

Periodic Research

Phytochemical Analysis and Antibacterial Activity in *Ricinus Communis* L.

Abstract

During the present investigation antibacterial activity of castor (*Ricinus communis*) and its oil, leaf, stem and seed extract were screened against pathogenic bacteria. One is gram positive bacteria (*Staphylococcus*) and other is gram negative bacteria (*Escherichia coli*). The ethanol and methanol extracts showed maximum zone of inhibition against gram positive and negative bacteria. These findings suggest that whole parts of plant are effective in antibacterial activity assay. In another study, phytochemical analysis of secondary metabolites was also carried out. However, further studies are needed to evaluate active compound and probability of medicinal benefits in chemotherapy among human.

Keywords: *Ricinus communis*, *Staphylococcus*, *Escherichia coli*, Antibacterial activity.

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Introduction

Natural Products are the rich source of biologically active compounds and today's many medicines are either obtained directly from natural source or were developed from a lead compound originally obtained from a natural source [1]. Ethnobotany has been deeply rooted in the Indian tradition and culture. Many medicines and their formulations have been based on rich ayurveda of ancient India. Many plants have been utilized to cure uncommon disorders. Among those plants castor is one the important plant. *Ricinus communis* (Castor oil plant) belong to the family Euphorbiaceae, is one of the medicinally important oil seed crop. Castor plant grows commonly in the tropical and warm region. It is a tall glabrous shrub or almost small tree, 2-4 m high, found in India. [2-3] In traditional system of medicines Euphorbiaceae plants are used to treat various microbial diseases such as diarrhea, dysentery, skin infections and gonorrhoea.[4] In Indian system of medicine the leaf, root and seed oil of this plant have been used for the treatment of the inflammation and used on liver disorders due to its [5] hypoglycemic[6] laxative properties[7].

The leaves are used for skin diseases and kidney; urinary bladder infections [8]. The seeds of *Ricinus Communis* have several traditional applications [9]. They are used with arguable success in the treatment of warts, cold tumors' indurations of the mammary glands, corns and moles [10]. Furthermore the toxic organic substance present in the plant. These compounds are mostly secondary metabolites such as alkaloids, steroids, tannins, phenol compounds, flavonoids resins, fatty acids and gums, which are capable of producing definite physiological action on body [11]. *Ricinus communis* is used medicinally in different countries and are a source of many potent and powerful drugs [12-15]. The seed contains 46-60 % oil and the only commercial source of ricinoleic acid. The oil is used as industrial lubricants, paints, coatings and plastics [16].

According to WHO (1993), 80% of the world's population is dependent on the traditional medicine and major part of the traditional therapies involves the use of plant extracts or their active constituents. With the continuous use of antibiotics, microorganisms have become resistant. This has created immense clinical problem in the treatment of infectious diseases. Therefore, there is a need to develop alternative antimicrobial drugs for the treatment of infectious diseases. One approach is to screen local medicinal plants for possible antimicrobial and antifungal properties.

Materials and Methods

Plant Materials

The leaf, stem, seeds of *Ricinus communis* were collected from various localities of Marathwada. They were shade dried at room temperature for 4-5 days. Fine powder was made from these plant parts. This powder was utilized for further studies.

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Preparation of Crude Extract in Solvents

Solvent extract were prepared in different solvents at room temperature by simple extraction method. Collected plant parts were shade dried and ground to fine powder using grinder mixture. Dried powder of each plant parts 10 gm was mixed in 100 ml of different extract solvent (ethanol, methanol, petroleum ether, chloroform etc.) in conical flask. The flasks were plugged with papers. All conical flasks were kept on shaker for 5 hours. Samples were then filtered and centrifuged at 5000 rpm for 15 minutes. The supernatant was collected and the solvent was evaporated at 45°C, 1ml in vacuum evaporator.

Phytochemical Analysis

Identification of the Chemical Constituents

Polyphenolic compounds, flavonoids, glycosides, saponins, tannins, and alkaloids were carried out using different solvent extracts (methanol, ethanol, petroleum ether, chloroform, aqueous).

Preliminary Phytochemical Analysis Saponins

About 0.5 ml of extract was taken with 5 ml distilled water and then heated to boil frothing (appearance of creamy miss of small bubbles) shows the presence of saponins.

Tannins

2-3ml extract was taken in test tube and 10% FeCl₃ (ferric chloride solution) was added in it. Dark blue or greenish grey coloration was observed. This is confirmation test for tannin.

Flavonoids

Aqueous extract was added in 10% Ferric chloride. A green precipitate indicates positive test. Secondly methanol extract + 10% NaOH + dil. HCl were added in test tube. Yellow solution turned colourless on addition of dil. HCl which indicates positive test.

Cardiac Glycosides

Methanol extract (2ml) + 3.5 % of FeCl₃ +Glacial acetic acid + 2ml of conc. H₂SO₄ was taken in beaker, reddish brown ring at inter phase is indicator of positive test.

Steroids and Terpenoides

Methanol extract (1ml+1ml chloroform +2-3 ml of acetic anhydride and 1-2 drops of conc. H₂SO₄ were added. Dark green colouration of the solution indicates the presence of steroids and pink or red colouration of the solution indicates that presence terpenoides.

Reducing sugar

Methanolic extract + 5 ml of equal volume of Fehling solution A and B boiled for 5 min. Red precipitate is indication positive test/ rusty brown.

Indole Alkaloids

Methanolic extract + conc. H₂SO₄ + potassium dichromate was taken in flask. Colour change is confirmation for the presence of indole alkaloids.

Anti-Microbial Assay

The gram negative bacteria (*E. coli*) and gram positive bacteria *Staphylococcus* were collected from Dept of microbiology, Govt. Institute of Science, Aurangabad.

In Vitro Determination of Antimicrobial Activity

Antibacterial activity was determined by agar disc diffusion method. The discs (6 mm diameter) impregnated with known concentration of the standards and extracts were placed on the surface of the Petri plates containing 20 ml of nutrient agar media for bacterial strains. The plates were incubated for 24 hrs. at room temperature. At the end of incubation, inhibition zones scale. The study was performed in triplicate and the mean values were presented.

The leaves, stem, seed, powder of Ricinus communis was checked for antibacterial activity with all extracts. However, the results of only methanolic and ethanolic extracts were active against the tested microorganisms. While the aqueous extract don't show any antimicrobial activity. The Minimum Inhibitory Concentration (MIC) and the diameters of the zone of inhibitions of the tested microbes are shown in Table 1. Ricinus

Extracts of different Plant parts	Sterols and Terpenoids	Tannins	Flavonoids	Indole Alkaloids	Carbohydrates and Glycosides	Saponins
Petroleum Ether						
Leaves	+++	++	+	++	++	++
Root	+	--	+	+	--	+
Stem	++	--	+	+	++	++
Seed	++	--	--	+	--	+
Ether Extract						
Leaves	++	--	++	++	++	++
Root	--	--	+	+	+	++
Stem	--	--	+	++	++	+
Seed	++	+	++	+	--	+
Chloroform Extract						
Leaves	+	++	+	+	+	+
Root	--	+	+	+	--	+
Stem	--	--	++	+	++	+

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Seed	+	--	+	+	++	+
Methanol Extract						
Leaves	++	++	+	+	--	+
Root	++	+	+	+	++	--
Stem	+	+	+	+	+	+
Seed	+	+	+	+	+	+
Aqueous Extract						
Leaves	--	+	+	--	+	+
Root	--	--	+	--	+	+
Stem	--	--	+	+	+	+
Seed	+	--	+	+	+++	+

The antibacterial activities of *R. communis* leaves, stem, seed suggest that the extract contains the effective active phytochemical responsible for the elimination of microorganisms.

Minimum Inhibitory Concentration (MIC)

MIC means the lowest concentration of an antimicrobial agent that appears to inhibit the growth of microorganism. Minimum inhibitory concentration was determined by Nutrient agar method. The study was performed in triplicate and the mean values were presented.

Result and Discussion

The plant parts of *Ricinus communis* leaves, root, stem, and seed, showed good activity against dermatophytic and pathogenic bacteria. The methanol, ethanol extract (100/200 ml) of leaves showed activity against selected bacteria. Stem extracts with ethanol, methanol (100/200 ml) also showed prominent activity against two pathogens. Aqueous root extracts showed no antibacterial activity against pathogen. The experiment was performed in triplicates (Table.1).

Phytochemical Study of *Ricinus Communis* in Various Solvents

Leaves and stem of *Ricinus communis* indicated the presence of alkaloids, flavonoids, steroid, terpenoids, cardiac Glycosides, saponins. As well as chloroform, methanol extract showed presence of flavonoids, steroid, terpenoids, saponins, glycosides and tannins.

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