Synthesis, Characterization and *in vitro* evaluation of Magnesium oxide Nanoparticles as Potential Anti-diabetic agents

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Abstract

The current study objective is to synthesize Mgo nanoparticles through green mediated means employing leaf extracts from Acalypha indica for biological purposes. The produced Mgo nanoparticles' optical, chemical and morphological characteristics were verified using UV–Vis spectroscopy, FT-IR, zeta potential, XRD and FE-SEM, in that order. Mgo nanoparticles' optical properties and functional groups were examined using UV visible absorption spectra and FT-IR analysis. The antibacterial evaluation of the generated Mgo nanoparticles was investigated using the disc diffusion method against microbial pathogens, including Gram-positive bacteria Staphylococcus aureus and Bacillus subtilis, as well as Gram-negative bacteria Escherichia coli and Klebsiella pneumoniae.

Amazing in vitro antibacterial, antioxidant and anti-diabetic properties of Mgo Nanoparticles synthesised using green assisted synthesis were noted. As a result, the current study concludes that environmentally safe, low-cost and green mediated magnesium oxide nanoparticles are the best materials for biological applications.

Keywords: Nanotechnology, Magnesium oxide Nanoparticles (Mgo NPs), Antimicrobial activity, Antioxidant activity and Anti-diabetic activity.

Introduction

The newest and most promising field of study in the contemporary medical sciences is nanotechnology. Technological advancements in biotechnology and nanotechnology have enabled the field of nanoparticle synthesis study. Nanoparticles (NPs) are widely employed in the therapies for diabetes, allergies, infections, cancer and swelling. Applications of NPs in variable sectors have been reported including medical[17], cosmetics[21], renewable energy[20], environmental clean-up, transportation[16]. Metal nanoparticles are employed extensively in the industrial, electronics and biomedical sectors and have significant optical, thermal and chemical properties. Until now, a variety of conventional techniques have been employed in the synthesis of nanoparticles such as co-precipitation, spray pyrolysis, sonochemical and combustion.

All of the above methods require costly tools, labor-intensive processes, hazardous materials and are not eco-friendly. To overcome these obstacles, a different method that makes use of green chemical approaches is utilized to produce nanoparticles[18]. Bio mediated materials, such as bacteria, fungus, plants and enzymes, have been used recently for green synthesis[8]. In addition to providing an abundance of active metabolites, plants are more environmentally friendly option than other materials for the manufacturing of nanoparticles in bulk[14]. A lot of emphasis has been paid to green synthesis as a dependable, environmentally acceptable and sustainable method for creating a wide range of nanomaterials. Growing interest is being shown in the alternative method of synthesising metal nanoparticles utilizing plant extract.

Certain properties of the metal nanoparticles that are obtained, like their antimicrobial activity and biocompatibility, are also dependent on the characteristics of the different phytochemicals that are present in the plant extracts that are used to synthesize them. Biomolecules found in plant extracts function as reducing and capping agents forming stable nanoparticles. Innovative uses for Mgo NPs have been found in electronics[7], adsorption[10], photocatalyst[22], ceramics[25], dielectric[23], chemical waste clean-up and detection of antibacterial and anticancer[19] agents. Furthermore, magnesium oxide nanoparticles are applied in the fields of biomedicine, agriculture and the environment to absorb harmful gasses, as well as for their antimicrobial, insecticidal and anti-film properties[11]. In the present study, the green synthesis of Mgo NPs is inexpensive, pollution free and environmentally safest material compatible for biological applications.

The Euphorbiaceae family includes *Acalypha indica*, a weedy annual herb plant[6] also known as Indian mercury or Indian copper leaf[15]. India is one among the countries where *Acalypha indica* is most frequently produced. The plant's leaves have been used to treat rheumatoid arthritis, syphilitic ulcers[9], scabies[13] and purgatives, diuretics and antihelmintics[24]. Despite having a variety of medical uses, *Acalypha indica* is utilized in the production of magnesium oxide nanoparticles (Mgo NPs).

Material and Methods

Collection of leaves: The *Acalypha indica* leaves are gathered in the vicinity of Sri Padmavati Mahila

Visvavidyalam, Tirupati. The leaves are collected, cleaned under running water from the faucet and rinsed with deionized water to remove dust and other particle matter. They are then allowed to dry at room temperature and are ground into a fine powder to provide a broad surface area for absorption.

Fig. 1*: Acalypha indica*

Preparation of *Acalypha indica* aqueous leaf extract: 20 grams of leaf powder and 200 ml of deionized water were combined in a 500 ml conical flask. The mixture was heated at 60°C for 30 minutes using a thermostatic water bath. The aqueous extract was obtained by filtering it through Whatmann no. 1 filter paper, once it had cooled to room temperature. The extract was kept in a refrigerator between 4 and 10°C for later use.

Phytochemical screening: Qualitative tests were done to know the metabolites present in *Acalypha indica* leaf aqueous extracts by using standard protocols with slight modifications[2].

Synthesis of Magnesium oxide Nanoparticles: *Acalypha indica* extract was used to make the Mgo NPs in the following manner: 30 milliliters of plant extract were placed in a 500 milliliter beaker and add 150 milliliters of freshly made 5 millimeter magnesium nitrate followed by addition of 10 milliliters of NaOH dropwise and the mixture was continually agitated using a magnetic stirrer at 70^0C for two hours. After centrifugation for 15 minutes at 10,000 rpm precipitate was formed and then dried for 12 hours at 50^0C in a hot air oven.

The finely ground powder obtained was subjected to a variety of characterization procedures in order to determine the size, shape, purity and other physicochemical elements of the nanoparticles. The synthesized Mgo NPs were characterized by using different techniques like UV- Visible spectrophotometer, FTIR, X- ray diffraction (XRD), Zeta potential and Scanning electron microscope (SEM). FTIR was used to evaluate the functional groups that are present in synthesized Mgo NPs and the sample was prepared by mixing the sample and KBR in the ratio of 1:6 and measured at 4000 $cm^{-1}$to 400 cm^{-1}. SEM is used to study the morphology and crystalline size of MgO NPs. The stability was known by using Zeta potential analyser. The absorbance of Mgo NPs was noted by using UV-Visible spectrophotometer. The crystalline nature of prepared Mgo NPs was measured by XRD.

Fig. 2: Dried leaves and Crushed Powdered leaves

Fig. 3: Schematic representation of green synthesis of Mgo NPs

Antimicrobial activity: Microbiological pathogens that cause human disease including Gram-positive bacteria *Staphylococcus aureus* and *Bacillus subtilis* and Gram-negative bacteria *Escherichia coli* and *Klebsiella pneumoniae* were subjected to antibacterial activity by disc diffusion method. The bacteria were inoculated in the nutrient broth. This activity made use of the disc diffusion assessment. On the LB agar plates, 0.1ml of each bacterial sample was spread out. Mgo NPs are placed in wells with the help of micropipette and then incubated for 24hr[5]. After incubation, Zone of inhibition was measured in mm.

DPPH scavenging technique: Using the usual approach with slight modifications, the DPPH scavenging activity test of plant extract was estimated[4]. An ethanol-based 0.1 mM DPPH solution was prepared. One milliliter of Mgo NPs at a varying concentration and an equal volume of ethanol were added to three milliliters of DPPH stock solution. Following 30 minutes of incubation, absorbance at 517 nm was measured. The ascorbic acid standard curve is used to quantify the antioxidant activity of the sample. The following formula was used to represent the DPPH scavenging capacity:

$$\% \text{ Inhibition} = \frac{\text{Blank} - \text{Sample}}{\text{Blank}} \text{X } 100$$

α-Amylase Inhibitory Activity: Mgo NPs may interact with alpha-amylase, either enhancing or inhibiting its catalytic activity. The activity is typically measured by quantifying the amount of reducing sugars (such as maltose and glucose) released from starch, using a colorimetric method (such as DNS assay). Different concentrations of Mgo NPs extracts were prepared in phosphate buffer using a 1 mg/ml stock solution. 500µl of α-amylase (0.5 mg/ml) was added to different concentrations of Mgo NPs and standard acarbose. The mixture was allowed to sit at room temperature for 10 minutes. 500µl of a 1% starch solution was added and the entire mixture was incubated for 10 minutes.

The reaction mixture was now treated to 1 ml of colouring DNS reagent (95 mM 3, 5-dinitrosalicylic acid and 30 gms NaK tartarate in 0.5M NaOH) to stop the enzymatic reaction. The reaction mixture was then heated for 15 minutes in a boiling water bath followed by addition of distilled water. To compute the absorbance of the colored extracts, buffer was replaced for each concentration of the enzyme in the sample set to create a blank. At 540 nm, the absorbance was calculated[3]:

% inhibition = (control absorbance- sample absorbance/control absorbance) ×100

α-Glucosidase Inhibitory Activity: The conventional protocol was followed with minor modifications for the alpha-glucosidase assay of the magnesium oxide nanoparticles[12]. For fifteen minutes at 37°C, samples and the standard (acarbose) at different concentrations were incubated inside a 96-well plate that contains 50 µL of phosphate buffer (100 mM, pH= 6.8) and 10 µL of alpha-glucosidase (1 U/mL). In brief, 20 µL of a 5 mM substrate (4-nitrophenyl β-d-glucopyranoside) was added to each well and the mixture was incubated for 20 minutes at 37°C. After incubation, add 100 µL of sodium carbonate (0.1 M) to each well to stop the reaction.

Sodium carbonate stops the reaction by raising the pH, which also develops the yellow color of p-nitrophenol (pNP). The absorbance of the samples was measured at 405 nm using a spectrophotometer. The intensity of the yellow color is proportional to the amount of pNP released, which correlates to the enzyme's activity:

% inhibition = (control absorbance- sample absorbance/control absorbance) ×100

Glucose Uptake by Yeast Cells: With minor modifications, this assay was conducted in accordance with Abubakar et al's[1] methodology. 1% suspension was developed by dissolving commercial baker's yeast in distilled water. Overnight, the suspension was kept at room temperature, 25°C. The yeast cell samples were centrifuged for five minutes at 4000 rpm on the next morning. This process was repeated with the addition of distilled water to the particle until a clear supernatant was obtained. 10% v/v solution of the yeast cells was made by combining ten parts of the clear supernatant fluids with ninety parts of distilled water. After that, the mixture was combined with 5 milligrams of one milliliter of glucose solution and it was heated to 37° C for ten minutes:

% inhibition = (control absorbance- sample absorbance/control absorbance) × 100

Results and Discussion

Phytochemical screening of *Acalypha indica* aqueous leaf extract shows the presence of phenols, flavonoids, glycoside, saponins, steroids, terpenoids, proteins and carbohydrates. Compounds like flavonoids and phenolic acids present in plant extracts facilitate the reduction of magnesium salts to form Mgo NPs. They also help in stabilizing the nanoparticles, preventing aggregation.

Characterization of Mgo NPs

UV-VIS Spectroscopy: The UV-VIS method was utilized to measure the absorbance of the manufactured Mgo NPs in order to confirm that Mgo NPs were effectively created. The UV-VIS spectrum was shown to have two peaks between 200 and 800 nm. The Mgo NPs are responsible for the largest peak which was seen at 250 nm.

XRD Analysis: The results of XRD show little amount of impurities for synthesized Mgo NPs. In this study diffraction peaks was observed at 28.4025 and 20.284. It is reported as amorphous in state.

Fourier transform infrared analysis (FTIR): The FTIR spectrum of the synthesized Mgo NPs shows six major bands shown in fig. 6. These FTIR characteristic bands correspond to different biologically active functional groups. Table 2 contains the functional groups and the mode of vibration. The vibrational frequency of the expansion and bending modes of Mgo NPs molecules can thus be determined by FTIR analysis.

SEM Analysis: The size and form of Mgo NPs were revealed by the SEM analysis. The SEM pictures generally showed that the morphological surfaces were composed of spherically shaped assemblies of nanoparticles. According to SEM scans, the nanoparticles were between 30 and 60 nm in size.

Particle size measurement of Mgo NPs: The mean and polydispersity index (PDI) of Mgo NPs produced with *Acalypha indica* were examined by the use of a dynamic light scattering approach. A 0.2 mV Zeta potential mean was used to evaluate the surface charge of the biosynthesized Mgo NPs.

Zeta potential of Mgo NPs: Mgo NPs produced from *Acalypha indica* have an average size of 22.4 nm. The Mgo NPs produced from *Acalypha indica* have a PDI value of 0.715.

Antimicrobial activity: Among the most adaptable techniques for determining sensitivity to antimicrobial drugs is the disc diffusion approach.

Table 1
Phytochemial screening of *Acalypha indica* leaf extract

S.N.	Phytochemicals	Aqueous extract
1.	Phenols	++
2.	Flavonoids	++
3.	Steroids	++
4.	Terpenoids	+
5.	Alkaloids	-
6.	Tannins	-
7.	Glycosides	+
8.	Proteins	+

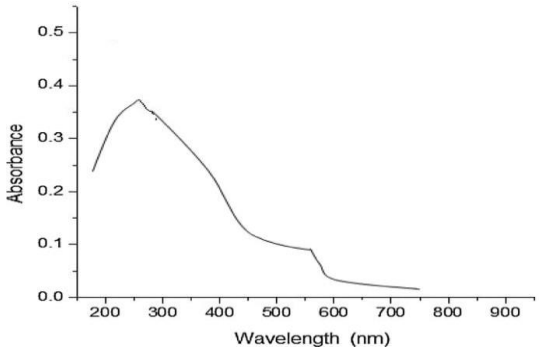

Fig 4: UV-VIS spectrum of the Mgo NPs

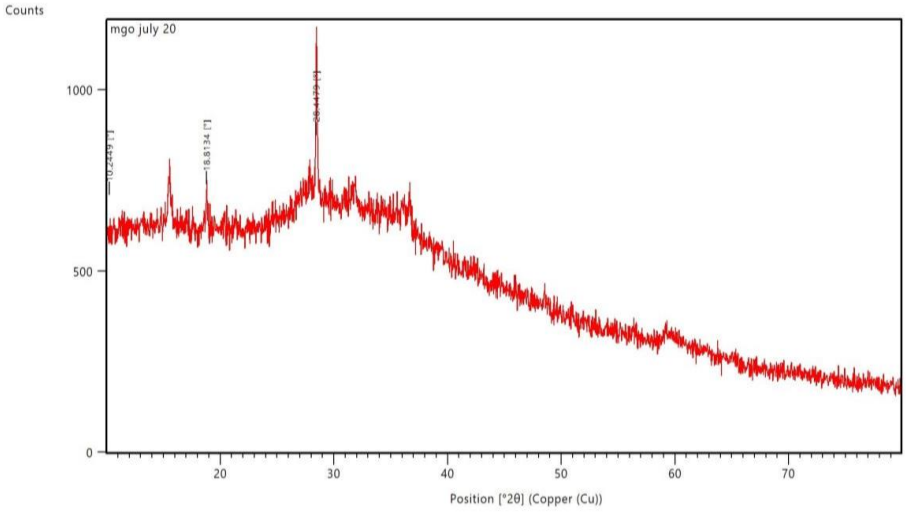

Fig. 5: XRD pattern of Mgo NPs

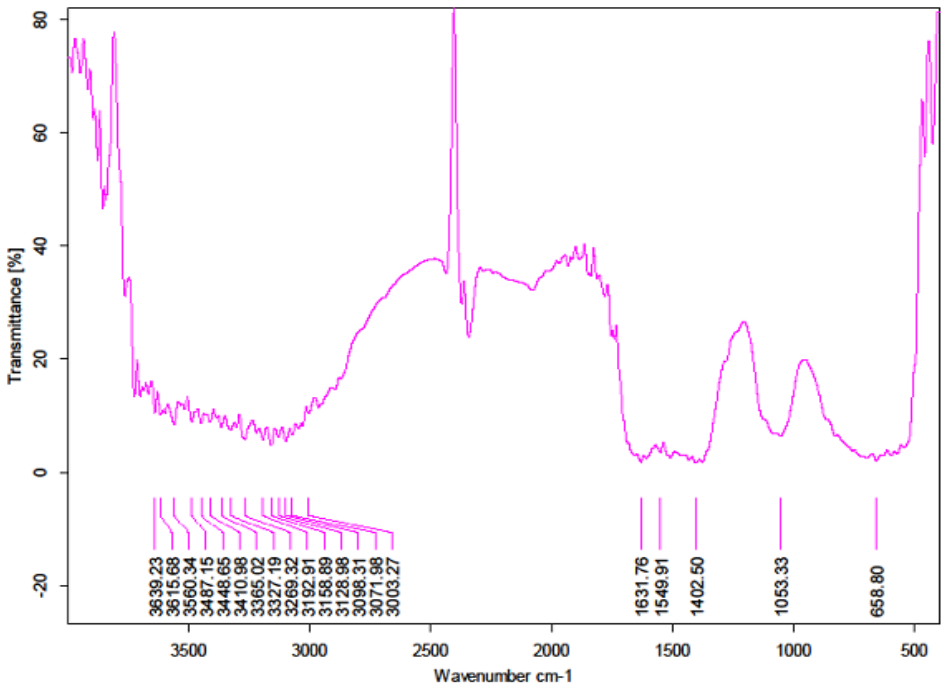

Fig. 6: FTIR spectrum of the synthesized Mgo NPs showing bands related to the capping molecule contained in the *Acalypha indica* extract and Mgo NPs.

Table 2
FTIR functional groups based on their values

S.N.	Frequency	Functional groups	Mode of vibration
1.	3639-3615	Phenols	OH-stretch
2.	3560.34	alcohol	O-H stretch
3.	3560-3410	Bonded amines and amides	N-H stretching
4.	3269-3192	Alkene	C-H Stretching
5.	3158-3128	Alkyne	C-H Stretching
6.	3098-3003	Alkene group.terminal(vinyl)	C-H stretching
7.	1631-1549	Alkenes group	C=C stretch
8.	1402-1053	Carboxylic acid	O-H Stretching
9.	658.80	Halogen compound(bromo-compound)	(C-C1)

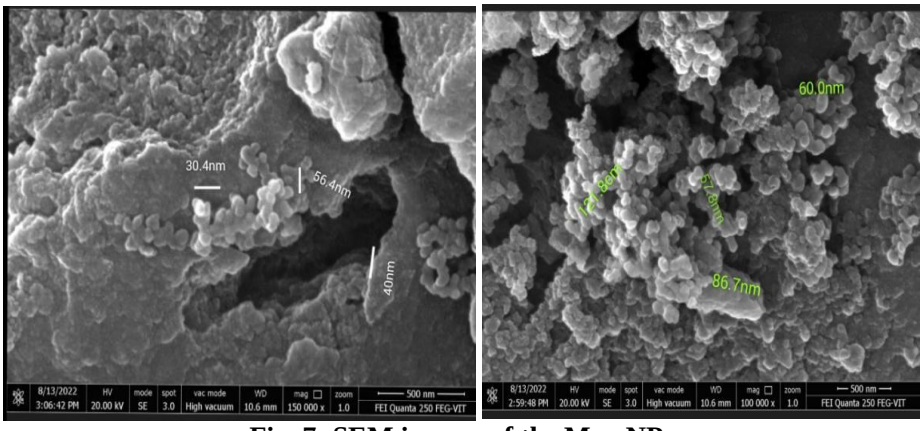

Fig. 7: SEM images of the Mgo NPs

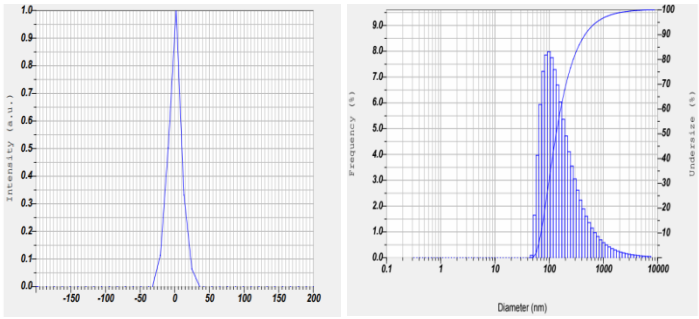

Fig. 8: Zeta potential and particle size of Mgo NPs

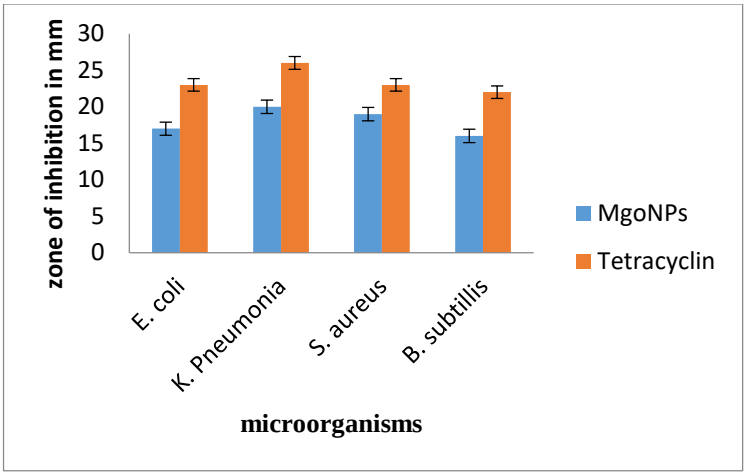

Fig. 9: Antimicrobial activity of Mgo NPs

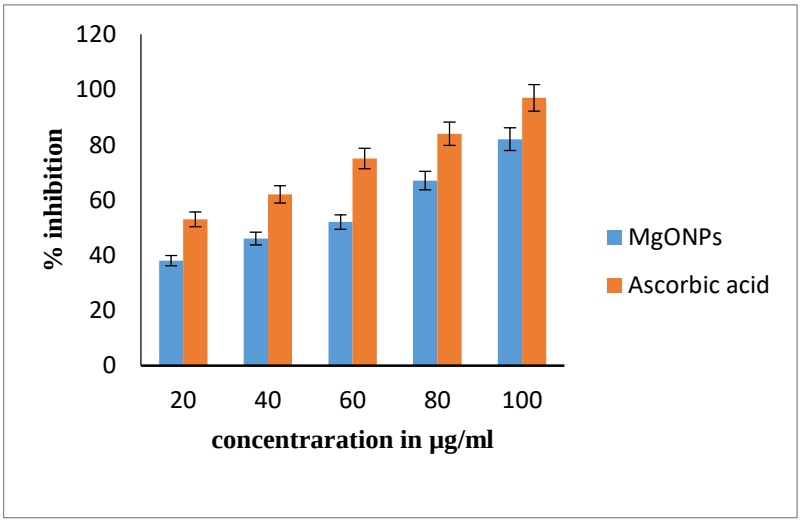

Fig. 10: DPPH activity of Mgo NPs

The procedure entails putting an antibiotic disc in the centre of the plate and applying paper discs saturated with antimicrobial agents to microorganisms on the agar medium's surface. The inhibitory zone is influenced by concentrations.

DPPH Assay: The 2-diphenyl-1-picrylhydrazyl (DPPH) assay was then used to assess the antioxidant activity of the Mgo NPs for this investigation. The assay typically involves the tested antioxidant molecules interacting with DPPH to convert it to DPPH-H. This results in a color shift, initially appearing deep purple with a strong absorption peak at 517 nm; however, after the antioxidant is added, the color changes to a pale yellow and finally becomes colorless. The degree of discolouration is a measure of the antioxidant chemicals' scavenging activity. Figure 10 displays the results for the scavenging potentials of the phytochemically generated Mgo NPs and the conventional (ascorbic acid) Mgo NPs. An IC50 value of 58.25 µg/mL was obtained by measuring the DPPH scavenging activity of Mgo NPs in order to evaluate their antioxidative potential.

α-Amylase Inhibitory Activity: The alpha-amylase inhibition assay is commonly used to evaluate the potential

of a compound to inhibit the enzyme alpha-amylase which breaks down starch into glucose. Inhibiting this enzyme can help control postprandial (after-meal) glucose levels, making it relevant in diabetes research.

α-Glucosidase Inhibitory Activity: The alpha-glucosidase inhibition assay is commonly used to evaluate the inhibitory potential of compounds against the enzyme alpha-glucosidase. Alpha-glucosidase is responsible for breaking down complex carbohydrates into glucose in the small intestine. Inhibiting this enzyme can help to manage postprandial blood sugar levels, which is especially relevant for diabetes research.

Glucose Uptake by Yeast Cells: Yeast cells (e.g. *Saccharomyces cerevisiae*) have transport systems that uptake glucose from the surrounding medium. The assay involves incubating yeast cells with glucose in the presence of Mgo NPs and measuring the amount of glucose remaining in the medium. A decrease in glucose concentration indicates uptake by the yeast cells. This can provide insights into their potential role in enhancing or inhibiting glucose uptake, which is relevant in diabetes research and metabolic studies. This methodology allows for the investigation of how Mgo NPs influence glucose uptake by yeast cells, providing insights into their potential metabolic or anti-diabetic effects.

Conclusion

Researchers' focus has switched from chemical to biological procedures because the former do not require the use of large machinery or equipment or a combination of harmful or toxic compounds that could endanger human health. Other inexpensive, widely available and plant- or microbe-mediated techniques are incorporated into the biological procedures. Because of the phytochemicals it contains, *Acalypha indica* has been used as a traditional medicinal plant. Currently, the biogenesis of the Mgo NPs in this study has been accomplished through the utilization of leaf extracts. Several microscopic and spectroscopic methods were used to confirm the produced Mgo NPs.

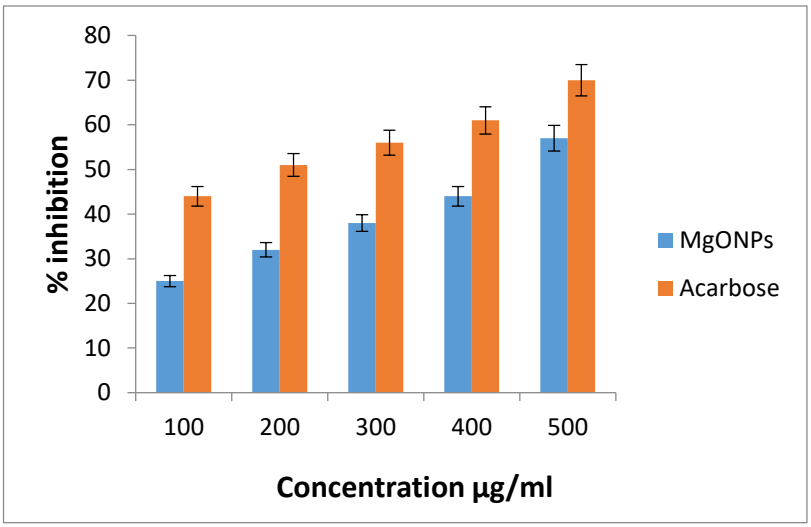

Fig. 11: α-Amylase Inhibitory Activity

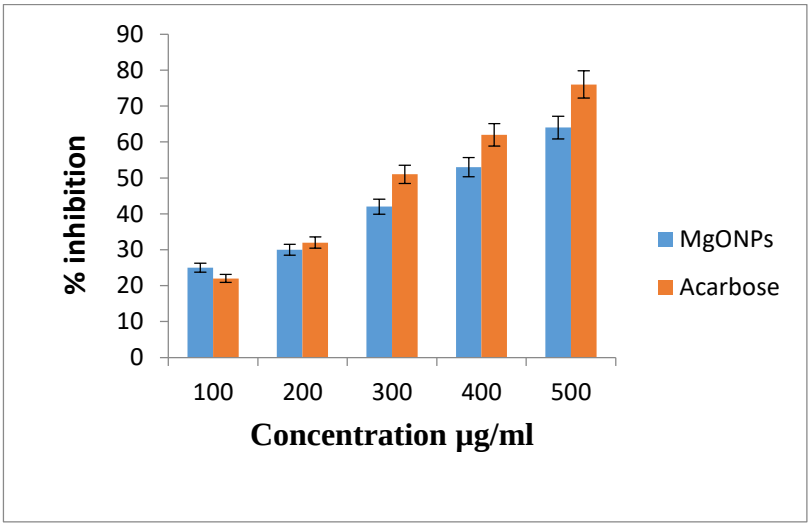

Fig. 12: α-Glucosidase Inhibitory Activity

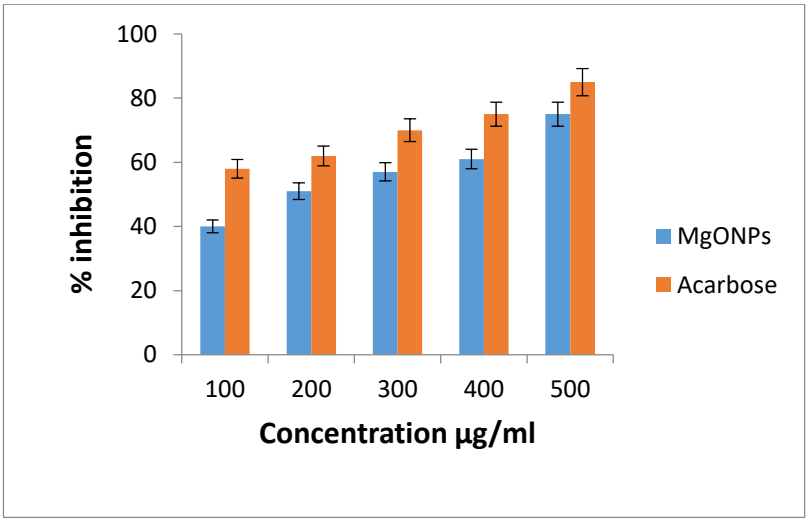

Fig. 13: Glucose Uptake by Yeast Cells

The generated Mgo NPs were assessed for their antioxidant and antimicrobial properties; as free radicals are known to cause a variety of human diseases. These nanoparticles show promising potential in *in vitro* antidiabetic studies, particularly in enzyme inhibition assays and glucose uptake studies. With further research and development, they could become an integral part of novel antidiabetic therapies. Mgo NPs will therefore be the ideal choice for therapeutic or medicinal purposes.

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References

1. Abubakar A.N., Lawal Z.B., Japeth E., Yunus I.O. and Garba R., *In vitro* antidiabetic potentials of crude saponins extract from Leptodenia hastata and Adansonia digitata leaves, *GSC Advanced Research and Reviews*, **6**, 61-66 **(2021)**

2. Adil M. et al, Phytochemical screening, HPLC analysis, antimicrobial and antioxidant effect of Euphorbia parviflora L. (Euphorbiaceae Juss.), *Scientific Reports*, **14**, 5627 **(2024)**

3. Bagyalakshmi J. and Haritha H., Green synthesis and characterization of silver nanoparticles using Pterocarpus marsupium and assessment of its in-vitro antidiabetic activity, *American Journal of Advanced Drug Delivery*, **5(3)**, 1–13 **(2021)**

4. Baliyan S., Mukherjee R., Priyadarshini A., Vibhuti A., Gupta A., Pandey R.P. and Chang C.M., Determination of antioxidants by dpph radical scavenging activity and quantitative phytochemical analysis of *Ficus religiosa*, *Molecules*, **27(4)**, 1-19 **(2022)**

5. Balraj B., Senthilkumar N., Vetha Potheher I. and Arulmozhi M., Characterization, antibacterial, anti-arthritic and *in-vitro* cytotoxic potentials of biosynthesized magnesium oxide nanomaterial, *Materials Science and Engineering*, **231**, 121–127 **(2018)**

6. Burkill H.M., The useful plants of West Tropical Africa, Royal Botanic Gardens, Kew, UK, **2**, 246 **(1985)**

7. Chen J., Lau Y., Coey J.M.D., Li M. and Wang J., High performance Mgo-barrier magnetic tunnel junctions for fexible and wearable spintronic applications, *Scientific Reports*, https://doi.org/10.1038/ srep42001 **(2017)**

8. De-Rui Di M.E., He Sun Z.Q. and Liu J., A new nano-cryosurgical modality for tumor treatment using biodegradable Mgo nanoparticles, Nanomedicine: Nanotechnology, *Biology and Medicine*, **8(8)**, 1233–1241 **(2012)**

9. Dhar M.L., Dha M.M., Dhawan B.N., Mehrotra B.M. and Ray B.N., Screening of Indian plants for biological activity: Part I, *Indian Journal of Experimental Biology*, **6(4)**, 232-247 **(1968)**

10. Ding J., Yu C., Lu J., Wei X., Wang W. and Pan G., Enhanced CO2 adsorption of Mgo with alkali metal nitrates and carbonates, *Applied Energy*, **263**, 1-7 **(2020)**

11. Fernandes M., Singh K.R.B., Sarkar T., Singh P. and Singh R.P., Recent applications of magnesium oxide (Mgo) nanoparticles in various domains, *Advanced Materials Letters*, **11(8)**, 1-10 **(2020)**

12. Govindappa M., Hemashekhar B., Manoj-Kumar A., Ravishankar Rai V. and Ramachandra Y.L., Characterization, antibacterial, antioxidant, antidiabetic, anti- inflammatory and antityrosinase activity of green synthesized silver nanoparticles using Calophyllum tomentosum leaves extract, *Results in Physics*, **9**, 400-408 **(2018)**

13. Gurib-Fakim A. Sewraj M. Gueho J. and Dulloo E., Medical ethnobotany of some weeds of Masuritius and Rodrigues, *Journal of Ethnopharmacology*, **39(3)**, 175-85 **(1993)**

14. Khan F.S.A., Mubarak N.M., Tan Y.H., Karri R.R., Khalid M., Walvekar R., Abdullah E.C., Mazari S.A. and Nizamuddin S., Magnetic nanoparticles incorporation into different substrates for dyes and heavy metals removal—A review, *Environmental Science and Pollution Research*, **27**, 43526–43541 **(2020)**

15. Kirtikar K.R. and Basu B.D., Indian Medicinal Plants, Volume I, 2nd ed., Dehradun, 314-317 **(1994)**

16. Matsukevich I., Lipai Y. and Romanovski V., Cu/Mgo and Ni/Mgo composite nanoparticles for fast, high-efficiency adsorption of aqueous lead(II) and chromium(III) ions, *Journal of Materials Science*, **56**, 5031–5040 **(2021)**

17. Mirtalebi S.S., Almasi H. and Khaledabad M.A., Physical, morphological, antimicrobial and release properties of novel Mgo-bacterial cellulose nanohybrids prepared by *in-situ* and *ex-situ* methods, *International Journal of Biological Macromolecules*, **128(1)**, 848-857 **(2019)**

18. Mohamed S.A., Prabhavathi G., Karunanithy M., Ayeshamariam A. and Jayachandran M., Green Synthesis of Nanoparticle by Plant Extracts-A New Aproach in Nanoscience, *Journal of Bio-nanoscience*, **12(3)**, 401–407 **(2018)**

19. Oginni O., Yakaboylu G.A., Singh K., Sabolsky E.M., Unaltosun G., Jaisi D., Khanal S., Shah A., Resources N. and States U., Phosphorus adsorption behaviors of Mgo modified biochars derived from waste woody biomass resources, *The Journal of Environmental Chemical Engineering*, **8(2)**, 1-11 **(2020)**

20. Papageridis K.N., Charisiou N.D., Douvartzides S., Sebastian V., Hinder S.J., Baker M.A., Alkhoori S., Polychronopoulou K. and Goula M.A., Continuous selective deoxygenation of palm oil for renewable diesel production over Ni catalysts supported on Al2O3 and La2O3–Al2O3, *RSC Advances*, **11**, 8569-8574 **(2020)**

21. Sambe A., Kuwayama T., Takenaka H., Yoshimune S. and Ishida K., The deodorant efficacy of MgO-SiO2 complex powder and application of the powder to antiperspirants, *Journal of Society of Cosmetic Chemists of Japan*, **29**, 55 **(1995)**

22. Scanning F.E., The effect of capping agent on the structural, optical properties and photocatalytic activity of Mgo nanostructures, *Physica B: Condensed Matter*, **583**, 1-7 **(2020)**

23. Selvi K.T., Mangai K.A., Priya M. and Sagadevan S., Investigation of the dielectric and impedance properties of Zno/Mgo nanocomposite, *Physica B: Condensed Matter*, **594**, 412355 **(2020)**

24. Varier V.P.S., Indian medicinal plants: a compendium of 500 species Orient Longman, Publication, Madras, India, 134 **(1996)**

25. Zhang C., Chen Y., Zhou M. and Li X., Achieving ultrahigh dielectric breakdown strength in Mgo-based ceramics by composite structure design, *Journal of Materials Chemistry C*, **7(26)**, 8120–8130 **(2019)**.

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