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Anti-microbial activity and Phytochemical Study of Ethanolic Seed Extract of *Abrus Precatorlus linn*

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ABSTRACT

Abrus precatorius linn (Fabaceae) is a climbing shrub widely distributed in most of the districts of the country in hedges and among bushes on open lands. The objective of this work is to evaluate Anti-microbial activity of ethanolic extract of red seeds of Abrus precatorius. Ampicillin is used as standard Anti-bacterial agent. The result of this study revealed that ethanolic extract and water extract of red seeds of Abrus precatorius exhibited a good inhibitory action against Escherichia coli, Staphylococcus aureus, Salmonella, Pseudomonas aeruginosa.

Key words: Abrus precatorius linn, Anti-microbial activity, Zone of inhibition

1. INTRODUCTION

Bacterial infections are among the most common infections in man, affecting a large population worldwide. About 80,000 species of plants are utilized for treating various diseases in different systems of Indian medicine. Many pharmaceutical companies show interest in plant derived drugs mainly due to the current wide spread belief that "Green Medicine" is safe and more dependable than the costly synthetic drugs. Since the last decade, the rise in the failure of chemotherapeutics and Anti-biotic resistance exhibited by pathogenic microbial infectious agents has led to the screening of several medicinal plants for their potential Anti-microbial activity (1, 2). Many herbs contain dozens of active constituents that combine to give the plant its therapeutic value. Abrus precatorius linn (*Fabaceae*) commonly known as Indian Liquorice, is a climbing shrub, found in sub - tropical regions in India. The seeds are acrid, bitter, astringent, purgative, aphrodisiac, applied locally for sciatica etc (3, 4). From literature review, it reveals that no work has been carried on the seeds of Abrus precatorius for Anti-microbial activity. The objective of the study is to evaluate Anti-microbial activity of ethanolic extract of red seeds of Abrus precatorius linn.

2. MATERIALS AND METHODS

2.1. Authentication of plant material

Abrus precatorius Linn Family Fabaceae, Voucher NO: SDIP, Ref No: 001 dated 10/10/2011 and authenticated by Dr. Madhavachetty, Botanist, Tirupati. The plant materials were dried in vacuum oven to 40° C.

2.2. Preparation of plant extract

Coarsely powdered seed material is successively extracted with 75% ethanol and distilled water separately for 24hrs in a round bottomed flask at room temperature by simple triple maceration method. Extracts were filtered through whiteman filter paper no.1. The filtrate was allowed to dry in a hot air oven, ethanolic and water extracts were scrapped, weighed and stored in air tight container at 4°C till further investigation. For evaluating the anti-microbial activity, both the extracts were prepared in the concentration of 10mg/ml.

Standard used for the activity:

Ampicillin suspension (micronized Ampicillin suspension) in the concentration of 1mg/ml.

2.3. Microorganism cultures used for the study

Standard cultures of Escherichia coli, Staphylococcus aureus, Salmonella, Pseudomonas aeruginosa.

2.4. Phytochemical evaluation

Phytochemical evaluation was carried out for all the extracts as per the standard methods (5, 6).

2.5. Detection of alkaloids

Extracts were dissolved individually in dilute hydrochloric acid and filtered. The filtrates were used to test for the presence of alkaloids.

2.5.1. Mayer's test

Filtrates were treated with Mayer's reagent (Pottasium mercuric iodide). Formation of a yellow cream precipitate indicates the presence of Alkaloids.

2.5.2. Wagner's test

Filtrates were treated with Wager's reagent (iodide in potassium iodide). Formation of brown/reddish brown precipitate indicates the presence of Alkaloids.

2.5.3. Dragendroff's test

Filtrates were treated with dragendroff's reagent (solution of potassium bismuth iodide). Formation of red precipitate indicates the presence of Alkaloids.

2.5.4. Hager's test

Filtrates were treated with Hager's reagent (saturated picric acid solution). Formation of yellow colored precipitate indicates the presence of Alkaloids.

2.6. Detection of carbohydrates

Extracts were dissolved individually in 5ml distilled water and filtered. The filtrates were used to test for the presence of carbohydrates.

2.6.1. Molisch's test

Filtrates were treated with 2 drops of alcoholic α – naphthol solution in a test tube and 2 ml of concentrated sulphuric acid was added carefully along the sides of the test tube. Formation of a violet ring at the junction indicates the presence of carbohydrates.

2.6.2. Benedict's test

Filtrates were treated with Benedict's reagent and heated on a water bath'.

2.6.3. Fehling's test

Filtrates were hydrolyzed with dilute HCl, neutralized with alkali and heated with Fehling's A & B solutions. Formation of red precipitate indicates the presence of reducing sugars.

2.7. Detection of saponins

2.7.1. Froth test

Extracts were diluted with distilled water to 20ml and this was shaken in a graduated cylinder for 15minutes. Formation of 1 cm layer of foam indicates the presence of saponins.

2.7.2. Foam test

Small amount of extract was shaken with little quantity of water. If foam produced persists for 10minutes it indicates the presence of saponins.

2.8. Detection of phytosterols

2.8.1. Salkowski's test

Extracts were treated with chloroform and filtered. The filtrates were treated with few drops of concentrated sulphuric acid, shaken and allowed to stand. Appearance of golden yellow color indicated the presence of triterpenes.

2.8.2. LibermannBurchard's test

Extracts were treated with chloroform and filtered. The filtrate were treated with few drops of acetic anhydride, boiled and cooled. Concentrated sulphuric acid was added carefully along the sides of the test tube. Formation of brown ring at the junction indicated the presence of phytosterols.

2.8.3. Tshugajeu test

Extracts were treated with chloroform and filtered. Excess of acetyl chloride and a pinch of zinc chloride was added, kept aside for some time till the reaction was complete and then warmed on water bath. Appearance of eosin red color indicates the presence of triterpenes.

2.9. Detection of phytosterols

2.9.1. Acetone-Water test

Extracts were treated with acetone. Small amount of water was added and shaken. Appearance of turbidity indicates the presence of resins.

2.10. Detection of phytosterols

To the extract, 1% gelatin solution containing sodium chloride was added. Formation of white precipitate indicates the presence of tannins.

2.11. Detection of flavonoids

2.11.1. Alkaline reagent test

Extracts were treated with few drops of sodium hydroxide solution. Formation of intense yellow color, which becomes colorless on addition of dilute acid, indicates the presence of flavonoids.

2.11.2. Lead acetate test

Extracts were treated with few drops of lead acetate solution. Formation of yellow color precipitate indicates the presence of flavonoids.

2.11.3. Shinoda test

To the alcoholic solution of extract, a few fragments of magnesium ribbon and concentrated HCl was added. Appearance of magenta color after few minutes indicates the presence of flavonoids.

2.11.4. Zinc hydrochloric acid reduction test

To the alcoholic solution of extract, a pinch of zinc dust and concentrated HCl was added. Appearance of magenta

color after few minutes indicates the presence of flavonoids.

2.10. Anti-microbial method

Agar medium was mixed with one liter of distilled water, enclosed in a screw cap container and autoclaved at 121°C for 15minutes. The medium was later dispersed into 90 mm sterile agar plates and left to set. The agar plates were incubated for 24hrs at 34°C to confirm their sterility. When no growth occurred after 24hrs, the plates were considered sterile. Standard cultures of human pathogenic organisms such as Escherichia coli, Staphycoccusaureus, Salmonella typhi and Pseudomonas aeruginosa were chosen for the study. E coli represents gram –ve bacteria, S aureus represents gram +ve bacterium and Pseudomonas aeruginosa is used to represent opportunistic and multi drug resistant bacteria.

3. RESULTS AND DISCUSSION

Cultures of the microorganisms from culture plates were scooped using a wire loop. A loopful was withdrawn and uniformly distributed on the surface of the 4 agar plated individually for each organism by streaking using a sterile swab. Four wells approximately 4mm in diameter and 2.5mm deep were made on the surface of the solid medium using a sterile borer. The plates were turned upside down and the wells labeled with a marker. All the extracts were separately dissolved in Dimethyl sulfoxide (DMSO) to get 10mg/ml solutions. Ampicillin in the concentration of 1mg/ml was prepared. Accurately 0.2 ml of the test (ethanolic and aqueous extracts) and standard solutions were transferred to the cups aseptically and labeled accordingly. The petri dishes are then incubated at 37 ± 1°C for 24hrs. The anti-bacterial activity of the test samples was assessed by measuring the zone of inhibition in mm. Preliminary phytochemical analysis showed the presence of alkaloids, carbohydrates, saponins, tannins, flavonoids like phyto constituents. Some of these components are responsible for anti-microbial activity. From Table 1, it could be agreed that Abrus precatorius seed extracts may be useful for anti-microbial activity against common human pathogenic organisms. Distilled water extracts also showed activity against the four organisms chosen for the study. Although it is an in-vitro study, these results may differ with in-vivo study which has to be done extensively against a wide range of microbial population.

Table 1. Anti-microbial activity of ethanolic and water extracts of seeds of Abrus precatorius

Organism	*Zone of Inhibition (mm)*		
	Ethanolic seed extract	Aqueous extract	Ampicillin
	Concentration: 10m/ml	10mg/ml	1mg/ml
	Dose: 0.2ml	0.2ml	0.2ml
Escherichia nosacoli	16	12	22
Staphylococcus aureus	18	14	19
Salmonella typhi	14	15	16
Pseudomonas aeruginosa	15	10	18

4. CONCLUSION

The result of this study revealed that ethanolic extract and water extract of red seeds of Abrus precatorius exhibited a good inhibitory action against Escherichia coli, Staphylococcus aureus, Salmonella, Pseudomonas aeruginosa.

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AUTHORS CONTRIBUTION

This work was carried out in collaboration among all authors.

CONFLICT OF INTEREST

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