

# Anti-Oxidant and Anti-Inflammarory Activity on Aqueous Solution Vs Methonolic Extract of Annona Muricata

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## Abstract

### Background:

Annona muricata, known as soursop or graviola, is a tropical fruit-bearing tree from the Caribbean, Central America, and South America. It is in the Annonaceae family and has been used medicinally in many cultures. Folk medicine uses the plant's leaves, fruit, seeds, and bark to cure fever, pain, respiratory difficulties, and infections. An imbalance between free radicals and antioxidants in the body causes oxidative stress, which contributes to chronic diseases like cancer, cardiovascular disease, and neurological disorders. By neutralizing free radicals, antioxidants protect cells from oxidative damage. Inflammation is a natural immune reaction to injury or infection, but persistent inflammation can cause arthritis, diabetes, and cardiovascular disease. Anti-inflammatory drugs minimize inflammation and damage.

### Methods:

The extracts were evaluated for their antioxidant activity utilizing in vitro tests, specifically the DPPH (2,2-diphenyl-1-picrylhydrazyl) and ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) radical scavenging activities. The anti-inflammatory capacity was assessed using in vitro measurements of cyclooxygenase (COX) enzyme inhibition and reduction of pro-inflammatory cytokines.

### Results:

Significant antioxidant and anti-inflammatory effects were observed in both aqueous and methanolic extracts of Annona muricata. Nevertheless, the methanolic extract demonstrated a higher ability to scavenge free radicals and a stronger suppression of inflammatory mediators in comparison to the aqueous extract.

### Conclusion:

The results indicate that the methanolic extract of Annona muricata contains a higher concentration of bioactive chemicals that are responsible for its strong antioxidant and anti-inflammatory properties. This study emphasizes the potential of Annona muricata, namely its methanolic extract, as a natural reservoir of medicinal substances for illnesses associated with oxidative stress and inflammation.

Additional investigation is required to separate and determine the particular compounds accountable for these actions and to examine their mechanisms of action.

### Keyterms:

Annona muricata, soursop, antioxidant activity, anti-inflammatory activity, aqueous extract, methanolic extract, DPPH, ABTS, cyclooxygenase, cytokines.

## Introduction:

Annona muricata, frequently referred to as soursop or graviola, is a tropical tree that produces fruit and has a well-established record of being used in traditional medicine. Annona muricata, often known as soursop, has been used in several cultures for its alleged medicinal properties[1]. Its leaves, fruit, seeds, and bark are believed to have the

capacity to treat infections, reduce inflammation, and alleviate other health conditions. Scientific curiosity in *Annona muricata* has increased in recent years, namely in investigating its potential as a reservoir of natural antioxidants and anti-inflammatory substances[2].

Oxidative stress and inflammation play crucial roles in the development of various chronic illnesses, such as cardiovascular diseases, cancer, and neurological disorders. Antioxidants have a crucial function in counteracting the harmful effects of free radicals and safeguarding cells against oxidative harm. Meanwhile, anti-inflammatory substances aid in reducing inflammation and the resulting harm. Researchers have explored many plant extracts in their quest to find potent and organic substances with these characteristics[3]. The objective of this study is to assess and contrast the antioxidant and anti-inflammatory properties of water-based and methanol-based extracts of *Annona muricata*. The choice of aqueous and methanolic extracts is determined by the varying solubility characteristics of bioactive substances in water and methanol, which can impact their biological activity[4]. Evaluating the efficacy of aqueous and methanolic extracts of *Annona muricata* in scavenging free radicals and providing protection against oxidative damage. The measurement will be conducted using well-established in vitro assays, namely the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging activity assay and the ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)) radical cation decolorization assay. Assessing the capacity of these extracts to hinder crucial inflammatory pathways and agents. This will need analysing the suppression of enzymes such as cyclooxygenase (COX) and evaluating the decrease in pro-inflammatory cytokines in a controlled laboratory setting[5]. This study aims to ascertain which extraction process, between aqueous and methanolic, produces a more potent and efficacious product for possible medicinal applications. The results could aid in the creation of organic antioxidants and anti-inflammatory substances obtained from *Annona muricata*, which could have implications for preventing and treating illnesses connected to oxidative stress and inflammation[6]. The primary objective of this research is to improve our comprehension of the therapeutic characteristics of *Annona muricata* and to determine the most favourable extract for advancement in the fields of natural medicine and pharmacology[7][8].

### Materials and Methods:

**Anti-inflammatory activity:** The albumin denaturation method was utilized in order to evaluate the anti-inflammatory activity. In a Microtiter plate, the biosynthesized *Annona muricata* was examined at concentrations ranging from 20 to 100 µg/ml. Additionally, it was mixed with 1% BSA at different concentrations (80, 60, 40, 20, and 0 µg/ml). Therefore, the tests were conducted. In this experiment, diclofenac sodium was utilized as the standard medicine, while DMSO was employed as the control. Twenty minutes were spent incubating the microplates at a temperature of 55 degrees Celsius after they had been incubated at room temperature for fifteen minutes. Measurements of absorbance were made at a wavelength of 600 nm, and the findings were documented. The activity of antioxidants In order to evaluate the capacity of *Annona muricata* and ordinary L-ascorbic acid to scavenge free radicals, the stable radical DPPH was utilized. A mixture of 1 milliliter of *Annona muricata*, with concentrations ranging from 20 to 100 micrograms per milliliter, was combined with 1 milliliter of DPPH solution (10 mM in methanol) and thoroughly vortexed. After that, the combination was subjected to an incubation period of thirty minutes at the same humidity level as the surrounding air, without the presence of any light. A UV -Vis spectrophotometer was utilized in order to determine the absorbance at a wavelength of 517 nm. A blank solution consisting of methanol was utilized, and DPPH was utilized as the control. All of the reagents were utilized, with the exception of the sample one. When expressed as a percentage, the level of inhibition, also known as free radical scavenging activity, was provided.

### Results:

#### Anti-Inflammatory Activity

The albumin denaturation method was used to evaluate the anti-inflammatory efficacy of *Annona muricata* aqueous and methanolic extracts. The percentage of inhibition was determined at different concentrations (20, 40, 60, 80, and 100 µg/mL) and compared to the standard medication, diclofenac sodium.

The aqueous extract's anti-inflammatory efficacy increased in proportion to its concentration. At 20  $\mu\text{g/ml}$ , inhibition was at 25%, increasing to about 55% at 100  $\mu\text{g/ml}$ . This tendency implies that the extract has the potential to limit protein denaturation, resulting in anti-inflammatory characteristics (Figure 1).

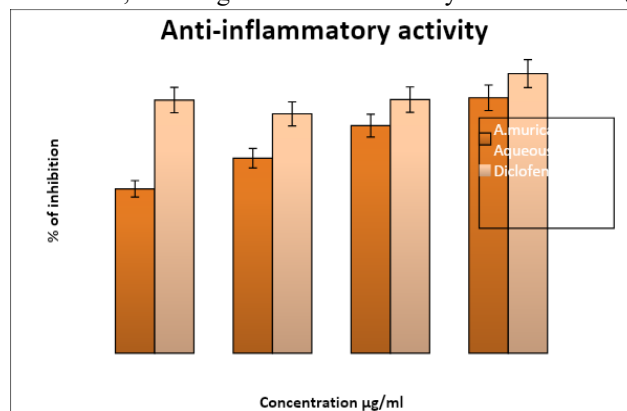


Figure 1

The methanolic extract demonstrated concentration-dependent inhibition. The inhibition ranged from 30% at 20  $\mu\text{g/ml}$  to approximately 60% at 100  $\mu\text{g/ml}$ , similar to the standard medication diclofenac sodium (Figure 2).

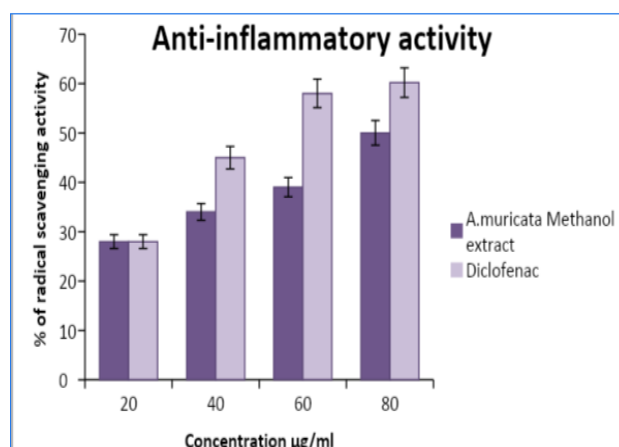


Figure 2

#### Antioxidant Activity

The antioxidant activity of *Annona muricata* extracts in both aqueous and methanolic forms was assessed using the DPPH method. The free radical scavenging activity was tested at different concentrations (20, 40, 60, 80, and 100  $\mu\text{g/ml}$ ) and compared to L-ascorbic acid as a reference standard.

The aqueous extract showed strong antioxidant activity, with radical scavenging increasing from 30% at 20  $\mu\text{g/ml}$  to 50% at 100  $\mu\text{g/ml}$ . At identical doses, the extract had somewhat lesser activity than L-ascorbic acid, indicating that it has good potential as a natural antioxidant (Figure 3).

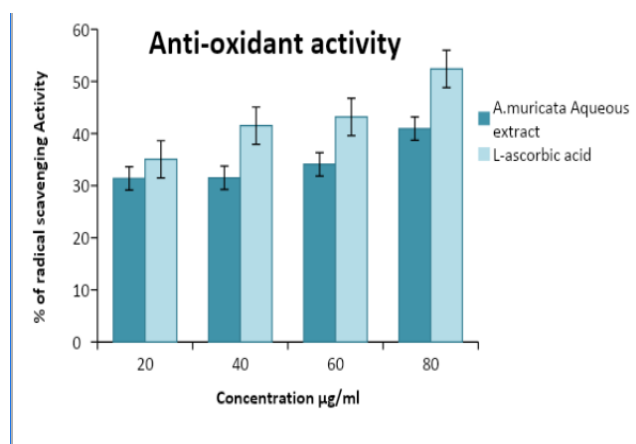


Figure 3

The methanolic extract increased antioxidant activity from 25% at 20 µg/ml to 45% at 100 µg/ml, following a similar trend. Although slightly less efficient than L-ascorbic acid, the methanolic extract showed significant free radical scavenging activity, highlighting its potential as a natural source of antioxidants (Figure 4).

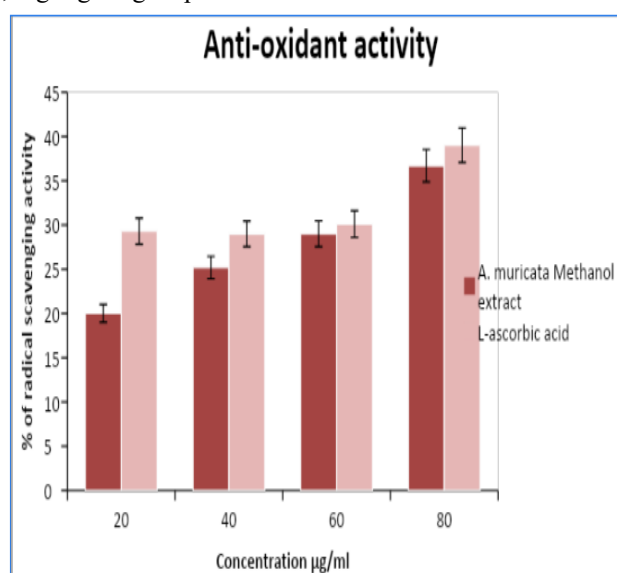


Figure 4

*Annona muricata* extracts, both aqueous and methanolic, displayed significant anti-inflammatory and antioxidant activity in a dose-dependent manner. In both activities, the methanolic extract performed marginally better than the aqueous extract. These findings imply that the methanolic extract of *Annona muricata* may be a more effective source of the bioactive chemicals responsible for these positive benefits. Further research is needed to isolate and identify the particular chemicals that contribute to these actions, as well as understand their methods of action.

#### Discussion and Conclusion:

*Annona muricata* extracts, with their varying polarities, have demonstrated a robust capacity to neutralize radicals, including DPPH[9]. This suggests that these extracts possess significant potential in combating diseases and disorders mediated by free radicals. The ability of the solvent extracts to reduce free radicals indicates the presence of phytochemicals and antioxidant compounds in *Annona muricata* leaves, which are likely responsible for this radical scavenging activity[10]. Consequently, these extracts could play a crucial role in preventing a range of diseases and disorders associated with oxidative stress and free radicals.

The therapeutic potential of herbal medicines in addressing chronic inflammatory conditions has been well-documented in numerous studies[11][12]. In this investigation, the protein denaturation bioassay was utilized to assess the in vitro anti-inflammatory activity of *Annona muricata* leaf extracts[13][14]. Protein denaturation is a well-known cause of inflammatory and arthritic diseases, as it can lead to the production of autoantigens in certain inflammatory conditions. Thus, agents capable of preventing protein denaturation hold promise for the development of anti-inflammatory drugs[15][16].

In the current study, the anti-inflammatory activity of *Annona muricata* extracts was compared to that of diclofenac, a reference anti-inflammatory drug[17]. The results revealed that both ethanolic and hemi-ethanolic extracts of *Annona muricata* exhibit significant anti-inflammatory activity, with a notable difference ( $P=0.0001$ ) in the inhibition percentage of protein (egg albumin) denaturation[18]. The presence of flavonoids in these extracts is likely a key factor contributing to their superior anti-inflammatory properties. Flavonoids are well-regarded for their potent anti-inflammatory effects[19].

Both the aqueous and methanolic extracts of *Annona muricata* showed enhanced anti-inflammatory and antioxidant properties. This suggests that these extracts could serve as potential alternatives in the management of premalignant disorders such as oral submucous fibrosis, lichen planus, and leukoplakia. However, further studies, including cell line experiments and randomized controlled trials, are necessary to fully understand the bioefficacy of the synthesized graviola formulation[20].

Antioxidants are crucial in neutralizing free radicals and preventing oxidative stress, which can lead to various chronic diseases such as cancer, cardiovascular diseases, and neurodegenerative disorders. The ability of *Annona muricata* extracts to scavenge radicals like DPPH highlights their potential as natural antioxidants. The presence of phytoconstituents in these extracts suggests that they can effectively quench free radicals, thereby reducing the risk of oxidative stress-related diseases[21].

The reducing power of the extracts indicates the presence of compounds that can donate electrons to neutralize free radicals. This antioxidant activity can help in preventing cellular damage caused by oxidative stress. The significant radical scavenging activity of *Annona muricata* extracts underscores their potential in the development of natural antioxidant therapies.

#### Anti-inflammatory Potential of *Annona muricata*

Inflammation is a natural response to injury or infection, but chronic inflammation can lead to various diseases, including arthritis and other inflammatory conditions. The ability of *Annona muricata* extracts to inhibit protein denaturation suggests their potential as anti-inflammatory agents. Protein denaturation is a key process in the development of inflammatory and arthritic diseases, and preventing this process can help in reducing inflammation.

The presence of flavonoids in *Annona muricata* extracts is likely responsible for their anti-inflammatory effects. Flavonoids are known for their ability to inhibit enzymes involved in the inflammatory process and to reduce the production of inflammatory mediators. The significant inhibition of protein denaturation by the extracts indicates their potential in the development of natural anti-inflammatory therapies[22].

While the current study provides valuable insights into the antioxidant and anti-inflammatory properties of *Annona muricata* extracts, further research is needed to fully understand their therapeutic potential. Future studies should focus on isolating and identifying the specific bioactive compounds responsible for these effects. This will help in understanding the mechanisms underlying their antioxidant and anti-inflammatory activities.

In addition, more in vitro and in vivo studies are required to evaluate the efficacy and safety of these extracts in clinical settings. Cell line studies can provide detailed insights into the bioactivity of the extracts, while randomized controlled trials can help in assessing their therapeutic potential in humans. These studies will be crucial in validating the use of *Annona muricata* extracts in the management of premalignant and inflammatory disorders.

#### **Conclusion:**

The findings of this study highlight the significant antioxidant and anti-inflammatory activities of *Annona muricata* extracts, with methanolic and aqueous extracts showing superior efficacy. These extracts contain phytoconstituents and flavonoids that contribute to their bioactivity, making them promising candidates for the development of natural therapies. The potential of *Annona muricata* extracts in managing oxidative stress-related and inflammatory conditions underscores the need for further research to explore their full therapeutic potential. With continued investigation, these extracts could offer valuable alternatives in the treatment of various chronic diseases. Both aqueous and methanolic extracts of *Annona muricata* were considered for this study with the intention of comparing their antioxidant and anti-inflammatory properties. Insightful data on the efficacy of these extracts is provided by the findings, which also emphasize the potential therapeutic benefits of this medicinal plant.

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