International Journal of Herbal Medicine
Available online at www.florajournal.com

I J H M International Journal of Herbal Medicine

E-ISSN: 2321-2187
P-ISSN: 2394-0514
IJHM 2019; 7(3): 36-40
Received: 16-03-2019
Accepted: 20-04-2019

Akshay Javalgikar
Assistant Professor, Department of Pharmacology, Sahyadri College of Pharmacy, Methwade, Maharashtra, India

Hasan Shaikh
Department of Pharmacology, Sahyadri College of Pharmacy, Methwade, Maharashtra, India

Monali Sargar
Department of Pharmacology, Sahyadri College of Pharmacy, Methwade, Maharashtra, India

Harshvardhan Survanshi
Department of Pharmacology, Sahyadri College of Pharmacy, Methwade, Maharashtra, India

Mukesh Rathod
Department of Pharmacology, Sahyadri College of Pharmacy, Methwade, Maharashtra, India

Correspondence
Akshay Javalgikar
Assistant Professor, Department of Pharmacology, Sahyadri College of Pharmacy, Methwade, Maharashtra, India

In vitro anti-inflammatory and anthelmintic activity of *Tectona grandis* leaves extract

Akshay Javalgikar, Hasan Shaikh, Monali Sargar, Harshvardhan Survanshi and Mukesh Rathod

Abstract
Ayurvedic system of medicine is one of the oldest systems in India. Herbs and herbal products, with their incredibly wide use throughout time and place, continue to provide real health benefits while maintaining safety profile. The conservative drug available in the marketplace treat inflammation and analgesia produces various side effects. For conquer these problems medicinal plants play a major role to alleviate many diseases related with inflammation and analgesia. *Tectona grandis* (Family - Lamiaceae) is one of the most famous timber plants in the world and is renowned for its dimensional stability, extreme durability and hard which also resists decay even when unprotected by paints and preservatives. Teak is the major exotic species found in tropical regions. It allays thirst, and acts as antihelmintic, expectorant and anti-inflammatory. The objective of present study was to evaluate in vitro anti-inflammatory activity and antihelmintic of ethanolic extracts of *Tectona grandis* leaves. The results of plant extracts were found to have significant (P<0.005) anti-inflammatory activity and showing effective against parasitic infections.

Keywords: *Tectona grandis*, anti-inflammatory activity, antihelmintic activity, phytochemical tests

1. Introduction
Medicinal plants have been playing an essential role in the development of human culture. As a source of medicine, Medicinal plants have always been at forefront virtually all cultures of civilizations. Medicinal plants are regarded as rich resources of traditional medicines and from these plants many of the modern medicines are produced. For thousands of years medicinal plants have been used to treat health disorders. [1]

Now a day's natural products are an integral part of human health care system because there is popular concern over toxicity and resistance of modern drugs. India is one of the 12 leading biodiversity centres with presence of over 45000 different Plant species. Plants are richest resource of drug of traditionary system of medicine, modern medicines, nutraceutical's, food supplements, folk medicines, pharmaceutical intermediates and chemical entities for synthetic drugs. The uses of traditional medicinal plants for primary health care have steadily increased worldwide in recent years.

Under investigation we identified Teak Plant (*Tectona grandis)* which is belonging to family Lamiaceae. [15] The whole plant is medicinally important and many repots claim to cure severe diseases according to an Indian traditional system of medicine. The survey reveals that the plant used in treatment of urinary discharge, bronchi disorder, cold and Headache. It is also used as Laxative, sedative, as diuretics, Anti-diabetics.

Tectona grandis Linn commonly known as Teak or Sagwan is one of the most famous timber in the world and is renowned for its dimensional stability. *Tectona* is major exotic species found in the topical region. It is also commonly found in India and South Asian countries. *Tectona grandis* is tropical hardwood tree species placed in the flowering plant belonging into family Lamiaceae was first described by Carl Linnaeus the younger in his 1782 work supplementum plantarum. In 1975 Harold Norman Moldeke published new description of four form of this species in journal phytologia. [7]

Plant has major constituent has various pharmacological activities like antibacterial, antioxidant, antifungal, anti-inflammatory, antipyretic, analgesic antidiuretics and hypoglycemic. The *Tectona grandis* flowers used in bronchitis, biliousness and urinary discharge. Both flowers and seed are used as diuretics the wood have expectorant, anti-inflammatory, anti-bilious, anthelmintic action. The bark is powerful Astringent used in bronchititis. Root is used for anuria and retention of urine, Nut oil used in the treatment of Scabies and other skin disease and also for promoting hair growth. [3] The Ayurvedic Pharmacopoeia of India recommends the heartwood in lipid disorder and also for treating

threatened abortion. The wood is rich in anthraquinones, naphthalene compound and triterpenic and hemi-terpenic compounds.

The phytochemical-pharmacological research work has recently yielded effective solutions to certain diseases which synthetic drug industry has failed to afford. Thus. the research of pharmacologically/ biologically active agents obtained by screening natural sources such as plant extracts had led to the detection of many pharmaceutically valuable drugs that play a key role in the treatment of human diseases. [12] On the basis of pharmacological review of *Tectona grandis* Linn plant. We emphasised on inflammation and helminthic activity.

Inflammation is a complex process, which is frequently associated with pain and involves occurrences such as: the increase of vascular permeability, increase of protein denaturation and membrane alteration. The management of inflammation related diseases is a real issue in the rural community; the population in these areas uses many alternative drugs such as substances produced from medicinal plants. [2, 4]

Infections with helminth are among the most widespread infections in humans and other domestic animals affecting a large number of world population. Parasitic diseases cause ruthless morbidity affecting principally population in endemic areas. The gastrointestinal helminths become resistant to currently available anthelmintic drugs.

Therefore, the present study was designed to evaluate the in-vitro anti-inflammatory and anthelminthic activity of *Tectona grandis* Linn.

2. Material and Methods

2.1 Collection of plant material

The leaves of *Tectona grandis* were collected from Sahyadri botanical garden, Sahyadri College of pharmacy, Methwade, Sangola. The plant was dried under shade then ground in to a uniform powder using a blender and stored in polythene bags at room temperature.

2.2 Preparation of drug extract

The extract was prepared by two methods-

2.2.1 Maceration process

The leaves of *Tectona grandis* were shade dried, after shade drying the leaves were transferred into glass bottle containing 500 ML of petroleum ether for 24 hours. After that remove the leaves from bottle, collect the solvent and allow to steam distillation, collect the drug extract, dried it, take down its weight and note.

2.2.2 Soxhlet extraction process

Take 500 gm of leaves of shade dried *Tectona grandis* was extracted with 60ml 90% ethanol and 40ml purified water allow to complete extraction process after collect the product, allow to evaporation on steam bath, collect the product and carried out its phytochemical tests. [6]

2.3 Phytochemical tests

2.3.1 Tests for alkaloids

Small quantities of methanolic extracts of all the drugs were treated with few drops of diluted hydrochloric acid and filtered. The filtrates of each extract were divided into four portions and the following tests were carried out -

1. Dragendorff's Test: - With Dragendorff's reagent (solution of potassium bismuth iodid-e) formed orange brown precipitate.
2. Mayer's Test: - With Mayer's reagent (potassium mercuric iodide solution) formed creamy precipitate.
3. Hager's Test: - With Hager's reagent (saturated picric acid solution) formed yellow precipitate.
4. Wagner's Test: - With Wagner's reagent (solution of iodine in potassium iodide) formed reddish-brown precipitate.

The formation of respective precipitates indicated the presence of alkaloids.

2.3.2 Tests for carbohydrates and reducing sugars

The extracts were dissolved in water and filtered. The filtrates were divided into several portions and were tested.

1. **Molisch's test:** To one portion of filtrates of various drugs' extracts, few drops of α-naphthol solution in alcohol were added and mixed well followed by concentrated sulphuric acid from the sides. Purple ring at the junction of two liquids indicated the presence of carbohydrates.
2. **Benedict's test:** To a set of filtrates of various drugs' extracts, added equal volumes of Benedict's reagent and heated in boiling water bath for 5min. The appearance of green, yellow or red color indicated the presence of reducing sugars.
3. **Fehling's test:** One ml each of Fehling's A and Fehling's B were mixed and heated for one minute and equal volumes of the filtrates were added and heated for 5-10min on a water bath. First yellow, then brick red precipitate indicated the presence of reducing sugars.

2.3.3 Tests for glycosides

The following tests were carried out to detect the presence of different types of glycosides.

1. Legal's Test: - To the methanolic extracts, added pyridine and sodium nitroprusside and development of pink or red color indicated the presence of cardiac glycosides.
2. Borntrager's Test: - The methanolic extracts were boiled with dilute sulphuric acid and filtered. To the cold filtrates equal volumes of chloroform were added. After thorough shaking the organic solvent layers were separated and ammonia solution was added. The change of ammonia layer to pink or red color indicated the presence of anthraquinone glycosides.
3. Foam Test: - Small quantities of drugs were shaken vigorously with water. Formation of persistent foam indicated the presence of saponin glycosides.
4. Guignard reaction or sodium picrate Test: - Soaked filter paper strips first in 10% picric acid and then in 10% sodium carbonate and dried. Drugs were taken in small bottles and the strips were suspended from the mouth of the container and the lids were tightly closed with portion of the strip stuck in the lid. The strips did not turn brick red or maroon indicating the absence of cyanogenetic glycosides.
5. Extracts when made alkaline did not show blue or green fluorescence indicating the absence of coumarin glycosides.

2.3.4 Tests for phenolic compounds and tannins

Small quantities of the methanolic extracts were treated with the following reagents and the appearance of corresponding endpoints indicated the presence of phenolic compounds and tannins.

1. With 5% Ferric chloride solution: Deep blue-black color.
2. With 10% lead acetate solution: White precipitate.

3. With 10% Potassium dichromate solution: Red precipitate.

2.3.5 Tests for flavonoids

i) Shinoda Test: - Methanolic extracts were extracted with 95% ethanol and hydrolysed by concentrated hydrochloric acid. Pink colour appeared after adding the magnesium turnings. Formation of yellow precipitates when lead acetate was added to the residues indicated the presence of flavonoids. [3]

2.4 Pharmacological screening activity

2.4.1 *In vitro* Anti- inflammatory activity: inhibition of albumin denaturation

The anti-inflammatory activity of *Tectona grandis* was studied by using inhibition of albumin denaturation technique which was studied according to Mizushima *et al*. [13] and Sakat *et al*. [14] followed with minor modifications. The reaction mixture was consisting of test extracts and 1% aqueous solution of bovine albumin fraction, pH of the reaction mixture was adjusted using small amount of 1N HCl. The sample extracts were incubated at 37ºC for 20 min and then heated to 51ºC for 20 min, after cooling the samples the turbidity was measured at 660nm (UV Visible Spectrophotometer) The experiment was performed in triplicate.

The Percentage inhibition of protein denaturation was calculated as follows: Percentage inhibition = (Abs Control – Abs Sample) X 100/ Abs control

2.4.2 Anti-helmentic activity

Evaluation of Anthelmintic Activity Using Earthworms. Earthworms, each of average length of 6 cm, were placed in Petri dishes containing 2mL of various drug concentrations, 50mg/mL, 100 mg/mL of solutions. Albendazole solution was used as reference standard drug and distilled water as control. The worms were observed for the motility after incubating at 37◦C. This was done after pouring the Petri dishes content in the wash basin and allowing the worms to move freely. By tapping the end of each worm with the index finger and applying a bit of pressure, the worms that were alive showed motility and those dead were non motile. The motile worms were returned to the respective Petri dishes containing drug solutions, and the incubation process was carried out again. In the control, the worms were viable for at least twelve days, which is similar to the findings reported earlier. The time taken for paralysis, motility activity of any sort, and death time of worms were observed and recorded after ascertaining that the worms did not move neither when shaken vigorously nor when dipped in warm water (50◦C) [5].

3. Results and Discussion

3.1 Phytochemical tests

Table 1: Phytochemical tests of *Tectona grandis* Linn

Sr. No.	Main Test	Sub Test	Observation	Inferance
1	Test for Alkaloids	1. Dragendrff's Test	Orange brown ppt	+
		2. Mayer's Test	Creamy ppt	+
		3. Hager's Test	Yellow ppt	+
		4. Wanger's Test	Reddish brown ppt	+
2	Test for carbohydrate	1. Molisch's test	Purple ring at the junction of two liquids	+
		2. Benedict's test	Yellow colour	+
		3. Fehling's test	No brick red ppt	-
3	Test for glycosides	1. Legal's Test	Red colour	+
		2. Borntrager's Test	Pink colour	+
		3. Foam Test	Foam is form	+
4	Test FOR PHENOLIC compounds and tannins	1. With ferric chloride	Deep blue colour	+
		2. With lead acetate	White ppt	+
		3. With potassium dichromate solution	Red ppt	+
5	Test for flavonoids	Shinoda test	Yellow ppt	+

Note: (+ indicates test positive, - indicates test negative)

Preliminary phytochemical screening has shown the presence of alkaloids, glycosides, carbohydrates, tannins, proteins, flavonoids in ethanolic extracts of plants.

3.2 *In vitro* anti –inflammatory activity

Table 2: Effect of EATG on heat induced protein denaturation

Sr. No.	Absorbance of controlled drug	Absorbance of sample
1.	1.996	0.633
2.	1.998	0.632
3.	1.997	0.634
Mean	1.997	0.633

Each value represents the mean ± SD. N=3, Experimental group were compared with control $p<0.05$, considered extremely significant.

EETG: Ethanol Extract of *Tectona grandis* Linn

The percentage inhibition was calculated by using following formula.

$$\% \, Inhibition = \frac{Absorbance\ of\ Controlled\ Drug - Absorbance\ of\ Sample}{Absorbance\ of\ Controlled\ Drug} \times 100$$

= (1.997-0.633/1.997) x 100
=68.3024%.

Protein Denaturation is a process in which proteins lose their tertiary structure and secondary structure by application of external stress or compound, such as strong acid or base, a concentrated inorganic salt, an organic solvent or heat. Most biological proteins lose their biological function when denatured. Denaturation of proteins is a well-documented cause of inflammation. As part of the investigation on the mechanism of the anti-inflammation activity, ability of plant extract to inhibit protein denaturation was studied. It was effective in inhibiting heat induced albumin denaturation. Maximum inhibition of 68% was observed at 200 µg/ml.

Aspirin, a standard anti inflammation drug showed the maximum inhibition 82% at the concentration of 100 µg/ml compared with control (Table 2).

As an inflammation is one of the body's nonspecific internal systems of defence. The response of a tissue to an accidental cut is similar to the response that results from other types of tissue damage, caused by burns due to heat, radiation, bacterial or viral invasion. When tissue cells become injured, they release kinins, prostaglandin and histamine. These work collectively to cause increased vasodilation (widening of blood capillaries) and permeability of the capillaries. This leads to increased blood flow to the injured site. These substances also act as chemical messengers that attract some of the body's natural defence cells a mechanism known as chemo taxis.

3.3 Anti-helmentic activity

Table 3: Antihelmintic activity of ethanolic extract of *Tectona grandis* Linn

Test Substance	Concentration (mg/ml)	Time taken for Paralysis (P) and Death (D) of worms in min	
		P	D
Normal Control	--	--	--
EETG	10	32.18± 0.89*	49.62 ± 1.06*
EETG	25	26.12 ±1.51*	36.00 ±1.14*
EETG	50	19.39 ± 0.83*	30.26 ±1.81*
Standard drug (Albendazole)	10	20.18 ± 0.57	48.39 ± 1.25

All the values are expressed as mean ± SEM (n=5), values are statistically significant at $p < 0.05$, * = $p < 0.05$ when compared with standard drug.

Fig 1: Test drug *Tectona grandis*Ethanolic extract

Fig 2: Standard drug Albendazole

From the results, it is observed that *Tectona grandis* Linn shown potent anthelmintic activity has taken long time for death of worms. *Tectona grandis* Linn was taken 19-32 min to bring paralysis and 30-49 min to bring death of worms. Standard drug is showing paralysis within 20 min and death is 48 min. The test drug shows the potent effect compared to standard drug.

4. Conclusion

In the present investigation we isolate different secondary metabolites from leaves extract of *Tectona grandis*. Steroids, tannins, glycosides, reducing sugar, terpenoids, flavonoids, saponins etc. which are responsible for different pharmacological actions like anti-inflammatory, anti-malarial, anti-helmentic, laxative, anticancer, antipyretic etc. out of them we focused on screening of anti-inflammatory and anti-helmentic properties. The extract fraction serves as free radical inhibitors as a primary oxidant and inhibited the heat induced albumin denaturation, proteinase activity.

This study gives an idea that the compound of plant *Tectona grandis* can be used as a lead compound designing a potent anti-inflammatory drug and this experiment was provided to natural environment to the earthworms and was used for evaluating the effect different doses of the drug on the viability of the preparacytic stages of the helmentics. [8] The wormicidal activity of ethanolic extract suggests that it is effective against parasitic infections of humans. Further, in future it is necessary to identify and isolate the possible active phytoconstituents responsible for the anthelmintic activity and study its pharmacological actions.

5. References

1. Dar RA, Shahnawaz M, Qazi PH. Natural product medicines: A literature update. J Phytopharmacol. 2017; 6(6):349-351.
2. Leelaprakash G, Mohan Dass S. *In vitro* anti-inflammatory activity of extract of *Tectona grandis*. International Journal of Drug Development and Research. 2011; (3):189-196.
3. Majumdar M, Nayeem N, Kamath JV, Asad M. Evaluation of Tectona grandis leaves for wound healing activity. Pakistan journal of pharmaceutical science. Pak J Pharm Sci. 2007; 20(2):120-4.
4. Practical pharmacognosy experiments and techniques by. Khandelwal KR, Nirali Prakashan.
5. Murugamani V, Raju L, Baskar Anand Raj V, Manjir Sarmakataki G. Girija Sankar, The New Method Developed for Evaluation of Anthelmintic Activity by Housefly Worms and Compared with Conventional Earthworm Method. ISRN Pharmacology, vol. 2012, Article ID 709860, 6 pages, 2012. https://doi.org/10.5402/2012/709860.
6. Sushilkumar Varma B, Sapana P. Giri. Study of wound healing activity of *tectona grandis* linn. Anc Sci Life. 2013; 32(4):241-244.
7. Available online https://en.wikipedia.org/wiki/Teak
8. Godghate A, Sawant R. Phytochemical analysis of leaves of *tectona grandis* Linn. International Journal of Pharma and Bio Sciences. 2014; 5(1):355-359.
9. Palanisamy K, Hegde M, Jae-Seon Yi. Teak (*Tectona grandis* linn): A Renowned commercial timber species. Journal of forest science. 2009; 25(1):1-24.
10. Nidavani R, Mahalakshmi AM. Pharmacology of tectona grandis linn. Short review. International Journal of Pharmacognosy and Phytochemical Research. 2014;

6(1):86-90.
11. Aradhana R, RaoKnv, David Banji, Chaithanya RK. A review on tectona grandis linn: chemistry and medicinal uses. Asian Pacific Journal of Tropical Medicine. 2011; 4(8):624-31.
12. Rastogi PR, Meharotra BN. In Compendium of Indian Medicinal Plants. Vol. I, 339; a) (1993) III: 194. PID, CSIR, New Delhi, India, 1990.
13. Mizushima Y, Kobayashi M. Interaction of anti-inflammatory drugs with serum proteins, especially with some biologically active proteins. J of Pharma Pharmacol. 1968; 20:169-173.
14. Sakat S, Juvekar AR, Gambhire MN. *In vitro* antioxidant and anti-inflammatory activity ofmethanol extract of *Oxalis corniculata* Linn. International Journal of Pharma and Pharmacological Sciences. 2010; 2(1):146-155.
15. Available online from https://indiabiodiversity.org/observation/show/1784090.