

“Aavarampoo’s (*Cassia auriculata*) Therapeutic Potential: Revealing the Secrets of Nature’s Anti-Inflammatory Elixir through Membrane Stabilization and Protein Denaturation”

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Abstract

Aavarampoo, scientifically known as Cassia auriculata, is a flowering plant belonging to the legume family, used in Ayurvedic medicine. Studies have shown that the phytochemical constituents of Aavarampoo, including flavonoids and tannins, contribute to its ability to modulate inflammatory responses in the body. Diclofenac is a well-known nonsteroidal anti-inflammatory drug (NSAID) it works by inhibiting cyclooxygenase (COX) enzymes, which play a key role in the inflammatory process. This makes it a suitable comparator when assessing the anti-inflammatory effects of Aavarampoo, which may have a different but potentially complementary mechanism. Given its widespread use in clinical practice, comparing Aavarampoo to diclofenac can provide relevant insights into its potential as a viable alternative or adjunct therapy in treating inflammation. Materials And Methods: This study is to explore Aavarampoo’s anti-inflammatory potential against Diclofenac by membrane stabilizing property (Maria dragan et al, 2016) and by protein denaturation method. Results: When assessed for membrane stabilizing activity, Aavarampoo exhibited commendable protective effects on human red blood cells (HRBC), demonstrating a maximum inhibition of 50.0% at a concentration of 500 µg. diclofenac exhibited significant inhibition of albumin denaturation, with a maximum percentage inhibition of 87.5% at 500 µg. In contrast, the Aavarampoo extract showed a progressive increase in inhibition, achieving 50.0% at the highest concentration tested. Conclusion: Aavarampoo possesses notable anti-inflammatory properties; further research could enhance our understanding of its mechanisms and broaden its applications in complementary health strategies.

Keywords: Anti-inflammatory Activity of Aavarampoo Vs Diclofenac, Albumin Denaturation Activity, Membrane Stabilizing Activity.

Introduction

Integrating Ayurveda with Allopathy can offer several advantages, Ayurveda focuses on treating the whole person—body, mind, and spirit—rather than just symptoms. This holistic perspective can complement allopathic treatments, which often target specific ailments. Both system of medicine emphasizes lifestyle changes, dietary modifications, and preventive care, which can enhance overall

health and reduce the incidence of diseases, thereby supporting treatments.

Ayurvedic principles consider individual constitution (Prakriti) and imbalances, leading to more tailored treatment plans that may improve outcomes in conjunction with allopathic approaches, which is Evidence-Based that is it relies on scientific research, clinical trials, and evidence to develop treatment protocols. Ayurvedic therapies often involve natural remedies, whereas allopathy

often aims to alleviate symptoms and manage diseases through treatments that are standardized and regulated, ensuring consistent dosages and protocols. It excels in emergency care and acute conditions, such as infections and trauma.

In general, Combining Ayurvedic practices with allopathic medicine can lead to innovative research and the development of new therapies, enhancing the medical field as a whole. Avarampoo (also known as *Cassia auriculata*) is widely used in Ayurveda for various diseases, it's been explored as antioxidant, and this study aims to bring to light the anti-inflammatory property. Few studies delve into the same, with focus on its membrane stabilizing and protein denaturation properties [1, 2]. These properties are often employed as key mechanisms to evaluate the plant's potential in managing inflammatory conditions [3]. This study authenticates further the anti-inflammatory property of Avarampoo extract (cold extraction) by comparing it with Diclofenac as standard.

Materials and Methods

The Avarampoo is extracted by Cold Extraction Method

10 gm of powder sample was weighed and soaked in 100 ml of ethanol. The extract was allowed to stand 72h and filtered using sterile filter paper. The filtrate was collected and incubated at room temperature for evaporation. Then measure the weight and find the yield by calculating.

Yield= initial weight - final weight.

Yield obtained = 0.56g

The cold extraction process yielded a noteworthy 0.56 g, indicating an efficient extraction of bioactive compounds.

Anti-Inflammatory Activities of Avarampoo (*Cassia Auriculata*)

Anti-Inflammatory by Membrane Stabilizing Property (Maria Dragan et al, 2016) [4]

The collected blood (1ml) was mixed with equal volume of Alsever solution (2 % dextrose, 0.8 % sodium citrate, 0.5 % citric acid and 0.42 % NaCl) and centrifuged at 3,000 rpm for 5 min. Discard the supernatant and collect the lower layer (RBC). RBC's were washed with isosaline (0.85% NaCl) (centrifuge 3000 rpm for 5 min) and collect 1 ml of RBC's and add 9ml of isosaline. The obtained solution is HRBC (hemoglobin RBC). Various concentrations of samples were taken in test tubes and made upto 1 ml using distilled water and to each tube, 1 ml of phosphate buffer, 2 ml hypo saline (0.36% NaCl) and 0.5 ml of HRBC suspension were added. Incubated at 37°C for 30 minutes, centrifuged at 3,000 rpm for 20 minutes. Absorbance of supernatant was read at 560 nm. Diclofenac (1mg/ml) was used as standard and a control was prepared without sample. The percentage (%) of HRBC protection was calculated using the following formula,

Percentage of Protection (%) = $100 - [(OD \text{ of sample} / OD \text{ of Control}) \times 100]$

In Vitro Anti-Inflammatory Activity by Albumin Denaturation Method (Raghavendra, et al, 2016) [5]

Different concentration of samples were taken in all the test tubes and made upto 1 ml using phosphate buffer saline (pH 6.4). Add 4ml of egg albumin to all the tube including control tube. Then the mixture were incubated at 37°C for 15 min and then heated at 72°C for 5 min. After incubation samples and control were allowed to cool for 10 min and the absorbance was measured at 660 nm. Diclofenac (1mg/ml) was used as standard and control was prepared without sample. The percentage inhibition of protein denaturation

(%) was calculated by using the following formula,

$$\text{Percentage of Inhibition (\%)} = \frac{(A \text{ control} - A \text{ sample})}{A \text{ control}} \times 100$$

Results

Anti-Inflammatory by membrane stabilizing property

Table 1 and 2 and Figure 1 shows the anti-inflammatory property of Avarampoo by membrane stabilizing activity.

Table 1. Membrane Stabilizing Activity of Diclofenac (Standard Blank= 0.46)

Sample Concentration (µg)	100	200	300	400	500
Diclofenac	0.33	0.28	0.24	0.19	0.16
% inhibition	28.3	39.2	47.9	58.7	65.3

Table 2. Membrane Stabilizing Activity of Avarampoo (Sample Blank= 0.46)

Sample Concentration (µg)	100	200	300	400	500
Avarampoo	0.29	0.26	0.22	0.20	0.18
% inhibition	19.5	27.8	38.9	44.5	50

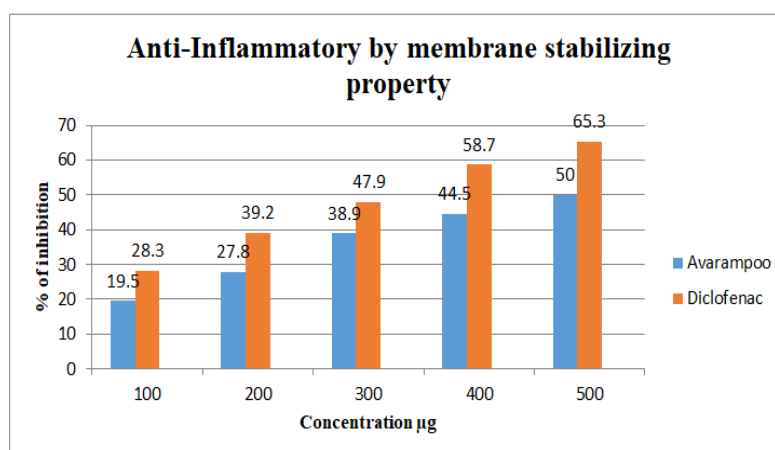


Figure 1. Anti-inflammatory Activity of Avarampoo by Membrane Stabilizing Activity at Different Concentrations

This study was based on “Maria Dragan et al. Anti-inflammatory properties of compounds with membrane-stabilizing effects: Implications for treatment strategies. Int J Mol Sci.2016”.

When the extract assessed for membrane stabilizing activity, Avarampoo exhibited commendable protective effects on human red blood cells (HRBC), demonstrating a maximum inhibition of 50.0% at a concentration of 500 µg. Although this was

slightly lower than the well-established standard, diclofenac, which achieved a 65.3% inhibition, the extract's ability to significantly reduce hemolysis underscores its therapeutic potential.

In Vitro Anti-Inflammatory activity by Protein Denaturation Method

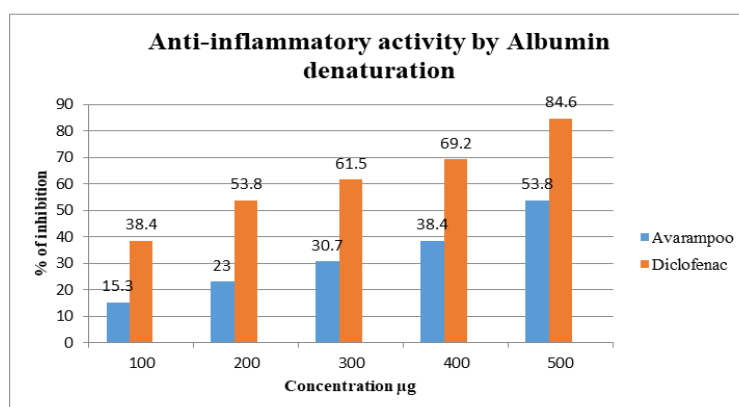
Table 3 and 4 and Figure 2 shows the anti-inflammatory property of Avarampoo by albumin denaturation activity.

Table 3. Albumin Denaturation activity of Diclofenac (Standard Blank= 0.13)

Sample Concentration (µg)	100	200	300	400	500
Diclofenac	0.08	0.06	0.05	0.04	0.02
% inhibition	38.4	53.8	61.5	69.2	84.6

Table 4. Albumin Denaturation Activity of Avarampoo (Sample Blank= 0.13)

Sample Concentration (µg)	100	200	300	400	500
Avarampoo	0.11	0.10	0.09	0.08	0.06
% inhibition	15.3	23.0	30.7	38.4	53.8

**Figure 2.** Anti-Inflammatory Activity of Avarampoo by Albumin Denaturation at Different Concentrations

This study was based on “Raghavendra, *et al.* (2016). "Anti-inflammatory potential of *Cassia auriculata*: Evaluation of protein denaturation inhibition." *Asian Pacific Journal of Tropical Medicine*”

The results indicate that diclofenac exhibited significant inhibition of albumin denaturation, with a maximum percentage inhibition of 87.5% at 500 µg. In contrast, the Aavarampoo extract showed a progressive increase in inhibition, achieving 50.0% at the highest concentration tested.

Although the initial concentrations of Aavarampoo did not yield measurable inhibition, the extract still demonstrated the ability to significantly reduce albumin denaturation, underscoring its potential as a natural anti-inflammatory agent.

Discussion

The anti-inflammatory properties of Avarampoo are often attributed to its ability to

inhibit protein denaturation, a process commonly associated with inflammation and disease conditions like arthritis. Denaturation refers to the loss of a protein's native structure due to external stress (e.g., heat, pH changes), leading to its inactivation. In inflammation, this process can trigger harmful responses in tissues.

Avarampoo extracts (aqueous or ethanolic) have bioactive compounds like flavonoids, tannins, and phenolic acids that can stabilize protein structures under stress. These compounds interact with proteins, preventing conformational changes that lead to denaturation. The activity can be measured using assays like heat-induced albumin denaturation or bovine serum albumin (BSA) denaturation assays. Avarampoo extracts are tested for their ability to inhibit denaturation under high temperatures or adverse conditions. Inhibition of protein denaturation is considered

an indirect measure of anti-inflammatory activity because the process mimics inflammatory mechanisms in vivo. Avarampoo has shown promise in reducing inflammatory markers and providing relief in inflammatory conditions. Venkatesh et al. (2014) investigated the membrane stabilizing activity of Avarampoo through in vitro assays on human red blood cell (RBC) membranes. The study demonstrated that Avarampoo extract significantly inhibited hypotonic solution-induced hemolysis (destruction of RBCs), which is a common model for assessing membrane stability. This suggests that Avarampoo has potential anti-inflammatory effects, as it stabilizes the cell membrane against oxidative damage [5]. Choudhury et al. (2017) reported that Avarampoo's ethanolic extract showed significant membrane stabilization in vitro. The extract exhibited the ability to reduce the release of hemoglobin from RBCs when exposed to heat-induced denaturation, pointing to its potential as an anti-inflammatory and protective agent in cellular membranes [6]. The mechanism for this membrane stabilizing effect is thought to be due to the presence of bioactive compounds like flavonoids and saponins, which are known to stabilize cellular membranes by interacting with phospholipids [7].

Protein denaturation is a process where proteins lose their native structure due to various factors, such as heat, pH changes, or oxidative stress. During inflammation, proteins are often denatured due to the release of inflammatory mediators, which can aggravate tissue damage. Inhibition of protein denaturation is one of the key methods used to assess anti-inflammatory potential. Raghavendra et al. (2016) examined the anti-inflammatory activity of Avarampoo by assessing its effect on protein denaturation using the heat-induced protein denaturation method [5]. The study showed that the aqueous extract of Avarampoo exhibited

significant inhibition of protein denaturation, suggesting that the plant extract can prevent the destabilization of proteins during inflammatory responses. The authors concluded that Avarampoo could be a potent anti-inflammatory agent by preventing protein denaturation, which is a hallmark of the inflammatory process. Kamaraj et al. (2019) used the protein denaturation method to further explore Avarampoo's anti-inflammatory activity. Their study confirmed that Avarampoo extract demonstrated a dose-dependent inhibition of protein denaturation, which could help reduce the inflammatory responses typically seen in conditions like arthritis and other inflammatory disorders [8]. The inhibitory effect on protein denaturation was attributed to the presence of compounds such as anthraquinones and flavonoids, which are known for their antioxidant and anti-inflammatory properties.

The inhibition of protein denaturation suggests that Avarampoo could reduce the production of inflammatory cytokines and free radicals, thus contributing to the overall anti-inflammatory response [9, 10, 11].

Conclusion

The studies reviewed indicate that Avarampoo (*Cassia auriculata*) shows strong anti-inflammatory activity, primarily through its membrane stabilizing properties and inhibition of protein denaturation. These mechanisms provide evidence that Avarampoo can be an effective natural therapeutic agent in managing inflammation-related disorders.

Future Directions

Further research should focus on:

1. Isolating specific active compounds responsible for these activities.
2. Conducting clinical trials to confirm its efficacy in humans.
3. Exploring synergistic effects with other anti-inflammatory agents.

4. These steps will be crucial in validating Avarampoo as a viable treatment option for inflammation and related diseases.

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Conflict of Interest

Nil.

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