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Phyto-assisted Synthesis and Assessment of Anti-Inflammatory and Antioxidant Properties of Copper and Graphene Oxide Nano Composite Reinforced with Amla Extract-An In-Vitro Study

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ABSTRACT

Background: conventional methods used for synthesis of nanoparticles use toxic chemicals, thus leading to toxicity in the environment. Hence, we need to shift to "green synthesis". Previously done have shown excellent antimicrobial properties of the copper and graphene oxide nanocomposite reinforced with amla extract. Hence, this study was conducted to assess the anti-inflammatory and antioxidant properties of copper and graphene oxide nanocomposite reinforced with amla extract. Copper causes cell wall lysis by leakage of intracellular substances hence is an excellent antimicrobial agent, graphene oxide for its barrier and structural strength hence these were chosen to create a nanocomposite.

Aim: Aim of the study was phyto-assisted preparation of nano copper with nano graphene oxide nanocomposite and evaluation of its anti-inflammatory and antioxidant properties.

Material and methods: Anti-inflammatory and antioxidant properties of the nanocomposite were assessed using Bovine Serum Albumin (BSA) and DPPH Assay respectively at 10 µL, 20 µL, 30 µL, 40µL, 50 µL.

Results: Values for anti-inflammatory property of nanoparticles were higher than the standard values at 40µL, 50 µL concentrations. Percentage of inhibition was highest at 40 µL (86%) and 50 µL (84.6%). The values for antioxidant property of nanoparticles were found to be higher than the standard values at concentrations except at 40µL, 50 µL. Percentage of inhibition was highest at 20 µL (86.2%).

Conclusion: Within the limits of the study, it can be concluded that copper and graphene oxide nanocomposite has exceptional anti-inflammatory and antioxidant properties and further can be incorporated in dental material or can be used to coat suture materials to improve their properties.

Key words: Copper, Characterisation, Graphene oxide, Green synthesis, Nanoparticle, Nanocomposite, Anti-inflammatory, Antioxidant properties

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INTRODUCTION

Nanotechnology is an emerging technology and has led to a new revolution in every field of science. Nanoparticles have gained a lot of importance in the research community in recent years. This technology has been used in the fields of optics, electronics, and biomedical and materials sciences [1]. Potent antimicrobial, anticancer, antioxidant agents, drug, and gene delivery, etc are some of the highlighted advantages of the nanoparticles in the recent years [2–4]. Nanotechnology deals with nanoparticles that are atomic or molecular aggregates characterized by size less than 100 nm. These are basic elements derived by modifying their atomic and molecular properties [5,6].

Several conventional methods are used for synthesis of nanoparticles like chemical reduction [7], laser ablation [8], inert gas condensation [9,10], sol-gel method [11]. Even though less time is utilized for synthesizing large quantities of nanoparticles using conventional physical and chemical methods, toxic chemicals are required as capping agents to maintain stability, thus leading to toxicity in the environment. "Green synthesis" offers numerous benefits of eco friendliness and compatibility for biomedical applications, where toxic chemicals are not used for the synthesis protocol. The use of agricultural

wastes [9] or plants and their parts [10], has emerged as an alternative to chemical synthetic procedures because it does not require elaborate processes such as intracellular synthesis and multiple purification steps or the maintenance of microbial cell cultures.

Copper nanoparticles (CuNP) are superior owing to their nontoxicity, biocompatibility, use in drug and bactericidal activity [12,13]. Contact killing property of copper was studied widely in recent years. Studies have shown that increased bacterial intracellular oxidative stress in the bacterial cell wall due to release of ions from the copper surface which results in bacterial cell lysis [14]. Synthesis of copper nanoparticles is highly technique sensitive due to its high incidence of oxide layer formation on the nanoparticle surface which will result in reduced antibacterial property [15].

Graphene oxide is known for its excellent mechanical strength, electrical conductivity and most importantly the barrier properties, also easy step-down preparation of graphene oxide nanoparticles makes it one of the most efficient carriers of nanoparticles in any nanocomposite. It is an atomically-thin, 2-dimensional (2D) sheet of sp2 carbon atoms in a honeycomb structure [16].

The objective of this study was to use amla fruit extract to synthesize copper and graphene oxide nanoparticles and to evaluate its anti-inflammatory and antioxidant properties as its excellent potency against oral aerobes was already proven in the previous studies [16]. Also the nanocomposite showed minimal cytotoxic effects.

MATERIAL AND METHODS

Preparation of amla extract

Freshly collected organic amla fruits were thoroughly washed multiple times in distilled water. Seed was taken out and the pulp was cut into small pieces using a sterile knife and was grounded into small particles by means of a mortar and pestle. Amla extract was prepared by 1 grams of amla pulp with 100 ml distilled water to make 1 molar solution of amla extract [16].

Synthesis of copper and graphene oxide (Cu-GO) nanocomposite

Nanocomposite synthesis was done by mixing 50 ml of both 1M solutions of copper and graphene oxide nanoparticles as mentioned in the previous steps. The nanocomposite solution was stirred overnight on an orbital shaker followed by a magnetic heated stirrer till colour change was observed. UV-vis spectrometric readings were taken hourly to check the synthesis of copper-graphene oxide nano composite. The resultant mixture was centrifuged and CuGO nanocomposite was obtained [16].

Anti-inflammatory activity

Test group

10 µL, 20 µL, 30 µL, 40 µL and 50 µL of the nanocomposite solution was taken in 5 test tubes respectively.

To each test tube 2 ml of 1% Bovine Serum Albumin (BSA) was added. 390 µL, 380 µL, 370 µL, 360 µL and 350 µL of distilled water was added to the test tube containing 10 µL, 20 µL, 30 µL, 40 µL and 50 µL of nanoparticles respectively.

Control group

2 mL of Dimethyl Sulphoxide (DMSO) was added to 2 mL of BSA solution.

Standard group

10 µL, 20 µL, 30 µL, 40 µL and 50 µL of Diclofenac Sodium was taken in 5 test tubes respectively.

To each test tube 2 mL of 1% Bovine Serum Albumin (BSA) was added. The test tubes were incubated at room temperature for 10 minutes.

Then they were incubated in water bath at 55°C for around 10 minutes. Absorbance was measured at 660 nm in UV Spectrophotometer.

% Inhibition was calculated using the following formula:

% of inhibition=Control OD−Sample OD Sample OD100

RESULTS

Anti-inflammatory assay showed the following values at the end of the study:

Anti-inflammatory property of nanoparticles was higher than the standard values at 40µL, 50 µL concentrations.

Percentage of inhibition was highest at 40 µL (86%) and 50 µL (84.6%) (Figure 1).

Antioxidant test showed the following values for the nanocomposite:

The values for antioxidant property of nanoparticles were found to be higher than the standard values at concentrations except at 40µL, 50 µL. Percentage of inhibition was highest at 20 µL (86.2%)(Figure 2).

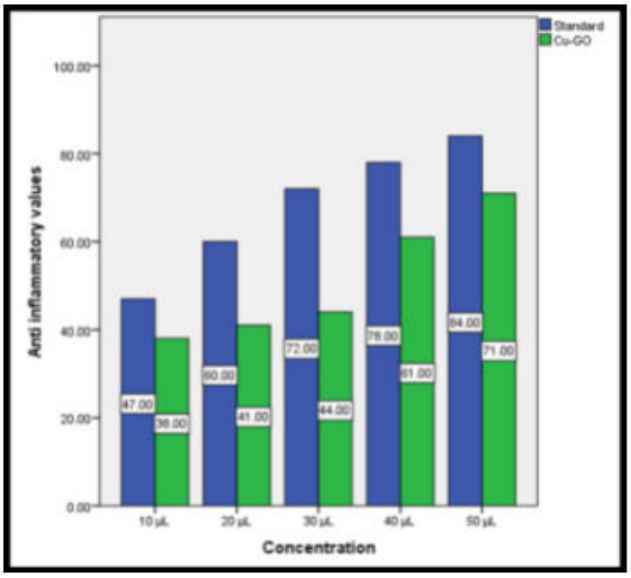

Figure 1: Bar diagram showing anti-inflammatory values at different concentrations for copper and graphene oxide nano composite. Where blue represents the control group and green represents the study group.

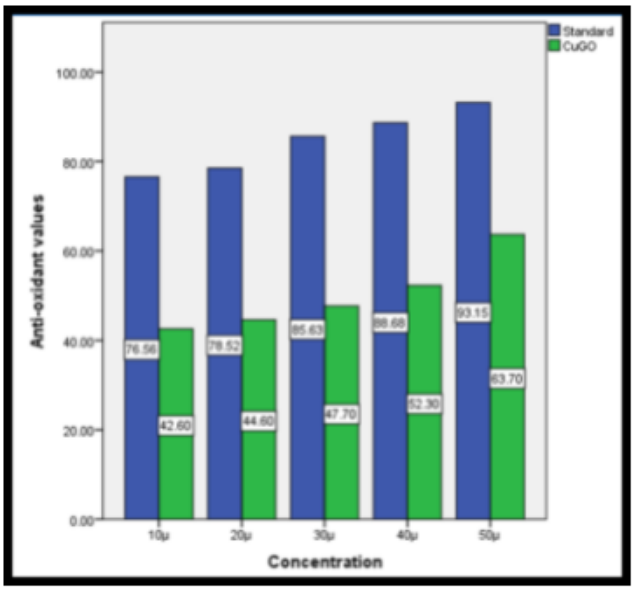

Figure 2: Bar diagram showing antioxidant values at different concentrations for copper and graphene oxide nano composite. Where blue represents the control group and green represents the study group.

DISCUSSION

Effective drug delivery systems with the ability to improve the therapeutic profile and efficacy of therapeutic agents is one of the key issues faced by modern medicine. Advances in nanoscience and nanotechnology, enabling the synthesis of new nanomaterials, have led to the development of several new drug delivery systems.

There has been a rapid evolution of nanoparticle synthesis recently as compared to the early part of the century [17]. Earlier conventional methods were used for the synthesis of the nanoparticles. Even though less time is utilized for synthesizing large quantities of nanoparticles using conventional physical and chemical methods, toxic chemicals are required as capping agents to maintain stability. These methods resulted in toxicity in the environment due to use of toxic chemicals. Green Synthesis method was proposed and is widely used all over the world to avoid the use of such toxic chemicals. It is an eco-friendly method as well as it is highly cost effective [18]. We therefore undertook this study to evaluate the cytotoxicity of the copper and graphene oxide nanocomposite reinforced with amla extract. Antibacterial properties of the same composite were found to be excellent against oral microbes in the previous studies [16].

Amla or Phyllanthus emblica is known since ancient times for its medicinal value and is commonly used in Ayurvedic medicine. Amla is a rich source of vitamin C which is a highly essential vitamin in maintenance of epithelial homeostasis. Superoxide radicals (O2–.) have been implicated in several pathological disorders and are responsible for elevated oxidative stress. Studies have shown that amla extract acts as a very good antioxidant by scavenging the reactive oxygen species and protects the antioxidant enzymes like SOD required for the cellular defence.

Copper has been known for its antibacterial activity. Copper nanoparticles impart the antioxidant effect by inhibition of chain reaction, decomposition of peroxides, binding of transition metal ion catalysts, radical scavenging activity and inhibition of continued hydrogen abstraction. The prop- erties of absorbing, neutralizing these free radicals or quenching singlet and triplet oxygen are few crucial factors that are responsible for the antioxidant activity. The highest antioxidant activity is attributed due to the presence of various bio-reductive groups of the phyto- chemicals present on the surface of the CuNPs. Copper not only is an excellent antioxidant but also an effective anti-inflammatory agent [12,19].

Remarkable physical–chemical properties, including a high Young's modulus, high fracture strength, excellent electrical and thermal conductivity, fast mobility of charge carriers, large specific surface area and biocompatibility are few of the major advantages of graphene nanoparticles. Graphene oxide is studied in the medicinal field for over 40 years for its potential for being the ideal drug delivery properties. Hence the combination of copper and graphene oxide with amla was selected to be tested for its anti-inflammatory and antioxidant property in the current study.

CONCLUSION

Within the limits of this study, it can be concluded that copper and graphene oxide nanocomposite has exceptional anti-inflammatory and antioxidant properties. With a possibility to be included in the dental materials to improve the properties.

CONFLICT OF INTEREST

The author declares that there are no conflicting interests.

REFERENCES

1. Rico CM, Majumdar S, Duarte-Gardea M, et al. Interaction of nanoparticles with edible plants and their possible implications in the food chain. J Agric Food Chem 2011; 59:3485–98.
2. Rajeshkumar S, Malarkodi C, Gnanajobitha G, et al. Seaweed-mediated synthesis of gold nanoparticles using Turbinaria conoides and its characterization. J Nanostructure Chem 2013; 3:44.
3. Malarkodi C, Rajeshkumar S, Paulkumar K, et al. Bactericidal activity of bio mediated silver nanoparticles synthesized by Serratia nematodiphila. Drug Invention Today 2013; 5:119-25.
4. Rajeshkumar S. Anticancer activity of eco-friendly gold nanoparticles against lung and liver cancer cells. J Genet Eng Biotechnol 2016; 14:195–202.
5. Daniel MC, Astruc D. Gold nanoparticles: Assembly, supramolecular chemistry, quantum-size-related properties, and applications toward biology, catalysis, and nanotechnology. Chem Rev 2004; 104:293–346.
6. Jamdagni P, Rana JS, Khatri P, et al. Comparative account of antifungal activity of green and chemically synthesized zinc oxide nanoparticles in combination with agricultural fungicides. J Nano Dimension 2018.
7. https://www.wiley.com/en-us/Materials+Science+and+Engineering%3A+An+Introduction%2C+10th+Edition-p-9781119405498
8. Snehal Yedurkar M, Kapil Punjabi D, Chandra Maurya B, et al. Biosynthesis of Zinc oxide nanoparticles using Euphorbia milii leaf extract-a green approach. Materials Today 2018; 5:22561–9.
9. Mohapatra S, Leelavathi L, Rajeshkumar S, et al. Assessment of cytotoxicity, anti-inflammatory and antioxidant activity of zinc oxide nanoparticles synthesized using clove and cinnamon formulation--an in-vitro study. J Evol Med Dent Sci 2020; 9:1859.
10. Chang H, Tsai MH. Synthesis and characterization of ZnO nanoparticles having prism shape by a novel gas condensation process. Rev Adv Mater Sci 2008; 18:734-43.
11. Li H, Wang J, Liu H, et al. Zinc oxide films prepared by sol–gel method. J Crystal Growth 2005; 275:e943–6.
12. Yusefi-Tanha E, Fallah S, Rostamnejadi A, et al. Particle size and concentration dependent toxicity of copper oxide nanoparticles (CuONPs) on seed yield and antioxidant defense system in soil grown soybean (Glycinemax cv. Kowsar). Sci Total Environ 2020; 715:136994.
13. Abdolhosseinzadeh M, Khodamoradi N. Synthesis & study of nano size copper oxide particle via chemical method. Adv Mater Res 2013; 829:187–91.
14. Abiodun-Solanke I, Ajayi D, Arigbede A. Nanotechnology and its application in dentistry. Ann Med Health Sci Res 2014; 4:S171–7.
15. Guidelli EJ, Ramos AP, Zaniquelli MED, et al. Green synthesis of colloidal silver nanoparticles using natural rubber latex extracted from Hevea brasiliensis. Spectrochim Acta A Mol Biomol Spectrosc 2011; 82:140–5.
16. Ketkar GN, Malaiappan S. Green preparation of nano copper (Cu) with nano graphene oxide (Go) nano composite characterization and antimicrobial activity against oral aerobic pathogens. Plant Cell Biotechnol Mol Biol 2020; 31-9.
17. Andersson M, Pedersen JS, Palmqvist AEC. Silver nanoparticle formation in microemulsions acting both as template and reducing agent. Langmuir 2005; 21:11387–96.
18. Chandran SP, Chaudhary M, Pasricha R, et al. Synthesis of gold nanotriangles and silver nanoparticles using Aloevera plant extract. Biotechnol Prog 2006; 22:577–83.
19. DeAlba-Montero I, Guajardo-Pacheco J, Morales-Sánchez E, et al. Antimicrobial properties of copper nanoparticles and amino acid chelated copper nanoparticles produced by using a soya extract. Bioinorg Chem Appl 2017; 2017:1064918.