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Research Article

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Antifungal Activities of Marine Polychaetes *Namalycastis fauveli*

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ABSTRACT

The crude extract of polychaete Namalycastis fauveli was screened for its Antifungal activities. Extracts were obtained from the whole body tissues of the animals to test against different pathogenic bacteria (Escherichia coli, Staphylococcus aureus and Salmonella typhi). The crude extracts were also fractionated and elutions were made with methanol. Eluted fractions were assayed for Antifungal activity followed the disc diffusion method. In polychaete Namalycastis fauveli fraction I (66%), the zone of inhibition against Staphylococcus aureus is significantly (P<0.05) recorded. In case of Bacillus substilus, it is significantly (P<0.05) observed in fraction I, V, and VI (31%) and III (21%), IV (12%) and fraction VII (7.81%). The zone of inhibition of E.coli is significant (P<0.05) observed in fraction VII (27%).

Keywords: Antimicrobial activities; Polychaetes; *Namalycastis fauveli*

INTRODUCTION

A number of terrestrial and marine organisms are used to fend off a wide range of microorganisms including bacteria and fungi by employing "Anti-Microbial Peptides (AMPs)". These AMP's are ribosomally synthesized from proteinogenic amino acids. In marine invertebrates AMPs are a primary component of innate immune mechanisms. Naturally occurring peptides are either synthesized by ribosomal machinery from 20 proteinogenic amino acids or by large enzymes and enzyme complexes called Non-Ribosomal Peptide Synthase [1]. Antimicrobial peptides (AMPs) that involved in marine invertebrate immunity are ribosomal peptides (gene-encoded peptides) and classified into: a) linear α-helical peptides, b) peptides with intramolecular disulfide bridges, c) β-sheet and small proteins, and d) peptides with one or two predominant amino acids [2-6]. The majority of AMPs are amphiphilic and cationic containing both hydrophilic and hydrophobic surfaces showing antimicrobial activity by forming pores in microbial membranes or disrupting membranes [5-7]. A total of 1,518 AMPs are listed in the second version of Antimicrobial Peptide Database, among which 442 peptides are antifungal [8]. Non-ribosomal peptides are found in sponges, molluscs and tunicates that are composed of unusual amino acids including D-amino acids and contain organic acids in addition to amino acids as cases of depsipeptides. These exhibits a wide range of biological activities such as antimicrobial, cytotoxic, and enzyme inhibitory.

Marine polychaetes (phylum Annelida) are useful to treat several patho-physiological conditions such as arthritis, osteoporosis, bone cancer etc. The bioactive compound has been isolated from a marine annelid, *Arenicola marina* by Mynderse [9]. The compound arenicins are 21 residue peptides which completely killed *E. coli* within 5 mins at a concentration of 5 μM probably by membrane permeablization [10]. The most important antimicrobial peptide (AMP) to be isolated from marine annelid is Hedistin. It is purified from the ragworm, *Nereis diversicolor*. Both native and synthetic hedistins are active against Gram-positive and Gram-negative bacteria [11]. Chain [12] isolated the potent bioactive compounds from marine polychaete *Glycera dibranchiate.* It revealed that coelomic fluid annelid exhibited Antifungal activity. Elayaraja [13] reported that the water, methanol and acetone extract from the whole body tissue of polychaete *Perinereis cultrifera* has potent Antifungal and antifungal activities. Among all the

extracts, methanol has shown maximum Antifungal activity against *Staphylococcus aureus* (8.0 mm) and minimum against *Klebsiella oxytoca* (1.0 mm) both in methanol and acetone extracts. For antifungal pathogens, methanol extract showed maximum activity against *Rhizopus* sp. (12.0 mm) and minimum against *Aspergillus niger* (2.0 mm) in water extract. Also, the trace activity was noticed in another fungal species such as *Mucor* spp. and *Aspergillus niger* in both water and acetone extracts. Similarly, Lovell [14] described the Antifungal activity from volatile halogenated secondary metabolites produced by the polychaetes. Benkendorff [15] tested the Antifungal activity 4 species of polychaetes against 4 marine bacterial pathogens. A review of literature on Antifungal activity of annelids particularly polycheates revealed that only a sketchy information is available. On the contrary, a large number of works is available in other groups of invertebrates. Considering the scarcity of information in this line, an attempt was made presently with a view to evaluate the potential Antifungal activities of polychaetes *Namalycastis fauveli* .

MATERIALS AND METHODS

Collection and Preparation of Samples

The Polycheate *Namalycastis fauveli* were collected during the lowtide of the intertidal area of the west coast of Ratnagiri, India. The collected samples were rinsed with sterile sea water to remove associated debris and salt. Methanol and Methylene chloride extract of the Polycheate *Namalycastis fauveli* was prepared. The organic extract was fractionated by the Thin Layer Chromatography on silica gel. The extracts were fractionated using. The solvent system used was chloroform: methanol (9:1). Zone of separation were observed under ultraviolet florescence using 230-240 nm and 250-270 nm lamp. Separated material was recovered from the plates by scraping and eluted with HPLC grade methanol. Methanol was removed by rotary evaporation under vacuum for using them for the Antifungal activity. Three species of pathogenic bacteria namely *Escherichia coli, Staphylococcus aureus,* and *Salmonella typhi* were used to screen the antifungal activity of polychaete extract. For screening Antifungal activity of polychaetes *Namalycastis fauveli* fractions Paper disc method was used Bauer [16]. This method was based on diffusion capacity of test chemicals through an agar medium. To determine the effect of the polychaete against bacteria (*Bacillus substilus, Bacillus aurius* and *Escherichia coli*) applied 20 µl of pure fraction to a sterile filter paper disc (6 mm in diameter). Allow to air dry for 1 hr to remove traces of the carrier solvent (methanol), than placed on a newly spread lawn of fungi and bacteria on each replicate plate. One disc treated with polychaete fractions concluded as experiment, along with the disc treated with methanol as control. Zone of inhibition (i.e., the distance from the edge of the filter paper disc to the growing edge of the microbes) is measured by using minimum inhibitory concentration (MIC). The growth area is almost round or oval. Hence, the growth area (mm) was measured using the mathematical formula D=2r.

RESULTS

Antifungal activity (zone of inhibition in mm) of fractionate extract of *Namalycastis fauveli*. The area of growth of control *Staphylococcus aureus* was a found to be 30 mm (Table 1). The area of growth in the experiment petridish applied with polychaete *Namalycastis fauveli* was found in fraction- I (10 mm), II, III, IV, V, VI and VII (30 mm). The zone of inhibition significantly ($P<0.05$) recorded in fraction I (66%). There was no zone of inhibition occurred against *Staphylococcus aureus* in polychaete fraction II, III, IV, V, VI and VII.

Table 1: The area of growth

Fractions	Growth of *Staphylococcus aureus*		% Inhibition	Growth of *Bacillus substilus*		% Inhibition	Growth of *E. coli*		% Inhibition
	control	Experimental		Control	Experimental		Control	Experimental	
I	30 mm	10 mm	66%	32 mm	22 mm	31%	32 mm	32 mm	0%
II	30 mm	30 mm	0%	32 mm	22mm	31%	32 mm	32 mm	0%
III	30 mm	30 mm	0%	32 mm	25 mm	21%	32 mm	31 mm	3%
IV	30 mm	30 mm	0%	32 mm	28 mm	13%	32 mm	32 mm	0%
V	30 mm	30 mm	0%	32 mm	22 mm	31%	32 mm	31 mm	3%
VI	30 mm	30 mm	0%	32 mm	25 mm	21%	32 mm	32 mm	0%
VII	30 mm	30 mm	0%	32 mm	28 mm	13%	32 mm	27 mm	15%

The area of growth of control *Bacillus substilus* was a found to be 32 mm (Table 1). The area of growth in the experiment petridish applied with polychaete *Namalycastis fauveli* was found in fraction- I, V, and VI (22 mm) and fraction- II (32 mm) in diameter, III (25 mm), IV and VII (28 mm). The zone of inhibition significantly ($P<0.05$) observed in fraction I, V, and VI (31%) and III (21%), IV (12%) and fraction VII (7.81%). There was no inhibition

occur against *Bacillus substilus* in polychaete fraction- II. Total growth level of control *E. coli* was a found to be 32 mm (Table 1). The area of growth inhibited by the polychaete *Namalycastis fauveli* was found in fraction- I, II, IV, and VI (32 mm) and fraction -III and V (31 mm), and fraction VII (27 mm). The zone of inhibition was recorded in fraction III and V (3%) not significant (P>0.05), however fraction VII (27%) is significant (P<0.05). There was no inhibition occur against *E. coli* in polychaete fraction- II, IV, VI, and I.

DISCUSSION AND CONCLUSION

Natural product drugs play a vital role in pharmaceutical care. Nature is an attractive source of new therapeutic candidate compounds as there a tremendous chemical diversity is found in millions of species of plants, animals, marine organisms and microorganisms. The results of minimum inhibitory concentrations of various fractions reveals that polychaete *Namalycastis fauveli* fraction I (66%), the zone of inhibition against *Staphylococcus aureus* is significantly (P<0.05) recorded. In case of *Bacillus substilus*, it is significantly (P<0.05) observed in fraction I, V, and VI (31%) and III (21%), IV (12%) and fraction VII (7.81%). The zone of inhibition of *E. coli* is significant (P<0.05) observed in fraction VII (27%). Similarly, Andra [17] studied the significance of the cyclic structure and found the arginine residues for the Antifungal activity of arenicin-1 and its interaction with phospholipid and lipopolysaccharide model membranes also screening of Antifungal activity in the oercula of gastropods [18,19]. Benkendorff observed the chemical defense in the egg masses of benthic invertebrates and assessed the Antifungal activity in 39 molluscus and 4 polychaetes [15]. Brogden has studied antimicrobial peptides and pore formers or metabolic inhibitors in bacteria [7]. Bulet also studied anti-microbial peptides from invertebrates to vertebrates [2]. Chain [12] conducted the study on Antifungal activity of the coelomic fluid from polychaetes *Glycera dibranchinata*. II Partial purification and biochemical characterization of the active factor and observed the same results as mentioned as above. Elayaraja screened methonol and acetone extract of polychaetes *Perineris cultrifera* for its antifungal and antifungal activities [13]. He stated that methanol shows maximum antifungal activity against *S. aureus* (8.0 mm) and minimum against *K. oxytoca* (1.0 mm) both in methanol and acetone extract which are in the same line as above. Lovell studied the antifungal activity of halogenated volatile secondary metabolites produced by polychaetes and noticed that these metabolites affects the rate respiration [14]. The marine worms are found in sediments indicating the requirement of antimicrobial strategy for their survival. AMPs have been isolated from polychaete and echiuroid worms. Arenicin-1 and -2 are 21-residue AMPs are isolated from coelomoycytes of the polychaete *Arenicola marina* showing no structural similarity to any AMPs reported [10]. Arenicins are cationic peptides having two antiparallel β-strands and one disulfide bond between Cys3 and Cys 20 [10]. Arenicin-1 has shown antifungal activity against *C. albicans, C. parasilosis, Malasseria furfur, Trichosporon beigelli* and *Trichophyton rubrum* with MICs of 4.5-9 μM comparable to that of mellitin by disrupting fungal phospholipid membranes [20]. The same screening was done by various workers [3,5,8,9,10,11,18,19] for the antimicrobial activity by using different marine crude extract. Baring these works, the present study could not be discussed at length as there were not many works in this line.

REFERENCES

[1] JA McIntosh; MS Donia; EW Schmidt. *Immunol Rev.* **2004**, 169-184.
[2] JA Tincu JA; SW Taylor. *Antimicrob Angents Chemother.* **2004**, 48, 3645-3654.
[3] REW Hancock; KL Brown; N Mookherjee. *Immunobiology.* **2006**, 11, 315-322.
[4] H Jenssen; P Hamill; REW Hancock. *Clin Microbiol Rev.* **2006**, 19, 491-511.
[5] MR Yeaman MR; NY Yount. *Pharmacol Rev.* **2003,** 55, 27-55.
[6] KA Brogden. *Rev Microbiol.* **2005**, 3, 238-250.
[7] G Wang G; X Li; Z Wang. *Nucleic Acids Res.* **2009**, 37, 933-937.
[8] JK Mynderse; M Moore; M Kashiwagi; T Norton. *Science.* **1997**, 196, 538-540.
[9] TV Ovchinnikova TV; GM Aleshina; SV Balandin; AD Krasnosdembskaya; ML Markelov; EI Frolova. *FEBS Lett*. **2004**, 577, 209-214.
[10] AD Tasiemski; F Schikorski; M le Croq; C Van camp; B Wichlacz; PE Sautiere. *Dev Comp Immunol.* **2007**, 31, 749-762.
[11] BM Chain; RS Anderson. *Biol Bull.* **1983**, 164, 41-49.
[12] S Elayaraja; P Murugesan; S Vijayalakshmi; T Balasubramanian. *Indian J Mar Sci*. **2010**, 39(2), 257-261.
[13] CR Lovell; C Steward; T Philips. *Mar Ecol Prog Ser*. **1999**, 179, 241-246.
[14] K Benkendorff; AR Davis; JB Bremmer. *J Invertibrates Pathol*. **2001**, 78, 109-111.
[15] AW Baur; TM Shemis; WHM Kirby. *Am J Clin Pathol.* **1966**, 45, 493.

[16] J Andra; MU Hammer; J Grötzinger; I Jakovkin; B Lindner; E Vollmer. *Biol Chem.* **2009**, 39, 337-349.
[17] W Pan; X Liu; F Ge; J Han; T Zheng. *J Biochem.* **2004**, 135, 297-304.
[18] WS Sung; SH Park; DG Lee. *FEBS Lett.* **2008**, 582, 2463-2466.
[19] C Park; DG Lee. *Biophys Acta.* **2009**, 1788, 1790-1796.
[20] TP Anand; EJK Patterson. *Phuket Mar Biol Center Spl Pub.* **2001**, 215-217.