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Determination of Composition, Antioxidant and Antibacterial Properties of Enzymatic Algae Extracts

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ABSTRACT:

Polyphenolic compounds extracted from various algae species have a wide range of biological activities. The aim of the study was to determine the antioxidant and antibacterial properties of enzymatic extracts of the algae *Fucus vesiculosus* and *Furcellaria lumbricalis*. Identification of algae collected from the Baltic Sea coast showed that they belonged to the species *F. lumbricalis* and *F. vesiculosus* L. The content of total phenolics in *F. vesiculosus* sample was found to be 40.16 ± 0.50 and 38.36 ± 0.40 mg/g in *F. lumbricalis* sample. Cellulase had the greatest effect on both *F. vesiculosus* and *F. lumbricalis* in enzymatic hydrolysis, with extraction yields of $40.16 \pm 1.00\%$ and $38.36 \pm 2.13\%$, respectively. The gallic acid content of *F. vesiculosus* was $176.05 \mu\text{g/g}$ and that of *F. lumbricalis* was $20.37 \mu\text{g/g}$. Among the algal extracts, the samples obtained by enzymatic extraction at 45°C for 12 h were the most efficient in scavenging DPPH radicals; the value for *F. vesiculosus* was $287.15 \pm 0.90 \mu\text{mol TE/g}$ and for *F. lumbricalis* was $273.36 \pm 0.90 \mu\text{mol TE/g}$. The activity of *F. lumbricalis* extracts against ABTS radicals was significantly lower than that of *F. vesiculosus* extracts. The extracts obtained using cellulase enzyme had the highest antimicrobial activity against *E. coli* (20.0 ± 0.5 mm), against *P. aeruginosa* (24.0 ± 0.5 mm), *B. subtilis* (18.0 ± 0.5 mm), and *C. albicans* (23.0 ± 0.5 mm). The key factors in the extraction of polyphenols are the choice of enzyme and optimal extraction conditions, which confirms the prospect of their use as polyphenolic components for the production of pharmaceuticals.

Keywords: brown and red algae, enzymatic extraction, chemical composition, polyphenols, biological activity.

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1. Introduction

Polyphenols are one of the most abundant groups of substances in the Plantae and Chromista kingdoms. Polyphenolic compounds extracted from various algae species have a wide range of biological activities, including antioxidant (Scalbert *et al.*, 2005; Hernández-Ledesma and Herrero, 2013; Wijesekara *et al.*, 2012), anti-inflammatory (Vo *et al.*, 2012), anticarcinogenic (Niedzwiecki *et al.*, 2016), antidiabetic (Lee and Jeon, 2013), inhibiting cardiovascular disorders, antihypertensive (Gómez-Guzmán *et al.*, 2018), antiviral (Nagahawatta *et al.*, 2022), and anti-allergic properties (Vo *et al.*, 2012). In addition to their role in inhibiting melanogenesis (Ding *et al.*, 2019), elastase and collagenase enzymes (Riani *et al.*, 2018), matrix metalloproteinases and hyaluronidase (Sanjeeva *et al.*, 2016) are important in the development of cosmeceutical products and pharmaceuticals. Due to their wide range of applications, seaweed-derived polyphenols are economically valuable candidates for mass production of various products.

Antioxidant and antimicrobial agents are the main drugs used to treat human and animal diseases. However, the ever-increasing number of new and expanding ranges of long-existing pathogens that have developed resistance to antimicrobial drugs in the world's population is becoming an increasingly significant global concern (Besednova *et al.*, 2020).

Globally, red and brown algae are the most studied in China. A team of scientists led by H. Zhou, a professor at Ningbo University's Kenneth Li Marine Biopharmaceutical Research Center, is conducting research on not only the commercial potential of red and brown algae, but also the pharmaceutical potential of using their polyphenols and phytosterols to find new drugs against Alzheimer's disease (Zhou, 2019).

Literature review shows that red and brown algae have global anti-neurodegenerative and antibacterial significance due to their unique cell wall composition. However, the available literature data do not lead to significant advances in this area of knowledge because they are predominantly descriptive, not supported by knowledge of the physiology and biochemistry of specific polyphenol-producing macroalgae strains, and geographically limited. Innovative approaches are needed to find cost-effective ways to isolate bioactive compounds, improve productivity, and elucidate the detailed mechanisms of anti-neurodegenerative and antibacterial effects (Zhou, 2019). The realization of the research on polyphenols of macroalgae of different strains will make a significant contribution not only to fundamental biology, but also to the development of relevant branches of applied sciences.

The study aimed to determine the antioxidant and antibacterial properties of enzymatic extracts of algae *Fucus vesiculosus* and *Furcellaria lumbricalis*. The results of the study will form the foundation of the technology for the cultivation of *F. vesiculosus* and *F. lumbricalis* macroalgae and the production of polyphenols, which will allow the production of polyphenols on a large scale with high added value.

2. Materials and methods

Collection and Drying of Brown and Red Algae Samples From Onshore and Offshore Areas of The Baltic Sea

Three sites (eastern part of the beach of Zelenogradsk, Zaostrovie settlement on the eastern side of the cape Gvardeysky, western part of the beach of Baltiysk) were selected for algae collection in the water area of Gdansk and Kaliningrad bays of the Baltic Sea. The size of each site was largely determined by the coastal landscape, where the intertidal zone at low tide (i.e., the area available for sampling) extended only 50 m perpendicular to the shoreline (Sukhikh *et al.*, 2024).

Samples of the macroalgae *Fucus vesiculosus* L. and *Furcellaria lumbricalis* were collected from the intertidal zone at these sites using nitrile gloves to prevent contamination. Macroalgae samples were washed in ambient seawater to remove adherent surface material, individually placed in labeled plastic bags, and transported to the laboratory at $5\pm 1^\circ\text{C}$. The samples were processed and dried under laboratory conditions for further studies. Algae samples were dried in a Being BO-30FL desiccator (Being, China) at a temperature of 40°C for 72 hours. The dried algae were then pulverized using a laboratory mill LMZ-M1 (NV-Lab, Novosibirsk, Russia). The crushed algae samples were stored at room temperature in a dry and dark place until biochemical testing (Malkov *et al.*, 2024).

Selection of Rational Parameters for Extraction of Polyphenols and Phytosterols from Brown and Red Algae Biomass

Phenolic extracts of macroalgae were prepared by combined extraction method (enzyme+ultrasound treatment).

The enzyme preparation Cellulase was used as a carbohydrase and Proteinase K was used as a peptidase. To increase the extraction efficiency, the algae were pretreated with ultrasound for 30 min as described in section 2.4, but instead of water and acidified solution, buffer systems with optimal pH for enzyme operation were used.

Enzymatic Extraction

Total extracts were obtained by enzymatic extraction, which can increase the yield of phenolic compounds by releasing phenolics bound to carbohydrates or proteins in cells (Wijesekara *et al.*, 2012). Cellulase enzyme preparation was used as carbohydrase and Protease K preparation was used as peptidase. To increase the extraction efficiency, the algae were pretreated with ultrasound for 30 minutes as described in section 2, but instead of water and acidified solution, buffer systems with optimal pH for enzyme operation were used: acetate buffer for cellulase (pH=4.5) and phosphate buffer for protease (pH=8.0). 100 mg of the enzyme preparation was added to the extraction mixture and incubated at 50°C for 3 hours. Constant stirring was maintained on a rotary shaker. Enzyme inactivation was performed by boiling the mixture for 5 min, after which the extracts were immediately cooled to 4°C on ice and centrifuged at 3800 rpm for 10 min to separate the supernatant. In parallel, a control experiment was performed without the addition of the enzyme preparation.

Study of the Component Composition of Extracts

Component and metabolomic analyses of *F. vesiculosus* and *F. lumbricalis* extracts were performed using HPLC. The process was performed on a Shimadzu Prominence LC-20AB chromatograph with a binary pump. SPD-M20A diode matrix detector. Zorbax 300SB-C18 4.6*250mm 5 μm column (Agilent). The separation was performed at 40°C in gradient elution mode. The mobile phase: eluent A – 0.1% trifluoroacetic acid in bidistilled water, B – acetonitrile. The flow rate was 1 mL/min and the analytical wavelengths were 254, 280 and 325 nm. Compounds were identified from retention times and spectra of individual standards. The concentration of the compounds was calculated from calibration curves. The components were identified by retention times and spectra of individual standards. The concentration of the compounds was calculated from calibration curves.

Determination of the Antioxidant Activity of the Extracts

To study the antioxidant activity (AOA) of *F. vesiculosus* and *F. lumbricalis*, methods based on the interaction of antioxidants with DPPH (2,2-diphenyl-1-picrylhydrazyl), ABTS (2,2-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) radicals, as well as on the determination of

the reducing ability in their interaction with Fe(III) complexes with photometric reagent - tripyridyltriazine, were used.

For determination of antioxidant activity by the DPPH method, 20 μL of extract or standard solution (Trolox) was mixed with 300 μL of freshly prepared 0.1 mmol/L solution of 2,2-diphenyl-1-picrylhydrazyl. The mixture was incubated for 40 minutes in the dark at room temperature. The decrease in optical density compared to the control consisting of 0.1 mmol/L solution of 2,2-diphenyl-1-picrylhydrazyl and the corresponding solvent used for extraction was recorded on a spectrophotometer (Shimadzu, Japan) at 515 nm. For the determination of antioxidant activity by the ABTS method, an ABTS radical solution was prepared. ABTS radical was generated by mixing aliquots of 7.0 mmol/L ABTS solution and 2.45 mmol/L potassium persulfate solution. The solution was incubated for 16 hours in a dark place at room temperature. For the reaction, 20 μL of the extract or standard (Trolox) was added to 300 μL of the prepared ABTS+ cation radical solution. The optical density was measured at 734 nm after the mixture was incubated for 15 min at 37°C in the dark. The sample with ABTS reagent and appropriate solvent was used as a control. When measuring antioxidant activity using DPPH, ABTS methods, solutions of Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) of known concentration were used as a standard solution. Results of the assays were expressed as μmol of trolox equivalents per gram of dry plant material ($\mu\text{mol TE}$). All spectrophotometric measurements were performed using a microplate reader (Eppendorf, Germany).

Determination of the antimicrobial activity of the extracts

Antimicrobial activity was determined by the disk diffusion method (on solid culture medium). The following test strains acquired from the State Research Institute of Genetics (Russia) were used as model microorganisms: Gram-negative bacteria (*Escherichia coli*, *Pseudomonas aeruginosa*) and Gram-positive bacteria (*Bacillus subtilis*), as well as yeast fungi of *Candida albicans* species.

The strains were cultivated on the dense nutrient medium LB (Dia-M, Russia). The test strain of microorganisms was sown on the agarized nutrient medium through a lawn into a Petri dish, and simultaneously a paper disk soaked with the tested extract was placed on the lawn. A concentration of 100 $\mu\text{g}/\text{disc}$ of the tested extract was used to determine the antimicrobial activity. A paper disk with appropriate solvent was used as negative control and a disk with antibiotic (kanamycin at a concentration of 30 $\mu\text{g}/\text{disc}$, ampicillin at a concentration of 10 $\mu\text{g}/\text{disc}$) was used as comparison preparation (positive control). The concentration of the microbial suspension was $1.5 \cdot 10^8$ CFU/mL. The diameter of the discs was 5.0 mm, and the thickness of the agar layer was 4.0 ± 0.5 mm. Petri dishes were incubated in thermostat TS-1/20 (Russia) at the temperature corresponding to the optimal growth temperature of each microorganism test strain for 24 ± 0.5 hours.

Statistical Processing

Statistical processing was performed using IBM SPSS Statistics 27 program (IBM, Armonke, NY, USA). Analysis of variance was performed using the Kraskell-Wallis test ($P < 0.05$), the Mann-Whitney criterion with Bonferroni correction for pairwise comparisons ($P < 0.016$), and Pearson's coefficient to test for the presence of correlation ($P < 0.05$). All experiments were repeated 5 times.

3. Results

The total extract yield and phenolic compound content for this extraction method are presented in Table 1.

Table 1: Performance of enzymatic extraction

| Enzyme preparation | Total yield, % | | phenolic content, % | |
|--------------------|----------------|----------------|---------------------|----------------|
| | F. vesiculosus | F. lumbricalis | F. vesiculosus | F. lumbricalis |
| Cellulase | 40.16±0.50 | 38.36±0.40 | 63.70±0.60 | 38.21±0.30 |
| Proteinase K | 28.77±0.30 | 26.49±0.30 | 51.34±0.50 | 27.11±0.20 |
| Control | 28.02±0.30 | 25.74±0.30 | 52.16±0.50 | 26.78±0.20 |

Table 2 shows the pigment content of macroalgae extracts using cellulase enzyme.

Table 2: Pigment content of F. vesiculosus and F. lumbricalis extracts using cellulase enzyme

| Pigment | Pigment content, mg/g | | | | |
|---------------|-----------------------|----------------|----------------|-----------------|-----------------|
| | Control | Cellulase | | Proteinase K | |
| | | F. vesiculosus | F. lumbricalis | F. vesiculosus | F. lumbricalis |
| Fucoxanthin | 0.220±0.00 2 | 0.523±0.015 | 0.411±0.012 | 0.330±0.00 2 | 0.280±0.00 2 |
| Violaxanthin | 0.010±0.00 2 | 0.080±0.002 | 0.036±0.001 | 0.020±0.00 2 | 0.016±0.00 2 |
| Anteroxanthin | bdl | bdl * | bdl | bdl | bdl |
| Zeaxanthin | 0.003±0.00 1 | 0.008±0.001 | 0.004±0.001 | bdl | bdl |
| Beta-carotene | 0.010±0.00 2 | 0.046±0.001 | 0.022±0.001 | 0.020±0.00 2 | 0.017±0.00 2 |

* bdl – below detection limit

When the metabolomic composition was studied, it was found that gallic acid was identified in all the extracts obtained. As an example, chromatograms are shown for extracts obtained by enzymatic extraction using cellulase enzyme (Figures 1-2).

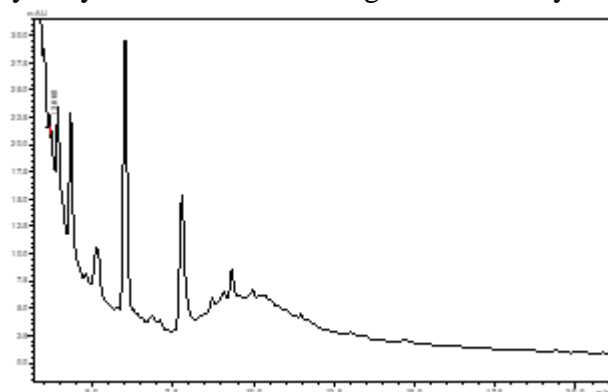


Figure 1: Chromatogram of F. vesiculosus extract

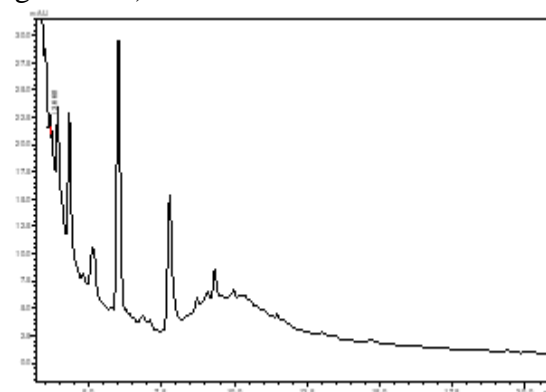


Figure 2: Chromatogram of F. lumbricalis extract

The results of the antioxidant activity study are shown in Table 3.

Table 3: Evaluation of the antioxidant activity of algae extracts

| AOA determination | AOA, μmol TE | | |
|-------------------|--------------|----------------|----------------|
| | Control | F. vesiculosus | F. lumbricalis |

| method | | Cellulase | Proteinase K | Cellulase | Proteinase K |
|--------|------------|-------------|-----------------|-------------|-----------------|
| DPPH | 90.92±0.70 | 287.15±0.90 | 159.35±0.70 | 273.36±0.90 | 120.16±0.70 |
| ABTS | 74.30±0.70 | 180,51±0.80 | 146.24±0.70 | 126,13±0.80 | 101.09±0.70 |

The results of the evaluation of the antibacterial activity of the algae extracts are presented in Table 4.

Table 4: Evaluation of the antibacterial activity of algae extracts

| Strain | Control | Diameter of lysis zone, mm | | | |
|----------------------|---------|----------------------------|-----------------|-----------------------|-----------------|
| | | <i>F. vesiculosus</i> | | <i>F. lumbricalis</i> | |
| | | Cellulase | Proteinase K | Cellulase | Proteinase K |
| <i>E. coli</i> | 7.0±0.5 | 20.0±0.5 | 7.5±0.5 | 10.0±0.5 | 6.0±0.5 |
| <i>P. aeruginosa</i> | 7.0±0.5 | 24.0±0.5 | 7.0±0.5 | 8.0±0.5 | 7.0±0.5 |
| <i>B. subtilis</i> | 6.0±0.5 | 18.0±0.5 | 0.0 | 6.0±0.5 | 0.0 |
| <i>C. albicans</i> | 5.3±0.5 | 23.0±0.5 | 0.0 | 2.0±0.5 | 0.0 |

4. Discussion

The main studies (Table 1) are aimed at the determination of substances of polyphenolic nature. The content of total phenolics using cellulase enzyme was found to be 38.21±0.30 mg/g in *F. vesiculosus* sample and 63.70±0.60 % in *F. lumbricalis* sample with total yield of extracts 40.16±0.50 mg/g and 38.36±0.40 mg/g, respectively. The results obtained indicate that the studied samples of red algae (*F. lumbricalis*) and brown algae (*F. vesiculosus*) are promising for further studies aimed at investigating the activity of polyphenols and phytosterols isolated from brown and red algae against Alzheimer's disease.

When studying the chemical composition of algae, it was found that the pigment complex of the studied samples includes carotenoids and chlorophyll. At the same time, the biomass of *F. vesiculosus* contains 1.9 times more carotenoids than the biomass of *F. lumbricalis*.

The greatest effect on both *F. vesiculosus* and *F. lumbricalis* in the extraction of polyphenolic compounds was cellulase, with extraction yields of 40.16±1.00% and 38.36±2.13%, respectively. The addition of peptidase did not significantly increase the total yield compared to the control. In the case of protease, no significant increase in total phenolic content was observed.

Analysis of the results showed that enzymatic extraction can increase the yield of phenolic compounds by releasing phenolics bound to carbohydrates or proteins in the cells (Habeebullah *et al.*, 2020).

The isolation and identification of various biologically active secondary metabolites from algae has been of recent scientific interest, with a particular focus on phenolic compounds and carotenoids. Carotenoids are some of the most important natural pigments with many health benefits, so at this stage the qualitative and quantitative composition of carotenoids present in the extracts of *F. vesiculosus* and *F. lumbricalis* were studied. The results presented in Table 2 allowed us to establish that the pigment profile of the studied samples is represented by beta-carotene, chlorophyll, fucoxanthin, violaxanthin and zeaxanthin. It was noted that the qualitative and quantitative content of the pigment profile can vary depending on the species of algae.

The gallic acid content of *F. vesiculosus* was 176.05 µg/g and that of *F. lumbricalis* was 20.37 µg/g. Gallic acid was also released when enzymatic extraction with enzymes was used. However, when peptidase was used, the extraction of gallic acid was lower at 127.07 and 14.07 µg/g, respectively.

Among the algae extracts, the samples obtained by enzymatic extraction using cellulase enzyme at 45°C for 12 h were the most efficient in scavenging DPPH radicals (Table 3); the value for *F. vesiculosus* was 287.15±0.90 µmol TE and for *F. lumbricalis* was 273.36±0.90 µmol TE, respectively. According to the ABTS method, the activity of *F. vesiculosus* and *F. lumbricalis* extracts obtained with the use of cellulase enzyme reached 180.51±0.80 µmol TE and 126.13±0.80 µmol TE, respectively. Analyzing the results presented in Table 3, it can be concluded that the best parameters for extraction of antioxidant complex from *F. vesiculosus* are room temperature and duration of 24 hours, and temperature 45°C with duration of 12 hours (cellulase enzymatic extraction). Under the above conditions, the activities of *F. vesiculosus* and *F. lumbricalis* extracts obtained with the proteinase K enzyme reached 159.35±0.70 µmol TE and 120.16±0.70 µmol TE, respectively, according to the DPPH method. The activity of *F. lumbricalis* extracts against ABTS radicals (Table 3) was significantly lower than that of *F. vesiculosus* extracts, which is in agreement with the results of total yield and total phenolic components.

Analyzing the data in Table 4, it can be concluded that the extracts obtained using cellulase enzyme have the highest antimicrobial activity against *E. coli* (20.0±0.5 mm), against *P. aeruginosa* (24.0±0.5 mm), *B. subtilis* (18.0±0.5 mm), and *C. albicans* (23.0±0.5 mm). The enzymatic extracts obtained using Proteinase K demonstrated activity exclusively against *E. coli* and *P. aeruginosa* in the case of the extract of *F. vesiculosus* and the extract of *F. lumbricalis*. The enzymatic extracts of *F. vesiculosus* exhibited minimal effects on *B. subtilis* and *C. albicans*.

The key factors in the extraction of polyphenols are the choice of enzyme and optimal extraction conditions.

The results correlate well with studies by leading scientists. The study (Jayawardhana *et al.*, 2023) described flavonoids as the most abundant subfamily of algal bioactive compounds. (Gómez-Guzmán *et al.*, 2018). These polyphenolic compounds can be isolated from various species of algae. Tannins are a family of phenolic metabolites that have special properties such as the ability to bind to proteins, large molecular size compounds, pigments, and metal ions (Hernández-Ledesma and Herrero, 2013). They have attracted considerable attention in recent decades because of their antioxidant capacity (Okuda and Ito, 2011). Phenolic acids possess a phenolic moiety and a resonance-stabilized structure with the ability to release protons, resulting in antioxidant activity through radical neutralization mechanisms. In addition, phenolic acids exhibit other health-protective effects such as antimicrobial, anti-inflammatory, anticancer and antimutagenic effects (Kumar and Goel, 2019). Stilbenes and lignans are small groups widely distributed in the plant and chromista kingdoms and can be identified as protective agents against protozoa, viral diseases and some forms of cancer (Tsopmo *et al.*, 2013).

Based on the mechanism of antioxidant activity, algal phenolic compounds can be classified as either radical scavengers or oxygen scavengers. Among these two mechanisms, radical scavenging is the most common and important method used to determine antioxidant activity (Fernando *et al.*, 2016). It is important to identify antioxidant agents that can be effectively utilized against free radicals. In general, seaweeds are constantly exposed to extreme environmental conditions that cause the accumulation of polyphenols to combat stress, inhibit oxidation, remove reactive oxygen species, and reduce DNA damage (Wei *et al.*, 2009). Thus, the most relevant studies include bioactive compounds isolated from seaweeds, especially polyphenolic compounds, which may play an important role in the pharmaceutical and cosmeceutical industries as photoprotectants due to their high potential for antioxidant activity (Wang *et al.*, 2015; Urquiaga and Leighton, 2000).

Farvin and Jacobsen (2013) reported that extracts of *Vertebrata fucoides* (formerly *Polysiphonia fucoides*) and *Fucus* spp. (Phaeophyceae) collected along the Danish coast

showed the best antioxidant activity among 16 different algae species. Antioxidant activity was measured using four in vitro antioxidant assays including 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity, reducing capacity, iron ion chelation and oxidation inhibition in a liposomal model system (Farvin and Jacobsen, 2013). Devi et al (2008) studied the in vitro antioxidant activity of 10 different algal species collected from Tamil Nadu, India. *Gelidiella acerosa*, a Rhodophyta species, has high antioxidant activity that may help prevent or slow the progression of several oxidative stress-related disorders (Devi *et al.*, 2008). According to a study by Shibata et al. (2007), polyphenols such as floroglucin (1,3,5-trihydroxybenzene), ecol, florofucofuroecol A, diecol, and 8,8'-biecol isolated from *Eisenia bicyclis*, *Ecklonia cava*, and *Ecklonia cava* subsp. Kurome (formerly *Ecklonia kurome*) (brown algae) showed significant activity against DPPH (50% effective concentration values: 12-26 μM) and superoxide anion (50% effective concentration values: 6.5-8.4 μM) and was more effective than α -tocopherol and ascorbic acid (Shibata *et al.*, 2007).

Another study showed that diecol, isolated from *Ecklonia cava*, is a potent source of photoprotective agent that showed significant protective effect against UV-B induced skin damage in human dermal fibroblasts. The results showed that diecol effectively reduced intracellular free radical activity while improving cell viability, which proved to be a powerful ingredient in the cosmeceutical industry (Wang *et al.*, 2021). Another study confirmed that fucoxanthin isolated from *Sargassum siliquastrum* (Phaeophyceae) has the ability to protect against UV-induced oxidative stress (Heo and Jeon, 2009).

Particulate matter is considered an environmental pollutant that has become a global problem due to oxidative stress that leads to apoptosis and tissue damage. The protective effect of *Caulerpa racemose* (Chlorophyta), a hexane fraction rich in clionasterol, against particulate matter-induced tissue damage was reported using human keratinocytes and a *Danio rerio* model. The results showed that the sample exhibited superior protective effects by reducing intracellular radical levels and mitochondrial radical levels. In addition, the *in vivo* results showed that clionasterol significantly reduced particulate-induced cell death, NO production and radical levels in a *Danio rerio* model (Liyanage *et al.*, 2022).

According to Kim et al. (2020), polyvinyl alcohol (PVA) hydrogels containing polyphenols from *Ishige okamurae* showed antibacterial effects in wound healing. The minimum inhibitory concentrations of polyphenols against *Staphylococcus aureus* and *Pseudomonas aeruginosa* were 128 $\mu\text{g/mL}$ and 512 $\mu\text{g/mL}$, respectively. In addition, the proposed PVA/polypheol hydrogels showed strong antibacterial effects compared with untreated groups of ICR mice *in vivo*. Thus, the data suggest that algal polyphenol hydrogels have great potential for use as antibacterial agents (Kim et al., 2020).

Based on the above studies, seaweed extracts can be considered as a potential source of therapeutic agents for wound healing and treatment of wound-related complications.

5. Conclusion

The samples collected for the study were found to belong to the red and brown algae division. Among the aqueous extracts of algae, the samples obtained by enzymatic extraction at 45°C for 12 h were the most efficient in absorbing DPPH radicals (Table 5); for *F. vesiculosus* the value was $287.15 \pm 0.90 \mu\text{mol TE}$, and for *F. lumbricalis* – $273.36 \pm 0.90 \mu\text{mol TE}$. The activity of *F. lumbricalis* extracts against ABTS radicals was significantly lower than that of *F. vesiculosus* extracts.

Extracts obtained using cellulase enzyme were shown to have the highest antimicrobial activity against *E. coli* ($20.0 \pm 0.5 \text{ mm}$), against *P. aeruginosa* ($24.0 \pm 0.5 \text{ mm}$), *B. subtilis* ($18.0 \pm 0.5 \text{ mm}$), and *C. albicans* ($23.0 \pm 0.5 \text{ mm}$).

It was found that the key factors in the extraction of polyphenols are the choice of enzyme and optimal extraction conditions. Thus, the results of the study of the biologically active properties of brown and red algae extracts confirm the prospects of their use as polyphenolic components for the production of pharmaceuticals.

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Conflict of Interest Declaration

The authors declare that they have NO affiliations with or involvement in any organization or entity with any financial interest in the subject matter or materials discussed in this manuscript.

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Author Contributions

Danil Malkov – performing research, analyzing and interpreting the data, and writing of the manuscript; **Elena Ulrikh** – interpreting the data, writing of the manuscript; **Svetlana Noskova** – developing a research methodology; **Alina Bakhtiyarova** – performing research; **Olga Babich** – conceiving, designing, and formalizing of the study. All authors accepted the final version.

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