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PHYTOCHEMICAL ANALYSIS, ANTIOXIDANT POTENTIAL AND ANTHELMINTIC ACTIVITY OF PETROLEUM ETHER AND ETHANOLIC FRUIT EXTRACT OF *AVERRHOA CARAMBOLA* AGAINST *PHERETIMA POSTHUMA*

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ABSTRACT

The present study aimed to evaluate the Antioxidant and Anthelmintic activity of fruit extract of *Averrhoa carambola*. The fresh fruits of *Averrhoa carambola* were collected and authenticated. The coarse powder was subjected to successive extraction by Soxhlet method using Petroleum ether and Ethanol as solvents. Further, the phytochemical constituents were identified by standard chemical tests. The Antioxidant activity of ethanolic extract was determined using DPPH method. The *In-vitro* Anthelmintic activities of both extracts at different concentration (25mg, 50mg and 100mg) were examined against *Pheretima posthuma*. The Vermifugal and Vermicidal effects were recorded in minutes. The results were expressed as Mean±SEM statistical analysis by One-Way ANOVA followed by Tukey's multiple comparison method. The present study revealed that increasing the concentration, increased the potency of extracts. The results indicated *Averrhoa carambola* exhibits Antioxidant and Anthelmintic activity and support the traditional usage of this fruit.

KEYWORDS

Averrhoa carambola, Pheretima posthuma, Albendazole, Vermifugal, Vermicidal and Mean±SEM.

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INTRODUCTION

As per WHO, Gastrointestinal Helminthic infections remain a major cause for deaths and are among the most widespread infections in humans especially due to poor sanitization facility. Infectious diseases remain major contributing factor of the debilitating poverty affecting the large portion of the world today[1]. It is the most prevalent worm-borne infection that affects the human body. The worms typically reside in the liver, gastrointestinal tract and other organs. Many Anthelmintic medications are currently available in

the market including Albendazole, Niridazole, Ivermectin and Praziquantel which are extensively used to treat helminthiasis. However, there are significant side effects associated with these medications including hepatotoxicity, appetite loss, lightheadedness, GIT disturbance, headaches and stomach discomfort[2]. Therefore, it is imperative to search for herbal Anthelmintic medications that are more effective while having the fewer possible adverse effects[3]. Also, many of these drugs are not recommended for pediatrics and pregnant ladies. Hence, plant derivatives are used in the discovery and development of Anthelmintic drugs[4].

Plants and its derivative form the basis for traditional system of medicine. Similarly, one such plant derivative (*Averrhoa carambola, Oxalidaceae*) was taken to evaluate its activity. During the Literature Survey, it was concluded that the activities of crude extracts of leaves, stem, and bark of *Averrhoa carambola* were investigated. But, there were no reports found on Anthelmintic activity of the fruit. Therefore, the present study aimed to examine the Anthelmintic activity as well as to confirm the Antioxidant potential which was previously reported.

The star fruit plant or *Averrhoa carambola* is medium-sized tree that belongs to the *Oxalidaceae* family and is characterized by its unusually beautiful star-shaped fruit[5]. It is rich in natural antioxidants like gallic acid and vitamin C[6]. *Averrhoa carambola* has slightly sour, acidic, juicy, crunchy, and sweet flavor. It has a golden-yellow color and resembles a star[7]. In general, star fruits are considered a rich source of natural phytochemicals like flavanoids, saponins, terpenes, alkaloids, tartaric acid, oxalic acid, citric acid, vitamin B1 and B2, carotene, fatty acids, fibers, polysaccharides and sterols along with an abundance of other nutrients like minerals, proteins, and vitamins[8].

METHODOLOGY

Plant sample

Fresh fruits of *Averrhoa carambola* were collected from the local region of Bangalore district, Karnataka. The fruits were identified and authenticated from Central Ayurveda Research Institute, Bangalore. The collected fresh fruits of *Averrhoa carambola* were washed in water, shade dried at room temperature and then pulverized to coarse powder and stored in air tight container.

Preparation of the extracts

The coarse powder was subjected to successive extraction by Soxhlet method using Analytical graded solvents procured from S. D. Fine/Qualigenf, Bangalore.

Soxhlet extraction

The dried, coarse powder of about 100g was placed inside the thimble and closed tightly. The successive extraction solvents (Petroleum ether and Ethanol) were placed in round bottom flask. The round bottom flask consisting of the 500ml solvent is heated; solvent evaporates, condenses and comes in contact with drug. The procedure was continued until the concentrated extract was obtained[11]. After extraction, the extracts were concentrated using Rotary flash evaporator and stored in Desiccators containing anhydrous calcium chloride that absorbs moisture content. Further, the phytochemical constituents of fruit extract of *Averrhoa carambola* were identified qualitatively by standard chemical tests.

Antioxidant Activity- DPPH method (1.1-diphenyl-2-picrilhidrazil)

The antioxidant potential of the ethanolic extract of *Averrhoa carambola* was determined by DPPH method. A radical solution was prepared by dissolving 2.4mg DPPH in 100ml methanol[12]. A test solution was added to methanolic DPPH. The mixture was shaken vigorously and kept in the dark for 30mins at room temperature. Absorbance of the reaction mixture was measured spectrophotometrically at 517nm. Absorbance of blank was also measured. All the determinations were performed in triplicate. The capability to scavenge the DPPH radical was calculated using the formula:

% of antioxidant activity = [(control absorbance-absorbance of sample)/ control absorbance] × 100[13].

Anthelmintic Modeling Procedure

The *Pheretima posthuma* (Indian earthworms) were collected from local regions of Bangalore district, Karnataka. The worms (*Pheretima posthuma)* were subjected to Anthelmitic activity by using Petroleum ether and Ethanol extracts of different concentrations with that of standard drug, (Albendazole).

All the test solutions and standard solutions were prepared freshly before the start of the experiment. Each solvent extracts of *Averrhoa carambola* fruit was diluted to obtain 25, 50 and 100mg/ml concentrations. Similarly, the standard drug, Albendazole was diluted to obtain 25mg/ml concentration. The earthworms were washed with normal saline and placed in petridishes at room temperature and all these dilutions were poured into the petridishes accordingly. The mean time of paralysis and death time of worms were recorded in terms of minutes. The time of paralysis was noted when no movement was observed and death time was recorded when the worms lost their complete motility and faded away their body colors[14].

RESULTS AND DISCUSSION

Anthelmintic of *Averrhoa carambola*

The fresh fruits of *Averrhoa carambola* were procured, dried and pulverized. The solvents such as Petroleum ether and Ethanol were used for extraction. The phytochemical screening revealed the presence of Alkaloids, Glycosides, Flavanoids, Triterpenoids, Tannins, Saponins, Polyphenols and Carbohydrates (Table No.1). The predominant effect of Albendazole is to bind to the β-tubulin and prevents its polymerization into microtubules, the cascade of cell division is corrupted at metaphase which ultimately leads to the death of individual cells and finally death of parasite[15].

Many studies revealed that the phenolics and flavanoids content contribute to the antioxidant activities[16,17]. Similarly, the Ethanolic extract of *Averrhoa carambola* is rich in antioxidants like polyphenols, flavanoids and carbohydrates which accounts for having the highest antioxidant potential compared to Petroleum ether extract (Table No.2).

The *In-vitro* Anthelmintic activity was performed and the mean time of paralysis (Vermifugal) and death time (Vermicidal) in minutes were recorded. The results were expressed as Mean±SEM statistical analysis by One-Way ANOVA followed by Tukey's multiple comparison method. From the results shown in Figure No.1(a) and 1(b), the fruit extracts of *Averrhoa carambola* exhibited Anthelmintic activity in dose dependent manner. The 100mg of the extracts demonstrated the paralysis and death of worms in a lesser time compared to 50mg and 25mg. Therefore, the study concludes that the doses of 50mg/ml and 25mg/ml showed relatively promising effect, whereas 100mg/ml showed significant activity when compared with the standard drug (Albendazole). Phytochemical analysis revealed presence of Flavanoids as one of the chemical constituents. Study declare that Polyphenolic compounds show Anthelmintic activity[18]. Similarly, is is possible that phenolic content in fruit extract of *Averrhoa carambola* produce similar effects.

Common Names of Star Fruit (*Averrhoa carambola L.*)[9,10]

S.No	Language	Name
1	English	Star Fruit, carambola
2	Kannada	Kamaraakshi
3	Hindi	Kamrakh
4	Telugu	Ambanamkaya
5	Tamil	Thambaratham
6	Bengali	Kamranga

Table No.1: Phytochemical components of *Averrhoa carambola*

Petroleum ether extract	**Positive results for**
	Alkaloids
	Glycosides
	Flavanoids
	Carbohydtares
Ethanol extract	**Positive results for**
	Alkaloids
	Glycosides
	Flavanoids
	Triterpenoids
	Tannins
	Saponins
	Polyphenols
	Carbohydrates

Table No.2: Antioxidant potential of *Averrhoa carambola*

S.No	**Ethanolic extract of *Averrhoa carambola***	**DPPH (% antioxidant activity)**
1	25mg	329.833±3.311
	50mg	356.166±3.710
	100mg	387.666±7.580
Petroleum ether extract of *Averrhoa carambola*		**DPPH (% antioxidant activity)**
2	25mg	184.166±3.970
	50mg	196.166±2.868
	100mg	199.66±2.804

Star fruits

Anthelmintic of *Averrhoa carambola*

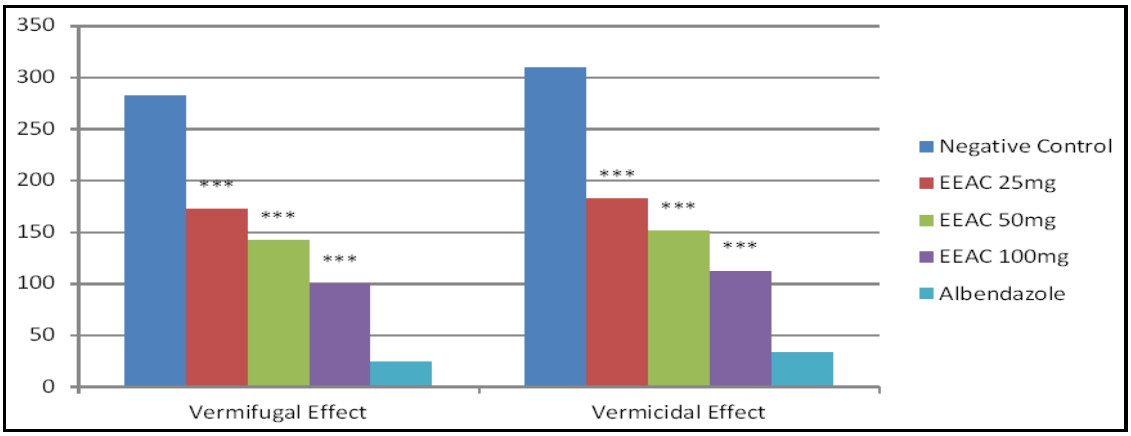

Figure No.1(a): Anthelmintic Activity of Ethanol extract of *Averrhoa carambola* (EEAC)

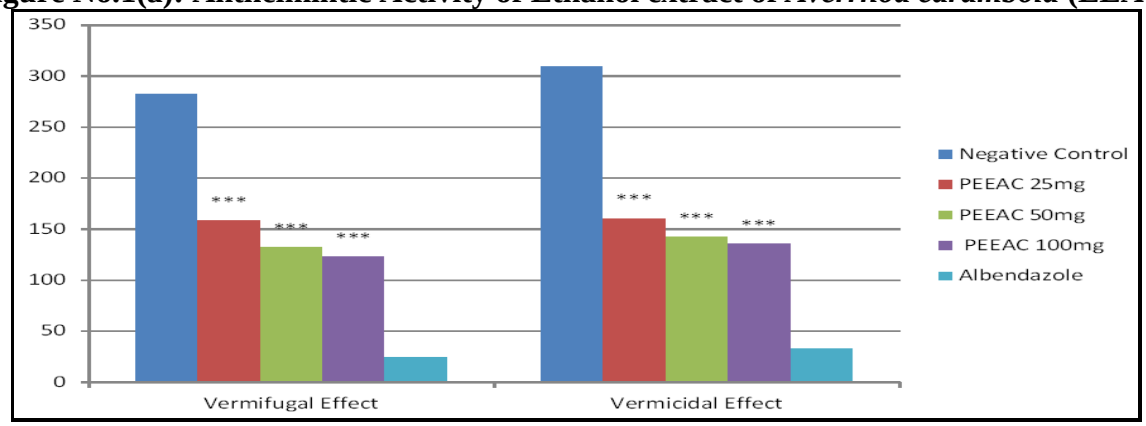

Figure No.1(b): Anthelmintic Activity of Petroleum ether extract of *Averrhoa carambola* (PEEAC)

CONCLUSION

The present study confirmed the traditional use of *Averrhoa carambola* as antioxidant and Anthelmintic medication. The majority of helminthiasis cases are managed using Anthelmintic medications that are currently available in the market. However, these medications have significant side effects. Therefore, herbal medicine is a complementary therapy that uses various plants and plant extracts for therapeutic purpose with lesser side effects. Thus, the screening of medicinal plants and their products for Anthelmintic activity continues to be of great scientific interest. Similarly, the present study enabled us to know the significant effect of *Averrhoa carambola* with minimum side effects.

Further detailed investigation is required to determine the particular active component responsible for the Anthelmintic activity, which may aid the society.

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CONFLICT OF INTEREST

We declare that we have no conflict of interest.

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