INVESTIGATION OF PHYTOCHEMICAL PROPERTIES OF THE METHANOLIC EXTRACT OF *ROSENVINGEA SPP.* FOUND IN THE NORTH-EASTERN REGION OF THE BAY OF BENGAL

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ABSTRACT

This study aimed to screen the phytochemical profile, antioxidant activity, and antibacterial activity of a 70% methanolic extract of *Rosenvingea sp*., which was collected from Bay of Bengal, Bangladesh. A total of five phytochemicals were detected in the *Rosenvingea* methanolic extract, including steroids, glycosides, flavonoids, alkaloids, and tannins.

The 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging experiment exhibited antioxidant activity though it is a weak antioxidant than standard ascorbic acid. The 70% methanol extract had an LC50 of 13.26 mg/mL in the Artemia lethality bioassay, whereas the positive control ($K_2Cr_2O_7$) had an LC50 of 59.97 µg/mL, indicating the non-toxicity in both Mayer's and Clarkson's indexes.

The agar disc diffusion method was used to investigate the antibacterial activity of a methanol extract of *Rosenvingea sp*. against Gram-positive and Gram-negative bacteria species (*Staphylococcus aureus, Bacillus cereus, Pseudomonas aeruginosa, Salmonella, and Klebsiella pneumonia*) which showed strong antibacterial activity mostly against *Staphylococcus aureus, Pseudomonas aeruginosa, Klebsiella pneumonia, and Salmonella* with zones of inhibition of 6.66±1.15mm, 5.66±0.57mm, 5.33±0.57 mm & 3.33±0.57 mm respectively. This study found *Rosenvingea* sp. from the Bay of Bengal to be a promising source of phytochemical, antioxidant, and antibacterial properties. However, further research is needed to establish its economic potential in the food and pharmaceutical industries.

KEYWORDS: *Rosenvingea,* Antibacterial, Antioxidant, Methanolic extract, DPPH, Cytotoxic, Phytochemical screening.

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Introduction

Many papers in recent years have focused on structurally novel and physiologically active metabolites discovered in marine sources. *Rosenvingea* spp. (Scytosiphonaceae, Phaeophyceae) are brown seaweeds defined by cylindrical to moderately compressed, dichotomous or alternatively branched, upright, hollow thalli with plurangia creating surface sori. Currently ten known *Rosenvingea* species are listed worldwide, including *R. sanctae-crucis* Børgesen, *R. fastigiated* Børgesen, *R. intricate* Børgesen, *R. orientalis* Børgesen, *R. floridana*, *R. endiviifolia*, *R. hatrangensis*, *R. antillarum*, *R. australis*.(Santiañez and West, 2019), and *R. stellata* Børgesen,(*World Register of Marine Species*, 2020). Brgesen (1914) was the first to identify the genus *Rosenvingea* (Phaeophyceae, Scytosiphonaceae), and their molecular research revealed the genus is consist of several species. (Kogame and Horiguchi, 1999; Jo, Kogame and Boo, 2006; West *et al.*, 2010; Lee, Hong and Boo, 2014; Huisman, Boo and Boo, 2018; Alim*et al.*, 2021). *Rosenvingea* sp. and their various habitats in the Arabian Sea are taxonomically characterized (Aisha and Shameel, 2016).

Brown algae from the Pacific ocean have been intensively studied for their culture and asexual life cycle (West *et al.*, 2010) and molecular techniques have been utilized to explore their potential for usage in food and medicine (Klochkova*et al.*, 2017). Additionally, microorganisms linked with marine macroalgae have been reported for a wide range of antibacterial activity and prospective therapeutic agents for pharmaceutical development (Kizhakkekalam and Chakraborty, 2019) as well as for increasing photosynthetic capacity. It has also been reported that endophytic microorganisms associated with marine algae aid to the growth promoting agents of plant (Tapia *et al.*, 2016) and has immense pharmacological value (Chowdhury KR, 2022). Brown algae have recently been recognized as a rich source of bioactive molecules such as phloroglucinol and its derivatives, which have anti-inflammatory, antiproliferative, antioxidant, antidiabetic, anti-allergic, and anti-aging properties. The potential role for neuroprotection, diverse antifungal effects including activity against fluconazole-resistant strains of fungi, and an anticancer impact on certain cancer cell lines are

additional health benefits that have been discovered (Abdel hamid *et al.*, 2018; Martins *et al.*, 2018).

According to World Health Organization (WHO) approximately 422 million people worldwide are affected with diabetes, which is one of the world's leading causes of mortality and is most prevalent in low-income countries. Brown seaweeds can help control diabetes as the prevalence of the disease rises (J., 2017; Ahsan, Islam and Hossain, 2020; Alim*et al.*, 2021). Leishmaniasis is a neglected tropical illness spread by female Phlebotomus and Lutzomyia sand flies and caused by protozoan parasites of the *Leishmania* genus. Marine algae have also been shown to exhibit antiprotozoan activity and have been proven to be particularly effective against Leishmaniasis (TchokouahaYamthe*et al.*, 2017).

Human acute promyelocytic leukemia cells (HL-60), human breast carcinoma cells (MCF-7), and human hepatocellular carcinoma cancer cell lines notably responded to the sulfated polysaccharide H3-a1 derived from edible brown seaweed (Wang *et al.*, 2010).

As an alternative to vegetables, seaweeds act as a storehouse for a large number of macronutrients and micronutrients. They are also a possible source of natural antioxidants, high-quality PUFA, and minerals (Kumar *et al.*, 2011). They are also used as a rich essential fatty acid source for mammals (Nogueira*et al.*, 2017). Aside from these, seaweed liquid fertilizer is gaining popularity since it contains crucial plant growth elements such as auxin, cytokinin, and gibberellins, which are essential for plant development, seed germination, and photosynthetic coloration (Thirumaran*et al.*, 2009).

In Bangladesh, 193 seaweed species have been discovered so far, with 19 of them being commercially important and connected with 94 genera (Md. Shirajul Islam Sarkar1, 2, Md. Kamal2* and Hossain2, 2016). Our study aimed to elucidate the prospects of the brown algae *Rosenvingea* and the methanolic extract of *Rosenvingea* from St. Martin Island of Bay of Bengal has been shown to have cytotoxic, antibacterial, and antioxidant properties. However, further research is required to understand their commercial significance.

Materials and Methods

Sample Collection and Processing

The samples of *Rosenvingea* were taken from St. Martin Island in Bangladesh, which is about 13 kilometers from the mainland and is roughly between 20°37′16.9" N and 267°41′11.256" W., The collecting area's water varied between 22 to 29 degrees Celsius and salinity 21.0 to 33.5PSU (Khan *et al.*, 2016; Alim*et al.*, 2021). To remove epiphyte, debris, and other wastes, the sample was rinsed with fresh seawater. It was then cleaned with distilled water and stored in a two-liter light-protective container filled with 50% ethanol. The World Register of Marine Species, Macroalgal Herbarium Portal, and Algae Base recognized the acquired sample ("algae base. *Rosenvingae*," 1914; *World Register of Marine Species*, 2020; *Macroalgal Herbarium Portal Homepage*, 2020; Alim*et al.*, 2021).The sample was removed from preservation, rinsed once more with MiliQ water, and cut into slices for shade-drying at 37°C (Model: JSON-030S,69, Geomsanggogae-GIL, Gongju-SI, Korea,32598). The dried sample was crushed in mortal pestle and then the powdered sample was maintained at room temperature or -20ºC until further use.

Extract preparations

Extract prepared by 70% methanol at concentration of 1gm/10mL(1:10) was maintained for 4 weeks at 25°C in a shaking incubator rotating at 150 rpm. The samples were filtered by double-layer filter paper (Whatman® qualitative filter paper, Grade 1 circles, diam.15 mm) after maceration (Mansuya*et al.*, 2010; Vimalkumar, Vilash, V. and Krishnakumar, 2014; Balachandran *et al.*, 2016; Gul*et al.*, 2017; Alim*et al.*, 2021).

Phytochemical screening

Using various approaches, like acetate test, ferric chloride test, Mayer's test, Salkowski's test, alkaline reagent test and foaming test, the qualitative phytochemical examination of phenolic compounds, tannins, alkaloids, steroids, steroidal glycosides, flavonoids, and saponins were carried out (Vimalkumar, Vilash, V. and Krishnakumar, 2014; Daisy A *et al.*, 2016; Gul*et al.*, 2017; Dahanayake *et al.*, 2019; Jagri, 2019; Ahsan, Islam and Hossain, 2020; Islam *et al.*, 2020; Alim*et al.*, 2021; Rahman *et al.*, 2021)

DPPH Scavenging Activity

A previously established approach was used to measure DPPH scavenging activity (Cahyana, Shuto and Kinoshita, 1992; Ponnanikajamideen, M. Malini, M., Malarkodi, C., Rajeshkumar, 2014; Das, 2015; Abdelhamid*et al.*, 2018; Alam, 2019; Ahsan, Islam and Hossain, 2020; Islam *et al.*, 2020; Tlili and Sarikurkcu, 2020; Alim*et al.*, 2021; Rahman*et al.*, 2021) with some modification. In a total amount of 100mL, 0.04mg/mL of DPPH (2,2-diphenyl-1-picrylhydrazyl) (Catalog no: SC-202591, Santa Cruz Biotechnology, USA) was dissolved in 95% methanol (Batch no: G16A/0816/2707/21, SD Fine Chem Ltd, Mumbai, India), 1.5 mL of sample (50µl, 100µL, 150µL, 200µL, 250µL and300µL of sample were added to 1450µl, 1400µl, 1300µl, 1250µL and 1200µL of sample solvents, respectively) mixed with 1.5mL of DPPH solution and kept in darkness for 1 hour at room temperature. The absorbance was then measured with a UV-VIS spectrophotometer at 517 nm (model: UV-1900, Shimadzu).

The following formula is used to compute the DPPH free radical scavenging activity.

% inhibition= {1- (A sample-Ablank)/ (A control-Ablank)} x 100

Where A sample= Absorbance of the sample (Sample dilution + DPPH solution)

A blank= Absorbance of blank for each sample dilution (sample dilution + DPPH solvent)

A control= Absorbance of control reaction (sample solvent + DPPH solution)

L-ascorbic acid was used as a positive control. All experiments were performed in triplicate.

Brine Shrimp Lethality Assay (BSLA)

The previously reported brine shrimp lethality experiment was utilized to investigate the cytotoxicity activity of the 70% methanolic extract (Lee, Min and Kho, 2002; M Alam Morshed1*, Azim Uddin1, Rahman Saifur2, Anik Barua3, Anwarul Haque2, 2011; Ogbole, Segun and Adeniji, 2017; Sm and M, 2019; Waghulde*et al.*, 2019; Ahsan, Islam and Hossain, 2020; Islam *et al.*, 2020; Alim*et al.*, 2021; Rahman*et al.*, 2021) with little modification. For 24 hours, 200 mg of

brine shrimp (*Artemiasalina*) eggs were hatched in 1.5 liters of direct seawater with continuous aeration in an appropriate jar lighted by a 60-watt incandescent lamp. 2.5, 5, 7.5, 10, 20 and 40mg/mL of air-dried sample were placed in 5mL of final concentration and 10 nauplii were transferred in each test tube. This formula was used to determine the number of dead nauplii after 24 hours.

Mortality (%) = (number of dead nauplii)/ (number of dead nauplii +number of alive nauplii) x 100

As a positive control, 45, 90, 135, 180, 225, and 270 µg/mL of potassium dichromate ($K_2Cr_2O_7$) were used. All experiment was carried out in triplicate.

Antimicrobial Activity

By using the agar well diffusion technique, the antimicrobial activity of the *Rosenvingea* extract was assessed against both gram-positive and gram-negative bacteria (Okeke*et al.*, 2001; Beuria, Santra and Panda, 2005; Devi *et al.*, 2011; Sathish Kumar and KokatiVenkataBhaskara, 2012; Omar, Al-Judaibiand and El-Gendy, 2018; Jagri, 2019; Ahsan, Islam and Hossain, 2020; Alim*et al.*, 2021; Rahman*et al.*, 2021). Incubation of bacterial strains in LB broth at 37°C for 18 hours was done in an incubator (Model – JSGI - 050T, JSR, Korea). Following incubation, all strains were inoculated with medium temperatures below 45°C and 70µl of methanolic extract of varying concentrations were placed into the appropriate wells. As a positive control, commercial antibiotic disc containing Tetracycline-30, Kanamycin-30, Ciprofloxacin-5, and Ampicillin-10 obtained from Bio Maxima S.A, Lublin, Poland were used and sample solvent (70% Methanol) was used as a negative control. The extracts were left to diffuse for an hour in petri plates with closed lids. The petri dishes were incubated overnight at 37°C or for 18 hours, after which the zone of inhibition surrounding the well was measured with a ruler and recorded for further computation. All tests were performed in triplicate.

Determination of Minimal Inhibitory Concentration (MIC)

The commercially available antibiotic discs Tetracycline 30, Kanamycin 30, Ciprofloxacin 5, Ampicillin 10 were used as a positive control, and 3x, 2x, and 1x dilution of 70% Methanol were used as a negative control. Sample concentration of 25, 50 and 100mg/mL were administrated to a volume of 70µL. The minimal inhibitory concentration was determined by the agar well diffusion method. The MIC was determined using the lowest concentration of *Rosenvingea* methanol extract and compared to a commercial antibiotic disc. All tests were performed in triplicate.

Statistical Analysis

In excel using ANOVA test, two samples tTEST, the half maximal inhibitory concentration IC_{50} calculated by linear regression, Lethal concentration 50 (LC_{50}) was calculated by Probit analysis. Mean ± SD was used for triplicate data and p values <0.05 for significance.

Results and Discussion

Phytochemical screening

The preliminary step to evaluate the existence of bioactive chemicals is phytochemical screening. Qualitative and quantitative variations in the presence of bioactive compounds in the same species have been reported, which may be caused by the use of different solvents, storage conditions, and ultimately biodiversity. In this study, an attempt has been made to compare the phytochemical screening of these seaweeds with previously published research (Ahsan, Islam and Hossain, 2020; Islam *et al.*, 2020; Alim*et al.*, 2021; Rahman*et al.*, 2021). Only 70% Methanolic solvents were used in a phytochemical investigation. The presence of Alkaloids, Steroids, Glycosides, Flavonoids, and Tannins was discovered in a 4-week macerated sample. (Table 1).

Table 1.Qualitative test of the phytochemical of *Rosenvingeasp.*on 70%nMethanol, (-): not detectable, (+): low quantities, (++): moderate quantities, (+++): high quantities.

No	Sample	Steroids	Glycosides	Flavonoid	Alkaloids	Tannins	Phenolic	Saponins
1	70% MeoH	+	+++	+	++	++	–	–

UV-Vis Spectrum

For the initial detection, 50ppm was used for the UV Visible Spectral analysis of a 70% methanol extract of *Rosenvingea* chosen from 1100 nm to 190 nm wavelengths by their wavelength and absorbance. From 190 nm to 1100 nm, the absorbance profile shows the presence of several phytochemicals at various wavelengths. *Rosenvingea* showed peak at 197.00 nm with the absorbance 0.261 which is the similar absorption spectra of flavonoids and their derivatives.(Rajeswari and Jeyaprakash, 2019; Alim*et al.*, 2021). So it can be said that *Rosenvingea* extract's has high flavonoid content.

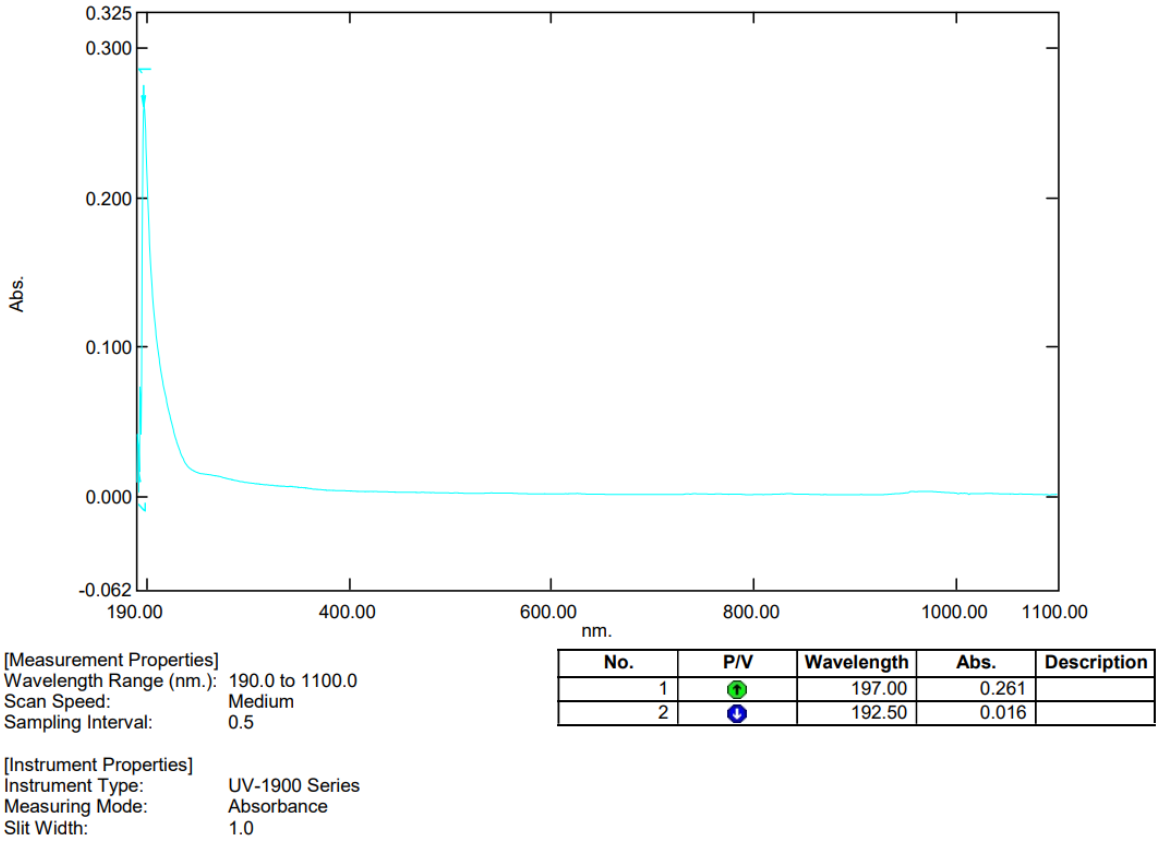
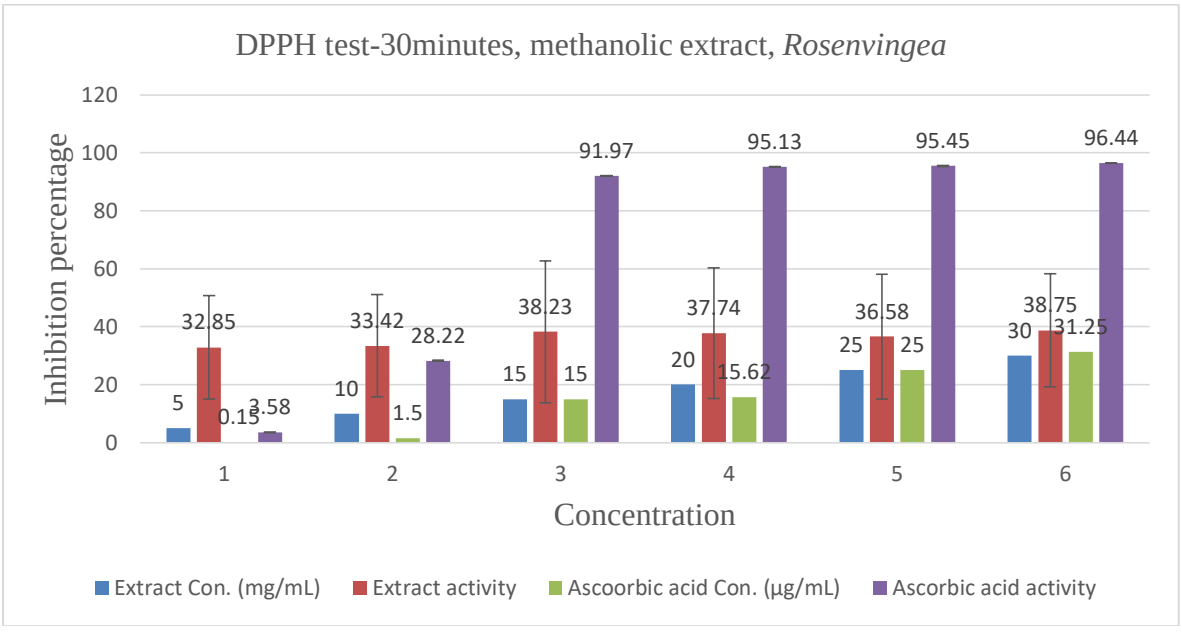


Figure 1. In a dual-beam spectrophotometer, the wavelength and absorbance of a 70% MeOH *Rosenvingea* extract were chosen from 1100nm to 190nm.

Antioxidants, DPPH Scavenging Activity

Numerous free radicals are produced in the human body as a result of normal essential metabolic processes; they are countered by free radical scavengers and antioxidants as part of homeostasis. (Alam, 2019; Alim*et al.*, 2021). It is commonly known for ages that seaweeds are high in antioxidants. (Cahyana, Shuto and Kinoshita, 1992). We performed a DPPH radical scavenging experiment to determine the potential involvement of this examined seaweed (Figure 2). In 30 minutes of 70% methanol extract at the concentration of 30mg/mL, the maximum scavenging activity of 38.75±19.47% inhibition was observed. The IC50 value was calculated in Excel using linear regression of extract and t-Test with type 1 and 2 tailed tests.



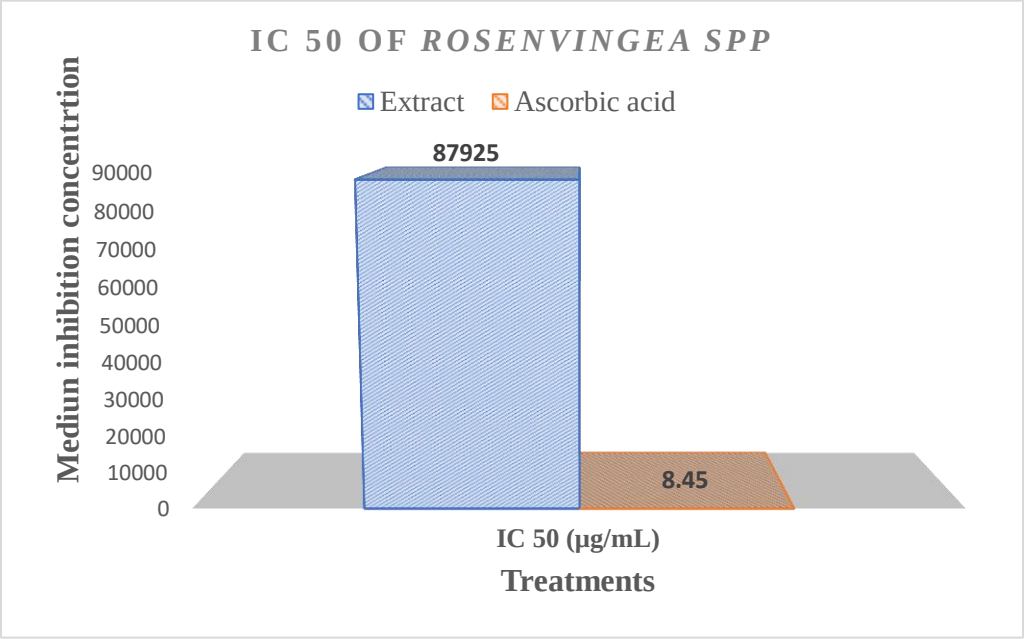


Figure 2. The antioxidant activity of a 70% methanolic extract of *Rosenvingea* was determined using the DPPH test.The percentage of inhibition was estimated using ascorbic acid as a positive control and IC-50.

We validated the presence of tannin and alkaloids in our phytochemical screening analysis, which would explain for the antioxidant activity based on the prior findings (Osawa, 1994; Khandare, 2012; Alim*et al.*, 2021). With an IC50 of 87925 µg/mL at 30 minutes and ascorbic acid at 8.45 µg/mL, it may be inferred that the methanolic extract contains less antioxidant activity than the positive control.

Cytotoxicity Assay:

The death rate was found to be 86.665.77% in a 70% methanol extract at a concentration of 40mg/mL. The median lethal concentration (LC50) in 70% methanol was 13260µg/mL, while the positive control ($K_2Cr_2O_7$) was 59.97µg/mL. The Probit regression analysed by Finney's chart and was employed in the LC50 and T-Test in Excel, using type 1 and 2 tail tests. (Figure 3)

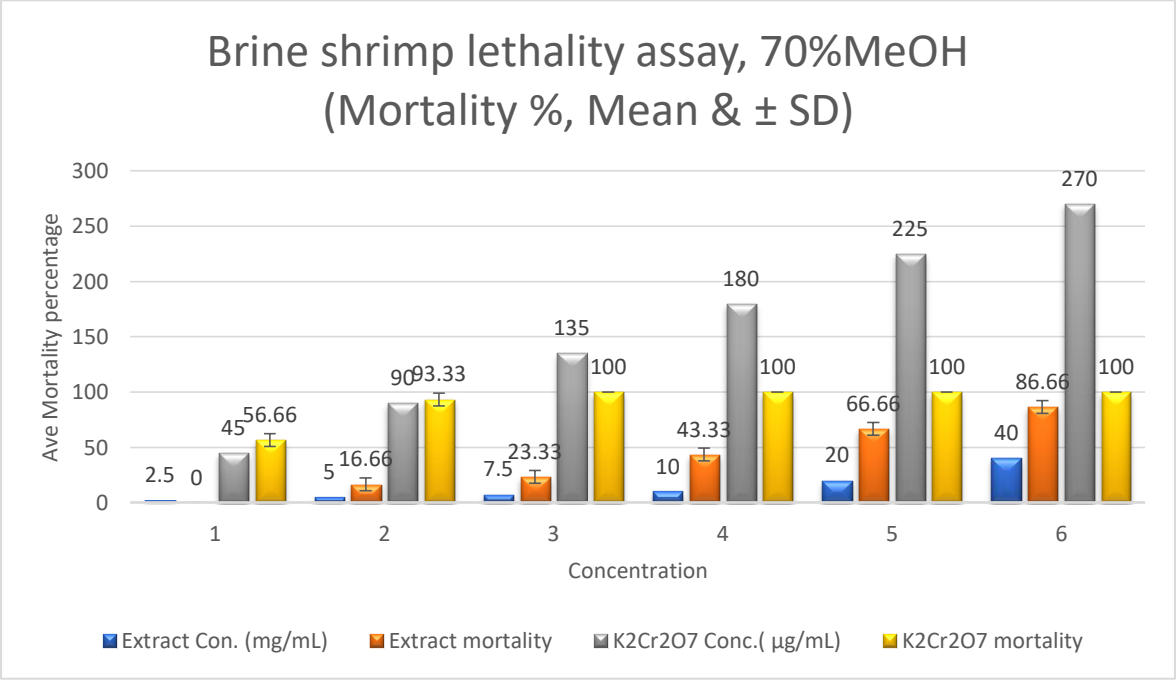


Figure 3. Brine shrimp lethality assay, **-A**. 70%Methanol extract performed percent of mortality with conc. mean &±SD, **-B.** Potassium Dichromate (K2Cr2O7) performed percent of mortality as a positive control with conc. mean &±SD, and a two-tailed t-test was performed. **-C**. Showed the values of Median Lethal concentration (LC50) between methanol & K2Cr2O7.No mortality observed in the negative control.

The toxicity level is defined as index 0-100, 100- 500 and 500-1000µg/mL as highly toxic, medium toxic and low toxic respectively in Clarkson's toxicity index (Clarkson *et al.*, 2004; Alim *et al.*, 2021) and median lethal concentration <1000µg/mL are toxic in Meyer's toxicity index (Meyer, Ferrigni and Putnam, 1982; Alim*et al.*, 2021). So, the methanolic extract has non toxic property compared to the positive control potassium dichromate (Table 2).

Table 2. Toxicity index based on Meyer's and Clarkson's Index where 70%MeOH is non-toxic level and potassium dichromate is toxic & highly toxic level.

No	Samples	LC 50	Toxicity Index	
			Meyer's Index	Clarkson's Index
			(<1000µg/mL)- Toxic	(0-100µg/mL)- Highly Toxic
				(100-500µg/mL)- Medium Toxic
				(500-1000µg/mL)- Low Toxic
				(1000µg/mL-above)- Non-Toxic
1	70%MeOH	13.26 mg/mL	Non-Toxic	Non-Toxic
2	Potassium Dichromate	59.97µg/mL	Toxic	Highly Toxic

Antibacterial activity

Methanolic extract was tested in vitro against three gram-positive and four gram-negative bacteria. The highest zone of inhibition was seen in one gram-positive and three gram-negative bacteria. *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Salmonella*, and *Klebsiella pneumonia* were inhibited with zones of inhibition of 6.66±1.15 mm, 5.66±0.57 mm, 3.33±0.57 mm & 5.33±0.57 mm respectively, while commercial ampicillin was resistant in gram negative bacteria, and Tetracycline, Ampicillin, Ciprofloxacin, and Kanamycin were utilized as positive controls, with sample solvent methanol serving as a negative control. All tests were carried out in triplicate (Table 3 and Figure 4).

Table 3. Zone of inhibition using different concentration of a 70% MeOH extract of *Rosenvingea*.

No:	Indicator Strain	Concentration mg/ml	Zone of Inhibition using sample (70% MeOH extract of *Rosenvingea)*	(+) Ve control	
			70%MeOH (Mean±SD)	Commercial disc	Zone of inhibition (Mean ±SD)

1	*Staphylococcus aureus*	2.5	-	Tetracycline (30)	21±1mm
		5	2.33±0.57mm	Ampicillin (10)	2.66±0.57mm
		10	6.66±1.15mm	Ciprofloxacin (5)	22±1mm
				Kanamycin (30)	15.66±0.57mm
2	*Bacillus cereus*	2.5	-	Tetracycline (30)	7±1mm
		5	-	Ampicillin (10)	Resistance
		10	-	Ciprofloxacin (5)	19.33±0.57mm
				Kanamycin (30)	15±1mm
3	*Staphylococcus hominies*	2.5	-	Tetracycline (30)	21±1mm
		5	-	Ampicillin (10)	3.66±0.57mm
		10	-	Ciprofloxacin (5)	22±1mm
				Kanamycin (30)	18.66±1.15mm
4	*Pseudomonas aeruginosa*	2.5	-	Tetracycline (30)	13.33±0.57mm
		5	-	Ampicillin (10)	Resistance
		10	5.66±0.57mm	Ciprofloxacin (5)	31±1mm
				Kanamycin (30)	28.66±1.15mm
5	*Salmonella*	2.5	-	Tetracycline (30)	2.33±0.57mm
		5	2±0mm	Ampicillin (10)	Resistance
		10	3.33±0.57mm	Ciprofloxacin (5)	4.66±0.57mm
				Kanamycin (30)	17.33±1.15mm
6	*Salmonella typhi*	2.5	-	Tetracycline (30)	20±1mm
		5	-	Ampicillin (10)	14.66±1.15mm
		10	-	Ciprofloxacin (5)	14.33±0.57mm
				Kanamycin (30)	17.33±0.57mm
7	*Klebsiella pneumonia*	2.5	-	Tetracycline (30)	7.33±0.57mm
		5	1.66±0.57mm	Ampicillin (10)	Resistance
		10	5.33±0.57mm	Ciprofloxacin (5)	11.33±1.15mm
				Kanamycin (30)	10.66±1.15mm

Each value is presented as mean ±SD, (-) = no zone of inhibition

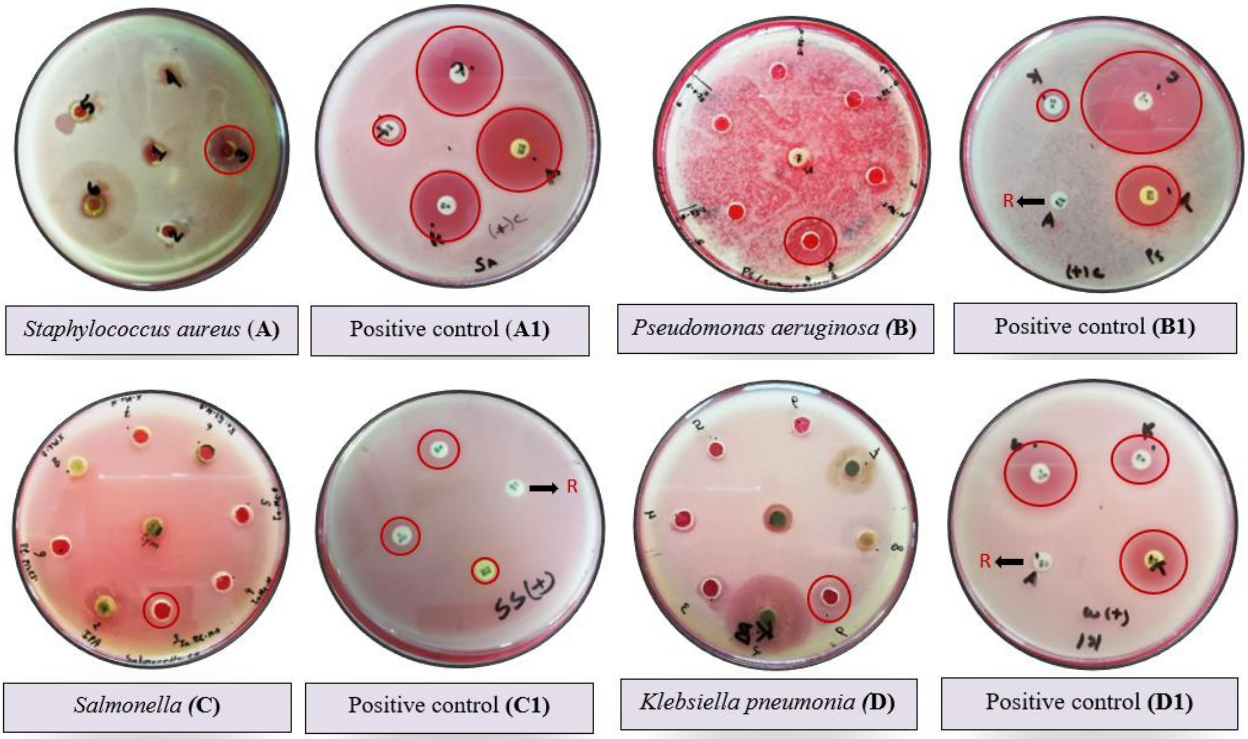


Figure 4.A. *Staphylococcus aureus*was showed clear zone of inhibition in **70%** Methanolic extract and positive control **A1**(Ampicillin, Kanamycin, Ciprofloxacin, Tetracycline) showed a clear zone of inhibition. **B.** *Pseudomonas aeruginosa*wherethe positive control Ampicillin was resistant and Ciprofloxacin showed a long-range zone of inhibition, fewer zone of Kanamycine in **B1**. **C.** *Salmonella* performed in Methanolic extract where the positive control showed fewer zone of inhibition; Ampicillin was resistantin**C1**. **D**. *Klebsiella pneumonia* performed in Methanolic extract where the positive control showed a medium-range zone of inhibition (Ampicillin) was resistant in figure **D1.**

As a result of the comparison investigation, it is clear that gram-negative bacteria were more susceptible to the methanolic extract than gram-positive bacteria, suggesting that it is a promising candidate for future antimicrobial research. Based on our phytochemical screening and earlier study, we may conclude that the presence of alkaloids is responsible for the antimicrobial action, which may disrupt bacterial Fts Z-Z ring formation and hinder bacterial cytokinesis, resulting in bactericidal activity.(Beuria, Santra and Panda, 2005; Alim*et al.*, 2021).

Temperature and the effectiveness of antimicrobials

To determine the impact of temperature optimization on *Klebsiella pneumonia*, the *Rosenvingea*70% Methanol extract was pre-incubated in the water bath for 30 minutes at a variety of temperatures, including 30°C, 60°C and 90°C. When treated at 60°C for 30 minutes with a commercial antibiotic disc used as a positive control, this extract demonstrated stability. However, at 90°C, the extract loss the effectiveness for antibacterial activity.(Table:04)

Table 4.

70% methanolic extract		
No	Temperature	Antimicrobial Potency
01	30	+
02	60	+
03	90	_

Minimum Inhibitory Concentration (MIC)

Rosenvingea methanolic extract MIC (Minimum Inhibitory Concentration) was shown to be larger than standard resistance antibiotic disc with *B. cereus, P. aeruginosa, Salmonella, K. pneumonia.* When the MIC showed the zone with the lowest concentration, the antimicrobial agent is usually more bacteriostatic. As a result, *Rosenvingea* methanol extract is likely more bacteriostatic than standard resistance antibiotics disc in varied situations. (Table-05)

Table 5. MIC (Minimum Inhibitory Concentration) compared with the commercial antibiotic disc.

70% Methanol extract of *Rosenvingea* (MIC)		
No	Indicator Strain	MIC according zone of inhibition
01	*Staphylococcus aureus*	5mg/mL
02	*Bacillus cereus*	Nill
03	*Staphylococcus hominies*	Nill
04	*Pseudomonas aeruginosa*	10mg/mL
05	*Salmonella*	5mg/mL
06	*Salmonella typhi*	Nill
07	*Klebsiella pneumonia*	5mg/mL

Conclusion

The phytochemicals contained in *Rosenvingea* brown seaweeds, such as steroids, glycosides, flavonoids, alkaloids, and tannins, were investigated in this study. Flavonoids, a group of natural substances with variable phenolic structures, are well known for their various health benefit. The high flavonoids containing *Rosenvingea* extract can be used as dietary antioxidant and anti-inflammatory supplements. Methanolic extract has lower antioxidant activity than ascorbic acid. *Rosenvingea*, a brown seaweed, also proved non-cytotoxic when compared to the positive control ($K_2Cr_2O_7$). Because of that, consuming and using it for different purpose may be safe. This extract also showed considerable antibacterial activity against both gram-positive and gram-negative pathogenic bacteria. The extract is thermotolerant and stays active in high temperature making it a promising candidate for next generation antibiotic and drug development. The data obtained from this study regarding the in vitro effects of *Rosenvingea* extract are promising and highlight their potentiality for commercial applications in the future.

Author contributions

MNH made the hypothesis, designed the experiments, supervised the work, analyzed the data, wrote and revised the final version of manuscript; MAA conducted the experiments, analyzed the data and wrote the initial draft of manuscript; NRZ did manuscript editing.

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