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EFFECT OF MICROWAVE-ASSISTED TREATMENT ON THE COMPOSITION OF SPRAY-DRIED *PORPHYRIDIVM CRUENTUM* EXTRACT

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ABSTRACT

Introduction	Microalgae are a valuable source of natural products with bioactive properties, with applications in biomedicine, the pharmaceutical industry, nutrition, and cosmetics. Among them, the red microalga <i>Porphyridium cruentum</i> is notable for its high content of biomolecules with antioxidant, anti-inflammatory, and immunomodulatory effects. Efficient extraction and preservation methods are essential to optimize the use of these biomolecules.
Material and methods	Biomass from <i>Porphyridium cruentum</i> was treated with microwaves (MW) at 180 W, 300 W, and 450 W for 10, 20, and 30 seconds, followed by aqueous extraction at 80 °C. The resulting extracts were spray-dried into powders at 100 °C. Both aqueous extracts and powders were analyzed for their composition (proteins, carbohydrates, phenolic compounds) and antioxidant activity.
Results	Moderate microwave treatment (180 W for 20–30 seconds) enhanced the extraction of proteins and carbohydrates while preserving high antioxidant activity. The resulting powders retained up to 90.96% of proteins, 95.81% of carbohydrates, and 74.91% of phenolic compounds, with only minimal losses in antioxidant activity after six months of storage.
Conclusions	These findings demonstrate that microwave treatment of <i>Porphyridium cruentum</i> biomass, followed by aqueous extraction and spray drying, is an effective strategy for obtaining and preserving microalgal bioactive compounds. This approach supports their potential applications in the pharmaceutical, nutraceutical, and cosmetic fields.
Keywords	<i>Porphyridium cruentum</i> , microwave-assisted pretreatment, bioactive composition, spray drying, powder stability.

EFFECTUL TRATAMENTULUI ASISTAT DE MICROUNDE ASUPRA COMPOZIȚIEI EXTRACTULUI DE *PORPHYRIDIVM CRUENTUM* USCAT PRIN PULVERIZARE

Introducere	Microalgele reprezintă o sursă valoroasă de produse naturale cu proprietăți bioactive, având aplicații în biomedicină, industria farmaceutică, nutriție și cosmetică. Microalga roșie <i>Porphyridium cruentum</i> se remarcă prin conținutul său bogat în biomolecule cu efecte antioxidante, antiinflamatoare și imunomodulatoare. Pentru valorificarea optimă a acestor biomolecule, este esențială aplicarea unor metode eficiente de extracție și conservare.
Material și metode	Biomasa de <i>Porphyridium cruentum</i> a fost tratată cu microunde (MW) la 180W, 300W și 450W pe durata a 10, 20 și 30 sec, apoi supusă extracției hidrice la 80°C. Pulberile au fost obținute prin atomizare la 100°C. În extracțele hidrice și în pulberile obținute s-a evaluat compoziția bioactivă (proteine, carbohidrați, compuși fenolici) și activitatea antioxidantă (teste ABTS și DPPH).
Rezultate	Tratamentul moderat cu microunde (180W timp de 20–30 sec) a favorizat extracția proteinelor și carbohidraților, menținând o activitate antioxidantă ridicată. Pulberile obținute au păstrat până la 90.96% din proteine, 95.81% din carbohidrați și 74.91% din compușii fenolici, cu pierderi minore ale activității antioxidante, după șase luni de păstrare.
Concluzii	Rezultatele cercetării demonstrează că tratamentul biomasei de <i>Porphyridium cruentum</i> cu microunde, urmat de extracția hidrică și uscarea extractului prin pulverizare, reprezintă o strategie eficientă pentru obținerea și conservarea compușilor bioactivi microalgali, facilitând utilizarea acestora în domeniul farmaceutic, nutraceutic și cosmetic.
Cuvinte-cheie	<i>Porphyridium cruentum</i> , tratare asistată de microunde, compoziția bioactivă, uscare prin atomizare, stabilitatea pulberii.

INTRODUCTION

Microalgae are a valuable source of natural products, characterized by their diverse bioactive composition. They have important applications across multiple fields, including biomedicine, the pharmaceutical industry, nutrition, and cosmetics (1). In addition, they contribute to sustainable and cost-effective solutions in health, food, and environmental protection (2–4).

The red microalga *Porphyridium cruentum* is a significant source of biologically active compounds with notable properties, including sulfated polysaccharides, polyunsaturated fatty acids, and functional proteins (5–7). The sulfated polysaccharides of *Porphyridium cruentum* exhibit antioxidant, anti-inflammatory, antimicrobial, and immunomodulatory effects. They stimulate the immune system and show antiviral activity by inhibiting viral replication and protecting host cells. Due to these properties, they are being investigated as complementary therapies for infections and autoimmune diseases (8). Polyunsaturated fatty acids, such as arachidonic acid and eicosapentaenoic acid, are essential for cardiovascular health and lipid metabolism, which makes them valuable in nutraceuticals and functional foods (9). The proteins of *Porphyridium cruentum* provide essential amino acids and bioactive peptides with antioxidant and immunostimulatory roles. These contribute to cellular health and may be applied in protein supplements with nutritional and therapeutic benefits.

Porphyridium cruentum is highly valued in the cosmetic industry for its exopolysaccharides, which provide hydration, antioxidant protection, and promote skin regeneration. Its proteins and polyunsaturated fatty acids further support skin elasticity and exert anti-aging effects, making them key ingredients in creams and skin care formulations (10).

Optimizing the use of *P. cruentum* bioactive compounds requires advanced biomass processing and extraction techniques. Among these, microwave-assisted extraction is particularly effective, as it achieves high yields while preserving the structural integrity of active molecules (11). Spray drying has also proven to be an efficient method for stabilizing and preserving extracts. By incorporating bioactive compounds into a dry matrix, it minimizes the risk of oxidation and degradation (12). This approach improves stability, extends shelf life, and facilitates both long-term storage and international distribution (13). In addition to protecting active molecules, these techniques enhance bioavailability, enabling straightforward integration into pharmaceutical, food, and cosmetic products. Modern processing and extraction technologies are therefore essential for converting microalgal bioactives into stable, valuable, and widely applicable industrial ingredients.

The purpose of this study is to evaluate the effect of microwave-assisted treatment of *P. cruentum* biomass on the bioactive composition of aqueous extracts and to assess their stability following spray drying.

MATERIAL AND METHODS

The study used the red microalga *Porphyridium cruentum* strain CNMN-AR-01, which is deposited in the National Collection of Non-Pathogenic Microorganisms at the Institute of Microbiology and Biotechnology, Technical University of Moldova. Biomass was produced by cultivating the microalga in Brody mineral medium. Cultivation took place in 1000 mL Erlenmeyer flasks containing 500 mL of medium under the following conditions: a temperature of 25–28°C, a pH of 6.8–7.2, and continuous illumination at 56 µmol

photons $\text{m}^{-2} \text{s}^{-1}$. After a 14-day cultivation cycle, the biomass was separated from the medium by centrifugation at 4000 rpm for 7 min (NÜVE NF-800, Ankara, Turkey). Excess salts were removed by washing with a 2.0% ammonium acetate solution. The biomass was then standardized to a concentration of 10 mg/mL for subsequent treatment and extraction.

Three types of raw material were prepared: native biomass (NBM) and frozen/thawed biomass (FTBM), both used as controls, and microwave-treated biomass, used as the experimental variant. For FTBM, biomass was frozen at -20°C (Snaigė AB, Alytus, Lithuania) and thawed at room temperature. This procedure was repeated six times, after which phycobiliproteins were removed by centrifugation at 4000 rpm for 7 min. For the experimental microwave treatment, 20 mL aliquots of standardized biomass (10 mg/mL) were irradiated in a Samsung microwave device (2450 MHz, Seoul, South Korea). The aliquots were exposed at power levels of 180W and 300W for 10, 20, and 30 seconds each. At 450W, exposure times were 10 and 20 seconds. Following microwave exposure, the biomass was centrifuged to separate the phycobiliproteins.

Bioactive compounds were extracted from control and experimental biomass samples with purified water at a 1:40 (g/mL) ratio. The mixture was homogenized and incubated at 80°C for 60 minutes in a water bath (GFL 1023, Burgwedel, Germany). After cooling to room temperature, the mixture was centrifuged at 4000 rpm for 7 minutes to separate the solid fraction. The supernatant, representing the crude aqueous extract, was collected, filtered to remove residual particles, and stored at -20°C .

Biochemical assays were performed using a UV-Vis spectrophotometer (80T, PG Instruments Ltd., Lutterworth, UK). The protein content of the microalgae biomass was determined by a modified Lowry method, and carbohydrate content was quantified using the anthrone reagent. Phenolic compounds were assessed with a modified Folin-Ciocalteu method. All results are expressed as g/100 g dry weight (DW). Antioxidant activity was evaluated using the 2,2'-azinobis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) assays (14), with results expressed as the percentage of radical inhibition per 10 mg DW. All reagents were purchased from Sigma-Aldrich Chemie GmbH (Taufkirchen, Germany).

Spray drying was performed using a UnoPex laboratory-scale spray dryer (Izmir, Turkey) with an integrated air compressor. The process was conducted at a constant inlet temperature of 100°C with extract solutions prepared at a 12% solid concentration. The resulting powders were collected and analyzed to evaluate the retention of biochemical components relative to the initial extracts.

To monitor stability, the spray-dried powders were stored in hermetically sealed, dark glass containers with silica gel stoppers at 4°C for 6 months to prevent light exposure and moisture absorption. For biochemical analysis, the powders were rehydrated in deionized water (1:10 m/v), homogenized for 30 minutes at room temperature, and centrifuged at 10,000 rpm for 5 minutes.

Statistical analyses were performed in three settings: (i) microwave-treated biomass extracts compared with controls (native biomass, NBM, and freeze-thawed biomass, FTBM); (ii) spray-dried powders compared with their corresponding aqueous extracts to assess compound retention; and (iii) powders stored for 1, 3, and 6 months compared with baseline values to evaluate biochemical stability over time. Results are reported as Mean $\pm$ Standard Deviation (SD). The percentages of protein, carbohydrates, and phenols, calculated on a dry weight, extract, or powder basis, were ana-

lyzed. Comparisons were made using Student's *t*-test. Statistical significance was defined as $p < 0.05$ at the 95% confidence interval (CI). All analyses were conducted in Microsoft Excel.

RESULTS

Figure 1 presents a detailed analysis of the percentage composition of proteins, carbohydrates, and phenolic compounds in aqueous extracts of *Porphyridium cruentum* biomass, depending on the processing method (microwave treatment of native biomass or repeated freeze-thaw cycles).

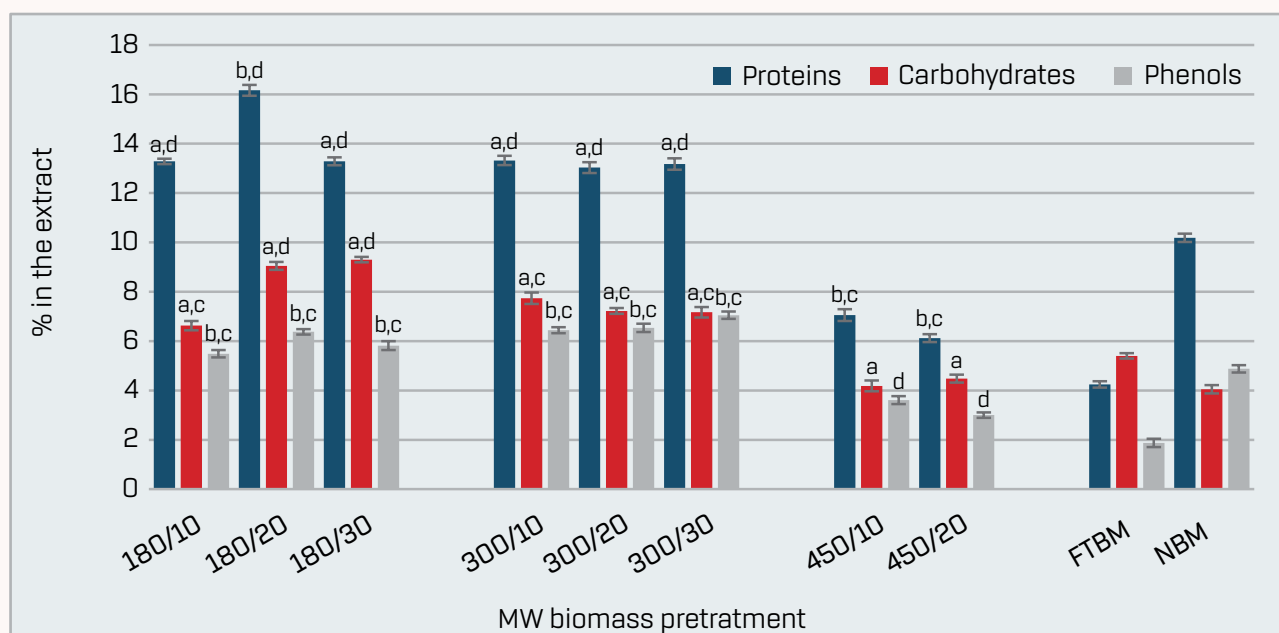


Figure 1. Composition of the aqueous extract obtained from *Porphyridium cruentum* biomass treated with MW (W/sec). Control: NBM - native biomass and FTBM-freeze-thaw biomass;

a, b: significantly different from NBM at $p < 0.01$ and $p < 0.001$, respectively;

c, d: significantly different from FTBM at $p < 0.01$ and $p < 0.001$, respectively.

The highest protein content (16.16%) was obtained from biomass treated with microwaves at 180 W for 20 seconds, corresponding to a 36.9% increase ($p < 0.001$) compared with untreated biomass and a 3.8-fold increase ($p < 0.001$) compared with biomass subjected to repeated freezing/thawing (10.19% and 4.25%, respectively). Extracts from biomass treated with microwaves at 300 W contained protein levels similar to those observed at 180 W with exposure times of 10 and 30 seconds. These values were 22.82% higher ($p < 0.01$) than in untreated biomass and 3.1-fold higher ($p < 0.001$) than after repeated freezing/thawing. In contrast, treatment of *Porphyridium* biomass at 450 W significantly reduced protein content, which was 44.38% lower ($p < 0.01$) than in untreated biomass.

The highest carbohydrate levels were obtained in extracts from biomass treated at 180 W for 20 or 30 seconds, ranging from 9.05% to 9.30%. These values were 2.2–2.3 times higher than in untreated biomass ($p < 0.001$) and 1.7 times higher than in frozen/thawed biomass ($p < 0.001$). At 300 W, microwave exposure time had no significant influence, and carbohydrate levels remained between 7.17% and 7.74%. In contrast, treatment at 450 W significantly reduced carbohydrate content, yielding values similar to those observed in untreated or frozen/thawed biomass.

The highest phenolic concentration was achieved in extracts from biomass treated at 300 W for 30 seconds, reaching 7.05%. In untreated biomass, the phenolic content was 4.88%, while frozen/thawed biomass contained significantly less ($p < 0.001$), at 1.88%. Treatment at 180 W resulted in values between 5.48% and 6.38%, indicating effective extraction. However, treatment at 450 W markedly reduced phenolic levels, with a maximum of only 3.61%.

Figure 2 presents the antioxidant activity (ABTS and DPPH assays) of aqueous extracts from *Porphyridium* biomass treated with microwave-assisted technology. This treatment significantly increases the extracts' capacity to neutralize free radicals, suggesting that process parameters may affect antioxidant efficiency.

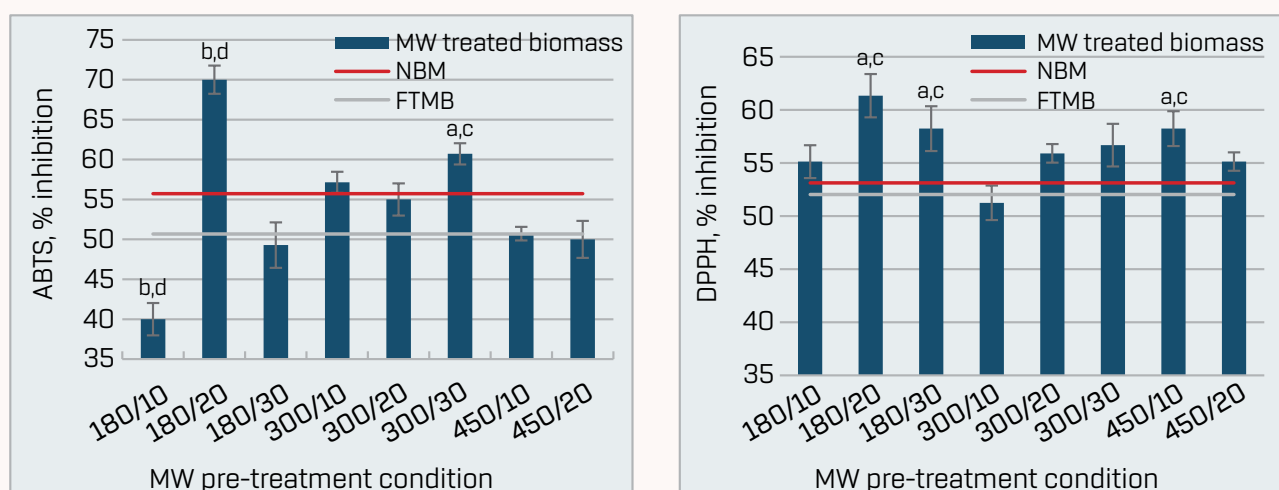


Figure 2. Antioxidant activity of the aqueous extract obtained from *Porphyridium cruentum* biomass treated with microwaves (W/sec). Control: NBM - native biomass and FTMB - freeze-thaw biomass.

a, b: significantly different from NBM at $p < 0.01$ and $p < 0.001$, respectively;
c, d: significantly different from FTMB at $p < 0.01$ and $p < 0.001$, respectively.

The highest antioxidant activity, as determined by the ABTS assay, was observed in extracts from biomass treated at 180 W for 20 seconds, showing 70.0% inhibition. Treatment at 300 W for 10 and 30 seconds resulted in ABTS inhibition levels of 57.1% and 60.7%, respectively. The lowest antioxidant activity, 40.0% inhibition, was obtained in extracts from biomass treated at 180 W for 10 seconds.

The DPPH assay revealed maximum antioxidant activity of 61.3% inhibition in extracts from biomass treated at 180 W for 20 seconds. The lowest activity, 51.2% inhibition, was recorded in extracts from biomass treated at 300 W for 10 seconds.

Extracts from native biomass demonstrated inhibition levels of 50.7% for ABTS and 52.0% for DPPH. In comparison, extracts from frozen/thawed biomass exhibited 55.7% ABTS inhibition and 53.6% DPPH inhibition.

Table 1 summarizes the protein, carbohydrate, phenolic content, and antioxidant activity of the spray-dried powder in comparison with the aqueous extracts obtained from *Porphyridium cruentum* biomass processed by different techniques.

Table 1. Composition of the spray-dried powder in comparison to the aqueous extracts from *Porphyridium cruentum* biomass, treated using various techniques.

Biomass treatment variant	Bioactive compound	Compound/activity in the extract	Compound/activity in powder	Recovering after drying
MW-180W/20 sec	Proteins (g/100g DW)	16.16±0.22	14.38±0.30*	88.99
	Carbohydrates (g/100 DW)	9.05±0.16	8.73±0.80	96.46
	Phenols (g/100g DW)	6.38±0.11	5.37±0.31**	84.17
	ABTS (% Inhibition)	70.00±3.2	66.52±3.4	95.03
	DPPH (% Inhibition)	61.33±4.1	54.55±2.5	88.95
MW-180W/30 sec	Proteins (g/100g DW)	13.28±0.16	12.08±0.80*	90.96
	Carbohydrates (g/100g DW)	9.30±0.11	8.91±0.84	95.81
	Phenols (g/100g DW)	5.82±0.18	4.36±0.22*	74.91
	ABTS (% Inhibition)	49.30±4.2	46.13±3.3	93.57
	DPPH (% Inhibition)	58.23±1.5	48.33±2.4*	82.99
MW-300W/30 sec	Proteins (g/100g DW)	13.17±0.23	11.58±0.52*	87.93
	Carbohydrates (g/100g DW)	7.17±0.21	6.82±0.85	95.12
	Phenols (g/100g DW)	7.05±0.15	5.54±0.21**	78.58
	ABTS (% Inhibition)	60.70±3.8	56.08±2.5*	92.39
	DPPH (% Inhibition)	56.60±3.1	50.77±2.8	89.70
NBM	Proteins (g/100g DW)	10.19±0.17	8.66±0.63*	84.99
	Carbohydrates (g/100g DW)	4.05±0.17	3.93±1.14	97.04
	Phenols (g/100g DW)	4.88±0.15	4.52±0.33	92.62
	ABTS (% Inhibition)	50.68 ±3.8	45.83±3.1	90.43
	DPPH (% Inhibition)	52.02±4.1	46.15±2.6	88.72
FTBM	Proteins (g/100g DW)	4.25±0.13	3.55±0.18*	83.53
	Carbohydrates (g/100g DW)	5.40±0.11	5.13±0.84	95.00
	Phenols (g/100g DW)	1.87±0.17	1.47±0.22*	78.61
	ABTS (% Inhibition)	55.71±2.5	50.11±3.3*	89.58
	DPPH (% Inhibition)	53.12±3.4	48.66±4.1	91.60

Data are presented as Mean ± (SD) of three independent experiments. The asterisks indicate statistically significant differences (* $p < 0.05$; ** $p < 0.01$) between spray-dried powders and their corresponding aqueous extracts (Student's t-test). MW: microwave-treated biomass; NBM: native biomass; FTBM: freeze-thaw biomass.

Following the spray-drying process, carbohydrates exhibited the lowest losses, ranging from 3.54% to 5.00%. In contrast, proteins showed significantly higher losses, between 9.03% ($p < 0.05$) and 16.47% ($p < 0.05$). In extracts obtained from biomass subjected to microwave treatment, protein losses were comparatively lower, ranging from 11.01% ($p < 0.05$) to 12.07% ($p < 0.05$). Phenolic compounds were the most affected, with losses in powders ranging from 15.83% ($p < 0.05$) to 25.09% ($p < 0.01$). Antioxidant capacity was also reduced: DPPH radical reduction decreased by 8.40%–17.01% ($p < 0.05$), while ABTS radical reduction declined by 4.94%–10.05% ($p < 0.05$).

Notably, proteins in powders from biomass treated with microwaves at 180W for 20 and 30 seconds retained 88.99% and 90.96% of their bioactive components, corresponding to losses of 11.01% ($p < 0.05$) and 9.04% ($p < 0.05$), respectively. The ABTS radical inhibition capacity of these powders decreased only moderately, by 4.97% and 6.43%, indicating good stability of the antioxidant compounds during spray drying. In contrast, losses were more pronounced in powders from extracts of biomass treated at 300W for 30 seconds. Here, the phenolic content decreased by 21.42% ($p < 0.01$), and the ABTS inhibition capacity decreased by 7.61% ($p < 0.05$). The powder from the native biomass best preserved carbohydrates, retaining 97.04% of the content found in the

aqueous extract prior to spray drying, which was a minimal loss of 2.96%. However, this powder showed a 15.01% ($p<0.05$) reduction in protein content. The highest losses were observed in the powder derived from the aqueous extract of frozen/thawed biomass, with phenolic compounds decreasing by 21.39% ($p<0.05$) and proteins by 16.47% ($p<0.05$).

The powders exhibiting the highest biological value were selected for a stability study over 1, 3, and 6 months. These included powders from biomass extracts treated with microwave-assisted technology at 180 W for 20 and 30 seconds and at 300 W for 30 seconds. Powders from native and frozen/thawed biomass extracts served as reference samples. Table 2 presents the results of the stability study, showing the biochemical composition and antioxidant activity of the powders derived from aqueous extracts of *Porphyridium cruentum* biomass treated using different techniques over a storage period of 1, 3, and 6 months.

Table 2. Stability of the powder from *Porphyridium cruentum* biomass during 1, 3, and 6 months of storage.

Biomass treatment variant	Bioactive compound	Compound/ activity in the initial powder	Weight of the compound after storage, % of initial content			Compound/ activity in powder
			1 month	3 months	6 months	
MW-180W/20 sec	Proteins (g/100g DW)	14.38±0.30	98.33	97.50	96.11	13.82±0.16
	Carbohydrates (g/100g DW)	8.73±0.80	94.16	93.13	91.87	8.02±0.18
	Phenols (g/100g DW)	5.37±0.31	98.14	97.21	93.67	5.03±0.31
	ABTS (% Inhibition)	66.52±3.4	98.38	97.75	97.11	64.60±2.6
	DPPH (% Inhibition)	54.55±2.5	98.92	98.00	98.45	54.05±3.2
MW-180W/30 sec	Proteins (g/100g DW)	12.08±0.80	98.25	98.34	96.52	11.46±0.11
	Carbohydrates (g/100g DW)	8.91±0.84	97.87	95.96	94.61	8.43±0.22
	Phenols (g/100g DW)	4.36±0.22	93.12	91.97	86.47	3.77±0.27
	ABTS (% Inhibition)	46.13±3.3	98.29	97.79	96.60	44.56±2.6
	DPPH (% Inhibition)	48.33±2.4	98.40	99.03	97.74	47.24±4.1
MW-300W/30 sec	Proteins (g/100g DW)	11.58±0.52	97.06	95.94	93.61	10.84±0.21
	Carbohydrates (g/100g DW)	6.82±0.85	97.51	95.75	92.82	6.33±0.17
	Phenols (g/100g DW)	5.54±0.21	96.03	91.34	82.49	4.57±0.44
	ABTS (% Inhibition)	56.08±2.5	97.86	96.50	95.44	53.52±5.2
	DPPH (% Inhibition)	50.77±2.8	97.85	97.45	96.44	48.96±3.1
NBM	Proteins (g/100g DW)	8.66±0.63	97.23	96.30	94.92	8.22±0.17
	Carbohydrates (g/100g DW)	3.93±1.14	97.96	94.40	91.09	3.58±0.15
	Phenols (g/100g DW)	4.52±0.33	95.35	88.72	73.45	3.32±0.22
	ABTS (% Inhibition)	45.83±3.1	95.51	94.85	94.55	43.33±3.6
	DPPH (% Inhibition)	46.15±2.6	97.83	97.53	95.67	44.15±4.1
FTBM	Proteins (g/100g DW)	3.85±0.18	88.73	86.20	98.03	3.48±0.18
	Carbohydrates (g/100g DW)	5.13±0.84	98.64	94.74	94.74	4.86±0.21
	Phenols (g/100g DW)	1.47±0.22	97.96	76.87	76.87	1.13±0.22
	ABTS (% Inhibition)	50.11±3.3	98.12	97.29	97.29	48.75±3.4
	DPPH (% Inhibition)	48.66±4.1	94.57	93.88	93.88	45.68±4.2

Data are presented as Mean ± (SD). MW- microwave-treated biomass; NBM - native biomass; FTBM - freeze-thaw biomass.

The highest protein stability after 6 months of storage was observed in powders obtained from biomass extracts treated using microwave-assisted technology at 180W for 20 seconds (96.11% stability) and 180W for 30 seconds (96.52% stability).

Carbohydrate stability remained high (>91%) over 6 months in all microwave-treated samples. The highest stability was in the untreated biomass powder, at 97.96% after 1 month and 91.09% after 6 months.

Antioxidant activity was largely preserved, remaining >95% in most variants after 6 months. In contrast, powders from biomass treated at 300W for 30 seconds and from frozen/thawed biomass exhibited significant losses of bioactive compounds. This was attributed primarily to a reduction in phenolic compounds, which declined by 17.51% to 23.16% ($p < 0.05$) during storage.

DISCUSSIONS

Microwave-assisted technology is an efficient and environmentally friendly method for releasing valuable components from microalgal biomass, including lipids, pigments, carbohydrates, vitamins, and proteins, either individually or as part of bioactive extract complexes (15, 16).

In this study, the intensity and duration of microwave treatment significantly influenced the biochemical composition of the aqueous extracts. Treating *Porphyridium* biomass at a lower power with a moderate exposure time (e.g., 180W for 20 sec) increased the protein and carbohydrate content compared to untreated or repeatedly frozen/thawed biomass. In contrast, more aggressive treatments (450W for 10 sec and 450W for 20 sec) substantially reduced these compounds, suggesting potential thermal degradation. Previous studies support the efficacy of microwave treatment for this purpose. For instance, one study demonstrated its efficiency in extracting proteins from *Chlorella vulgaris* using 600W for 20 seconds (17). Similarly, microwave-assisted extraction has been used to obtain carbohydrates from *Scenedesmus* sp., with the highest yield achieved at 1075W under direct acid extraction conditions for 22 minutes (18).

The analysis of phenolic content revealed that these compounds varied significantly in aqueous extracts depending on the treatment, with the maximum values observed in biomass treated at 300W for 30 seconds. This suggests that microwave-assisted extraction enhances the release of these metabolites. A similar finding was reported in a study of *Chlorella vulgaris* biomass, where a 300W treatment for 14 minutes was optimal for extracting phenolic compounds into ethanol (19). Therefore, microwave-assisted technology can optimize the yield of bioactive compounds, depending on the extraction method and intended application.

Antioxidant assays (ABTS and DPPH) indicated that biomass treated at 180W/20 sec exhibited the highest inhibition, suggesting this condition optimally releases antioxidant compounds into aqueous extracts. Conversely, more intense microwave treatments (450W/10 sec and 450W/20 sec) reduced antioxidant activity, likely due to the degradation of active metabolites. This is consistent with reports that optimized microwave-assisted extraction enhances antioxidant activity in microalgae; for example, a power of 380W yielded a significant DPPH inhibition of 17.58% in *Chlorella vulgaris* (20). Thus, moderate microwave treatment of *Porphyridium cruentum* biomass produced extracts with superior antioxidant activity compared to those from native (untreated) or repeatedly frozen/thawed biomass. These results confirm that the strategic selection of microwave power and exposure time is crucial for enhancing the bioactive potential of the aqueous extracts.

Spray drying effectively transforms microalgal biomass into fine, easily dispersible powders, making it a popular technique for various applications (3). In a study on *Scenedesmus acuminatus*, the loss of pigments during processing was influenced by the inlet and outlet air temperatures, as well as the

suspension's solid content. Despite this, the method did not significantly alter the content of proteins, carbohydrates, or lipids, demonstrating its overall effectiveness for preserving these key bioactive compounds (12).

In this study, the atomization of aqueous extracts from *Porphyridium cruentum* biomass caused only minimal losses in bioactive compounds, with maximum reductions of 11% for proteins, 5% for carbohydrates, and 39% for phenolic compounds. These losses were influenced by the biomass pretreatment. Extracts from repeatedly frozen-thawed, native, and 450W microwave-treated biomass showed the greatest degradation. The best results were obtained with powders from aqueous extracts of biomass treated with 180W microwaves for 20 and 30 seconds. These powders provided a balance between protein preservation (losses of 9.04–11.01%) and carbohydrate retention (losses of 2.96–4.19%), alongside minor losses in antioxidant activity (4.97–7.61%). For comparison, one study demonstrated that the bioactive stability of *Chlamydomonas reinhardtii* powder containing a recombinant protein was highly dependent on storage temperature. The protein was relatively stable at -80°C, losing approximately 38% over 27 months. Degradation was more pronounced at +4°C, with a 50% loss, and severe at room temperature, where a 92% loss was recorded, indicating this condition is unsuitable for long-term storage (21).

Therefore, an efficient strategy for preserving bioactive compounds from the red microalga *Porphyridium cruentum* is to use moderate-intensity microwave-assisted technology, followed by extraction and spray drying. This approach enhances the stability of the final product, and optimizing the process parameters can ensure both bioactive efficacy and long-term stability.

CONCLUSIONS

1. Microwave-assisted treatment significantly alters the extraction process and biochemical composition of aqueous extracts from the red microalga *Porphyridium cruentum*. A moderate regimen of 180W for 20–30 seconds increases the yield of proteins and carbohydrates, while a more intense treatment at 450W leads to their degradation.
2. Treating *Porphyridium cruentum* biomass with microwaves at 300W for 30 seconds enhances the extraction of phenolic compounds. Antioxidant activity is also influenced by the pretreatment parameters, peaking in extracts derived from biomass treated at 180W for 20 seconds.
3. Spray drying effectively preserved proteins and carbohydrates, with minimal losses of 9.04–11.01% and 2.96–4.19%, respectively. Phenolic compounds, however, were the most vulnerable, with losses of 15.83–25.09%.
4. After six months of storage, the powders retained a stable bioactive composition, with only a minimal reduction in antioxidant activity (4.97–7.61%). These results confirm that spray drying is an efficient method for preserving bioactive compounds from the red microalga *Porphyridium cruentum*.

CONFLICT OF INTEREST The authors deny any conflict of interest in the publication of this material.

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