



Preliminary Phytochemical Screening and Antibacterial Studies of Leaf Extract from *Argemone mexicana* Linn.

S. Komathi*, J. Mohamed Nasrudheen¹, P. Saundharya¹, M. Hibana¹ and K. Kavimani¹

*Associate professor – PG and Research Department of Biotechnology, Hindustan College of arts and science (Autonomous), Coimbatore, Tamilnadu, India.

¹III-B.Sc., Biotechnology, Hindustan College of Arts and Science (Autonomous), Coimbatore, Tamilnadu, India.

Received: 25 Feb 2019 / Accepted: 20 Mar 2019 / Published online: 01 Apr 2019

*Corresponding Author Email: komathipolur@gmail.com

Abstract

The aim of the present study was to evaluate phytochemical constituents, antibacterial activities of methanol and chloroform leaf extract of *Argemone mexicana* Linn. The extracts were subjected to qualitative tests by screening of phytochemicals that present in the extracts and to quantitative tests by evaluation of antibacterial activity using well agar diffusion method. The evaluation of antibacterial activity were done against standard bacterial strains (*Klebsiella pneumonia* and *Pseudomonas aeruginosa*) respectively. Phytochemical screening showed the presence of alkaloids, flavonoids, tannins, phenolics and Saponins.

Keywords:

Argemone mexicana Linn, Extraction, Antibacterial, Phytochemical.

INTRODUCTION:

Medicinal Plants have been identified and used throughout the human history. Plants have the ability to synthesize a wide variety of chemical compounds that are used to perform important biological functions, and to defend against some pathogens such as bacteria and fungi. It is also effective as an insecticide. At least 12,000 such compounds have been isolated so far, a number estimated to be less than 10% of the total. Chemically compounds in plants mediate their effect on the human body through processes identical to those already well understood for the chemical compounds in conventional drugs, thus

herbal medicines do not differ greatly from conventional drugs in terms of how they work. This enables herbal medicines to be effective as conventional medicines. The use of plants as medicines have been mentioned in traditional medicines. Ethnobotany (the study of traditional human uses of plants) is recognized as an effective way to discover future medicines. In 2001, researchers identified 122 compounds used in modern medicine which were derived from "ethnomedical" plant source, 80% of these have had an ethnomedical use identical or related to the current use of active elements of the plant. Many of the pharmaceuticals currently available

to physicians have a long history of use as herbal remedies, including aspirin, digitalis, quinine and opium.^[1]

India is one of the leading countries in Asia in terms of the wealth of traditional knowledge system related to the use of plant species. India is also known to harbor a rich diversity of higher plant species about 17,000 species of which 7500 are known as medicinal plants. Such a huge number of medicinal plant species has allowed the evolution of many systems of herbal medicines. Ayurveda is arguably the oldest medicinal system in Indian subcontinent.

Argemone mexicana L. is a species of poppy found in Mexico and now has widely naturalized in the United States, Ethiopia, India and Bangladesh. In Mexico, the seeds are considered as an antidote to snake venom. In India, the smoke of the seeds is used to relieve toothache. The fresh yellow, milky seed extract contains protein-dissolving substances, effective in the treatment of warts, cold sores, cutaneous infections, skin diseases, itches and also dropsy and jaundice. *A. mexicana* L. is widely distributed throughout Bangladesh.

The whole plant is analgesic, antispasmodic, possibly hallucinogenic and sedative. It contains alkaloids similar to those in the opium poppy (*Papaver somniferum*) and so can be used as a mild painkiller. Owing to its various ethnopharmacological properties, the present investigation was undertaken to evaluate the antibacterial potential of *A. mexicana* Linn stem extracts against a range of food-borne pathogenic bacteria with the possible use as a natural antimicrobial agent in pharmaceutical or food industries.

MATERIALS AND METHODS:

Collection of Plant Materials and Identification

Fresh leaves of *Argemone mexicana* Linn. were used in this study, which were collected from the back side of Hindusthan college of Arts and Science, Coimbatore, Tamil Nadu - 641028.

The collected plant material was identified as *Argemone mexicana* Linn. (Papaveraceae) by Scientist 'E', Botanical Survey of India, Southern Regional Centre, Coimbatore, Tamil Nadu – 6410031

Preparation of Plant Extracts

The fresh leaves of *Argemone mexicana* Linn. Were collected and washed with running tap water followed by distilled water. About 10g of leaves was grind to a

fine paste in 30 ml of solvents such as Methanol and Ethanol. About 10g of leaves were grind to a fine paste in 60ml of chloroform, then transfer the content in the fresh centrifuge tube and allow to centrifuge at 5000 rpm for 2 minutes. After centrifugation the aqueous solution was transferred into a clean and dry tube or beakers then covered with aluminum foil. The solutions were left to stand at room temperature for 24 hours. The filtrate was used for antimicrobial test.

Antibacterial activity

Preparation of the medium

Antibacterial activities of plant extract were tested by agar diffusion method. The culture plates were prepared by pouring 20 ml of Muller Hinton Agar (MHA) medium into sterile petri plates. The inoculum suspension was spread uniformly over the agar medium using sterile cotton swabs to get uniform distribution of bacteria. Wells (6mm in diameter) were punched in the agar.

Different leaf extracts of *Argemone mexicana* Linn. which were Methanol and Chloroform with different concentrations such as 25, 50, 75, 100 µl were used. The test microorganisms: *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* were used for the antibacterial studies.

Agar Well Diffusion method

The antibacterial effect of Methanol and chloroform extracts of *Argemone mexicana* Linn. leaves on three bacterial organisms (*Staphylococcus aureus*, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa*) were determined using agar well diffusion method. The Muller Hinton Agar (MHA) medium (Hi-Media, Mumbai) which was already swapped with the test organism was incubated at 37°C for 16 - 18 hours. The zone of inhibition in mm was measured and the results were given in the table.

Phytochemical Analysis of Leaves extract From *Argemone Mexicana*

Phytochemical tests were performed to identify the active chemical compounds such as Alkaloids, Flavonoids, Tannins, Saponins and phenolic compounds in different extracts such as Methanol and Chloroform by following procedure.^[4]

Reagents Required

Mayer's reagent, Ferric chloride, 5% NaOH, 10% HCl, lead acetate solution, Distilled water.

Preparation of Reagents

Preparation of Ferric chloride solution

5g of ferric chloride was dissolved in 100 ml of Distilled water.

Preparation of Lead acetate solution

10g of Lead acetate powder was dissolved in 100 ml of Distilled water.

Preparation of 5% NaOH

5g of NaOH was dissolved in 100 ml of Distilled water.

Preparation of 10% HCl

10 ml of concentrated HCl was diluted with 90 ml of Distilled water

Procedure

Test for Alkaloid (Mayer's test)

Take 2 ml of extract in a clean and dry test tube. Add two drops of Mayer's Reagent by the side of the test tube. A white or creamy precipitate indicates the test positive.

Test for Flavonoids (10% HCl, 5% NaOH test)

Take 1 ml of extract in a clean and dry test tube. Add 1 ml of 10% HCl and 5% NaOH into the test tube. The yellow yellow color solution solution turns into colorless indicates the test positive.

Test for Saponins (Foam test)

1 ml of extract was diluted with the same amount of distilled water. The suspension was shaken well. A 2 cm layer of Foam indicates the test positive.

Test for Tannins (Ferric chloride test)

Take 1 ml of extract in clean and dry test tubes. Add 2 drops of neutral 5% Ferric chloride solution. A dark green colour indicates the test positive.

Test for Phenolic Compounds (Lead acetate test)

1 ml of extract in a clean and dry test tube. Add 2 drops of lead acetate solution. Formation of white precipitate indicates the test positive.

RESULTS AND DISCUSSION:

Antibacterial Activity

The antibacterial effect of three extracts like Methanol and Chloroform of *Argemone mexicana* linn leaves on three microorganisms like *Staphylococcus aureus*, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* were determined using agar well diffusion methods.

The zone of inhibition representing the antimicrobial activity for all the solvents of *Argemone Mexicana* linn are represented in the Fig: 1-3 and Table: 1-2.

Table 1: Antibacterial activity of Methanol extract of *Argemone mexicana* Linn (Fresh leaf)

S.NO	MICROORGANISMS	ZONE OF INHIBITION (mm)			
		25µl	50µl	75µl	100µl
1	<i>Pseudomonas aeruginosa</i>	11	13	15	16
2	<i>Klebsiella pneumoniae</i>	9	13	15	18

Pseudomonas aeruginosa (Methanol Extract)

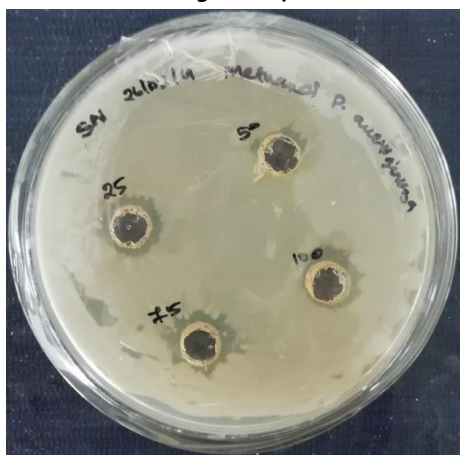


Figure1: *Klebsiella pneumonia* (Methanol Extract)

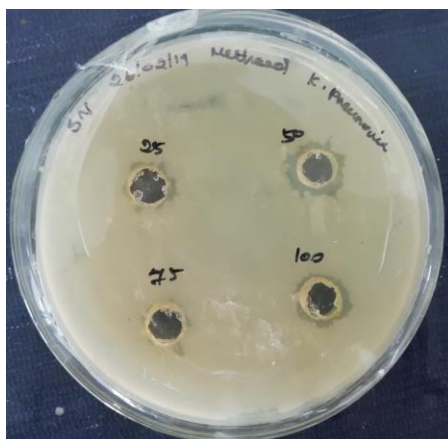


Figure 2

Table 2: Antibacterial activity of Chloroform extract of *Argemone mexicana* Linn (Fresh leaf)

S.NO	MICROORGANISMS	ZONE OF INHIBITION (mm)			
		25 µl	50 µl	75 µl	100 µl
1	<i>Klebsiella pneumoniae</i>	1	9	12	15

Klebsiella pneumonia (Chloroform extract)



Figure 3

Phytochemical Analysis:

The phytochemical screening of *Argemone mexicana* Linn. revealed the presence of various bioactive secondary metabolites which might be responsible for this medical attribute. Results showed five phytochemicals screened were present. They are Alkaloids, Flavonoids, Saponins, Tannins and Phenolic

compounds. In all, more Phytochemicals that is Alkaloids, Flavonoids, Tannins and phenolic compounds were found present in Methanol. Chloroform extract showed the presence of three Phytochemicals such as Flavonoids, Saponins and Phenolic compounds. The results were shown in Table: 3 & 4 and Figure: 4 & 5.

Table 3: Phytochemical analysis of Methanol extract of *Argemone mexicana* Linn (Fresh leaf)

Phytochemicals	Methanol extracts
Alkaloids	+
flavonoids	+
Saponins	-
Tannins	+
Phenolic compounds	+

+ = Present; - = Absence

Phytochemical analysis of Methanol extract



Figure 4

Table 4: Phytochemical analysis of Chloroform extract of *Argemone mexicana* Linn (Fresh leaf)

Phytochemicals	Chloroform extracts
Alkaloids	-
Flavonoids	+
Saponins	+
Tannins	-
Phenolic Compounds	+

+ = Present; - = Absence

Phytochemical Analysis of Ethanol Extract



Figure 5

CONCLUSION:

Finally, it can be concluded from the study that the leaves of *Argemone mexicana* Linn has potent antibacterial and Pharmacological activity. Further studies can be carried out to separate potential pharmacological compounds using column chromatography, HPLC and NMR spectroscopy. From the above results, the presence of the identified Phytochemicals makes the leaf Pharmacologically active. The medicinal values of the plant leaves may be related to their constituent phytochemicals.

ACKNOWLEDGEMENT

We would like to express my deepest sense of thanks and gratitude to our guide Dr. S. Komathi, M.Sc.,M.Phil.,Ph.D.,PGDBI., Associate Professor, Department of Biotechnology, Hindustan College of Arts and Science (Autonomous) Coimbatore. For her sagacious guidance, great encouragement and useful suggestion through the period of our work.

REFERENCE

1. Ayesha Husna S., Dr. V. JayasankarReddy "A review on *Argemone mexicana*". International journal of pharmacological research,7(9) : 170-174. .(2017).

2. Liaw, P.P., Lee Chang, Y.C., Hsieh, P.W., Chang, F.R., K.H., Wu., R.R. "Two new protopines *Argemone mexicanines* A and B and the anti-HIV alkaloid 6-acetyldehydrochloroznine from formosan *A. mexicana*", Department of chemical engineering, Kao Yuan Institute of Technology, Kaohsiung, Taiwan, Roc. (2003).
3. Kausar M. Ansari., Alok Dhawan., Subash K. Khanna., Mukul Das. "In vitro DNA damaging potential of sanguinarine alkaloid, isolated from argemone oil, using alkaline comet assay in mice". Food and chemical toxicology. 147-153. (2005).
4. Khade, K.A. "Report on antimicrobial activity and phytochemical screening of *Argemone mexicana* linn". Scholarly journal of Agricultural science vol.5(6), pp-210-214. (2015).
5. Md. Shahedur Rahman., Md. Faizus Salehin., Md. Abu Hrba Mostafa Jamal., Anzana Parvin., Md. Khasrul Alam. "Antibacterial activity of *Argemone mexicana* linn against water borne microbes". Research journal of medicinal plant 5(5): 621-626, (2011).