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RESEARCH ARTICLE
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Antioxidant, Anti-Inflammatory And Antidiabetic Activity Of Oolong Tea Mediated Strontium Nanoparticles

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ABSTRACT

Background: Oolong tea is enriched with polyphenols which are natural compounds that are useful in the synthesis of nanoparticles. In addition to polyphenols, oolong tea is also blessed with caffeine and theanine, which has influence over the size and shape of the nanoparticles. Oolong tea is also a natural, sustainable and ecosystem friendly source.

Aim: To synthesize and study the antioxidant, anti- Inflammatory and anti diabetic activity of oolong tea mediated strontium nanoparticles.

Materials And Methods: Oolong tea extract was made and the strontium nanoparticles were synthesized by adding the extract to strontium chloride solution. 2,2-Diphenyl-1-picrylhydrazyl (DPPH) and Hydrogen peroxide (H2O2) assays were conducted to assess the antioxidant activity along with Egg albumin, bovine serum albumin assays were performed to analyze the anti-inflammatory activity. The anti- diabetic property was evaluated through alpha amylase inhibition

Results: In DPPH assay it was observed that the percentage of inhibition is approximately 65% (the lowest) in the concentration of 10µL and approximately 90%(the highest) in the concentration of 50µL. Similarly, in H2O2 assay it was observed that the percentage of inhibition is approximately 51% in the concentration of 10µL and approximately 77% with the concentration of 100µL. It was observed in both the DPPH and H2O2 assay that the percentage of inhibition increases as the concentration increases. The same was observed for EA and BSA assays and in the antidiabetic assay.

Conclusion: Oolong tea mediated strontium nanoparticles was evaluated to have strong antioxidant, anti- inflammatory and anti diabetic properties, which qualifies the material as a potential regenerative biomaterial for diabetes induced bone pathologies.

Keywords: *DPPH assay, H2O2 assay, EA assay, BSA assay*

INTRODUCTION

Polyphenols, which are organic substances that have been discovered to be useful in the creation of nanoparticles, can be found in oolong tea. Metal ions can be reduced by polyphenols to generate metal nanoparticles by binding to the metal ions. Particularly oolong tea has been found to possess significant quantities of polyphenols, making it an attractive source for the creation of nanoparticles. Along with polyphenols it also has other ingredients that can affect the properties of nanoparticles. For instance, it has been discovered that the caffeine and theanine in oolong tea affect the size and form of the nanoparticles. The fact that oolong tea is a natural, sustainable source and has an environmentally benign composition is another benefit of using it to synthesize nanoparticles.

Diabetes mellitus can be treated by antidiabetic activities that have power over the glucose levels in the blood, specifically Type 2 diabetic patient's plasma glucose levels can be effectively reduced by oolong tea. (1). Strontium nanoparticles are one of the nanoparticles that have strong anti diabetic activities because patients with diabetes have also benefited from the use of strontium nanoparticles as they can provide control of the release of insulin and so control the pathophysiology of diabetes.(2)Inhibiting glucose absorption and digestive enzymes are two ways that dietary polyphenols use insulin to prevent and treat Type 2 diabetes mellitus. (3)

Oolong teas contain a number of low molecular weight antioxidants that might have health promoting effects. The most prevalent antioxidants in the diet are polyphenols. They might consume up to 1 g of food daily in total, which is substantially more than all other classes of known dietary antioxidants and phytochemicals combined.(4) Oolong tea has a significant amount of total polyphenols and antioxidant properties. Herbs and herbal polyphenols are effective in regulating oxidation and preventing oxidation-related damage. (5)

Polyphenols are essential in the treatment and prevention of illnesses with underlying inflammatory disorders. However, more research needs to be done on how polyphenols affect various inflammatory cellular processes. One of the main ingredients in oolong tea, polyphenols, has been found to have anti-inflammatory properties. In particular, it has been discovered that polyphenols suppress the inflammatory cytokine production that plays a role in the progression of chronic inflammation, including TNF-alpha and IL-6. It has been demonstrated that caffeine present in oolong tea inhibits the activity of inflammatory cells and lowers inflammatory cytokine production.

This research is needed because the current medication has many side effects, careful monitoring of all patients is needed for the current medications as well as clinical prudence.

This study familiarizes herbal based treatments medicines for antioxidants, ant- inflammatory and anti diabetic activities of oolong tea. This study will give a method for antioxidants, anti-inflammatory and anti diabetic activity of oolong tea mediated with strontium nanoparticles. The aim of the study is to analyze the antioxidant, anti- inflammatory and antidiabetic activity of oolong tea mediated strontium nanoparticles. Our team has extensive knowledge and research experience that has translate into high quality publications(Sathivel et al. 2008; Sekar et al. 2019; Rajeshkumar et al. 2019; Lakshmi et al. 2015; Felicita et al. 2012; Thejeswar and Thenmozhi 2015; Saravanan et al. 2021; Menon and Thenmozhi 2016; Sahu et al. 2014; Wang et al. 2019)

MATERIALS AND METHODS

Biosynthesis of strontrium nanoparticles mediated by oolong tea

10.4g of oolong tea was measured and added with 100ml of distilled water. It was then kept on the mantle for 10-15 minutes to heat. While the oolong tea mixture was being heated the strontium solution was made by weighing 0.532g of strontium added with 70ml of distilled water.

After the oolong tea was heated it was filtered to separate the tea leafs to obtain the oolong tea extract. 30ml of this oolong tea extract was then mixed with the strontium solution. This new mixture was then covered with an aluminium foil and kept on the shaker for overnight for 24 hours to observe the colour change. This solution was thdm centrifuges for 10 minutes at 2000rpm, to conduct our DPPH, H2O2, BSA, EA and alpha amylase inhibition assays.

Assessment of anti-oxidant property of the SnNP

Dpph Assay

DPPH assay was used to test the antioxidant activity of biogenic synthesized strontium nanoparticles. Diverse concentrations (10µL,20µL,30µL,40µL,50µL) of Oolong tea leaf extract interceded strontium nanoparticle was mixed with 1 ml of 0.1 mM DPPH in methanol and 450 µl of 50 mM Tris HCl buffer (pH 7.4) and incubated for 30 minutes. Later, the reduction in the quantity of DPPH free radicals was assessed dependent on the absorbance at 517 nm. Ascorbic acid was used as standard. The percentage of inhibition was determined from the following equation,

% inhibition= Absorbance of control- Absorbance of test sample × 100

Absorbance of control

H2o2 Assay

All solutions were prepared freshly.1.0mL of the reaction mixture contained 100µL of 28mM of 2-deoxy-2-ribose (dissolved in phosphate buffer,pH 7.4), 500µL solution of various concentrations of the oolong tea (10µL,20µL,30µL,40µL,50µL) 200µL of 200µM Fecl3 and 1.04mM EDTA (1:1 v/v),100µL H2O2(1.0mM) and 100µL ascorbic acid(1.0mM).After an incubation period of 1 hour at 37°C the extent of deoxyribose degradation at about 532nm against the blank solution . Vitamin E was used as a positive control.

Assessment Of Anti-Inflammatory Property Of The Snnp

Egg Albumin Denaturation Assay

A 5ml solution was made with which consisted of 2.8ml of freshly prepared phosphate buffered saline of pH - 6.3, 0.2 ml of egg albumin extracted from hens egg. Specific concentrations were prepared separately for Oolong tea as (10µL,20µL,30µL,40µL,50µL). Diclofenac sodium was used as the positive control.. Then the mixtures were heated in water bath at 37ºC for 15 minutes. After which the samples were allowed to cool down to room temperature and absorption was measured at 660 nm.

Albumin Denaturation Assay

The anti-inflammatory activity for Oolong tea gel was tested by the following convention proposed by Muzushima and Kabayashi with specific alterations (Pratik Das et al.,2019). 0.05 mL of Oolong tea gel of various fixation (10µL,20µL,30µL,40µL,50µL)was added to 0.45 mL bovine serum albumin(1% aqueous solution) and the pH of the mixture was acclimated to 6.3 utilizing a modest quantity of 1N hydrochloric acid. These samples were incubated at room temperature for 20 min and then heated at 55 °C in a water bath for 30 min. The samples were cooled and the absorbance was estimated spectrophotometrically at 660 nm. Diclofenac Sodium was used as the standard. DMSO is utilized as a control.

Percentage of protein denaturation was determined utilizing following equation,

% inhibition= Absorbance of control- Absorbance of sample×100

Absorbance of control

RESULTS

Anti-inflammatory activity

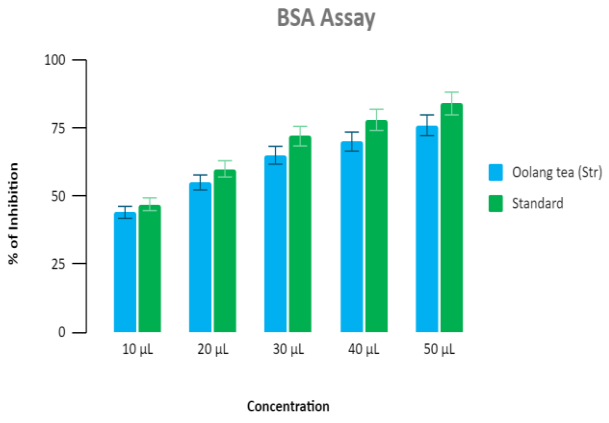

FIGURE 1: BSA assay

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The concentration was increased gradually, and the percentage of inhibition of oolong tea when mediated with strontium nanoparticles was read while comparing it with the commercially available antibiotics. The concentrations being 10µL, 20µL, 30µL, 40µL, 50µL and the percentage of inhibition being observed as approximately 48%, 62%, 70%, 73%, and 75% respectively. It was also noted that the commercially available antibiotics were slightly less effective then the oolong tea when it is mediated with strontium nanoparticles. The percentage of inhibition increases as the concentration increases in BSA assay.

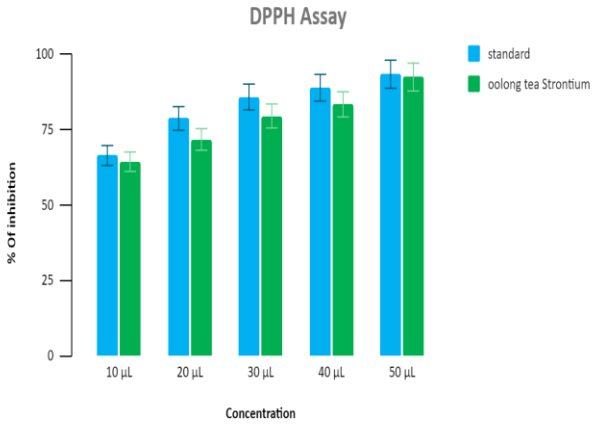

FIGURE 3: DPPH assay

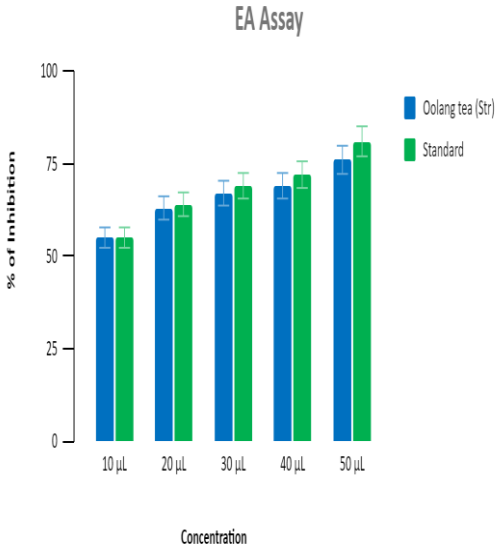

FIGURE 2: EA assay

Gradually raising the concentration, the percentage of oolong tea inhibition caused by strontium nanoparticles was measured and compared to that of commercially available antibiotics. The concentrations were 10µL, 20µL, 30µL, 40µL, and 50µL, respectively, and the percentage of inhibition was 52%, 55%, 65%, 73%, and 74%. Additionally, it was discovered that when used in conjunction with strontium nanoparticles, commercially available antibiotics were somewhat less effective than oolong tea. In the EA assay, the percentage of inhibition rises as the concentration does.

Antioxidant activity

The proportion of oolong tea inhibition brought on by strontium nanoparticles was calculated and contrasted to that of commercially available antibiotics while gradually increasing the concentration. The concentrations were 10µL, 20µL, 30µL, 40µL, and 50µL, with an observed inhibition percentage of around 55%, 60%, 74%, 76%, and 80%, respectively. Additionally, it was found that strontium nanoparticles mediated by oolong tea were just as effective as antibiotics now available on the market. The proportion of inhibition increases with concentration in the DPPH assay.

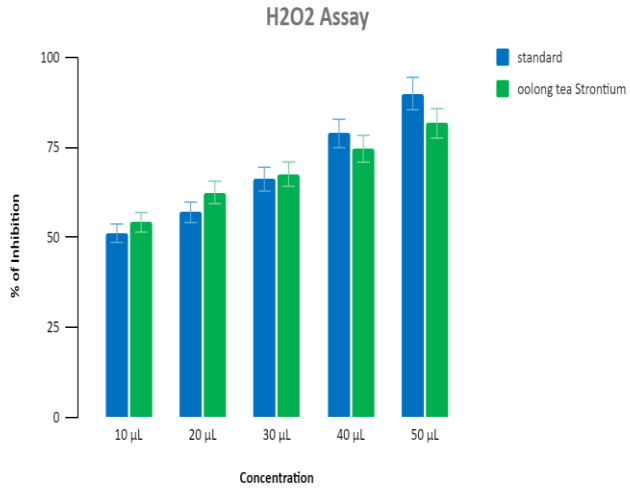

FIGURE 4: H2O2 assay

When comparing it to commercially available antibiotics, the concentration was gradually increased while reading the percentage of oolong

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tea inhibition when mediated with strontium nanoparticles. The concentrations were 10µL, 20µL, 30µL, 40µL, and 50µL, respectively, with an observed inhibition percentage of around 52%, 60%, 68%, 72%, and 77%. The commercially available antibiotics were also found to be less effective than oolong tea when they were mediated with strontium nanoparticles up to a concentration of 30 L; however, once this concentration was reached, it was found that the antibiotics were actually more effective than oolong tea's antioxidant activity. In general, when the concentration rises in the H2O2 assay, the percentage of inhibition rises.

Antidiabetic activity

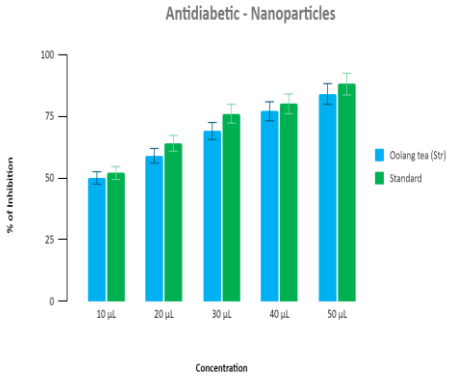

Figure 5(a)

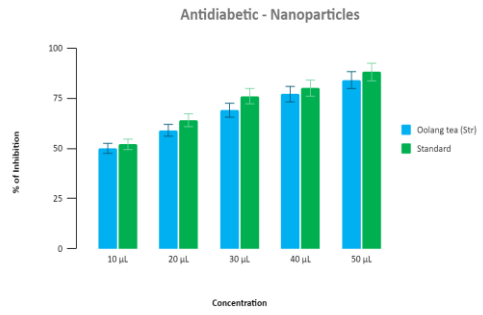

Figure 5(b)

FIGURE 5(A & B): alpha amylase inhibition assay

As the concentration was steadily increased, the percentage of oolong tea inhibition caused by strontium nanoparticles was estimated and compared to that of commercially available antibiotics. For the concentrations of 10µL, 20µL, 30µL, 40µL, and 50µL, the observed percentages of inhibition were 50%, 54%, 60%, 74%, and 78%, respectively. Additionally, it was shown that when strontium nanoparticles were used as a mediating agent, commercially available antibiotics were more effective than oolong tea. In the alpha amylase inhibition assay, the proportion of inhibition rises with concentration.

DISCUSSION

The term "green synthesis of nanoparticles" refers to the production of nanoparticles using sustainable and natural resources rather than hazardous or toxic chemicals. Green synthesis of nanoparticles is an eco-friendly process that uses sustainable and natural resources to create biocompatible nanoparticles that are suitable for use in biomedical applications. It is an economical method since it uses easily available natural resources, including plant extracts, and involves straightforward procedures that do not call for expensive equipment or specialized knowledge, making it open to a wider range of researchers. In overall, green synthesis of nanoparticles is a methodology that offers a number of advantages over traditional methods of nanoparticle synthesis, including being sustainable, biocompatible, affordable, scalable, and diversified.

In a previous study done on oolong tea ethanol extract by using MTS tetrazolium concluded that oolong tea ethanol extract had the potential to use as anti- inflammatory drugs. (6)Similarly, when oolong tea extract was mediated with strontium nanoparticles it showed strong anti-inflammatory activity. In a study done on the antioxidant activity of a polysaccharide isolated from oolong tea, they're results showed evidence of oolong tea polysaccharide being effective in antioxidant activities on hydroxyl radical scavenging assay and inhibition assay of lipid peroxidation (7)similarly, when oolong tea extract went through DPPH and H2O2 assay when being mediated with strontium nanoparticles it showed strong antioxidants activities when being compared with the commercially available antioxidants.

A study conducted on oolong tea concluded that it effectively inhibited the activity of inflammation when oolong tea was mediated with selenium nanoparticles (8)similarly, when oolong tea was mediated with strontium

nanoparticles in this study it showed strong anti-inflammatory activities.In a study done previously, oolong tea was fermented at a high degree, the results showed that the oolong tea produces higher contents of antioxidants and alpha- glucosidase inhibitory activities making them have good antidiabetic activity. (2) similarly, when oolong tea was mediated with strontium nanoparticles it showed good antidiabetic activity.

A previous study that was studied on various tea such as green tea, black tea, fermented, unfermented and semi fermented rooibos along with oolong tea compared their antioxidant activities. The results showed that oolong tea had the third highest antioxidant activity. (9)similarly, when oolong tea was mediated with strontium nanoparticles it showed strong antioxidant activity.

CONCLUSION

Only a little amount of research has been done on the possible advantages of oolong tea with strontium nanoparticles for oxidative stress, inflammation, and diabetes. Studies have been done on the individual impacts of strontium nanoparticles and oolong tea, though. We can conclude that the strong antioxidant, anti-inflammatory, and anti-diabetic activities of strontium nanoparticles mediated by oolong tea make the substance a viable regenerative biomaterial for diabetes-related bone diseases. The benefits and dangers of this combination need to be further investigated.

CONFLICT OF INTEREST

There is no conflict of interest

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