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Research Article

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Antibacterial activity of marine red alga *Hypnea musciformis*

Shareef Khan M, Sridharan M. C.* and Abdul Nazar Y.

Department of Plant Biology and Plant Biotechnology, The New College, (Autonomous) Chennai 600 014, India.

ABSTRACT

Chloroform, methanol, chloroform: methanol (2:1 v/v), hexane and petroleum ether extracts of Hypnea musciformis from the coast of Kanyakumari were tested in vitro for their antibacterial activities against Escherichia coli, Salmonella typhi, Pseudomonas aeruginosa, Staphylococcus aureus and Proteus mirabilis with the disc diffusion method. Chloroform: methanol was the best solution for extracting the effective antibacterial materials from the Hypnea musciformis used in this experimental and compared with standard drug, Tetracycline. In the present study show the importance of in producing new bioactivity compounds having antibacterial activity.

Key words: Antibacterial activity, *Hypnea musciformis* and alga extract

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INTRODUCTION

Marine species have been used in a wide array of traditional remedies and provided a good source of antimicrobial activity. Algae are rich in phytochemical ingredient such as agar, carragenean and alginate, which were utilized in the field of medicine and pharmacy for decade [1]. The seaweeds have a unique place in traditional medicine of maritime nation as vermifuges, aesthetics and antibiotics in the treatment of cough, wounds, gout, goiter, hypertension, veneral diseases, cancer and a variety of other sickness [2-3]. The production of microbial inhibitory a substance from marine species was noted as early as in 1917 [4]. Resolution to the growing crisis of antibiotic resistance and their side effects are the breakthrough for search of new antimicrobial compounds from natural resources. Since then, several studies have been carried out to identify novel antimicrobial compounds from marine sources.

In the present study was undertaken to evaluate antimicrobial activity of the *Hypnea musciformis* However against selected strains of both Gram-positive and Gram-negative bacteria. *Hypnea musciformis* belongs to the order; Gigartinales of Rhodophyceae, Rhodophycophyta.

EXPERIMENTAL SECTION

Sample was collected from the sea coast of Kanyakumari, Tamil Nadu, India in the form of dry sample. Alga sample were cleaned at epiphytes and necrotic parts were removed. Sample was rinsed with sterile water to remove any associated debris. Sample was kept under sunshade for 7 days.

Crude extract preparation

For preliminary investigation, 20.0 g dry powdered material was extracted with 200 ml of desired solvent [chloroform, methanol, chloroform: methanol (2:1 v/v), hexane and petroleum ether] in cold maceration method using aspirated bottle and the extract thus obtained was dried *in vacu.*

Test Microorganisms

Clinical isolates of *Escherichia coli*, *Salmonella typhi, Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Proteus mirabilis* were gifted by Dr Agarwal's eye Hospital, Chennai, used as the test organisms. Disc diffusion method Bauer [5] was adopted for the determination of antibacterial activity of the extract residues. From the stock cultures of various test organisms, inoculum was prepared by subculturing each of the organisms on Muller-Hinton agar at 37^{o}C. Seeding of Muller-Hinton agar plates was done using the 24 hr culture with a cotton swab under aseptic conditions. The discs loaded with extract residues were aseptically placed on top of the seeded medium and gently pressed to ensure contact. The plates were then incubated at 37^{o}C. After overnight incubation, the plates were observed for zones of inhibition. Tetracycline was used as standard drug.

RESULTS AND DISCUSSION

The antibacterial activity of the crude solvent extracts of the *Hypnea musciformis* five solvent systems namely chloroform, methanol, chloroform: methanol (2:1 v/v), hexane and petroleum ether. The bacteria *Pseudomonas aeruginosa* were used as test organism (Table.1). In this preliminary investigation, the alga extract prepared with a mixture of chloroform: methanol (2:1 v/v) proved to be more effective than the other solvent system used in inhibiting the growth of *Pseudomonas aeruginosa* on Muller-Hinton agar plates. Petroleum ether exhibit only 77% maximum activity against the test organism (Table.1). While in methanol and chloroform of the alga were able to exhibit only 35 to 50% maximum activity against the test organism (Table.1). The hexane extract appeared to be poor growth of *Pseudomonas aeruginosa*. Based on these observations, further experiments on the antibacterial activities of the *Hypnea musciformis* were carried out to chloroform: methanol (2:1 v/v) extracts.

The chloroform: methanol (2:1 v/v), extract of the *Hypnea musciformis* were prepared as described earlier and testes at a concentration of 700 µg/disc by disc diffusion method against five pathogenic bacteria namely, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Proteus mirabilis*, *Salmonella typhi and Escherichia coli*. The results are presented in Table.2. The extract residues of alga recorded maximum activity against *Pseudomonas aeruginosa* with an inhibition zone of 5.4 mm for *Hypnea musciformis* which are quite high (Table.2). *Proteus mirabilis* and *Staphylococcus aureus* were also effectively inhibited by the extract residues of the *Hypnea musciformis* (Table.2). The extract residue of *Hypnea musciformis* recorded minimum activity against *Eschechia coli* and *Salmonella typhi* 42.5 to 53.7% (Table .2). The antibacterial activity of the chloroform: Methanols (2:1 v/v) extract residues of the *Hypnea musciformis* against the five pathogenic bacteria was compared with the standard antibiotics (Tables.2).

Table 1: Antibacterial activity of the crude solvent extracts of the *Hypnea musciformis*

S.No	Solvent used for extraction	Antibacterial activity (% maximum activity)
		Hypnea musciformis
1.	**Hexane**	20
2.	**Chloroform**	35
3.	**Methanol**	50
4.	**Petroleum Ether**	77
5.	**Chloroform: Methanol (2:1 v/v)**	100

Algae are eukaryotic organisms inhabited in salty sea water and is recognized to synthesize several bioactive compounds which show antimicrobial property [6].The present study was focused on *Hypnea musciformis* for the antibacterial activity against bacteria.

Marine genus synthesizes active constituents which are used in traditional and complementary medicine. Different varieties of marine algae were reported to contain active ingredients that can cure diseases. Nowadays, higher percentage of population prefers to use remedies of natural origin for curing illness as these claimed to produce less side effect [7]. Which clearly explain the activity of pertroleum ether and methanol extracts against *Eschechia coli* [8]. The antifungal, antiviral and antibacterial activities of saponins are well documented [9-10].

The present study differs from the previous study since the antibacterial activity was evaluated using chloroform: methanol (2:1 v/v) extract residues of the *Hypnea musciformis.* On evaluating the antibacterial property of, *Hypnea musciformis* the alga proved to be a potent antibacterial agent. The finding of this study also paves the way for further research to identify the specific bioactive compounds that is responsible for its claimed antibacterial activity. Maximum activities (zone of inhibition) for *Pseudomonas aeruginosa* were 5.4 mm (for *Hypnea musciformis* extract)

Table 2: Antibacterial activity of the crude petroleum extracts residue of the *Hypnea musciformis*

S.No	Test bacteria	Zone of inhibition (mm) ± S.E.	
		Hypnea musciformis	Tetracycline
1.	*Pseudomonas aeruginosa*	5.4 ± 0.057 (100)	3.2 ± 0.041
2.	*Staphylococcus aureus*	4.4 ± 0.145 (81.4)	2.4 ± 0.030
3.	*Proteus mirabilis*	3.8 ± 0.088 (70.3)	2.1± 0.037
4.	*Salmonella typhi*	2.9 ± 0.058 (53.7)	2.0± 0.029
5.	*Escherichia coli*	2.3 ± 0.033 (42.5)	1.2 ± 0.057

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