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# Effect of *Mastacembelus armatus* Epidermal Mucus Extracts on Selected Urinary Tract Infection Pathogens

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**ABSTRACT: The developing bacterial resistance counter agents of antimicrobials have led to the research for alternatives to traditional antibiotics. The fish mucus comprises different antimicrobial representatives that serve as a first defence line to counter numerous pathogens of invading. The modern work was considered for evaluating the outcome of *Mastacembelus armatus* epidermal mucus on selected urinary tract infection (UTI) causing bacteria (*Staphylococcus epidermidis, staphylococcus hominis, and proteus mirabilis, Enterobacter cloacae, Escherichia coli, Enterococcus avium, Pseudomonas aeruginosa*) and fungi (*Candida albicans*). The selected UTI pathogens were treated with acetic acid, Aqueous and organic epidermal mucus subtracts gathered in the container of *Mastacembelus armatus* at 12.5, 25 and 50 along with 100µg/mL. The acidic mucus extract showed novel antibiotic activity against the selected UTI pathogens, followed by aqueous and organic mucus subtracts. Moreover, the acidic epidermal mucus extract of *Mastacembelus armatus,* at 100 µg/mL, displayed the highest zone of inhibition for *Escherichia coli,* followed by *Candida albicans, Pseudomonas aeruginosa*, *Staphylococcus epidermidis*, *Proteus mirabilis, Staphylococcus hominis, Enterobacter cloacae* and *Enterococcus avium*, respectively. Moreover, the values of MIC extended between 25 and 50 µg/mL, as well as the amounts of MBC scaled from 100 µg/mL and 50 µg/mL. For acidic epidermal mucus extract against selected UTI pathogens. The current findings presented that the epidermal mucus of acidic "*Mastacembelus armatus* processes" novel antimicrobial actions counter pathogens of UTI.**

**Keywords:** Fish mucus, Activity of Antimicrobial, Tract of urinary infection, *Mastacembelus armatus.*

## INTRODUCTION

Tract infection of the urinary is the place of pathogenic bacteria seize the bladder and urethra, obstruct urine move through reasoning inflammation, cloudy urine and intolerable pain as well as haematuria and nocturia (Mishra *et al*., 2017). It is prevalent mainly in babies, women as well as older people. At least 1 in 20 male and 1 in 2 females may collect a UTI in the middle of their lifetime (Flores-Mireles *et al*., 2015). In addition, the agents of causative due to UTIs are "*gram positive*" and "*gram negative*" bacteria as well as "certain fungi". In addition, it, the most naturally come out on top agent of infection in favour of UTIs is "uropathogenic Escherichia coli" and it maintained through "Klebsiella pneumoniae" "staphylococcus hominis", and "proteus mirabilis" and "Enterobacter cloacae", and "Escherichia coli", *Enterococcus avium, Pseudomonas aeruginosa and Candida spp* (Maithili *et al*., 2019). Antibiotics like "sulfamethoxazole" and "trimethoprim", "ciprofloxacin" are between the naturally suggested therapeutics considering UTIs. Yet, the antibiotic rise resistance as well as top rates of recurrence about natural infections significantly affect our society (Mishra *et al*., 2017). Such situations of critical strained antibiotics find power from synthetic resources, and natural elements are an interesting area to expand in developing new antibiotics because of their negligible cost effectiveness and toxicity on synthetic drugs. Between animal dependent elements, and skin mucus, a fish by element has been embraced as a major source in the reason for elements of antimicrobial (Kuppulakshmi *et al*., 2008).

The mucosal fish surface, for the immune first line defence, in addition it, this role as a barrier of immunological which serves a resistance about powerful counter infections and pathogens (Go *et al*., 2019; Salinas *et al*., 2021). The mucus produced with cells of goblet in the middle of epithelium of mucus contains kinds of active compound of biologically, as an example, "*lectins*", "*Lysozyme*" and "*photolytic enzymes*" and "*c reactive protein*" (Villarroel *et al*., 2007). Different molecules of antimicrobials have been discovered in the middle of external enzymes, pore forming glycoproteins, fish mucus, and "antibacterial peptides" and so on (Brinchmann, 2016).

The increasing bacterial resistance against antimicrobial agents has induced the search for alternative antimicrobial resources. Thus, antimicrobial peptides in

the fish mucus have the potential to develop new therapeutic agents. *Mastacembelus armatus* (*M. armatus*) is one of the biggest economically and spiny eel major food fish within countries related to Asia (Anu Prasanna *et al*., 2011). This is an important fish whose epithelial surface secretes a huge amount about mucus (it stands between 0.5% and 1.0% about body weight) constrained by other telecosts. The protection of mucus in the skin gathered from suspended particles and pathogens, in addition to that other mucin substance, has the efficient consideration of antimicrobial and noxious elements (Knouft *et al*., 2003). Thus, the present work was designed to evaluate the antimicrobial activity of different epidermal mucus extracts (acetic acid, organic and aqueous extracts) of freshwater fish *Mastacembelus armatus* against selected UTI bacteria (*Staphylococcus epidermidis, "Staphylococcus hominis", "Proteus mirabilis", "Enterococcus avium", "Enterobacter cloacae", "Escherichia coli", "Pseudomonas aeruginosa")* and fungi (*Candida albicans).*

## MATERIALS AND METHODS

*A. Sample collection*

The faithful live freshwater *Mastacembelus armatus*, old stands around 6 months, and weight of 500 gms of single, were gathered from Thamirabarani river in Tharuvi village, Tamilnadu and Tirunelveli dt was completed by certain modifications maintaining the method (Subramanian *et al.,* 2008). Previous mucus collection, fish were contained in the middle of water with in a sample tray for 1 hour time duration. In addition, one hour later, mucus discharge upon the surface of the epidermal fish body was received as samples. One spatula of sterile fish was implemented to scrape mucus from the body surface of the dorsal side. Moreover, the mucus has not been grabbed on the side of the ventral to reduce intestinal as well as sperm contamination. The grasped fish mucus was restored at the temperature of 4 °C for the reason of using it next time.

*B. Preparation of mucus extracts*

The mucus specimen was spitted into three divisions subtracted individually by aqueous, organic solvents and acetic acid. The aqueous extract was prepared by mucus sample was mixed thoroughly with an equal amount of sterilised Saline is physiological along with centrifuges at 5000 rpm due to time duration of 15 m at the temperature of 25 degree centigrade. The supernatant was implemented as contact of aqueous (Tyor *et al*., 2016; Kumari *et al*., 2019). To prepare acidic mucus extract, 5 ml mucus was combined by 5 ml about 3% acetic acid, and the combine form was contained in the middle of boiling heat water, and ice cooled, and centrifuged, homogenized at 8000 rpm and the taking duration it 35 min and the temperature is 40 °C. The supplement filtered and collected utilising a 0.22 µm filter of syringe. In addition, the filter was implemented as acidic mucus subtracts (Kumari *et al*., 2011). For the preparation of organic extract, the mucus sample was Melted within 95% ethanol, as well as the pellet was subtracted after centrifugation about solution, that was vaporised below a vacuum under 40 °C. The substracts was melted in the middle of water which is distilled along with partitioned by dichloromethane. As well as, the DCM situation was gathered, suspended and dried in the middle of percentage around 5% "diethyl sulfoxide", which was used as organic mucus extract (Hellio *et al*., 2002). The different solvent mucus extracts were analysed for antimicrobial activity and store at 4°C for extra analysis.

**Bacterial strains.** The selected UTI bacteria such as, "*Staphylococcus epidermidis"* [MG911002], *"Enterobacter cloacae"* [MG913261], *"Enterococcus avium"* [MG913259], *"Escherichia coli"* [MG913260], *Pseudomonas aeruginosa* [MG907041], *"Staphylococcus hominis"* [MG907042], and fungi *Candida albicans* [MG91325) were gifted from Department of Microbiology, Dr. S.S. Maithili, Assistant Professor, AVS College in Arts and Science, Tamil Nadu, Salem.

*C. Preparation about the culture of microorganism*

All the microorganism of test were protected upon nutrient plates of agar and blood agar, and the strains of bacteria were then laid for the time of 24 hours, as well as the strains of fungal were laid for 48 hours at the temperature of 37 degree centigrade in the position of inverted, the growth gathered was then fixed in the middle of refrigerator at the temperature of 4 degree c till implied.

*D. Screening of Antimicrobial*

Initially screening in the reason of antibacterial and antifungal action of all three mucus of epidermal extracts (organic, acetic acid and aqueous) prepared from *Mastacembelus armatus* was carried out against selected UTI pathogens using the British Society for Antimicrobial Chemotherapy (BSAC) standardised disc susceptibility testing method (Andrews, 2005). All the Petri plates were filled with pouring MHA in the reason of bacteria, MHA supplement by 4% sodium chloride on behalf of MRSA along with SDA used for filamentous fungi. After that, the plates were endorsed to solidify along with used in the test of susceptibility. A 100 µL of bacterial and/or fungal suspension containing 108 CFU/mL of bacteria or 104 spore/mL of fungi was swab upon peak of the solidify onto the media of respective and was sanctioned to dry designed for 10 minutes.

Briefly, different conscious (12.5, 25, 50 along with 100 µg/mL) of mucus extract, such as organic, acetic acid and aqueous, were impregnated onto a 6 mm disc, separately. The discs by mucus subtracts were located on top of the intermediate using sterilized forceps as well as gently pushed to make sure contact through the inoculate agar outside. Ciprofloxacin (10 µg/disc) in favor of bacteria and Ketoconazole (50 µg/disc) meant for fungi were used as positive controls. Finally, the inoculated plates were incubated for 24 h (37°C) for all bacterial strains and 30°C for 72-96 h for selected fungi strains. The consequences were record by calculating the zones of expansion inhibition adjacent the disc. In addition, Clear shyness zones just about the discs indicate antimicrobial commotion. All figures on antimicrobial motion are the normal of triplicates analysis.

*E. MIC, MBC and MFC value of acidic epidermal mucus extract*

The epidermal acidic mucus subtract presented appropriate activity of antimicrobial for all selected strains. Therefore, the assay was carried out in acidic extract against the selected strains. The smallest amount inhibitory attention (MIC), as well as the lowest amount bactericidal attention (MBC) of acidic mucus take out, were perform from side to side the micro broth strength system using the "*96-well microtiter plate*" (Poly propylene plate) as explained by Sharma *et al.,* (2009). The dissimilar concentration of acidic mucus extract (6.25 to 200 µg/ml) was prepared and loaded in 96 well microtiter plate using carrier solvent, and then it was evaporated below sterile situation. After that, 100 µl about bacterial ($1\times10^6$ CFU/ml) along with fungal ($1\times10^6$ cell/ml) suspension was extra to each well. The microtiter plates with inoculum were incubated intended for 24 h at 37°C for bacterial strain as well as 96hrs on behalf of fungal strains. The assay was in three replicates. The MIC principles were engaged as the smallest possible concentration of acidic mucus extract in the health of the microtiter plate that presented no turbidity after incubation. As well as, the MBC and minimum fungal concentration (MFC) was single-minded through streaking every well containing a inoculums loopful in a nutrient agar plate. The least attentiveness of acidic take out showing no observable growth on subculturing was unavailable as MBC/MFC and value (Prakash *et al*., 2016).

*F. Statistical Analysis*

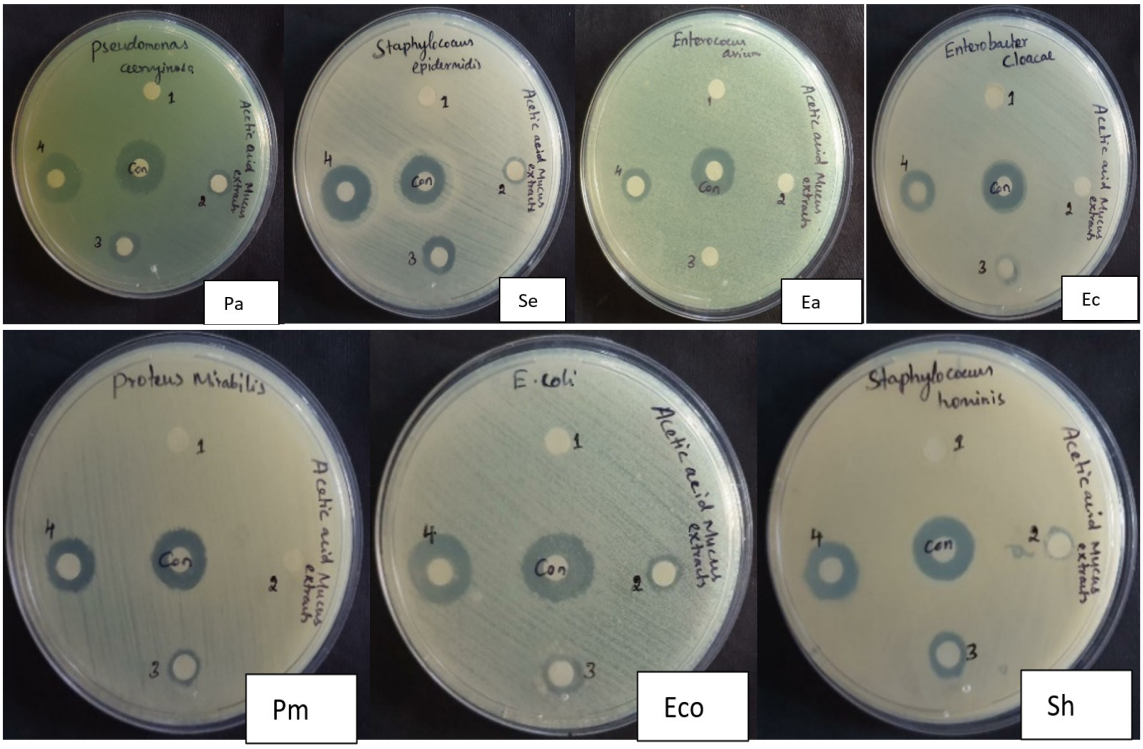
The results are expressed in mean ± SD. Then one-way analysis of variance (ANOVA) was carried out for the results to determine the significant differences ($P < 0.05$) between the means. The studies were carried out using SPSS software 16 (SPSS Inc., Chicago, IL).

## RESULTS

*A. Antimicrobial effect of different mucus extract*

The antibacterial and antifungal effects of different solvents epidermal mucus extract (aqueous, organic and acetic acid) were examined against selected UTI bacteria such as *Staphylococcus hominis Staphylococcus epidermidis*, *Proteus mirabilis, Enterobacter cloacae, Escherichia coli, Enterococcus avium*, *Pseudomonas aeruginosa* and fungi *Candida albicans* at different concentration like12.5, 25, 50 and 100 µg/mL (Fig. 1-3). Further, the antibacterial activity of different mucus extracts was compared with Ciprofloxacin (standard drug) at a 10 µg/ml concentration. The acidic mucus extract showed novel antibiotic activity against all selected UTI pathogens, followed by organic and aqueous mucus extract.

In the acidic epidermal mucus extract of *Mastacembelus armatus* (100 µg/mL), the highest zone of inhibition was observed against *Escherichia coli,* followed by *Pseudomonas aeruginosa*, *Staphylococcus epidermidis*, *Proteus mirabilis, Staphylococcus hominis, Enterobacter cloacae* and *Enterococcus avium*, respectively (Table 1).



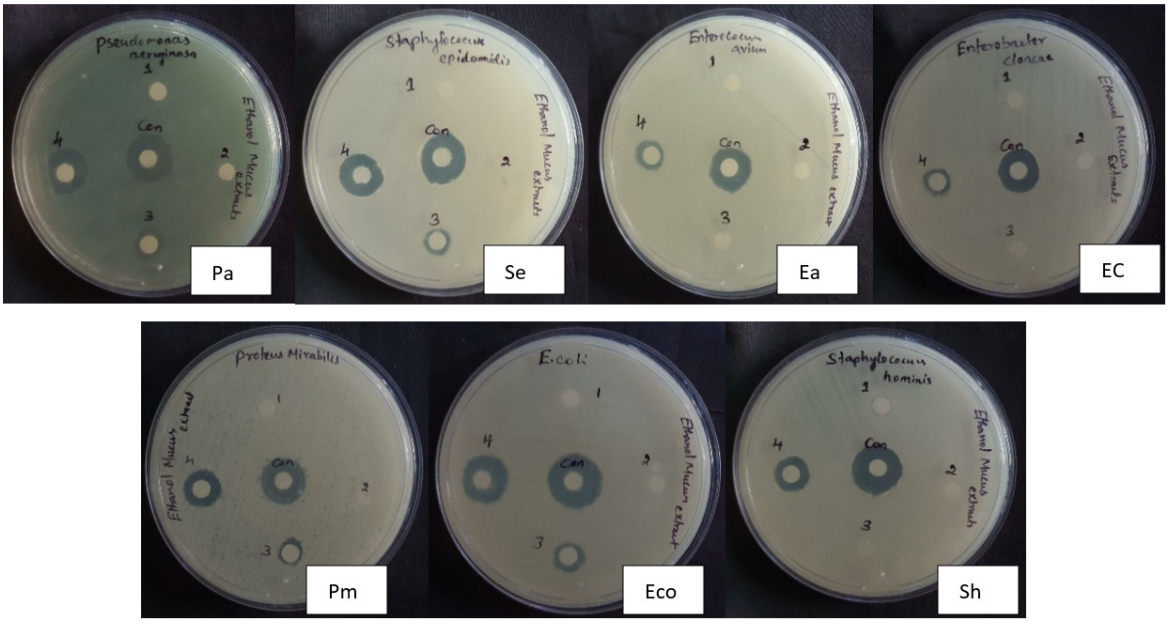

Pa: *Pseudomonas aeruginosa,* Se: *Staphylococcus epidermidis,* Ea: *Enterococcus avium,* Ec: *Enterobacter cloacae,* Pm: *Proteus mirabilis,* Eco: *Escherichia coli,* Sh: *Staphylococcus hominis;* 1: 12.5 µg/mL, 2: 25 µg/mL, 3: 50 µg/mL, 4: 100 µg/mL and Con: Ciprofloxacin 10 µg/mL.

**Fig. 1.** Antibacterial activity of acidic epidermal mucus extract of freshwater fish *Mastacembelus armatus* against selected UTI bacteria.

**Table 1: Antibacterial activity of acidic epidermal mucus extract of freshwater fish *Mastacembelus armatus* against selected UTI bacteria.**

| Bacteria tested | The concentration of extract and zone of inhibition (mm) | | | | |
|---|---|---|---|---|---|
| | Ciprofloxacin 10 µg/mL | 100 µg/mL | 50 µg/mL | 25 µg/mL | 12.5 µg/mL |
| *Staphylococcus hominis* | 17.6 ± 0.74a | 17.2a ± 0.32 | 10.2 ± 0.56b | 7.3 ± 0.12c | NZI |
| *Staphylococcus epidermidis* | 16.6 ± 0.36a | 16.7 ± 0.28a | 12.1 ± 0.28b | 8.6 ± 0.26c | NZI |
| *Proteus mirabilis* | 16.8 ± 0.56a | 13.4 ± 0.44b | 8.2 ± 0.16c | NZI | NZI |
| *Enterobacter cloacae* | 15.2 ± 0.42a | 10.5 ± 0.26b | 7.5 ± 0.08c | NZI | NZI |
| *Escherichia coli* | 18.2 ± 0.64a | 18.6 ± 0.46a | 13.5 ± 0.42b | 9.3 ± 0.24c | NZI |
| *Enterococcus avium* | 15.5 ± 0.24a | 9.5 ± 0.23b | 7.4 ± 0.12c | NZI | NZI |
| *Pseudomonas aeruginosa* | 17.3 ± 0.46a | 17.0 ± 0.38a | 12.9 ± 0.2b | 9.2 ± 0.28c | 7.2 ± 0.23d |

NZI: No zone of inhibition. All the values were presented as mean□±□SEM (*n*□=□3), and significantly different (P < 0.05) were determined between the standard drug and each concentration of mucus extract by one-way ANOVA



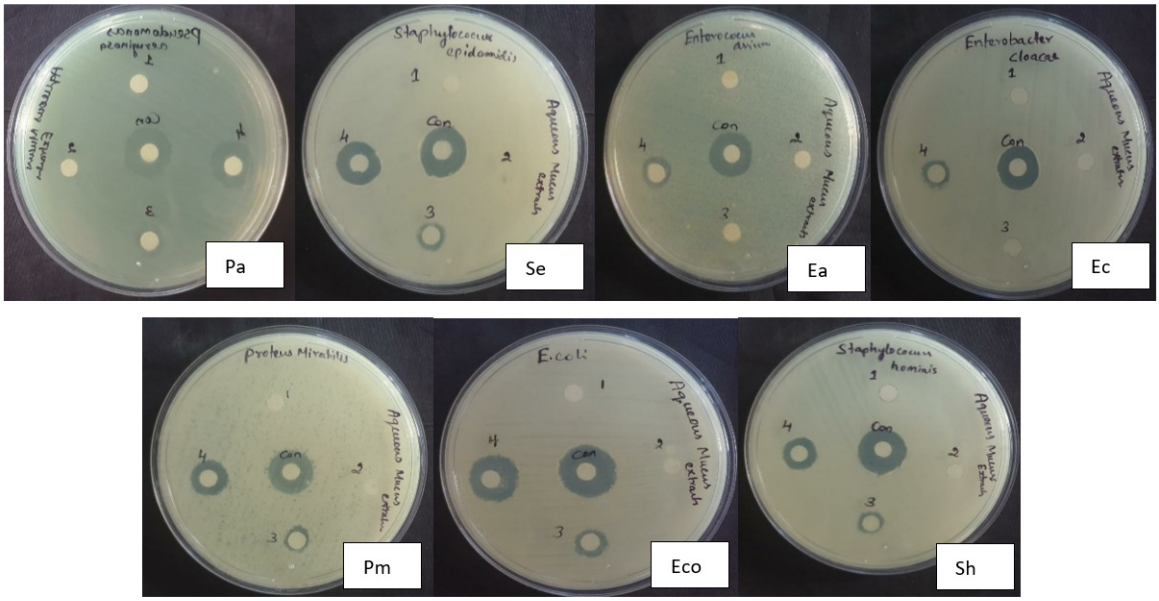

Pa: *Pseudomonas aeruginosa,* Se: *Staphylococcus epidermidis,* Ea: *Enterococcus avium,* Ec: *Enterobacter cloacae,* Pm: *Proteus mirabilis,* Eco: *Escherichia coli,* Sh: *Staphylococcus hominis:* 1: 12.5 µg/mL, 2: 25 µg/mL, 3: 50 µg/mL, 4: 100 µg/mL and Con: Ciprofloxacin 10 µg/mL.

**Fig. 2.** Antibacterial activity of organic epidermal mucus extract of freshwater fish *Mastacembelus armatus* against selected UTI bacteria.

**Table 2: Antibacterial activity of organic epidermal mucus extract of freshwater fish *Mastacembelus armatus* against selected UTI bacteria.**

| Bacteria tested | The concentration of extract and zone of inhibition (mm) | | | | |
|---|---|---|---|---|---|
| | Ciprofloxacin 10 µg/mL | 100 µg/mL | 50 µg/mL | 25 µg/mL | 12.5 µg/mL |
| *Staphylococcus hominis* | 17.4 ± 0.48a | 12.4 ± 0.28b | 7.2 ± 0.34c | NZI | NZI |
| *Staphylococcus epidermidis* | 16.4 ± 0.42a | 14.5± 0.16b | 7.6 ± 0.33c | NZI | NZI |
| *Proteus mirabilis* | 16.7 ± 0.52a | 13.4 ± 0.42b | 8.2 ± 0.15c | NZI | NZI |
| *Enterobacter cloacae* | 15.4 ± 0.36a | 8.5 ± 0.26b | NZI | NZI | NZI |
| *Escherichia coli* | 18.4 ± 0.63a | 15.1 ± 0.48b | 9.5 ± 0.12c | 7.1 ± 0.15d | NZI |
| *Enterococcus avium* | 15.2 ± 0.26a | 7.5 ± 0.15b | NZI | NZI | NZI |
| *Pseudomonas aeruginosa* | 17.4 ± 0.42a | 14.6 ± 0.25b | 8.9 ± 0.25c | 6.7 ± 0.28d | NZI |

NZI: No zone of inhibition. All the values were presented as mean□±□SEM (*n*□=□3), and significantly different (P < 0.05) were determined between the standard drug and each concentration of mucus extract by one-way ANOVA



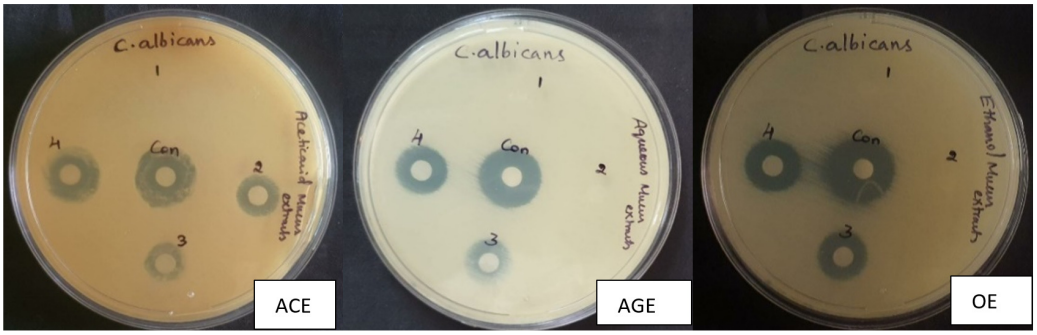

Pa: *Pseudomonas aeruginosa,* Se: *Staphylococcus epidermidis,* Ea: *Enterococcus avium,* Ec: *Enterobacter cloacae,* Pm: *Proteus mirabilis,* Eco: *Escherichia coli,* Sh: *Staphylococcus hominis;* 1: 12.5 µg/mL, 2: 25 µg/mL, 3: 50 µg/mL, 4: 100 µg/mL and Con: Ciprofloxacin 10 µg/mL.

**Fig. 3.** Antibacterial activity of aqueous epidermal mucus extract of freshwater fish *Mastacembelus armatus* against selected UTI bacteria.

**Table 3: Antibacterial activity of aqueous epidermal mucus extract of freshwater fish *Mastacembelus armatus* against selected UTI bacteria.**

| Bacteria tested | The concentration of extract and zone of inhibition (mm) | | | | |
|---|---|---|---|---|---|
| | Ciprofloxacin 10 µg/mL | 100 µg/mL | 50 µg/mL | 25 µg/mL | 12.5 µg/mL |
| *Staphylococcus hominis* | 17.5 ± 0.58a | 11.4 ± 0.45b | 6.5 ± 0.8c | NZI | NZI |
| *Staphylococcus epidermidis* | 16.7 ± 0.46a | 12.8 ± 0.26b | 7.9 ± 0.14c | NZI | NZI |
| *Proteus mirabilis* | 16.6 ± 0.38a | 11.9 ± 0.36b | 7.6 ± 0.25c | NZI | NZI |
| *Enterobacter cloacae* | 15.4 ± 0.32a | 8.2 ± 0.18b | NZI | NZI | NZI |
| *Escherichia coli* | 18.5 ± 0.64a | 13.7 ± 0.23b | 8.4 ± 0.12c | NZI | NZI |
| *Enterococcus avium* | 15.1 ± 0.28a | 7.3 ± 0.16b | NZI | NZI | NZI |
| *Pseudomonas aeruginosa* | 17.6 ± 0.36a | 13.4 ± 0.18b | 8.2 ± 0.24c | NZI | NZI |

NZI: No zone of inhibition. All the values were presented as mean□±□SEM (*n*□=□3), and significantly different ($P < 0.05$) were determined between the standard drug and each concentration of mucus extract by one-way ANOVA




ACE: Acidic extract, AQE: Aqueous extract, OE: Organic extract; 1: 12.5 µg/mL, 2: 25 µg/mL, 3: 50 µg/mL, 4: 100 µg/mL and Con: Ketoconozole (50 µg/mL)

**Fig. 4.** Antifungal activity of different epidermal mucus extract of freshwater fish *Mastacembelus armatus* against *Candida albicans.*

**Table 4: Antifungal activity of different epidermal mucus extract of freshwater fish *Mastacembelus armatus* against *Candida albicans*.**

| Fungus tested | Ketoconozole (50 µg/mL) | 100 µg/mL | 50 µg/mL | 25 µg/mL | 12.5 µg/mL |
|---|---|---|---|---|---|
| Acidic epidermal mucus extract | | | | | |
| *Candida albicans* | 20.2 ± 0.62[a] | 18.2 ± 0.86[a] | 12.5 ± 0.34[b] | 8.4 ± 0.24[c] | NZI |
| Organic epidermal mucus extract | | | | | |
| *Candida albicans* | 20.1 ± 0.93[a] | 12.6 ± 0.52[b] | 8.5 ± 0.18[c] | NZI | NZI |
| Aqueous epidermal mucus extract | | | | | |
| *Candida albicans* | 20.4 ± 0.86[a] | 12.9 ± 0.28[b] | 8.7 ± 0.24[c] | NZI | NZI |

NZI: No zone of inhibition. All the values were presented as mean ± SEM ($n$ = 3), and significantly different ($P < 0.05$) were determined between the standard drug and each concentration of mucus extract by one-way ANOVA

The organic epidermal mucus extract displayed the highest zone of inhibition against *Escherichia coli,* followed by *Pseudomonas aeruginosa*, *Staphylococcus epidermidis*, *Proteus mirabilis, Staphylococcus hominis, Enterobacter cloacae* and *Enterococcus avium*, respectively (Table 2). The aqueous epidermal mucus extract presented the maximum zone of inhibition against *Escherichia coli,* followed by *Pseudomonas aeruginosa*, *Staphylococcus epidermidis*, *Proteus mirabilis, Staphylococcus hominis, Enterobacter cloacae* and *Enterococcus avium*, individually (Table 3). In fungi, the prominent zone of inhibition was represented against *Candida albicans* at a concentration of 100µg/mL of acidic epidermal mucus extract (Table 4). The best antifungal effect was observed for acidic extract, followed by organic and aqueous epidermal mucus extract (Fig. 4). As the concentration of epidermal mucus extract increased, the zone of inhibition also significantly increased against the selected UTI pathogens. The good antibiotic effect of different epidermal mucus extracts was ordered: acetic acid, ethanol and aqueous. Therefore, further investigation was performed only on the acidic epidermal mucus extract of *Mastacembelus armatus.*

### MIC, MBC MFC values of acidic epidermal mucus extract

The MIC and MBC values of acidic epidermal mucus extract of *Mastacembelus armatus* was analysed at a concentration of 6.25, 12.5, 25, 50, 100 & 200 µg/mL) against UTI pathogens such as *Escherichia coli, Pseudomonas aeruginosa*, *Staphylococcus epidermidis*, *Proteus mirabilis, Staphylococcus hominis, Enterobacter cloacae and Enterococcus avium*and *Candida albicans* results were summarised in Table 5. The MIC values ranged from 25 to 50 µg/mL of acidic epidermal mucus extract against selected UTI pathogens. The MIC value was 25 µg/mL for *Escherichia coli, Staphylococcus epidermidis* and *Pseudomonas aeruginosa*and 50 µg/mL of acidic epidermal mucus extract for *Proteus mirabilis, Staphylococcus hominis, Enterobacter cloacae and Enterococcus avium.* The MBC value was between 50 µg/mL and 100 µg/mL for selected UTI bacterial strains. The MBC value was 50 µg/mL for *Escherichia coli, Staphylococcus epidermidis* and *Pseudomonas aeruginosa*and 100 µg/mL of acidic epidermal mucus extract for *Proteus mirabilis, Staphylococcus hominis, Enterobacter cloacae and Enterococcus avium*, respectively. The *Candida albicans* of MIC and MFC were 25 and 50 µg/mL of acidic epidermal mucus extract, respectively.

## DISCUSSION

Several potent antibiotics are available for treating UTIs, but increasing drug resistance among bacteria has made therapy of UTIs difficult. Bacteria have the genetic ability to transmit as well as acquire resistance to drugs (Maithili *et al*., 2019).

**Table 5: MIC and MBC values obtained for epidermal mucus acetic acid extract of freshwater fish *Mastacembelus armatus* against selected UTI pathogens**

| Microbes | Concentration (µg/mL) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 6.25 | 12.5 | 25 | 50 | 100 | 200 | MIC | MBC |
| *Staphylococcus hominis* | +++ | +++ | ++ | + | - | - | 50 (µg/mL) | 100 (µg/mL) |
| *Staphylococcus epidermidis* | +++ | ++ | + | - | - | - | 25 (µg/mL) | 50 (µg/mL) |
| *Proteus mirabilis* | +++ | +++ | ++ | + | - | - | 50 (µg/mL) | 100 (µg/mL) |
| *Enterobacter cloacae* | +++ | +++ | ++ | + | - | - | 50 (µg/mL) | 100 (µg/mL) |
| *Escherichia coli* | +++ | ++ | + | - | - | - | 25 (µg/mL) | 50 (µg/mL) |
| *Enterococcus avium* | +++ | +++ | ++ | + | - | - | 50 (µg/mL) | 100 (µg/mL) |
| *Pseudomonas aeruginosa* | +++ | ++ | + | - | - | - | 25 (µg/mL) | 50 (µg/mL) |
| *Candida albicans* | +++ | ++ | + | - | - | - | 25 (µg/mL) | 50 (µg/mL) |

Excellent growth - (+++), Moderate growth- ( ++ ), MIC value ( + ) , MBC value ( - )

The fish skin mucus performs its role by continuous production and sloughing off, preventing the pathogens' attacks, and containing several antimicrobial factors, such as proteins, lysozyme, immunoglobulin, and lectins (Dash *et al*., 2018). Thus, the present study was conducted to examine the antimicrobial activity of different solvents like acetic acid, organic and aqueous epidermal mucus extract of freshwater fish *Mastacembelus armatus* against selected UTI bacteria such as *Staphylococcus hominis, Staphylococcus epidermidis, Proteus mirabilis, Enterobacter cloacae, Escherichia coli, Enterococcus avium, Pseudomonas aeruginosa* and selected fungi *Candida albicans*.

In the disc diffusion method, the inhibition zones were observed around the wells. This indicated the antibacterial activities of bioactive extract whereas the absence of zones of inhibition around each well-signified resistance (Choyam *et al*., 2015). In the present investigation, we analysed the antimicrobial effect of acetic acid, organic and aqueous epidermal mucus extract of freshwater fish *Mastacembelus armatus* against selected UTI pathogens at various concentrations like 12.5, 25, 50 and 100 µg/mL. From the three extracts, the acidic epidermal mucus extract of *Mastacembelus armatus* at 100 µg/mL showed the highest zone of inhibition against *Escherichia coli,* followed by *Pseudomonas aeruginosa*, *Staphylococcus epidermidis*, *Proteus mirabilis, Staphylococcus hominis, Enterobacter cloacae* and *Enterococcus avium*, respectively. Likewise, a 100 µg/mL concentration of acidic epidermal mucus extract showed antifungal activity against *Candida albicans*. The finding indicates that many proteins/bioactive compounds are present in the fish mucus, which exerts strong resistance to invading pathogens. These bioactive compounds/proteins have shown antimicrobial activities against selected human pathogenic bacteria and fungi via copious mechanisms, e.g., interrupting microbial membranes, weakening cellular mechanisms, controlling biofilm formation, inhibiting bacterial capsule production, and reducing microbial toxin production. The present results were supported by Uthayakumar *et al.,* (2012). They stated that the epidermal mucus extract of *Mastacembelus armatus* displays antimicrobial activity against *Escherichia coli, Vibrio cholera, Staphylococcus aureus, Salmonella typhi*, *Klebsiella pneumonia* and *Candida albicans.*

The MBC is complementary to the MIC. But, the MIC test demonstrates the lowest level of the antimicrobial agent that significantly inhibits growth. Whereas, the MBC is the lowest level of antimicrobial agent that induces microbial death (Ashraf *et al*., 2018; Atef *et al*., 2019). In the present investigation, the MIC values ranged from 25 to 50 µg/mL, and the MBC value ranged from 50 µg/mL and 100 µg/mL of acidic epidermal mucus extract against selected UTI pathogens. The *Candida albicans* of MIC and MFC were 25 and 50 µg/mL of acidic epidermal mucus extract, respectively. The MBC values obtained for the acidic mucus extracts against the selected UTI pathogens are higher than MIC, indicating that the extracts are bacteriostatic at lower concentrations and bactericidal at higher concentrations. This effect could be attributed to the synergic impacts exerted by compounds which act on different bacterial targets. These results were further reinforced by Abareethan (2021), who reported that fish epidermal mucus from *Labeo rohita* possesses MIC and MBC values of 100µg/ml against *E. coli* and *V. cholera*, respectively.

## CONCLUSION AND FUTURE SCOPE

The present study presented the acidic mucus extracts of *Mastacembelus armatus* possess novel antimicrobial activity against selected UTI pathogens than organic and aqueous mucus extracts. This indicates the presence of antimicrobial bioactive compounds / peptides / proteins in the mucus of *Mastacembelus armatus.* Therefore, further investigation is required to identify and isolate the individual antimicrobial compounds / peptides / proteins from the mucus of *Mastacembelus armatus.*

**Competing interests:** No conflict of interest.

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