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# ANTIMICROBIAL ACTIVITY OF VARIOUS TISSUE EXTRACTS AND COELOMIC FLUID OF STARFISH, *PROTOREASTER LINCKII* (BLAINVILLE, 1830)

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**ARTICI**

**Key words**

Antimicrobial, Coelomic fluid, Lectin, *Protoreaster linckii*, Starfish

**ABSTRACT**

Antimicrobial potential of solvent extracts of diverse tissues and coelomic fluid lectin of starfish, *Protoreaster linckii* was evaluated against five human pathogenic bacteria, two fungal pathogens and five fish pathogenic bacteria. All the tested pathogens were sensitive to the butanol and ethyl acetate extracts of *P. linckii* tissues except the crude coelomic fluid which revealed sensible activity against fish pathogenic bacteria, *Staphylococcus aureus* and fungi *Aspergillus niger* and *Aspergillus flavus*. Human pathogenic bacteria, *S. aureus*, *E. coli* and *P. aeruginosa* were sensitive to the chloroform extracts of digestive gland, gonad and tube feet. Correspondingly the digestive gland extract and coelomic fluid were effective against the fish pathogenic bacteria and human pathogenic fungi. Acetone extracts of digestive gland, tube feet and gonad exhibited good to moderate activity against most of the pathogens tested. Methanolic extract of *P. linckii* was found to be less effective in inhibiting the growth of human pathogenic bacteria whereas the other tested pathogens were sensitive to the extract. Among the different solvents used for extraction, butanol showed better antimicrobial activity against the pathogens.

## INTRODUCTION

Echinoderms are a opulent source of natural bioactive products and contain antibacterial, antifungal, antiviral, antitumor, anticoagulant, cytotoxic, haemolytic, antithrombotic and even anti-HIV agents [1, 2, 3, 4, 5]. Among echinoderms, the research on starfish has expanded rapidly over the past few years, which has been prompted by the discovery of a variety of unique structures and the interest in their pharmacological properties [6]. Sea stars are benthic free living echinoderms and have evolved with rich sources of bioactive metabolites such as steroidal glycosides, steroids, anthraquinones, alkaloids, glycolipids and phospholipids [7,8,9]. A variety of antimicrobial factors, including steroidal glycosides[10], polyhydroxylated sterols[11], naphthoquinone pigments[12], lysozymes[13,14], complement like substances[15]and antimicrobial peptides[16] have also been isolated from echinoderms. Li *et al* [17] has reviewed many AMPs (antimicrobial peptides) from echinoderms which has strong antimicrobial activity against bacterial, fungal and viral pathogens. Coelomocytes mediate the cellular responses to immune challenges through phagocytosis, encapsulation, cytotoxicity and the production of antimicrobial agents. In addition, a variety of humoral factors found in coelomic fluid, including lectins, agglutinins, lysins, acute phase proteins and antimicrobial factors[18,19] are

important in host defense against pathogens and other foreign substances. Anbukkarasu *et al* [20] observed both antibacterial and antifungal activity in the starfish, *Luidia maculata*. Haug *et al* [21] detected antibacterial activity in the body wall, eggs, intestinal organs, cell free coelomic fluid and the coelomocytes of starfish, *Asterias rubens*. The crude extracts and fractions of starfish, *Stellaster equestris* had remarkable antimicrobial activities against human bacterial pathogens[22]. Jung[23] reported that the fractions obtained from *Asterias amurensis* and *Asterina pectinifera* showed strong activity against various human bacterial and fungal pathogens. Coelomocyte extract from the starfish, *Asterias rubens* showed antimicrobial activity and several proteins/ peptides with molecular mass of around 2 kDa was isolated[24,25]. The methanol and ethyl acetate extracts of starfish, *Astropecten indicus* showed high antimicrobial activity against pathogenic bacteria[26]. Prabha *et al* [27] accounted that the body wall extract of sea star *Pentaceraster affinis* showed considerable activity against *Shigella flexneri*, *Acinetobacter* sp. and moderate activity against *Streptococcus pyogenes*. Since echinoderms are known to possess antimicrobial effect, an attempt was undertaken to study the antimicrobial activity of different tissue extracts and coelomic fluid of the starfish, *Protoreaster linckii*.

## MATERIALS AND METHODS

### Experimental animal and Sample Collection

The starfish, *Protoreaster linckii* was collected by dip net fishing from in and around Kanyakumari district. Coelomic fluid was collected by cutting off the tip of the arm. Digestive glands, gonad, tube feet and arm were carefully dissected, pooled and stored at -20ºC prior to extraction.

### Preparation of extracts for antimicrobial activity

Starfish tissue extracts were prepared following the method of Karthikeyan *et al* [28] with slight modification. 1 g each of digestive glands, gonad, tube feet and arm were soaked in 10 volumes (v/w) of 70% methanol, acetone, chloroform, butanol and ethyl acetate and kept for three days at room temperature. After three days, the tissues were homogenized, extracted and again kept for three days. The extracts were filtered through Whatman No 1 filter paper, concentrated by evaporating in room temperature to give a dark gummy mass and used for the antimicrobial agar disc diffusion assay.

### Test microorganisms and culture media

Test microorganisms, fish pathogenic Gram positive bacteria (*Staphylococcus aureus*) and Gram negative bacteria (V*ibrio harveyi*, *Aeromonas hydrophila*, *Pseudomonas aeruginosa*, *Proteus mirabilis*) were obtained from Inbiotics, Nagercoil. Human pathogenic Gram positive bacteria [*Staphylococcus aureus* (MTCC 3160)], Gram negative bacteria [*Escherichia coli* (MTCC 443), *Pseudomonas aeruginosa* (MTCC 424), *Klebsiella pneumoniae* (MTCC 3040), *Proteus vulgaris* (MTCC 744)] and human fungal pathogens *Aspergillus niger* (MTCC 1344) and *Aspergillus flavus* (MTCC 7589) were purchased from Microbial Type Culture Collection and Gene Bank, IMTECH, Chandigarh and were used for antimicrobial assay of tissue extracts. Bacterial strains were cultured in Muller Hinton Agar and fungal strains were cultured using Sabouraud Dextrose Agar.

### Antimicrobial assay

*In vitro* antimicrobial assay was carried out by disc diffusion technique[29]. Solvent extracts of digestive glands, gonad, tube feet, arm and coelomic fluid of starfish *Protoreaster linckii* were used for the assay. Sterile discs of 6 mm were impregnated with extracts of 50 µl and allowed to dry at room temperature and extract loaded discs were placed on agar

plates seeded with microorganisms and incubated at 37°C for 24 hours. The susceptibility of the test organisms were determined by the diameter of the zone of inhibition and then measured in mm. The amikacin discs were used as a positive control for bacteria, flucanazole for fungi and solvent discs were used as a negative control. All the extracts were tested in triplicate and the results were expressed as mean ± SD of three independent values.

**RESULTS**

**Antimicrobial Activity**

Antibacterial and antifungal activity of crude tissue extracts of starfish *P. linckii* was screened against five human pathogenic bacteria and two fungi and five fish pathogenic bacteria. Among the five extracts (methanol, acetone, chloroform, butanol and ethyl acetate) of digestive glands, gonad, tube feet, arm and coelomic fluid tested, butanol extract was found to be effective against the pathogens. All the tested human and fish pathogenic bacteria were sensitive to the butanol extracts of starfish tissues and exhibited maximum zone of inhibition except the crude coelomic fluid. The fungal pathogens *A. niger* and *A. flavus* recorded a high inhibitory zone with all the tested extracts (Table 1). Ethyl acetate extracts of starfish tissue showed antimicrobial activity against most of the human pathogenic bacteria tested. Digestive glands and gonad recorded a maximum zone of inhibition against fish pathogenic bacteria, *S. aureus*, *V. harveyi* , *A. hydrophila* and *P. mirabilis*. The ethyl acetate treated coelomic fluid showed high activity only with fish pathogenic bacteria *P. aeruginosa* (14 mm) while *S. aureus* was sensitive to crude coelomic fluid (11 mm). Similarly the growth of *A. niger* and *A. flavus* was inhibited only by digestive gland and crude coelomic fluid (Table 2). Chloroform extracts of digestive glands, gonad and tube feet showed moderate activity against *S. aureus* while gonad and tube feet exhibited good activity against *E. coli*. *P. aeruginosa* was sensitive to digestive gland and gonad. Among the fish pathogenic bacteria tested, antimicrobial activity was noted with gonad extract. *V. harveyi* and *A. hydrophila* was sensitive to tube feet and *P. aeruginosa* to digestive gland. The crude coelomic fluid was effective against *A. niger* and *A. flavus* (11 mm and 12 mm) and a maximum zone of inhibition of was observed with digestive gland extract against *A. flavus* (15 mm) (Table 3). The acetone extract of digestive glands showed a maximum zone of inhibition of 12 mm against human pathogenic bacteria *S. aureus*, 13 mm and 11 mm against fish pathogenic bacteria *S. aureus* and *P. mirabilis* respectively. Human pathogenic bacteria *P. aeruginosa* and fish pathogenic bacteria *P. mirabilis* were highly sensitive to the gonad extract. Tube feet extracts inhibited the growth of human pathogenic bacteria *S. aureus*, *K. pneumoniae* and *P. vulgaris* while the arm extract showed moderate activity against *E. coli* and *P. aeruginosa*. Among the human fungal pathogens tested, the growth of *A. flavus* was highly inhibited by arm (18 mm), crude coelomic fluid (15 mm) and tube feet (11 mm) whereas *A. niger* was inhibited by crude coelomic fluid (12 mm) and tube feet (13 mm) (Table 4). Methanolic tissue extracts of starfish, failed to inhibit the growth of human pathogenic bacteria except the tube feet that showed trace activity against *E. coli*. Among fish pathogenic bacteria, *S. aureus* was sensitive to digestive gland and crude coelomic fluid and *A. hydrophila* to gonad (17 mm). Gonad exhibited good activity against *A. niger* (16 mm) and arm had trace activity against *A. flavus* (Table 5).

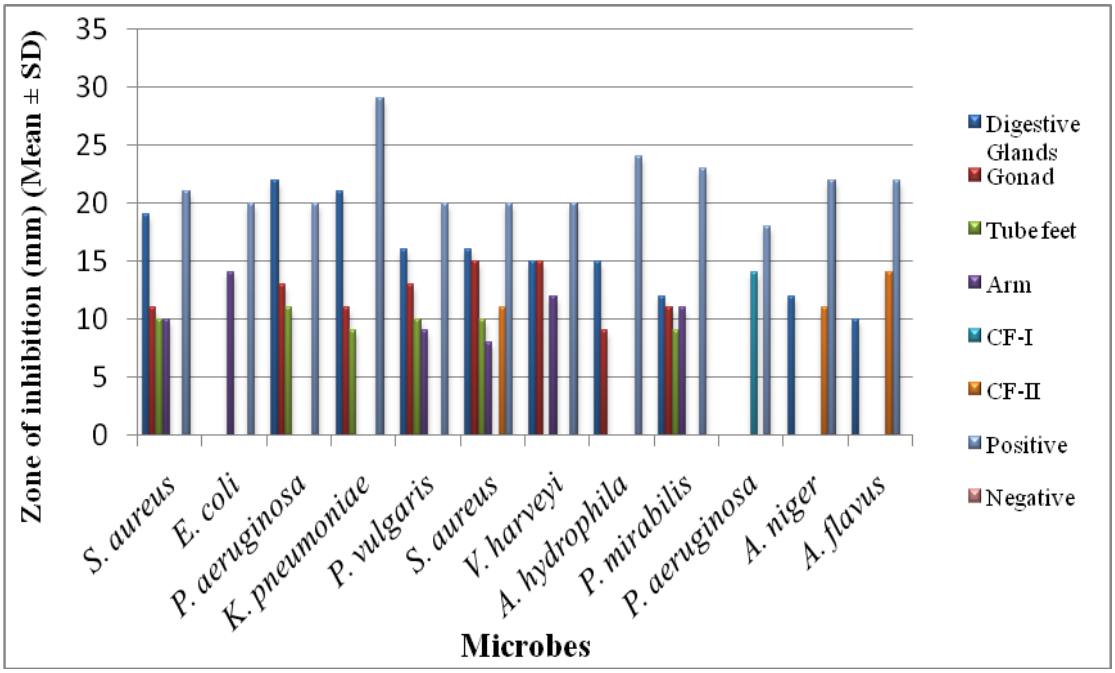


**Fig. 1. Antimicrobial activity of butanol extract of starfish, *P. linckii***

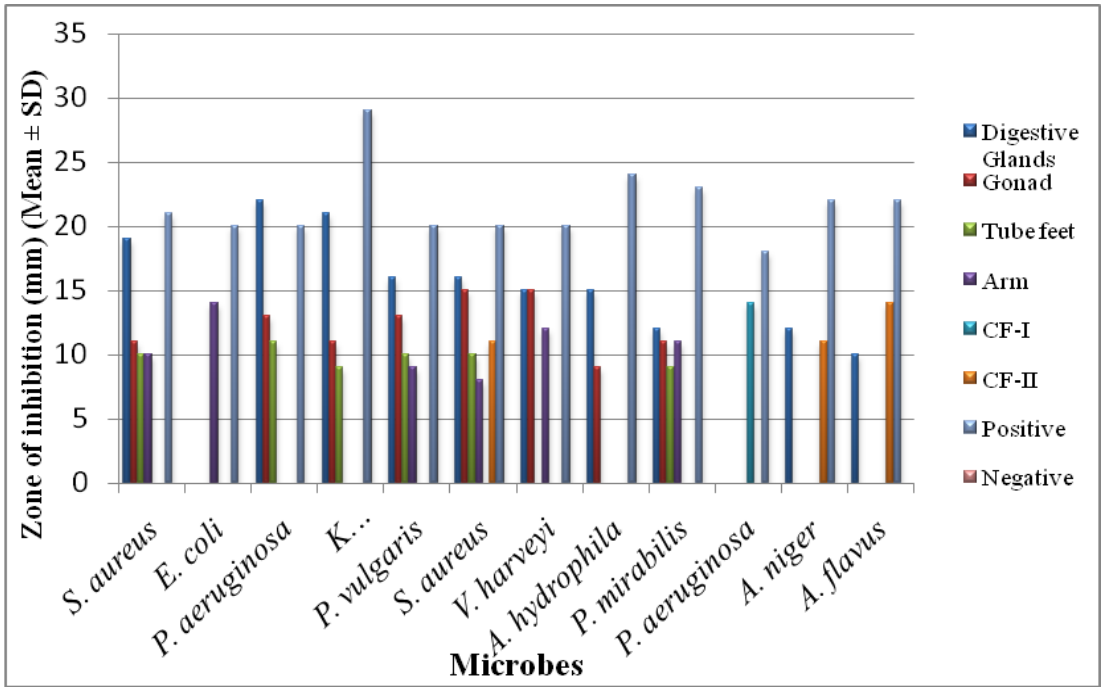


**Fig. 2. Antimicrobial activity of ethyl acetate extract of starfish, *P. linckii***

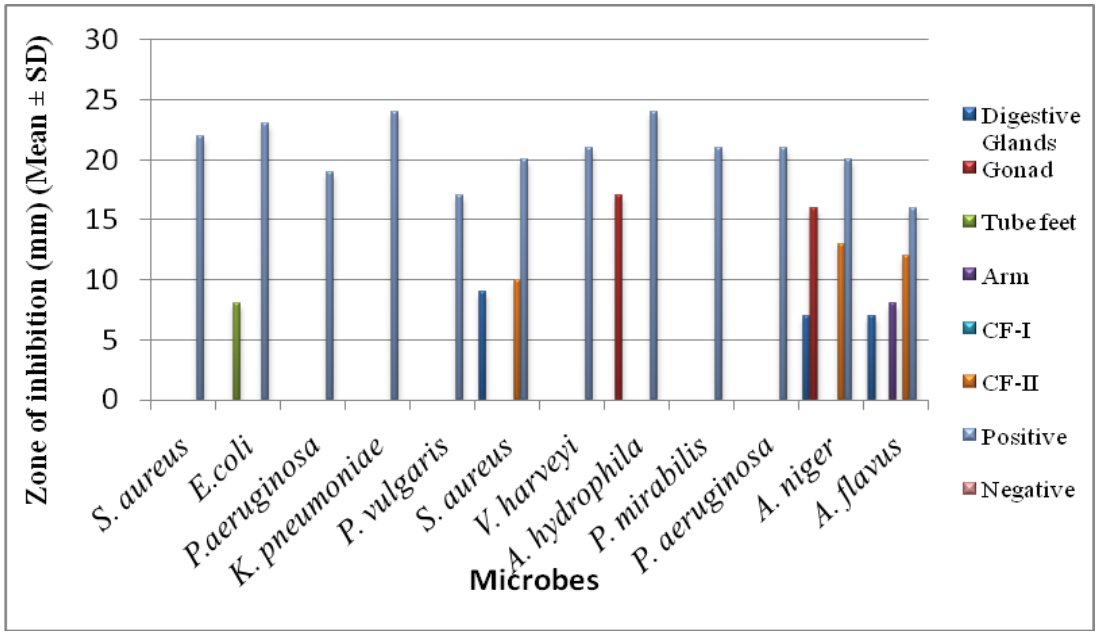


**Fig. 3: Antimicrobial activity of chloroform extract of starfish, *P. linckii***

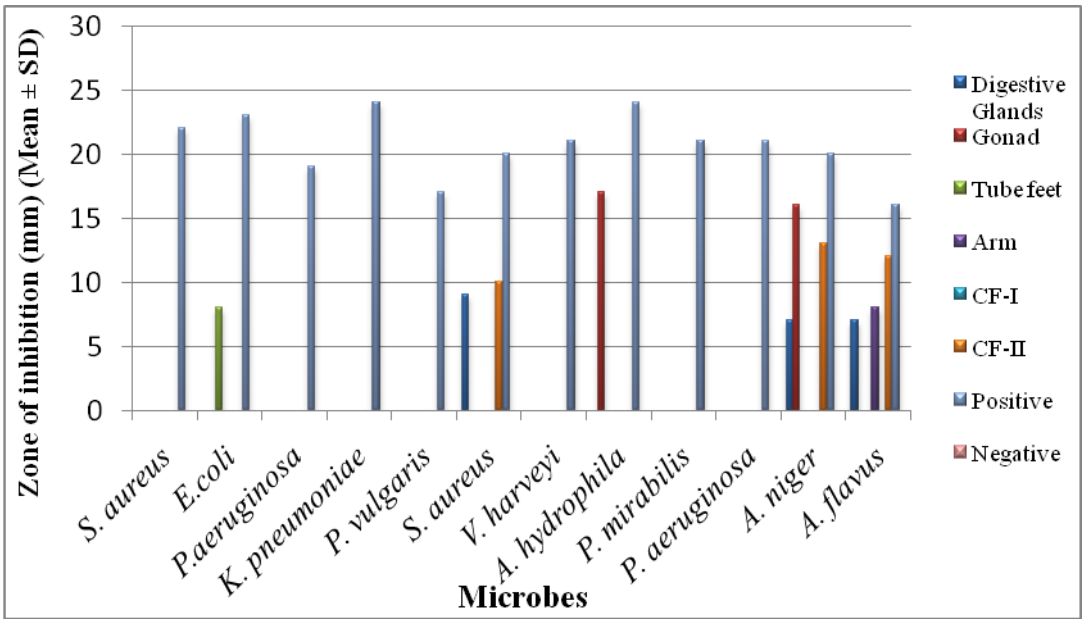


**Fig. 4: Antimicrobial activity of acetone extract of starfish, P. linckii**

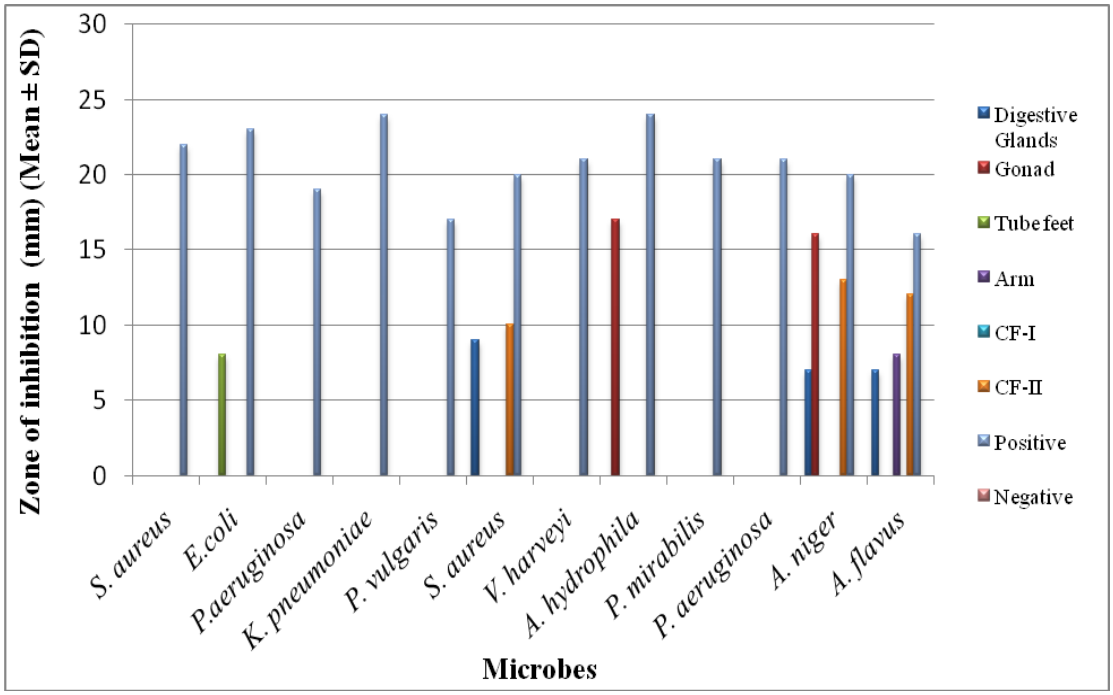


**Fig. 5: Antimicrobial activity of methanolic extract of starfish, P. linckii**

**Table 1. Antimicrobial activity of butanol extract of starfish, *P. linckii***

| Microbes | | Zone of inhibition (mm) (Mean ± SD) | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Digestive glands | Gonad | Tube feet | Arm | CF- I | CF- II | Positive | Negative |
| Bacteria (human pathogens) | *S. aureus* | 7 ± 0.5 | 6 ± 1 | 7 ± 1.5 | 16 ± 0.5 | 12 ± 1 | - | 23 ± 0.5 | - |
| | *E. coli* | 12 ± 1.1 | 11 ± 1 | 10 ± 1 | 11 ± 1 | 8 ± 1.5 | - | 15 ± 1 | - |
| | *P. aeruginosa* | 9 ± 1 | 12 ± 1.5 | - | 10 ± 1.5 | 10 ± 1.1 | - | 20 ± 1.1 | - |
| | *K. pneumoniae* | 17 ± 1 | 16 ± 1.5 | 12 ± 0.5 | 11 ± 1.5 | 13 ± 0.5 | - | 30 ± 1 | - |
| | *P. vulgaris* | 13 ± 1 | 12 ± 1.5 | 13 ± 0.5 | 12 ± 1 | 8 ± 1 | - | 22 ± 0.5 | - |
| Bacteria (fish pathogens) | *S. aureus* | 21 ± 1.1 | 14 ± 1 | 16 ± 1 | 15 ± 1.5 | 12 ± 0.5 | 13 ± 1 | 28 ± 0.5 | - |
| | *V. harveyi* | 12 ± 0.5 | 15 ± 1 | 10 ± 0.5 | 11 ± 1.1 | - | - | 27 ± 0.5 | - |
| | *A. hydrophila* | 10 ± 2 | 7 ± 1.1 | 10 ± 1.1 | 13 ± 1 | 11 ± 1.5 | - | 26 ± 1 | - |
| | *P. mirabilis* | 13 ± 1.5 | 17 ± 1 | 10 ± 0.5 | 9 ± 1.5 | 11 ± 1 | - | 30 ± 1.1 | - |
| | *P. aeruginosa* | 17 ± 1 | 14 ± 1.5 | 12 ± 0.5 | 11 ± 1 | 15 ± 1 | - | 21 ± 1 | - |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| **Fungi (human pathogens)** | ***A. niger*** | 17 ± 0.5 | 14 ± 0.5 | 13 ± 0.5 | 12 ± 0 | 10 ± 0.5 | 11± 0.5 | 25 ± 1 | - |
| | ***A. flavus*** | 15 ± 1 | 10 ± 1 | 12 ± 0.5 | 14 ± 1 | 10 ± 0.5 | 11 ± 1.1 | 21 ± 1 | - |

*CF- I = coelomic fluid butanol treated, CF- II = crude coelomic fluid without solvent treatment*

## Table 2. Antimicrobial activity of ethyl acetate extract of starfish, *P. linckii*

| **Microbes** | | **Zone of inhibition (mm) (Mean ± SD)** | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | **Digestive glands** | **Gonad** | **Tube feet** | **Arm** | **CF- I** | **CF- II** | **Positive** | **Negative** |
| **Bacteria (human pathogens)** | ***S. aureus*** | 19 ± 1 | 11 ± 1 | 10 ± 1 | 10 ± 1 | - | - | 21 ± 1.7 | - |
| | ***E. coli*** | - | - | - | 14 ± 0.5 | - | - | 20 ± 1.5 | - |
| | ***P. aeruginosa*** | 22 ± 1 | 13 ± 1.5 | 11 ± 1.5 | - | - | - | 20 ± 1.5 | - |
| | ***K. pneumoniae*** | 21 ± 1.1 | 11 ± 1.5 | 9 ± 1.1 | - | - | - | 29 ± 0.5 | - |
| | ***P. vulgaris*** | 16 ± 1 | 13 ± 1 | 10 ± 1.1 | 9 ± 0.5 | - | - | 20 ± 1.1 | - |
| **Bacteria (fish pathogens)** | ***S. aureus*** | 16 ± 0.5 | 15 ± 0.5 | 10 ± 0 | 8 ± 0 | - | 11 ± 0.5 | 20 ± 1.1 | - |
| | ***V. harveyi*** | 15 ± 1 | 15 ± 1.5 | - | 12 ± 0.5 | - | - | 20 ± 0 | - |
| | ***A. hydrophila*** | 15 ± 1 | 9 ± 0.5 | - | - | - | - | 24 ± 1.1 | - |
| | ***P. mirabilis*** | 12 ± 1 | 11 ± 1 | 9 ± 1.1 | 11 ± 1.1 | - | - | 23 ± 1 | - |
| | ***P. aeruginosa*** | - | - | - | - | 14 ± 1.1 | - | 18 ± 1 | - |
| **Fungi (human pathogen)** | ***A. niger*** | 12 ± 1 | - | - | - | - | 11 ± 0.5 | 22 ± 1 | - |
| | ***A. flavus*** | 10 ± 1 | - | - | - | - | 14 ± 0.5 | 22 ± 0.5 | - |

CF- I = coelomic fluid ethyl acetate treated, CF- II = crude coelomic fluid without solvent treatment

## Table 3. Antimicrobial activity of chloroform extract of starfish, *P. linckii*

| **Microbes** | | **Zone of inhibition (mm) (Mean ± SD)** | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | **Digestive glands** | **Gonad** | **Tube feet** | **Arm** | **CF- I** | **CF- II** | **Positive** | **Negative** |
| **Bacteria (human** | ***S. aureus*** | 14 ± 1 | 11 ± 1.5 | 13 ± 1 | - | - | - | 21 ± 0.5 | - |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| **pathogens)** | ***E. coli*** | | 12 ± 0.5 | 11 ± 1 | - | - | - | 16 ± 1 | - |
| | ***P. aeruginosa*** | 8 ± 0.5 | 14 ± 1.5 | - | - | - | - | 19 ± 1 | - |
| | ***K. pneumoniae*** | - | - | - | - | - | - | 21 ± 1 | - |
| | ***P. vulgaris*** | - | 10 ± 0.5 | 9 ± 0.5 | - | - | - | 21±1 | - |
| **Bacteria (fish pathogens)** | ***S. aureus*** | - | - | - | - | - | 11 ± 1 | 20 ± 0.5 | - |
| | ***V. harveyi*** | - | 10 ± 1.1 | 11 ± 1 | 12 ± 1 | - | - | 17 ± 1.5 | - |
| | ***A. hydrophila*** | - | 7 ± 1.1 | 12 ± 0.5 | - | - | - | 22 ± 0.5 | - |
| | ***P. mirabilis*** | - | 12 ± 2 | - | 11 ± 0.5 | - | - | 15 ± 1 | - |
| | ***P. aeruginosa*** | 10 ± 0.5 | 11 ± 1.5 | - | - | - | - | 17 ± 1.1 | - |
| **Fungi (human pathogens)** | ***A. niger*** | - | - | - | - | - | 11 ± 0.5 | 21 ± 1.1 | - |
| | ***A. flavus*** | 15 ± 1 | - | - | - | - | 12 ± 1 | 21± 0.5 | - |

CF- I = coelomic fluid chloroform treated, CF- II = crude coelomic fluid without solvent treatment

## Table 4. Antimicrobial activity of acetone extract of starfish, *P. linckii*

| **Microbes** | | **Zone of inhibition (mm) (Mean ± SD)** | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | **Digestive glands** | **Gonad** | **Tube feet** | **Arm** | **CF- I** | **CF-II** | **Positive** | **Negative** |
| **Bacteria (human pathogens)** | ***S. aureus*** | 12 ± 1 | - | 10 ± 0.5 | - | - | - | 21 ± 1.1 | - |
| | ***E. coli*** | - | - | - | 11 ± 1.5 | - | - | 14 ± 0.5 | - |
| | ***P. aeruginosa*** | - | 11 ± 1.5 | - | 9 ± 1 | - | - | 20 ± 1.5 | - |
| | ***K. pneumoniae*** | 7 ± 1.5 | - | 10 ± 1.5 | - | - | - | 24 ± 1 | - |
| | ***P. vulgaris*** | 9 ± 0.5 | - | 11 ± 0.5 | - | - | - | 20 ± 1.1 | - |
| **Bacteria (fish pathogens)** | ***S. aureus*** | 13 ± 1 | - | - | - | - | 10 ± 0 | 22 ± 1 | - |
| | ***V. harveyi*** | - | - | - | - | - | - | 12 ± 0.5 | - |
| | ***A. hydrophila*** | 9 ± 1.5 | - | - | - | - | - | 24 ± 1 | - |
| | ***P. mirabilis*** | 11 ± 0.5 | 13 ± 1.5 | - | - | - | - | 20 ± 0.5 | - |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | ***P. aeruginosa*** | - | - | - | - | - | - | 17 ± 0.5 | - |
| **Fungi (human pathogens)** | ***A. niger*** | 6 ± 0.5 | 7 ± 0.5 | 13 ± 1 | - | - | 12 ± 1 | 22 ± 1 | - |
| | ***A. flavus*** | 8 ± 1 | - | 11 ± 1 | 18 ± 0.5 | - | 15 ± 1 | 22 ± 0.5 | - |

CF- I = coelomic fluid acetone treated, CF- II = crude coelomic fluid without solvent treatment

**Table 5. Antimicrobial activity of methanolic extract of starfish, P. linckii**

| **Microbes** | | **Zone of inhibition (mm) (Mean ± SD)** | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | **Digestive glands** | **Gonad** | **Tube feet** | **Arm** | **CF-I** | **CF-II** | **Positive** | **Negative** |
| **Bacteria (human pathogens)** | ***S. aureus*** | - | - | - | - | - | - | 22 ± 0.5 | - |
| | ***E. coli*** | - | - | 8 ± 0.5 | - | - | - | 23 ± 1 | - |
| | ***P. aeruginosa*** | - | - | - | - | - | - | 19 ± 1 | - |
| | ***K. pneumoniae*** | - | - | - | - | - | - | 24 ± 1.5 | - |
| | ***P. vulgaris*** | - | - | - | - | - | - | 17 ± 1.1 | - |
| **Bacteria (fish pathogens)** | ***S. aureus*** | 9 ± 0.5 | - | - | - | - | 10 ± 0.5 | 20 ± 0.5 | - |
| | ***V. harveyi*** | - | - | - | - | - | - | 21± 1.5 | - |
| | ***A. hydrophila*** | - | 17 ± 1 | - | - | - | - | 24 ± 0.5 | - |
| | ***P. mirabilis*** | - | - | - | - | - | - | 21 ± 1 | - |
| | ***P. aeruginosa*** | - | - | - | - | - | - | 21 ± 1 | - |
| **Fungi (human pathogens)** | ***A. niger*** | 7 ± 0 | 16 ± 0.5 | - | - | - | 13 ± 1 | 20 ± 1.1 | - |
| | ***A. flavus*** | 7 ± 0.5 | - | - | 8 ± 0.5 | - | 12 ± 1.1 | 16 ± 1.5 | - |

CF- I = coelomic fluid methanol treated, CF- II = crude coelomic fluid without solvent treatment

## DISCUSSION

From our results it was interesting to note that butanol and ethyl acetate extract were highly efficient in inhibiting the growth of the pathogens when compared to other solvents. Chloroform, acetone and methanol extracts of digestive gland, gonad, tube feet and arm were also effective against selective pathogens. Thus our studies clearly indicate that the solvents are efficient in extracting the bioactive compound from the samples. Layson *et al* [30] studied the effects of aqueous, methanol, chloroform and hexane extracts of seven echinoderm animals and almost all sample extracts showed

antibacterial activity. Sri kumaran *et al* [31] had reported the antibacterial activity of whole body extract of *P. linckii* against human bacterial and fungal pathogens and fish bacterial pathogen and observed that microbes were sensitive to methanolic and n-butanol extracts of *P. linckii*. Similarly fungi *A. niger* was found to be sensitive to methanol and water extracts of *Asterina pectinifera*[32]. Thus it is clearly evident that the type of solvent plays a key role in antimicrobial activity. Among the tissue extracts tested, the body tissues like digestive gland, gonad, tube feet and arm were more effective against the pathogenic bacteria and fungi. Haug *et al* [21] reported antibacterial activity of three echinoderm species *Asterias rubens, Strongylocentrotus droebachiensis* and *Cucumaria frondosa* and highest activity was found in the body wall, eggs and intestinal organs of *A. rubens*, shells and coelomocytes of *S. droebachiensis* and eggs of *C. frondosa*. Body wall extracts of echinoderms displayed antimicrobial activity against marine bacteria[33]. The antimicrobial activity may be due to the antimicrobial compounds, including steroidal glycosides, polyhydroxylated sterols, lysozymes, complement- like substances or antimicrobial peptides which have been reported from echinoderms [18, 34, 35, and 36]. The possible antimicrobial effect with tissues may also be due to the antimicrobial peptides (AMPs), small molecular weight proteins of the innate immune response, with a broad spectrum of antimicrobial activities against bacteria, viruses and fungi [37]. AMPs are likely to be attracted and attach to the negatively charged bacterial surfaces because of their cationic nature. However, once the peptides come into contact with the outer leaflet of the cell membrane and the peptide/ lipid ratio increases, the peptides start forming multimers or self-associating on top of the membrane[38] allowing them to permeate by pore formation.

Antifungal assay demonstrated a maximum zone of inhibition against *A. niger* and *A. flavus*. *Aspergillus* species are ubiquitous fungi which can cause a variety of clinical syndrome, especially in immunosuppressed patients and as agents of foetal systemic infections and have therefore gained considerable public health importance[39]. Analysis of cell wall composition of *A. niger* showed the presence of six sugars, glucose, galactose, mannose, arabinose, glucosamine and galactosamine[40]. According to Pinto *et al.*[41] the cell wall of *Aspergillus* species is composed of a number of unique interconnected polysaccharides, including chitin and a variety of glucans. Results of Leal *et al.* [42] showed there was expression of N-acetyl-D-glucosamine, L-fucose and D-galactose on the cell wall surface of *A. flavus* and *A. niger*. Thus from our studies it could be implicated that the bioactive compounds in the tissue extracts may possess the ability to recognize the saccharide moieties present on the cell wall and may cause fungal cell wall polymer degradation or damage to cellular ribosomes or may inhibit the cell cycle[43]. Thus our results indicate the starfish, *P. linckii* as an effective antibacterial and antifungal agent.

## CONCLUSION

The pathogens are highly sensitive to the butanol and ethyl acetate extract of tissue of starfish, *P. linckii*. Body tissues like digestive gland, gonad, tube feet and arm could be used as an antimicrobial agent.

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