Asian J Biol Life Sci., 2025; 14(1):48-55.
https://ajbls.com

Research Article

Investigating *in vitro* Antioxidant and Anti-Inflammatory Activities of *Fromia indica* Extracts: A Pilot Study

Kavisha Kavindi Perera, Varuni Karunika Gunathilake*

Department of Zoology, Faculty of Applied Sciences, University of Sri Jayewardenepura, Nugegoda, SRI LANKA.

ABSTRACT

Background: Sea Stars are a highly abundant source of bioactive compounds but have been relatively understudied compared to other invertebrates. This study examines anti-inflammatory and antioxidant properties of zoo chemicals of *Fromia indica* marine sea star crude extract. **Materials and Methods:** *F. indica* identification was based on morphological characteristics and the Sea Star Crude Extract (SCE) was prepared by methanol and dichloromethane extraction and subsequently tested for the presence of zoo chemicals, *in vitro* radical scavenging activity by, 2, 2-diphenyl-1-picrylhydrazy (DPPH), and peroxide radicals and anti-inflammatory properties by protein denaturation. Each assay was triplicated, and the results were compared with reference drugs or chemicals where necessary. **Results:** The findings showed the presence of tannins, saponins, phenols, quinones, sterols, alkaloids, flavonoids, and unsaturated sterols, while anthraquinones were absent. Additionally, the IC_{50} values for the DPPH and H_2O_2 radical scavenging assays were 5.67 µg/mL and 3.81 µg/mL, respectively, both surpassing the values obtained for the standard drug Ascorbic acid (IC_{50}-21.66 µg/mL and 9.54 µg/mL, respectively). The *in vitro* anti-inflammatory activity indicated a dose-dependent negative linear correlation in protein denaturation, yielding an IC_{50} value of 6.22 µg/mL compared to the value obtained for standard drug ascorbic acid (95.59 µg/mL). **Conclusion:** Cumulatively, *F. indica* crude extract contained zoo chemicals, which may result the potential antioxidant properties. Further comprehensive bioactivity-guided fractionation is highly recommended.

Keywords: Antioxidant properties, DPPH assay, *Fromia indica*, Starfish.

Correspondence:
Dr. Varuni Karunika Gunathilake
Department of Zoology, Faculty of Applied Sciences, University of Sri Jayewardenepura, Nugegoda, SRI LANKA.
Email: varunig@sjp.ac.lk

Received: 14-11-2024;
Revised: 06-01-2025;
Accepted: 24-03-2025.

INTRODUCTION

Echinoderms are becoming increasingly important source of bioactive compounds that could be used in developing new medicines and therapeutic applications.[1] Starfish who belong to this phylum possess some unique characteristics which help to distinguish them from others known as Asteroids. The production of bioactive secondary metabolites in Sea Stars is quite fascinating. Peptides, glycosides, steroids, and lipids are some of the common compounds that can be found in them which possess remarkable antioxidant activity, effectively counteracting the harmful effects of free radicals and protect cells against oxidative damage. These chemical compounds are important for safeguarding starfish from potential predators as well.

Fromia indica which belongs to the family *Goniasteridae* is recognized for its unique pattern. They are often referred to as the Indian Starfish or Zebra Star. This species is identifiable by their distinctive shape, featuring a flattened body with short, triangular, and sturdy arms. The orange to brown color, the somewhat rounded nodules, and the black reticulation between the nodules set *Fromia indica* apart from other species. Perrier in 1869 reported about *Fromia indica* in his report for the first time and identification was done by A. H. Clark in 1949.[2] Since its first discovery, it has been reported on several instances from Madagascar, Andaman and Fiji Islands. Most recent record was from Sri Lanka, during a survey done at shell bay, Trincomalee.[3]

Sea Stars have not received as much attention as other marine invertebrates like sponges and mollusks, despite the growing interest in marine natural products. The exploration of Sea Stars and their bioactive compounds not only holds promise for therapeutic applications but also offers valuable insights into unexplored mechanisms of action and the potential identification of new molecular targets in drug development. This paper investigates the antioxidant and anti-inflammatory activities of *F. indica* Sea Star species, emphasizing their potential therapeutic applications and the biochemical mechanisms that drive these effects. This will lay the groundwork for further investigations and potential applications in the realm of biomedicine.

OPEN ACCESS

ScienScript

DOI: 10.5530/ajbls.20251755

Publishing Partner : ScienScript Digital. [www.sciencript.com.sg]

MATERIALS AND METHODS

Chemicals and reagents

The anti-inflammatory drug Voltaren® 50 was obtained from Union Chemists Pvt. Ltd., in Colombo, Sri Lanka. All other chemicals utilized in the study were of analytical grade or specified in other sections.

Collection, identification, and preparation of crude extract from sea stars

The Faculty of Graduate Studies granted ethical approval at the University of Sri Jayawardenepura, Sri Lanka. (FGS/ERCAS/2022/10/01), for the use of animals in research.

Live, healthy adult starfish specimens were sourced from a commercial aquarium in Wadduwa, Sri Lanka (Star Fish Paradise, 237, A2 Colombo-Galle Main Rd). Specimens were transported in ambient seawater and temporarily maintained in an outdoor recirculating tank at a controlled temperature of 28°C for 2 days. Following a two-day acclimatization period, the starfish were euthanized by rapid chilling at -20°C before proceeding with extraction.

Each specimen (weighing 180 g) was finely diced and incubated for 72 hr in a methanol and dichloromethane solution (1:1 v/v; Sigma-Aldrich, St. Louis, MI, USA) at 25°C to obtain the Sea Star Crude Extract (SCE). The mixture was subsequently filtered through Whatman No. 1 filter paper (LINCO, India) and concentrated via rotary evaporation (BUCHI R-124, Germany) at 40°C. The yield percentage of the crude extract was determined, and then the sample was dissolved in a 5% (v/v) ethanol solution to create the required concentrations for anti-inflammatory and radical-scavenging tests.

Qualitative Zoo-Chemical Analysis

A qualitative analysis of the SCE was performed to detect the presence of various zoo-chemicals, including anthraquinones, quinones, sterols, tannins, terpenoids, flavonoids, unsaturated sterols, alkaloids, phenols, and saponins. This analysis was performed following established protocols with minor modifications.[4-7]

Alkaloids Test

Test for alkaloids was performed by Wagner's Test.

About 20 mg (0.02 g) of SCE was mixed with 2 mL of 2N dilute HCl and then filtered. A few drops of Wagner's reagent were added to the resulting 2 mL filtrate. Samples showing turbidity or precipitation were recorded as positive.

Anthraquinones Test

A few drops (1-2) of 10% KOH (v/v) were added to 1 mL of SCE. Following agitation, the solution was examined for a red color appearance, indicating the presence of anthraquinones.

Flavonoids Test

This was performed by ammonia test.

To begin, 30 mL of ethyl acetate was combined with 2 mL of the sample and heated in a water bath for 3 min. After allowing the mixture to cool, it was filtered. Next, 4 mL of the filtered solution was mixed with 1 mL of a 10% dilute ammonia solution. The layers were allowed to separate, and the appearance of a yellow color in the ammonia layer signified the presence of flavonoids.

Quinones Test

A 10% (v/v) NaOH solution was precisely added to 1 mL of SCE, and the appearance of yellow, red, or purple color was observed to confirm the presence of quinones.

Saponins Test

The test for saponins was performed by foam test.

A glass vial containing around 1 mL of SCE was shaken vigorously. The presence of saponins was determined by observing the solution for its formation maintain the froth for at least 10 min.

Sterol Test

For the sterol test, 2 mL of the SCE was combined with 1 mL of chloroform, followed by the gradual addition of concentrated H_2SO_4 along the sides of the container. A reddish-brown color appearing in the chloroform layer indicated the presence of sterols.

Tannin Test

The Braemer test was conducted by adding a few drops of 1% (v/v) $FeCl_3$ to 1 mL of the SCE. The appearance of a blue-black color signified gallic tannins, while a greenish-brown color indicated tannins in general.

Table 1: Results obtained for qualitative zoo-chemical analysis.

Zoo chemical	Results
Saponins	+++
Tannins	+
Unsaturated sterols	++
Alkaloids	+
Anthraquinones	-
Quinones	++
Sterols- Salkwoski test	+
Phenols	+
Flavonoids	+
Terpenoids	+

The tested zoo-chemical was found in significant amounts, yielding a positive result within 5 min (+++); detected in moderate quantities, showing a positive result between 5 to 10 min (++); found in trace amounts, resulting in a positive outcome from 10 to 15 min (+); and completely absent (-).

Terpenoid Test

Approximately 1 mL of the SCE extract was mixed with 2 mL of concentrated H_2SO_4 in a boiling tube and heated in a water bath at 40°C for 2 min. A brown to red color in the solution confirmed the presence of terpenoids.

Test for Unsaturated Sterols

To detect unsaturated sterols, 1-2 drops of concentrated H_2SO_4 were carefully added dropwise along the wall of the container holding 1 mL of SCE. The presence of a red ring at the interphase indicated unsaturated sterols.

Phenol Test

Three drops of 10% (v/v) $FeCl_3$ solution were added to 3 mL of the SCE. The appearance of a blue-violet or greenish color indicated the presence of phenols.

Antioxidant assays

Antioxidant assays were carried out for the SCE. Ascorbic acid was used as the standard drug in all antioxidant experiments.

2,2-Diphenyl-1-Picryl-Hydrazil (DPPH) assay

To conduct the experiment, 100 µL of various concentrations of SCE (2.0, 1.5, 1.0, 0.5, and 0.25 µg/mL) or the reference compound, ascorbic acid (5.0, 3.0, 2.0, 1.0, and 0.5 µg/mL), was mixed with 100 µL of 125 µM DPPH solution and incubated for 30 min at room temperature in the dark. The reduction in color from deep violet to pale yellow was observed by measuring absorbance at 517 nm with a microplate reader (model: s/n MR05405, USA). A control was prepared using DPPH and 5% ethanol.

The percentage inhibition of the DPPH radicals was calculated using the following formula:

$$\text{Percentage inhibition (\%)} \frac{\text{Abs control} - \text{Abs sample}}{\text{Abs control}} \times 100$$

(Abs control-absorbance of negative control, Abs sample-absorbance of test sample),

The IC_{50} for 50% DPPH radical scavenging has been calculated for both SCE and standard drug.[8]

Peroxide radicals scavenging assay

For the peroxide radical scavenging assay, 850 µL of SCE (10.0, 6.0, 4.0, 3.0, and 2.0 µg/mL) or ascorbic acid (20.0, 10.0, 7.0, 5.0, and 4.0 µg/mL) was combined with 150 µL of 4 mM hydrogen peroxide solution in PBS (pH 7.4). Absorbance was measured at 230 nm after 10 min. A solution containing 5% ethanol served as the negative control.

Percentage inhibition of peroxide radicals was calculated using the following formula:

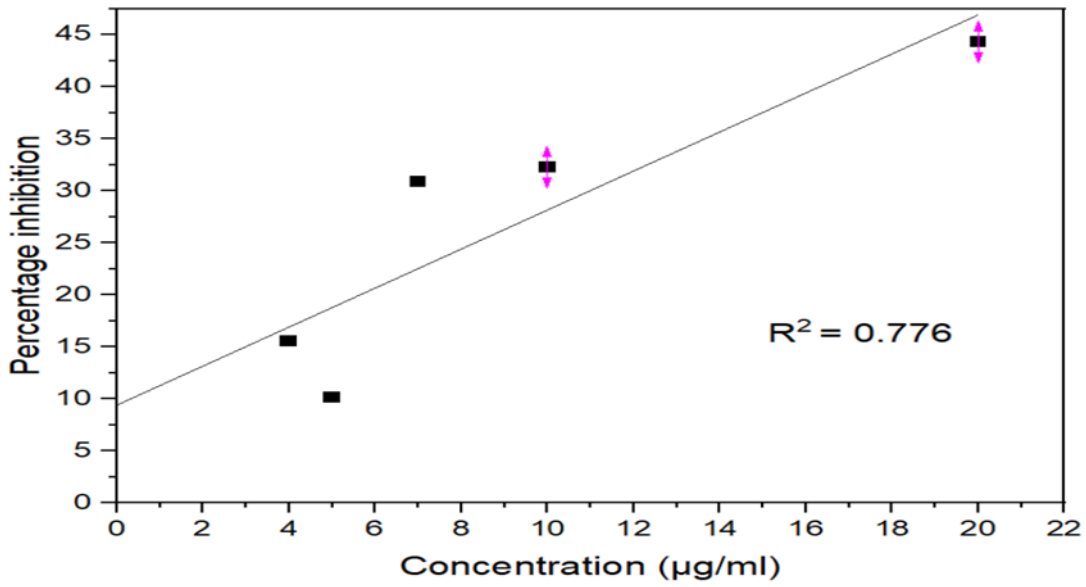
$$\text{Percentage inhibition (\%)} \frac{\text{Abs control} - \text{Abs sample}}{\text{Abs control}} \times 100$$

(Abs control-absorbance of negative control, Abs sample-absorbance of test sample),

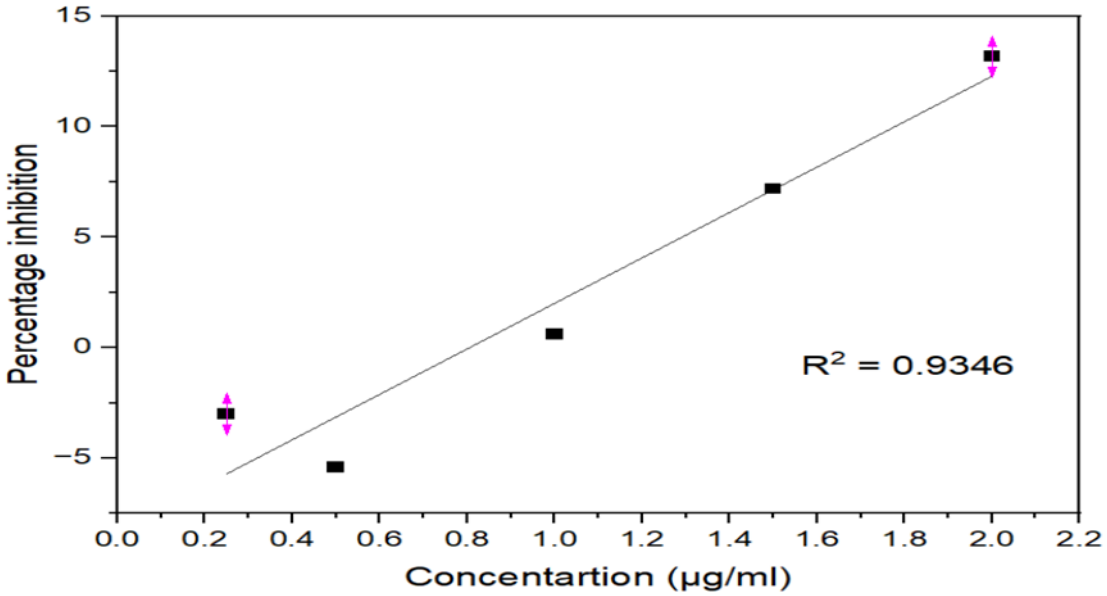
The IC_{50} for 50% peroxide radical scavenging was calculated for both SCE and standard drug.[9]

Statistical analysis

Each assay was triplicated, and results were presented as mean±Standard Error Mean (SEM). The graphs were created with OriginPro®2023b, and the equations from each graph were



A



B

Figure 1: Percent free radical scavenging activity against H2O2 radicals with increasing concentrations. (A) Standard drug ascorbic acid (20.00, 10.00, 7.00, 5.00, and 4.00 µg/mL) and (B) SCE (10.00, 6.00, 4.00, 3.00, and 2.00 µg/mL). The scavenging activity increased with increasing concentration in both Standard drug ascorbic acid and SCE and had strong positive linear relationships with concentration (r=0.906, p=0.034 and r=0.959, p=0.010 respectively). Ascorbic acid and crude extract had IC_{50} values of 9.54 µg/mL and 3.81 µg/mL, respectively. Data are presented as mean±SEM (n=3).

used to calculate the LC_{50} and IC_{50} values for each test. For each linear regression model, the coefficient of determination (R^2) was calculated to indicate the goodness-of-fit measure. Minitab® 20.4 statistical software was used to analyze the results. The form and magnitude of the correlation between the two variables were assessed using Pearson's product-moment correlation. To compare the mean values of each treatment group and controls, a one-sample t-test was performed. The applied parametric tests had a 95% confidence interval, and statistical differences were significant for a p value <0.05.

RESULTS

Qualitative Zoo chemicals

The results of the major zoo chemicals are indicated in Table 1, Saponins were present in considerable amounts whereas tannins were present in trace amounts. Moderate amounts of quinones, phenols, flavonoids, sterols, alkaloids, and unsaturated sterols were present while anthraquinones were absent in the Sea Star Crude Extract (SCE) (Table 1).

Antioxidant activity of the SCE by *in vitro* antioxidant assays

DPPH scavenging activity

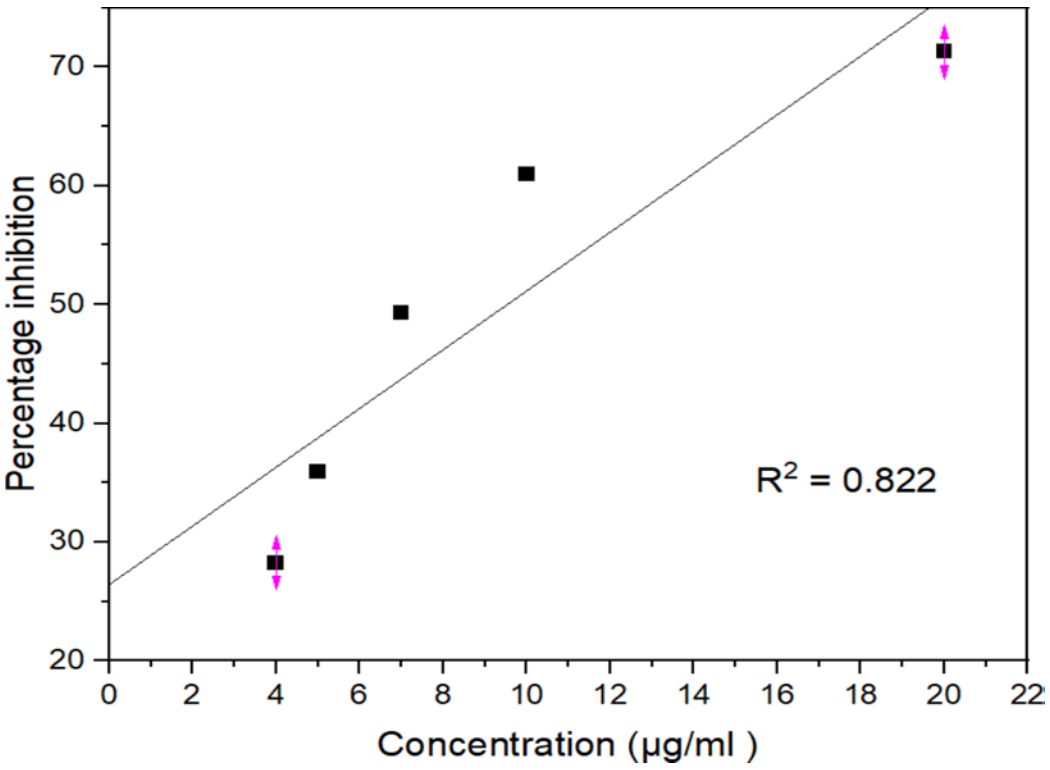
The *in vitro* scavenging activity against DPPH free radicals of the ascorbic acid and SCE is given in Figure 1. The color change from purple to yellow was recorded for both ascorbic acid and SCE. The scavenging activity increased with increasing concentration in both Standard drug ascorbic acid and crude extract and had strong positive linear relationships with concentration (r=0.881, p=0.049 and r=0.967, p=0.007, respectively). Ascorbic acid and SCE resulted an IC_{50} value of 21.66 µg/mL and 5.67 µg/mL, respectively, indicating the SCE is more potent than the reference drug (Figure 1).

Peroxide radicals scavenging activity

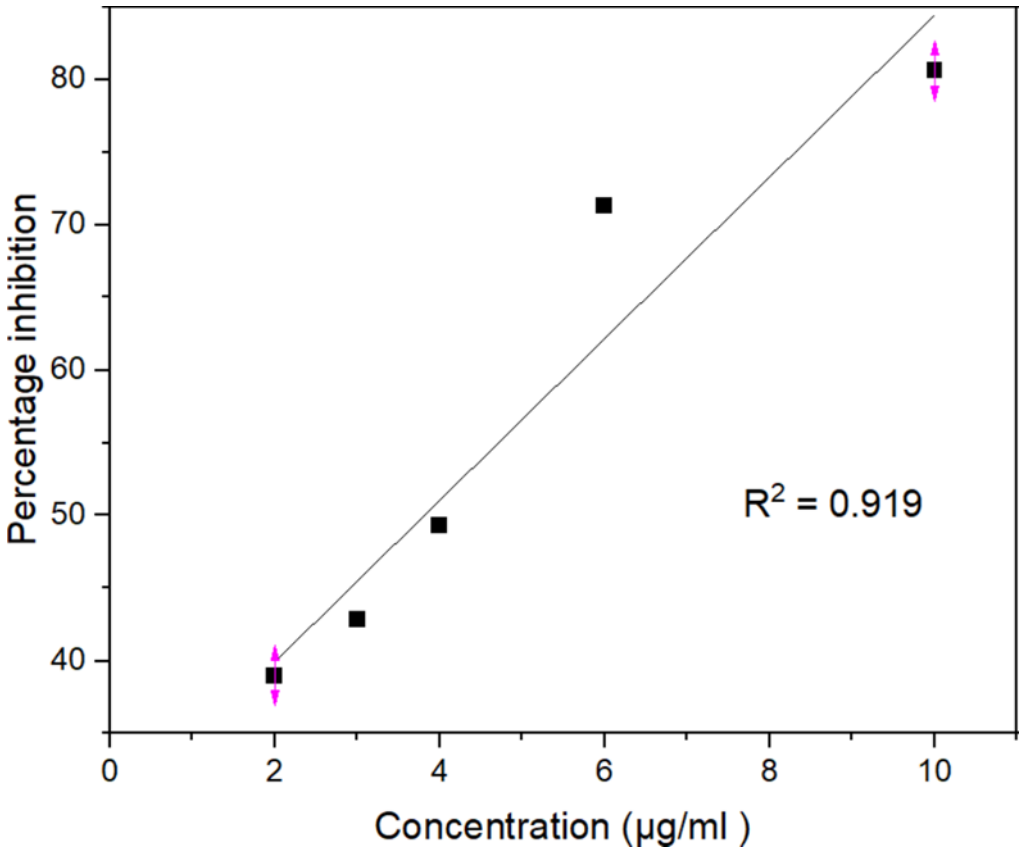
The hydrogen peroxide scavenging activity of standard drug Ascorbic acid and SCE at varying concentrations is given by Figure 2. Strong positive relationship of inhibition was observed in both the standard drug ascorbic acid and SCE with increasing concentration. (r=0.906, p=0.034 and r=0.959, p=0.010 respectively). Ascorbic acid and SCE resulted an IC_{50} values of 9.54 µg/mL and 3.81 µg/mL, respectively indicating that the SCE is more potent than the reference drug. According to IC_{50} values, SCE requires a lower concentration to scavenge peroxide radicals compared to standard drug ascorbic acid (Figure 2).

In vitro anti-inflammatory activity of SCE

The inhibitory effects of diclofenac sodium and the Standardized Crude Extract (SCE) at different concentrations are illustrated in Figures 3 and 4. The standard drug, diclofenac sodium, demonstrated a dose-dependent inhibitory effect, characterized by a strong positive linear correlation (r=0.992, p=0.001). In contrast, the crude extract exhibited a significant negative linear correlation between concentration and its inhibitory effect



A



B

Figure 2: Percent free radical scavenging activity against H2O2 radicals with increasing concentrations. (A) Standard drug ascorbic acid (20.00, 10.00, 7.00, 5.00, and 4.00 µg/mL) and (B) SCE (10.00, 6.00, 4.00, 3.00, and 2.00 µg/mL). The scavenging activity increased with increasing concentration in both Standard drug ascorbic acid and SCE and had strong positive linear relationships with concentration (r=0.906, p=0.034 and r=0.959, p=0.010 respectively). Ascorbic acid and crude extract had IC_{50} values of 9.54 µg/mL and 3.81 µg/mL, respectively. Data are presented as mean±SEM (n=3).

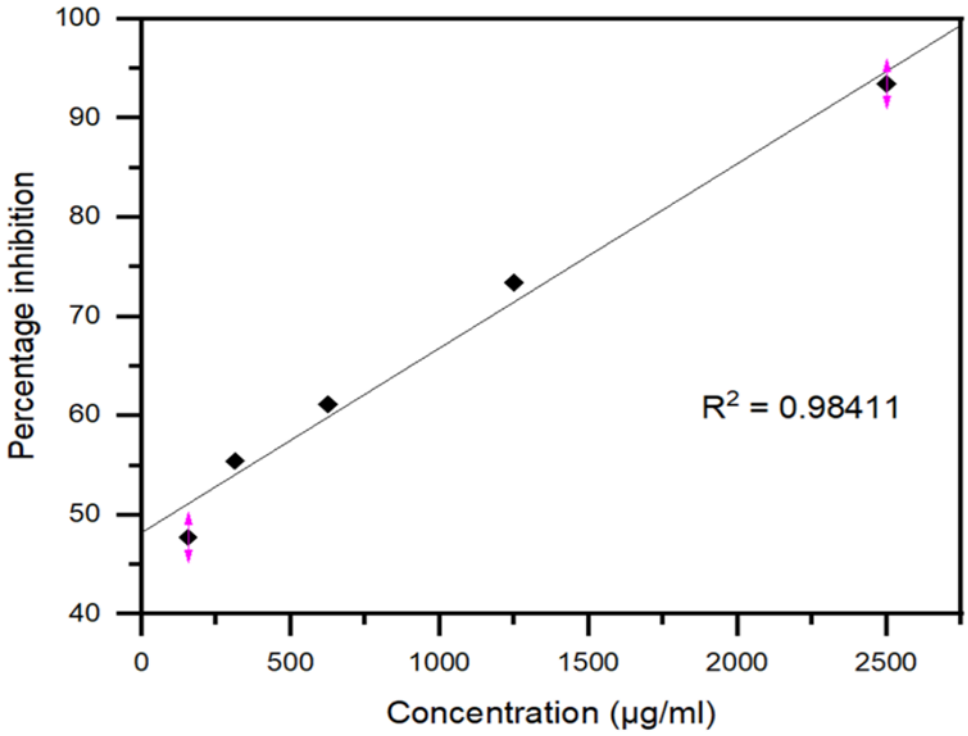


Figure 3: The percentage inhibition of standard drug diclofenac sodium of egg albumin denaturation with increasing concentrations. A dose dependent inhibitory effect was observed for standard drug diclofenac sodium with a strong positive linear correlation. (r=0.992, *p*=0.001) with an IC_{50} value of 95.59 µg/mL. Data are presented as mean±SEM (*n*=3).

(r=-0.990, *p*=0.001). The calculated IC_{50} values for ascorbic acid and SCE were 95.59 µg/mL and 6.22 µg/mL, respectively (Figures 3 and 4).

DISCUSSION

The limited research on sea stars in drug discovery has resulted in a scarcity of data on the bioactive properties of sea star crude extracts, particularly those from the Indian Ocean. While previous studies have predominantly investigated *Acanthaster planci*, which is known for its rich metabolite content.[10,11] The present study is the first to examine the bioactive potential of *Fromia indica*, a common Indian Ocean species and a popular aquarium species in Sri Lanka. This investigation focused on evaluating the anti-inflammatory, antioxidant activities, and toxicity of the *F. indica* Sea Star Crude Extract (SCE).

Our study revealed significant levels of saponins, sterols, phenols, and flavonoids in *F. indica* crude extract, alongside moderate quantities of alkaloids, terpenoids, quinones, and unsaturated sterols. Saponins, specifically asterosaponins containing sulfate groups, are prominent within the order Valvatida, to which *F. indica* belongs.[12] Previous research on *Fromia milleporella,* a congener, reported steroidal glycosides of the saponin group, highlighting the potential cytotoxicity of this class of compounds.[13] The sterols, unsaturated sterols, and saponins identified in *F. indica* may therefore possess cytotoxic effects, indicating its potential as a candidate for cancer-related drug development.[14,15] Additionally, saponins can modulate pro-inflammatory and anti-inflammatory cytokine responses, supporting the observed anti-inflammatory properties of *F. indica* extract.[16]

Flavonoids, known for their ability to neutralize various free radicals, are naturally occurring antioxidant compounds.[17] The considerable flavonoid content in *F. indica* extract affirms its antioxidant capacity. Notably, phenolic compounds in marine species, such as brittle stars, have been reported in trace amounts and are responsible for antioxidant properties.[18] Similarly, studies have identified phenols in *A. planci* and *Linckia laevigata*, attributing these compounds to chemical defenses against predation.[17] Although moderate flavonoid levels were detected in *F. indica*, further quantification is recommended to better understand its antioxidant profile.

Reactive oxygen and nitrogen species are well-known contributors to oxidative damage in human cells, leading to pathologies such as cancer, diabetes, atherosclerosis, and chronic inflammation.[19] The radical scavenging potential of SCE was evaluated against two radicals, DPPH and peroxide, demonstrating a strong positive correlation between concentration and radical scavenging with IC_{50} values of 5.67 µg/mL and 3.81 µg/mL, respectively. These values are notably lower than those of the standard antioxidant, indicating the potent radical scavenging ability of SCE. DPPH, a stable radical commonly used in antioxidant assays, revealed an IC_{50} value for

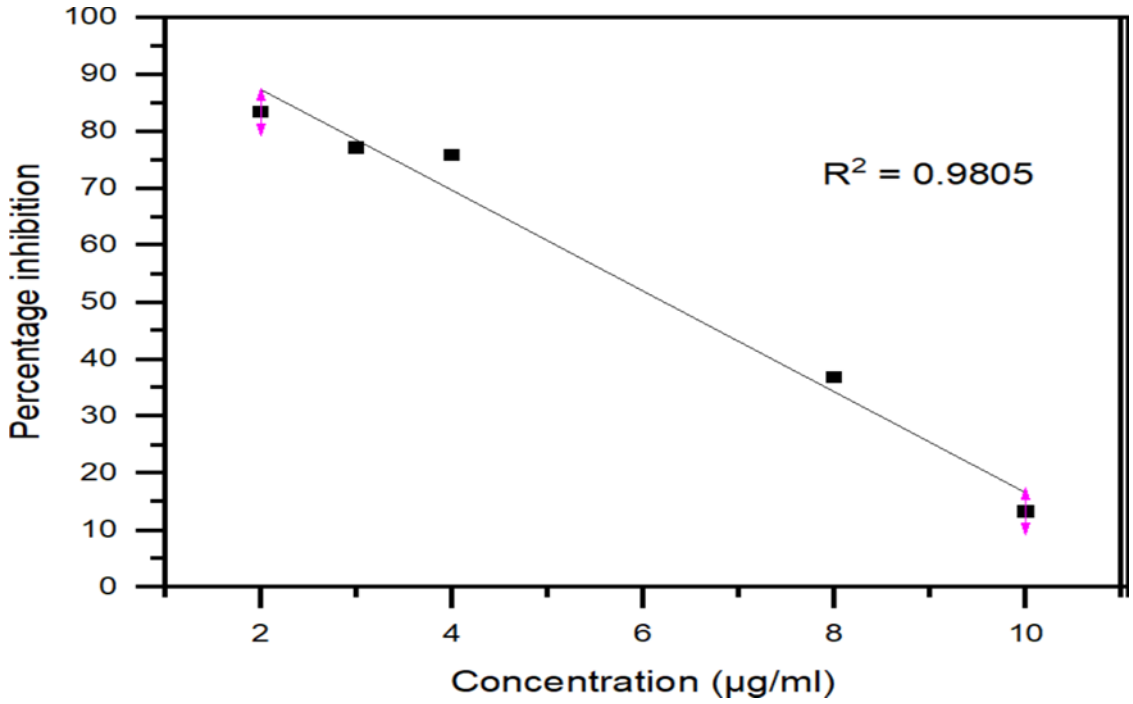


Figure 4: The percentage inhibition of SCE of egg albumin denaturation with increasing concentrations (2.00, 3.00, 4.00, 8.00 and 10.00 µg/mL). A dose dependent inhibitory effect was not observed for SCE with a strong negative linear correlation. (r=-0.990, p=0.001) with an IC_{50} value of 6.22 µg/mL. Data are presented as mean±SEM (n=3).

F. indica comparable to that of *A. planci* (6.58 µg/mL), a species also within the Valvatida order assay.[20,21]

Inflammation, often resulting from tissue damage in conditions like atherosclerosis, fever, and cardiovascular issues, can be assessed effectively using *in vitro* anti-inflammatory models due to their adherence to the 3Rs principle (replacement, reduction, and refinement).[22] The present investigation employed *in vitro* experiments to assess the ability of SCE to prevent protein denaturation. The results indicated that SCE effectively inhibited the denaturation of egg albumin induced by heat, exhibiting an IC_{50} value that was lower than that of the standard drug used for comparison. Although the literature on related species within Valvatida supports anti-inflammatory properties,[16,23] the present study observed a negative linear correlation in inhibition percentage with increasing extract concentration, suggesting dose-dependent anti-inflammatory activity of SCE.

The exploration of bioactive compounds from sea stars remains underdeveloped compared to other marine sources, likely due to challenges and costs associated with their collection and extraction. In Sri Lanka, marine starfish research is still in its early stages, in contrast to global progress, which leaves a considerable research gap. *Fromia indica*, first reported by J.O.E. Perrier in 1869,[24] has since been recorded across a wide range of Indo-Pacific locations, including Australia, Fiji, Indonesia, and Madagascar. Despite occasional mentions in literature detailed taxonomic records are limited, with only one comprehensive description published in 2018 in ZooTaxa[25] up-to-date but there are several studies which has mentioned about this particular species.[26,27]

CONCLUSION

In conclusion, the SCE from *Fromia indica* demonstrated considerable bioactivity with a promising yield. The zoo-chemical analysis confirmed the presence of key bioactive groups, including saponins, phenols, and flavonoids, which likely underlie the observed antioxidant and anti-inflammatory activities. This initial exploration highlights the therapeutic potential of *F. indica* and suggests further bioactivity-guided fractionation to isolate and characterize its active compounds for potential pharmacological applications.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

SCE: Sea Star Crude Extract; **DPPH:** 2, 2-diphenyl-1-picrylhydrazy; **NaOH:** Sodium hydroxide; **H_2SO_4:** Sulfuric acid; **$FeCl_3$:** Ferric chloride; **HCl:** Hydrochloric acid; **KOH:** Potassium hydroxide; **PBS:** Phosphate Buffered Saline.

FUTURE PERSPECTIVES

Further studies based on bioactivity guided fractionation is highly recommended. Moreover, bioassays for hemolytic effects and evaluation of cytotoxicity using animal and human cell lines are further recommended.

SUMMARY

Crude extracted obtained from *Fromia indica* sea star revealed its bioactive properties with a considerable yield. The qualitative zoo-chemical analysis revealed the presence of saponins, phenols, and flavonoids, which could explain the observed bioactivities. Evaluation of anti-oxidant and anti-inflammatory properties revealed its potential use in pharmacology for the development of potent cytotoxic drugs.

REFERENCES

1. Gomes AR, Freitas AC, Duarte AC, Rocha-Santos TAP. Chapter 1-Echinoderms: A Review of Bioactive Compounds With Potential Health Effects. In: Atta-ur-Rahman, editor. Studies in Natural Products Chemistry, Elsevier; 2016;49:1-54. https://doi.org/10.1016/B978-0-444-63601-0.00001-6.
2. Perrier JOE. Investigations on the Pedicellariae and the Tube Feet of Asteroids and Urchins. Herizos Press; 1869. https://doi.org/10.5038/bin.books.1012.
3. Rajasuriya A. Survey Report on Marine Biodiversity of Shell Bay, Trincomalee n.d.
4. Kuruppuarachchi S, Gunathilake V. Zoo-chemical profiling, *in vivo* toxicity and *in vitro* anti-inflammatory properties of Luffariella herdmani marine sponge extract 2022;5:269-82. https://doi.org/10.5455/jabet.2022.d114.
5. Walag AMP, Rosario RMD, Canencia OP. Zoochemical Composition of Selected Sea Stars Collected from the Coastal Waters of Carmen, Agusan Del Norte, Philippines. Asian Journal of Biological and Life Sciences 2019;8. https://doi.org/10.5530/ajbls.2019.8.10.
6. Farnsworth NR. Biological and phytochemical screening of plants. J Pharm Sci 1966;55:225-76. https://doi.org/10.1002/jps.2600550302.
7. Muthiyan R, Ponraj P, Muniswamy K, Bhattacharya D, Kundu A, Sunder J, *et al.* Zoo chemical profile analysis and cytotoxicity screening of five marine sponge species collected from Andaman and Nicobar Islands. Indian Journal of Geo-Marine Sciences 2020;49.
8. Prastya ME, Astuti RI, Batubara1 I, Wahyudi 2 and A. T. Indian Journal of Pharmaceutical Sciences n.d.
9. Gülçin İ, Huyut Z, Elmastaş M, Aboul-Enein HY. Radical scavenging and antioxidant activity of tannic acid. Arabian Journal of Chemistry 2010;3:43-53. https://doi.org/10.1016/j.arabjc.2009.12.008.
10. Lazzara V, Arizza V, Luparello C, Mauro M, Vazzana M. Bright Spots in the Darkness of Cancer: A Review of Starfishes-Derived Compounds and Their Anti-Tumor Action. Marine Drugs 2019;17:617. https://doi.org/10.3390/md17110617.
11. Lee Y-C, Moon J-Y. Introduction to Bionanotechnology. Singapore: Springer; 2020. https://doi.org/10.1007/978-981-15-1293-3.
12. Dong G, Xu T, Yang B, Lin X, Zhou X, Yang X, *et al.* Chemical Constituents and Bioactivities of Starfish. Chemistry and Biodiversity 2011;8:740-91. https://doi.org/10.1002/cbdv.200900344.
13. Levina EV, Kalinovsky AI, Levin VS. New steroid glycosides from the starfish *Fromia milleporella*. Russ J Bioorg Chem 2006;32:84-8. https://doi.org/10.1134/S1068162006010092.
14. Claereboudt E, Eeckhaut I, Lins L, Deleu M. How different sterols contribute to saponin tolerant plasma membranes in sea cucumbers. Scientific Reports 2018;8. https://doi.org/10.1038/s41598-018-29223-x.
15. Bouabdallah S, Al-Maktoum A, Amin A. Steroidal Saponins: Naturally Occurring Compounds as Inhibitors of the Hallmarks of Cancer. Cancers (Basel) 2023;15:3900. https://doi.org/10.3390/cancers15153900.
16. Stonik VA, Kicha AA, Malyarenko TV, Ivanchina NV. Asterosaponins: Structures, Taxonomic Distribution, Biogenesis and Biological Activities. Marine Drugs 2020;18:584. https://doi.org/10.3390/md18120584.
17. Walag AM, Rosario R. Total Flavonoids Content, Total Phenolics Content, and Antioxidant Activities of *Acanthaster planci* and *Linckia laevigata* collected from Carmen, Agusan del Norte, Philippines. Malaysian Journal of Biochemistry and Molecular Biology 2020;23:77-85.
18. Baharara J, Amini E. Phytochemical Screening, Antioxidant Effect and Down Regulation of TGF-β Induced by *Ophiocoma erinaceus* Brittle Star Crude Extract. Zahedan J Res Med Sci 2015;17. https://doi.org/10.17795/zjrms-5194.
19. Ekin S, Bayramoglu M, Goktasoglu A, Ozgokce F, Kiziltas H, Ekin S, *et al.* Antioxidant activity of aqueous and ethanol extracts of *Crataegus meyeri* Pojark leaves and contents of vitamin, trace element. Journal of the Chilean Chemical Society 2017;62:3661-7. https://doi.org/10.4067/s0717-97072017000403661.
20. Lee C-C, Hsieh HJ, Hsieh C-H, Hwang D-F. Antioxidative and anticancer activities of various ethanolic extract fractions from crown-of-thorns starfish (*Acanthaster planci*). Environmental Toxicology and Pharmacology 2014;38:761-73. https://doi.org/10.1016/j.etap.2014.08.021.
21. Kumar YN, Poong S-W, Gachon C, Brodie J, Sade A, Lim P-E. Impact of elevated temperature on the physiological and biochemical responses of *Kappaphycus alvarezii* (Rhodophyta). PLOS ONE 2020;15:e0239097. https://doi.org/10.1371/journal.pone.0239097.
22. Leelaprakash G, Dass SM. *In vitro* anti-inflammatory activity of methanol extract of *Enicostemma axillare*. International Journal of Drug Development and Research 2011;3.
23. Thao NP, Luyen BTT, Koo JE, Kim S, Koh YS, Cuong NX, *et al.* Anti-inflammatory components of the Vietnamese starfish *Protoreaster nodosus*. Biol Res 2015;48:12. https://doi.org/10.1186/s40659-015-0002-2.
24. Perrier E. Recherches sur les pédicellaires et les ambulacres des astéries et des oursins. Académie de Paris; 1869.
25. Mah CL. New genera, species and occurrence records of Goniasteridae (Asteroidea; Echinodermata) from the Indian Ocean. Zootaxa 2018;4539:1-116. https://doi.org/10.11646/zootaxa.4539.1.1.
26. James PSBR, Thomas PA, Pillai CSG, Achari GPK. Catalogue of Types and of Sponges, Corals, Polychaetes, Crabs and Echinoderms in the Reference Collections of the Central Marine Fisheries Research Institute. CMFRI Bulletin 1969;7:1-66. http://eprints.cmfri.org.in/567/ (accessed October 29, 2023).
27. Samuel VKD, Krishnan P, Sreeraj CR, Chamundeeswari K, Parthiban C, Sekar V, *et al.* An updated checklist of Echinoderms from Indian waters. Zootaxa 2017;4354:1-68. https://doi.org/10.11646/zootaxa.4354.1.1.

Cite this article: Perera KK, Gunathilake VK. Investigating *in vitro* Antioxidant and Anti-Inflammatory Activities of *Fromia indica* Extracts: A Pilot Study. Asian J Biol Life Sci. 2025;14(1):48-55.