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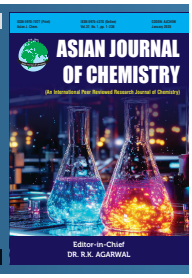

Exploring the Pharmaceutical Potential of *Meretrix casta* (Gmelin, 1791) (Mollusca: Bivalvia)

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Meretrix casta, a marine mollusk, has been recognized traditionally for its nutritional and medicinal properties. This study aims to investigate the pharmacological potential of *M. casta* extracts, specifically focusing on the antioxidant, hemolytic, anti-inflammatory and antimicrobial activities. The antioxidant activity, assessed *via* hydrogen peroxide scavenging and phosphomolybdate assays, revealed concentration-dependent inhibition, with the methanol extract showing 61.04% inhibition at 100 µg/mL compared to 61.19% for ethyl acetate, while the standard ascorbic acid exhibited 87.80%. Anti-inflammatory activity was evaluated using heat-induced hemolysis, hypotonicity-induced hemolysis and protein denaturation assays. Both extracts demonstrated significant anti-inflammatory effects, with the ethyl acetate extract achieving 85.40% inhibition of hemolysis, closely matching acetylsalicylic acid's 90.50% and methanol extract showing 87.60% at 100 µg/mL. Antibacterial and antifungal assays demonstrated significant inhibitory effects against pathogenic bacteria and fungi, with the methanolic extract frequently exhibiting higher efficacy. These findings highlight the therapeutic potential of *M. casta* extracts as natural bioactive agents. Future investigations should aim to isolate and characterize the specific bioactive compounds underlying these pharmacological effects and to explore their mechanisms of action in detail. Such studies could pave the way for new therapies derived from marine biodiversity, addressing various health challenges.

Keywords: *Meretrix casta*, Natural antioxidants, Medicinal marine mollusks, Therapeutic marine resources.

INTRODUCTION

Mollusks have long been integral to human diets and traditional medicine, particularly in coastal regions where they are abundant. Among them, *Meretrix casta*, commonly known as the clam, holds a prominent place due to its nutritional and medicinal properties [1]. The selection of *M. casta* for this research is based on its historical use in folk medicine and the increasing interest in marine organisms as sources of bioactive compounds [2]. This species, found primarily along the shores of Asia, has been consumed for centuries not only for its protein-rich meat but also for its therapeutic benefits, which are well-documented in traditional healing practices [3].

M. casta possesses significant nutritional value, offering a rich supply of essential nutrients like high-quality proteins, omega-3 fatty acids and vital vitamins and minerals, including zinc, iron and magnesium [4]. These components make the clam a valuable food item for maintaining overall health. Omega-3 fatty acids are known to promote cardiovascular health, reduce inflammation and support cognitive function. The presence of antioxidants in the clam also helps in combating oxidative stress, which is linked to various chronic diseases [5].

Clam meat is believed to have detoxifying properties and is consumed to support liver function in several coastal communities in India [6]. Additionally, the clam is thought to promote digestion and folk healers have recommended its consumption to alleviate digestive discomfort, improve nutrient absorption and maintain gastrointestinal health [7]. Furthermore, *M. casta* is considered an aphrodisiac in some cultures, where it is believed to enhance vitality, energy and sexual health [8]. This reputation may be linked to its high zinc content, a mineral critical for reproductive health and hormone balance. The clam has also been applied externally in the form of pastes or extracts to treat skin infections and wounds [9]. Its presumed antimicrobial and anti-inflammatory properties make it a common remedy for minor cuts, abrasions and skin irritations. Traditional practitioners have used clam-based treatments to accelerate wound healing, reduce inflammation and prevent infections [10]. Despite these traditional uses, there is still a significant gap in scientific understanding of the specific bioactive compounds responsible

for these effects. Hence, the present work was planned to evaluate the pharmaceutical properties of *M. casta* extracts, focusing on their antimicrobial, antioxidant and anti-inflammatory properties. Understanding these aspects could lead to the development of novel therapeutic agents derived from *M. casta*, contributing not only to modern medicine but also to validating its traditional uses.

EXPERIMENTAL

Study area: The yellow clam, *M. casta*, was collected from a site approximately 300 meters offshore from the coastline of Rameswaram, India. The coastal region of Rameswaram, located at approximately 9.2876º N latitude and 79.3129º E longitude, provides an essential environment for numerous marine species, including *M. casta*. These clams are typically found in the intertidal zones, favouring areas with sandy or muddy bottoms that are abundant along the shores of costal area of Rameswaram, India.

Collection and identification of *M. casta*: Live specimens of *M. casta* were collected by hand from the above coastal region. Identification was conducted by examining their morphological characteristics using a standard field guide and cross-referencing with the World Registry of Marine Species (WoRMS). After identification, the specimens were preserved in appropriate solvents and transported to the laboratory for further analysis. Upon reaching the lab, the shells were carefully removed with a sharp blade to avoid damaging the flesh. The flesh samples were then thoroughly cleaned with multiple tap water rinses to remove surface contaminants and debris, followed by a final rinse with distilled water to ensure all impurities were eliminated.

Preparation of crude extract: The whole flesh sample was homogenized in 100 mL of ethyl acetate and methanol separately for about 24 h using a homogenizer to ensure adequate extraction of the bioactive chemicals present in *M. casta*. After homogenization, the samples were centrifuged at 1000 rpm (OS20-S, D Lab, Romania) for 20 min at room temperature. The supernatant obtained from the ethyl acetate extract and methanol extract was then concentrated under reduced pressure using a rotary evaporator (RE 100-S, D Lab, Singapore). The concentrated extracts were stored separately at 4 ºC until further analysis was performed [11].

Hydrogen peroxide scavenging assay: The capacity of the *M. casta* extracts to neutralize H_2O_2 was evaluated using the hydrogen peroxide scavenging assay [12]. In this assay, 40 mM H_2O_2 solution was prepared in a phosphate buffer with a pH of 7.4. The various concentrations of *M. casta* extract were added to H_2O_2 solution and the reaction mixture was incubated at room temperature for 10 min. After incubation, the absorbance of the mixture was measured at 230 nm. The percentage of H_2O_2 neutralized by the extracts was determined by comparing the absorbance to that of a control reaction, in which the solvent was used as blank and the activity was calculated with the following formula:

$$H_2O_2 \text{ scavenging activity } (\%) = \frac{A_C - A_S}{A_C} \times 100$$

where A_C = absorbance of control, A_S = absorbance of sample.

Total antioxidant assay: The total antioxidant capacity of the *M. casta* extract was evaluated using the phosphomolybdate assay method [13]. This assay is based on the reduction of molybdenum(VI) to molybdenum(V) by antioxidants present in the sample, resulting in the formation of a green-coloured phosphate/molybdenum(V) complex under acidic conditions. The reagent for the assay was prepared by dissolving 4 mM ammonium molybdate in 0.6 M sulfuric acid and adding 28 mM sodium phosphate. The various concentrations of *M. casta* extracts were diluted in ethanol and mixed with the reagent. The mixture was then incubated at 95 ºC for 90 min. After incubation, the samples were cooled to room temperature and their absorbance was measured at 695 nm. The antioxidant capacity of the extracts was quantified by a calibration curve using ascorbic acid as a standard.

Hemolysis assay: The anti-hemolytic activity of *M. casta* extracts was assessed using a hemolysis assay on red blood cells (RBCs) [14]. Fresh, anticoagulated blood treated with EDTA was collected from the healthy volunteer and centrifuged at 10,000 rpm for 10 min at 4 ºC to separate the RBCs from plasma and buffy coat. The RBCs were washed several times with cold phosphate-buffered saline (PBS, pH 7.4) until a clear supernatant was obtained. A 2% v/v RBC suspension in PBS was prepared for the experiment. In assay, this RBC suspension was exposed to different concentrations of *M. casta* extracts. Controls were set up with untreated RBCs as a negative control and RBCs treated with a known hemolytic agent as a positive control. These mixtures were incubated at 37 ºC for 2 h. After incubation, the mixtures were centrifuged at 1,000 rpm for 10 min and the supernatant was carefully transferred for analysis. The hemolysis activity was determined by measuring the absorbance of the supernatant at 540 nm. The percentage of hemolysis was calculated using the following formula:

$$\text{Hemolysis activity } (\%) = \frac{A_C - A_S}{A_C} \times 100$$

where A_C = absorbance of control, A_S = absorbance of sample.

Anti-inflammatory assay: To evaluate the anti-inflammatory activities of *M. casta* extracts, three distinct assays were employed: heat-induced hemolysis, hypotonicity-induced hemolysis of RBCs and inhibition of egg albumin denaturation [15]. The heat-induced hemolysis assay by isolating red blood cells (RBCs) from anticoagulated blood and washing them with phosphate-buffered saline (PBS). The RBCs were then prepared into a 2% solution in PBS, to which different concentrations of *M. casta* extracts were added. The RBC-extract mixtures were exposed at 50 ºC for 30 min to induce hemolysis. After heating, the mixtures were cooled and centrifuged at 1000 rpm for 10 min and the supernatant was analyzed at 540 nm to measure haemoglobin release, indicating hemolysis. The percentage of heat induced hemolysis was calculated using the following formula:

$$\text{Heat-induced hemolysis } (\%) = \frac{A_C - A_S}{A_C} \times 100$$

where A_C = absorbance of control, A_S = absorbance of sample.

For the hypotonicity-induced hemolysis assay, the same initial steps were followed to prepare a 2% RBC suspension in PBS. This suspension was added with different concentrations of the extracts and a hypotonic saline solution (0.36% NaCl) was added. The mixture was incubated at room temperature for 30 min and then centrifuged at 1000 rpm for 10 min. The absorbance of the supernatant was measured at 540 nm to assess hemolysis. The percentage of hypotonicity induced hemolysis was calculated using the following formula:

$$\text{Hypotonicity-induced hemolysis (\%)} = \frac{A_C - A_S}{A_C} \times 100$$

where A_C = absorbance of control, A_S = absorbance of sample.

In the egg albumin denaturation assay, fresh egg albumin was mixed with different concentrations of the extracts diluted in PBS. This mixture was heated in a water bath at 70 ºC for 15 min to induce protein denaturation. Once heating was completed and the mixture was cooled at room temperature, the turbidity was measured at 660 nm. The percentage of egg albumin denaturation was calculated using the following formula:

$$\text{Egg albumin denaturation (\%)} = \frac{A_C - A_S}{A_C} \times 100$$

where A_C = absorbance of control, A_S = absorbance of sample.

Antimicrobial activity: The antimicrobial activity of *M. casta* extracts was systematically evaluated through both antibacterial and antifungal assays [16]. To assess the antibacterial properties, the disk diffusion method was used. In this method, sterile paper disks were soaked with the *M. casta* extracts and placed on Mueller-Hinton agar plates that had been pre-inoculated with various bacterial strains. These strains included methicillin-resistant *Staphylococcus aureus* (MRSA) (collected from Chettinadu Hospital), *Salmonella typhi* (collected from Chettinadu Hospital), *Escherichia coli* (MTCC443), *Staphylococcus aureus* (MTCC3160), *Enterococcus faecalis* (MTCC-439) and *Pseudomonas aeruginosa* (MTCC424). The plates were incubated at 37 ºC for 24 h. After incubation, the antibacterial efficacy of the extracts was determined by measuring the zones of inhibition, which are clear areas around the disks where bacterial growth was suppressed.

For the antifungal activity, the disk diffusion method was used. In this method, sterile paper disks were loaded with the *M. casta* extracts and placed on Potato Dextrose agar plates that had already been inoculated with fungi like *Aspergillus niger* (MTCC 4325), *Candida albicans* (KT831886) and *Candida tropicalis* (MTCC184). The plates were then incubated under conditions that were ideal for fungal growth. After incubation, the antifungal effectiveness was assessed by measuring the clear zones around the disks. To ensure the accuracy and reliability of the findings, both the antibacterial and antifungal assays were performed in triplicate.

RESULTS AND DISCUSSION

Hydrogen peroxide scavenging activity: The results indicate that both the ethyl acetate and methanol extracts of *M. casta* exhibit concentration-dependent inhibitory activity, with the standard ascorbic acid showing higher inhibition across all concentrations compared to the extracts (Fig. 1). At the lowest concentration of 20 µg/mL, the ethyl acetate extract showed an average inhibition of 34.04%, while the methanol extract showed 36.91% and the ascorbic acid showed 67.95%. At the highest concentration of 100 µg/mL, the ethyl acetate extract exhibited 61.19% inhibition, the methanol extract 61.05% and the ascorbic acid showed the highest inhibition at 87.80%. These findings suggest that inhibition increases with concentration for both extracts and the standard. The ascorbic acid standard consistently demonstrated the highest inhibition, indicating its strong antioxidant activity. The methanol extract generally showed slightly higher inhibition percentages compared to the ethyl acetate extract at most concentrations. These results suggest significant antioxidant properties of *M. casta* extracts, warranting further studies to explore the specific bioactive compounds responsible for this activity. When compared with other molluscan species, similar antioxidant properties have been observed. For instance, the antioxidant defense system in wedge clams showed variable enzyme activities, indicating an inherent mechanism to cope with environmental stressors [17]. Similarly, the antibacterial and hemolytic activities in different solvent extracts of marine bivalve freshwater mussel (*Lamellidens marginalis*) suggest the presence of bioactive compounds with potential pharmacology properties [18]. The antioxidant activity in molluscan species can be attributed to various factors. Primarily, the marine environment, characterized by exposure to various stressors like pollution and temperature changes, necessitates robust antioxidant systems in these organisms [19]. The antioxidant compounds may synergize with the organism's innate defense mechanisms to neutralize oxidative stress, thereby maintaining cellular homeostasis. Furthermore, the diet and habitat of these mollusks greatly influence their antioxidant profile. Species like *M. casta*, which thrive in diverse ecological settings, tend to accumulate a range of bioactive compounds, contributing to their antioxidant capacity [20]. This is evident in studies where Indian marine green mussel extracts showed potential anti-inflammatory properties, indicating the presence of bioactive compounds [21]. The comparative analysis highlights the significant antioxidant potential of *M. casta* extract, akin to other molluscan species. These findings open avenues for further exploration of marine mollusks as sources of natural antioxidants and their potential application in pharmacology and food industries.

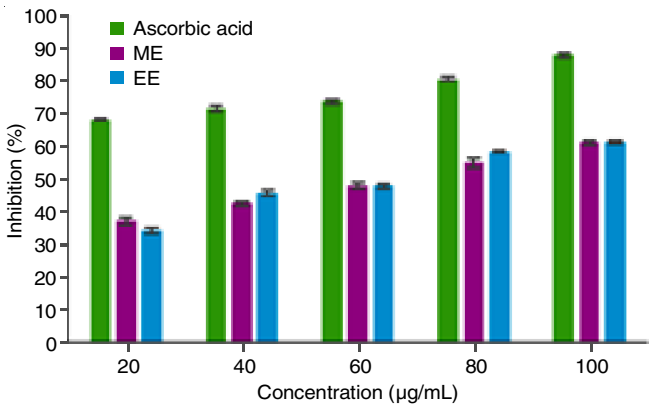

Fig. 1. Hydrogen peroxide scavenging activity of *M. casta* extract

Total antioxidant activity: The phosphomolybdate assay was used to evaluate the total antioxidant capacity of ethyl acetate and methanol extracts of *M. casta*, with ascorbic acid serving as a standard (Fig. 2). From the optical density (OD) of ascorbic acid, the equivalent antioxidant activity of the extracts was calculated. At the lowest concentration of 20 µg/mL, the ethyl acetate extract showed an average absorbance of 7.235, which corresponds to an antioxidant activity equivalent to 0.34 µg/mL of ascorbic acid. The methanol extract at the same concentration exhibited an average absorbance of 6.420, equivalent to 0.34 µg/mL of ascorbic acid. At the highest concentration of 100 µg/mL, the ethyl acetate extract demonstrated an average absorbance of 37.383, equivalent to 1.403 µg/mL of ascorbic acid, while the methanol extract showed an average absorbance of 43.432, also equivalent to 1.403 µg/mL of ascorbic acid. The data indicates that the methanol extract typically displayed greater antioxidant activity than the ethyl acetate extract across various concentrations. Nonetheless, both extracts demonstrated considerable antioxidant capabilities, suggesting the need for additional research to identify the specific bioactive compounds responsible for this effect. In comparison, studies on Asiatic hard clam (*Meretrix meretrix*), a close relative in the bivalve family, revealed antibacterial and antioxidant properties, underscoring the potential of bivalves as sources of bioactive compounds [22]. Such bioactive compounds in *M. casta* are consistent with the observed antioxidant activity. Evaluating other molluscan species like Mediterranean mussel (*Mytilus galloprovincialis*) highlighted the role of environmental factors and genetic variability in influencing antioxidant activities. This suggests that the observed antioxidant properties in *M. casta* might also be influenced by similar factors [23]. Marine sponges, another group within the marine ecosystem, have been recognized for their rich bioactive compounds. For instance, the sponge black pinnacle sponge (*Neopetrosia compacta*) exhibit significant anti-inflammatory activities, often associated with antioxidants. This comparison indicates a widespread occurrence of antioxidant activities across different marine organisms [24]. The variability in antioxidant activity within *M. casta* extracts and across different marine species can be attributed to the diversity of bioactive compounds, environmental adaptations and genetic factors. These findings reinforce the importance of marine biodiversity in the search for natural antioxidants.

Hemolysis activity: The hemolysis assay was used to evaluate the hemolytic activity of ethyl acetate and methanol extracts, with 1% SDS serving as a positive control (Fig. 3). At the lowest concentration of 20 mg/mL, the ethyl acetate extract showed an average inhibition of 0.3131%, while the methanol extract showed 0.2325%. At the highest concentration of 100 mg/mL, the ethyl acetate extract demonstrated an average inhibition of 1.5407%, while the methanol extract showed 1.1353%. The 1% SDS positive control exhibited consistently high inhibition, with values around 98.21%, confirming the assay's validity. These findings indicate that neither the ethyl acetate nor the methanol extracts of *M. casta* exhibit significant hemolytic activity across the tested concentrations. The low percentages of inhibition suggest that the compounds present in both extracts do not interact strongly with red blood cell membranes to cause hemolysis. This is a favourable outcome for their potential use in therapeutic applications, as it indicates a lower risk of cytotoxicity to red blood cells. The lack of hemolytic activity in these extracts highlights their potential safety as components in formulations intended for human use. These findings align with the cytotoxic potentials observed in other marine bivalves, such as mediterranean mussel (*Mytilus galloprovincialis*), suggesting the presence of bioactive compounds capable of disrupting cell membranes [25]. Comparative studies on bivalves like pacific oyster (*Crassostrea* spp). And windowpane oyster (*Placuna placenta*) indicates a complex balance between beneficial antioxidants and potential cytotoxic effects [26]. Furthermore, marine sponges, known for their rich bioactive compound repertoire, exhibit similar hemolytic activities linked to anti-inflammatory and antimicrobial actions, as seen in species like compact petrosiid sponge (*Neopetrosia compacta*) and sulphur sponge (*Suberites domuncula*) [27]. The hemolytic activity in *M. casta* extracts underscores the need to further explore the specific bioactive compounds responsible for this activity, which could have significant implications in biomedical applications

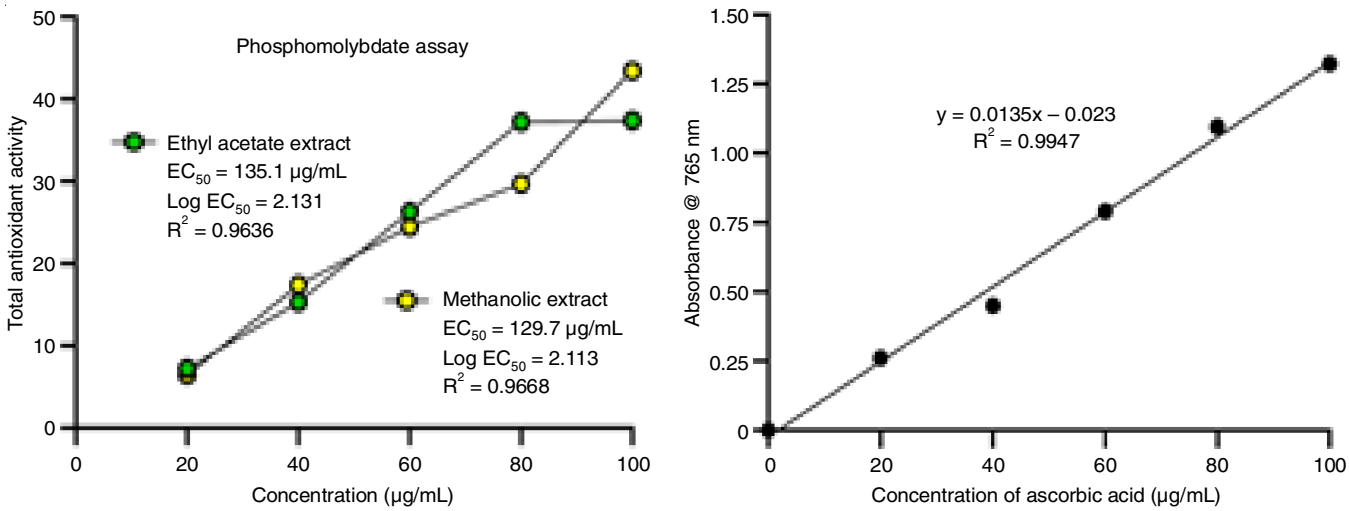

Fig. 2. Total antioxidant activity of *M. casta* extract

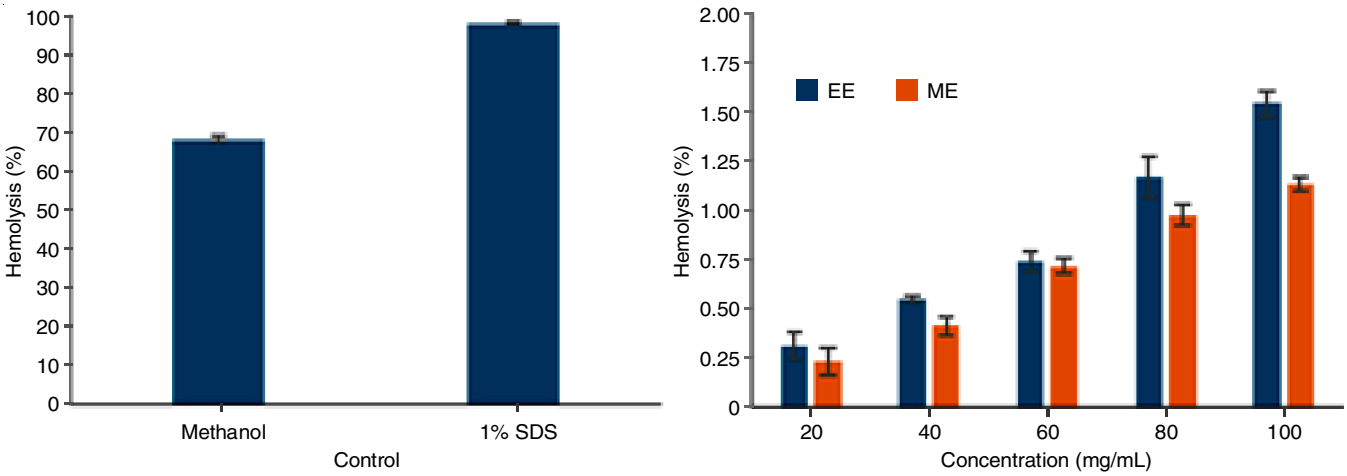

Fig. 3. Hemolytic activity of *M. casta* extract

such as developing novel anticancer drugs. Understanding the molecular mechanisms behind these compounds could lead to significant advancements in biomedicine.

Anti-inflammatory activity: The study investigated the anti-inflammatory properties of *M. casta* extract by evaluating its effects through three assays: heat-induced hemolysis, hypotonicity-induced hemolysis and protein denaturation, comparing the results to acetyl salicylic acid (ASA) as standard. In the heat-induced hemolysis assay, at a concentration of 100 µg/mL, the ethyl acetate extract (EE) exhibited significant protection against hemolysis with an inhibition percentage of 85.40 ± 2.35%, closely matching ASA's 90.50 ± 2.60%, while the methanolic extract (ME) achieved 87.60 ± 2.50% (Fig. 4). Similarly, in the hypotonicity-induced hemolysis assay, the EE demonstrated substantial inhibitory effects on hemolysis at 83.50 ± 2.30%, compared to ASA's 88.90 ± 2.45% and the ME showed 85.90 ± 2.40% (Fig. 5). In the protein denaturation assay, the EE displayed a robust capacity to inhibit protein denaturation, achieving 91.50 ± 2.30% inhibition at 100 µg/mL, comparable to ASA's 98.10 ± 2.45%, while the ME achieved 93.90 ± 2.35% (Fig. 6). These findings suggest that *M. casta* extract, particularly its ethyl acetate and methanolic extracts, possesses potent anti-inflammatory properties, with efficacy similar to that of the standard anti-inflammatory drug ASA, especially at higher concentrations. This indicates the potential of *M. casta* extract as a natural anti-inflammatory agent. Notably, species such as Asiatic hard clam (*M. meretrix*) have been recognized for their anti-inflammatory potential, indicative of the rich bioactive compound diversity in marine environments [28]. This diversity, also seen in organisms like green mussels (*Perna viridis* L.), highlights the therapeutic potential of marine derived compounds. The consistency of the anti-inflammatory activity in *M. casta* across different assays, despite the lack of significant variation in inhibitory activity across concentrations, suggests that the extract's bioactive components might exert their effects through multiple mechanisms [29]. This implies that even at lower concentrations, the compounds present in the extracts are sufficiently active to induce a response which does not significantly amplify with increasing concentration within the tested range. Marine fungi, such as *Emericellopsis*

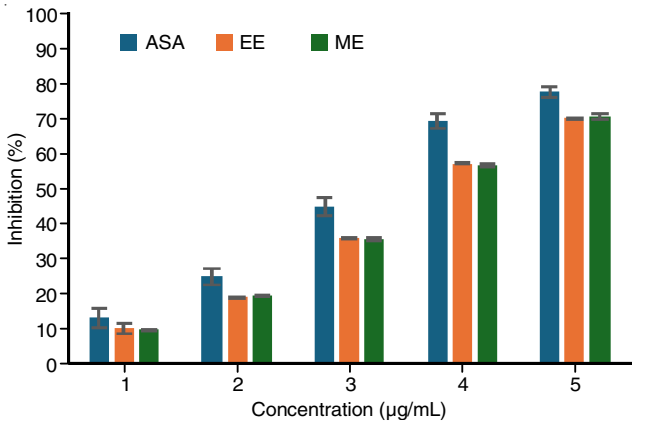

Fig. 4. Heat-induced hemolysis of RBC membrane activity of *M. casta* extract

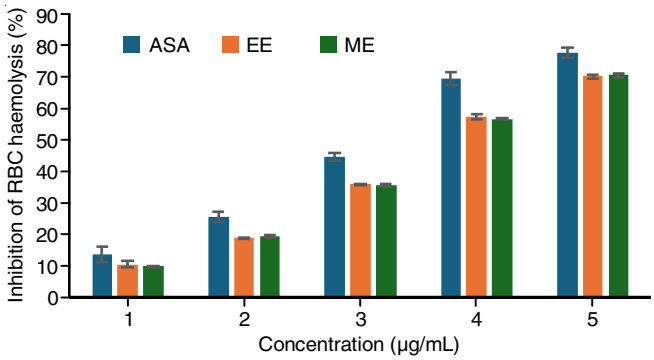

Fig. 5. Hypo tonicity induced hemolysis of RBC membrane activity of *M. casta* extract

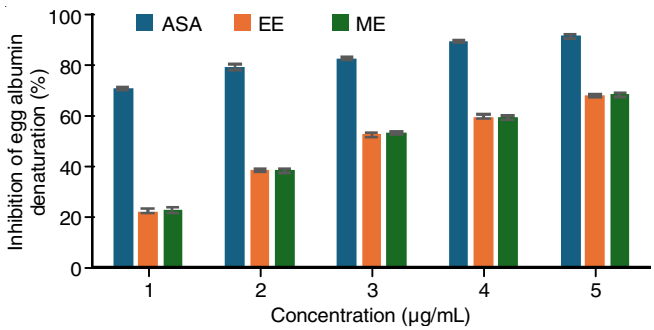

Fig. 6. Effect of *M. casta* extracts on egg albumin denaturation

cladophorae and *Zalerion maritima*, have also been explored for their anti-inflammatory lipid extracts, further supporting the notion that the marine ecosystem is a rich source of bioactive compounds with potential health benefits [30]. These findings underscore the importance of exploring marine biodiversity for novel therapeutic agents. Given the limitations of current synthetic anti-inflammatory drugs, natural compounds from marine sources like *M. casta* offer promising alternatives [31]. Future research should focus on isolating and characterizing the specific bioactive compounds in these marine extracts, understanding their mechanisms of action and exploring their efficacy and safety in clinical settings. Such studies could pave the way for new, more effective, potentially safer anti-inflammatory treatments.

Antimicrobial activity: The antibacterial activity of *M. casta* extracts was evaluated against various bacterial strains. For methicillin-resistant *S. aureus* (MRSA), the ethyl acetate extract produced inhibition zones of 13.67 mm at 25 mg/mL and 17.33 mm at 50 mg/mL, while the methanol extract showed zones of 13 mm and 17.33 mm, respectively. In case of *S. typhi*, the EE extract's inhibition zones were 13.33 mm and 15.67 mm and the ME extract exhibited zones of 14 mm and 16.33 mm for the two concentrations. *E. faecalis* responded with inhibition zones of 14 mm and 17.33 mm for the EE extract and 14.33 mm and 19.33 mm for the ME extract. Notably, *S. aureus* showed significant activity for the EE extract with inhibition zones of 24.33 mm and 26.33 mm and for the ME extract with zones of 24.67 mm and 27.33 mm. *E. coli* demonstrated inhibition zones of 21.67 mm and 23.67 mm for the EE extract and 21.33 mm and 24.33 mm for the ME extract. Finally, for *P. aeruginosa*, the inhibition zones of EE extract were 17.33 mm and 19.67 mm, while the ME extract showed zones of 16.67 mm and 20.67 mm (Fig. 7). The observed antibacterial activity of *M. casta* extracts aligns with findings in similar marine organisms.

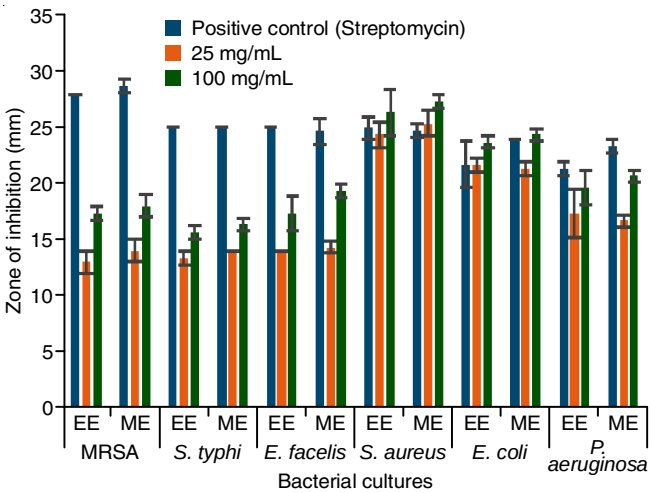

Fig. 7. Antibacterial activity of *M. casta* extract

The antifungal activity of *M. casta* extracts, as indicated by the zone of inhibition against various fungal species, demonstrates the potential of marine organisms to produce bioactive compounds with antifungal properties. Using ethyl acetate (EE) and methanol (ME) as solvent extracts, the study compared their efficacy with ketoconazole, a standard antifungal agent. For *A. niger*, the control (ketoconazole) exhibited a mean inhibition zone of 18.67 mm, while *M. casta* extracts showed promising activity, especially the methanol extract with mean zones of 16.67 mm at 200 mg/mL and 17 mm at 250 mg/mL. This efficacy mirrors findings in other marine organisms where extracts have shown significant antifungal activities against *Aspergillus* species. For *C. albicans*, ketoconazole exhibited a mean zone of 16 mm, whereas the *M. casta* extracts, particularly at 250 mg/mL, showed significant activity (Fig. 8). This is similar to the antifungal properties of marine algae, which have demonstrated effectiveness against *Candida* species. For *Candida tropicalis*, the methanol extract of *M. casta* exhibited increased activity, especially at the higher concentration, similar to the antifungal effects observed in marine fungi isolated from marine environments. Although less potent than ketoconazole, the observed antifungal activity of *M. casta* extracts is significant, considering their natural origin and the growing issue of antifungal resistance. Research on the antimicrobial activities of bivalve's Asian gold clam. (*Corbiculid* Clam), golden clam (*Corbicula fluminea*) and cyrenid clam highlights the potential of these species as sources of antimicrobial compounds [32]. Studies emphasized the broad antibacterial activity of marine bivalves, including *M. casta*, against pathogens like *E. coli* and *S. typhi* [33]. Moreover, marine sponges have shown substantial antibacterial properties, indicating a rich source of antimicrobial compounds in the marine ecosystem. Research has documented the antibacterial activity of endosymbiotic fungi from marine sponges, further supporting the diverse bioactive potential in these environments [34]. These comparative insights suggest that the antimicrobial properties of *M. casta* extracts might be attributed to unique bioactive compounds, warranting further research to isolate and characterize these compounds for potential therapeutic applications. Sponges such as those from black pinnacle sponge (*Neopetrosia*) and sea sponge (*Suberites*) have been studied for their antimicrobial compounds [35]. Studies highlighted the potential of these marine sponges from the Ratnagiri coast of India in producing novel antimicrobial agents [35]. Research on endosymbiotic fungi from marine sponges, including species like *Emericellopsis cladophorae* and *Zalerion maritima*, shows how these organisms can be sources of unique antibacterial compounds [36]. The comparative analysis underscores the importance of marine biodiversity, particularly marine bivalves like *M. casta*, sponges, fungi and algae, as potential sources of new antimicrobial compounds. These organisms, adapted to unique marine environments, have developed a variety of chemical defenses, including antimicrobial agents that hold promise for addressing the global challenge of antibiotic-resistant bacteria.

The fact that the methanol extract showed slightly higher activity suggests a difference in the composition and concentration of bioactive compounds in the extracts. The antifungal activity exhibited by *M. casta* extracts, as observed against various fungal species, can be effectively compared with the anti-fungal properties of extracts from other marine and terrestrial animals, emphasizing the broad spectrum of bioactive

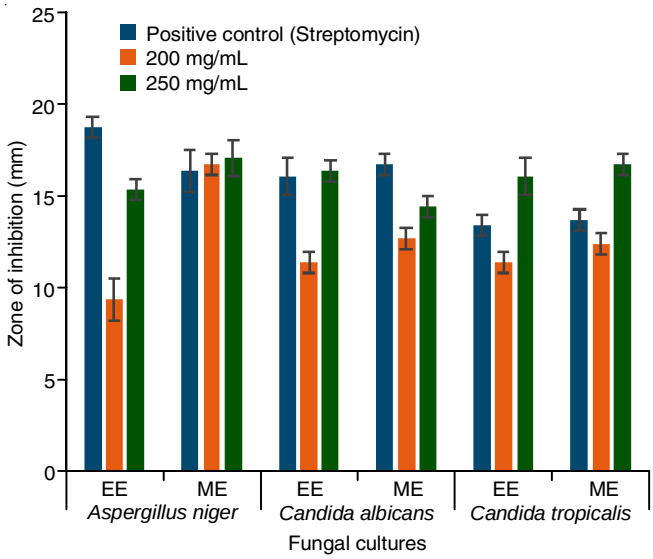

Fig. 8. Antifungal activity of *M. casta* extract

compounds in these organisms. For instance, extracts from the marine mollusk Akoya pearl oyster (*Pinctada fucata*) have been studied for their antifungal properties [37], particularly against fungi like *Candida albicans* [38]. The antifungal activities observed in *Pinctada fucata* extracts suggest a common trait among marine mollusks, including *M. casta*, to produce effective antifungal compounds [39]. Marine sponges, such as those from Golden Sponge (*Aplysina aerophoba*) and Caribbean chicken-liver sponge (*Chondrilla nucula*), are renowned for their diverse bioactive compounds with antifungal properties [40]. These sponges have been shown to exhibit significant activity against various fungal species, including *Aspergillus niger* and *Candida albicans*, paralleling the findings in *M. casta* [41]. Antifungal properties have also been identified in marine crustaceans like *Carcinus maenas* (European green crab) extracts. These extracts have shown activity against pathogenic fungi, suggesting the presence of bioactive antifungal compounds in crustaceans as well [42]. The comparison with other marine and terrestrial animals highlights the vast and largely untapped potential of animal-derived extracts as sources of novel antifungal agents. These organisms, adapted to unique ecological niches, produce a variety of chemical defences that can be harnessed for developing new antifungal therapies, especially in the face of emerging drug-resistant fungal strains [43].

Conclusion

This study underscores the significant bioactive potential of *Meretrix casta* extracts, demonstrating a wide range of properties, including antioxidant, hemolytic, anti-inflammatory, antibacterial and antifungal activities. The increase in antioxidant activity with concentration in ethyl acetate and methanol extracts aligns with findings in other marine organisms, indicating a rich presence of antioxidative compounds within the marine ecosystem. The dose-dependent hemolytic activity suggests potential for developing novel anticancer drugs, as these compounds can disrupt cell membranes. Moreover, the consistent anti-inflammatory properties, coupled with effective antibacterial and antifungal activities, highlight the therapeutic potential of *M. casta*. These results emphasize the importance of marine biodiversity, particularly marine mollusks, in discovering natural bioactive compounds. Future research should aim to isolate these compounds, understand their mechanisms of action and evaluate their efficacy for potential pharmaceutical applications. This approach could lead to the development of new therapies addressing various health challenges, including antibiotic-resistant infections and cancer.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

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