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IN VITRO EVALUATION OF ANTIBACTERIAL PROPERTIES OF *JATROPHA CURCAS*

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Abstract

Jatropha curcas is a Bio-diesel plant known for various medicinal uses. The present study was conducted to determine the antibacterial properties of crude extracts of leaves, stem, roots, bark and latex of *Jatropha curcas* plant extracted using various solvents like, water (room temperature and elevated temperature), methanol (room temperature and elevated temperature), ethanol (room temperature and elevated temperature), ethyl acetate and acetone. Extracts were used in a final concentration of 100 mg/ml for assessment of antibacterial properties against two gram negative bacteria namely *Escherichia coli* and *Pseudomonas aeruginosa* and a gram positive bacteria namely *Staphylococcus aureus* using agar well diffusion method. The standard antibiotic Tetracycline was used in a final concentration of 1 mg/ml. Latex of *J. curcas* showed maximum inhibitory zone among all the extracts prepared.

INTRODUCTION

Jatropha curcas (*Euphorbiaceae*) is a biodiesel plant grown in various parts of India and other tropical countries[1]. *J. curcas* is mainly used for the production of biodiesel. The biodiesel is obtained from its seeds. During initial 4-5 years the seeds are obtained in small amount and plant parts like leaves, branches etc. are harvested for rapid growth of the plant. These parts are generally discarded but these can be used for the production of the herbal antibiotics.

Traditional medicine using plant extracts has proved health coverage for over 80% of the world's population, especially in the developing world[2]. The development of resistance among the available pathogens against the antibiotics could also be overcome by usage of these traditionally used medicines.

Previously *J. curcas* has been studied for its antimicrobial properties[3-6]; antidiarrhoeal properties[7], giving a hint that there are some phytochemical definitely present in the species which are responsible for its use in various diseases.

Previous interests and benefits of herbal drugs lead us to design the present investigation with an aim to assess the antibacterial properties of various plant parts of *J. curcas*.

MATERIALS AND METHODS

Plant Samples

Leaves, bark, stem, roots and latex of *Jatropha curcas* were collected from plots near Gomati Nagar Railway station, Lucknow after proper investigation with by experts from MRD Life Sciences, Lucknow. All the parts were washed with tap water and later rinsed with distilled water, dried in sun light and ground with the help of a grinder. The latex was stored at 4°C.

Pathogens

One Gram positive bacteria namely *Staphylococcus aureus* and two Gram negative bacteria namely *Escherichia coli* and *Pseudomonas aeruginos*a available at MRD Life Sciences (P) Ltd., Lucknow, collected from IMTECH, Chandigarh, were sub cultured and used throughout the project work.

Preparation of Plant Extracts

A. For Hot Extracts

Leaves, bark, stem and roots of *Jatropha curcas* were used for preparation of hot

water, hot ethanol and hot methanol. 4 gm of powdered samples (leaves, bark, stem and roots) were dissolved in 40 ml of distilled water, 80% methanol and 70% ethanol. Hot extracts namely hot water (at 85°C), hot ethanol (at 65°C), hot methanol (at 75°C) were kept in water bath for 90 minutes. After that the extracts were filtered through a watt man filter paper in bowls. Then the bowls were kept in hot air oven at 50°C till the extracts got dried. Then the dried extracts were collected in centrifuge tubes. After that the extracts were dissolved in DMSO to get a final concentration of 100 mg/ml.

B. For Room Temperature Extracts

Leaves, bark, stem and roots of *Jatropha curcas* were used for preparation of water, ethanol, methanol, ethyl acetate and acetone extracts. 4 gm of powdered samples (leaves, bark, stem and roots) were dissolved in 40 ml of distilled water, 80% methanol, 70% ethanol, ethyl acetate and acetone. The extracts were kept in dark for 2-3 days at room temperature. After that the extracts were filtered through a watt man filter paper in the bowls. Then the bowls were kept in hot air oven at 50°C till the extracts got dried. After that the extracts were dissolved in DMSO making the extract concentration 100 mg/ml. 8 Different extracts were prepared for each part used and the list is given in.

Antibiogram Analysis

Nutrient agar media was prepared by dissolving all its components in distilled water. Petri plates and NA media was autoclaved. After that media was poured into the autoclaved Petri plates in the LAF room and left for solidification. Then after solidification 20 µl of three pathogens were spread on respective Petri plates and wells (8mm) were bored with the help of a sterile borer. After that 50 µl of antimicrobial samples (100 mg/ml), standard antibiotic tetracycline (1 mg/ml) and autoclaved DMSO were loaded into the wells. The plates were kept in incubator for 24 hrs, at 37°C. Then the zone of inhibition of sample was compared with that of tetracycline. All the tests were performed in triplicates.

RESULTS

Antibiogram Analysis

Table 1-5 below shows antibiogram analysis of various plant extracts.

DISCUSSION

The medicines obtained from plants (herbal medicines) are valuable and available resources for the primary health care system. The plant kingdom holds many species of plant containing antimicrobial properties and can be used for medicinal purpose. Though large number of plants is being used for their antimicrobial properties but more pharmacological investigation is needed.

From the tabulated results it was found that latex of *J. curcas* showed a good response against *E. coli, P. aeruginosa* and *S. aureus* and ethanol extract of roots showed a satisfying activity against *E. coli* and *P. aeruginosa*. Ethanol extracts of Leaves showed an average inhibitory zone against *S. aureus*. The antibacterial activity of the extracts was compared with standard antibiotic tetracycline (1 mg/ml).

Water extract of leaves, acetone, hot methanol, hot ethanol, ethanol (room temp.) and water extracts of bark did not give satisfactory result against bacterial pathogens.

Namuli *et al.*, 2011[8], in their study showed that stem, bark, root and kernel meal of *J. curcas to* contain compounds with antibacterial activities. The results indicate the potential of *J. curcas* as a source of antibacterial compounds

Igbinosa *et al.*, 2009[9], during their *in vitro* investigation of antimicrobial activity of crude ethanolic, methanolic and water extracts of the stem bark of *Jatropha curcas* showed activities with zones of inhibition ranging from 5 to 12, 8 to 20 and 0 to 8 mm for ethanol, methanol and water extracts respectively.

CONCLUSION

It can be concluded from the above research work that latex, roots ethanol extracts can be very good source for herbal drug and can be explored further for the extraction of antibacterial compound by more sophisticated procedures for extraction in order to increase the yield. Future work includes the pharmacological investigation of the solvent extracts.

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Table 1

Antibiogram Analysis of *J. curcas* Latex against Different Bacterial Pathogens

Sr. No.	ZOI against *P. aeruginosa* (mm)		ZOI against *S. aureus* (mm)		ZOI against *E. coli* (mm)	
	By Latex	By Tetracycline	By Latex	By Tetracycline	By Latex	By Tetracycline
1.	15.5	24	20.5	20.5	16.5	23.5

Table 2

Antibiogram Analysis of *J. curcas* Bark Extracts against Different Bacterial Pathogens

Sr. No.	Extracts	ZOI against *P. aeruginosa* (mm)		ZOI against *S. aureus* (mm)		ZOI against *E. coli* (mm)	
		By Extract	By Tetracycline	By Extract	By Tetracycline	By Extract	By Tetracycline
1.	Hot water	0	24	11.5	20.5	0	23.5
2.	Water (Room temp.)	0	24	0	20.5	0	23.5
3.	Hot methanol	0	24	0	20.5	0	23.5
4.	Methanol (room temp.)	0	24	14	20.5	0	23.5
5.	Hot ethanol	0	24	0	20.5	0	23.5
6.	Ethanol (room temp.)	0	24	12.5	20.5	0	23.5
7.	Ethyl acetate	0	24	0	20.5	0	23.5
8.	Acetone	0	24	0	20.5	0	23.5

Table 3

Antibiogram Analysis of *J. curcas* Leaf Extracts against Different Bacterial Pathogens

Sr. No.	Extracts	ZOI against *P. aeruginosa* (mm)		ZOI against *S. aureus* (mm)		ZOI against *E. coli* (mm)	
		By Extract	By Tetracycline	By Extract	By Tetracycline	By Extract	By Tetracycline
1.	Hot water	14	24	0	20.5	14	23.5
2.	Water (Room temp.)	0	24	0	20.5	0	23.5
3.	Hot methanol	15	24	0	20.5	16	23.5
4.	Methanol (Room temp.)	0	24	13	20.5	0	23.5
5.	Hot ethanol	14	24	12	20.5	14	23.5
6.	Ethanol (Room temp.)	0	24	12.5	20.5	0	23.5
7.	Ethyl acetate	11.5	24	0	20.5	0	23.5
8.	Acetone	0	24	0	20.5	12.5	23.5

Table 4

Antibiogram Analysis of *J. curcas* Stem Extracts against Different Bacterial Pathogens

S. No.	Extracts	ZOI against *P. aeruginosa* (mm)		ZOI against *S. aureus* (mm)		ZOI against *E. coli* (mm)	
		By Extract	By Tetracycline	By Extract	By Tetracycline	By Extract	By Tetracycline
1.	Hot water	0	24	0	20.5	0	23.5
2.	Water (room temp.)	0	24	0	20.5	0	23.5
3.	Hot methanol	0	24	0	20.5	0	23.5
4.	Methanol (room temp.)	0	24	0	20.5	0	23.5
5.	Hot ethanol	0	24	0	20.5	0	23.5
6.	Ethanol (room temp.)	11	24	0	20.5	12	23.5
7.	Ethyl acetate	0	24	0	20.5	0	23.5
8.	Acetone	0	24	0	20.5	0	23.5

Table 5

Antibiogram Analysis of *J. curcas* Roots Extracts against Different Bacterial Pathogens

S. No.	Extracts	ZOI against *P. aeruginosa*		ZOI against *S. aureus*		ZOI against *E. coli*	
		By Extract	By Tetracycline	By Extract	By Tetracycline	By Extract	By Tetracycline
1.	Hot water	0	24	12.5	20.5	0	23.5
2.	Water (room temp.)	0	24	12	20.5	13	23.5
3.	Hot methanol	0	24	13.5	20.5	0	23.5
4.	Methanol (room temp.)	0	24	15	20.5	14.5	23.5
5.	Hot ethanol	0	24	16.5	20.5	14	23.5
6.	Ethanol (room temp.)	18	24	19	20.5	20.5	23.5
7.	Ethyl acetate	0	24	0	20.5	0	23.5
8.	Acetone	0	24	0	20.5	0	23.5

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