the term called nanotechnology after 15 years in 1974. Some recent studies on nanotechnology in ocean department; Multidrug-resistant (MDR) pathogenic bacteria have become dangerous in bringing intermittent outbreaks in public health and continually spreading nosocomial infection from the addition of antibacterial. By that green synthesis of gold nanoparticles using aqueous extracts of cell-free nitrate fixing bacteria, the blue-green algae called *Anabaena spiroides*. The sample was collected from the brackish water, Odisha.

These synthesized AuNPs were tested to be used as antibacterial agents against the MDR bacteria. The synthesized AuNPs were characterized by UV-V spectrophotometry, SEM-XRD, and ART-FTIR

analysis. The presence of AuNPs is confirmed at the absorption peak at 538nm by UV-v spectrophotometry and FTIR analysis. Some *Anabaena spiroides* AuNPs were monitored for antibacterial activity against an MDR pathogenic bacterium. The bacterial strains were collected from the clinical samples namely *Klebsiella oxytoca* and *Streptococcus pyogenes* and in vitro, the individual antibiograms of those bacteria were known the record MIC values are 20, 25 and 30mg. (Chinmayee Priyadarshini mandhata et al., 2021)

Green synthesis of gold nanoparticles using aqueous extracts of *Anabaena spiroides*. Synthesized AuNPs were tested to be used as antibacterial agents against the MDR bacteria (*Klebsiella oxytoca*, MRSA and *Streptococcus pyogenes*). The synthesized AuNPs were characterized by UV-V spectrophotometry, SEM-XRD, and ART-FTIR analysis. The presence of AuNPs is confirmed at the absorption peak at 538nm by UV-v spectrophotometry and FTIR analysis. Some *Anabaena spiroides* AuNPs were monitored for antibacterial activity against an MDR pathogenic bacterium. The bacterial strains were collected from the clinical samples namely *Klebsiella oxytoca* MRSA and *Streptococcus pyogenes* and in vitro, the individual antibiograms of those bacteria were known the record MIC values are 20, 25 and 30mg (Mandhata et al., 2021). Selenium nanoparticles were also known as a bioactive compound. In this study, the green synthesis of silver nanoparticles was done by using sodium selenite on *Spirulina plantensis* for the synthesis of silver nanoparticles in various cultivation conditions including PH and illumination schedule variation. The characterization of silver nanoparticles was done by FT-IR and XRD. Size and zeta and potential measurements and antioxidant activities of silver nanoparticles were evaluated by DPPH and FRAP arrays. Selenium nanoparticles were in the size range of 145 ± 6 and 171 ± 13 nm. The antioxidant activity of selected selenium nanoparticles was higher than sodium selenite. It shows that green synthesis is a safe and eco-friendly cellular consistent method to scale up this selenium nanoparticles production. (Shohreh Alipour et al., 2021)

In this study, the biosynthesis of gold nanoparticles by marine microbes and their application in the biomedical field were tested. Material chemical like HAuCl₃, DPPH, FeSO₄, 2,2'-bipyridyl, tris-HCL buffer with pH 7.4 was brought from sigma Aldrich. Media brought from Hi-media PVT ltd. Nutrient agar was used on isolated bacteria for better growth. For cell culture, the human colon cancer cell line (HAC-7) was cultured with DME antibiotic-antimycotic solution under 5% CO₂ at 37 deg C. AgNPs was synthesised extracellularly. For the characterization of AuNPs done by UV-visible analysis, surface Plasmon resonance was observed at a wavelength of 530nm which indicated that the bacteria in synthesizing AuNPs after incubation of 24hrs. For the identification of morphological system of AuNPs was done TEM. Analysis showed that the comparison between synthesized AuNPs was irregular in

shapes with a size range of 10-100nm. FTIR also measured some functional groups that helping in reducing and stabilizing agents for formulated gold NPs in peaks at 1644cm^{-1} for alkenes, 1399cm^{-1} for CH_3 bend, 3542cm^{-1} for alcohol. Antioxidant activity of gold nanoparticles was tested by DPPH assay with a reagent that produces a violet solution that with ethanol solvent because of transfer of free electron. This method of using microbes mediated synthesis of AuNPs is considered a non-toxic method and easy for scale-up and economically stable. Finally, TEM and SEM showed that synthesized AuNPs were irregular in shapes with sizes ranging from 100-150nm. These AuNPs were also biocompatible in nature and also depicted their non-cytotoxic properties compared to other nanoparticles. (Rajeshkumar shunmugan et al., 2020)

padina boryana mediated green synthesis of crystalline palladium nanoparticles as potential nanodrug against multi drug resistant bacteria and cancer cells. In this study the peculiar nano-specific features of palladium nanoparticles that offer invaluable possibilities for clinical treatment is tested. The use of algal mediated could be the propertive substitute for the development of multi drug resistance in pathogenic bacteria and the prevalence of cancer. Here the green synthesized pd-nps from *padina boryana* and its anti microbial,antibioflim and anticancer activities were tested. Aqueous extract of *padina boryana* were prepared, dried and powdered. The facile one pot fabrication method using green PB-extract were used to prepare pd-NPs.and prepared in sterile DIW procured from Sigma Aldrich,USA. Physicochemical characterization of PB extract-capped pd-nps were done by UV-visible spectroscopy,EDX and SEM analysis. By using double beam operation of PerkinElmer lambda 35 spectrophotometer were checked for the absorption fo UV visible range of Pb-extract , Na_2PdCl_4 , PB extract-capped Pd-NPs. Structure shape and size were determined by tramission electron microscopes used for the determination of average diameter and shape of pd-nps at 200 kV. Measurement of surface area of pb capped – pd-nps were done by Brunauer Emmett teller (BET) usinf autosorb io mp/XR surface analysis. To determine the surface functional group and compound, FTIR analysis of pb extract and pd-nps was recorded in attenuated total reflectance (ATR) mode on the Perkin elmer system 2000 instrument for the characterization of cilinical bacterial pathogen the samples were collected from sluid and suputum of immunocompromised patient diagnosed with inspiratory infections. Antibiotic resistant isolated culture were molecularly caracterized by partially seqencing16sRNA gene using universal primers 785F and 907R and sequence. Finally the synthesised pd-nps were small in size, spherical and crystalline in nature which are biologically effective in the range of $31.25\text{--}125\text{ }\mu\text{g/ml}$ against multi drug resistance in pathogenic bacteria *S. aureus* showed a Gram-positive reaction whereas, *E. fergusonii*, *A. pittii*, *.P. aeruginosa*, *A. enteropelogenes*, and *P. mirabilis* were found Gram-negative. So this study suggest the possible role of PB- capped pd-extract capped pd-nps for successful clinical management od MDR pathogens and breast cancer cells.(Hana sonbol et al., 2021) This is the study to report the first to use brown microalgae

cystoseira bacteria to synthesis gold nanoparticles by eco friendly. In fact a one-pot synthetic route method. A sample of cystoseira bacteria was collected from the lower intestinal rocky shore in the new cot in Spain. for Gold Wave synthesis HAuCl₄ was from Merck for the preparation of secured extract sample were cut into pieces and then boiled for 15 min with the preparation of 1g/ml of mili-Q-water extract is centrifuged by Beckman water microfuge 16 at 4500rpm for 10 mins and the obtained extract was stored at 4°C for later use. There(CB) extract was characterized by a standard curve was built with ascorbic acid with concentrations of 50 and 400 mg/l. By the method, CB extract showed a high reducing activity value of 583.1 ± 17.1 mg ascorbic acid per gram. For the synthesis of gold nanoparticles. In 75ml of aqueous solution of HAuCl₄ (0.01m) was added to the CB extract by 1ml. Gold synthesis was confirmed by a colour change in Ph changing from 5.4 in the CB extract to 4.5 in AuCB solution. Optimum concentration was chosen by trial and error method and the 10nm in Loke 0.4mm after the analysis UV-spectra should SPR absorption band at 532nm which confirm the synthesis of Gold nanoparticle. Z potential measurement showed equal to the -30.1 ± 20 mv was obtained from AuCB which shows the Gold nanoparticle stable colloidal suspension with negatively charged particle. To better characterise Tem images were obtained which shows that most of Nps were embedded in the CB matrix, which is due to high angle annular dark field images. The average particle single of GNPs was 8.4 ± 2.2 nm and FIRC studio showed that the NP, were poly crystalline in water. Also FTIR showed that a high value of CB extract could have a fast Nucleation process, spectrum showed 3402 cm⁻¹ very broad peak which corresponds to nit stretching vibration of free OH group or polyphenol and polysaccharides. The cytotoxic activity of CBAU and the CB extract was tested by using two human colour cancer cells called caco2 and HT-29 and also on healthy cell line (Pcs-201-010 CRS) Effect of Non-significance antiproliferative of CB extract was detected at the dose of any for CaCo-2 F (4,15)=1.506, p=0.25047 and pcs-201-010 [F(4,15)=0.2079, (p=0.9345) And with the of HT-29 the height dose is only 1g/ml had the effect of p≤0.0001 significant value effect of caco2-2 cells by doses are 400,200,100 and 50µm. In HT-29 cells the significant value doses are $11.75 \pm 3.94\%$ 12.87 ± 0.36 , 18.98 ± 3.57 , $25.8 \pm 7.29\%$ and $21.74 \pm 5.3\%$ IC₅₀ value of CaCo2 were 79.03 µm and for HT-29 in 49.61 µm. when the treatment of AuCB at IC₅₀ does 48-fold included an early apoptotic cell in CaCo2 in ($15.90 \pm 7.7\%$) when the created control is ($0.33 \pm 0.57\%$) But only 3 fold at HT-29 with (8.03 ± 1.31) in $3.09 \pm 1.82\%$. And also synthesized AuCB exhibited high biocompatibility on the healthy cell lines. This study concluded that induced apoptotic activity of AuCB was assessed by the extrinsic and mitochondrial pathway and also suggest significant potential for the treatment of colorectal cancer. (W.Gonzalez Ballesteros et al., 2017)

In this study, Anticancer activity on A549 lung cancer of Biosynthesized AgNPs and their morphological damage was tested. Biosynthesized AgNPs from green marine algae called Caulerpa taxifolia which can produce enormous bioactive compounds and more bioactivities needed chemicals

like sodium nitrate(AgNO_3) sodium hydroxide (NaOH) D_2O was purchased from suresh scientific@co, Tamilnadu India cancer cells were obtained from king institute of prevention and medicine, Guindy, Chennai Tamilnadu DMes, Ao/EBR fluorescence dye was purchased from mirk, India, the cultured cells were kept maintained at 37degree C with 50pco2 Incubator at Tissue culture plate for seaweed extract preparation seaweed was left dried for 30 days and ground using a mortar and pestle ground powder was dissolved in 1000ml of millipos water and boiled at 80Degree C for about 20 minutes and filtered by using whatman no-1 filter paper. By using HPLC the purified extract was recovered. After that HPLC extract was analyzed the HPLC-ms to get complete phytochemical and anticancer derivatively. In the third fraction of HPLC extract, some already reported anticancer compound was identified (2,4-Di test-butyl-phenol) Tetracosenoic acid, 9-H-pyrido [3,4-6] indole, Bis (2-Ethylhexyl) phthalate, neo-phytadiene. This study suggests that the anti-cancer compound can act as a reducing agent in the synthesis, of Agnp UV-visible spectral analysis indicates that Agnlps was formed from the reaction mixture by noticing a colour change from yellow to reddish-brown in purified seaweed extract and AgNO_3 solution who Uv absorption peak of AgNPs in 420nm (310-490nm) SPR. powder XRD analysis results indicate that Agnlp was crystalline in nature and it been derived from seaweed extract SEM analysis with Morphological image that the AgNPs was in uniform-sized particles, with dispersed spherical in shape. The confirmation of AgNPs image By TEM showed that seaweed synthesized Agnlp were small in size which can able to damage the cancer cells through ROS generation or mitochondrial damage or cell cycle has Also shown that Agnlp was 10 and 100nm along with polydispersed spherical in shape with small size but with large surface area. For anti-cancer experiment before by using a Dimethyl thoizoldisphenyltetragolium bromide(MTT) a cytotoxicity assay to detect the anti-cancer effect of synthesized Agnlps And also exhibited excellent nanoparticle for A549 lung cancer cells with effectiveness at 70kgm/co concentration. At same concentration 900 of inhibition was shown with gradually dear of growth and in increased concentration who show high turbidity at 96 cell plate with the addition MTT assay in A549 plate formazan was rapidly formed and continued with a color change which used to identify the control cells with no turbidity or color formation with IC59 value of 40mg/ml which exhibited the inhibition value of ~52% against A549 cells Hence this study result that biosynthesis Agnp has more anti-cancer against A549 lung cancer cells.(Danjie zhang et al., 2020)

3. AIM:

To synthesis gold nanoparticles from marine seaweed

OBJECTIVES:

Collection of Marine seaweeds from Rameshwaram district Tamil Nadu

Synthesis gold nanoparticles by using different Seaweeds methods.

Characterization of Gold nanoparciels using Various instruments

Material methods

Gold Chloride preparation

Gold chloride has been purchased from sigma Aldrich.

303mg of AuCl_3 power form is mixed up with 1000ml of distilled water, and kept wrapped with aluminum foil in a dark place for further use.



Three number of seaweed were purchased from local algae collection centre in Mandapam coastal region, tamilnadu. 1kg of each seaweed was brought up. Each sample was washed with clean tap water once again and shade dried. After that completely dried seaweed was ground using electric mixture to fine powder. And stored in an air tight container.

SEAWEED SAMPLE

Seaweed name: *Turbinaria conoides*

Class: *phaeophyceae*

Order: *fucales*

Family: *sargassaceae*

Genus: *Turbinaria*



Figures :2 Collection of Seaweed and Preparation of Seaweed Extract

PREPARATION OF SEAWEED EXTRACT

1g of seaweed powder is mixed in 100ml of distilled water and kept in water bath for about 30mins to get the extract. After 30mins the cooled extract was filtered using NO.1 whatman filter paper. Filtered extract is used for the synthesis of gold chloride

SYNTHESIS OF GOLD NANOPARTICLES

For the synthesis of gold nanoparticles, in 5 test tubes AuCl_3 solution and seaweed extract were added in the concentration of 9ml-1ml, 8ml-2ml, 7ml-3ml, 6ml-4ml, 5ml-5ml in order to know which concentration has high efficiency in synthesizing gold particles. Mostly 8ml-2ml, 7ml-3ml has given the best result. The formation of gold nanoparticles were confirmed by the colour change of the solution from pale brown to dark purple and dark pink. So the sample were prepared in bulk volume to take further characterizations.



FIGURE 4: 1) after mixing sample and gold chloride solution. 2) formation of gold nanoparticles after 1hr after mixing.

ANTIMICROBIAL ACTIVITY

Antimicrobial activity was checked by using agar diffusion method. Preparation of agar medium: nutrient agar medium was prepared by mixing with 100ml of distilled water and mixed sterilized in autoclave. The sterilized media was poured into instant sterilized Petri dishes and allowed it to solidify at room temperature in laminar flow. Preparation of pathogen inoculums: each 2 pathogen were freshly made 12hrs inoculums from preserved plates. Each sample of pathogens were swabbed into to prepared plates by using cotton swab and sample were loaded into each 5mm wells respectively. After incubation period inhibition zone indicates the antimicrobial activity.

CHARACTERIZATION OF SILVER NANOPARTICLES

UV(UV-VISIBLE SPECTROSCOPY)

The characterization of gold nanoparticle was observed by the colour changes. Gold chloride was diluted 9ml of AuCl_3 and 1ml of sample 8ml of AuCl_3 and 2ml of sample 7ml of AuCl_3 and 3 ml of sample 6ml of AuCl_3 and 4 ml of sample 5ml of AuCl_3 and 5ml of sample kept in incubation for 72 hours and the colour change were observed and scanned at wavelength of 520nm to 550nm in uv spectroscopy to determine absorption of the sample

FTIR (Fourier Transform Infrared Spectroscopy)

To identify the potential biomolecules responsible for the conversion of AuCl₃ to AuCl₂, FT-IR spectra were collected for turbinaria. The FTIR spectrum reported from the freeze-dried powder of gold nanoparticle, formed after 72hr of incubation with seaweed aqueous extract. The characteristic bands are related to the chemical and physical changes of the OH stretching vibration.

SEM(SCANNING ELECTRON MICROSCOPE)

The morphology and shape of these gold nanoparticles were carried out using SEM After 2 hours of exposure to 10⁻³ M aqueous gold chloride solution, nanoparticles biosynthesized from *turbinaria.c* biomass displayed the colloidal form of the particles in solution, which microprecipitated. on the surface of the experimental seaweed's biomass During the. The SEM investigation of gold nanoparticles in this study, in addition to being interesting, was also useful. It was also microprecipitated when it was present in colloidal form in solution. on the biomass's surface, which were clearly showing that The reduction of Au⁺ ions produces nanoparticles that are bonded together. To the cell's outer surface Biosynthesis of silver nanoparticles produced from *turbinaria.c* were dominated by cubical and spherical formations with homogeneous shape. The more luminous cubical and spherical region of the electrons scattering in the back.

TEM (TRANSMISSION ELECTRON MICROSCOPE)

TEM is a technique for determining the size and particle dispersion of gold nanoparticles. The magnitude and distribution of the group Observations of biosynthesized gold nanoparticles were made using the micrograph of silver complex solution drop-coated sheets with *turbinaria.c* an experimental seaweed extract The TEM is a term that refers to a. The production of spherical shaped particles was visible in images. Gold nanoparticles are found in aggregates with diameters ranging from 5 to 10 nanometers as depicted in The size of gold nanoparticles on average *turbinaria* has a diameter of 32nm used the extract as a reducing and antioxidant agent of capping The form of metal nanoparticles has changed dramatically.

XRD (X-RAY DIFFRACTION)

The X-ray diffraction (XRD) method was used to confirm the creation of a single-phase compound. The XRD pattern of synthetic AuNPs was examined and compared to that of natural AuNPs.

Results

Antimicrobial activity:

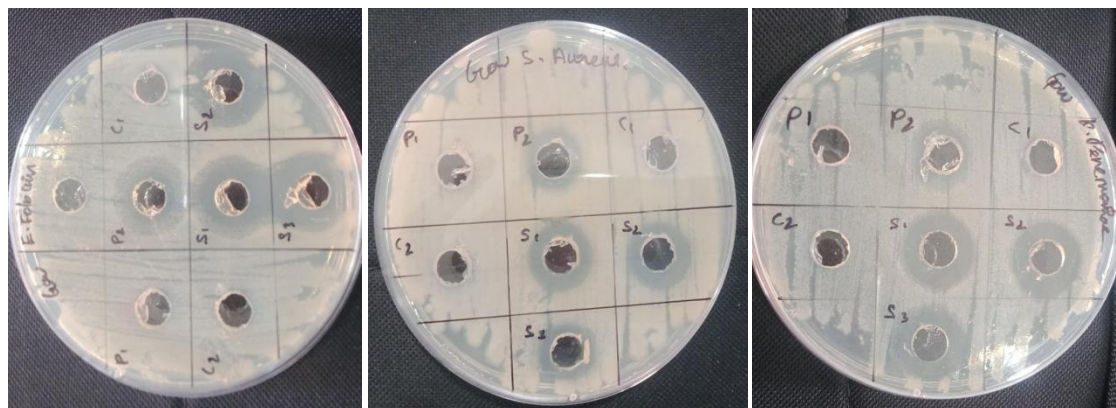
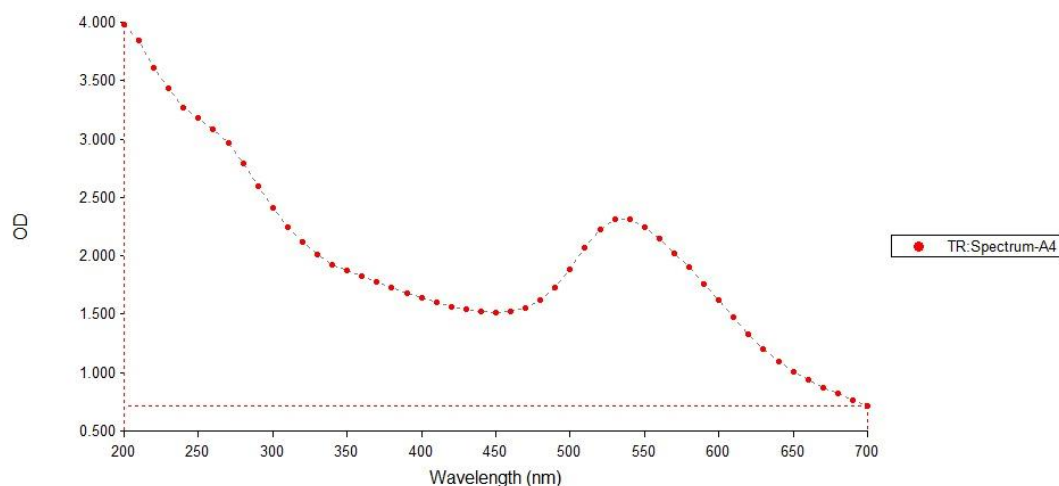


FIGURE 5: inhibition zone formed at different concentration of seaweed synthesized gold nanoparticles.

CHARCTERIZATION OF GOLD NANOPARTICLES:

UV-Visible spectroscopy: The localized surface Plasmon resonance wavelength of gold nanopartciles was observed at 520nm which confirms the formation of gold nanoparticles.



Fourier Transform Infrared Spectroscopy

Fourier transform infrared spectroscopy is used to identify the potential biomolecules responsible for the conversion of AuCl_3 into AuCl_2 . We extract and manufacture gold nanoparticles. The FTIR spectrum reported from the freeze-dried powder of seaweed synthesized gold nanoparticles, formed after 72 hr of incubation with seaweed aqueous extract. The characteristic bands are related to the chemical and physical changes of the OH stretching vibration.

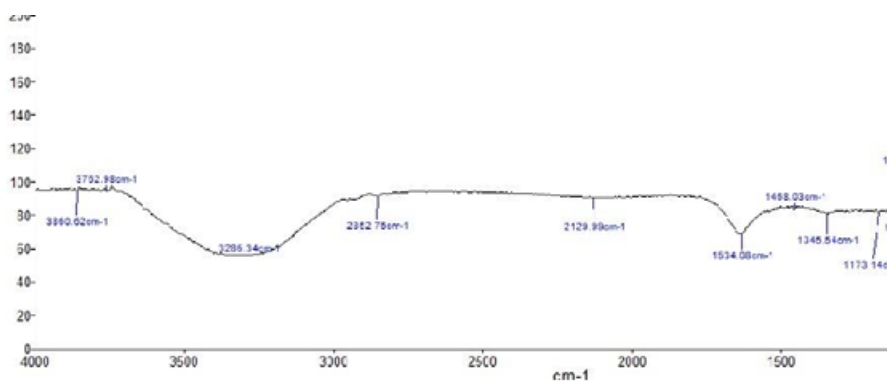


Figure: 7 FTIR result of synthesized gold nanoparticles.

SCANNING ELECTRON MICROSCOPE

The shape and morphological study of these synthesized gold nanoparticles were carried out using SEM. After 2 hours of exposure to 10^{-3} M aqueous gold chloride solution, nanoparticles biosynthesized from *thurbinaria conoides* biomass displayed the colloidal form of the particles in solution, which microprecipitated on the surface of the experimental seaweed's biomass. During the SEM investigation of gold nanoparticles in this study, in addition to being interesting, was also useful. It was also micro precipitated when it was present in colloidal form in solution on the biomass's surface, which were clearly showing that the reduction of Au^+ ions produces nanoparticles that are bonded together. To the cell's outer surface. Biosynthesis of gold nanoparticles produced from *Sargassum.w* were dominated by cubical and spherical formations with homogeneous shape. The more luminous cubical and spherical region of the electrons scattering in the back. The particle size of the gold nanoparticles is between 50-90nm.



Figure: 8 SEM result of gold nanoparticles.

TRANSMISSION ELECTRON MICROSCOPE

TEM is a technique for determining the size and particle dispersion of gold nanoparticles. The magnitude and distribution of the group. Observations of biosynthesized gold nanoparticles were made using the micrograph of silver complex solution drop-coated sheets with *turbinaria.c* an experimental seaweed extract. The TEM is a term that refers to a. The production of spherical shaped particles was visible in images. Gold nanoparticles are found in aggregates with diameters ranging from 5 to 10 nanometers as depicted in. The size of gold nanoparticles on average *turbinaria* has a diameter of 32nm used the extract as a reducing and antioxidant agent of capping. The form of metal nanoparticles has changed dramatically.

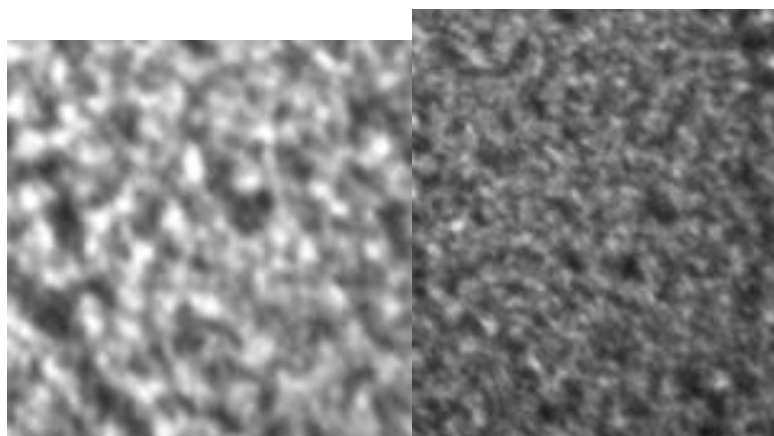


Figure: 9 .TEM result of gold nanoparticles.

X-RAY DIFFRACTION

The X-ray diffraction (XRD) method was used to confirm the creation of a single-phase compound. The XRD pattern of synthetic AuNPs was examined and compared to that of natural AuNPs.

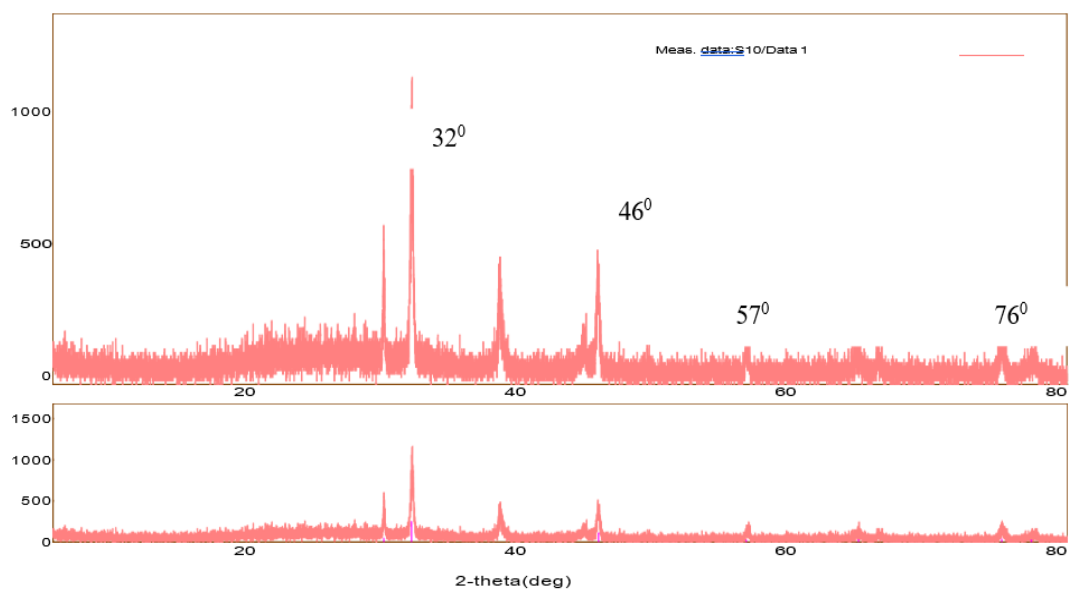


Figure: 10: XRD result of gold nanoparticles.

Discussion

Antibacterial activity: In each 3 plates s1 concentration has reached the higher inhibition zone of 32mm on *E. faecalis* pathogen which cause urinary tract infection, wound infection and intra-abdominal infection. By this result it clearly indicates that this biosynthesized gold nanoparticles from the seaweed *turbinaria* is having efficiency on wound healing property.

REFERENCES

1. Hamouda, R.A., Hussein, M.H., Abo-elmagd, R.A. et al. Synthesis and biological characterization of silver nanoparticles derived from the cyanobacterium *Oscillatoria limnetica*. *Sci Rep* 9, 13071 (2019).
2. Yadvinder Singh ., Sandeep Kaushal ORCID and Ramandeep Singh Sodhi et al. Biogenic synthesis of silver nanoparticles using cyanobacterium *Leptolyngbya* sp. WUC 59 cell-free extract and their effects on bacterial growth and seed germination. DOI: 10.1039/D0NA00357C *Nanoscale Adv.*, 2020, 2, 3972-3982
3. Gaurav Sharma, Nakuleshwar Dut Jasuja, Manoj Kumar, Mohammad Irfan Ali, "Biological Synthesis of Silver Nanoparticles by Cell-Free Extract of *Spirulina platensis*", *Journal of Nanotechnology*, vol. 2015, Article ID 132675, 6 pages, 2015.
4. Ramya, V. P., & Muralitharan, G. (2019). Biosynthesis of silver nanoparticles using *Calothrix membranacea* KLR006 and characterization of its antimicrobial properties. *Research Journal of Life Sciences, Bioinformatics, Pharmaceutical and Chemical Sciences*, 5(2), 237.
5. Mishra, Bharti, Abhishek Saxena, and Archana Tiwari. "Biosynthesis of silver nanoparticles from marine diatoms *Chaetoceros* sp., *Skeletonema* sp., *Thalassiosira* sp., and their antibacterial study." *Biotechnology Reports* 28 (2020): e00571.
6. Jayashree Jena, Nilotpala Pradhan, Bishnu Prasad Dash, Prasanna Kumar Panda, Barada Kanta Mishra, Pigment mediated biogenic synthesis of silver nanoparticles using diatom *Amphora* sp. and its antimicrobial activity, *Journal of Saudi Chemical Society*, Volume 19, Issue 6, 2015, Pages 661-666, ISSN 1319-6103.
7. Darwesh, O.M.; Matter, I.A.; Eida, M.F.; Moawad, H.; Oh, Y.-K. Influence of Nitrogen Source and Growth Phase on Extracellular Biosynthesis of Silver Nanoparticles Using Cultural Filtrates of *Scenedesmus obliquus*. *Appl. Sci.* 2019, 9, 1465.

8. Ramanathan, Tevan & Jayakumar, Saravanan & Sahimi, Nor & Iqbal, Nur & Zapri, Iffah & Govindan, Natanamurugaraj & Ab. Rahim, Mohd Hasbi & Ichwan, Solachuddin & Maniam, Gaanty. (2017). BIOSYNTHESIS OF SILVER NANOPARTICLES USING MARINE MICROALGAE ISOCHRYDIS sp. Journal of Chemical Engineering and Industrial Biotechnology. 2. 1-12. 10.15282/JCEIB-V1-02.29/9/2017/2.2.
9. Mandhata, C. P., Sahoo, C. R., Mahanta, C. S., & Padhy, R. N. (2021). Isolation, biosynthesis and antimicrobial activity of gold nanoparticles produced with extracts of *Anabaena spiroides*. Bioprocess and Biosystems Engineering, 1-10.
10. Tevan, R., Jayakumar, S., Sahimi, N. H. A., Iqbal, N. F. A., Zapri, I., Govindan, N., ... & Maniam, G. P. (2017). BIOSYNTHESIS OF SILVER NANOPARTICLES USING MARINE MICROALGAE ISOCHRYDIS sp. Journal of Chemical Engineering and Industrial Biotechnology, 2(1), 1-12.
11. San Keskin, N.O., Koçberber Kılıç, N., Dönmez, G., Tekinay, T., 2016. Green Synthesis of Silver Nanoparticles Using Cyanobacteria and Evaluation of their Photocatalytic and Antimicrobial Activity. JNanoR 40, 120–127.
12. El-Belely, E. F., Farag, M., Said, H. A., Amin, A. S., Azab, E., Gobouri, A. A., & Fouda, A. (2021). Green synthesis of zinc oxide nanoparticles (ZnO-NPs) using *Arthrospira platensis* (Class: Cyanophyceae) and evaluation of their biomedical activities. Nanomaterials, 11(1), 95.
13. Bin-Meferij MM, Hamida RS. Biofabrication And Antitumor Activity Of Silver Nanoparticles Utilizing Novel Nostoc sp. Bahar M. Int J Nanomedicine. 2019 Nov 22;14:9019-9029. doi: 10.2147/IJN.S230457. PMID: 31819416; PMCID: PMC6881505.
14. Bin-Meferij, M. M., & Hamida, R. S. (2019). Biofabrication and antitumor activity of silver nanoparticles utilizing novel nostoc sp. Bahar M. International journal of nanomedicine, 14, 9019.
15. Shohreh Alipour, Sara Kalari, Mohammad Hossein Morowvat, Zahra Sabahi, Ali Dehshahri, "Green Synthesis of Selenium Nanoparticles by Cyanobacterium *Spirulina platensis* (abdf2224): Cultivation Condition Quality Controls", BioMed Research International, vol. 2021, Article ID 6635297, 11 pages, 2021.
16. Sreemanti Banerjee, Abhishek Bhattacharya, Piya Roychoudhury, Anjan K. Dasgupta, Moumita Dutta & Ruma Pal (2021) *Arthrospira platensis* (Cyanobacteria) – a potential biofactory for fluoromagnetic nanoiron production, Phycologia, 60:1, 62-72.
17. Pytlik, Nathalie, et al. "In vivo uptake of gold nanoparticles by the diatom *Stephanopyxis turris*." Algal research 39 (2019): 101447.

18. Lauritano, C., Martín, J., de la Cruz, M., Reyes, F., Romano, G., & Ianora, A. (2018). First identification of marine diatoms with anti-tuberculosis activity. *Scientific reports*, 8(1), 1-10.
19. Younis, N.S., El Semaary, N.A. and Mohamed, M.E., 2021. Silver nanoparticles green synthesis via cyanobacterium *Phormidium* sp.: characterization, wound healing, antioxidant, antibacterial, and anti-inflammatory activities. *European review for medical and pharmacological sciences*, 25(7), pp.3083-3096.
20. Elkomy RG. Antimicrobial screening of silver nanoparticles synthesized by marine cyanobacterium *Phormidium formosum*. *Iran J Microbiol.* 2020 Jun;12(3):242-248. PMID: 32685121; PMCID: PMC7340607.
21. Mandhata, C. P., Sahoo, C. R., Mahanta, C. S., & Padhy, R. N. (2021). Isolation, biosynthesis and antimicrobial activity of gold nanoparticles produced with extracts of *Anabaena spiroides*. *Bioprocess and Biosystems Engineering*, 1-10.
22. Sonbol, H., Ameen, F., AlYahya, S. et al. *Padina boryana* mediated green synthesis of crystalline palladium nanoparticles as potential nanodrug against multidrug resistant bacteria and cancer cells. *Sci Rep* 11, 5444 (2021).
23. Rajeshkumar Shunmugam, Sri Renukadevi Balusamy, Venkat Kumar, Soumya Menon, T. Lakshmi, Haribalan Perumalsamy, Biosynthesis of gold nanoparticles using marine microbe (*Vibrio alginolyticus*) and its anticancer and antioxidant analysis, *Journal of King Saud University - Science*, Volume 33, Issue 1, 2021, 101260, ISSN 1018-3647.
24. Martínez-Cabanas M, López-García M, Rodríguez-Barro P, et al. Antioxidant Capacity Assessment of Plant Extracts for Green Synthesis of Nanoparticles. *Nanomaterials (Basel)*. 2021;11(7):1679. Published 2021 Jun 25.
25. Araya-Castro, K.; Chao, T.-C.; Durán-Vinet, B.; Cisternas, C.; Ciudad, G.; Rubilar, O. Green Synthesis of Copper Oxide Nanoparticles Using Protein Fractions from an Aqueous Extract of Brown Algae *Macrocystis pyrifera*. *Processes* 2021, 9, 78.
26. Sonbol, H., Ameen, F., AlYahya, S. et al. *Padina boryana* mediated green synthesis of crystalline palladium nanoparticles as potential nanodrug against multidrug resistant bacteria and cancer cells. *Sci Rep* 11, 5444 (2021).
27. Aletayeb P, Ghadam P, Mohammadi P. Green synthesis of AgCl/Ag₃PO₄ nanoparticle using cyanobacteria and assessment of its antibacterial, colorimetric detection of heavy metals and antioxidant properties. *IET Nanobiotechnol.* 2020 Oct;14(8):707-713.

28. Rajeshkumar Shunmugam, Sri Renukadevi Balusamy, Venkat Kumar, Soumya Menon, T. Lakshmi, Haribalan Perumalsamy, Biosynthesis of gold nanoparticles using marine microbe (*Vibrio alginolyticus*) and its anticancer and antioxidant analysis, Journal of King Saud University - Science, Volume 33, Issue 1, 2021, 101260, ISSN 1018-3647.
29. Martínez-Cabanas, María & López-García, Marta & Rodríguez-Barro, Pilar & Vilariño, Teresa & Lodeiro, Pablo & Herrero, Roberto & Barriada, José & sastre de vicente, Manuel. (2021). Antioxidant Capacity Assessment of Plant Extracts for Green Synthesis of Nanoparticles. Nanomaterials. 11. 1679. 10.3390/nano11071679.
30. Selvi, B & Madhavan, J & Santhanam, Amutha. (2016). Cytotoxic effect of silver nanoparticles synthesized from *Padina tetrastomatica* on breast cancer cell line. Advances in Natural Sciences: Nanoscience and Nanotechnology. 7. 035015.
31. Rajagopal, Gopalan & Nanthagopal, Manivannan & Mahalingam, Sundararajan & Kumar, A. & Samuthirarajan, Senthilkumar & N, Mathivanan & Sakkanan, Ilango. (2021). Biocompatibility assessment of silver chloride nanoparticles derived from *Padina gymnospora* and its therapeutic potential. Nano Express.
32. González-Ballesteros N, Prado-López S, Rodríguez-González JB, Lastra M, Rodríguez-Argüelles MC. Green synthesis of gold nanoparticles using brown algae *Cystoseira baccata*: Its activity in colon cancer cells. Colloids Surf B Biointerfaces. 2017 May 1;153:190-198.
33. Zhang, Danjie & Govindan, Ramachandran & Mothana, Ramzi & Siddiqui, Nasir & Ullah, Riaz & Almarfadi, Omer & Manoharan, Natesan. (2020). Biosynthesized silver nanoparticles using *Caulerpa taxifolia* against A549 lung cancer cell line through cytotoxicity effect/morphological damage. Saudi Journal of Biological Sciences.
34. Bhimba, Valentin. (2012). Anticancer Activity of Silver Nanoparticles Synthesized by the Seaweed *Ulva lactuca* Invitro. Open Access Scientific Reports. 1. 1-4. 10.4172/scientificreports.242.
35. Venkatesan J, Kim SK, Shim MS. Antimicrobial, Antioxidant, and Anticancer Activities of Biosynthesized Silver Nanoparticles Using Marine Algae *Ecklonia cava*. Nanomaterials (Basel). 2016 Dec 6;6(12):235.
36. Zada S, Ahmad A, Khan S, Yu X, Chang K, Iqbal A, Ahmad A, Ullah S, Raza M, Khan A, Ahmad S, Fu P. Biogenic synthesis of silver nanoparticles using extracts of *Leptolyngbya* JSC-1 that induce apoptosis in HeLa cell line and exterminate pathogenic bacteria. Artif Cells Nanomed Biotechnol. 2018;46(sup3):S471-S480.

37. Acharya, Diptikanta & Satapathy, Sagarika & Yadav, Kanti & Somu, Prathap & Mishra, Gitanjali. (2021). Systemic Evaluation of Mechanism of Cytotoxicity in Human Colon Cancer HCT-116 Cells of Silver Nanoparticles Synthesized Using Marine Algae *Ulva Lactuca* Extract.