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Investigation of Invitro Anthelmintic Activity of Tecoma Stans on Pheretima

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ABSTRACT:
Herbal medicine has long been valued for its safety and efficacy. Tecoma stans (L.) Kunth, a member of the Bignoniaceae family, is traditionally used for its anti-diabetic, digestive, and anthelmintic properties. This study investigates its phytochemical, antioxidant, and anthelmintic potential. Ethanolic leaf extracts were prepared using the Soxhlet method and screened for bioactive compounds, confirming the presence of flavonoids, phenols, steroids, and triterpenoids. Albendazole is a commonly used anthelmintic drug known for its effectiveness in treating parasitic worm infections. The anthelmintic activity was assessed in Indian earthworms, comparing the extract's efficacy with Albendazole. The study results confirmed its ability to induce paralysis and death in worms. While the ethanol extract of Tecoma stans also showed anthelmintic properties, Albendazole acted more rapidly, demonstrating greater efficiency. Results demonstrated significant biological activity, supporting Tecoma stans as a promising natural therapeutic agent.

I. INTRODUCTION

1.1 Herbal Medicine

Plants have been the basis of different traditional medicinal systems throughout the world and continue to provide mankind with new remedies. World Health Organization (WHO) defines traditional medicine (including herbal drugs) as therapeutic practices that have been in existence, often for hundreds of years, before the development and spread of modern medicine and are still in use today. Herbal medicine is the synthesis of therapeutic experience of generation of practicing physicians of indigenous systems of medicine. The traditional preparations include medicinal plants, minerals, organic matter etc. Herbal drugs have been in use in Indian, Chinese, Syrian, Roman, Egyptian, Greek texts for thousands of years as per recorded evidences. The Indian texts include Rigveda, Atharvaveda, Charak Samhita and Sushruta Samhita. Folk medicines also play a vital role in healthcare system of ethnic people. Thus herbal medicines have been derived from rich traditions of ancient civilizations and scientific heritage.[1,2,3] Herbal medicines serve as major remedy in traditional system of medicine, even in 21st century these are the primary source of health care system in rural areas and poor countries. According to WHO, about 80% of the world populations still depend on herbal medicines for primary health care. Herbal medicine practices continue still today because of their biomedical superiority over modern medicine.

1.1.1 Importance of herbal medicine

Herbal medicines are used for their safety, efficacy, cultural acceptability and lesser side effects. The chemical constituents present in plants are a part of the physiological functions of living system and hence they are believed to have better compatibility with the human body. These drugs are made from renewable resources of raw materials by eco-friendly processes and will bring economic prosperity.

1.2 Antioxidants:

Anti-oxidants are substances capable to end up free radicals and prevent them from causing cell damage. Free radicals are capable causing a wide number of health problems which include cancer, heart diseases, and gastric problems etc. Antioxidants cause protective effect by neutralizing free radicals, which are toxic byproducts of natural cell metabolism. Foods generally enhance antioxidant levels because foods contain a lot of antioxidant substances.

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Fruits and vegetables are loaded with key antioxidants such as vitamin A, C, E, beta-carotene and important minerals, including selenium and zinc. Herbs also serve as antioxidants. Phytoconstituents are also important source of antioxidant and capable to stop the free radical chain reactions.

Antioxidant molecule react with single free radicals and are capable too neutralize free radicals by donating one of their own electrons, ending the carbon-stealing. A variety of components act against free radicals to neutralize them from both endogenous and exogenous in origin.[12] These include

- Endogenous enzymatic antioxidants.
- Non enzymatic, metabolic and nutrient antioxidants.
- Metal binding proteins like ferritin, lactoferrin, albumin and ceruloplasmin.
- Phytoconstituents and phytonutrients.

The endogenous defense system includes different enzymes like superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx), glutathione reducates (GR), and non enzymatic defense system included vitamin E, vitamin C and reduced glutathione (GSH). Metabolic antioxidants are the endogenous antioxidants, which produced by metabolism\ in the body like lipoid acid, glutathione, L-ariginine, coenzyme Q10, melatonin, uric acid, bilirubin, metal-chelating proteins, transferrin etc., while nutrient antioxidants includes exogenous antioxidants, which cannot be produced in the body but provided through diet or supplement viz. trace metals (selenium, manganese, zinc), flavonoids, omega-3 and omega-6 fatty acids etc. Vitamin E and C are the nonenzymatic antioxidants exist within normal cells as well as they can be supplied through diet. radicals and to treat disorders leads to oxidative stress has proved to be clinically effective and relatively less toxic than the existing drugs. Therefore it is demand of time to uses drugs from plant sources or Phytoconstituents to prevent and / or to treat oxidative stress.

1.3 Tecoma Stans:

The plant Tecoma stans (L.) Kunth belongs to family Bignoniaceae and commonly known as "Piliya" in Rajasthan is a dicotyledonous herb popularly grown for its flowers as an ornamental /garden plant in normal gardens and temples. It is also known as Bignonia stans L. It has wide range of medicinal and pharmacological applications. Almost all parts (leaves, root, flower, seed, fruit, bark) of the plant is reported for its medicinal use. Tecoma stans is a herbal medicine used for treatment of diabetes, digestive problems, control of yeast infections, as powerful diuretic, vermifuge and tonic. Preliminary phytochemical screening of this plant revealed the presence of tannins, flavonoids, alkaloids, quinones and traces of saponins and amino acids. This review supports all updated information on its phytochemical and pharmacological activities and its traditional uses.

Tecoma stans is an ornamental plant is an erect, branched, sparingly hairy or nearly smooth shrub, about 2 to 4 meters in height. Its leaves are opposite, odd-pinnate, and up to 20 cm in length, with 4 to 5 leaflets. Leaflets are lanceolate to oblong lanceolate, 6 to 13 cm long, pointed at both ends, and toothed at the margins. Flowers are yellow, faintly scented, borne in short, dense, terminal clusters. Calyx is green, 5 to 7 mm long and 5-toothed. The capsules are linear, compressed, 15 to 20 cm long, 6 to 8 mm wide, pointed and hanging from the branches. Seeds are numerous, less than 2 cm long, 7 mm wide and furnished with a transparent wing. It is widely distributed in cultivation. Tecoma is a genus of 14 species of shrubs or small trees in the trumpet vine family, Bignoniaceae. Twelve species are from the Americas, while the other two species are African. The American species range from the extreme southern United States through Central America and the Antilles south through Andean South America to northern Argentina. The generic name is derived from the Nahuatl word tecomaxochitl, which was applied by the indigenous peoples of Mexico to plants with tubular flowers. Its chemical constituents are phytosterols, alkaloids, quinines, amino acids, monoterpenes, triterpene, glycosides, phenols, flavonoids, saponins, and tannins. Roots are reported to be diuretic, tonic, anti-syphilitic and vermifuge, decoction of flowers and bark are used for stomach pains, the whole plant is used in the treatment of diabetes. In Guadalajara, roots used for making beer.

1.4 Helminthiasis

Helminthiasis, also known as worm infection, is any macroparasitic disease of humans and other animals in which a part of the body is

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infected with parasitic worms, known as helminths. There are numerous species of these parasites, which are broadly classified into tapeworms, flukes, and roundworms. They often live in the gastrointestinal tract of their hosts, but they may also burrow into other organs, where they induce physiological damage.

Soil-transmitted helminthiasis and schistosomiasis are the most important helminthiases, and are among the neglected tropical diseases.[14]This group of helmianthiases have been targeted under the joint action of the world's leading pharmaceutical companies and non-governmental organizations through a project launched in 2012 called the London Declaration on Neglected Tropical Diseases, which aims to control or eradicate certain neglected tropical diseases by 2020.

Helminthiasis has been found to result in poor birth outcome, poor cognitive development, poor school and work performance, poor socioeconomic development, and poverty.Chronic illness, malnutrition, and anemia are further examples of secondary effects.

Soil-transmitted helminthiases are responsible for parasitic infections in as much as a quarter of the human population worldwide.[19] One well-known example of soil-transmitted helminthiases is ascariasis.

1.4.1 **Symptoms:**

The signs and symptoms of helminthiasis depend on a number of factors including: the site of the infestation within the body; the type of worm involved; the number of worms and their volume; the type of damage the infesting worms cause; and, the immunological response of the body. Where the burden of parasites in the body is light, there may be no symptoms.

Certain worms may cause particular constellations of symptoms. For instance, taeniasis can lead to seizures due to neurocysticercosis.

II. PLANT PROFILE

2.1 Tecoma Stans:

Scientific Name : Tecoma stans

Family **:** Bignoniaceae[64]

Fig:-1.Tecoma stans plant

2.1.1 Taxonomical Classification:

- **Family:–** Bignoniaceae
- **Domain:–** Eukayiota
- **Kingdom:-** Plantae–Plant
- **Subkingdom–Angiosperm**:– Seeds are cover
- **Phyllum-Tracheobionta:–** Vascular plant
- **Subphylum:-** Euphyllophytina
- **Super division:-** Spermatophyta
- **Division:–** Magnoliophyta(Eudicots)
- **Class:–** Magnoliopsida-Dicotyledons
- **Subclass:-** Asteridae
- **Order:–** Scrophularials
- **Genus:-** Tecoma Juss
- **Species :-** Tecoma stans (L) Juss.
- **Kunth:–** Yellow trumpetbush.

2.1.3 Synonyms:

Ginger-thomas, Yellow trumpet/ Yellow bells/ Yellow-elder.

2.1.4 Vernacular Name:

- Telugu– Pachagotla
- Hindi – Piliya/ Pila kaner
- English– Yellowbells
- Kannada– Koranekelar
- Tamil– Sonnapatti

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- Bengali– Chandaprabha
- Marathi– Ghanti ful[65].

2.1.5 Parts used :
Stem, Flowers, Leaves, Bark.

2.1.7 Chemical constituents:
Therapeutically important active principle of Tecoma stan is Tecomine (the alkaloids isolated from the plant harvested in Egypt) was shown to be one of the compounds responsible for the hypoglycemic action given the interest in substances able to treat type II diabetes. The two other alkaloids isolated, namely 5β-Hydroxyskitanthine, early called Base C, and Boschniakine were inactive both in vivo and in vitro assays. Other chemical constitutes are phytosterols, alkaloids, quinones, amino acids, monoterpenes, triterpene , glycosides, phenols, flavonoids, saponins, and tannins.

2.1.8 Medicinal action:
Diabetes, digestive problems ,control of yeast infections ,as powerfuldiuretic, vermifuge and tonic.

2.1.9 Traditional Uses:
Almost all the parts of Tecoma stans are of medicinal importance and used traditionally for the treatment of various ailments. South America and Latin America used traditionally for reducing blood glucose. The Tecoma stans leaves, and roots have been used for a variety of purposes in herbal medicine. Bark shows smooth muscle relaxant, mild cardiotonic and chloretic activity.

III. REVIEW OF LITERATURE

3.1 Earlier work done on tecoma stans.

- Aguilar-santhamaria L,et.al evaluated invivo and invitro intestinal alpha-glucodiases inhibition as possible as model of diabetes type-.
- S.Kameswaran et.al. reported ethanolic extract of flower of tecoma stans on acetic acid induced colitis in albino rats.
- Shanmukha ittagi et.al. reported cardioeffective effect of hydroalcoholic extracts of tecoma stans flowers against isoproterenol induced myocardial infraction in rats[71].
- Ramachandran rajamuragan et.al. investigated on antimicrobial activity,antioxidant ,phytochemical of ethanol extract of tecomas stans.

IV. AIM AND OBJECTIVE

- Research on free radicals and to find new antioxidant phytochemicals have arouse significant interest among scientist in past decade ,their broad range of effect in biological system have drawn attention of many researches. There are extensive evidences to implicte free radicals in the development of chronic and acute diseases such as cancer ,cardiovascular diseases, diabetes, damage caused by reactive oxygen or nitrogen species leads to the pathological changes which are associated with many diseases. Many synthetic antioxidants such as butylated hydroxyl amisole [BHA] are very effective and commonly used industrial processing but responsible for potential health risk such as carcinogenicity, inflammations.
- Tecoma stans is a medicinal plant throught out the world and widely used treat diuretics, antisyphilitics, antibacterial, antispasmodic, woundhealing. anticancer, diabeties[type-2].Therefore ethanol extracts of Tecoma stans leaves, barks, roots have been used for a variety of purposes in herbal medicines. Bark shows smooth muscle relaxant, mild cardiotonic and chloretic activity. The ethanol, acetone and ethyl acetate extracts of tecoma stans. were assessed for diuretics activity. In addition, measured the antioxidant activity and determined content of total phenolic and flavonoids in the extract in order to correlate them with the assayed activities.
- Thus in the current research the ethanol extract of the plant was investigated for the anthelmintic activity.

V. PLAN OF WORK

- Preparation of ethanol extract of Tecoma stans.
- Preliminary Phytochemical screening
- In-vitro antioxidant studies of Hydrogen peroxide radical scavenging method of ethanol extract of Tecoma stans.
- Antihelmentic activity of ethanolic extract of Tecoma stans.

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VI. MATERIALS AND METHODS

6.1. Collection of Plant Material:

Tecoma stans.leaves were procured in February 2025, from Minjur locality in Chennai.

6.3. Chemicals and Reagents:

Ethanol, Hydrogen peroxide(H_2O2), sodium hydroxide (NaOH), $FeCl_3$solution, conc.H_2SO_4, conc.HCL, sulfur powder, Zinc dust, picric acid, KOH,

6.4. Instruments Used:

- Hot air oven (ASHOK UNITED SCIENTIFIC COMPANY)
- Hot plate (Aarson industries)
- Precision weighing balance (Contech 0.1mg precision)
- Sonicator (LC)
- Weighing balance.[WENSAR]

6.5. Preparation of plant extracts:

Leaves of Tecoma stans. were shade dried under room temperature for one week and leaves were powdered. The finely powdered leaves were kept separately in an air tight container until the time of use. About 50 gms of finely powdered leaves were extracted with ethanol using Soxhlet apparatus and finally solvent was evaporated and concentrated by using distillation apparatus. These leaf extract were used for phytochemical screening, antioxidant and anti-helmenthetic activities

6.6. Qualitative phytochemical screening of ethanol extract of tecoma stans.

Ethanol extract of Tecoma stans. were screened for their chemical constituents. Phytochemical screening was done as explained in literature.[99,100,101]A small amount of dried extract was used to determine the alkaloids, carbohydrates, phenols, flavonoids, steroids and triterpenoids using the following methods

6.7.5 Procedure:

1. To the 4 ml of various concentrations of sample and standard solutions add 0.6 ml of 40mM H_2O_2 (0.136mg of 30% H_2O_2) in triplicate manner i.e. each concentration in 3 series of test tubes.
2. An equal amount of sample and phosphate buffer of pH 7.4 and H_2O_2 were mixed and was used as control.
3. Ascorbic acid was used as standard for comparison.
4. After incubation for 10 min. in dark, absorbance was recorded at 230nm.
5. % scavenging activity was calculated using the formula.

$$I\% = \left[\left(\frac{A_C - A_S}{A_C}\right)\right] \times 100$$

Where

A_C**:**Absorbance control

A_S**:** Absorbance standard/extract.

6. The effective concentration of sample required to scavenge H_2O_2 radical by 50% (IC_{50} value) was obtained by linear regression analysis of dose-response curve plotting between % inhibition on Y-axis and concentration on X-axis.

6.8. In- vivo anti-helmenthetic activity

6.8.1 Experimental Animals:

Experiments were performed Earth Warms (150-250 gms). The Earth warms were housed for one day before starting with 9:00 AM to 12:00 PM the experiment. The Earth warms are taken in three plate lets 1.Test 2.Standard 3.control

1.TEST:- Plant extract, normal saline
2.STANDARD:- normal saline+Albendazole
3.CONTROL;- saline only

6.8.2 .Procedure for Anthelmintic Activity

The anthelmintic activity was evaluated on adult Indian earthworms by the reported methods with slight modification. Three groups of approximately equal sized Indian earthworms consisting of six earthworms in each group were released in to 10 ml of each sample as follows: vehicle (1% gum acacia in normal saline), Albendazole (10 mg/ml) and, normal saline ethanol extracts (10 mg/ml) of Tecoma stans. were used. Normal saline was used as control. Observations were made for the time taken to paralysis and/or death of individual worms. Paralysis was said to occur when the worms do not revive even in normal saline water. Death was concluded when the worms lose their motility followed with fading away of their body color.

6.9 Statistical analysis

The data on biological studies were reported as mean ± Standard deviation (n = 6). For determining the statistical significance, standard error mean

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VII. RESULTS AND DISCUSSION

7.1. Physical characteristics of extracts:

The Ethanol extract of Tecoma stans.was thick Yellow color, sticky in nature and the percentage yield of the extract was found to be 35.66% w/w.

7.2. Preliminary phytochemical screening of extracts:

Qualitative phytochemical screening was carried out using several tests and results revealed that Ethanol extract of Tecoma stans. contains phenols, flavonoids and absence of alkaloids, carbohydrates and steroids & triterpenoids.

Table 7:Qualitative phytochemical screening of Ethanol extract of Tecoma stans:-

Sl.No.	Test	Ethanol
1.	Carbohydrates	+
2.	Alkaloids	-
3.	Glycosides	+
4.	Tannins	+
5.	Steroids	+
6.	Triterpenoids	+
7.	Volatile oils	-
8.	Fats and fixed oils	-
9.	Flavanoids	+
10.	Polyphenols	+
11.	Saponins	+

Ethanol extract of Tecoma stans leafs contains phenols, flavonoids, steroids, triterpenoids and absence of alkaloids.

7.3. Hydrogen peroxide scavenging activity of extracts:

Table 8: H_2O_2 radical scavenging activity of Ethanol extracts of Tecoma stans.

S. No	Sample	Concentration	Absorbance (Mean±SEM)	Percentage inhibition (Mean±SEM)	IC_{50}
1	Ethanol extract	20 µg/ml	0.049±0.002	64.41±1.623	49 µg/ml
		40 µg/ml	0.059±0.001	71.16±0.710	
		60 µg/ml	0.066±0.003	74.84±7.110	
		80 µg/ml	0.098±0.002	77.09±1.781	
		100 µg/ml	0.116±0.002	82.72±0.707	
2	Ascorbic acid	20 µg/ml	0.053±0.001	44.60±0.566	53 µg/ml
		40 µg/ml	0.061±0.001	52.94±0.566	
		60 µg/ml	0.075±0.004	72.38±1.928	
		80 µg/ml	0.096±0.003	76.15±1.278	
		100 µg/ml	0.181±0.002	81.86±1.020	

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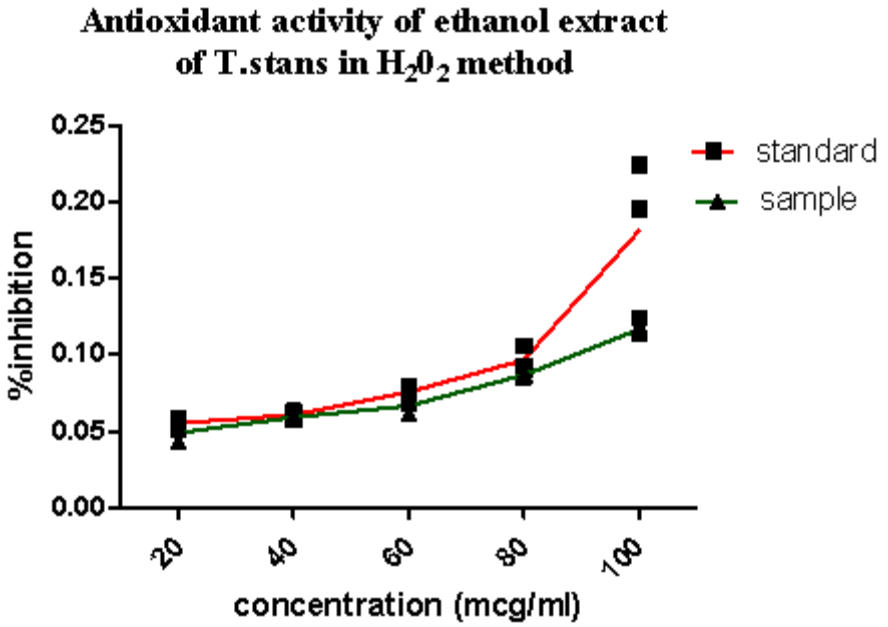

Figure 3: H_2O_2 scavenging activity of Ethanol, extracts of Tecoma stans.
Values are expressed as the Mean±SEM, n=3**.**

The extracts was able to neutralize H_2O_2in a concentration dependent manner at a concentration range of 20-100µg/ml. Ethanol (IC_{50}=49 µg/ml) extract shows better antioxidant activity than the standard ascorbic acid (IC_{50}=53 µg/ml) and the percentage scavenging activity of ethanol and ascorbic acid values are 82.72% and 81.86% respectively. Ethanol extracts IC_{50}values was 49µg/ml and 53µg/ml with percentage inhibition values of 72.70% and 63.72% respectively.

7.4 ANTHELMINTIC ACTIVITY:

Table 9: Anthelmintic Activity of Ethanol Extracts of Tecoma stans

slno	Group	Time of Paralysis	Time of death
1	Extract (10mg/ml)	101.33±34.21	161.66±21.13
2	Standard (10mg/ml)	70.66±16.36	101.16±18.22

Values were represented as Mean±SEM (n=6)

The antihelmintic activity of Tecoma stans was tested on Pheretima posthuma by taking standard drug as Albendazole and control as normal saline and the paralysis time and death time was noted.

The results indicate that the ethanol extract of Tecoma stans at a concentration of 10mg/ml caused paralysis in the worms after an average time of 101.33 minutes, with subsequent death occurring at an average time of 161.66 minutes. Comparatively, albendazole, the standard anthelmintic agent, exhibited a shorter time to paralysis (70.66 minutes) and death (101.16 minutes) under the same conditions.

Albendazole is a well-known anthelmintic medication commonly used to treat parasitic worm infections. Its efficacy in causing paralysis and death in the worms was evident from the results. Although the ethanol extract of Tecoma stans demonstrated anthelmintic activity, albendazole exhibited a faster onset of action.

These findings suggest that the ethanol extract of Tecoma stans holds promise as an anthelmintic agent. Further studies are warranted to explore its potential as an alternative or adjunct therapy for parasitic worm infections and to elucidate its mechanism of action compared to standard drugs like albendazole.

International Journal of Pharmaceutical Research and Applications
Volume 10, Issue 2 Mar – Apr 2025, pp: 170-178 www.ijprajournal.com ISSN: 2456-4494

Extract treated group

Standard drug treated group

Figure 4: Petri plates with earthworms treated with Tecoma stans leaf extract

VIII. CONCLUSION

- The present work revealed that the extracts of leafs of Tecoma stans .by phytochemical screening of ethanol extracts contains flavonoids and phenols and steroids & triterpenoids and it may be possess significant antioxidant activity.
- Ethanol extracts exhibit antihelmanthitics activity at the dose of 500mg/kg when compared to standard which may be due to the presence of high phenolic content and high flavonoid content and other constituents.
- Present study also indicates that the possible antioxidant mechanism of the extract can be due to hydrogen or electron donating and

direct free radical scavenging activity of the extracts, but exact antioxidant mechanism and identification of antioxidant Phytoconstituents should be further studied.

The results justified the use of leaf extracts in several cardio tonic, anti-inflammatory, skin diseases, antioxidant and antihelmanthitics diseases traditionally. We suggest that the leafs of the plant can be viewed as the potential source of natural antioxidant and anti-helmanthitic can afford precious functional components

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