International Journal of Environmental Sciences
ISSN: 2229-7359
Vol. 11 No. 23s, 2025
https://www.theaspd.com/ijes.php

Anti Inflammatory Activity Of Silver Nanoparticles Synthesized Using Acai Berry Fruit Extract

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Abstract

Introduction: *There are numerous health advantages and medicinal uses for the palm Eutrepe oleracea Martius(açai).Nanoparticles is an excellent antibacterial agents used everywhere.Inflammation is a vast process with the pro inflammatory cytokines blocking lymphocyte molecule.*

Aim and Objective: *To synthesize Antiinflammatory activity of silver nanoparticles synthesized using Açai berry fruit extract.*

Methods and materials:*Invitro laboratory study.Materials used in conical flask,funnel,heater,filtering cloth,Açai berry powder and distilled water.*

Assay used to check the anti-inflammatory activity is Albumin denaturation/Bovine Serum Albumin (BSA Assay) and Egg Albumin Assay(EA) is used.Diclofenac Sodium is used as control and DMSO is used as standard.

Result: *Increasing the concentration shows increase in the percentage of inhibition of Açai berry fruit extract which is usually lower than the standards further indicates that the açai berry fruit extract that is being prepared have anti inflammatory effect in it.*

Conclusion: *Higher the concentration higher the production of extract which shows that the extract we synthesize is showing anti inflammatory.*

Keywords: *Egg albumin assay(EA),cytokines,lymphocytes,Euterpe oleracea Maritus,Bovine Serum Albumin Assay,Diclofenac Sodium,DMSO.*

INTRODUCTION:

There are numerous health advantages and medicinal uses for the palm Euterpe oleracea Martius (açai).In many regions of Brazil, açai has a long history of use as a staple meal and as a medicinal plant.Have relatively high content of polyphenolic,high anti oxidant,anti inflammatory,anti proliferative and cardio protective properties.Silver nanoparticles is an excellent anti bacterial ability ranging from home and disinfectant and to water purification.Anti inflammatory activity of AgNP potential clinical applications through postoperative peritoneal adhesion model.(1,2)

Traditional uses for Brazil palm include the treatment of fevers,skin issues,digestive problems and bacterial infections.Healht benefits that can typically only be substantially or to a very limited degree claimed.The extract of Açai berry is to present inflammation and oxidative stress(3) as a bio character for soil conditioner with anti adipogenic effects useful in preservation of hepatic steatosis and type 2 diabetes.

In this study,we investigated the anti inflammatory effect of silver nanoparticles from the Açai berry extract where this açai berry consist of dietary fibers and also decrease the environmental impact of industrial processing.Selection of these nanoparticles is a therapeutically active biomolecule that improves the biological applicability in our daily life.(4)).The metals is a nano particle had a huge impact in various range of field like optoelectronics,energy conversion and biomedical uses.This particle which is nano meter in single yielded very large amount with quantitative yield without the need of sorting and arranging it.It exhibited a outstanding ability of closed shell electron configuration with a large energy gap between the highest occupied molecular orbital and lowest unoccupied molecular orbital(5)

Inflammation is a vast process with pro inflammatory cytokines with it playing main role such as blocking lymphocyte costimulatory molecule or deplete B lymphocyte.Through a number of methods, inflammation serves as a physical reaction that defends against stress, illness, and injury. It is well recognised that the

oxidative stress that results from an inflammatory response can promote insulin resistance and diabetes,(6) raise the risk of atherosclerosis, induce coronary heart disease, cause lesions in the brain associated with Alzheimer's disease, and stimulate the growth of tumours.Inflammation is a major contributing factor to several disorders, hence treating the inflammatory condition may be a useful therapeutic strategy(Vikneshan et al.2020). The majority of clinically significant medications are either steroidal or nonsteroidal chemical anti-inflammatory.It helps in response to phagocytosis and intracellular killing of pathogen and the control of infections..Anti biotics play a major role to subside the inflammation and assist in repairing the particular tissue or organ.(7)
Our team has extensive knowledge and research experience that has translate into high quality publications (8–17)(18)
AIM: To synthesize the anti inflammatory activity of silver nanoparticles using açai berry fruit extract.

OBJECTIVES:
-Creating an anti inflammatory process for the extract that is being taken.
-New concept of adding silver nanoparticles to it which will produce further enhancement.
-To fulfill the stability of AgNPs in different exposure to media or environmental relevant conditions.
-To fulfill some interesting traditionally used plants that has not been investigated yet. Anti anti bacterial bacterial cells

MATERIALS AND METHODS:
Study was conducted in vitro laboratory.Sample is collected from Açai berry fruit extract.
Material- conical flask , funnel , heater , filtering cloth , acai berry powder, distilled water.
Procedure:

- Take Acai Berry powder is dissolved in a 100 ml of distilled water .
- Boil for 15-20 mins temp 60-70 degree celsius in a heating mantle.
- Keep the cotton cloth in an hot air oven for 10-25 minutes.
- Take 70ml of distilled water and add that to the silver nitrate crystals of (1 millimetre=0.0169 gm) in quantity .This is made to prepare the silver nanoparticles.
- Now add 30ml of Acaiberry fruit extract solution to the silver nitrate solution.
- Take this mixture of solution in a centrifuging tube of 6 in 14ml.Having that in 8000rpm for 10 mins will finally result in separating of pellate and supernatant.Centrifugation is done.
- UV Nanometer spectrum will be reading the Centrifugation.

Figure 1: Acaiberry fruit extract

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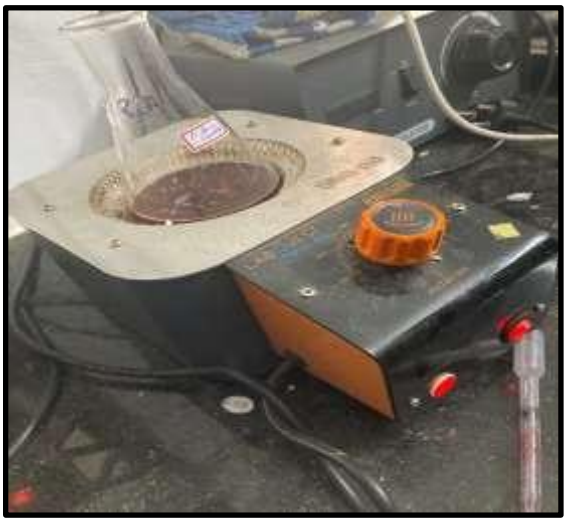

Figure 2:Boiling the extract

Figure 3:Prepared extract with silver nitrate crystals

Figure 4:Centrifugation

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Figure 5: Resulting Supernatant

Anti inflammatory activity:

ALBUMIN DENATURATION ASSAY:

The anti-inflammatory activity for Solanum torvum gel was tested by the following convention proposed by Muzushima and Kabayashi with specific alterations (Pratik Das et al.,2019). 0.05 mL of Solanum torvum 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.After cooling the samples,the absorbance at 660 nm was calculated spectrophotometrically. Diclofenac Sodium was used as the standard. DMSO is utilized as a control.

The following equation was used to calculate the percentage of protein denaturation,

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

EGG ALBUMIN DENATURATION ASSAY:

A 5ml solution was made which was comprised 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 Syzygium caryophyllatum 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.

Statistical analysis:

Utilization of SPSS Software(23.0)for appropriate statistical analysis and excel software.

ANOVA-All the obtained data will be statistically analysed.A p value less than 0.05 was considered as statistically significant.

RESULT:

BSA Assay:

Concentration (in micro litre)	Acaiberry(Ag) (% of inhibition)	Standard
10	40	47
20	53	60
30	65	72
40	71	78
50	77	84

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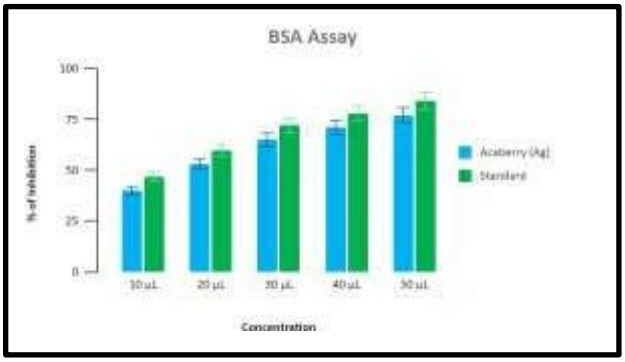
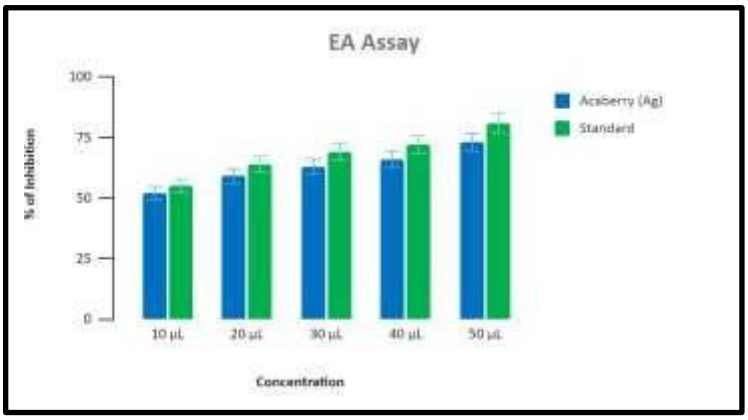


GRAPH- 1

Graph 1 represents the BSA Assay where x-axis represents concentration and y-axis represents the percentage of inhibition.On increasing the concentration of Acaiberry extract the percentage of inhibition of it gradually increases.Further stating that Açaiberry extract have inhibitory effect with it.

EA Assay:

Concentration (Micro litre)	Acaiberry(Ag)	Standard
10	52	55
20	59	64
30	63	69
40	66	72
50	73	81



GRAPH-2

Graph-2 represents the EA Assay where x-axis represents concentration and y-axis represents percentage of inhibition where on increasing the concentration percentage of inhibition increases gradually and standard is usually higher than the concentration i.e obtained.
Therefore comparing ,Ag concentration the standard there is not much difference in percentage of inhibition.Increasing of concentration causes increase in percentage of inhibition which tells that the extract we obtained have anti inflammatory activity within it.

DISCUSSION:

Graph 1 discusses that obtaining for 10 micro litre of concentration the Ag inhibition is 40% which is lesser than the standard i.e 47% .Then for 20 micro litre of concentration we obtained 53% of Ag compared to 60% of standard,for 30 micro litre of concentration we got 65% of Ag comparing to standard which is of 72%,For 40 micro litre of concentration we obtained 71% of Ag comparing to the 78% of standard.And finally for 50 micro litre of concentration,we obtained 77% of Ag concentration to 84% of standard.

Graph 2 discusses for 10 micro litre of concentration the Ag % of inhibition was 52% and standard is significantly increased for 55% likewise for 20 micro litre of concentration,it was 59% of Ag and 64% of standard,for 30 micro litre of concentration,it was 63% of Ag and 69% of standard,for 40 micro litre of concentration,it was 66% of Ag and 72% of standard and for 50 micro litre of concentration,it was 73% of Ag and 81% of standard(19).

Anti inflammatory activity of extract from the fruits where previous articles which stated with similar findings is the Anti inflammatory of fruits,herbs and spice,food chemistry.(20)Anti inflammatory effects of Phytochemical from fruits,vegetables and food legumes:Arevias,Critical review.(21)Anti inflammatory effect of plant based food and their constituents(22).These articles state that inflammation plays an important role in various diseases with high prevalence of disease such as rheumatoid arthritis,asthma and atherosclerosis.

Previous articles which stated with opposing finding is inhibition of inflammatory mediators by polyphenolic plant extracts in human CaCo2 cells(23).Plant derived polyphenolic attenuate lipopolysaccharide induced nitric oxide and tumor necrosis factor,which state that nitrogen activated protein kinases(MAPK) involved in transduction cascade that play a key role in inflammatory response((20,23).Lipopolysaccharide released in bacterial infection include expression of pro inflammatory cytokines and lead to neuronal damage in CNS and septic shock in periphery.

Silver nanoparticles functions.Biofunctionalized silver nanoparticles:advances and prospects,colloids and surfaces and silver nanoparticles in environment,Environmental source:Process and impact as a new generation of anti microbial,biotechnology advances stated that unique sized nanoparticles a promising field for biomedical and silver is remarkable in optical,electrical and anti microbial(5).The article which cited previous literature with opposing findings are green synthesis of silver nanoparticles:an approach to overcome toxicity,environmental toxicology and pharmacology,pharmaceutical aspects of silver nanoparticles,artificial cells,nanomedicine and biotechnology stated that they are expensive and can have toxicity and can have possible toxicological challenge(24).For a putative anti-inflammatory effect mechanism, the ability of the produced silver nanoparticles to block albumin denaturation has been studied. Protein denaturation has a well-established role in arthritic responses and the occurrence of tissue damage during inflammation. Results show that silver nanoparticles made through synthetic means were successful in preventing thermally induced albumin denaturation at all tested concentrations, demonstrating their capacity to prevent protein denaturation associated with inflammation(25).

Skin wound healing effects and action mechanism of açai berry water extracts((26–32)).It is with similar findings are optimisation of microwave and ultrasound extraction method of açai berries in term of highest content of phenolic compounds and a UHPLC method for rapid separation and quantification of anthocyanin in açai berry,stated that traditionally açai berry is a natural remedy for a number of disorders like influenza,cold and inflammatory activities(33).

Açai berry as a source of bio active phytochemicals(34)) and enhancing thermal stability and biocompatibility of açai fruit polyphenolic through electrohydrodynamic encapsulation into electroprayed particles stated that Açai berry contains high amount of mono saturated fats and dietary fiber.Açai berry may contribute(35) to weight loss by increasing metabolism,through there are no substantial studies to support this.

FUTURE SCOPE:

To produce the Açai berry extract with further anti oxidant,anti microbial and with cytotoxic effect.So that the product being produced will be enhanced in future view.

CONCLUSION:

Higher the concentration higher the production of extract which shows that the extract we synthesize is showing anti inflammatory property.

LIMITATIONS:

- It haven't fulfilled the stability of AgNPs in different exposure media or environmental relevant conditions and to predict health effect in humans.
- It haven't fulfill some interesting traditionally used medicinal plants which was not investigated yet.
- Silver nanoparticles will expose to environment in different dimensions.

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