Sahel Journal of Life Sciences FUDMA (SAJOLS)
March 2026 Vol. 4(1): 348-359
ISSN: 3027-0456 (Print)
ISSN: 1595-5915 (Online)
DOI: https://doi.org/10.33003/sajols-2026-0401-40

Research Article

Phytochemical Profiling, Antioxidant Potentials, and Antimicrobial Properties of Locally-Isolated *Chlorococcum* and *Scenedesmus* Species: Sustainable Sources of Bioactive Compounds

***Samuel S. A.[1], Yisa A. G.[1], Ibrahim Z. A.[2], Omada S.[1], and Naibi H. A.[1]**

[1]Department of Biological Sciences, National Open University of Nigeria
[2]Department of Zoology, University of Ilorin, Nigeria
****Corresponding Author's email*: chatwitsadisam@gmail.com; Phone: +2348068470115**

ABSTRACT
Microalgae thrive in diverse habitats and possess adaptive mechanisms that enable them to produce unique bioactive compounds. These compounds have gained increasing attention due to their potential applications in the food, medicinal, pharmaceutical, nutraceutical, and cosmetic industries. Despite the growing interest in microalgal bioactive compounds global research on locally-isolated species in Nigeria remains limited. This study evaluated the phytochemical composition, antioxidant potential, and antimicrobial properties of *Chlorococcum* sp and *Scenedesmus* sp ethanol extracts. Phytochemical analysis confirmed the presence of alkaloids, flavonoids, saponins, steroids, and phenolic compounds, which exhibit various biological activities. Antioxidant assays revealed concentration-dependent free radical scavenging activities, with the highest DPPH inhibition percentages recorded at 43.1% for *Chlorococcum* sp and 47.58% for *Scenedesmus* sp at 1 mg/mL. Reducing power assays confirmed significant antioxidant activity, with absorbance values of 0.52 mg AAE/g DW for *Chlorococcum* sp and 0.56 mg AAE/g DW for *Scenedesmus* sp. The reducing power of ethanol extracts also correlated significantly with inhibition capacity, indicating strong antioxidant potential. The antimicrobial evaluation demonstrated the inhibitory effects of ethanol extracts against *Escherichia coli* and *Staphylococcus aureus*, with inhibition zones of 26.0 mm for *Scenedesmus* sp and 20.0 mm for *Chlorococcum* sp at 500 mg/mL. The MIC values of 125 mg/mL and 250 mg/mL for *Scenedesmus* sp and *Chlorococcum* sp respectively, revealed their potency as antibacterial agents. These findings highlight the potential of *Chlorococcum* sp and *Scenedesmus* sp as valuable natural sources of bioactive compounds for pharmaceutical and nutraceutical applications.

Keywords: Antimicrobial; Antioxidant; *Chlorococcum* sp; *Scenedesmus* sp; Phytochemical

Citation: Samuel, S.A., Yisa, A.G., Ibrahim, Z.A., Omada, S., & Naibi, H.A. (2026). Phytochemical Profiling, Antioxidant Potentials, and Antimicrobial Properties of Locally-Isolated *Chlorococcum* and *Scenedesmus* Species: Sustainable Sources of Bioactive Compounds. *Sahel Journal of Life Sciences FUDMA*, 4(1): 348-359. DOI: https://doi.org/10.33003/sajols-2026-0401-40

INTRODUCTION

Microalgae exist in various habitats such as terrestrial, freshwater, marine, brackish and wastewater environments in Nigeria (Salaam *et al*., 2019; Abdel-Karim *et al*., 2020). They can survive in a robust environment due to their adaptive survival strategy, such as the production of distinctive bioactive compounds. In recent years, bioactive compounds produced by microalgae have been very useful in the food, medicinal, pharmaceutical, nutraceutical, cosmetic, aquaculture and bioenergy industries (Shaima *et al*., 2016; Zabed *et al*., 2020; Shaima *et al*., 2022). Currently, the economically feasible production of high-value bioactive compounds has become a significant area in algae biotechnological research (Russell *et al*.,2022; Leon-

Vaz *et al*., 2023). Several microalgae, particularly the chlorophytes are greatly diverse and contain variety of bioactive products like phenolic, carotenoids, polyunsaturated fatty acids, and amino acids (Elshobary *et al*., 2020; Singh *et al*., 2020). Numerous studies on microalgae have shown their potential for antioxidant and antimicrobial activity for example *Oscillatoria* sp (Seddek *et al*., 2019; Nainangu *et al*., 2020) *Chlorella* sp (Shaima *et al*., 2022) and *Chlorococcum* sp (Shaima *et al*., 2022). These bioactive compounds possess a wide range of biological activities such as antioxidant, antimicrobial, antiviral, anti-inflammatory, antiprotozoal and anticancer (Gogineni and Hamann, 2018; Marrez *et al*., 2019). In Nigeria, the bioactive compounds in microalgae are not well studied (Salaam *et al*., 2019; Adeniyi-Martins *et al*., 2023). More so, knowledge about locally isolated microalgae bioactive compounds and their antioxidant and antimicrobial activities is scarce. Antioxidants play an important role in inhibiting and scavenging radicals, therefore protecting humans and animals against infections and degenerative diseases (Seddek *et al*., 2019). The restriction of synthetic antioxidants (butylated hydroxyl anisole (BHA) and butylated hydroxyl toluene (BHT) due to toxicity, induced DNA damage and increased antibiotic resistance indicate an urgent need for newly isolated antioxidant antibacterial bioactive compounds (Yap *et al*., 2019; Shaima *et al*., 2022). This study aimed to explore the bioactive compounds, antioxidant and antimicrobial properties of locally isolated *Scenedesmus* sp and *Chlorococcum* sp for subsequent application in medicinal and pharmaceutical industries.

MATERIALS AND METHODS

Microalgae species and and culture

Axenic cultures of *Chlorococcum* sp and *Scenedesmus* sp were sourced from the Phycology Laboratory at the Department of Botany, Ahmadu Bello University, Zaria. Identification of species was done using morphological and taxonomical standard methods according to (van Vuuren *et al*., 2006). Furthermore, experimental cultures of *Chlorococcum* sp and *Scenedesmus* sp were sustained in BG11 medium at a pH of 7.4. These cultures were kept under-regulated laboratory conditions, including a light intensity of 40 µmol m^{-2} s^{-1}, a photoperiod of a 16:8hour light-to-dark cycle, and a temperature maintained at 25.0 ± 2 °C. To maintain sterility, the BG11 medium was autoclaved at 121 °C for 30 minutes. The organisms were acclimatised through multiple culture transfers during their exponential growth stage to ensure accurate physiological outcomes. The biomasses of algal cultures were harvested at the end of exponential growth phase by centrifugation at 5000 rpm for 15 mins. The Cell pellets were rinsed three times and resuspended in sterilized distilled water to remove traces of growth medium followed by centrifugation at 5000 rpm for 15 mins after each wash. The collected biomass of *Chlorococcum* sp and *Scenedesmus* sp was then allowed to dried in the laboratory.

Measurement of Growth rate

The absorbance of the cultures was read at 750 nm with a VIS spectrophotometer (Spectrumlab 752s) at regular interval of 2 days to monitor their growth. The specific growth rate (K) of *Chlorococcum* sp and *Scenedesmus* sp was calculated according to Liang *et al*. (2013), using the following formula:

$$K = \frac{InN_2 - InN_1}{T_2 - T_1}$$

N_1 and N_2 are the average cell densities of *Chlorococcum* sp and *Scenedesmus* sp at T_1 and T_2, respectively.

Microalgae extract preparation

Fifty grams (50 gm) each of *Chlorococcum* sp and *Scenedesmus* sp was weighed into two separate 5000ml Erlermeyer flasks and 400ml of ethanol (solvent) was added into each container. The content was well mixed and kept in bioshaker for 24 hours. After 24 hours, the content in each flask was filtered using Whattman no.1 filter paper and then concentrated by evaporation in the water bath at 40 ^{0}C.

Screening of bioactive compounds

The ethanol extracts of the *Chlorococcum* sp and *Scenedesmus* sp were subjected to phytochemical analysis described by Andima *et al*. (2014) used for chemical identification of different phytochemical components.

Tests for tannins, Phenolic and Flavonoids

Tannins was determined by using ferric chloride test where 5 ml solution of the extract was taken in a test tube. Then 1 ml of 5% ferric chloride solution was added. Greenish-black precipitate was formed and indicated the presence of tannins. On addition of 5%

$FeCl_3$ solution to the extract, deep blue-black colour appeared to confirm the presence of phenolic. For flavonoids three drops of sodium hydroxide was added to 2 mL of extract. A deep yellow colour appeared, which gradually become colourless when a few drops of dilute HCl was added.

Test for Saponins, Steroids and Alkaloids

One milliliter (1 ml) of extract was diluted with distilled water to 20 ml and shaken for 15 minutes, forming a one-centimeter foam layer indicating saponins. Steroids were identified using a sulfuric acid test: 1 ml of chloroform extract mixed with 1 ml of sulfuric acid produced a red color. For alkaloids, Wagner's test involved mixing 2 ml of the extract with 0.2 ml of dilute hydrochloric acid, followed by 1 ml of iodine solution, resulting in a reddish-brown precipitate that confirmed their presence.

Determination of total phenolic content (TPC)

TPC of the microalgae extracts was determined using the Folin–Ciocalteu method, as modified by Kumar *et al*. (2008). Briefly, 0.1 mL of each extract (10 mg mL-1) was mixed with 0.6 mL of distilled water and 0.5 mL of Folin–Ciocalteu reagent, followed by 1.5 mL of sodium carbonate (20% in distilled water). Final volume was made up to 10 mL with distilled water. The mixtures were incubated for 30 min at room temperature in the dark. Absorbance was measured at 760 nm. TPC was expressed in mg GAE g-1 dry weight (dw) using a GA calibration curve.

Antioxidant Activity

DPPH Free radical scavenging assay

The DPPH free radical scavenging was measured using a modified method from Sultana *et al*. (2008). A 5.0 ml solution of 0.025 g/l DPPH in ethanol was mixed with 1.0 ml of extract at 25 g/ml dry matter. The mixture was shaken and left in the dark at room temperature for 30 minutes. Absorbance was measured at 515 nm using a UV-visible spectrophotometer (Shimadzu UV-1601PC). The percentage of DPPH scavenging for each test sample concentration was calculated using a specific formula.

$$\% \text{ DPPH} = (\text{Abs control} - \text{Abs sample})/ (\text{Abs control}) \text{ x } 100$$

Where Abs control is the absorbance of DPPH solution without extracts

Reducing power assay

The reducing power of crude extracts from *Chlorococcum* and *Scenedesmus* species was assessed using Sultana *et al*. (2008). Crude extracts (5 ml) were combined with 0.2 M phosphate buffer (pH 6.6) and 1% potassium ferricyanide, incubated at 50°C for 20 min. After adding 1% trichloroacetic acid (TCA), the mixture was centrifuged at 3000 rpm for 10 min. The supernatant was mixed with distilled water and 0.1% FeCl3 (1:1:2 ratio), and absorbance was measured at 700 nm. Higher absorbance indicated greater reducing power, expressed as mg ascorbic acid/g dry weight (mg AAE/g DW), with ascorbic acid as the standard, and the test performed in triplicate.

Antioxidant activity of crude extracts of the *Chlorococcum* sp and *Scenedesmus* sp

The ferric thiocyanate (FTC) method was adapted from Sultana *et al*. (2008) to assess the antioxidant activity of microalgae extracts from *Chlorococcum* and *Scenedesmus* species Each 2.5 ml extract was combined with 2.5 ml of 95% ethanol, 4.1 ml of linoleic acid (2.51% V/V in ethanol), 8 ml of 0.05 M phosphate buffer (pH 7.0), and 3.9 ml of distilled water. This mixture was stored in the dark at 4 °C. Afterward, 0.1 ml of the solution was mixed with 9.7 ml of 75% ethanol and 0.1 ml of 30% ammonium thiocyanate, followed by 0.1 ml of 20 mM ferrous chloride in hydrochloric acid. The absorbance of the resulting red solution was measured at 500 nm after 3 minutes and subsequently at 24-hour intervals until the control reached maximum absorbance. The percentage inhibition of linoleic acid peroxidation was then calculated.:

$$\% \, \boldsymbol{Inhibition} = \mathbf{100} - \frac{\boldsymbol{Absorbance\ increase\ of\ sample\ x\ 100}}{\boldsymbol{Absorbance\ increase\ of\ blank}}$$

Antimicrobial activity assay

The antibacterial evaluation was performed using the well and agar disc diffusion methods according to Alsenani *et al*. (2017). Antibacterial activity of the ethanolic extract of the *Chlorococcum* sp and *Scenedesmus* sp was evaluated by the cup plate agar diffusion method (Alsenani *et al*., 2017). The plates were incubated at 37^0C for 24 hours. A well was made in each of the culture plates and filled with 20 µl of 50 µg/ml of chloramphenicol as control. Antimicrobial activity was determined by measuring the zone of inhibition around each well (excluding the diameter of the well). All the experiments were carried out in triplicate.

Determination of Minimum Inhibitory Concentration (MIC)

The antimicrobial activity of *Chlorococcum* sp and *Scenedesmus* sp were further tested using the

minimum inhibitory concentration (MIC). The Minimum Inhibitory Concentration of the extracts was determined for the microalgae extracts against the test organisms (*Staphylococcus aureus* and *Escherichia coli*) using the broth dilution technique. (Patra *et al*., 2009). All the experiments were carried out in triplicates.

Determination of Minimum Bactericidal Concentration (MBC)

To determine the MBC, for each set of test tubes in the MIC determination, a loopful of broth was collected from those tubes that did not show any bacterial growth in the MIC and inoculated on sterile Nutrient agar by streaking. All the plates were then incubated at 37^{O}C for 24h. After the incubation period, the concentration at which no visible growth occurred on the plate was noted as the Minimum Bactericidal Concentration (MBC) (Patra *et al*., 2009). All the experiments were carried out in triplicates.

Data Analysis

All experiments were conducted in triplicate and the results were expressed as mean ±standard deviation (SD). The means of all the parameters were examined for significance using one and two-way analysis of variance (ANOVA). Pearson correlation test was used to study the correlation between % inhibition and ferric-reducing power activity. Significant differences were considered at $p<0.05$. All data were analysed using R software version 4.4.4.

RESULTS

The growth rate of *Chlorococcum* sp and *Scenedesmus* sp

The microalgae (*Chlorococcum* sp and *Scenedesmus* sp) biomass grown in BG 11 medium was determined as optical density at 750 nm (Figure.1). The stationary phase of *Chlorococcum* sp. and *Scenedesmus* sp was stated on day 24 of the incubation. Cultures were allowed to grow for 27 days (Figure 2).

Bioactive compounds of *Chlorococcum* sp and *Scenedesmus* sp

The phytochemical alkaloids, flavonoids, saponins, steroids and phenol were detected in*Chlorococcum* sp and *Scenedesmus* sp ethanol extract. However, tannins bioactive metabolites were not observed in *Chlorococcum* sp and *Scenedesmus* sp extract (Table.1).

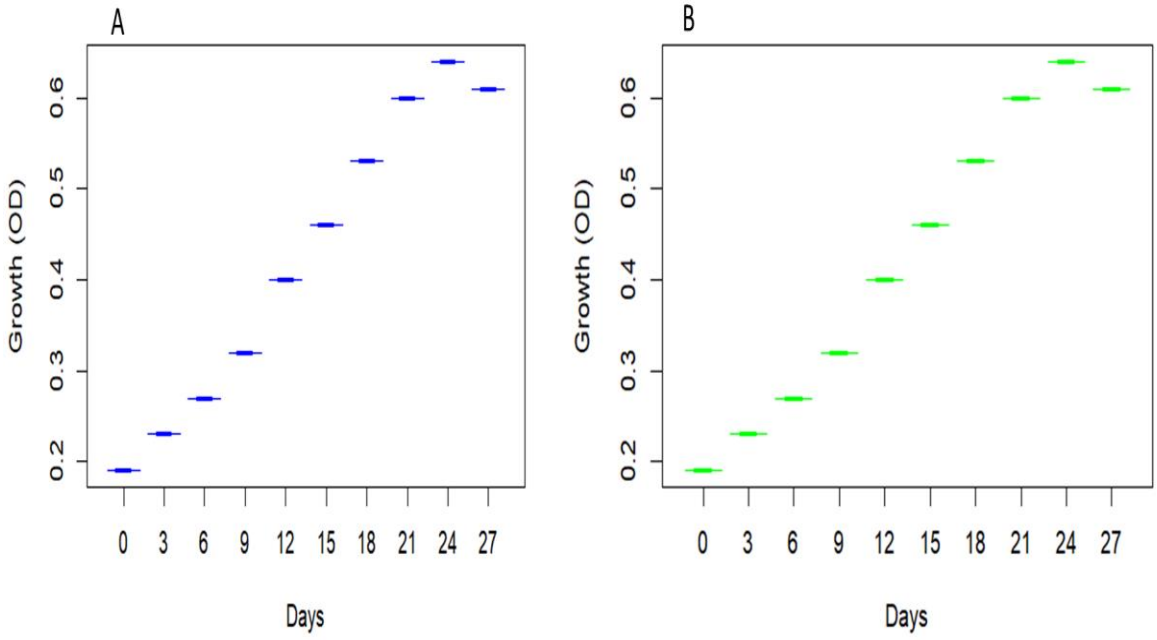


Figure 1: Growth curve of A) *Chlorococcum* sp and B) *Scenedesmus* sp grown in BG 11 medium

Table 1: Qualitative phytochemicals analysis of ethanol extract of *Chlorococcum* sp and *Scenedesmus* sp

Phytochemicals	*Chlorococcum sp*	*Scenedesmus sp*
Alkaloids	+	+
Flavonoids	+	+
Tannins	-	–
Saponins	+	+
Steroids	+	+
Phenol	+	+

Keys: + = Present, - = Absent

Diphenyl-1-picrylhydrazyl (DPPH) scavenging radical activity

The DPPH in vitro assays of *Chlorococcum* sp and *Scenedesmus* sp extract at four different concentrations compared with an ascorbic acid standard showed the free radical scavenging activity. The *Chlorococcum* and *Scenedesmus* species showed the highest scavenging activity ($p<0.05$), with an inhibition percentage of 43.1% and 47.58%, respectively, at a 1 mg/ml concentration. Additionally, 0.7mg/mL concentration of *Chlorococcum* sp and *Scenedesmus* sp extract showed significant ($p<0.05$) inhibition of 35.42% and 39.85% respectively (Figures 3 and 4).

Reducing power and total antioxidant capacity

The results in Figure 5 and 6 revealed that 1 mg/ml ethanol extract of *Scendesmus* sp and *Chlorococcum* sp showed the highest reducing power capacity of 0.56 and 0.52 mg AAE/g DW respectively while 0.1 mg/ml concentration recorded the lowest activity values of 0.31 and 0.32 mg AAE/g DW. The microalgae *Scenedesmus* sp and *Chlorococcum* sp showed a concentration-dependent reducing power within a concentration range of 0.1, 0.3, 0.5, 0.7 and 1.0 mg/ml. *Scenedesmus* sp recorded the highest reducing power than the *Chlorococcum* sp (Figure 6).

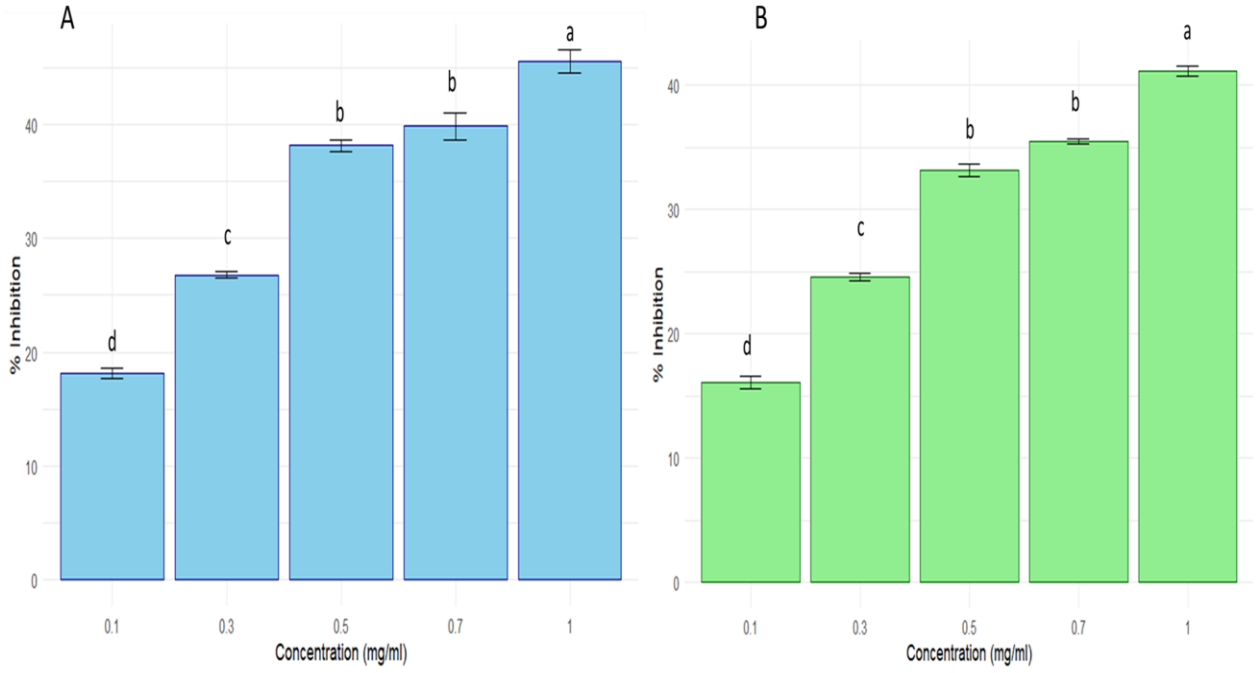


Figure 2: DPPH (1, 1-diphenyl- 2- picryhydrazyl) radical scavenging activity of A) *Scendesmus* sp and B) *Chlorococcum* sp at different extract concentrations

Error bars represent standard deviations based on a sample size of 3 replicates. Bars having different alphabets are significantly different ($P < 0.05$).

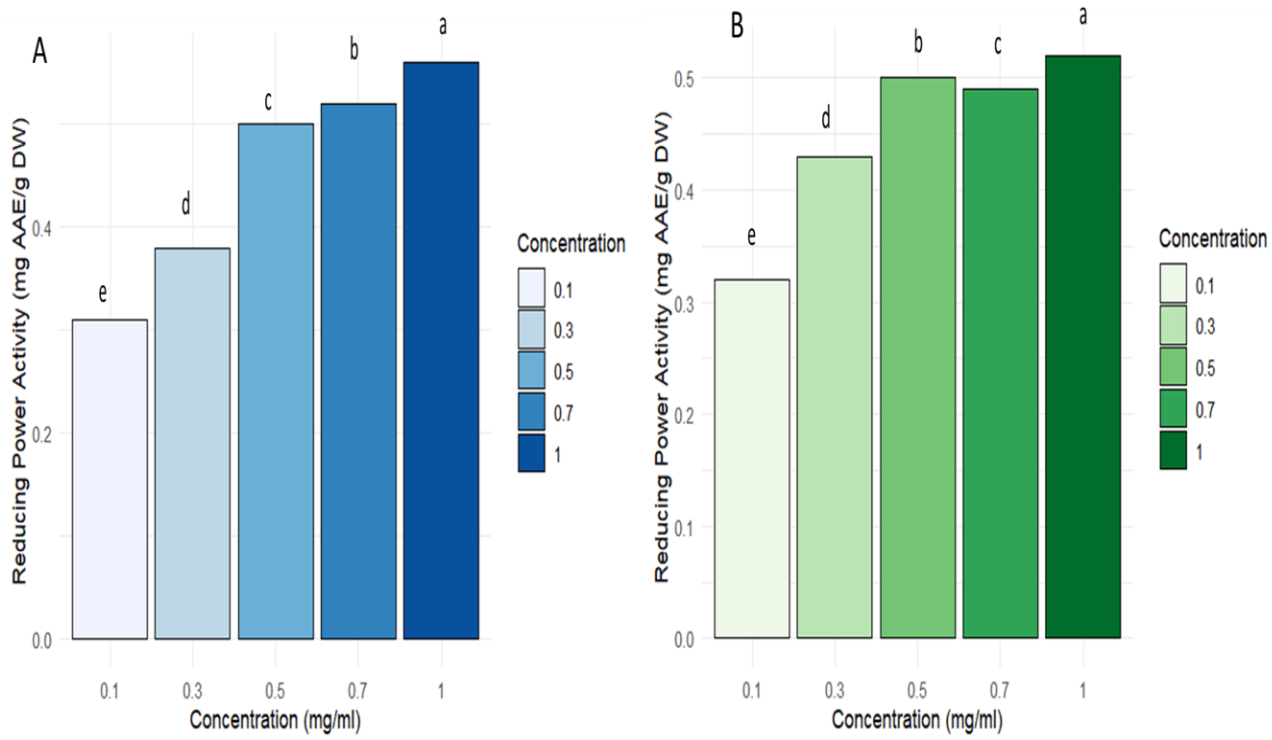


Figure 3: Reducing power activity of the ethanol extract of A) *Scenedesmus* sp and B) *Chlorococcum* sp at different concentrations

Bars having different alphabets are significantly different (P < 0.05)

Antimicrobial Activity

The antimicrobial activity of the ethanol extract of *Scenedesmus* sp showed significant (p<0.05) inhibition against two different gram-negative bacterium (*Escherichia coli*) and positive bacterium (*Staphylococcus aureus).* The extract of *Scenedesmus* sp and *Chlorococcum* sp at 500 mg/ml showed the highest (p<0.05) zone inhibition diameter of 26mm and 20mm respectively against *Escherichia coli*than that of the control (chloramphenicol). However, at 500 mg/ml,the extract of *Scenedesmus* sp and *Chlorococcum* sp revealed a zone inhibition diameter of 18mm and 17mm against *Staphylococcus aureus.*The findings showed that the antibacterial activities of *Scenedesmus* sp and *Chlorococcum* sp extract increased with increasing concentrations against *Escherichia coli* and *Staphylococcus aureus* (Figures7 and 8).The antibacterial activity of *Scenedesmus* sp against *Staphylococcus aureus* and *Escherichia coli* was higher than that of *Chlorococcum* sp against the same bacteria.

Minimum inhibitory concentration

The results showed that *Scenedesmus* sp had the minimum inhibition concentration (MIC) of 125 mg/ml against *Escherichia coli* and MIC of 250mg/ml against *Staphylococcus aureus* which correlate with the control (chloramphenicol) while the extracts of *Chlorococcum* sp had a higher minimum inhibitory concentration of 250 mg/ml against both *Escherichia coli* and *Staphylococcus aureus* (Table 2).

Minimum Bactericidal concentration

The microalgae *Scenedesmus* sp had the minimum bactericidal concentration (MBC) of 250 mg/ml against *Escherichia coli* and MBC of 500mg/ml against *Staphylococcus aureus,* which correlate with the control (Chloramphenicol) while the extracts of *Chlorococcum* sp had a higher minimum bactericidal concentration against of 500 mg/ml against *Escherichia coli* and *Staphylococcus aureus* (Table 3).

Correlation heatmap

All the values in the matrix range from 0.88 to 1.00, representing strong positive correlations between all pairs of variables. *Scenedesmus* and *Chlorococcum* species show dose-dependent increases in both %

inhibition and reducing power. This correlation matrix strongly showed that increasing concentration of the algal extracts enhances both their antioxidant capacity (reducing power) and biological inhibition (% inhibition), particularly in *Scenedesmus* (Table 4).

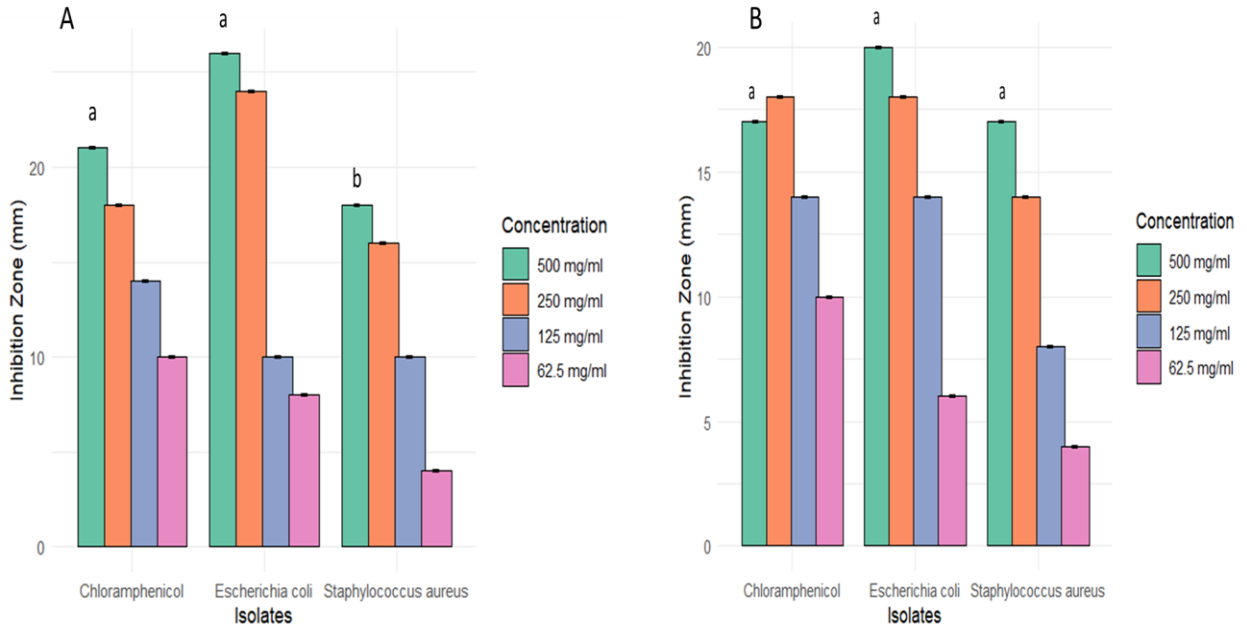


Figure 4: Antimicrobial activity diameter zone of inhibition of A) *Scenedesmus* sp and B) *Chlorococcum* sp against *Escherichia coli* and *Staphylococcus aureus* at four different concentrations

Bars represent the mean ± SD for n = 3. Bars having different alphabets are significantly different (P < 0.05).

Table 2: Minimum Inhibitory Concentration of *Scenedesmus* sp and *Chlorococcum* sp

Isolates	500 (mg/mL)	250 (mg/mL)	125 (mg/mL)	62.5 (mg/mL)
***Scenedesmus* sp**				
Staphylococcus aureus	-	-	+	+
Escherichia coli	-	-	-	+
Chloramphenicol	-	-	-	+
***Chlorococcum* sp**				
Staphylococcus aureus	-	-	+	+
Escherichia coli	-	-	+	+
Chloramphenicol	-	-	-	+

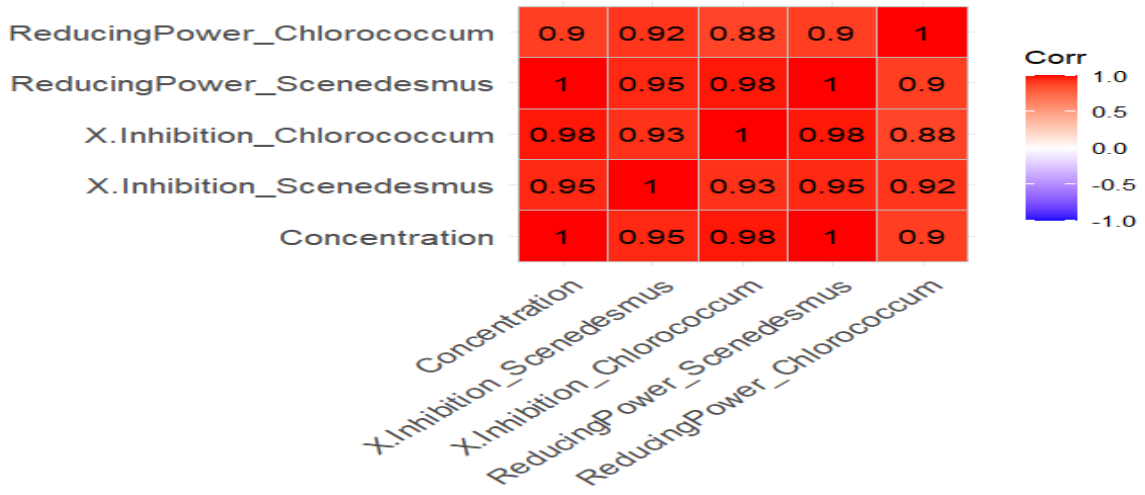
Keys+ = Growth, - = No growth

Table 3: Minimum Bactericidal Concentration of *Scenedesmus* sp and *Chlorococcum* sp

Isolates	500 (mg/mL)	250 (mg/mL)	125 (mg/mL)	62.5 (mg/mL)
***Scenedesmus* sp**				
Staphylococcus aureus	-	+	+	+
Escherichia coli	-	-	+	+
Chloramphenicol	-	-	+	+
***Chlorococcus* sp**				
Staphylococcus aureus	-	+	+	+
Escherichia coli	-	+	+	+
Chloramphenicol	-	-	+	+

Keys+ = Growth, - = No growth

Table 4: Correlation values between Concentration, % Inhibition and Reducing Power for *Scenedesmus* sp and *Chlorococcum* sp



DISCUSSION

Microalgae can produce a wide range of bioactive compounds with antimicrobial and antioxidant properties (Singh *et al*., 2020). They serve as a continuous and consistent source of natural products, because they can be cultivated in bioreactors on a large scale. The value of microalgae as a source of natural antimicrobials and antioxidants is further enhanced by the relative ease of purification of target compounds (Abdel-Karim *et al*., 2020). In this study ethanol extract of *Chlorococcum* sp and *Scenedesmus* sp revealed several phytochemical compounds such as alkaloids, flavonoids, saponins, steroids and phenol which showed a wide range of activity. Alkaloids have shown cytotoxic activity due to the presence of microtubule-interfering agents that can bind to beta-tubulin, thus inhibiting the formation of the mitotic spindle fibre required for cell division (Solanki *et al*., 2008). More so, flavonoids, steroids and phenol have antimicrobial, antioxidant, anti-inflammatory and antiviral activities. Phenolic compounds possess specific physical, chemical and biological activities that make them useful as drugs. Also, phenolic compounds act as antioxidants by chelating metal ions, preventing radical formation and improving the antioxidant endogenous system (Dai and Mumper, 2010; Singh *et al*., 2020). The antibacterial activity in phenolic compounds could be related to the capability of these substances to alter cell permeability (Alshuniaber *et al*., 2021). Also, these compounds interact with proteins and enzymes in the microbial cell membrane, resulting in disruption of cellular function or genes. Steroids on the other hand, play a vital role in nutrition, herbal medicine and cosmetics (Okwu, 2001; Abdel-Karim *et al*., 2020). Saponins are also known to have antifungal and antibacterial properties. The bioactive compounds derived from microalgae such as flavonoids, phenol, saponins and steroids etc., have received considerable attention in recent years due to their diverse pharmacological properties, including antimicrobial, antioxidant and anticancer activity (Bajpai, 2016; Shaima *et al*., 2022). Similar studies also revealed the presence of bioactive compounds such as phenol in *Chlorella* sp (Sawant and Mane, 2018; Shaima *et al*., 2022), flavonoids, alkaloids and phenols in *Chlorella vulgaris* (Abdel-Karim *et al*., 2020), palmitic and linolenic acids found in *Chlorococcum minutum* (Mostafa *et al*., 2020) and astaxanthin, produced from *Haematococcus pluvialis* is unique among carotenoids and provides a superior antioxidant defence (Shah *et al*., 2016).

Antioxidants are compounds that delay antioxidation by inhibiting the formation of free radicals or by interrupting the propagation of harmful free radicals (Singh *et al*., 2020). Antioxidant compounds can play favourable roles in food preservation and human health (Wilson *et al*., 2017). In this present study, the antioxidant capacity of *Chlorococcum* sp and *Scenedesmus* sp. showed significant ($p<0.05$) variation depending on the concentrations. *Chlorococcum* sp and *Scenedesmus* sp recorded the highest scavenging activity with an inhibition

percentage of 43.1% and 47.58%, respectively, at a 1 mg/ml concentration. This study showed that the highest reducing power was1 mg/ml ethanol extract of *Chlorococcum* sp. and *Scenedesmus* sp at 0.52 and 0.56 mg AAE/g DW, respectively. The effect of microalgae bioactive compounds acting as antioxidants on DPPH radicals was due to their potential to donate hydrogen to free radicals and reduce to non-reactive species (Davoodbasha *et al*., 2018; Elshobary *et al*., 2020). However, Jayshree *et al*. (2016) recorded the free radical scavenging ability of the acetone extract of *C. vulgaris*at 50.8% and the methanol extract of *C. vulgaris* was 92.57% at 1000 µg/ml. Similarly, Moustafa *et al*. (2020) showed that methanol extract had the highest reducing power followed by the ethanol and acetone extract of *Chlorococcum minutum*. The reducing activities of the methanol extract were mainly related to the presence of reducing aldehydes or ketones. Our study also indicates that the DPPH radical scavenging activity and reducing power of *Scendesmus* sp and *Chlorococcum* varied significantly at different extract concentrations. The correlation coefficients (R) between % inhibition and reducing power were substantially high (0.95 and 0.88, respectively). Both *Scenedesmus* and *Chlorococcum* sp exhibit increases in both % inhibition and reducing power in a dose-dependent manner, indicating that their antioxidant capabilities enhance with higher concentrations. The significant correlations between variables suggest that % inhibition and reducing power may represent similar or interconnected biological mechanisms. Based on stronger correlations, it appears that *Scenedesmus* might respond slightly better to increasing concentrations regarding antioxidant activity compared to *Chlorococcum* sp.

In this study, the ethanol extracts of *Chlorococcum* sp and *Scenedesmus* sp showed antimicrobial activity against the two pathogens, based on the diameter of the inhibition zones. The zone diameter of inhibition of ethanol extracts of *Scenedesmus* sp and *Chlorococcum* sp at 500 mg/ml and 250mg/ml against *Escherichia coli* shows that they both met the standard of the antibiotic used as the positive control (chloramphenicol) with 26.0±0.0 mm and 20±2.0 mm, respectively. Additionally, *Staphylococcus aureus* showed significant zone diameter of inhibition of 18mm and 17mm against *Scenedesmus* sp and *Chlorococcum* sp respectively at a concentration of 500 mg/ml. Similarly, Marrez *et al*. (2017) revealed that *Chlorococcum* sp fractions FIV and FVII had antibacterial activity against *Staphylococcus aureus* ATCC 13565, *Bacillus cereus* EMCC 1080, *Salmonella* typhi ATCC 25566, *Pseudomonas aeruginosa* NRRL B-272, and *Klebsiella pneumoniae* LMD 7726. Our study also revealed that *Scenedesmus* sp. had MIC of 125 mg/ml against *Escherichia coli* and MIC of 250mg/ml against *Staphylococcus aureus* while the extracts of *Chlorococcum* sp had a higher MIC of 250 mg/ml against both *Escherichia coli* and *Staphylococcus aureus*. Correspondingly, Lauritano *et al*. (2016) showed that *Oscillatoria* sp crude extracts hada maximum antibacterial activity of 156.24 mg/ml against *S. aureus*.Nianangu *et al*. (2020) also reported that MeOH: CHCl3 (70:30) extract of *Oscillatoria* sp. SSCM01 exhibits the strongest antibacterial activity against *Staphylococcusl aureus* and *Salmonella typhi* with minimum inhibition concentration of 31.2 µg/mL and 7.8 2 µg/mL, respectively In this study the MIC value of 125mg/ml was observed for *Staphylococcus aureus* and *Escherichia coli* under *Scenedesmus sp* extract and 250 mg/ml under *Chlorococcum* sp. The low MIC value suggested that its biochemical content has a strong antimicrobial activity since it can largely inhibit microbial growth at low concentrations (Moustafa *et al*., 2020). More so, the minimum bactericidal concentration (MBC) values of the microalgae extracts showed antibacterial potency against *Escherichia coli* and *Staphylococcus aureus*. This study showed a significant ($p<0.05$) difference in the antimicrobial activity towards the two bacterial strains.

CONCLUSION

The findings of this study demonstrate the significant bioactive potential of *Chlorococcum* sp and *Scenedesmus* sp, particularly in their antimicrobial and antioxidant properties. The presence of key phytochemicals such as alkaloids, flavonoids, steroids, phenols, and saponins suggests their potential application in pharmaceutical and nutraceutical industries. The strong antioxidant activity observed highlights their capability to mitigate oxidative stress, while their antimicrobial effects against *Escherichia coli* and *Staphylococcus aureus* suggest their relevance in combating microbial infections and antibiotic resistance. The observed MIC values further support their efficacy at varying

concentrations, positioning them as promising candidates for pharmaceutical development. Given their adaptability and ease of cultivation, these microalgae offer a sustainable source for bioactive compounds. Future research should focus on optimizing extraction methods and assessing their broader applications in biomedicine and biotechnology.

Author contributions

Conceptualization: SSA, NHA; Experimentation and data collection: NHA, SAS, OS; Data analysis: SSA; Interpretation of findings: SSA, YAG, IZA; Writing and revision of manuscript: NHA SSA, YAG, IZA, OS; Supervision: SSA, YAG, OS

Declaration of competing interests

The authors confirm that they have no known financial interests or personal associations that may potentially impact the conclusions or analysis of the findings presented in this study.

Funding

This research did not receive any specific grant support from public, private, or non-profit funding organizations.

REFERENCES

Abdelrheem, D.A., Rahman, A.A., Elsayed, K.N.M., Ahmed, S.A., (2020). GC/MS spectroscopic approach, antimicrobial activity and cytotoxicity of some marine macroalgae from Qusier and Marsa Alam Seashore (Red Sea). Egypt. *Egyptian Journal of Aquatic Biology and Fish*eries, 24 (3), 125–144. https://doi.org/10.21608/ ejabf.2020.88670.

Adeniyi-Martins, O., Fasuba, O. A., Bolajoko, A. A., Nwankwo, D. I., & Adesalu, T. A. (2023). Antioxidant and antibacterial activities of Oscillatoria sp. and Chlorella sp. *Journal of Scientific Research and Development*, *22*(1), 126-140. https://jsrd.unilag.edu.ng/article/view/2364

Alsenani, F., Tupally, K. R., Chua, E. T., Eltanahy, E., Alsufyani, H., Parekh, H. S., & Schenk, P. M. (2020). Evaluation of microalgae and cyanobacteria as potential sources of antimicrobial compounds. *Saudi Pharmaceutical Journal*, *28*(12), 1834-1841. https://doi.org/10.1016/j.jsps.2020.11.010

Alshuniaber, M.A., Krishnamoorthy, R., Al-Qhtani, W.H., (2021). Saudi Journal of Biological Sciences Antimicrobial activity of polyphenolic compounds from Spirulina against food-borne bacterial pathogens. *Saudi J. Biol. Sci.* 28 (1), 459 464. https://doi.org/10.1016/j.sjbs.2020.10.029.

Andima, M., Bernard, S., Owor, R., and Edward, A. (2014). Preliminary qualitative analysis of phytochemical constituents of the Endemic Aloe tororoana Raynolds in Tororo, Eastern Uganda. *Global Advanced Research Journal of Agricultural Science*, 3(3), 2315-5094.

Bajpai, V.K., (2016). Antimicrobial bioactive compounds from marine algae: A mini review. *Indian Journal of Geo-Marine Sci*ences, 45, 1076–1085.

Dai, J., and Mumper, R. J. (2010). Plant phenolics: Extraction, analysis and their antioxidant and anticancer properties. *Molecules*, 15(10).

Davoodbasha, M., Edachery, B., Nooruddin, T., Lee, S. Y., & Kim, J. W. (2018). An evidence of C16 fatty acid methyl esters extracted from microalga for effective antimicrobial and antioxidant property. *Microbial Pathogenesis*, *115*, 233-238. https://doi.org/10.1016/j.micpath.2017.12.049

Elshobary, M. E., El-Shenody, R. A., Ashour, M., Zabed, H. M., & Qi, X. (2020). Antimicrobial and antioxidant characterization of bioactive components from *Chlorococcum minutum*. *Food Bioscience*, *35*, 100567. https://doi.org/10.1016/j.fbio.2020.100567

Gogineni, V., Hamann, M.T., (2018). Marine natural product peptides with therapeutic potential: Chemistry, biosynthesis, and pharmacology. *Biochim. Biophys. Acta* 1862 (1), 81–196. https://doi.org/10.1016/j.bbagen.2017.08.014.

Jayshree, A., Jayashree, S., and Nallamuthu, T. (2016). Chlorella vulgaris and Chlamydomonas reinhardtii: Effective antioxidant, antibacterial and anticancer mediators. *Indian Journal of Pharmaceutical Sciences*, 78, 575-581.

Kumar, K. S., Ganesan, K., & Rao, P. V. S. (2008). Antioxidant potential of solvent extracts of Kappaphycus alvarezii (Doty) Doty - An edible seaweed. *Food Chemistry*, 107(1), 289-295

Lauritano, C., Andersen, J.H., Hansen, E., Albrigtsen, M., Escalera, L., Esposito, F., Helland, K., Hanssen, K.Ø., Romano, G., Ianora, A., (2016). Bioactivity screening of microalgae for antioxidant, anti-inflammatory, anticancer, anti-diabetes, and antibacterial activities. *Frontiers of Marine Sci*ences, 3, 1–12. https://doi.org/10.3389/ fmars.2016.00068.

León-Vaz, A., León, R., Vigara, J., & Funk, C. (2023). Exploring Nordic microalgae as a potential novel source of antioxidant and bioactive compounds. *New*

Biotechnology, *73*, 1-8.https://doi.org/10.1016/j.nbt.2022.12.001

Liang, F., Wen, X., Geng, Y., Ouyang, Z., Luo, L., Li, Y., (2013). Growth rate and biomass productivity of chlorella as affected by culture depth and cell density in an open circular photobioreactor. *Journal of Microbiology and. Biotechnol*ogy, 23, 539–544.

Marrez, D.A., Naguib, M.M., Sultan, Y.Y., Higazy, A.M., (2019). Antimicrobial and anticancer activities of *Scenedesmus obliquus* metabolites. *Heliyon* 5 (3), e01404. https://doi.org/10.1016/j.heliyon.2019.e01404.

Marrez, D.A., Sultan, Y.Y., Embaby, M.A., (2017). Biological activity of the cyanobacterium Oscillatoria brevis extracts as a source of nutraceutical and bio-preservative agents. *International Journal of Pharmacology,* 13 (8), 1010–1019.

Nainangu, P., Antonyraj, A. P. M., Subramanian, K., Kaliyaperumal, S., Gopal, S., & Renuka, P. S. (2020). In vitro screening of antimicrobial, antioxidant, cytotoxic activities, and characterization of bioactive substances from freshwater cyanobacteria Oscillatoria sp. SSCM01 and Phormidium sp. SSCM02. *Biocatalysis and Agricultural Biotechnology*, *29*, 101772. https://doi.org/10.1016/j.bcab.2020.101772

Okwu, D. E. (2001). Improving the nutritive value of Cassava Tapioca meal with local spices. *Journal of Nutraceuticals, Functional & Medical Foods*, 3(4), 43-50.

Patra, J.K., Patra, A.P., Mahapatra, N.K., Thatoi, H.N., Das, S., Sahu, R.K., Swain, G.C., 2009. Antimicrobial activity of organic solvent extracts of three marine macroalgae from Chilika Lake, Orissa. India. Malays. *Journal of Microbiol*ogy, 5, 128–131. https://doi.org/10.21161/mjm.15409.

Russell C, Rodriguez C, Yaseen M. (2022). High-value biochemical products & applications of freshwater eukaryotic microalgae. *Science of Total Environment,* 809:151111. https:// doi.org/10.1016/j.scitotenv.2021.151111

Salaam, A. M., Adebayo-Tayo, B. C., & Ajibade, A. A. (2019). Phytochemical analysis, antioxidant, antibacterial potentials and chemical composition of methanol extract of Oscillatoria sp. *Nigerian Journal of Pharmaceutical Research*, *15*(2), 219-227. https://doi.org/10.4314/njpr.v15i2.9

Sawant, S.S., Mane, V.K., 2018. Nutritional profile, antioxidant, antimicrobial potential, and bioactives profile of Chlorella emersonii kj725233. *Asian Journal of Pharmacy Clinical Res*earch, 11, 220–225. https://doi.org/10.22159/ajpcr.2018. v11i3.21990

Seddek, N. H., Fawzy, M. A., El-Said, W. A., & Ahmed, M. M. R. (2019). Evaluation of antimicrobial, antioxidant and cytotoxic activities and characterization of bioactive substances from freshwater blue-green algae. https://doi.org/10.30955/gnj.002949

Shah, M., Mahfuzur, R., Liang, Y., Cheng, J.J., Daroch, M., (2016). Astaxanthin-producing green microalga Haematococcus pluvialis: from single cell to high value commercial products. *Frontier of Plant Sciences,* 7, 531.

Shaima, A. F., Yasin, N. H. M., Ibrahim, N., Takriff, M. S., Gunasekaran, D., & Ismaeel, M. Y. (2022). Unveiling antimicrobial activity of microalgae Chlorella sorokiniana (UKM2), Chlorella sp.(UKM8) and Scenedesmus sp. (UKM9). *Saudi Journal of Biological Sciences*, *29*(2), 1043-1052. https://doi.org/10.1016/j.sjbs.2021.09.069

Shaima, A.F., Gires, U., Asmat, A., (2016). Algicidal activity of Aeromonas hydrophila sdPS-7 isolate against toxic marine dinoflagellate Alexandrium minutum KB-5. *Malays. Journal of. Microbiology,* 12, 24–29.

Singh, S. K., Kaur, R., Bansal, A., Kapur, S., & Sundaram, S. (2020). Biotechnological exploitation of cyanobacteria and microalgae for bioactive compounds. In *Biotechnological production of bioactive compounds* (pp. 221-259). https://doi.org/10.1016/B978-0-444-64323-0.00008-4

Solanki, R., Khanna, M., and Lal, R. (2008). Bioactive compounds from marine actinomycetes. *Indian Journal of Microbiology,* 48(4), 410-431.

Sultana, B., Anwar, F., & Iqbal, S. (2008). Effect of different cooking methods on the antioxidant activity of some vegetables from Pakistan. *International Journal of Food Science and Technology*, *43*(3), 560-567.

van Vuuren, S. J., Taylor, J., & van Ginkel, C. (2006). Freshwater algae. *Sch Environ Sci Dev Bot*.

Wilson, D., Nash, P., Buttar, H., Griffiths, K., Singh, R., De Meester, F., Horiuchi, R., Takahashi, T., (2017). The role of food antioxidants, benefits of functional foods, and influ ence of feeding habits on the health of the older person: an overview. *Antioxidants,* 6, 81

Yap, W.-F., Tay, V., Tan, S.-H., (2019). Decoding coding antioxidant and antibacterial potentials of Malaysian green seaweeds: Caulerpa racemosa and Caulerpa lentillifera. *Antibiotics,* 8, 152. https://doi.org/10.3390/antibiotics8030152.

Zabed, H. M., Akter, S., Yun, J., Zhang, G., Zhang, Y., & Qi, X. (2020). Biogas from microalgae: Technologies, challenges and opportunities. *Renewable Sustainable Energy Review*, 117, Article ID 109503; doi; 10.1016/j.rser.2019.109503