Green Synthesis of *Guilandina bonduc* Seed Extract Mediated Silver Nanoparticles and Its Anti-Inflammatory Activity

Mohammed Adil A.M[1], R. Pranav Vikram[2], S. Rajeshkumar[3], Pradeep manigandan[4]

[1] Department of Cardiology, Saveetha medical College and hospitals, Saveetha institute of medical and technical sciences, Saveetha University, Chennai, Tamil Nadu, India

[2] Department of Cardiology, Saveetha medical College and hospitals, Saveetha institute of medical and technical sciences, Saveetha University, Chennai, Tamil Nadu, India

[3] Nanobiomedicine Laboratory, Centre for Global Health Research, Saveetha Medical College and Hospitals,SIMATS, Saveetha Nagar, Thandalam, Chennai, Tamil Nadu, India

[4] Nanobiomedicine Laboratory, Centre for Global Health Research, Saveetha Medical College and Hospitals, SIMATS, Saveetha Nagar, Thandalam, Chennai, Tamil Nadu, India

Abstract: Green synthesis is a promising approach for the synthesis of silver nanoparticles (AgNPs) that is environmentally friendly and sustainable. In this study, we report a green method for the synthesis of AgNPs using *Guilandina bonduc* seed extract. The seed extract was used as a reducing and capping agent for the synthesis of AgNPs. The prepared nanoparticles were evaluated for its anti-inflammatory activity using Bovine serum albumin denaturation assay and Egg albumin denaturation assay. The AgNPs also showed significant anti-inflammatory activity in vitro. The results show that the green synthesised silver nanoparticles have excellent anti-inflammatory activity. The results of this study suggest that the green synthesised AgNPs have potential application in the treatment of inflammation. These findings also highlight the potential of *Guilandina bonduc* seed extract as a source of natural compounds for the green synthesis of AgNPs with biological activity.

Keywords: Green synthesis, *Guilandina bonduc*, seed extract, silver nanoparticles, anti-inflammatory.

1. INTRODUCTION

Nanotechnology is the new field of technological innovation on their application in human biology and medicine. Nanotechnology helps in the improvement of potentially revolutionary treatments for cancer, critical diseases and also drug improvements with reduction of side effects (Wanisa Abdussalam-Mohammed, W., 2019). The application of nanotechnology in the medical sector is referred to as Nanomedicine. Nanoparticles have potential applications in the field of medical sciences including new diagnostic tools, imaging agents and methods, targeted drug delivery, pharmaceuticals, bio implants and tissue engineering (Rifaath et al., 2023). The aim of nanotechnology in the medical sciences is to develop new materials and methods to detect and treat diseases in a targeted, precise, effective and lasting way, with the ultimate goal of making medical practice safer and less intrusive (Malik S et al., 2023).

Green nanoparticle synthesis has been achieved using environmentally acceptable plant extract and eco-friendly reducing and capping agents (Shanmugam R et al., 2023). Nanoparticles produced by plants are more stable and the rate of synthesis is faster than in the case of microorganisms. Moreover, the nanoparticles are more varied in shape and size in comparison with those produced by other organisms (Siavash Iravani 2011).

Nonresolving inflammation is a major driver of disease. Perpetuation of inflammation is an inherent risk because inflammation can damage tissue and necrosis can provoke inflammation (Ebenezer JL et al.,2022). The inflammatory response is particularly vulnerable to changes in relevant environmental factors, a few examples of which are changes in microbial exposure, diet, stress, toxins and physical activity (Rajeshkumar S et al., 2021). Chronic inflammatory

conditions such as systemic lupus erythematosus, inflammatory bowel disease, rheumatoid arthritis, ankylosing spondylitis, psoriatic arthritis, polymyositis/dermatomyositis, (I Roifman et al.,2011). An increase in lower gastrointestinal injury and clinical events with non-selective-NSAIDs appears relatively consistent across the heterogeneous collection of trials (Laine L et al., 2006). *Guilandina bonduc* from the Fabaceae family was used for improving wound, fever, tumor, hydrocele, hernia, smallpox, toothache, inflammation, and as astringent, anthelmintic, antidiabetic, and antimalarial agent in traditional medicine (Narges Pournaghi 2021). The seeds of the G. bonduc have the ability to cure fever and it has shown to possess tonic and antipyretic properties (Srinivasan P et al., 2023). The aim of the present study is to investigate the anti-inflammatory activity of *Guilandina bonduc* seed extract mediated silver nanoparticles using Bovine serum albumin denaturation assay and Egg albumin denaturation assay.

2. MATERIALS AND METHODS

2.1. PREPARATION OF SILVER NANOPARTICLES:

Guilandina bonduc seed extract was collected from Saveetha Herbal Garden and was shade dried and grinded to powder. 1g *Guilandina bonduc* seed powder was added to 100ml of distilled water in a beaker and kept for boiling in a heating mantle at 60°C to 70°C for 20 mins. The extract was then filtered using muslin cloth. 1 mM of silver nitrate was mixed with 80mL distilled water. 20 mL of filtered seed extract was added to the precursor solution and was kept in an orbital shaker at 110 RPM. The solution was then centrifuged at 8000 RPM for 10 minutes. After centrifugation, the pellet was collected and kept in the refrigerator for further use.

2.2. Anti-inflammatory activity

2.2.1. ALBUMIN DENATURATION ASSAY:

The anti-inflammatory activity for *Guilandina bonduc* seed extract mediated silver nanoparticles as tested by the following convention proposed by Muzushima and Kabayashi with specific alterations (Pratik Das et al.,2019). 0.05 mL of *Guilandina bonduc* seed extract mediated silver nanoparticles 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 utilising following equation,

$$\% \text{ inhibition} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100$$

2.2.2. EGG ALBUMIN DENATURATION ASSAY:

A 5ml solution was made 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 Guilandina bonduc seed extract mediated silver nanoparticles as (10µL,20µL,30µL,40µL,50µL). Diclofenac sodium was used as the positive control.. Then the mixtures were heated in a 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.

Percentage of protein denaturation was determined utilising following equation,

$$\% \text{ inhibition} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100$$

3. RESULT AND DISCUSSION

The figures 1-4 clearly shows the preparation of plant extract and silver nanoparticels synthesis and colour formation. The formation of dark brown colour indicates the synthesis of silver nanoparticles using green route (Dharman etal., 2023, Indumathy et al., 2023, Tharani et al., 2023). The figure 5 (a) and (b) shows the anti-inflammatory activity of silver nanoparticles synthesized using *G. bonduc.*

In a BSA assay, seed extract of *G. bonduc* mediated silver nanoparticles shows 80 % and 43 % inhibition at 50 µL and 10 µL concentration. In an EA assay, seed extract of *G. bonduc* shows 75 % and 52 % inhibition at 50 µL and 10 µL concentration. In BSA assay, *Guilandina bonduc* seed extract mediated silver nanoparticles shows slightly lower anti inflammatory activity when compared to standard (Diclofenac) which proves that the extract has anti inflammatory properties. Similarly, in EA assay, *Guilandina bonduc* seed extract mediated silver nanoparticles shows slightly lower anti-inflammatory activity when compared to standard (Diclofenac) which proves that the extract has anti-inflammatory properties.

Figure 1: Boiling of *Guilandina bonduc* seed extract using heating mantle.

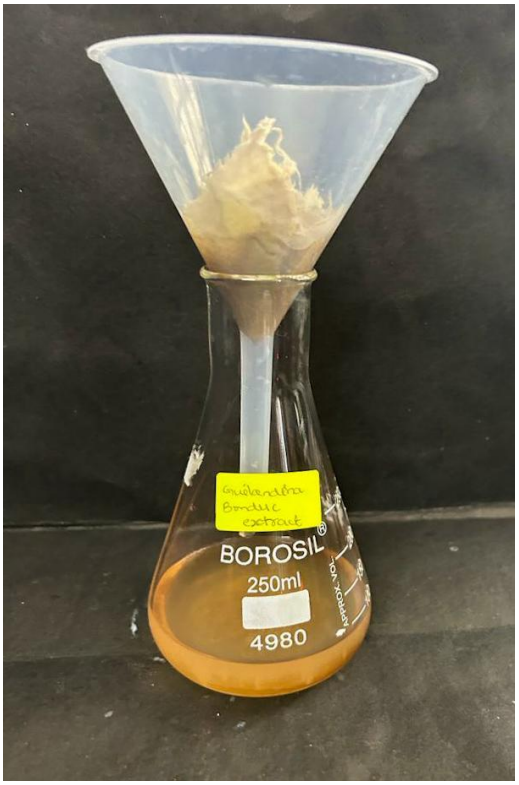


Figure 2: Filtering of *Guilandina bonduc* seed extract using funnel and filtering cloth

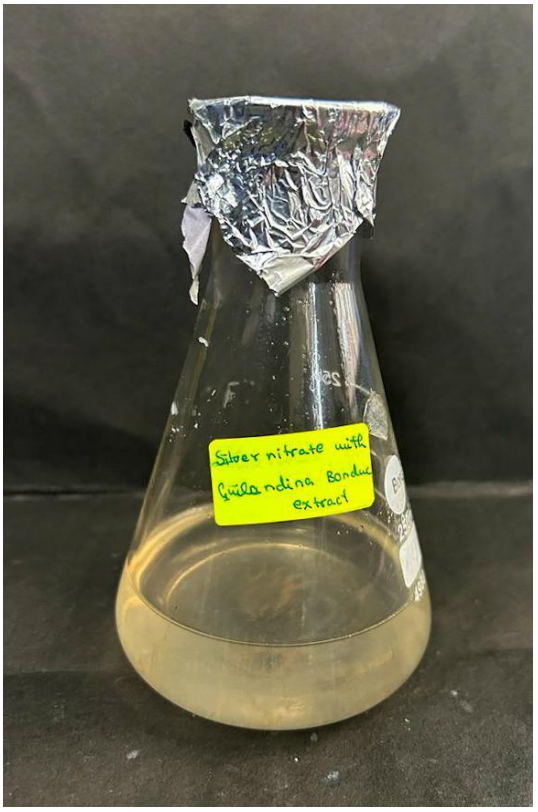


Figure 3: Silver nanoparticles synthesized using *Guilandina bonduc* stem extract.

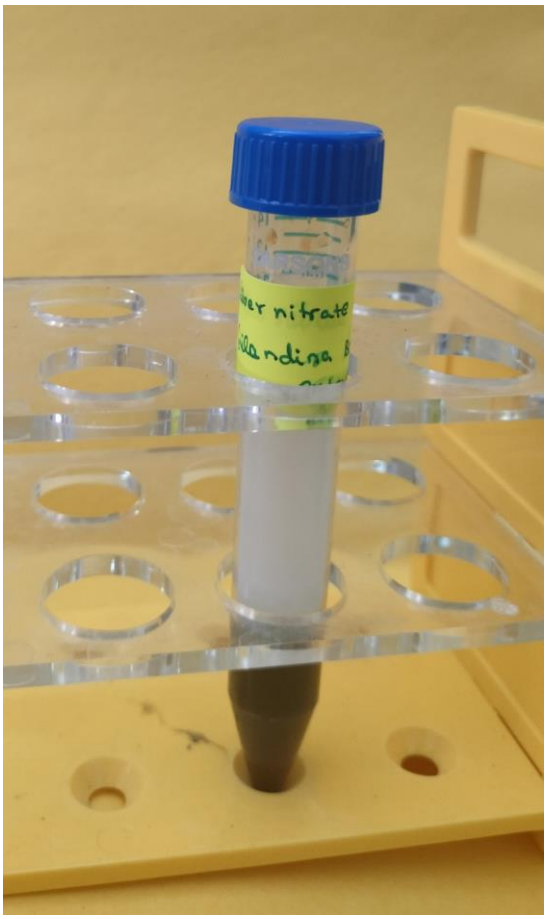
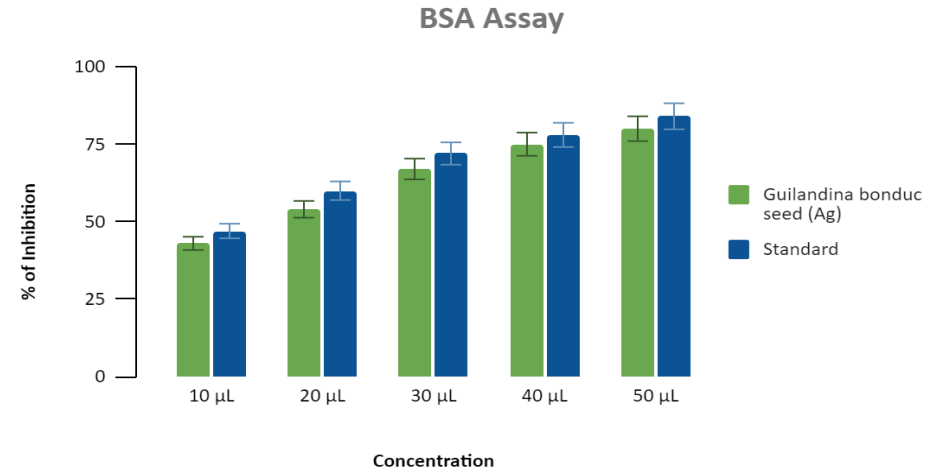
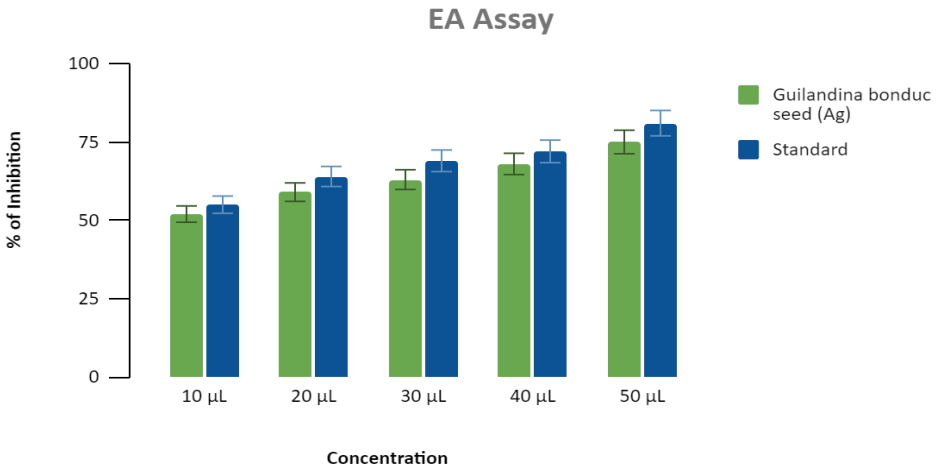


Figure 4: Collected silver nanoparticles after centrifugation



(a)



(b)

Figure 4: Antioxidant activity of the *G. bonduc* mediated silver nanoparticles (a) Bovine serum albumin denaturation assay (b) Egg albumin denaturation assay

Results of previous studies clearly indicate that the oil of *C. bonducella* seeds could be a potential source for use as anti-inflammatory, antipyretic and analgesic age (Shukla S, et al. 2010). The interesting anti-inflammatory potential of the new extracts and their nanoparticles could also be relevant to the development of new, effective anti-inflammatory agents (David L et al., 2014). Nanotechnology has shown healthcare improvements including potentially revolutionary treatments for cancer, critical diseases and also drug improvements with reduction of side-effects (Abdussalam-Mohammed, W., 2019).

4. CONCLUSION

The green synthesis of silver nanoparticles using *Guilandina bonduc* seed extract is a feasible and eco-friendly method. The study has several important implications. First, it demonstrates that green synthesis is a viable method for the production of silver nanoparticles. This is significant because green synthesis methods are more environmentally friendly than traditional methods, which often use toxic chemicals. Second, the study shows that silver nanoparticles synthesized using *Guilandina bonduc* seed extract have good anti-inflammatory activity. This suggests that the nanoparticles may be useful for the treatment of inflammatory diseases. Third, the study provides a foundation for further research on the anti-inflammatory properties of silver nanoparticles.The anti-inflammatory activity of the nanoparticles was only evaluated in vitro. Further studies are needed to investigate the potential of the nanoparticles as anti-inflammatory agents in vivo.

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