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Effect of *Lawsonia inermis* L. Leaves Extract on In-Vitro Phyto-Nematicidal Activity on *Meloidogyne incognita Race-II*

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ABSTRACT

Medicinal plants are represent the mostly old days form of medication, used for thousands of years in traditional medicine in many countries around the world. The empirical knowledge are transmitted over the centuries within human communities for about their beneficial effects. Lawsonia inermis Linn. is commonly known as henna, which is recognized in traditional use of medicine. It consists of various types of phytoconstiuents like flavonoids, coumarins, triterpenoids, steroids, xanthones. It is mostly utilised for cosmetic purpose only but it also have various use as like as headache, bronchitis, syphilitis, amenorrhea, diseases of the spleen, dysuria, bleeding disorder, skin diseases, diuretic, antibacterial, antifungal, anti-amoebiasis, astringent, anti-hemorrhagic, hypotensive effect. With having these all benefits, this plant is not widely utilized. This study gives a view mainly on the nematicidal activity of L. inermis on okra plant.

Keywords: Henna; Taxonomy; Phytochemistry, Traditional uses

INTRODUCTION

India has a rich heritage of knowledge on plant based drugs both for use in preventive and curative medicine (*Nair and Chandra,2005*). The earliest mention of medicinal use of plants in Hindu culture is founds in "Rig veda", to have been written between 4500-1600 B.C. and supposed to be the oldest repository of human knowledge (*Rastogi and Mehrotra,2002*). According to the world health organization, 2003 about 80% of the population of developing countries being unable to afford pharmaceutical drugs rely on traditional medicines, mainly plant based, to sustain their primary health care needs (*Goyal, B.R. et.al;2008*). Herbal medicines are in great demand in the developed as well as developing countries for primary healthcare because of their wide biological and medicinal activities, higher safety margins and lesser costs (*Cragg ,1997 and Padma ,2005*). A plant used all over the world. This plant is commonly known as henna or mhendi and abundantly available in tropical and subtropical areas. Ancient history of india describes its diverse uses and also plays appreciable role in ayurvedic or natural herbal medicines (*Lavhate and Mishra ,2007*).

Henna Lawsonia inermis L., belongs to lythraceae, also known as the loosestrife family. The molecular formula and weight of Lawsone is C10H6O3 and 174.15, respectively. Henna is cultivated by many farmers for cosmetic and pharmaceutical purposes, it belongs to the group of plants that are popular in nature and all parts of the plant (root, stem, leaf, flower pod and seed) are of great medicinal important (*Abdelraouf et al. 2011*). The most important part of the plant is the leaf which contained a coloring compound called Lawson. The Lawson is a red orange dye molecule, also known as hennotannic acid (*Gibbons et al. 2004, Awek and Tapapul 2005, Emin and Mehmet 2012*).

Henna has been used cosmetically and medicinally for over around 9,000 years. Traditionally in India, mehndi is applied to hands and feet in the form leaf paste or powder is widely used for decorating hands, nails and feet with patterns. Also used as hair dye because it nourishes to become healthy hair. Henna symbolizes fertility. Its use became popular in India because of its cooling effect in the hot Indian summers. Henna plants all body parts are i.e. leaves, flowers, seeds, stem bark and roots are used in traditional medicine to treat a variety of ailments as rheumatoid arthritis, headache, ulcers, diarrheoa, leprosy, fever, leucorrhoea, diabetes, cardiac disease, hepatoprotective and colouring agent. (Chetty, 2008 and Chopra et al;1956 Reddy, 1988).It is used for alleviating

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jaundice, skin diseases, venereal diseases, smallpox and spermatorrhoea. Flowers are very fragrant and used to extract a perfume, which is used as base for local scents. An infusion of the flowers is a valuable application to bruises. Decoction of the flowers is describes as an emmenagogue. Seeds are deodorant. Powered seeds with real ghee (clarified butter) are effective against dysentery. Seeds in powered form are good medicine for liver disorders and associated problems. The bark is applied in the form of a decoction to burns and scalds. It is given internally in a variety of affections, such as jaundice, enlargement of the spleen, calculus, as an alternative in leprosy and obstinate skin affections. Root is considered as a potent medicine for gonorrhoea and herpes infection. Root is astringent may be pulped and used for sore eyes. Pulped root may also be applied to the heads of children for boils. Cambodians drink a decoction as a diuretic. Decoction of the root generally in combination with prepared indigo as a powerful abortifacient. The root is supposed to be useful in treatment of hysteria and nervous disorder. (Chetty, 2008 and Chopra et al;1956 and Reddy, 1988).

TAXONOMY

Lawsonia inermis L. (Lytheraceae) is a perennial shrub commonly called Henna (English), Mehndi (Urdu & hindi). It is 2-2.5 m tall shrub, its foliage resembles to that of Pomegranate tree (Hannan, 1997). Dicot, shrub. Stems erect, brown, glabrous. Leaves simple, glabrous, sessile, opposite, lanceolate, margin entire, apex acuminate. Inflorescence cymose. Flowers

white, hermaphrodite, actinomorphic, sepals (4), united, valvate, petals (4), free, twisted, stamens (8), united, found in pairs, epipetalous, ovary superior. Fruits brown capsules. It is used to dye fabrics like (silk, wool and leather). For this reason, henna should be free from any contaminants such as pesticide residues; and disinfectants used for preventing the spread of pests and bacteria. When concentrations of pesticide residues exceed the permissible level, the risk to human health comes from direct or indirect exposure to pesticide residues in primary or derived agricultural products as it could lead to several potential toxic effects, carcinogenic and biological effects. (Nagham Said, 2020). Its Seeds have hard and thick coat its colour is brownish (Makhija et al. 2011).

Classification:-

Kingdom: Plantae.

Division: Magnoliophyta.

Class: Magnoliopsida.

Order: Meyrtales.

Family: Lythraceae.

Genus: Lawsonia.

Species: Lawsonia inermis L.

Table 1. Medicinal use and local use of Henna

A) Medicinal Uses	B) Local uses
1) Skin diseases..	1) Skin dye
2) Fever.	2) Hair dye
3) Smallpox.	3) Leather dye
4) Headache..	4) Wool dye

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5) Burns.	5) Textile industry
6) Leprosy.	
7) Leishmania.	
8) Dysentery.	

MATERIAL AND METHODOLOGY

Soil Samples:

Okra soil samples were used for collection of M. incognita race II from Nematology Field MPKV Rahuri.

Sieves:

20, 60, 200 and 350 mesh sizes sieves were used for washing and filtration of soil sample and roots of rhizospheres.

Chemicals:

Alcohol grades, xylene, glycerine, chemical Nematicides, the micro plots Methanol, hexane, acetone, Formalin, 2% Dimethyl Sulphoxide (DMSO), D.W, Neem cake, Whatman filter paper No.1, Carbofuron, nailpolish, Silica gel 60-120 mesh, Silica gel column chromatography, etc are used for present study. All the research work was carried out in Department of Zoology, D. B. F. Dayanand College of Arts and Science Solapur (M.S.), India. A survey of soil and root system of vegetable crop of Okra plants for disease incidence and for the occurrence from farmers fields around the Akkalkot regions.

Methods

Collection of nematode from infected soil sample Soil sample from infected fields of okra of 1 kg soil collected out of 200 cc soils was washed thoroughly and processed using Cobb's sieving and Decanting method. Followed by modified Baermann's funnel methods. It was used to study seasonal incidence, prevalence of M. incognita race II.

Extraction of nematodes

Pure cultures of okra are continuously grown in micro plots. Whenever nematode is required it is taken the soil from root zone region and processed by Cobb's Sieving and Decanting method. Identification of root-knot nematode species *Meloidogyne incognita race II* Species was identified on the basis of perennial pattern method. The nematode M. incognita race II was identified by Dr. Pallavi Pallande madam and Vinod Pawar sir Plant Nematologist Nematology section Department of Agricultural Entomology, Mahatma Phule Krishi Vidypeeth Rahuri, Dist. Ahmednagar, (M.S.) India.

Preparation of crude extracts

The fresh collected plants parts were washed thoroughly with running tap water and then rinsed with distilled water. Washed plant material was shade dried for 5 to 7 days. It cut into small pieces and pulverized in a mechanical blender. The dried plant materials were fine powdered with the help of electric grinder and preserved separately for further use.

Aqueous methanol extract preparation

Extracts were prepared in cold extraction methods described by (Gilani et al., 2004). Powered plant material were soaked in methanol solution in the round bottom flask. 1 kg of dried powdered plant materials were soaked in 1500 ml of methanol solution. It was kept at room temperature for about 4 to 5 days after which the filter was

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collected in a large Petri dish with the help of Whatman filter paper No.1 the obtained filter was let for evaporation.

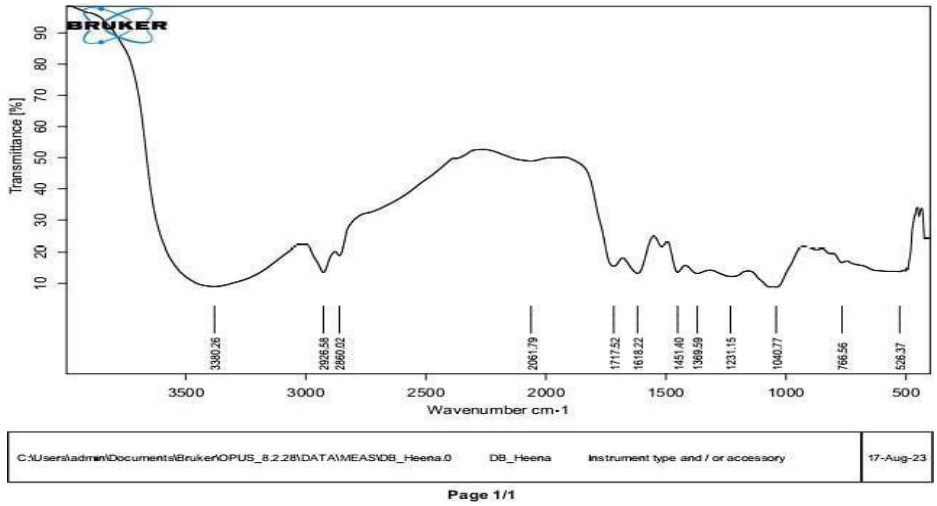
Fractions separated by Column chromatography

Extracts obtained were mixed with silica gel (60-120 mesh) and eluted by using Petroleum ether and methanol. By using column chromatography techniques different fractions A, B, C, D, E and F were collected. Eluted bioactive fractions were further used for in- vitro antinematicidal activity.

In-vitro phyto-nematicidal testing method

In-vitro phyto-nematicidal testing method here made total 4 group, in each petridish contains about 100 second stage juvenile larvae one group standard (Control), with distilled water, second group with standard (control), Nimitz chemical as nematicides, Third group control standard (control), plants bio pesticide Neem cake and fourth group with different extracts of medicinal plants. The extracts were diluted with 2% DMSO. Time of immobile phyto-nematodes was noted in 24 hours and 48 hour time intervals.

The IR spectra were measured in CHCI3 and 1H-NMR spectra were for solutions in CCI3.



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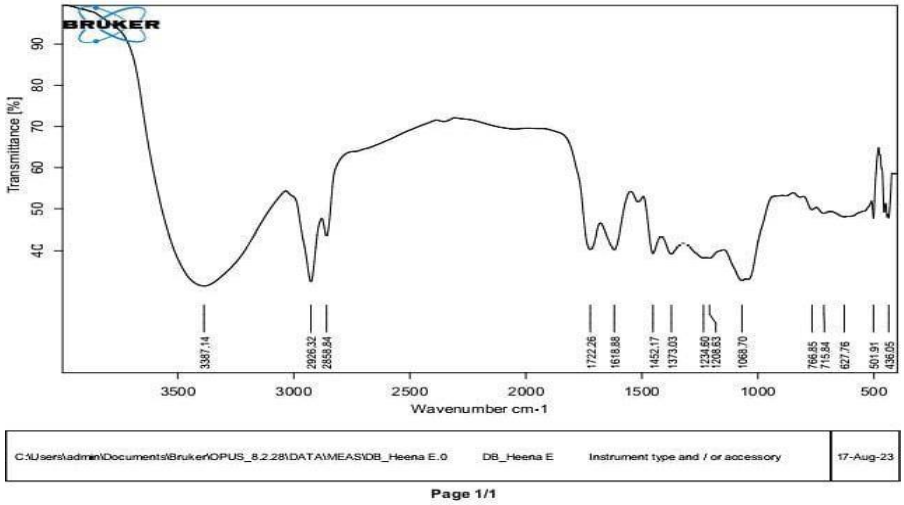

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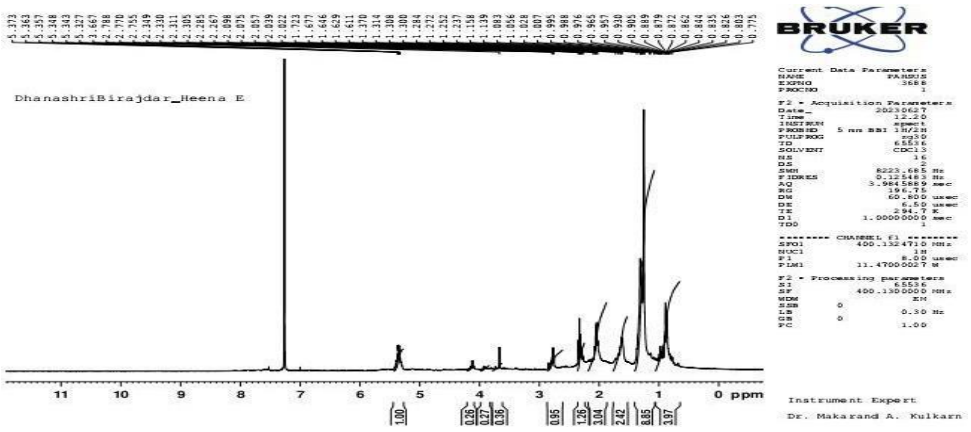

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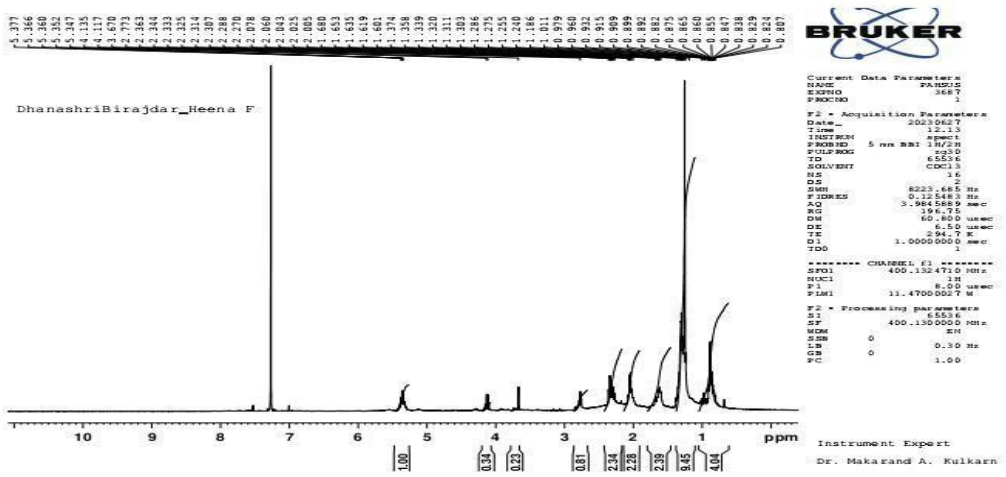

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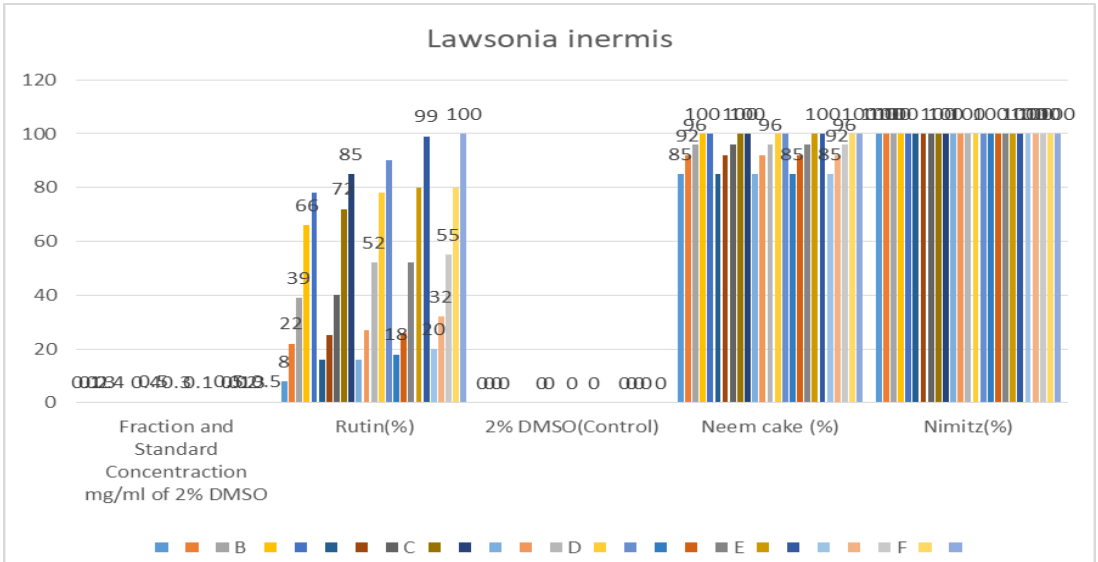
RESULT

The isolated *Lawsonia inermis* compounds were tested in vitro against root-knot nematodes isolated from roots of okra plant (Cobb, 1918). Lawsone shows activity of Nematicidal impact on M. incognita Race-II. The details given in following table.



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As per data here would be Lawsonia inermis concentraction impact on M. incognita was 0.3 con. showed 50% mortality of nematodes, 0.4 con. showed 75% mortality of nematodes, 0.5 showed 99% mortality of nematodes. Natural compound had impact on *in-vitro* mortality of M.incognita. Utilize the natural pesticides rather than the chemical pesticides. Also gives the beneficial impact on soil which improves soil fertility and provide good yield quality of okra.

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