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Anti Microbial Activity of the Cnidarian Blue Button *Porpita porpita* (linneaus, 1758)

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Abstract: *Porpita porpita*, commonly known as the blue button, is a marine invertebrate. The present study was undertaken to characterize antimicrobial activity of methanol, butanol, ethanol and chloroform extract from the P. porpita. Antimicrobial properties of the blue botton *P. porpita* were tested against 10 pathogenic bacteria and 10 pathogenic fungi. This study represents the first report of antimicrobial activity of *Porpita porpita* collected from Pazhayar of South east coast of India. Results from the growth inhibition assays guided a fractionation scheme to find active compounds. The extract of antibacterial agent of Erythromycin showed activity against all the bacterial strains tested. The maximum activity showed against *Klebsiella pneumonia* (16mm). The lowest zone of inhibition was found in *Escheria.coli* (12mm). The extract of antifungal agent of Fluconazole showed activity against all the fungal strains tested. The maximum activity was found against *A. niger* (13mm). Rest of them showed no activity. It was concluded that the central disc region of *P. porpita* extracted using butanol solvent extraction exhibits anti bacterial and antifungal activity against human pathogens.

Key words: *Porpita porpita* · Central disc region · Methanol · Antibacterial and Antifungal activity

INTRODUCTION

It is well known that infectious account for high proportion of health problems. Microorganisms have developed resistance to many antibiotics and this has created many clinical problems in the treatment of infectious diseases [1]. This resistance has increased due to indiscriminated use of commercial antimicrobial drugs commonly used in the treatment of infectious diseases. This opens the door to search for new antimicrobial substances from various sources.

Cnidarians were for a long time grouped with Ctenophores in the phylum Coelenterata, but increasing awareness of their differences caused them to be placed in separate phyla. Cnidarians are said to have enormous antimicrobial activities, though yet not so much exploited. *P. porpita*, commonly known as the blue button, is a marine organism consisting of a colony of hydroid found in topical waters from California to the tropical Pacific. In this study, the cnidarian, *Porpita porpita* is used as the study animal to check its anti microbial properties.

Shapo *et al.* [2] examined the antimicrobial activity in the gorgonian *Leptogorgia virgulata* (common seawhip) from South Carolina waters and confirmed that partially-purified *L. virgulata* fractions collected from HPLC-MS fractionation were shown to have antimicrobial activity using *Micrococcus luteus* and *Vibrio harveyii*. They found that there is a bio active substance that is an active constituent of the innate immune system in *L. virgulata*.

Shinji Kasahara *et al.* [3] experimented on the nervous system evolved with the cnidarians. When assessing antibacterial activity in the freshwater polyp *Hydra*, they observed a strong correlation between the number of neurons present and the antibacterial activity. Tissue lacking neurons had a drastically enhanced antibacterial activity against Gram-positive (*Bacillus subtilis*) and Gram-negative (*E. coli*) bacteria.

Futher more, Anselmo *et al.* [4] described antimicrobial peptides from marine invertebrates as a new frontier for microbial infection control.

The potential of marine invertebrates as a source of biologically active products is largely unexplored. Hence, a broad, based screening of marine organisms for bioactive compounds is necessary. In the present study antimicrobial activity in the central disc region of *Porpita porpita* against human pathogens was investigated.

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MATERIALS AND METHODS

Collection and Extraction: Live specimens of *P.porpita* were collected from the Pazhayaar coastal region (Lat 11° 21'32.27" N; long 79° 49'24.92" E). All specimens were collected with a spatula, placed in a recipient with sea water, transported to the laboratory.

The central disc region was removed from the animal with fine scissors. The region was suspended in the solvents over night and homogenated using clean Mortar and Pestle. The homogenate centrifuged at 5000 rpm for 15 mins and the supernatant was collected and the extract was stored in refrigerator.

Microbial Strains Used: Antibacterial activity of central disc region was determined against 10 bacterial strains viz., *Vibrio parahemolyticus, V. cholerae, Salmonella typhi, Pseudomonas aeruginosa, Proteus mirabilis, Salmonella paratyphi, E.coli, S.aureus, Klebsiella pneumonia andK.oxytoca aw well as* 10 fungal strains viz., *A.niger, C. albicans, A.flavus, Mucor sp, A.alternaria, Pencillium sp, Rhizopus, T. rubrum, T.mentagarophytes* and *E.floccosum.* These clinical bacterial and fungal strains were obtained from the department of medical microbiology (Raja muthaiah medical college and hospital) Annamalai University, Annamalai nagar.

Antimicrobial Assay: The spectrum of antibacterial and antifungal activitiy was studied by using the techniques of Juan Luis Morales-Landa *et al.* [5]. Antibacterial and antifungal activity was expressed in terms of diameter of zone of inhibition was measured in mm using Vernier caliper or scale and recorded.

RESULTS

Biological Screening: Antibacterial activity in central disc region extraction of *P. porpita* was used for the present study. The positive control was also used. Investigation against a range of ten different bacterial strains was used of which both, gram-positive bacteria and gram negative bacteria were present.

Antibacterial Activity: The zone of inhibition in different bacterial strains against *P. porpita* extraction is shown in Table 1. Among the various strains maximum diameter of (16 mm) zone of inhibition was recorded in *K.pneumonia*

Table 1: Antibacterial activity of *Porpita porpita*

			Zone of Inhibition (mm)			
	Name of the Bacteria	Activity of Agent Erythromycin (C)	Chloroform	Butanol	Methanol	Ethanol
1	*Vibrio parahemolyticus*	+Ve	-	-	-	-
2	*V. cholerae*	+Ve	-	-	-	-
3	*Salmonella typhi*	+Ve	-	-	-	-
4	*Pseudomonas aeruginosa*	+Ve	-	-	-	-
5	*Proteus mirabilis*	-Ve	-	-	-	-
6	*Salmonella paratyphi*	+Ve	-	-	-	-
7	*E.coli*	+Ve	-	12	-	-
8	*S.aureus*	+Ve	-	-	-	-
9	*Klebsiella pneumonia*	-Ve	-	16	-	15
10	*K.oxytoca*	+Ve	-	14	-	-

Table 2: Antifungal activity of *Porpita porpita*

			Zone of Inhibition (mm)			
S.No	Name of the Fungus	Activity of Agent Flucunozole ©	Chloroform	Butanol	Methanol	Ethanol
1	*A.niger*	+Ve	-	13	-	-
2	*C.albicans*	+Ve	-	-	-	-
3	*A.flavus*	+Ve	-	-	-	-
4	*Mucor sp*	+Ve	-	-	-	-
5	*A.alternaria*	-Ve	-	11	-	-
6	*Pencillium sp*	+Ve	-	-	-	-
7	*Rhizopus*	+Ve	-	-	-	-
8	*T.rubrum*	+Ve	-	12	-	-
9	*T.mentagarophytes*	-Ve	-	-	-	-
10	*E.floccosum*	+Ve	-	-	-	-

strain in butanol and ethanol extraction (15mm) and also strains of *E.coli* (12mm) and *K.oxytoca* (14mm) in the butanol extraction. No clear zones were formed in the other strain that was used to check the antibacterial activity.

Antibacterial Activity of Positive Control Erythromycin: The extract of antibacterial agent of Erythromycin showed activity against all the bacterial strains tested. The maximum activity was showed against *K. pneumonia* (16mm).

Antifungal Activity: The antifungal activity of the extraction of *P.porpita* against the fungal strains was performed by the disc diffusion technique. The maximum activity was observed in *A.niger* (13mm), *T.rubrum* (12mm) and in *A.alternaria* (11mm) only in the butanol extracts (Table 2).

Antifungal Activity of Positive Control of Fluconazole: The extract of antifungal agent of Fluconazole showed activity against all the fungal strains tested. The maximum activity was showed against *A. niger* (13mm).

DISCUSSION

Shinji Kasahara *et al.* [3] studied the antibacterial activity of Hydra polyp against Gram-positive (*Bacillus subtilis*) and Gram-negative (*E. coli*) bacteria compared to control tissue. A promising novel use of antimicrobial peptides as effective drugs in human and veterinary medicine could be based on their unusual properties and synergic counterparts as immune response humoral effectors, in addition to their direct microbicidal activity. This has been seen in many other marine proteins that are sufficiently immunogenic to humans, not necessarily in terms of antibody generation but as inflammation promoters and recruitment agents or immune enhancers [4].

In the present investigation, pronounced antimicrobial activity has been observed against some bacterial strains. The butanol extraction of the central disc region of *P. porpita* shows activity against both bacterial and fungal strains. In antibacterial activity the maximum diameter of zone of inhibition was recorded in *K. pneumonia* strain in butanol and ethanol extracts. In the antifungal activity, the central disc region extraction of *P. porpita* against the fungal strains showed maximum activity against *Aspergillus niger* in the butanol extract. Antimicrobial, antiprotozoal and toxic activities of crude extracts obtained from six cnidarian species was found to be most active against the yeast *Candida albicans* and the protozoan *Giardia lamblia* [6].

Most recently, a novel antimicrobial peptide was purified from the mesoglea of the jellyfish *Aurelia aurita*, one of the most common and widely recognized types of jellyfish found near the coasts in the Atlantic, Arctic and Pacific Oceans [7]. Aurelin, a 40-residue antimicrobial peptide with a molecular mass of 4296.95 Da was shown to exhibit activity against Gram-positive (*Listeria monocytogenes*, strain EGD) and Gram-negative (*Escherichia coli*, strain ML-35p) bacteria.

Antimicrobial peptides of animal origin may be an effective alternative or additive of conventional antibiotics for therapeutic use. The recent identification of highly active antimicrobial peptides in hydra and jellyfish show that antimicrobial peptides from marine and freshwater cnidarians may represent a largely unexploited resource that can afford the design of new antibiotics with broad-spectrum antimicrobial activity [7].

These preliminary analyses of the marine invertebrate, *Porpita porpita,* indicates that the central disc region of it may contain antimicrobial peptide, the antimicrobial assay done may serve as a preliminary data. In conclusion, further studies are required. especially on butanol solvent extraction system to examining the main compound or strain of *Porpita porpita* that may have antimicrobial property, which might prove to be of high use in the pharmaceutical industry as a component of antibiotics.

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