Evaluation of antibacterial activity of cyanobacterium *Nostoc humifusum* Carmichael ex Bornet & Flahault

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ABSTRACT

Cyanobacteria represent a group of highly diverse, widely distributed and ecologically important prokaryotes. Freshwater cyanobacteria are known to synthesize a variety of biologically active and structurally diverse secondary metabolites that have potential pharmaceutical applications. The antibacterial activity of the methanolic extract of the cyanobacterium *Nostoc humifusum* against two bacteria *Staphylococcus aureus* and *Pseudomonas aeruginosa* were studied. The antibacterial activity was determined by agar disc diffusion method on solid nutrient agar medium. The methanolic extract of *Nostoc humifusum* showed significant antibacterial activity against the two tested bacteria by expressing various zones of inhibition. The present study reveals the presence of potential antibacterial compounds in the selected cyanobacterium *Nostoc humifusum*.

Key words: *Nostoc*, antibacterial activity, methanolic extract.

I. INTRODUCTION

Cyanobacteria are the oldest and most diverse group of gram negative photosynthetic prokaryotes. They are widely distributed and ecologically important group of microorganism. Cyanobacteria have gained a lot of attention in recent years because of their potential applications in biotechnology. They are rich sources of structurally novel and biologically active metabolite, which are shown to exhibit antibacterial (Ghasemi *et al.*, 2003), antifungal, anticancer or cytotoxic (Kwan *et al.*, 2010), antimalarial (Linington *et al.*, 2007) and other pharmacological activities. Cyanobacteria from local habitats seem to be a source of potential new active substances that could contribute to reduction of the number of bacteria, fungi, viruses and other microorganisms (Mundt *et al.*, 2001). Several studies reporting the antibacterial activity of cyanobacterial extracts have been performed during last decade (Ostenswik *et al.*, 1998; Skulberg, 2000; Biondi *et al.*, 2008).

The aim of the present study was to investigate the antibacterial activity of *Nostoc humifusum* in methanolic extract.

II. MATERIALS AND METHODS

2.1 Collection of sample

Water samples of cyanobacteria were collected from Poomala damsite, Thrissur District, Kerala.

2.2 Isolation and culture conditions

The cyanobacteria were grown in nitrogen free BG-11 medium. The pH of the medium was maintained at 7.4. The cultures were incubated at a temperature of 24 ± 2°C under continuous illumination of 1500-2000 lux for 12 hours light and 12 hours darkness. Standard plating and streaking techniques were employed for obtaining pure culture.

2.3 Identification of the cyanobacterium

The Cyanobacterium was examined under Leica DM 1000 compound microscope and photographs were taken. The identification the cyanobacteria was done with standard taxonomic manuals of Desikachary (1959), Prescott (1982) and Anand (1989).

2.4 Preparation of cyanobacterial culture crude extract.

The cyanobacterial culture was harvested after fifteen days of growth by centrifugation at 5000 rpm for ten minutes. 2 g of the pellet was weighed, added 100 ml methanol and 4 ml of acetic acid and was evaporated to dryness by heating in a water bath. The resulting concentrate was resuspended in 2 ml of methanol. 100µl of the crude extract was used for bioassay.

2.5 Test bacteria

In the present study *Staphylococcus aureus* (MTCC-737) and *Pseudomonas aeruginosa* (MTCC-424) were used for testing antibacterial activity.

2.6 Antibacterial assay

Antibacterial assay of the crude extract was tested by agar disc diffusion method. The inoculum of test bacteria was spread on nutrient agar plates with a cotton swab. Filter paper discs of 5 mm diameter saturated with 100 µl of the extract dried and placed on the agar plates. Also a filter paper disc saturated with aqueous extract was placed. Plates were incubated at 37°c for 24 hours. The diameter of the zone of inhibition were measured and recorded. Antibacterial activity of the extract was compared with standard Tetracycline antibiotic disc.

III. RESULTS AND DISCUSSION

Antibacterial activity of methanolic extract of *Nostoc humifusum (fig.A &B)* was tested invitro against gram positive bacteria *Staphylococcus aureus* and gram negative bacteria *Pseudomonas aeruginosa*. The methanol extract showed better antibacterial activity against both the strains of bacteria (Table 1). Antibacterial activity was evaluated as the diameter of inhibition zone formed as a result of disc diffusion assay method. The methanol extract showed a larger inhibition zone (1.96cm) against *Pseudomonas aeruginosa (fig. C)*. Aqueous extract do not showed any inhibitory activity against the respective bacterial species. Thus the result proved that methanol was a potent solvent for extracting antibacterial agents from *Nostoc humifusum*.

Productions of antimicrobial compounds by cyanobacteria are well established. Most of them are secondary metabolites and may be extracellular (Jaki *et al.*, 1999) or intracellular (Asthana *et al.*, 2006). Antimicrobial activity of *Nostoc sp*. have been reported by many authors (Shweta *et al.*, 2012 ; Farag *et al.*, 2014).The present study proved that the cyanobacterium *Nostoc humifusum* is a potential source of antibacterial bioactive compounds. Further detailed research is needed for the isolation, identification and characterization of the active compounds responsible for the antibacterial activities.

Table 1: Antibacterial activity exhibited by *Nostoc humifusum*

Target Microorganism	Diameter of inhibition zone(cm)		
	Methanol	Water	Control(tetracycline)
Staphylococcus aureus	1.36 ± 0.10	-	2.5
Psuedomonas aeruginosa	1.96 ± 0.15	-	2.5

CONCLUSION

Cyanobacteria are promising sources of antibiotics and other pharmacologically active compounds. This preliminary study has established the antibacterial activity of *Nostoc humifusum*. The antibacterial activity shown by cyanobacteria depends on several factors like pH, growth conditions and the solvent used for preparing the extract. Detailed studies are required to characterize the compounds responsible for antibacterial activity.

ACKNOWLEDGEMENT

The support provided by the Head of the Department of Botany, University of Calicut is gratefully acknowledged.

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