Phytochemical Screening and Anthelmintic Activity of Psidium guajava (L.) leaves Extract

Soumyaranjan Biswal[1] *, Anup Kumar Dash[2], Anupam Sahoo[3], Chandrika Kumari Bodala[4]

[1,2] Assistant Professor, Department of Pharmacology, Gayatri Institute of Science and Technology, Regeda, Gunupur 765022 , Odisha, India.
[3] Associate Professor, Department of Pharmaceutics, Gayatri Institute of Science and Technology, Regeda, Gunupur 765022 , Odisha, India.
[4] Assistant Professor, Department of Pharmaceutics, Gayatri Institute of Science and Technology, Regeda, Gunupur 765022 , Odisha, India

Abstract:

As per WHO , there are very less anthelmintic drugs are used in Helminthes. For parasitic infection the anthelmintic from natural source may play an important role. Hence, this study was undertaken to evaluate the anthelmintic efficacy of Psidium guajava (L.) leaves extract against parasites. Phytochemical screening was carried out on ethanolic extract of Psidium guajava (L.) and then screened for its anthelmintic property against earth worm Pheretima posthuma. The effect of Psidium guajava leaves Extract and its fractions on the anthelminthic activity of piperazine citrate were also determined. PGE showed a dose-dependent anthelmintic activity against Pheretima posthuman. Anthelmintic activity of aqueous and alcoholic extract of leaves of Psidium guajava were evaluated separately with different concentraction on adult earth worm and compared with the standard drug piperazine citrate. The result found that extract exhibited, dose dependent action and inhibition of spontaneous motility (paralysis) and death of earth worm.

Key words: 1. Psidium guajava , 2.Anthelmintic activity, 3.Earthworm, 4.Phytochemicals, 5.Pheretima posthuman

Introduction :

The Herbal medicinal system is one of the oldest medicinal system, which comprises thousands of medical concepts and hypothesis. it is the complete medical system which have been proving to be real blessing to the people. Ayurvedic medicine is highly effective; but mode of action, pharmacology, pharmacokinetics, and many activity are still not fully explored. However, the efforts are needed to explore the ayurvedic medicines for their potency, safety, and efficacy in order to bring them to market. [1,2,3]

Psidium guajava (common name-guava) is belongs to *Myrtaceae* family which is grown in tropical and many areas like India, Bangladesh, Florida, and West Indies. The plant containing different bioactive constituents like phenolic compounds, flavonoids, gallic acid, catechin, epicatechin, rutin, narigenin, kaempferol . In traditional system different parts of the plant parts like root, bark, leaves and fruits have been

used for different disease condition like diarrhea, dysentery, wounds, ulcers, toothache, cough, vomiting, sore throat etc. The plant have also hepatoprotective, antioxidant, anticancer, antispasmodic, anti-inflammatory, antimicrobial, anti hyperglycemic, analgesic action.[4,5,6,7]

Fig. 1: Psidium guajava plant **Fig. 2: Psidium guajava plant Leaves** **Fig. 3: Psidium guajava Dry Leaves**



Taxonomic classifications [1]

Kingdom: Plantae
Phylum: Tracheophyta
Class : Magnoliopsida
Subclass: Rosidae
Order: Myrtales
Family : Myrtaceae
Subfamily: Myrtoideae
Genus: Psidium
Species : Guajava

Vernacular name :[8,9]

Sanskrit: Perala, amratafalam
English: Lemon Guava, Apple Guava
Hindi : Amrud
Odia: Pijuli
Bengali: Peyara
Gujarati: Jamrukh
Marathi: Peru
Kannada: Sibehannu
Telugu: Jamapandu
Malayalam: Perakka
Tamil: Koyya

Materials and Methods:[10,11,12,13]

Materials

The plant of Psidium guajava Leaf's was collected in the month of January from the Gunupur Rayagada, odisha. Then the plants were dried under shade separately (temperature 30-40°C). Air dried plant material was ground to powder and the plant materials were then used for further investigations. For anthelmintic study adult earthworm pheretima posthuma were procured from the local area of gunupur, odisha.

Methods

Extraction of plant material:

About 25 gm of dry crushed material was extracted with Ethanol by continuous hot percolation using soxhlet apparatus. The extraction was continued for 10 hrs. the extracts were concentrated by distilling the solvent and the extracts were dried on water bath. Then consistency, color, appearance of the extracts and their percentage yield were noted.

Figure : 4 Psidium guajava leaves extraction by soxhlet apparatus.

Extractive value

Extractive value determines the amount of active constituents extracted with solvents from given amount of herbal material. It is usually calculated as alcohol soluble and aqueous soluable extractive value.The % yield of ethanolic and aqueous extracts was calculated as,

$$\% \text{ Yield of Extract: } \frac{\text{Weight of dried extract}}{\text{Weight of plant material}} \times 100$$

% Yield of Ethanolic Extract: 2.19 / 25 × 100 = 8.76

% Yield of Ethanolic Extract: 2.19 / 25 × 100 = 7.42

Preliminary phytochemical screening:[1,4,5,6]

Phytochemical analysis of the different extract of stem bark was carried out using standard methods. The plant materials were checked for the presence of various active constituents. Like carbohydrates, protein, phenolic, tanin, saponins, flavonoids, terpenes, steroids and alkaloids **(Table-3).**

Materials: Water, Alcohol, 70 % alcohol, extracts, Reagents: mayer's, wagner's, sulphuric acid, 10 % ammonia, pyridine, sodium nitropruside, acetic anhydride, ferric chloride, 10 %, 5% lead acetate, 10 % ammonium hydroxide solution. [2,3,6]

Animal:

Adult earthworm pheretima posthuma were procured from the local area of gunupur, odisha. The length of the pheretima posthuma is 9 to 10 cm. The earthwarm were washed with normal saline and divided into different group, each group containing five animals. The different extracts and standard solution (Piperazine citrate) were freshly prepared before performing the experiments. Then different concentrations (25, 50, 75 and 100 mg/ml in distilled water) of test sample of extracts (Ethanol, aqueous) were prepared and poured into different Petri dishes. The pheretima posthuma were transferred one by one into the petri dishes. The observations were made for the time taken to paralysis and death of individual worms were noted. The time taken for paralysis was noted when any movement of worm could not be observed. Death was included when the worms lost their motility followed by white secretions and fading away of their body colour.
Statistical analysis of the data were expressed as mean ± standard error mean (SEM)

Result and Discussion

The preliminary phytochemical investigation revealed that the presence of different phytoconstituents like Phenolics, tannins, steroids, flavonoids, saponins, terpenoids, glycosides etc . Table 3. The presence of above phytoconstituents may be responsible to show a potent anthelmintic activity.

The extractive values of psidum guajava leaves were recorded during the study. The extractive values for ethanolic, aqueous extract were found to be 8.76% and 7.42%respectively.

The peak anthelmintic activity exhibited by the ethanolic extract at highest concentration (100 mg/ml) which takes 10.30 ± 0.03 minute for paralysis and 19.30 ± 0.03 minute for death of the worms. Potency of the extract was inversely proportional to the paralysis (vermifuge) and death (vermicidal) time of the worms. Different extract of psidium guajava possess several medicinal properties, has been extensively researched for various pharmacological properties.

Table no-1 Macroscopy of *Psidium guajava (L.)*

Part of plant	Morphology	Observation
Leaves	Colour	Green
	Odour	Characteristic
	Taste	Herbaceous
	Texture	smooth

Table 2. Preliminary phyto profile of *Psidium guajava (L.)*

Sr. no.	Solvent	Color	Consistency	%Yield w/w
1	Ethanol	Dark Green	Slight Sticky	8.76
2	Aqueous	Dark Green	Sticky	2.12

Table 3. Phytochemical screening of extracts of *Psidium guajava* (L.)

Chemical constituents	Ethanol Extract	Aqueous Extract
Carbohydrate	+	+
Protein	-	-
Phenolics & Tannins	+	+
Saponins	+	+
Flavanoids	+	+
Terpenes	+	-
Steroids	-	-
Alkaloids	+	+

'+' presence '-' absence

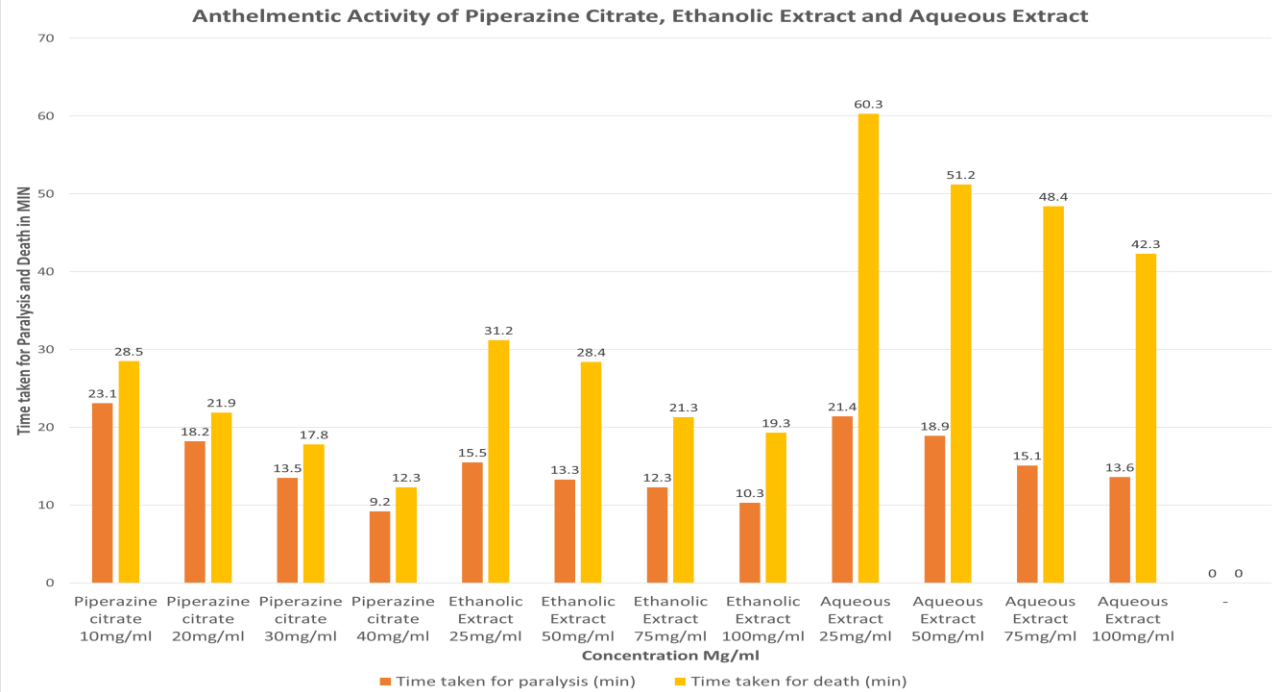
Table: 4 Paralysis and death time of ethanolic extract, aqueous extract and Piperazine citrate

Test substance	Concentration (mg/ml)	Time taken for paralysis in min	Time taken for death in min
Piperazine citrate	10	23.10±0.04	28.50±0.02
	20	18.20±0.02	21.90±0.04
	30	13.50±0.01	17.80±0.06
	40	09.20±0.02	12.30±0.03
Ethanolic Extract	25	15.50±0.06	31.20±0.02
	50	13.30±0.02	28.40±0.04
	75	12.30±0.04	21.30±0.06
	100	10.30±0.03	19.30±0.03
Aqueous Extract	25	21.40±0.06	60.30±0.03

	50	18.90±0.06	51.20±0.01
	75	15.10±0.03	48.40±0.02
	100	13.60±0.04	42.30±0.03
Control	-	-	-

Figure : 5 Anthelmintic activity of Piperazine citrate and different Psidium guajava leaves extract in Indian earthworm.

Figure : 4 Anthelmintic activity of Piperazine citrate, Ethanolic Extract and Aqueous Extract of Psidium guajava leaves



Conclusion

The present study showed that the Leaves extract of ***Psidium guajava*** possess anthelmentic activity. The ethanolic extract (100mg/ml) of the leaves extract possess highest anthelmintic activity then the Aqueous extract.

Flavonoids, tannins, saponins, alkaloid and steroids are the main chemical constituent that are present in the leaves of ***Psidium guajava*** which may be responsible for anthelmentic activities.

Acknowledgments:
The authors would like to thank msanagement of gayatri institute of science and technology, gunupur for their kind support during the studies.

References:

1. Reddy SN, Yeedulapally SR, Palusam GV, Pathi N, Gunala MSG, Bakshi V, Boggula N. Anthelmintic Activity and Phytochemical Evaluation of Psidium Guajava Leaves. Journal of Pharmaceutical and Scientific Innovation. 2018 June 17;7(3): 82-87.
2. Shah D, Shah SS, Khan I, Khan JM, Nabi M, Zeb J, Imad J, Khan I, Shah I, Khan MW. A review of Phytochemistry, Traditional Uses and Antibacterial Potential of Psidium guajava leaves Extract. Letters in Applied Nano Bio Science. 2020 January 19;10(3) :2506-2514.
3. Cerio ED, Verardo V, Ceravaca AMG, Gutierrez AF, Carretero AS. Health Effects of Psidium guajava L. Leaves. International Journal of Molecular Sciences. 2017 April 24; 18, 897 ; 1-31.
4. Pradhan DK, Mishra MR, Hota A. Phytochemical and Comparative Anthelmintic Activity of Psidium guajava L. Leaves extract against Pheretima posthuma. Journal of Pharmaceutical Research. 2021 November 24;20(3):5–8.
5. Anbuselvi S, Rebecca J. The Phytochemical Biochemical and Antimicrobial Activty of Psidium Guajava Leaf Extract. Journal of Pharmaceutical Science and Research. 2017 ;9(12): 2431-2433

6. Hussain S, Naseer S, Naeem N, Pervaiz M, Rahman M. The Phytochemistry & medicinal value of Psidium guajava Extract (guava). Clinical Phytoscience. 2018; 4:32 .
7. Calagui RT. The Anthelmintic activity of Chrysophyllum cainito and Psidium guajava ethanolic bark extracts against Ascaridia galli of chicken. International Journal of Biosciences. 2021 Sepetember 30; 19 (3): 141-147.
8. Uzzaman S, Akanda KM, Mehjabin S, Parvez GM. A review on a Nutritional Fruit : Guava. Bio core. 2018 December 3; 1(1): 1-8.
9. Prabhudesai AP, Biyani D, Umekar MJ. Psidium guajava: Multipurpose Medicinal Herb. International Journal of Pharmaceutical Science Review & Research (IJPSRR). 2019 November 3; 59(1); 125-132
10. Silva LP, Debiage RR, Júnior JLB, Silva RMG, Peixoto ECT. In vitro anthelmintic activity of Psidium guajava extract against gastro-intestinal sheep nematodes. Biological sciences. 2020 June 17;92(2).
11. Metwally AM, Omar AA, Harraz FM, Sohafy SM. The Phytochemical investigation and antimicrobial activity of Psidium guajava L. leaves. Pharmacognosy Magazine.2010 July 30 ; 6 (23): 212-218.
12. Chand S, Netish KK, Saiful I, Rana A, Safia A, Nazmus S, Mir IIW. Antioxidant, Analgesic & Antimicrobial Activities of Different Fractions From Methanolic Extract Of Psidium Guajava L. Leaves. International Journal of Pharmaceutical Sciences and Research. 2020 June 1;11(6): 2733-2738.
13. Kaware MS, Ismail MS. Antimicrobial Activity of Guava Leaf Extracts Against Some Clinical Bacteria Isolates. Dutse Journal of Pure and Applied Sciences (DUJOPAS).2020 June; 7(2): 245-254.
14. Melo C, Corneja N, Cruz V, Alsaidi S, Rodriguez GC, Ramirez AG, Sorel V, Bonnaire T, Zhang S, Zydowsky TM, Priano C , Romero JAF, Koroch A. Antioxidant and Antimicrobial Activity of Commercial Samples of Guava Leaves (Psidium guajava). Journal of Medicinally Active Plants. 2020 March 31; 9 (1).
15. Das M, Goswami S. Antifungal and Antibacterial Property of Guava (Psidium guajava) Leaf Extract. International Journal of Health Sciences & Research. 2019 February ;9(2):39-45