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Comprehensive Profiling of Phenolic Compounds and Antioxidant Activity in Marine Macroalgae from Sabratha Coast

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ABSTRACT

The growing demand for natural antioxidants underscores the need to explore under-investigated marine resources. This study examines the biochemical diversity and antioxidant potential of three dominant macroalgae species *(Fucus vesiculosus, Jania sp., and Laurencia sp.)* collected from the Libyan coast (Sabratha). Extraction was carried out using aqueous, ethanol, ethanol/water, methanol, and acetone solvents to assess solvent-dependent efficiency. The ethanolic extract of *Fucus vesiculosus* showed the highest total phenolic content *(92.5 ± 7.5 mg GAE/g)*, while the acetone extract of *Jania sp.* exhibited the strongest DPPH radical scavenging activity *(75.82% at 40 µg/mL)*. These results highlight the key role of solvent polarity in enhancing bioactivity and indicate the presence of potent non-phenolic antioxidants. This study provides baseline quantitative data on Libyan marine algae and highlights their potential for nutraceutical and functional food applications under Mediterranean environmental conditions.

تحليل شامل للمركبات الفينولية والنشاط المضاد للأكسدة في الطحالب البحرية الكبيرة من ساحل صبراتة، ليبيا

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يسلّط الطلب المتزايد على مضادات الأكسدة الطبيعية الضوء على أهمية استكشاف الموارد البحرية غير المدروسة. تهدف هذه الدراسة إلى تقييم التنوع الكيميائي الحيوي والنشاط المضاد للأكسدة لثلاثة أنواع من الطحالب الكبيرة (*Fucus vesiculosus* و *Jania sp.* و *Laurencia sp.*) من ساحل صبراتة. استُخدمت مذيبات مختلفة (الماء، الإيثانول، الإيثانول/الماء، الميثانول، والأسيتون) لدراسة تأثير قطبية المذيب على كفاءة الاستخلاص. أظهر المستخلص الإيثانولي لطحلب *Fucus vesiculosus* أعلى محتوى فينولي كلي *(92.5 ± 7.5 ملغم مكافئ حمض الغاليك/غرام)*، بينما سجّل مستخلص الأسيتون لطحلب *Jania sp.* أعلى نشاط مضاد للأكسدة *(75.82%)*. تؤكد النتائج أهمية قطبية المذيب وتشير إلى وجود مركبات غير فينولية فعالة، كما تبرز إمكانات الطحالب الليبية في تطبيقات المكملات الغذائية والأغذية الوظيفية.

INTRODUCTION

Marine macroalgae are increasingly recognized as sustainable sources of bioactive compounds, particularly phenolics, which play a vital role in antioxidant defense mechanisms (Cadar et al., 2025) Their biochemical composition varies across brown, red, and green algae, influenced by ecological and environmental factors such as salinity, temperature, and pollution (Chen et al., 2024) Globally, species such as Fucus vesiculosus, Jania sp., and Laurencia sp. have been studied for their phenolic content and antioxidant activity, yet the Libyan coast remains largely unexplored.

The originality of this study lies in its focus on the Sabratha region, where unique Mediterranean conditions may induce distinct metabolite profiles. Fucus vesiculosus is known for phlorotannins and fucoidans, Janiasp. for sterols and terpenoids, and Laurencia sp. for halogenated metabolites with strong antioxidant potential (Hernández et al., 2025) By evaluating solvent-dependent extraction efficiency, this research aligns with principles of green extraction (ethanol/water systems) and addresses industrial scalability and bioavailability challenges (Zhao et al., 2025).

The aim

1. Quantify the total phenolic content (TPC) in three Libyan macroalgal species.
2. Assess antioxidant activity using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay under different solvent systems.
3. Compare the findings with recent global studies (2023–2025) to highlight regional biochemical differences.

Discuss the implications for the nutraceutical and functional food industries.

Study area

In August 2024, samples of *Fucus vesiculosus*, *Jania* sp., and *Laurencia* sp. collected from Sabratha Beach, Libya. This beach is an ecologically rich area, abundant in seaweed and characterized by high biodiversity. The geographical boundaries of the sampling sites are shown in Figure 1. The collected algal samples were taxonomically identified at the Department of Ecology, Faculty of Science, Sabratha University, Sabratha, Libya. (Errayes *et al*., 2025).

Figure (1) Geographical boundaries of samples collected from the beach of Sabratha city.

MATERIALS AND METHODS

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1. Sample Collection and Preparation

Fresh specimens were rinsed thoroughly with seawater to remove epiphytes and sand, followed by washing with distilled water. Samples were air-dried at room temperature (25 ± 2 °C) for 72 hours, then ground into fine powder using a mechanical grinder. The powdered material was stored at −20 °C until extraction.

2. **Extraction Procedure**

Five extraction solvent systems were utilized: aqueous extract (100% deionized water), ethanol, ethanol/water mixture (70:30, v/v), methanol, and acetone. For each extraction procedure, 1.0 g of powdered algal material was combined with 20 mL of solvent at a solid-to-solvent ratio of 1:20 (w/v). The mixtures were then incubated under orbital shaking at 150 rpm for 24 h at 25 °C. The extracts were centrifuged at 5000 rpm for 10 min, and the resulting supernatants were filtered using Whatman No. 1 filter paper. The solvents were subsequently removed under reduced pressure with a rotary evaporator at 40 °C. The dried residues were reconstituted in their respective solvents to obtain a final concentration of 10 mg/mL prior to analysis. All extraction procedures were carried out in triplicate.

Extracts were centrifuged at 5000 rpm for 10 minutes, and supernatants were filtered through Whatman No.1 paper. Solvents were evaporated under reduced pressure using a rotary evaporator at 40 °C. Dried extracts were redissolved in the respective solvents to a final concentration of 10 mg/mL for analysis. All extractions were performed in triplicate.

3 **Phytochemical Screening**

The tested extracts were screened for alkaloids, flavonoids, tannins, saponins, Phenolic compounds, turbinoids, glycosides, and steroids (Harborne, 1998).

4 Total Phenolic Content (TPC)

The total phenolic content (TPC) of the three different extracts obtained from each macroalgal species was estimated following the method described by Heffernan et al. (2014, 2015).

The antioxidant activity of extracts from Fucus vesiculosus, Jania sp., and Laurencia sp. was evaluated using commonly used assays: DPPH (Sánchez-Bonet et al., 2025; Gil-Martínez et al., 2025).

RESULTS AND DISCUSSION

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4. **Discussion**

4.1 Phytochemical of Selected Marine Algae

Table 1 shows the results of the qualitative phytochemical examination of three species of marine algae. The presence of many phytochemicals has been tested using standard procedures, and the results are qualitatively expresse.

Table1. The results of the qualitative phytochemical examination of species of marine algae

Species	Aqueous Extract	Ethanol Extract	Ethanol/Water Extract
Fucus vesiculosus	52.5 ± 7.5^{b}	92.5 ± 7.5^{a}	77.5 ± 7.5
Laurencia sp.	30.0 ± 5.0^{b}	47.5 ± 7.5^{a}	42.5 ± 7.5
Jania sp.	15.0 ± 5.0^{b}	25.0 ± 5.0^{a}	23.0 ± 5.0

4.2 **Total Phenolic Content (TPC)**

Table 2. Total phenolic content (TPC) in three algae species (mean± SD, n = 3).

Phytochemicals	Fucus vesiculosus	Janya .P	Laurencia p.
Flavonoids	+++	++	++
Alkaloids	+	±	+
Phenolic compounds	+++	++	+++
Glycosides	±	±	±
Tannins	++	+	±
Saponins	+	±	±
Turbinoids	++	+	+++
Steroids	±	±	+

Key: +++ = High Attendance, ++ = Moderate Attendance, + = Low Attendance, ± = Tracking or Uncertain Presence

The TPC values varied significantly among species and solvents (ANOVA, F = 47.71, $p < 0.001$). Ethanol proved to be the most effective solvent for *Fucus vesiculosus*, yielding 92.5 ± 7.5 mg GAE/g, significantly higher than aqueous (52.5 ± 7.5 mg GAE/g) and ethanol/water extracts (77.5 ± 7.5 mg GAE/g). Superscript letters indicate statistically significant differences ($p < 0.05$) within each species.

4.3 Antioxidant Activity (DPPH Assay)

Antioxidant activity also varied significantly among solvents and species (ANOVA, $p < 0.001$). For *Fucus vesiculosus*, methanol extracts showed the highest inhibition (77.67% at 40 µg/mL), followed by acetone (71.77%) and ethanol (60.73%). In contrast, *Jania sp.* exhibited maximum activity in acetone extracts (75.82%), while *Laurencia sp.* also favored acetone (69.69%)

Table 3. DPPH radical scavenging activity % 40 µg/ mL

Species	Methanol Extract	Acetone Extract	Ethanol Extract
Fucus vesiculosus	77.67 ± 2.1^{a}	71.77 ± 1.9^{ab}	60.73 ± 2.5^{b}
Jania sp.	70.92 ± 2.3^{ab}	75.82 ± 2.0^{a}	54.60 ± 2.8^{b}
Laurencia sp.	64.54 ± 2.4^{ab}	69.69 ± 2.2^{a}	53.86 ± 2.7^{b}

Note: Superscript letters (a, b, c, d) indicate statistically significant differences (p < 0.05) within each row (between species) or column (between solvents).

This behavior is consistent with the presence of phlorotannins, whose extraction is favored by semi-polar solvents such as ethanol.

The obtained values are slightly lower than those reported in previous studies, which may be attributed to geographical and environmental variability affecting phenolic biosynthesis. *Laurencia* sp. displayed moderate TPC levels, with ethanolic extracts outperforming aqueous systems, in agreement with literature data. In contrast, *Jania* sp. showed the lowest TPC values, likely due to its calcareous structure limiting solvent penetration.

Overall, ethanol proved to be the most efficient solvent for phenolic extraction, followed by ethanol/water mixtures and water. Brown algae exhibited significantly higher phenolic contents than red algae, although the latter remain biologically relevant sources of antioxidant-related compounds.

4.4 **Antioxidant activity**

Antioxidant activity of algae extracts Tables (2), (3) and (4) showed that the antioxidant activity of the study algae varied in the resulting values. Fucus vesicuiosus Compared to vitamin C (ascorbic acid), we find that the methanol extract of algae is higher than acetone, while ethanol is lower.

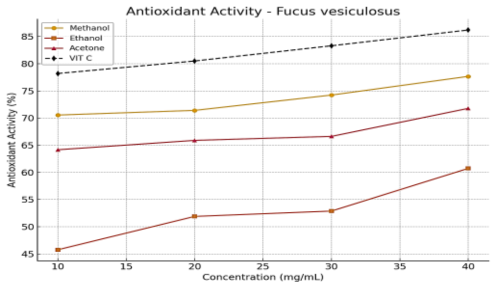

Figure 2: DPPH inhibition (%) at different concentrations for Fucus vesiculosus extracts.

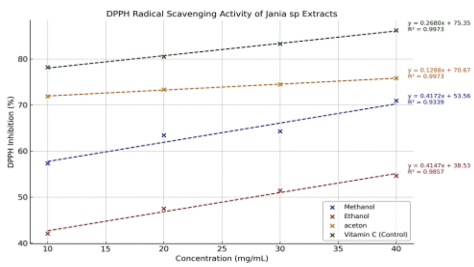

Figure 3: DPPH inhibition (%) at different concentrations for Jania sp extracts.

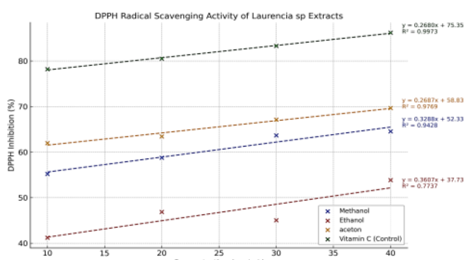

Figure 4: DPPH inhibition (%) at different concentrations for Laurencia sp.

The results demonstrate a clear variation in both phenolic content and antioxidant activity, influenced by the algal species and the extraction solvent's polarity. The DPPH inhibition (%) at different concentrations for the three species is conceptually illustrated in Figure 2, which combines the individual inhibition curves for better comparative analysis.

Effect of Solvent Polarity on Extraction Efficiency: The efficiency of extracting bioactive compounds is highly dependent on the solvent's polarity, as it dictates the solubility of specific metabolites. In this study, **ethanol** and **methanol** were particularly effective for *Fucus vesiculosus*. This is attributed to the presence of phlorotannins, which are polar polyphenolic compounds unique to brown algae. The semi-polar nature of ethanol allows for better penetration of the algal cell wall and stabilization of these molecules (Holdt & Kraan, 2011). Conversely, acetone yielded the highest antioxidant activity for *Jania* sp. and *Laurencia* sp. This suggests that the primary antioxidants in these red algae might be less polar, such as halogenated terpenoids and bromophenols, which are more soluble in acetone than in highly polar aqueous or alcoholic systems(El Gamal, 2010; Tierney et al., 2013).

Interspecific Variation in Bioactive Profiles: The brown alga *Fucus vesiculosus* consistently exhibited higher TPC (92.5 mg GAE/g in ethanol) compared to the red algae *Jania* sp. and *Laurencia* sp. This is consistent with global trends where brown algae are known to accumulate high concentrations of phlorotannins as a defense mechanism against UV radiation and herbivory (Wang et al., 2009). The lower TPC in *Jania* sp. can be explained by its calcareous structure, which not only acts as a physical barrier to solvent extraction but also represents a different evolutionary strategy where structural defense (calcification) is prioritized over chemical defense (phenolics). However, the high antioxidant activity of *Jania* sp. in acetone (75.82%) despite lower phenolics indicates a significant contribution from non-phenolic antioxidants, highlighting the biochemical diversity of Mediterranean macroalgae (Jiménez-Escrig et al., 2001).

Regional Significance: The unique environmental conditions of the Sabratha coast, including specific salinity and temperature fluctuations, likely contribute to the distinct metabolite profiles observed. While the TPC values are slightly lower than some tropical species, the high antioxidant efficiency at low concentrations (40 µg/mL) suggests a high quality of bioactive compounds, making these species promising candidates for the nutraceutical industry.

4.5 Comparative Insights

Brown algae (*Fucus vesiculosus*) consistently exhibited higher phenolic content compared to red algae (*Jania sp.* and *Laurencia sp.*), confirming previous global findings (Tierney et al., 2013).The strong antioxidant activity of acetone extracts in red algae suggests the presence of non-phenolic compounds such as bromophenols and halogenated terpenoids (Zhang et al., 2020).

(salinity, temperature fluctuations) may explain the moderate but distinct antioxidant profiles compared to algae from Asian waters (Zhang et al., 2020).

4.6 Statistical Analysis

All experiments were conducted in triplicate (n = 3). Data were expressed as mean ± standard deviation (SD). Statistical differences among species and solvents were analyzed using one-way ANOVA followed by Tukey's post hoc test, with significance set at $p < 0.05$. Analyses were performed using SPSS v.25 software.

5. Conclusion

This study presents original quantitative data on the phenolic content and antioxidant activity of three marine macroalgae (*Fucus vesiculosus*, *Jania* sp., and

Laurencia sp.) collected from the underexplored Sabratha coast, Libya. The findings confirm that extraction efficiency and antioxidant performance are strongly influenced by both algal taxonomy and solvent polarity.

Ethanolic extracts of *Fucus vesiculosus* exhibited the highest total phenolic content, supporting ethanol as an effective and relatively green solvent for recovering polar and semi-polar phenolics. In contrast, *Jania* sp. and *Laurencia* sp. demonstrated notable antioxidant activity in acetone extracts despite lower phenolic levels, indicating the contribution of non-phenolic bioactive compounds and reflecting the biochemical diversity of these species.

While the study is limited by single-season sampling and the absence of compound-level characterization, it provides reliable baseline data specific to the Libyan Mediterranean coast. These results highlight the potential of these algae as natural antioxidant sources and support their relevance for future nutraceutical and functional food applications. Further studies should prioritize seasonal assessment, metabolomic profiling, and evaluation of bioavailability to facilitate industrial exploitation.

Conflicts of interest

The authors declare that they have no conflicts of interest, financial or otherwise, that could have influenced the research presented in this paper.

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