Project Report On-

"CARBOHYDRATE POLYMER BASED SILVER NANOPARTICLE FOR BIOMEDICAL APPLICATION"

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B.Sc 6th Semester, 2023 (3 Year)

Department of Chemistry

JIS University

2023

Certificate

This is to certify that **Ms. Anjali Kumari** has completed her project on the topic entitled "Carbohydrate polymer based Silver Nanoparticle for Biomedical Application" for the fulfillment of her BSc. degree. This is the record of the candidate's own effort and work under my guidance and supervision. I wish her all success in life.

Date:

Place: Agarpara

Acknowledgement

I would like to express my special thanks to my teacher **Dr. Piyali Jana** as well as our **HOD Partha Pratim Ghosh** who gave me the golden opportunity to do this wonderful project on the topic Carbohydrate polymer based Silver Nanoparticle for Biomedical Application, which also helped me in doing a lot of Research and I came to know about so many new things I am really thankful to them.

I am thankful to **JIS UNIVERSITY** for providing me with the required resources and facilities that helped me to complete my research and also to **Bose Institute** for helping me with DLS

Any attempt at any level can't be satisfactorily completed without the support and guidance of my parents and friends.

I would like to thank my parents who helped me a lot in gathering different information, collecting data and guiding me from time to time in making this project, despite of their busy schedules they gave me different ideas in making this project unique.

Thanking you,

Anjali Kumari

B.Sc 6th Semester, 2023 (3 Year)

Contexts

Abstract

In our present work, by greeen synthesis method we prepare guar gum based silver nano particle. For this method guar gum used as a reducing as well as stabilising agent .We prepare GG Ag NPS` by different pH i.e at acidic, neutral and basic pH by this synthesis procedure. After formation of silver nano particle these are characterised by UV–vis spectroscopy, Fourier transform infrared (FTIR). By DLS instrument the average dynamic size of the Guar gum silver nanoparticle (GG AgNps) was determined as well as zeta potential was measured by the same instrument to see the surface charge.

1. Introduction :

In recent years, metal nanoparticles (NPs) have been investigated extensively to understand their physical and chemical properties, due to their potential applications in emerging areas of nano-science and nano-technology. In the nano size regime, metal NPs have received special attention because of their characteristic optical, electronic and catalytic properties [1-4]. Generally, the preparation of metal NPs involves the reduction of metal ions with a suitable reducing agent, such as hydrazine, dimethyl formamide (DMF), and sodium borohydride ($NaBH_4$). All of these are highly reactive chemicals and will pose potential environmental and biological risks. Following the principles of "green chemistry", the primary challenges in this regard are the maximization of usage of environmentally friendly materials and adoption of sustainable processes in the generation of nano-sized metal particles. It is now well established that polymers are excellent host materials for the preparation of metal NPs and serve also as a surface-capping agent when those NPs are embedded or encapsulated in a polymer. Due to large reserves, biodegradability and eco-friendly "green" processing [5,6] the use of biopolymer such as starch [5] , alginate [7,8] chitosan [9,10] and cellulose [11] in research and industry has significantly increased. The biopolymer scan provide a size-confined micro-environment where the reduction of metal ions into NPs can be achieved by biopolymer itself via adsorption coupled reduction pathways [12,13] or by external assistance via chemically [9,14] photochemically [8] by heating [15] by laser ablation or by high-energy radiation. Some biopolymers (such as chitosan) not only show a better ability to stabilize the resulting metal NPs by anchoring them, but also act as the reducing agent for the surrounding metal ions [20] but this in situ reduction requires heating and controlled pH.

Guar gum (GG) is an edible carbohydrate polymer extracted from the seeds of *Cyamopsist etragonoloba* and is considered as a polysaccharide with one of the highest molecular weights of all naturally occurring water soluble polymers. It is a nonionic, branched-chain polymer, consisting of straight-chain mannose units joined by β-D-(1-4) linkages having a-D-galactopyranose units attached to this linear chain by (1-6) linkages. Galactose and mannose are the repeating units in GG. Compared with native GG, sulfated or phosphorylated GG shows better antioxidant activities. Grafting GG with acrylamide irradiated by microwaves can be used as a better drug delivery system in colon. GG has a strong hydrogen bond forming tendency in water which makes itan excellent thickener and stabilizer. GG has also a strong tendency to form gel in the presence of borax, an efficient crosslinker for polymers bearing hydroxyl groups [19] .These characteristic Senable GG to entrap, protect and stabilize the synthesized metal NPs by acting as an excellent surface capping agent . GG solution is stable over time, not prone to coagulation over a wide range of salinity and pH. Therefore, GG can be used to effectively improve stability and mobility of zero valent iron NPs used for in situ remediation of groundwater [20] .

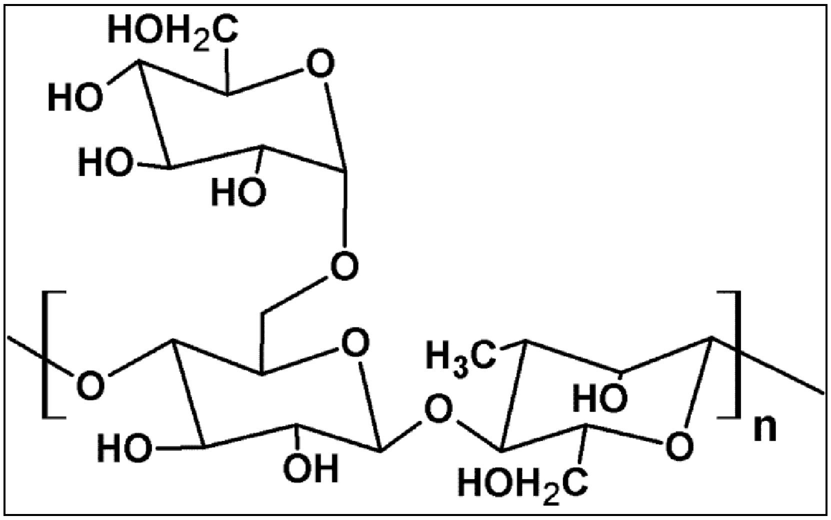


Figure 1: Chemical Structure of Guar gum

Actually, GG has been demonstrated to be a more effective choice for stabilising iron NPs than starch and alginate. Furthermore, GG are possible substitutes that might be extensively used in a number of biotechnology and environmental protection applications due to their intrinsic biocompatibility and biodegradability in the presence of particular enzymes and microorganisms. Based on the foregoing context, we describe in this study a completely green method for the direct synthesis and stabilisation of metallic Ag NPs, using GG as both a reducing agent and a stabiliser for the in situ creation of Ag NPs [19].

By using the reduction technique, silver nanoparticles based on guar gum are further modified. Guar gum (GG) is first purified using conventional methods before being used to create silver nanoparticles from AgNO3 by reducing method with the variation of GG as well as by variation of Silver Nitrate concentration in a specific pH and temperature. The resulting materials are then characterised for biomedical applications using DLS, Zeta Potential, IR, UV, etc.

2. Literature Review-

Lei Dai, Ben Nadeau, Xingye An, Dong Cheng, Zhu Long, Yonghao Ni [33] report of a new paradigm using natural polymer (guar gum, GG) and sodium borohydride ($NaBH_4$), for the preparation of silver nanoparticles (AgNPs)- containing smart hydrogels in a simple, fast and economical way. $NaBH_4$ performs as a reducing agent for AgNPs synthesis using silver nitrate ($AgNO_3$) as the precursor. Meanwhile, sodium metaborate ($NaBO_2$) (from $NaBH_4$) behaves as a cross-linking agent between GG molecular chains. The AgNPs/GG hydrogels with excellent viscoelastic properties can be obtained within 3 min at room temperature without the addition of other cross-linkers. The resultant AgNPs/GG hydrogels are flowable and injectable, and they possess excellent pH/thermal responsive properties. Additionally, they exhibit rapid self-healing capacity. This work introduces a facile and scale-up way to prepare a class of hydrogels that can have great potential to biomedical and other industrial applications.

ES Abdel-Halim, MH El-Rafie, Salem S Al-Deyab ,[32] Guar gum/polyacrylamide graft copolymer was prepared in the presence of potassium bromate/thiourea dioxide as initiation system. The so-prepared and separated guar gum/polyacrylamide graft copolymer was used for preparation of silver nanoparticles through reduction of silver nitrate under certain conditions.Using the composite or the separated graft copolymer in nanosilver preparation, the UV–vis spectra showed high improvement in the absorption intensity at 45 min and the peaks acquire ideal bell shape, which means that the reduction power got higher. Also no decrease in the peak intensity was noticed after 60 min, which means that the graft copolymer has better stabilization efficiency than each individual component alone.

Nida Khan, Deepak Kumar, Pramendra Kumar [34] Guar gum/Gelatin/Silver nanocomposite (GG/Gl/Ag-N-composite) was synthesized thoroughly in greenway via simple in situ method by maltose sugar reduction using the varying concentration of $AgNO_3$ solution. The synthesized composite was characterized by Fourier Transform Infrared (FTIR) spectroscopy, Scanning Electron Microscopy (SEM), Transmission Electron Microscopy (TEM) and X-rays diffraction (XRD) analysis. The synthesized composites were tested for antibacterial activity performed by well diffusion method using tetracycline as reference drug. The GG/Gl/Ag-Ncomposites were ascertained to exhibit outstanding antibacterial property against both Gram-positive Staphylococcus aureus and Gram-negative Escherichia coli and Pseudomonas aeruginosa. It was found that the sample GG/Gl/Ag-N3 exhibited maximum antibacterial activity [zone of inhibition against E.Coli,(14.5 mm) S. aureus (14.5 mm) and P Aeruginosa (15 mm)] among all synthesized composite samples.

Neha Pal, Madhu Agarwal, Ragini Gupta [35] Polysaccharide-based guar gum (GG) biopolymer is

used via a hydrothermal process to synthesize silver nanoparticles. The GG biopolymer act as a reducing and stabilizing agent. Moreover, GG was used for preparing peel-off masks to provide the desired consistency of formulation and synthesis of nanoparticles as an antibacterial agent. This work presents the novel GG/Ag nanoparticles peel-off gel and evaluates the antibacterial efficiency. The synthesized Ag-nanoparticles analyzed by UV–spectroscopy reflect a prominent peak at 413 nm. The size and distribution of nanoparticles were examined by TEM images obtained from the 6 to 18 nm range. We demonstrate the efficiency of peel-off facial gel as an antibacterial and preservative-free cosmetic product at different temperature ranges. The RSM study was used for parameter optimization of peel-off gel for extrudability, spreadability, and drying time by employing a CCD. The results show that the optimized GG, PVA, and ethanol concentration were 3.47, 8.30, and 5.80 w/w%, respectively, with 0.02 w/w% Ag nanoparticles. The peel-off gel antibacterial activity against Escherichia coli (11 ± 0.1 mm), Staphylococcus aureus (10 ± 0.3 mm), and Propionibacterium acnes (11 ± 0.3 mm). The peeloff gel was prepared from natural ingredients; due to this, it is nontoxic for human skin.

3. Experimental Procedure-

3.1. Materials

Sodium hydroxide and GG powder were purchased from SRL. After being purified, GG has been utilized. From Qualigens, silver nitrate has been purchased. The creation of all aqueous solutions used deionized water.

3.2. Methods

3.2.1.Purification of Guar Gum (GG)

The commercial guar gum was purified by taking Crude guar gum (10 g) was stirred in 250 ml of 5% of NaOH solution in distilled water for 24 h at room temperature with continuous stirring. Then the solution pH was adjusting at 7 by acetic acid and then purified it by 70 % ethanol wash. Then the sample is dried in the hot air oven at 50^{o} C. The yield of the pdt was 80%.

3.2.2.Preparation of Silver Nitrate Solution

To prepare 50 ml of 20 milimolar silver nitrate solution –

Molecular Weight of $AgNO_3$ = 169.87

For 1000 ml 1 (M) $AgNO_3$ solution 169.87 gm of $AgNO_3$ is required.

For 50 ml of 40 milimolar $AgNO_3$ solution the amount of silver nitrate required is

$= 169.87 \times 20 \times 10^{-3} / 20$

= **0.1698 gm** of solid $AgNO_3$ is dissolved in 50 ml of distilled water to prepare desired AgNO3 solution.

3.2.3. Synthesis of GG -Ag NPs at different pH variation :

In order to achieve the best size and size distribution of Ag nanoparticles, GG Ag NPs were synthesized at various GG weight concentrations. This study focused on controlling the reaction parameters. The concentration of reactants, such as guar gum and AgNO3, has an impact on this optimization process. Ag NPs were created using guar gum as a stabilizing and reducing agent.

Table. 1. Preparation of GG AgNps solution by varying different pH :

Amount of GG taken	Volume Of Water in ml	Volume of $AgNO_3$ in ml
1%	8.5	1.5

4. Result & Characterization-

4.1 FTIR-

FTIR spectroscopy (Model-Alpha, Bruker, Germany) with 8 consecutive scans at 1 cm-1 resolution and a frequency range of 4000-600 cm-1 was used to obtain the FTIR spectra of GG and GGAgNps.Zetasizer Nano ZS (Malvern Instrument, UK) was used to measure the zeta potential value and the size of the GG, GGAgNps particles under various GG concentration, silver nitrate concentration, and time variation variations.By using a nanoliter cell and an UV-vis spectrophotometer (OPTIZEN POP BIO, Mecasys), nanoparticle formation was verified. Zetasizer Nano ZS (Malvern Instrument, UK) UV-Vis spectroscopy is another method used to confirm the polyethyleneimine polymer coating.

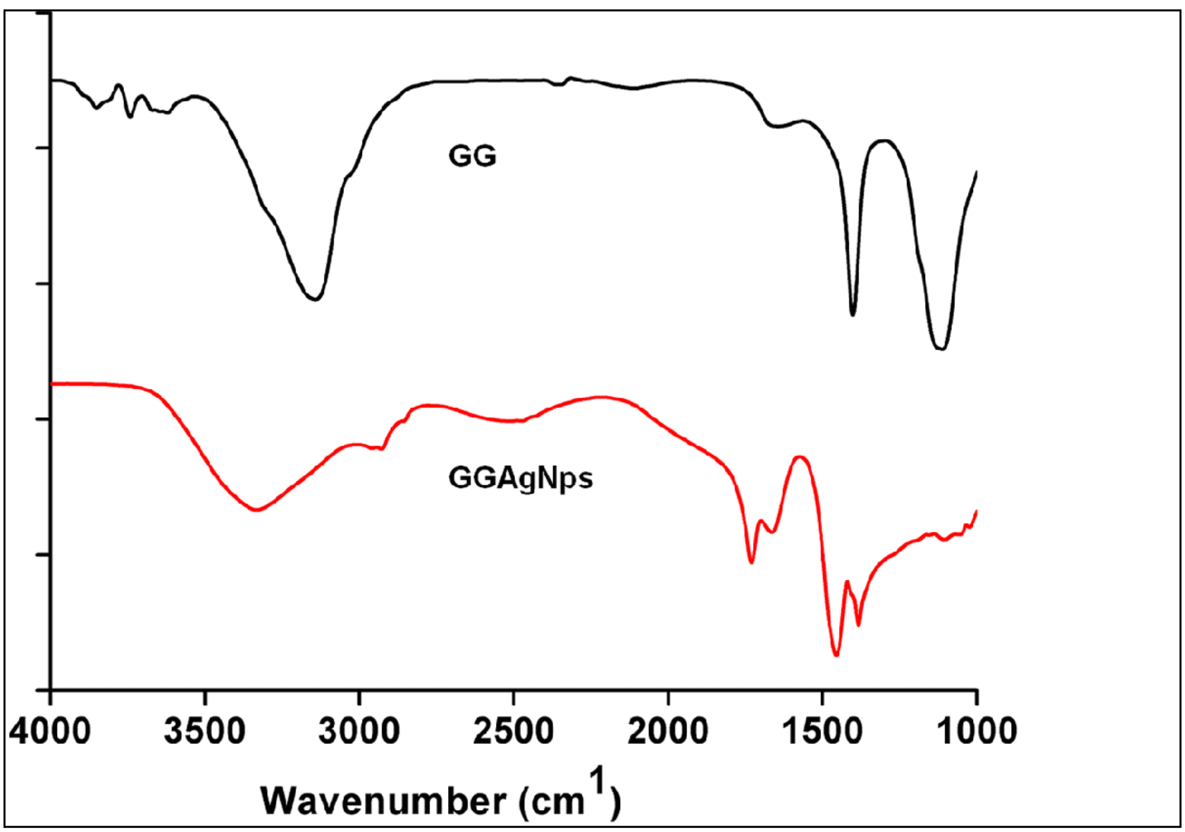
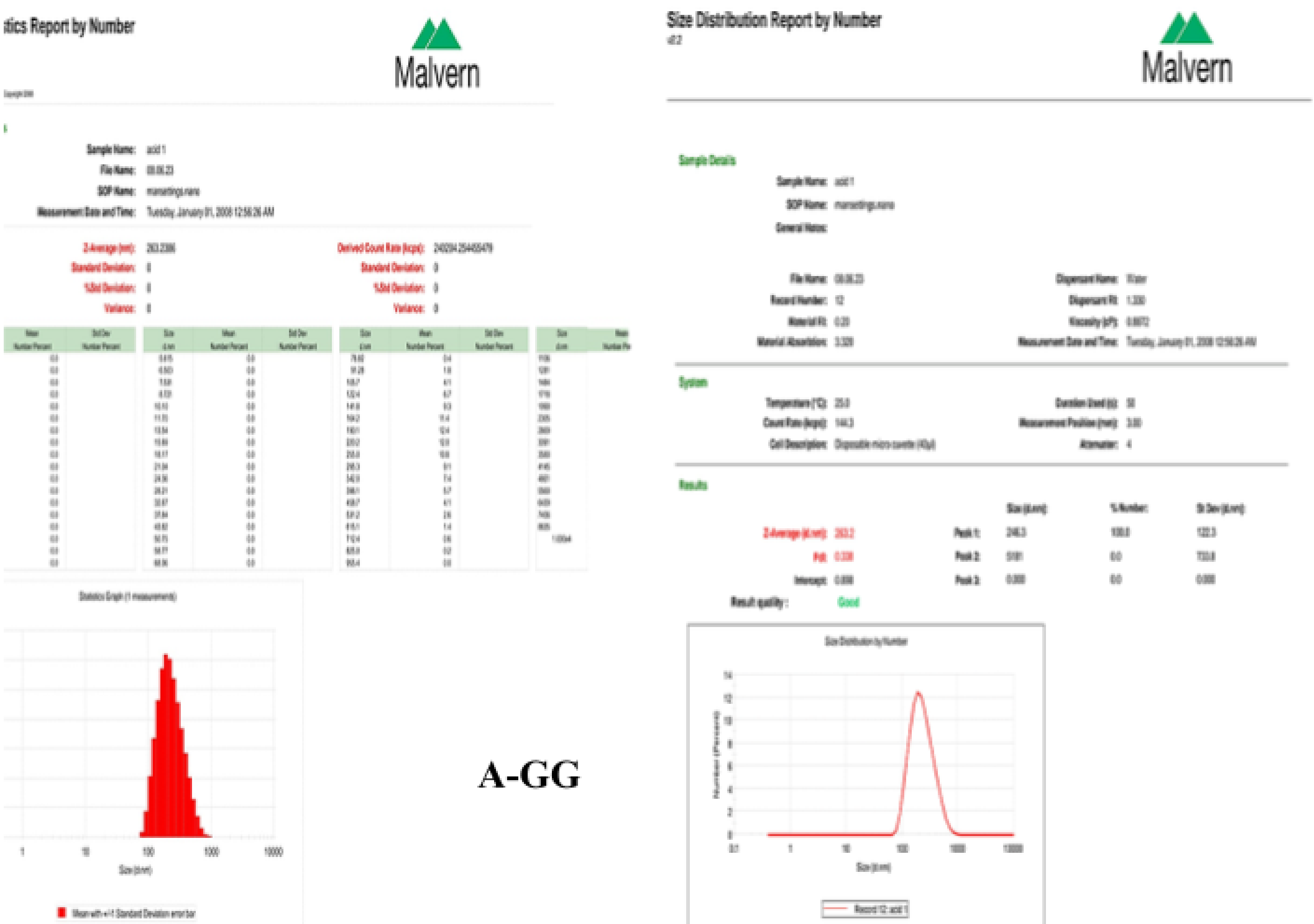


Figure.2 FTIR for GG and GG AgNPs

In the current study, purified GG was used as a stabilizing and reducing agent to make silver nanoparticles. After GG-AgNps were created for additional cationic modification, PEI was coated on the nanoparticle for straightforward ionic interaction. Fig. 2 shows the FTIR spectrum of GG and GG-AgNps. The spectrum of pure GG exhibits absorption bands at approximately 3386, 2928, and 1150 cm-1 because of the presence of O-H, C-H, and C-O stretching vibrations, as shown in Fig. 6. The absorption band near 1025 cm-1 was brought on by the glyosidic linkage of the pyranose ring of GG.The band at 3426 cm-1 shifted to 3457 cm-1 in the presence of Ag, and in Ag/GG, the band was wider than in GG, as shown in the figure below. These findings unequivocally demonstrate the interaction between Ag and the —OH group of GG. Similar results were previously reported in the synthesis of silver nanoparticles stabilized by GG backbone.

4.2 Dynamic Light Scattering (Size distribution)

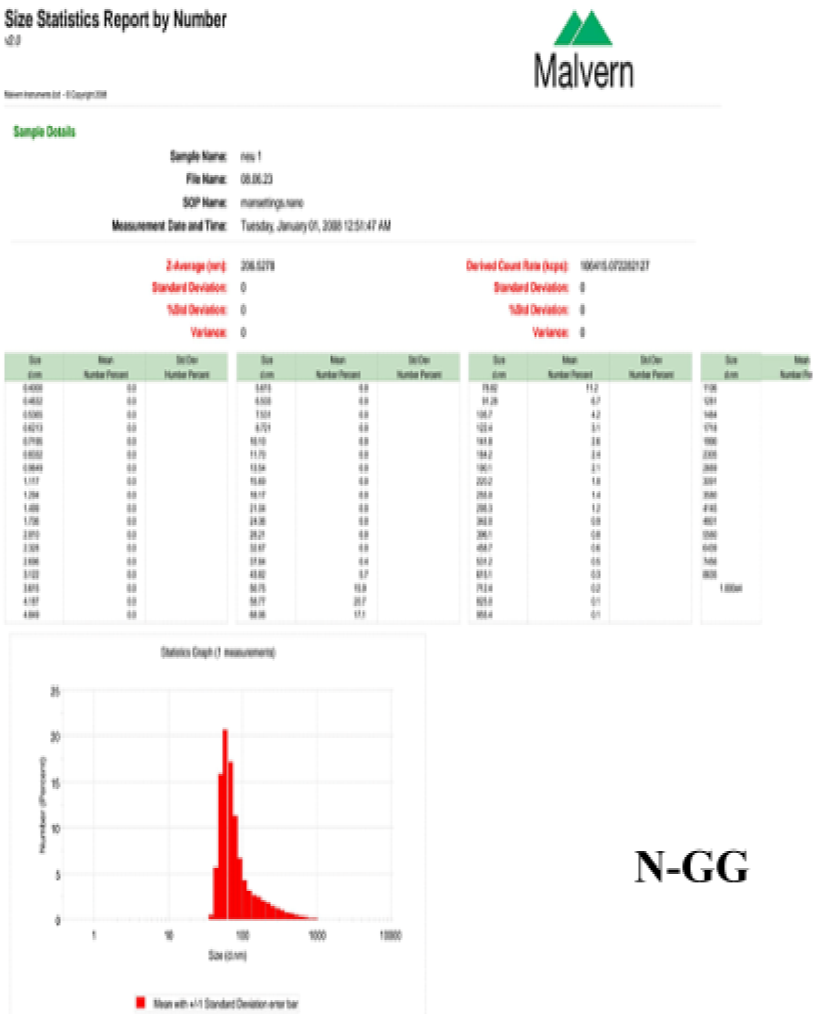
Depending on the particle size, the dielectric medium, and the chemical environment, AgNPs absorb differently. observation of this peak related to the surface Plasmon is renowned for its wide range of metal nanoparticles, which have sizes between 2 and 100 nm. We evaluated the stability of biologically produced AgNPs. The physicochemical characterization of prepared nanomaterials is essential for the analysis of biological activities using radiation scattering techniques. A scale ranging from submicron to one nanometer can be used by DLS to analyze the size distribution of small particles in suspension or solution. Dynamic light scattering is a method that depends on the interaction of light with particles. Narrow particle size distributions can be measured using this method, especially in the 2-500 nm range. One of the techniques used most frequently for characterizing nanoparticles is DLS. In order to measure the light scattered from a laser that passes through a colloid, DLS primarily uses Rayleigh scattering from the suspended nanoparicles. The hydrodynamic size of the particles can then be calculated by examining how the intensity of the scattered light changes over time. Any nanomaterial's characterization in solution is necessary to assess its toxic potential. In order to ascertain particle size and size distributions in aqueous or physiological solutions, DLS is primarily used.



A-GG

Fig 3 A: Particle size of acidic silver nano(A-GG) particle

From the above Fig 3 A, it is clear that statistical and size distribution results are the same. The size distribution between the range of 100 to 1000 is the same.



N-GG

Fig 3 B: Particle size of Neutral silver nanoparticle (N-GG)

According to the UV plot for GG-$AgNO_3$ neutral nanoparticles it gave a peak around 460 nm , but its intensity is lesser than the basic condition. For that reason, the particles are formed. According to the distribution graph Fig 3 B, Almost 50% of particles fall under the range of less than 100.

B-GG

Fig 3 C: Particle size of Basic silver nanoparticle (B-GG)

According to the UV plot for GG-AgNO3 basic nanoparticles it gave a peak around 420 nm , its intensity is very higher. For that reason, the concentration of particles is higher. According to the distribution graph Fig 3 C, the size of the polymeric nanoparticles is in the range of 50-100 nm . Fully 100 % of particles fall under the range of less than 100 , so in this case highest GG-AgNO3 nanoparticles are formed.

4.4 UV-Visible Spectroscopy -

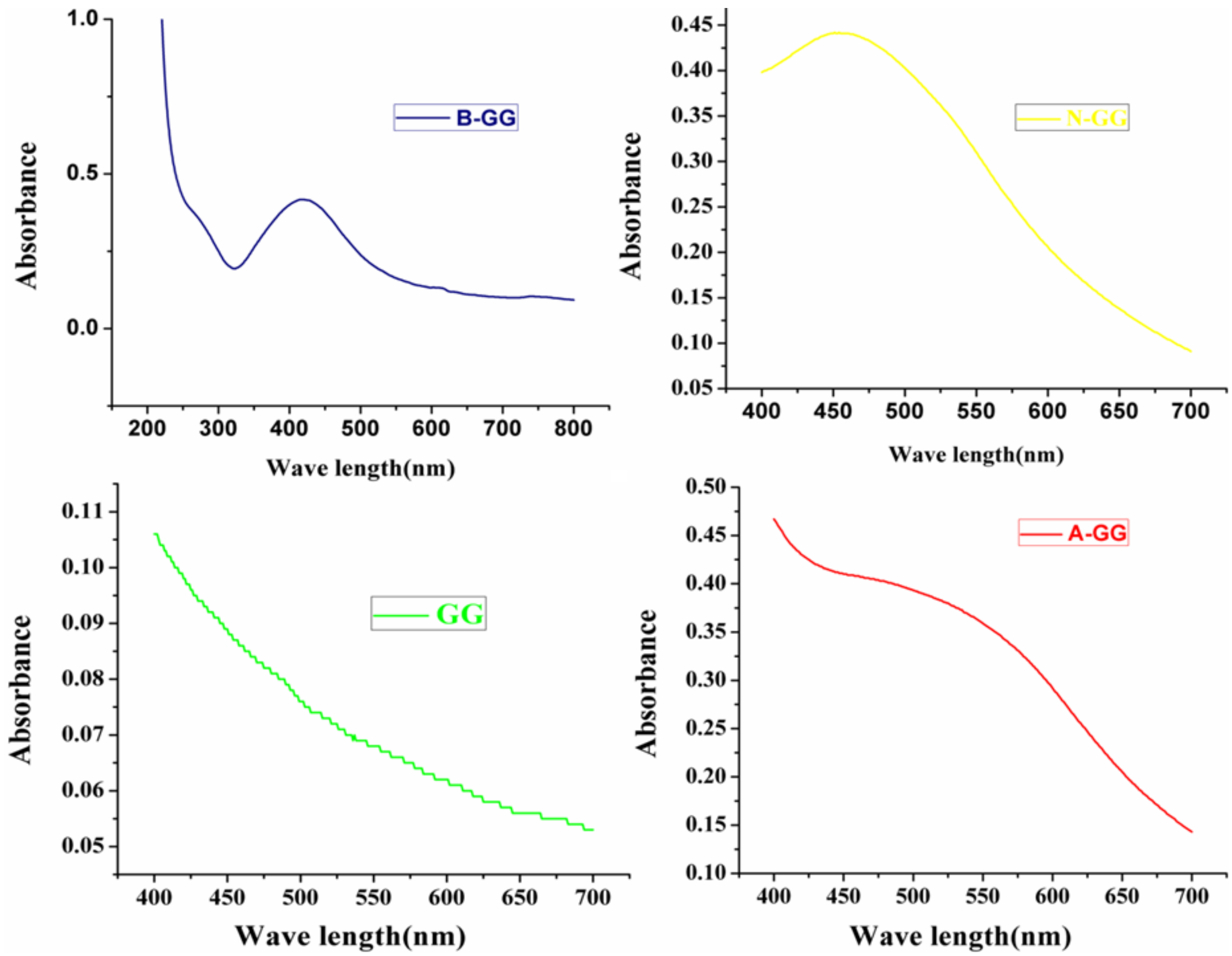
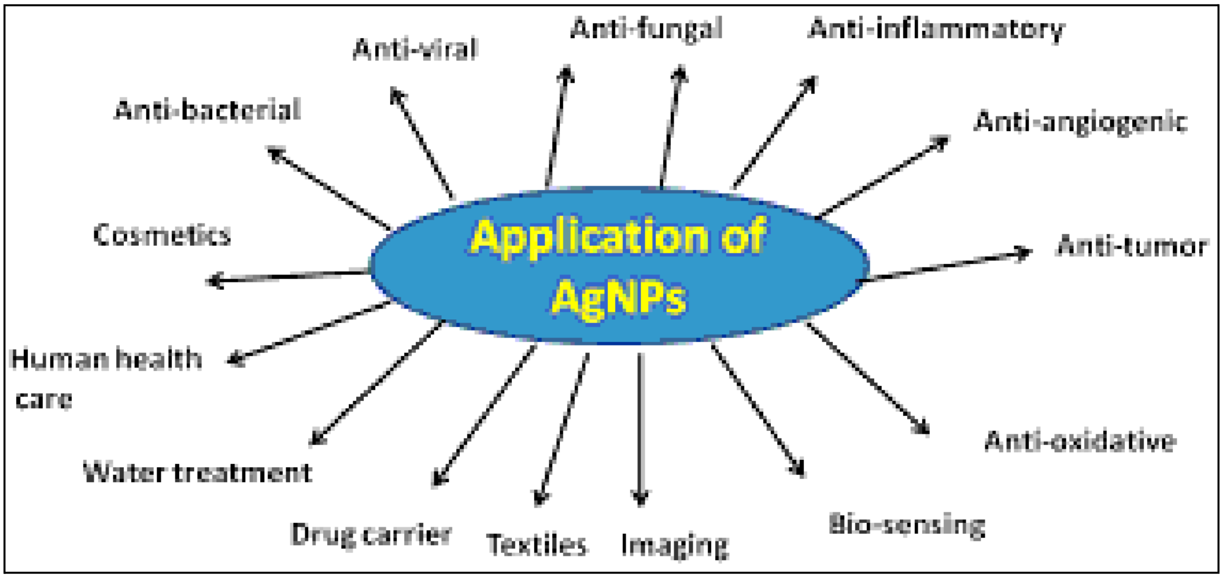


Fig4: UV spectra of GG (Guargum), A-GG(Acidic silver nanoparticle), N-GG(Neutral silver nanoparticle), B-GG(Basic silver nanoparticle).

UV-vis spectroscopy is a very useful and reliable technique for the primary characterization of synthesized nanoparticles which is also used to monitor the synthesis and stability of AgNP.

In addition, UV-vis spectroscopy is fast, easy simple, sensitive, selective for different types of NPs, needs only a short period time for measurement, and finally a calibration is not required for particle characterization of colloidal suspensions . In AgNPs, the conduction band and valence band lie very close to each other in which electrons move freely. UV-visible spectra of the synthesized composite and that of the polymer are shown in Fig. 4. The Spectra represents the absorption bands corresponding to polymer mixture GG-AgNO3 nanoparticles by varying pH . The B-GG(Basic silver Nanoparticles) showed the absorption band in the region 420–450nm.The appearance of band in the region 400–500, indicates the presence of silver nanoparticles and it gives high intense colour. The exact position of the band depends on the size of nanoparticles which can be confirmed from previous literature review. The absorption band (0.5) is maximum at wavelength value 420 nm about, is observed for Basic GG/Ag-NO3 nanoparticles sample. The absorption band (0.45) is observed at wavelength 460 nm for neutral GG/Ag-NO3 nanoparticles sample . For pure GG no absorption band is seen and for A-GG(Acidic silver nanoparticles) slight absorption band at 0.36 at wavelength of 540 nm is observed. From this result we can conclude that in basic GG/Ag-NO3 nanoparticles sample there is high absorption band so, the production of GG/Ag-Nps is maximum in basic GG/Ag-NO3 nanoparticles sample and it gives highly intense colour.

5. Application Of Silver Nanoparticle –

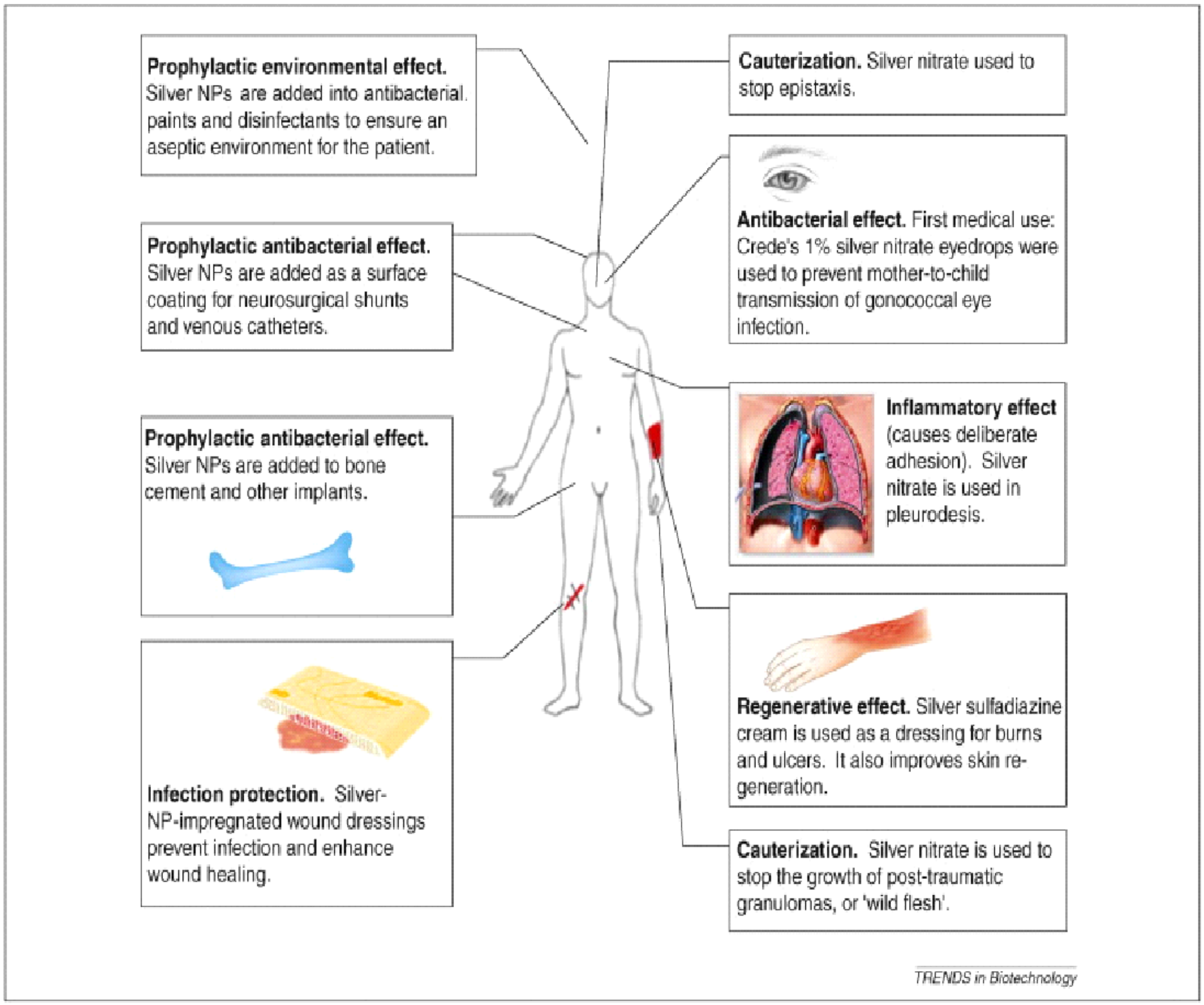


Some application of AgNPs

Because of their attractive and distinctive nano-related characteristics, such as their potent intrinsic antimicrobial effectiveness and non-toxic makeup, silver nanoparticles have attracted a great deal of interest in the biomedical field. AgNPs have a wide range of potential uses in this field, but recently, impressive attention and efforts have been focused on their promising applications as nanomaterials for wound dressing, tissue scaffolding, and protective clothing [21,22]. AgNPs' inherent physical and chemical properties, such as maintaining their nanoscale size, enhancing their dispersion and stability, are some fundamental aspects related to their specific antimicrobial properties, and avoiding aggregation [22]. There are many studies which experimentally proved that the anti-pathogenic activity of AgNPs is better than that exhibited by silver ions [23]

As important as a drug's inherent therapeutic effects are in medicine, its pharmacokinetics and pharmacodynamics [25]. Nanoparticles have attracted a lot of attention in the design and development of new and improved drug-delivery systems because the specific and selective delivery and action of therapeutic agents has emerged as one of the most researched topics for enhancing current human healthcare practice [26]. A number of therapeutic molecules, including anti-inflammatory [27, 28], anti-oxidant [29], antimicrobial [31], and anticancer [30] bio substances, have been tested as viable carriers in AgNP-based Nano systems.

AgNPs play a significant role in the creation and application of novel biomedicinal strategies due to their distinctive physiochemical properties and bio functional features, such as anti-inflammatory, anti-angiogenesis, antiplatelet, antiviral, antifungal, and antibacterial activities [25]. Recent studies have closely examined the anticancer effects of AgNPs in a variety of human cancer cell lines, including endothelial cells, IMR-90 lung fibroblasts, U251 glioblastoma cells, and MDA-MB-231 breast cancer cells [22,23]. By using energy-driven internalization pathways, AgNPs have the innate ability to bind to and easily penetrate mammalian cells [21]. AgNPs also have a unique fluorescence, which makes them desirable candidates for dose-enhancement and detection applications in X-ray irradiation applications [26]



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6. Conclusion

This review thoroughly covered the synthesis, characterization, and bio-applications of guar gum-based silver nanoparticles, with a focus on the anticancer activity and its mechanisms as well as therapeutic approaches for cancer using AgNPs. Guar gum was used as a green reducing agent in this review. Due to the common side effects of chemotherapy and radiation therapy, recent academic and industrial research has looked into the possibility of using AgNPs as a next-generation anticancer therapeutic agent. Despite the fact that AgNPs are crucial for clinical research, a number of factors need to be taken into account, such as the source of the raw materials, the production process, stability, biodistribution, controlled release, accumulation, cell-specific targeting, and finally human toxicological concerns. Undoubtedly one of the most intriguing methods for treating cancer is the development of AgNPs as anti-angiogenic molecules. Poor delivery and the issue of drug resistance can be solved by it. Additionally, it might open up a new door for other angiogenic conditions like endometriosis, diabetic retinopathy, and obesity. GG is a readily available, reasonably priced, and environmentally friendly green biopolymer, and the preparation work required for this study is surprisingly easy. It can offer a simple and environmentally friendly method for producing metallic nanoparticles, antimicrobial substances, catalysts, and other useful materials.

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