

In Vitro Antimicrobial Activity of Benzene and Chloroform Extract of *Mucuna pruriens*

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ABSTRACT

The investigation aimed to carry out the In-vitro antimicrobial activities of the benzene and chloroform extract of *Mucuna pruriens* seeds. The antimicrobial activity of the extract was determined by the agar well diffusion method against various gram positive, gram negative and spore forming microorganisms and fungi. The chloroform and benzene extract of *M. pruriens* shown antimicrobial activity against the tested microorganisms *E.coli*, *Candida albicans*, *Rhodococcus rhodochrous* and *Arthrobacter protothormiae* but chloroform extract shown broad-spectrum antimicrobial activity against *Arthrobacter protothormiae*. The results obtained in the study shows that chloroform extract of *M. pruriens* has more antimicrobial activity against *Arthrobacter protothormiae* because it shown large diameter of Zone of Inhibition (ZI).

Keywords: *Mucuna pruriens*, benzene, chloroform, antimicrobial.

INTRODUCTION

Plants were used for treating various diseases in an age old practice in a large part of the world especially in developing countries were dependence on traditional medicine for variety of diseases¹. Long before the advent of modern medicine herbs were main stream remedy for nearly all ailments. Microorganisms and medicinal plants are rich sources (remedies for nearly all ailments) of secondary metabolites, which were the potential sources of useful drug and other useful bioactive products². There was a great interest in plants with antimicrobial properties, had revived as a result of current problems such as resistance associated with the use of antibiotics³. The primary benefits of using plant derived medicine derived that they were relatively safer than synthetic alternative offering profound therapeutic benefits and more affordable treatment⁴. Usually these natural products extracted and isolated from plants⁵. These extracts show various medicinal properties, among which one is the antimicrobial property⁶. Over the past several years, intensive efforts have been made to discover clinically useful antimicrobial drugs, which have been reviewed by many researchers⁷.

Mucuna pruriens plant is commonly called as common cowitch, velvet bean, cowhage, kapikachhu and naikaranam. It is a constituent of more than 200 indigenous formulations⁸. The fruit of the plant is thick and leathery. *Mucuna pruriens* belongs to the family Leguminosae⁹. *Mucuna pruriens* is an annual climbing legume indigenous to tropical regions, especially Africa,

India and West Indies. In India it is found in the foothills of the Himalayas, the plains of West Bengal, Madhya Pradesh, Karnataka, Kerala and Andhra Pradesh¹⁰. The plant is infamous for its extreme itchiness produced on contact, particularly with the young foliage and the seed pods. The plant is an annual, climbing shrub with long vines that can reach over 15 m in length¹¹. Seeds contain alkaloid constituents mucanadine, mucunine, prurienidine, purienine¹², epoxy fatty acids such as cis-12,13-epoxyoctadec-trans-9-cis-acid, cis-12,13-epoxyoctadec-trans-9-enoic acid¹³.

Mucuna pruriens has also shown to exhibit neuroprotective effect by increase brain mitochondrial complex-I activity and significantly restoring dopamine and norepinephrine levels in Parkinsonism animal model¹⁴. *Mucuna pruriens* possess a wide range of pharmacologic activities such as antimicrobial activity¹⁵, anti-protozoal activity¹⁶, anti-inflammatory activity¹⁷, neuroprotective activity¹⁴, anti diabetic activity¹⁸, antioxidant activity¹⁹. Literature has shown that the biological activities of plant extracts essential oils and their individual pure components and has documented the inhibitory activity of these substances against the growth of various fungi²⁰. There is a general growing trend among consumers for more natural than synthetic products in a whole range of industries, including food and drink, cosmetics, agricultural and pharmaceuticals²¹⁻²².

MATERIALS AND METHODS

Table 1. Names of bacterias with catalog number.

Bacterium	Catalog number
<i>Escherichia coli</i>	MTCC78 pBR
<i>Condida albicans</i>	MTCC227
<i>Rhodococcus rhodchrous</i>	MTCC265
<i>Arthrobacter protophormaie</i>	MTCC2682

Table 2. Percentage yield of benzene extract of *Mucuna pruriens*

Initial weight (gm)	Final Yield (gm)	% Yield
30	1.59	5.29%

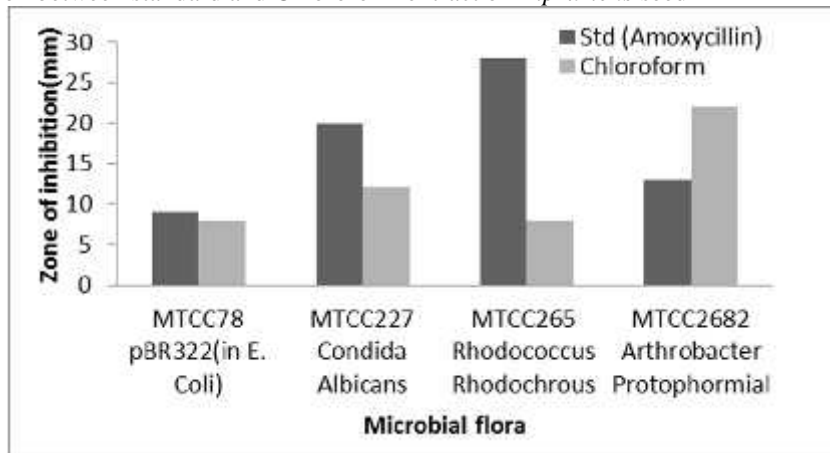
Table 3. Percentage yield of chloroform extract of *Mucuna pruriens*

Initial weight (gm)	Final Yield (gm)	% Yield
30	1.20	4.0%

Table 4. Zone of Inhibition: area of bacterial culture that was inhibited by the benzene and chloroform extracts of *mucuna pruriens*.

Sr. No.	Microbial Flora	Solutions [Zone of Inhibition (mm)]		
		Std (Amoxicilline)	Chlorofom	Benzene
1	MTCC78 pBR322(in E. Coli)	9 mm	8 mm	8 mm
2	MTCC227 Candida Albicans	20 mm	12 mm	8 mm
3	MTCC265 Rhodococcus Rhodochrous	28 mm	8 mm	10 mm
4	MTCC2682 Arthrobacter Protophormiae	13 mm	22 mm	8 mm

Figure 1. Comparison between standard and Chloroform extract of *M.pruriens* seed



Plant Material and Microbial flora: Fresh beans of *Mucuna pruriens*(Genus- *Mucuna*, Family- Fabaceae) were collected from the rural area (Kaliar in Roorkee). The Plant material was thoroughly washed in water and dried plant material was grinded to powder and 30gms of the dried plant material was used for the extraction. Microbial flora: [obtained from Microbial Type Culture Collection & Gene Bank (MTCC), Institute of Microbial Technology, Chandigarh, India] see (Table 1.). Standard antibiotic solution: Antibiotic used: Suprimox, Amoxicillin 250mg vial

Suprimox: INJ from Ranbaxy (Rexcel).
Dose: 10mg/ml
Vehicle used: Water for injection

Assays
Extraction Process:
Extraction with Benzene:
The dried seeds were crushed to reduce size which subjected to soxhlation was exhaustively extracted with benzene for 6 hours. Collect the extract in beaker and evaporate the solvent in hot-air oven at 40-50°C to

Figure 2. Comparison between standard and Benzene extract of *M.pruriens* seed

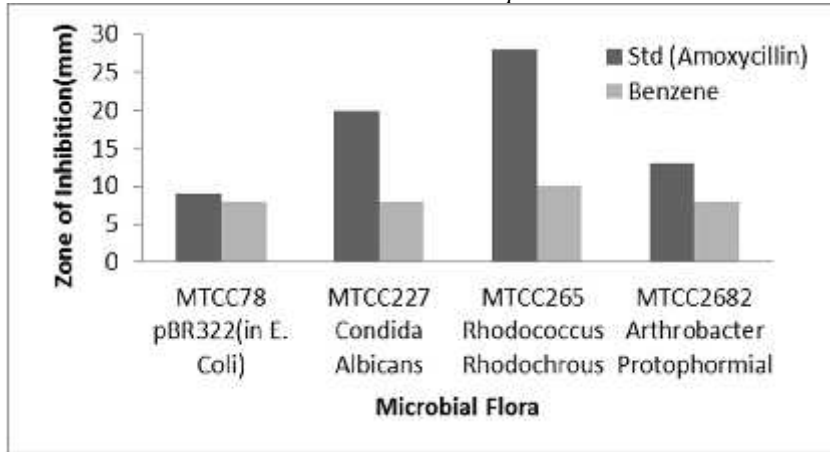
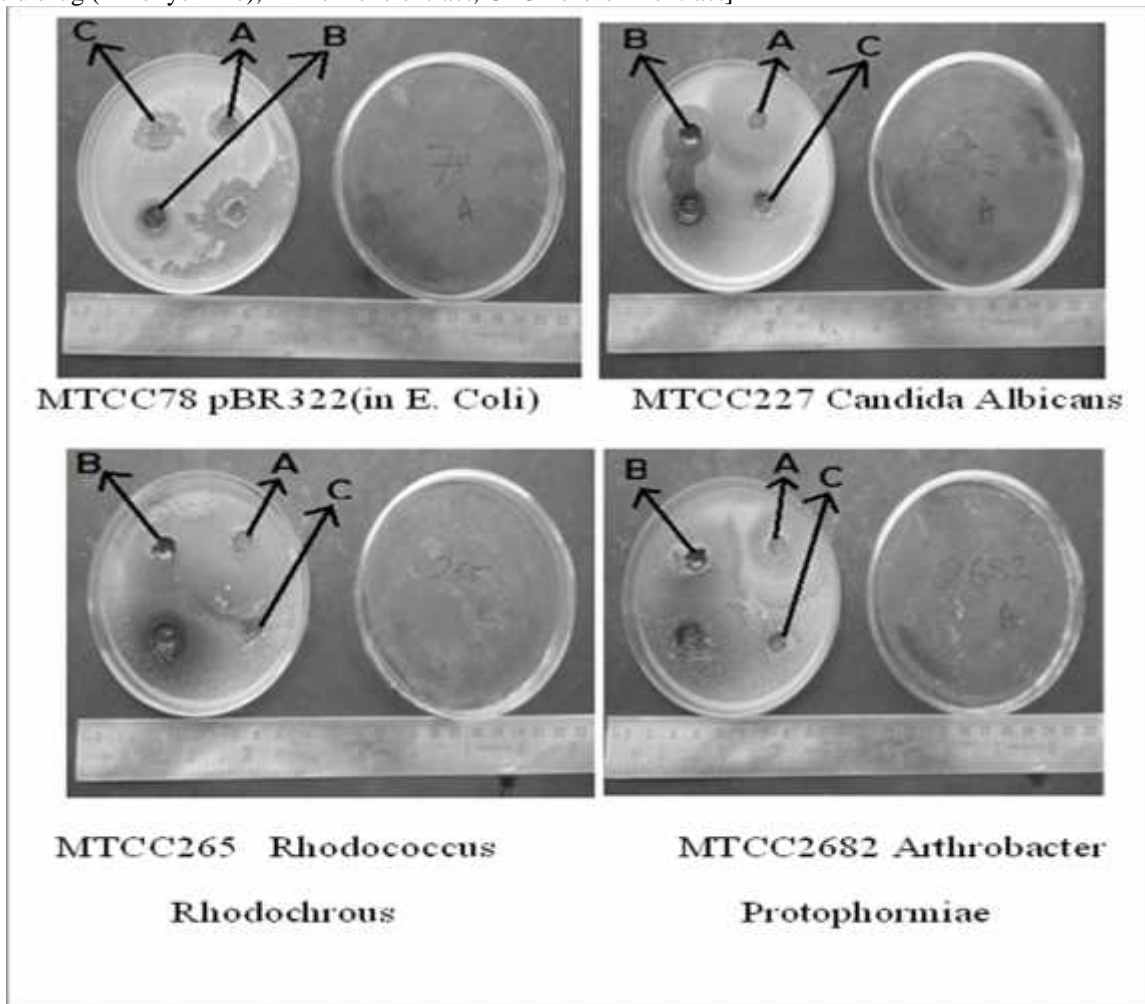


Figure 3. Zone of bacterial flora inhibited by the extract of *mucuna pruriens* and standard drug i.e. Amoxycilline [A- Standard drug (Amoxycilline), B- Benzene extract, C- Chloroform extract]



obtained dried contain. Stand the dried extract[1.590gm] in closed container. Percentage yield of benzene extract of *Mucuna pruriens* was 5.29% (Table 2.).

Extraction with chloroform

The dried seeds were crushed to reduce size which subjected to soxhlation was exhaustively extracted with

chloroform for 6 hours. Collect the extract in beaker and evaporate the solvent in hot-air oven at 40-50°C to obtained dried contain. Stand the dried extract[1.20gm] in closed container. Percentage yield of chloroform extract of *Mucuna pruriens* was 4.0% (Table 3.).

Antimicrobial Activity of Mucuna Pruriens: A part from the studied of phytochemistry, the plant extract was also

subjected to anti microbial studies. To carry out these studies Pour plate method has been chosen. To assay the activity the pour plate method was carried out using plant pathogenic microorganisms including various bacterias. Finally determined the zone of inhibition.

RESULTS AND DISCUSSION

Mucuna pruriens commonly known as Atmagupta widely used Ayurvedic medicine in India. In the present days the whole plant *Mucuna pruriens* are used for antimicrobial properties for extracting plant metabolites against plant pathogenic bacteria and fungi by using agar diffusion methods and the zones of inhibition were calculated and note down in the tabular form.

The chloroform and benzene extract of *Mucuna pruriens* seed has shown antimicrobial activity against *Arthrobacter protophormiae*, *E. coli*, *Condida albicans* and *Rhodococcus rhodochrous*.

Zone of inhibition was measured (Table 4. & Figure 3.) and compared activity of benzene extract and chloroform extract of *Mucuna pruriens* with standard drug (Figure 1.& 2.).

CONCLUSION

The chloroform extract of *Mucuna Pruriens* inhibit the growth of some of the tested micro-organisms (gram positive & gram negative). The Chloroform extract at a concentration exhibited the antimicrobial effect against all the tested micro-organisms. The extract showed strong antimicrobial activity against *Arthrobacter protophormial* and showed the minimum activity against *E. coli*, *Condida albicans*, *Rhodococcus rhodochrous* in compared to standard solution of antibiotic (Amoxicilline).

The Benzene extract of *Mucuna Pruriens* inhibit the growth of some of the tested micro-organisms (gram positive & gram negative). The Benzene extract at a concentration exhibited the antimicrobial effect against all the tested micro-organisms. The extract shown minimum antimicrobial activity *Arthrobacter protophormial*, *E. coli*, *Condida albicans*, *Rhodococcus rhodochrous* in compared to standard solution of antibiotic (Amoxicilline).

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