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Journal of Applied Pharmaceutical Science

ISSN: 2231-3354 Received on: 05-08-2011 Accepted: 10-08-2011

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Investigation of antibacterial activities of ethanol extracts of *Musa paradisiaca* Lam

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ABSTRACT

In the present work, the roots of *Musa Paradisiaca* Lam. (Family: Musaceae) were investigated for preliminary phytochemical screening and antibacterial activity. Preliminary phytochemical screening of ethanol extract of the roots revealed the presence of various classes of compounds like alkaloids, glycosides, steroids, flavonoids, saponins, reducing sugars and tannins. The ethanol extract of the roots of *Musa Paradisiaca* Lam. was tested by using agar disc diffusion method for antimicrobial assay. The ethanol extract of *Musa Paradisiaca* Lam. showed moderate in-vitro antibacterial activity against both gram positive (*B. Megaterium, B. Subtilis, S. aureus*) and gram negative (*E. coli, P. aeruginosa, S. dysenteriae, S. typhi, Vibrio cholerae and S. flexneri*) bacteria with the zones of inhibition ranging from 10.53 ± 0.37 to 12.42 ± 0.85 mm at concentration of 500 µg/disc. Thus the findings revealed the medicinal potential of *Musa Paradisiaca* Lam. against various infectious diseases to develop a drug.

Key words: Antimicrobial assay, antibacterial activity, zones of inhibition, Musa paradisiaca.

INTRODUCTION

Musa paradisiaca L. (Musaceae) an important source of food in the world, has also been reported to be popularly used as anti-venom (Borges et al., 2005). Stem juice is also used in nervous affectations like epilepsy, hysteria and in dysentery and diarrhoea. Several oligosaccharides comprising fructose, xylose, galactose, glucose and mannose occur naturally in banana, making it an excellent prebiotic for the selective growth of beneficial bacteria in the intestine (Gibson, 1998). Roots of M. paradisiaca are anthelmentic and flowers are astringent. Fruits are mild laxative. It aids in combating diarrhoea and dysentery and promotes healing of intestinal lesions in ulcerative colitis. It is also useful in celiac disease, constipation and peptic ulcer (Mallick et al., 2007). In southern Nigeria various parts of *M. paradisiaca* are used as hypoglycemic agents. Long term administration of the powdered banana is known to slightly alter some hematologic parameters in rats (Costa et al., 1999). The use of plant extracts and phytochemicals, both with antimicrobial properties, can be a great significance in therapeutic treatments. In the last few years, a number of studies have been conducted in different countries to prove such efficiency (Kubo et al., 1993; Shapoval et al., 1994). The present study included Musa paradisiaca Lam. used in folk medicines due to lack of scientific data especially on antibacterial activities.

MATERIALS AND METHODS

Plant materials and preparation of the extract

The fresh plant selected for the study was collected from Dulahazara, Chittagong, Bangladesh on March in 2009 at day time. The roots of *Musa paradisiaca* Lam. (Family: Musaceae) were cut into very small pieces and dried at room temperature under shade. The roots were ground into a coarse powder with the help of a suitable grinder. The powder was stored in an airtight container and kept in a cool, dark and dry place until the analysis was commenced. Ground plant materials (200gm) were taken in a clean, flat bottomed plastic container and soaked in 1400ml of 99.8% ethanol. The container with its contents was sealed and kept for a period of 14 days accompanied by continuous shaking with the shaker. The extracts were filtered through a Millipore filter (0.25 μ m). The resulting filtrate was concentrated under reduced pressure and then transferred into a well labelled sterile container.

Test organisms

The test microorganisms included *Bacillus megaterium*, *Vibrio cholera, Escherichia coli, Bacillus subtilis, Staphylococcus aurious, Shigella dysenteriae, Shigella flexneri and Salmonella typhi* were clinical strains obtained from the stock culture of the microbiology laboratory, Department of Pharmacy, BGC Trust University Bangladesh. Stock cultures of bacteria were maintained on Nutrient Agar and all cultures were sub-cultured monthly and subsequently stored at 4°C.

Standard drug

Gentamycin was used as a standard drug in this research work and the drug was received as a gift from Incepta Pharmaceuticals Limited, Dhaka, Bangladesh.

Antimicrobial assay

The antimicrobial assay was done using the agar disc diffusion method. The nutrient agar media and nutrient broth were used to demonstrate the antibacterial activity and to make subculture of the test organisms.

Sterilization of different equipments and media

Media, petri dish and other glassware were sterilized by autoclaving at a temperature of 121°C and then, all of these were kept in the laminar air flow for 30 minutes. The UV-light was also switched on before working in the laboratory.

Preparation of subculture media and determination of zone of inhibition

The agar medium was distributed among the conical flasks and the bacteria were placed. After mixing very quickly, the mixture was placed in the respective petridishes. Then, the petridishes were placed in the incubator at 37^{0} C for 18 to 24 hours to assure the growth of bacteria. In sterile medium, the filter paper discs were impregnated with known amount of test substances by using micropipette and dried. These discs were placed in the

petridishes which contain suitable agar medium and then placed for incubation at 37^{0} C for 18 hours. If the sample has antimicrobial activity, it will inhibit the growth of microorganisms by giving clear, distinct zone called "zone of inhibition". The antimicrobial activities were determined by the width of the zone of inhibition in millimetre (mm).

Phytochemical screening of extracts

The phytochemical screening of the extracts was done on the ethanol extracts using standard procedures described by Trease and Evans (1989). The following qualitative tests were carried out:

Test for alkaloids

Two tests were performed to identify the presence of alkaloids in *Musa paradisiaca* Lam. **Mayer's Test**: Under this test, 2ml of the extract and 0.2ml of dilute hydrochloride acid were taken in a test tube. Then, 1ml of Mayer's reagent was added. A yellowish buff precipitate is indicative of the presence alkaloids. **Dragendroff's Test**: In case of this test, 2ml of the extract and 0.2ml of dilute hydrochloride acid were taken in a test tube. Then, 1ml of Dragendroff's reagent was added. Observation of an orange-brown precipitate was taken to indicate the presence of alkaloids.

Test for glycosides

A small amount of the alcoholic extract was taken in 1ml of water in a test tube and a few drops of aqueous NaOH were added. A yellow coloration indicates the presence of glycosides.

Test for steroids

1ml of the ethanolic extract was taken in a test tube and 1ml of sulphuric acid was added. The formation of red colour solution indicates the presence of steroids.

Test for gums

5 ml of the extract was taken and then Molish reagent and sulphuric acid were added. The formation of red-violet ring at the junction of two liquids indicates the presence of gums.

Test for flavonoids

A few drops of concentrated hydrochloride acid were added to a small amount of an alcoholic extract of the plant material. Immediate development of a red colour indicates the presence of flavonoids.

Test for reducing sugars

2ml of an aqueous extract of the plant material was added to 1ml of a mixture of equal volume of Fehling's solution A&B and was boiled for 5 minutes on a boiling water bath. The formation of brick-red color precipitate shows the presence of reducing sugars.

Test for tannins

5ml of the extract was taken in a test tube and then 2ml of 5% FeCl₃ solution was added. A greenish-black precipitate indicates the presence of tannins.

Test for saponins

1ml of the extract was diluted with distilled water to 20 ml and shaken in a graduated cylinder for 15 minutes. The formation of one centimetre layer of foam indicates the presence of saponins.

RESULTS AND DISCUSSION

The antimicrobial activities of *Musa paradisiaca* Lam. were performed by agar diffusion method using a paper disc. The ethanol extracts of the plant produced a significant antibacterial activity against both gram positive and gram negative bacteria. The results on investigation of antimicrobial activities of ethanol extracts of roots of *Musa paradisiaca Lam*. are summarized in Table 1. Activity of ethanol extracts of roots of the plant against both gram positive and gram negative bacteria showed an indication that the extracts of the plant were of broad spectrum. The zone of inhibition of ethanol extract of the plant at concentration of 500μ g/disc ranged from 10.53 ± 0.37 to 12.42 ± 0.85 where as the standard drug, Gentamycin at concentration of 30μ g/disc had higher zones of inhibition ranging from 23.55 ± 0.76 mm to 27.10 ± 0.60 mm (**Table 1**).

Table 1: Antimicrobial activities of ethanol extract of *Musa paradisiaca* Lam. and standard drug, Gentamycin.

Test Organisms	Blank	Zone of Inhibition (mm)			
		Ethanol extract of <i>Musa</i> <i>paradisiaca</i> Lam. (500µg/disc)	Standard drug Gentamycin (30 µg/disc)		
Gram positive					
Bacillus megaterium	-	12.42 ± 0.85	25.67 ± 0.88		
Bacillus subtilis	-	12.07 ± 0.59	27.10 ± 0.60		
Staphylococcus aureus	-	10.53 ± 0.37	25.42 ± 0.17		
Gram negative					
Escherichia coli	-	11.48 ± 0.48	25.10 ± 0.72		
Salmonella typhi	-	10.55 ± 0.42	24.27 ± 0.37		
Shigella dysenteriae	-	11.10 ± 0.12	23.72 ± 0.69		
Vibrio cholerae	-	10.70 ± 0.12	23.55 ± 0.76		
Shigella flexneri	-	10.55 ± 0.20	23.93 ± 0.45		

Data were represented as Mean \pm SD of triplicate determination

 Table 2: Preliminary phytochemical screening of ethanol extract of the roots of Musa paradisiaca Lam.

Alkaloid	Glycosides	Steroid	Gums	Flavonoids	Saponin	Reducing sugars	Tannins
+	+	+	-	+	+	+	+

Key: + (plus) indicates present and - (minus) indicates absent

The qualitative phytochemical tests showed that *Musa paradisiaca* Lam. contained alkaloids, glycosides, steroids, gums, flavonoids, saponin, reducing sugars and tannins. **Table 2** showed the results of the qualitative analysis of the phytochemical components of the plant.

CONCLUSION

Thus from our findings, it is concluded that the studied plant can be a potential source of useful antibacterial drug. Further studies are however recommended on the plant in order to isolate, identify, characterize and elucidate the structure of the bioactive compounds from the medicinal plant *Musa paradisiaca* Lam.

ACKNOWLEDGEMENT

All of the authors are very grateful to Professor M Mohi Uddin Chowdhury, Head of the Department of Pharmacy of Southern University Bangladesh for providing the experimental bacterial strains.

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