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IN VITRO EVALUATION OF ANTIBACTERIAL PROPERTIES OF *RICINUS COMMUNIS*

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ABSTRACT

Cold and hot water, methanol, ethanol, ethyl acetate, acetone, and hexane extracts of leaves, stem and root of *Ricinus communis* in a final concentration of 500mg/ml were evaluated for their antibacterial properties against pathogenic microorganisms such as *Pseudomonas aeruginosa, Staphylococcus aureus, and Escherichia coli* using agar well diffusion method. In case of leaf methanolic extract was most effective followed by ethanolic and acetone extracts. Hexane extract of stem was most effective followed by acetone and cold aqueous extracts. Acetone, hexane and ethyl acetate extracts of root were most effective.

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Key Words
Antibacterial properties, *Ricinus communis,* Pathogens, solvent extraction, Agar well diffusion.

INTRODUCTION

People are in a "what to do situation" because of the side effects caused due to the prolonged use of the drugs commonly used today particularly for the treatment of pathogenic microorganisms. Alternative medicines are the need of the hour. The most effective way can be the use of tradional medical system that is the use of herbal medicines which were used long back and are even today being used in the villages. According to WHO (1993), 80% of the world's population is dependent on the traditional medicine and a major part of the traditional therapies involves the use of plant extracts or their active constituents [1].

Ricinus communis (Euphorbiceae) called in India as Arandi is a soft wooden small tree, wide spread throughout tropics and warm temperature regions of the world [2]. The seeds of the plant contain alkaloid ricinine and toxalbumine ricin. They yield a fixed oil, which is used chiefly for medicinal purposes. Though castor plant or its oil is not a food, yet it is one of the most commonly used oil all over the skin, alleviating swelling and pain.

Castor oil chiefly consists of ricinoleate of glycerol or triricinolein with a small quantity of palmitin and stearin. Unlike most fixed oils, castor oil possesses the remarkable property of mixing with absolute alcohol and glacial acetic acid in all proportions. The glycerides of ricinoleic acid in castor oil are mainly responsible for its purgative effect. It can be helpful in Lumbago, rheumatism and sciatica, boils and swellings etc. **[3].**

However the seed and oil have been explored a lot very little literature is available on the medicinal values of other plant parts a few being alcoholic extract of the leaf which has been reported to be hepatoprotective in rats [4-6], methanolic extracts of the leaves have been reported to be antimicrobial. The extract has been reported to be non toxic [7].

Looking at the medicinal importance and current research work on *Ricinus communis* the present study has been designed to evaluate the plant parts (leaf, stem, root) for their antibacterial properties.

MATERIALS AND METHODS:

Sample Collection

Ricinus communis whole plant was collected from nearby locality after proper identification and brought to the laboratory, plant parts leaf, stem and root were separated washed and dried. Dried samples were ground into fine powder by the help of grinder.

Pathogenic Strains

Three pathogenic strains namely *Pseudomonas aeruginosa, Staphylococcus aureus, and Escherichia col*i, available at MRD LifeSciences (P) Ltd., Lucknow availed from IMTECH, Chandigarh, were subcultured and used throughout the study.

Extract Preparation

Antimicrobial metabolites from all the plants parts were extracted in various solvents as cold and hot aqueous, methanol, ethanol, ethyl acetate, acetone, and hexane. For hot aqueous extraction 5gm plant material was soaked in 50ml of hot water and kept in boiling water bath for 2 hours, after that it was filtered in a weighed petri plate and kept in hot air oven for drying, dried extract was dissolved in double volume of DMSO (DimethylSulfoxide) thus giving the final concentration of 500mg/ml. For rest of the extracts 5gm of ground powder was soaked in 50ml of the respective solvents for 3-4 days, thereby filtered in weighed petri plate and dried in hot air oven, the dried extract was dissolved in double volume of DMSO thus giving the final concentration of extract to 500mg/ml.

Antibacterial Susceptibility Assay

Antibacterial susceptibility assay was carried out by agar well diffusion method of **[8]** wherein sterile Nutrient agar plates were prepared and spreaded with 25 µl of pathogen, there after 4 wells (8mm) were dug by the help of sterile borer, 1$^{st\ and}$ 2 nd well were filled with 50 µl of extract, 3rd well with 50 µl of the standard antibiotic Tetracycline and 4th well with sterile distilled water. Plates were incubated at 37ºC for 24 hours and observed for zone of inhibition. All the susceptibility tests were performed in triplicates.

RESULTS

Antibacterial susceptibility assay of the various solvent extracts was carried out against pathogens and the results revealed that all the extracts of leaf are more or less effective, but stem extracts are nearly ineffective, ethyl acetate and acetone extracts of roots are most effective rest all extracts of root are nearly ineffective. Detailed results can be seen in **Table 1-3** below.

Table 1: Antibacterial Susceptibility Assay of *Ricinus communis* Leaf Extracts.

S.NO.	Extracts	Diameter of ZOI against *P. aeruginosa*		Diameter of ZOI against *S. aureus*		Diameter of ZOI against *E. coli*	
		By Extract	By Tetracycline	By Extract	By Tetracycline	By Extract	By Tetracycline
1.	**Cold aqueous**	12mm	28mm	0	23mm	0	19mm
2.	**Hot aqueous**	12mm	26mm	12mm	24mm	0	22mm
3.	**Methanolic**	15mm	17mm	**23mm**	23mm	12mm	18mm
4.	**Ethanolic**	12mm	17mm	14mm	23mm	**18mm**	24mm
5.	**Ethyl acetate**	12mm	25mm	12mm	22mm	0	20mm
6.	**Acetone**	0	25mm	0	18mm	**18mm**	22mm
7.	**Hexane**	0	20mm	0	17mm	0	20mm

Table 2: Antibacterial Susceptibility Assay of *Ricinus communis* Stem Extracts.

S.NO.	Extracts	Diameter of ZOI against *P. aeruginosa*		Diameter of ZOI against *S. aureus*		Diameter of ZOI against *E. coli*	
		By Extract	By Tetracycline	By Extract	By Tetracycline	By Extract	By Tetracycline
1.	**Cold aqueous**	0	22mm	**12mm**	17mm	0	18mm
2.	**Hot aqueous**	0	22mm	0	17mm	0	18mm
3.	**Methanolic**	0	22mm	0	17mm	0	18mm
4.	**Ethanolic**	9mm	23mm	0	15mm	0	20mm
5.	**Ethyl acetate**	9mm	23mm	0	15mm	0	20mm
6.	**Acetone**	0	23mm	**10mm**	18mm	9mm	20mm
7.	**Hexane**	0	23mm	0	18mm	**13mm**	22mm

Note: Well diameter= 8mm, ZOI in case of D/W in each case = 0mm.

Table 3: Antibacterial Susceptibility Assay of *Ricinus communis* Root Extracts.

S.NO.	Extracts	Diameter of ZOI against *P. aeruginosa*		Diameter of ZOI against *S. aureus*		Diameter of ZOI against *E. coli*	
		By Extract	By Tetracycline	By Extract	By Tetracycline	By Extract	By Tetracycline
1.	**Cold aqueous**	12mm	20mm	11mm	25mm	0	20mm
2.	**Hot aqueous**	0	20mm	0	25mm	0	20mm
3.	**Methanolic**	0	20mm	0	25mm	0	20mm
4.	**Ethanolic**	0	22mm	0	20mm	0	23mm
5.	**Ethyl acetate**	0	22mm	15mm	20mm	0	23mm
6.	**Acetone**	**18mm**	22mm	**20mm**	20mm	**17mm**	23mm
7.	**Hexane**	0	25mm	12mm	17mm	0	20mm

Note: Well diameter= 8mm, ZOI in case of D/W in each case= 0mm.

DISCUSSION

Ricinus communis extracts were found to be active against used pathogens and this could be the drugs without side effects for the future.

The methanol, ethanol and acetone extracts of leaves showed good activity against used pathogens, similar results have been reported earlier by [9]. Maximum zone of inhibition of 19 mm by ethanolic extracts of leaf (5gm/ml) was reported by [10], methanol, ethanol and acetone extracts in concentration (0.5gm/ml) much lesser than them showed zones of inhibition ranging from 15mm- 23mm.

Extracts of stem were also assessed for their antibacterial properties, hexane, acetone, cold aqueous, ethyl acetate and ethanol were found to be effective most effective being hexane extract showing a zone of inhibition of 13mm against *E. coli.* This study is one of the few on antibacterial susceptibility of stem extracts of *Ricinus communis.*

Methanol, petroleum ether and hexane extracts of *Ricinus communins* roots have been evaluated earlier for antibacterial properties by **[1]** and methanolic extracts showed maximum zone of 18mm, in the present study methanol extracts did not show any zone but acetone, ethyl acetate and hexane extracts were effective giving zones of inhibition of 15- 20mm.

CONCLUSION

Based on the above research work it can be concluded that *Ricinus communis* can be a very good source for herbal drugs and specially the solvents like methanol, ethanol, acetone and hexane can be explored further for the extraction of antimicrobials by more sophisticated procedures for extraction in order to increase the yield.

Further work also includes the further pharmacological investigation of the solvent extracts and also the investigation of phytochemicals responsible for antimicrobial activity.

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