



Anthelmintic activity of *Lawsonia inermis* leaf extracts against *Pheretima posthuma* and *Ascaridia galli*

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ARTICLE DETAILS	ABSTRACT
<p><i>Article history:</i> Received on 2 March 2021 Modified on 25 March 2021 Accepted on 29 March 2021</p> <hr/> <p><i>Keywords:</i> <i>Lawsonia inermis</i> Linn, Anthelmintic Activity, <i>Ascaridia Galli</i>, <i>Pheretima Posthuma</i>.</p>	<p>Plants play an important role on the earth, and humans depend on them for their medicinal properties. Medicinal plants are commonly used in the health-care delivery system, either as a single drug or in combination. Helminthes infections are among the most common infections in humans, affecting a significant portion of the global population. Although the majority of helminth infections are confined to tropical areas, they pose a significant health risk and lead to the prevalence of malnutrition, anaemia, eosinophilia, and pneumonia. Parasitic diseases cause a high incidence of morbidity in people living in endemic areas. The anthelmintic activity of ethyl acetate and ethanolic extracts of <i>Lawsonia inermis</i> Linn leaves against <i>Pheretima posthuma</i> and <i>Ascaridia galli</i> is currently being investigated. In the bioassay, various concentrations were used, and the worms were paralysed and died after a certain period of time. Both extracts were found to have significant anthelmintic activity.</p>

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INTRODUCTION

In India, mehndi is usually applied to the hands and feet [1]. Because of its cooling effect in the hot Indian summers, it became popular in India [2,3]. *Lawsonia inermis* Linn (Family: Lythraceae), also known as henna, is a plant that grows mostly in subtropical and tropical climates and is used all over the world [4-8]. It has been used as a beauty dye for over 9000 years. Carbohydrates, phenolics, flavanoids, saponins, proteins, alkaloids, terpenoids, quinones, coumarins, xanthenes, fat, resin, and tannins were contained in *Lawsonia inermis* phytochemical study. 2-hydroxy-1,4-naphthoquinone was also present [9-13]. *Lawsonia inermis* was used to isolate a number of alkaloids, naphthoquinone derivatives, phenolics, and flavonoids. Antibacterial, antifungal, antiparasitic, molluscicidal, antioxidant, hepatoprotective, central nervous, analgesic, anti-inflammatory, antipyretic, wound and burn healing, immunomodulatory, antiurolithiatic, antidiabetic, hypolipidemic, antiulcer, antidiarrhoeal, diuretic, anticancer,

and many other pharmacological effects were found in *Lawsonia inermis* [14-19].

Traditional medicine uses henna leaves, flowers, seeds, stem bark, and roots to treat a number of illnesses, including rheumatoid arthritis, headaches, ulcers, diarrhoea, leprosy, fever, diabetes, heart disease, hepatoprotective, and colouring agent. Henna leaf has an orange-red dye and leaf paste or powder is widely used for decorating hands, nails and feet with patterns. It's also used to hair dye [20-27]. In India, mehndi is usually applied to the hands and feet. The sodium content of 25 populations of *Lawsonia inermis* collected from the coastal oases of Gabès in Tunisia ranged from 0.08 to 0.69 % in leaves and 0.08 to 0.46 % in stems, according to mineral analysis. Potassium levels in the leaves ranged from 0.16 to 0.47 %, while potassium levels in the stems ranged from 0.15 to 0.81 %. Calcium levels in the leaves ranged from 0.2 to 0.41 %, while calcium levels in the stems ranged from 0.11 to 0.47 % [28].

The magnesium content in the leaves ranges from 0.09 to 0.23 %, while the magnesium content in the stems ranges from 0.03 to 0.11 %. The leaves have a phosphorus content of 2.57 to 6.29 %, while the stems have a phosphorus content of 2.73 to 9.84 percent. The copper content in the leaves ranges from 0.06 to 1.87 %, while the copper content in the stems ranges from traces to 11.27 % [29-32].

Zinc levels in the leaves ranged from 0.47 to 2.92 %, while zinc levels in the stems ranged from 0.2 to 7.39 %. The iron content in the leaves ranges from 4.03 to 28.77 %, while the iron content in the stems ranges from 1.17 to 15.85 %. The manganese content in the leaves ranges from 0.27 to 1.28 %, while the stems have a manganese content of 0.14 to 0.95 %. The nitrogen content of the leaves ranged from 0.14 to 4.72 percent, and the stems from 0.17 to 0.56 %. Henna is a symbol of fertility. Because of its cooling effect in the hot Indian summers, it became popular in India [33-38].

Rheumatoid arthritis, headaches, ulcers, diarrhoea, leprosy, fever, leucorrhoea, diabetes, heart disease, hepatoprotective, and colouring agent are all handled with henna leaves, flowers, seeds, stem bark, and roots in traditional medicine. Henna leaves produce an orange-red dye, and the paste or powder is commonly used to make designs on the hands, nails, and feet. It's also used to dye hair. The microwave extraction method has proven to be more accurate and reliable than the soxhlet extraction method. A typical technique is soxhlet extraction, which is a continuous solvent extraction process. Soil, sediments, sludge, polymers and plastics, pulp and paper, biological tissues, textiles, and food samples are all collected using extraction systems [39].

Microwaves, in contrast to soxhlet extraction, use a smaller amount of solvent and analyse and extract at a much faster rate, according to studies [40]. An outbreak of coronavirus disease (COVID-19) triggered by the novel severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) poses an unparalleled challenge in the development of effective medicines for prevention and treatment. The oral cavity has been suggested as a possible reservoir for COVID-19 transmission due to the close proximity to the patient during dental treatment, the high generation of aerosols, and the detection of SARS-CoV-2 in saliva. Someday, you might be

able to make your own medicines at home. This is due to the fact that researchers have developed a 3D printer using clear, readily available medicinal active agents fed into a drug delivery system [41]. The main mode of gastrointestinal parasite control today is commercial anthelmintic. However, widespread anthelmintic resistance, scarcity, and high costs, particularly for low-income farmers in developing countries, necessitated the creation of alternative helminth control methods [42]. Traditional herbal dewormers, as well as other alternative approaches, are gaining popularity. The study's aim is to determine *Lawsonia inermis* Linn's anthelmintic potential [43].

MATERIALS AND METHODS

Collection of Plant Material

Lawsonia inermis Linn was obtained from Karad, Maharashtra, India. The plant was identified and authenticated by Department of botony, Yashwantrao Chavan College of Science, Karad.

Preparation of Plant Extract

Shade drying was done for almost a month as to avoid chemical degradation due to sunlight. Grinding of the dried material was done, with the aid of a grinder and converted into coarse powder. Extraction of *Lawsonia inermis* Linn was done by microwave extraction method further filtered and excess solvent present was evaporated and dried extract were collected and subjected for activity studies.

Animals

Indian adult earthworms (*Pheretima posthuma*) were collected from water logged areas and *Ascardia galli* (nematode) worm were obtained from freshly slaughtered fowls (*Gallus gallus*).

Evaluation of Anthelmintic Activity

The anthelmintic assay was carried as per method of Ajaiyeoba et al., with minor modifications. The anthelmintic activity was evaluated on adult Indian earthworm *Pheretima posthuma* worm due to its anatomical and physiological resemblance with the intestinal round worm parasites of human beings. *Ascardia galli* (nematode) worms are easily available in slaughtered fowls and it can be used as a screening model for anthelmintic drugs as advocated earlier. Three different concentrations, each of crude methanolic and ethyl acetate extract (10, 50, 100 mg/mL in distilled water) were and six worms (same type) were placed in it. This was done for both type of

worms. Observations were made for the time taken to cause paralysis and death of the individual. Mean time for the paralysis (P) in min was noted when no movement of any sort could be observed, except when the worm was shaken vigorously; time of death (D) in min was recorded after ascertaining the worms neither moved when shaken vigorously nor when dipped in warm water (50°C). Albendazole (10 mg/mL) was included as reference compound [40, 44].

RESULTS AND DISCUSSION

As shown in Table 1, ethyl acetate and ethanolic extract exhibited anthelmintic activity in dose-dependent manner giving shortest time of

paralysis (P) and death (D) with 100 mg/mL concentration. The ethyl acetate extract of *Lawsonia inermis* Linn caused paralysis of 9 min. and time of death of 25 min. While ethanolic revealed paralysis of 8 and 21 min. respectively against the earthworm *Pheretima posthuma*. The reference drug Albendazole showed the same at 20 and 55 minutes, respectively. *Ascaridia galli* worms also showed sensitivity to the extract of *Lawsonia inermis* Linn. The ethanolic extract caused paralysis in 11 min, death in 20 min and the ethyl acetate extract displayed P and D in 10 and 17 min, respectively. Albendazole did the same at 16 and 38 min.

Table 1: Anthelmintic activity of methanolic and ethyl acetate extract of *Lawsonia inermis* Linn

Test subs	Concentrations (mg/mL)	Time taken for paralysis (P) and death(D) of worms in min			
		P. posthuma		A. galli	
		P	D	P	D
Control	-	-	-	-	-
Ethyl acetate extract	10	25.09 ± 0.34	79.0 ± 0.31	28.43 ± 0.66	51.34 ± 0.72
	50	18.12 ± 0.43	41.08 ± 0.15	15.92 ± 0.41	31.39 ± 0.55
	100	09.41 ± 0.20	25.4 ± 0.18	11.30 ± 0.10	20.87 ± 0.71
Ethanolic extract	10	29.92 ± 0.56	60.71 ± 0.10	41.45 ± 0.12	60.18 ± 0.92
	50	25.59 ± 0.02	44.78 ± 0.92	30.96 ± 0.11	55.12 ± 0.85
	100	08.34 ± 0.21	21.98 ± 0.67	10.12 ± 0.25	17.81 ± 0.44
Standard (Albendazole)		20.44 ± 0.55	55.12 ± 0.82	16.48 ± 0.91	38.61 ± 0.71

CONCLUSION

In comparison to the reference drug albendazole, leaf extract of *Lawsonia inermis* Linn. not only demonstrated paralysis, but also induced worm death in a shorter period at higher concentrations of 100 mg/mL. The extracts included alkaloids, saponins, flavonoids, triterpenes, tannins, and steroids, according to phytochemical analysis. Tannins have been shown to have anthelmintic properties. Polyphenolic compounds are what tannins are chemically. It's probable that tannins present in *Lawsonia inermis* Linn., extracts have a similar effect. Tannins are said to have an anthelmintic effect since they may attach to free proteins in the gastrointestinal tract of the host animal or glycoprotein on the parasite's cuticle, causing death. Further research is underway to classify the phytoconstituents that may be responsible for anthelmintic activity.

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