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Comparative evaluation of amino acid profiles, fatty acid compositions, and nutritional value of two varieties of head water *Porphyra yezoensis*: "Jianghaida No. 1" and "Sutong No.1"

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ABSTRACT

Comparative nutritional analysis of *Porphyra yezoensis* strains "Jianghai No. 1" and "Sutong No.1" revealed significant differences in crude protein, crude fat, crude fiber, crude ash, and total sugar. Both strains contained 16 amino acids, with alanine as the highest and histidine the lowest content. Methionine was determined to be the first limiting amino acid for both strains in both amino acid score and chemical score assessment. They also featured 24 fatty acids, differing notably in four saturated fatty acids and five unsaturated fatty acids. All 12 mineral elements were present, notably differing in sodium, magnesium, potassium, calcium, iron, and zinc. The "Jianghai No. 1" strain stands out with its nutrient-rich profile, featuring high protein content, low fat, and abundant minerals, which could potentially command higher market prices and generate greater economic benefits due to its superior nutritional, and set a strong foundation for its future large-scale promotion and cultivation.

1. Introduction

Macroalgae are recognized as a nutritious food due to their composition and health benefits (Shannon & Abu-Ghannam, 2019). While Asian nations have long used seaweeds as food, Western countries primarily employ them in the food, pharmaceutical, and cosmetic industries as gelling agents and colloids (Peñalver et al., 2020). Seaweeds offer essential nutrients like proteins, vitamins, minerals, and dietary fiber. Red algae boast higher protein content (average 18.8 g/100 g) compared to green and brown varieties (Gamero-Vega et al., 2020). However, essential amino acid levels differ among types, with brown algae ranging from 22% to 44%, green algae from 26% to 32%, and red algae from 14% to 19% (Fleurence, 1999). Notably, red algae contain essential fatty acids like linolenic and linoleic acids, with concentrations reaching up to 22% and 11%, respectively. They also exhibit higher proportions of eicosapentaenoic (EPA) and docosahexaenoic (DHA) fatty acids, comprising up to 48.4% of total fatty acids (Gamero-Vega et al., 2020). The health properties of seaweed are attributed to compounds like polyphenols, polysaccharides, sterols, and other bioactive molecules, known for their antioxidant, anti-inflammatory, anti-cancer, and anti-diabetic attributes (Gamero-Vega et al., 2020; Jiao et al., 2019; Wang et al., 2021). Macroalgae are rich sources of essential minerals like Mg, Fe, Mn, Cu, Zn, Ca, and K, making them valuable for food fortification (Lozano Muñoz & Díaz, 2020; Meng et al., 2022). These marine vegetables have high mineral content, often exceeding that of terrestrial plants, and can contribute significantly to daily mineral intake (Guo et al., 2022). Additionally, macroalgae are indeed an excellent source of various vitamins, including vitamin A, C, E and K, and several B-vitamins (such as thiamine, riboflavin, niacin, folate, and vitamin B12) (Drăgan et al., 2023; Guo et al., 2022; Reddy et al., 2023). These minerals and vitamins also play crucial roles in maintaining human health and are beneficial for various bodily functions. Hence, algae are seen as

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Abbreviations: TAA, total amino acids; EAA, essential amino acids; NEAA, non-essential amino acids; UAA, umami amino acids; SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids.

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a valuable natural source due to their diverse biological activities and potential use as functional ingredients in various technological applications to create functional foods.

Porphyra yezoensis, commonly known as nori, falls under the Rhodophyta division and is prized for its nutritional and economic value (Yang et al., 2022). Esteemed for its delightful taste and nutrient richness, it enjoys popularity in many countries (Li et al., 2017). Researchers have isolated and purified polysaccharides from P. yezoensis, revealing their immunomodulatory and antioxidant activities (Wang et al., 2021). Antimicrobial peptides with potent activity were also identified from this seaweed (Jiao et al., 2019). However, recent expansion in P. yezoensis farming has faced challenges like harmful algae (such as Ulva prolifera, Sargassum horneri, naviculoid diatom species, and other algal species.) and pathogenic diseases (such as abnormal leafy thalli, porphyra red rot disease, and porphyra filamentous thalli yellow blotch disease), leading to reduced yields (Wang et al., 2020). Hence, the search for nutrient-rich *P. yezoensis* strains with adaptability to adverse conditions and robust growth potential becomes crucial. There are two new strains of P. yezoensis ("Jianghaida No. 1" and "Sutong No.1") artificially selected by different organizations that have caught our attention. These two new strains not only grow fast and have strong disease resistance, but also have good adaptability for cultivation. This study compares and evaluates amino acid profiles, fatty acid compositions, and nutritional value of these two strains of P. yezoensis that with excellent culture performance. It will provide a foundation for enhancing the nutritional quality of P. yezoensis products and addressing the evolving consumer demands, facilitating future large-scale promotion and cultivation endeavors.

2. Materials and methods

2.1. Experimental materials

"Jianghaida No. 1" *P. yezoensis* is a novel strain derived from indigenous wild *P. yezoensis* found in Lianyungang, following numerous generations of cultivation. On the other hand, "Sutong No. 1" *P. yezoensis* was collected from a specific cultivation zone located in Ganyu District, within Lianyungang City, Jiangsu Province. Postharvest, the samples underwent multiple rinses in seawater, drying in an oven at 60 °C for 24 h until achieving a consistent weight, grinding using a high-speed grinder, and subsequent storage in dry culture dishes for future use.

2.2. Routine nutrient content determination

The assessment of crude ash employs the incineration method. Crude protein determination follows the Kjeldahl method. The measurement of crude fat involves the Soxhlet extraction method (Liu et al., 2018). Lastly, the determination of crude fiber is conducted through the acid-alkali digestion method as per GB/T 5009.10–2003.

2.3. Amino acid composition determination

According to the high-performance liquid chromatography method outlined in GB/T 30987–2020, the percentage quantification involves the area normalization method.

To execute this method, take an appropriate sample quantity into a hydrolysis tube, add 20 mL of 1 + 1 HCL, and place it in an oven at 110 °C for 22–24 h. After cooling, extract 100 μ L of the clear liquid, vacuum dry it in an oven, derivatize it under nitrogen protection, adjust to 0.5 mL, and filter before analysis. Determination of amino acid composition by high performance liquid chromatography (HPLC) (Agilent 1260, Agilent , USA), and the specific detection method is as follows:

- (1) Chromatographic Column: C18 SHISEIDO (4.6 mm \times 250 mm \times 5 $\mu m);$
- (2) Injection Volume: 10 µL;
- (3) Column Temperature: 40 °C;
- (4) Wavelength: 254 nm;
- (5) Mobile Phase: A, 0.1 M Anhydrous Sodium Acetate aqueous solution (97%) mixed with Acetonitrile (3%), adjusted to pH 6.5; B: Acetonitrile: Water = 80: 20 (ν/v).

The free amino acid content is calculated according to the following formula: $W{=}^{\underline{(C-C_0)^*V^*N}}_{\underline{m}}$

W - is the content of the target substance in the sample (mg/kg);

C - is the concentration of the target substance in the sample determination solution (mg/L);

 C_0 - is the concentration of the target substance in the blank control (mg/L);

V - is the volume of the preparation (mL);

N - is the dilution factor;

m - is the amount of the sample (g).

2.4. Fatty acid composition determination

In accordance with the internal standard method outlined in GB 5009.168–2016 for determining fatty acids, and the analysis of fatty acid composition was conducted using gas chromatography–mass spectrometry (GC–MS) instrument (Trace1310 ISQ, Thermo, USA).

The specific experimental procedures are as follows: First step for sample preparation, weigh the sample, add 95% ethanol and water, mix thoroughly, then add 10 mL of 8.3 mol/L hydrochloric acid for hydrolysis. Second step for hydrolysis, mix homogenized sample with pyrogallic acid, zeolite, 95% ethanol, and 10 mL HCl. Heat at 70-80 °C for 40 min with intermittent shaking. Cool to room temperature. Third step for fat extraction, add 95% ethanol and ether/petroleum ether solution. Shake, allow separation, collect ether layer. Repeat extraction thrice. Evaporate solvent and dry extract at 100 \pm 5 °C for 2 h. Fourth step for saponification and methylation of fat, add 2% NaOH/methanol solution, heat at 85 °C for 30 min. Add 14% BF3/methanol solution, heat 85 °C for 30 min. Cool, extract with n-hexane, filter through 0.45 µm membrane. Fifth step for analysis of fatty acid methyl esters, analyze the fatty acid methyl esters using the GC-MS conditions below, matching spectra against the National Institute of Standards and Technology (NIST) database (https://www.nist.gov/). Finnaly, the percentage of each fatty acid in the sample was calculated using the area normalization method.

The GC–MS analysis condition was shown below:

- (1) Chromatography column: HP-88 (100 m \times 0.25 mm \times 0.20 μ m);
- (2) Heating procedure: hold at 100 °C for 15 min, increase to 190 °C at 15 °C/min, hold for 25 min, then increase to 235 °C at 2.5 °C/min, hold for 4 min;
- (3) Injector temperature: 240 °C; Carrier gas flow rate: 1.0 mL/min;
- (4) Shunt ratio: no shunt;
- (5) Mass spectrometry conditions: ion source temperature 280 $^\circ\text{C};$
- (6) Transfer line temperature: 280 $^{\circ}$ C;
- (7) Solvent delay time: 5.00 min;
- (8) Ion source: Electron ionization source 70 eV.

Calculate the content of each fatty acid in the sample according to the formula: $W = \frac{C^* V^* M}{m} * K \cdot 10^{-4}$.

"W" refers to the content of each fatty acid present in the sample, measured in milligrams per kilogram (mg/kg);

"C" refers to the concentration of fatty acid methyl ester within the sample test liquid, measured in milligrams per liter (mg/L);

"V" refers to the volume of the fixed container, in milliliters (mL);

"k" refers to the conversion coefficient of each fatty acid methyl ester to fatty acid;

"N" refers to the dilution factor;

" 10^{-4} " refers to the unit conversion coefficient; "m" refers to the mass of the sample (g).

2.5. Mineral content determination

The content of 12 inorganic elements, including Fe, Zn, Se, Cu, Mn, As, Cd, K, Na, Mg, and P, is determined using the GB 5009.268–2016 method employing inductively coupled plasma mass spectrometry (ICP-MS) instrument (iCAPQ, Thermo, USA) (Tanvir et al., 2020). The specific procedure is as follows: First, an appropriate amount of the sample is weighed into a specially designed polytetrafluoroethylene digestion vessel, and 5 mL nitric acid is added. After pretreatment, the vessel is sealed and heated for digestion. The temperature is gradually increased to 190 °C according to the preset multi-stage heating program and maintained for a period of time until complete digestion, then naturally cooled. Next, the digestion solution is transferred and diluted to volume to prepare the test solution. Finally, the test solution is introduced into the ICP-MS for elemental detection, and the elemental content is calculated according to the formula:

$$W = \frac{(C - C_0) * V * N}{m}.$$

where:

W——the content of the target in the sample, unit: mg/kg;

C——the concentration of the target in the sample measurement solution, unit: mg/L;

 C_0 — the concentration of the target in the blank control, unit: mg/L;

V——Constant volume, unit: mL;

N-dilution factor;

m——the sample size of the sample, unit: g.

2.6. Evaluation of amino acid nutritional value

Referring to evaluation of the nutritional value of pickled Chinese cabbage (Fleurence, 1999), the formulas for amino acid score (AAS) and chemical score (CS) are as follows (a) and (b) and (c):

- (a) AAS = (amino acid content in the detected sample protein, mg·g⁻¹) / (corresponding amino acid content in the FAO/WHO scoring pattern, mg·g⁻¹);
- (b) CS = (amino acid content in the detected sample protein, mg·g⁻¹)
 / (corresponding amino acid content in egg protein, mg·g⁻¹);
- (c) Amino acid content in the detected sample protein $(mg.g^{-1}) = {(content of a certain amino acid in the sample) / (crude protein content in the sample)} × 6.25 × 1000.$

2.7. Data analysis

The data underwent analysis through Excel software (Microsoft, USA) to compute the mean and standard deviation. An independent sample *t*-test was conducted utilizing SPSS Statistics 26.0 software (IBM, USA), where significance was set at P < 0.05, and high significance at P < 0.01. All results are expressed as mean \pm standard error (mean \pm SE).

3. Result

3.1. Routine nutrient analysis

The analysis of the conventional nutritional components of two types of *P. yezoensis*, namely "Jianghaida No.1" and "Sutong No.1", shows that there is a highly significant difference in crude protein, crude fiber, crude ash, and crude fat between individual samples of the two types. The protein and crude fiber content of "Jianghaida No.1" is significantly higher than that of "Su tong No.1" (P < 0.01), while the crude ash and crude fat content of "Su tong No.1" are significantly higher than that of "Jianghaida No.1" (P < 0.01). In addition, there is a significant difference in total sugar content between the two types, with "Sutong No.1" significantly higher than "Jianghaida No.1" (P < 0.05) (Table 1).

3.2. Amino acid composition analysis

Two varieties of *P. yezoensis*, namely "Jianghaida No.1" and "Sutong No.1", were analyzed for their amino acid composition, revealing the presence of 16 amino acids; however, tryptophan was not detected. Among these, 7 are essential amino acids crucial for the human body, while the remaining 9 are non-essential. Both strains exhibit the highest content of alanine, followed by glutamic acid, while histidine demonstrates the lowest concentration. Notably, "Sutong No.1" displays a significantly higher level of glutamic acid in non-essential amino acids compared to "Jianghaida No.1" (P < 0.05). Moreover, no significant difference was observed in the total sum of UAA (Umami Amino Acids) or the ratio of UAA to TAA (Total Amino Acids) between the two strains (P > 0.05) (Table 2).

3.3. Evaluation of amino acid nutritional value

In Table 3, assessed under the AAS model as the benchmark, both "Jianghaida No.1" and "Sutong No.1" strains display the highest scores for valine in essential amino acids, followed by threonine. Conversely, methionine scores the lowest for both strains. Notably, the scores for several amino acids such as leucine, threonine, valine, lysine, phenylalanine, tyrosine, isoleucine, and methionine are significantly higher in "Sutong No.1" compared to "Jianghaida No.1" (P < 0.01).

Under the CS model, as shown in Table 4, "Jianghaida No.1" and "Sutong No.1" *P. yezoensis* both exhibit threonine as the top-ranking amino acid, followed by valine, with methionine scoring the lowest for both strains. Likewise, various amino acids' content in "Sutong No.1" demonstrates a significant elevation compared to "Jianghaida No.1" (P < 0.01).

3.4. Fatty acid composition analysis

The fatty acid analysis of "Jianghaida No. 1" and "Sutong No. 1" *P. yezoensis* (Table 5) unveiled the presence of 24 different fatty acids. Among these, the ratio of C16:0 to saturated fatty acids emerged as the highest, followed by C23:0, while C21:0 exhibited the lowest ratio. In the category of monounsaturated fatty acids, C20:1 displayed the highest ratio, followed by C18:1n-9c. The ratio of C20:5n-3 to poly-unsaturated fatty acids ranked the highest, followed by C22:6n-3 registering the lowest ratio.

There are 10 saturated fatty acids, and the total saturated fatty acid content of "Sutong No.1" is significantly higher than that of "Jianghaida No.1" (P < 0.05). Among them, C16:0 and C12:0 of "Sutong No.1" are significantly higher than those of "Jianghaida No.1" (P < 0.05), and C14:0 and C18:0 are extremely significantly higher than those of

Table 1

Nutrient composition analysis of *P. yezoensis* in "Jianghaida No. 1" and "Sutong No. 1" (%, dry basis).

Variety	Jianghaida No. 1	Sutong No. 1
Crude protein	$41.79\pm0.23~^{\rm A}$	$33.71\pm0.31^{\rm B}$
Total sugar	$29.33 \pm 1.07^{\rm a}$	$32.30\pm0.50^{\rm b}$
Crude fiber	3.13 ± 0.15 $^{ m A}$	$1.93\pm0.06^{\rm B}$
Ash	$4.28\pm0.13~^{\rm A}$	$10.24\pm0.03^{\rm B}$
Crude fat	$0.30\pm0.03~^{\rm A}$	$0.53\pm0.01^{\text{B}}$

Notes: Values in the same row marked with different capital letters indicate an extremely significant difference (P < 0.01), while those marked with different lowercase letters indicate a significant difference (P < 0.05). This convention applies throughout, and the values are mean of 3 replicates.

Table 2

Analysis of amino acid composition of "Jianghaida No. 1" and "Sutong No. 1" head water *P. yezoensis* (%, dry based).

Amino acid	Jianghaida No. 1	Sutong No. 1
Val [#]	$\textbf{2.48} \pm \textbf{0.08}$	$\textbf{2.44} \pm \textbf{0.02}$
Met [#]	0.55 ± 0.03	0.60 ± 0.04
Trp [#]	_	-
Thr [#]	1.78 ± 0.06	1.83 ± 0.06
Lys [#]	1.59 ± 0.05	1.54 ± 0.06
Leu [#]	2.81 ± 0.08	2.83 ± 0.03
Phe [#]	1.54 ± 0.05	1.54 ± 0.02
Ile [#]	1.67 ± 0.05	1.62 ± 0.02
Asp*	2.85 ± 0.15	2.89 ± 0.13
Ser*	1.54 ± 0.07	1.59 ± 0.02
Glu*	$2.93\pm0.05^{\rm a}$	$3.05\pm0.04^{\rm b}$
Gly*	1.84 ± 0.08	1.93 ± 0.10
Arg	2.22 ± 0.07	2.09 ± 0.06
Ala*	3.37 ± 0.09	3.32 ± 0.04
Pro	1.85 ± 0.04	1.89 ± 0.02
Tyr*	0.85 ± 0.05	0.83 ± 0.03
His	0.53 ± 0.01	0.52 ± 0.01
\sum TAA	29.63 ± 0.95	30.49 ± 0.39
∑EAA	11.65 ± 1.09	12.4 ± 0.14
\sum NEAA	17.98 ± 0.51	18.09 ± 0.26
∑UAA	13.39 ± 0.39	13.59 ± 0.20
$\sum EAA / \sum TAA$	39.26 ± 2.67	40.68 ± 0.13
$\sum EAA / \sum NEAA$	64.87 ± 7.08	68.57 ± 0.38
\sum UAA/ \sum TAA	45.21 ± 2.04	44.58 ± 0.1

Note: # represents essential amino acid; * represents umami amino acids; – represents undetectable; \sum TAA represents total amino acids; \sum EAA represents total essential amino acids; \sum NEAA represents total non-essential amino acids; \sum UAA represents total umami amino acids. The values are mean of 3 replicates.

Table 3

Evaluation of essential amino acid composition in the fronds of "Jianghaida No.1" and "Sutong No.1" *P. yezoensis* based on AAS model.

Essential amino acids	FAO/WHO Recommended model/(mg·g ⁻¹)	Jianghaida No. 1	Sutong No. 1
		AAS	AAS
Leu	440	$0.95\pm0.03~^{\text{A}}$	$\begin{array}{c} 1.19 \pm \\ 0.00^{\text{B}} \end{array}$
Thr	250	1.06 ± 0.04 $^{\text{A}}$	$\begin{array}{c} 1.36 \ \pm \\ 0.04^{\text{B}} \end{array}$
Val	310	$1.20\pm0.04~^{\text{A}}$	$\begin{array}{c} 1.46 \ \pm \\ 0.01^{B} \end{array}$
Lys	340	$0.70\pm0.02~^{\text{A}}$	$\begin{array}{c} 0.84 \pm \\ 0.03^{\mathrm{B}} \end{array}$
Phe and Tyr	380	$0.94\pm0.03~^{\text{A}}$	$\begin{array}{c} 1.15 \pm \\ 0.01^{\text{B}} \end{array}$
Ile	250	$1.00\pm0.03~^{\text{A}}$	$\begin{array}{c} 1.20 \ \pm \\ 0.02^{\mathrm{B}} \end{array}$
Met	220	$0.37\pm0.02~^{\text{A}}$	$\begin{array}{c} 0.50 \pm \\ 0.03^{\text{B}} \end{array}$

"Jianghaida No. 1" (*P* < 0.01).

Ten saturated fatty acids were identified, with the total saturated fatty acid content significantly higher in "Sutong No.1" than in "Jianghaida No.1" (P < 0.05). Particularly, C16:0 and C12:0 in "Sutong No.1" showed significantly higher levels compared to "Jianghaida No.1" (P < 0.05), and C14:0 and C18:0 exhibited extremely significant differences compared to "Jianghaida No. 1" (P < 0.01).

Regarding monounsaturated fatty acids, no significant difference was observed in the total content between the two *P. yezoensis* types (*P* > 0.05), nor in each individual monounsaturated fatty acid (*P* > 0.05). Nine polyunsaturated fatty acids were identified. The total polyunsaturated fatty acid content and the total unsaturated fatty acid content were significantly higher in "Jianghaida No. 1" than in "Sutong No.1". Specifically, C20:2, C18:3n-6, and C20:5n-3 were significantly higher in "Jianghaida No. 1" (*P* < 0.05), whereas C22:6n-3 in "Sutong No. 1" was notably higher than in

Table 4

Evaluation of essential amino acid composition in the fronds of "Jianghaida No.1" and "Sutong No.1" *P. yezoensis* based on CS model.

Essential amino acids	Egg protein recommended model/(mg·g ⁻¹)	Jianghaida No. 1	Sutong No. 1
		CS	CS
Leu	534	$0.79\pm0.02^{\text{ A}}$	$\begin{array}{c} 0.98 \pm \\ 0.00^{\text{B}} \end{array}$
Thr	292	$0.91\pm0.03~^{\text{A}}$	$\begin{array}{c} 1.16 \ \pm \\ 0.03^{\text{B}} \end{array}$
Val	410	$0.90\pm0.03~^{A}$	$\begin{array}{c} 1.10 \ \pm \\ 0.01^{B} \end{array}$
Lys	441	$0.54\pm0.02~^{A}$	$\begin{array}{c} 0.65 \pm \\ 0.03^{\text{B}} \end{array}$
Phe and Tyr	565	$0.63\pm0.02~^{\text{A}}$	$\begin{array}{c} 0.78 \pm \\ 0.01^{B} \end{array}$
Ile	331	$0.75\pm0.02\ ^{\text{A}}$	$\begin{array}{c} 0.91 \ \pm \\ 0.01^{B} \end{array}$
Met	386	$0.21\pm0.01~^{\text{A}}$	$\begin{array}{c} 0.29 \pm \\ 0.02^{B} \end{array}$

Table 5

Analysis of fatty acid components in "Jianghaida No. 1" and "Sutong No. 1" head water *Porphyra yezoensis* (%, dry based).

Fatty acid	Jianghaida No. 1	Sutong No. 1
C12:0	$0.03\pm0.00~^{\rm A}$	$0.06\pm0.01^{\rm b}$
C14:0	$0.20\pm0.01~^{\rm A}$	0.25 ± 0.01^{B}
C15:0	0.22 ± 0.01	0.24 ± 0.01
C16:0	41.74 ± 0.38^{a}	43.01 ± 0.5^{b}
C17:0	0.08 ± 0.01	$\textbf{0.10} \pm \textbf{0.01}$
C18:0	1.11 ± 0.01 $^{ m A}$	$1.14\pm0.01^{\rm B}$
C20:0	0.06 ± 0.00	$\textbf{0.06} \pm \textbf{0.01}$
C21:0	0.02 ± 0.00	$\textbf{0.03} \pm \textbf{0.01}$
C22:0	0.05 ± 0.00	$\textbf{0.04} \pm \textbf{0.03}$
C23:0	1.85 ± 0.04	1.75 ± 0.01
∑SFA	$45.36\pm0.42^{\rm a}$	$46.67 \pm 0.51^{\rm b}$
C16:1	0.32 ± 0.01	0.32 ± 0.00
C18:1n-9c	2.44 ± 0.02	$\textbf{2.46} \pm \textbf{0.03}$
C20:1	5.77 ± 0.02	5.73 ± 0.11
C22:1n-9	1.01 ± 0.07	$\textbf{0.93} \pm \textbf{0.02}$
C24:1	0.33 ± 0.03	0.21 ± 0.16
∑MUFA	$\textbf{9.87} \pm \textbf{0.06}$	9.65 ± 0.26
C18:2n-6c	1.88 ± 0.02	1.86 ± 0.03
C18:3n-3	0.28 ± 0.03	$\textbf{0.3} \pm \textbf{0.03}$
C18:3n-6	$0.22\pm0.01^{\rm a}$	$0.2\pm0.01^{\rm b}$
C20:2	$1.35\pm0.02^{\rm a}$	$1.30\pm0.02^{\rm b}$
C20:3n-3	0.47 ± 0.54	$\textbf{0.15} \pm \textbf{0.01}$
C20:3n-6	$2.14\pm0.03~^{\rm A}$	$1.99\pm0.01^{\text{B}}$
C20:4n-6	1.85 ± 0.04	1.75 ± 0.01
C20:5n-3	$36.52\pm0.15^{\rm a}$	$36.00\pm0.26^{\rm b}$
C22:6n-3	$0.08\pm0.02^{\rm a}$	$0.13\pm0.02^{\rm b}$
∑PUFA	44.78 ± 0.36^{a}	$43.68\pm0.25^{\rm b}$
\sum MUFA+ \sum PUFA	$54.65\pm0.42^{\text{a}}$	$53.34\pm0.50^{\rm b}$

Notes: \sum SFA represents total saturated fatty acids; \sum MUFA represents total monounsaturated fatty acids; \sum PUFA represents total polyunsaturated fatty acids; \sum MUFA+ \sum PUFA represents total unsaturated fatty acids. The values are mean of 3 replicates.

"Jianghaida No. 1" (P < 0.05). However, C20:3n-6 in "Jianghaida No.1" exhibited an extremely significant difference compared to "Sutong No. 1" (P < 0.01).

3.5. Mineral element composition analysis

After conducting mineral testing on both "Jianghaida No. 1" and "Sutong No. 1" strains of *P. yezoensis*, the analysis revealed the detection of 12 inorganic elements (Table 6). Sodium (Na) emerged as the most abundant mineral element in both strains, followed by potassium (K). However, four elements such as cadmium (Cd), arsenic (As), selenium (Se), and copper (Cu), were detected at relatively low levels in both strains (<0.01 mg·g⁻¹).

Table 6

Analysis of mineral element composition in "Jianghaida No. 1" and "Sutong No. 1" head water *P. yezoensis* (mg·g⁻¹, dry based).

inorganic elements	Jianghaida No.1	Sutong No. 1
Na	$10.73\pm0.06^{\rm A}$	$\textbf{7.83} \pm \textbf{0.4}^{B}$
Mg	$2.92\pm0.06^{\rm A}$	$3.12\pm0.01^{\rm B}$
K	$8.71\pm0.04^{\rm A}$	$4.62\pm0.23^{\rm B}$
Ca	$2.52\pm0.06^{\rm A}$	$3.86\pm0.02^{\rm B}$
Mn	0.01 ± 0.00	0.01 ± 0.00
Fe	$0.28\pm0.00^{\rm A}$	0.49 ± 0.00^{B}
Zn	$0.03\pm0.00^{\rm A}$	0.04 ± 0.00^{B}
Р	2.71 ± 0.09	2.65 ± 0.03
As	<0.01	< 0.01
Se	<0.01	< 0.01
Cu	<0.01	< 0.01
Cd	<0.01	< 0.01

Notes: The values are mean of 3 replicates.

The mass fraction of sodium (Na) and potassium (K) in "Jianghaida No. 1" *P. yezoensis* was notably higher than in "Sutong No. 1" (P < 0.01). Conversely, the content of four inorganic elements, such as calcium (Ca), magnesium (Mg), iron (Fe), and zinc (Zn), was significantly higher in "Sutong No. 1" compared to "Jianghaida No. 1" (P < 0.01).

4. Discussions

4.1. Analysis of differences in conventional nutritional components

This study examined the fundamental nutritional components of two variations of *P. yezoensis*, namely, "Jianghaida No. 1" and "Sutong No.1". The findings revealed significant disparities in crude protein, crude fat, crude fiber, crude ash, and total sugar content between the two types. The crude protein content observed in "Jianghaida No. 1" and "Sutong No. 1" exceeded the range typically reported for red algae, 20% to 32% (Yanshin et al., 2021). Notably, the crude protein content of both seaweed varieties surpassed that of *Grateloupia turuturu* (23%) (Denis et al., 2010), *Hypnea japonica* (19.0%) and *Hypnea charoides* (18.4%) (Wong & Cheung, 2000), and *Gracilaria changgi* (6.9%) (Norziah & Ching, 2000). Specifically, while the crude protein content of "Sutong No. 1" closely resembled that of white soybean (33.8%), "Jianghaida No. 1" exhibited an even higher crude protein content, surpassing white soybean by approximately 8%, reaching 41.79% (Hwang et al., 2013).

The crude ash content of "Sutong No. 1" closely resembled the results found in *Porphyra tenera* and *Porphyra haitanensis* (8.78% to 9.07%) (Hwang et al., 2013), whereas "Jianghaida No. 1" exhibited a notably lower crude ash content compared to "Sutong No. 1". Moreover, the crude fat content of "Sutong No. 1" surpassed that of "Jianghaida No. 1", yet both varieties of *P. yezoensis* demonstrated lower crude fat content in comparison to *P. tenera* and *P. haitanensis* (Hwang et al., 2013). In terms of crude fiber, "Jianghaida No. 1" showed significantly higher levels than "Sutong No. 1". Notably, crude fiber holds substantial importance in human metabolism. Furthermore, the significant disparity in total sugar content between "Sutong No. 1" and "Jianghaida No. 1" is notable. Given their richness in sugars and the thickening properties of their polysaccharide components, they serve as valuable food additives (Wang et al., 2021).

4.2. Analysis of amino acid composition differences

The analysis of amino acid composition in the two types of *P. yezoensis* revealed that "Jianghaida No. 1" exhibited a mass fraction of total amino acids ranging from 28.68% to 30.58%, while "Sutong No. 1" showed a range of 30.10% to 30.88%, indicating a similar mass fraction between the two. The ratio of essential amino acids to total amino acids for both seaweed types fell within the range of 36.59% to 41.93% and 40.55% to 40.81%, respectively. This ratio ($\Sigma EAA/\Sigma TAA$) closely aligned with the FAO/WHO standard of around 40%. Furthermore, the

ratio of essential amino acids to non-essential amino acids was observed to be between 57.79% to 71.95% and 68.19% to 68.95%, respectively, aligning closely with the FAO/WHO standard (\sum EAA/ \sum NEAA) of approximately 60% (Norziah & Ching, 2000). These findings indicate a reasonable distribution of various amino acids in both seaweed types, highlighting their high nutritional value and abundance in high-quality protein.

"Jianghaida No. 1" and "Sutong No. 1" showcased the highest content of alanine (ranging from 3.28% to 3.46%), followed by glutamic acid (2.88% to 2.98%, 3.01% to 3.09%) and aspartic acid (2.70% to 3.00%, 2.76% to 3.02%), with histidine showing the lowest content (0.52% to 0.54%, 0.51% to 0.53%). These findings align with the amino acid composition ratios of *G. changgi* (Norziah & Ching, 2000). Among these amino acids, "Jianghaida No. 1" and "Sutong No. 1" contained four distinct flavorful amino acids: Glu, Asp, Ala, and Gly (Hwang et al., 2013). Notably, Glu and Asp significantly contribute to the flavor profile of seaweed (Mišurcová et al., 2014; Paiva et al., 2014).

The mass fractions of umami amino acids ranged from 13.00% to 13.78% and 13.39% to 13.79%, respectively, constituting 42.51% to 48.04% and 43.36% to 45.81% of the total amino acids in both seaweed types, contributing to their delightful taste. Although there was no significant difference in aspartic acid content between the two strains (P > 0.05), "Sutong No. 1" exhibited a significantly higher glutamate content than "Jianghaida No. 1". Glutamic acid serves multiple functions, including enhancing mental reaction speed, bolstering the body's immunity, maintaining acid-base balance, and protecting intestinal mucosa (Chen et al., 2021).

4.3. Analysis of differences in the evaluation of nutritional value of amino acids

High-quality proteins necessitate a comprehensive spectrum of amino acids with rich content and a balanced distribution. Essential amino acids, in particular, play a crucial role in evaluating the nutritional value of food proteins (Norziah & Ching, 2000). Therefore, this study analyzes $\sum EAA / \sum TAA$ and $\sum EAA / \sum NEAA$. Yet, for a comprehensive assessment of the protein nutritional value of the two seaweeds, it's essential to integrate both amino acid score (AAS) and chemical score (CS) methodologies (Luo et al., 2017). The findings reveal that the amino acid scores and chemical scores of "Sutong No. 1" significantly surpass those of "Jianghaida No. 1". Considering AAS, the values of six amino acids (Leu, Thr, Val, Phe + Tyr, Ile) in "Sutong No. 1" exceed 1, indicating higher mass fractions of these amino acids in "Sutong No. 1" compared to the FAO/WHO standard model. However, only three amino acids (Thr, Val, Ile) in "Jianghaida No. 1" have values >1, though all amino acid mass fractions surpass the FAO/WHO standard model. This suggests that the proportion distribution of amino acids in "Sutong No. 1" is more reasonable, indicating a higher nutritional value. Considering CS as the benchmark, "Sutong No. 1" exhibits Thr and Val values >1, meeting FAO/WHO standards. Conversely, none of the amino acid values in "Jianghaida No. 1" reach 1, all falling below FAO/WHO standards. Combining AAS and CS, "Sutong No. 1" demonstrates amino acid values >1 in Thr and Val, indicating relatively rich content of these two amino acids. Among the 16 amino acids, Methionine (Met) exhibits the lowest content, serving as the first limiting amino acid, followed by Lysine as the second limiting amino acid. In summary, the amino acid composition and distribution in "Sutong No. 1" are more balanced and reasonable, resulting in a higher protein nutritional value compared to "Jianghaida No. 1".

4.4. Analysis of fatty acid composition differences

In the fatty acid composition of the two strains, "Jianghaida No. 1" and "Sutong No. 1", significant differences were observed in the total saturated fatty acids in *P. yezoensis*. Among the ten saturated fatty acids analyzed, palmitoleic acid stood out with the highest content at 41.74%

and 43.01%, respectively. Palmitic acid, known for its therapeutic properties in chronic diseases like metabolic syndrome, diabetes, and inflammation, has been identified for its potential in treating wound inflammation (Mozaffarian et al., 2013; Wu et al., 2012).

Significant differences were noted in the total unsaturated fatty acids between the two strains, with "Jianghaida No. 1" exhibiting a higher content, constituting 54.65% of the total fatty acid composition. While there was no significant difference in the total monounsaturated fatty acids between the two strains, this component represented the lowest proportion within the fatty acid composition. However, notable differences were observed in the total polyunsaturated fatty acids, where EPA emerged as the primary content at 36.52% and 36.00%, respectively, closely resembling the EPA content of 39% found in the analysis of *Porphyra umbilicalis* and *Porphyra amplissima* (Blouin et al., 2006). As an essential component of nori fatty acids, EPA plays a crucial role in reducing blood pressure, promoting smooth muscle movement, and relaxing blood vessels to prevent atherosclerosis (Bhatt et al., 2021). Consequently, both these strains of *P. yezoensis* exhibit high edible value and beneficial health effects.

4.5. Analysis of differences in mineral composition

Minerals, essential for human health and disease prevention, are naturally occurring inorganic substances found abundantly in various foods, aiding daily metabolic activities and functioning as vital micronutrients (Ciudad-Mulero et al., 2022). A myriad of analytical techniques have been employed for the determination of metal ions and trace elements, including ion chromatography (IC) (Muhammad, Zhang, Asif, et al., 2020; Muhammad, Zhang, Subhani, et al., 2020), ion chromatography coupled with fluorescence/UV detector (Muhammad et al., 2021), steam distillation ion chromatography (SD-IC) (Muhammad et al., 2023), and inductively coupled plasma mass spectrometry (ICP-MS) (Tanvir et al., 2020). In this study, ICP-MS method was employed for mineral content determination in these two strains of *P. yezoensis*. Both "Jianghaida No. 1" and "Sutong No. 1" strains of *P. yezoensis* exhibit the presence of 5 macroelements (Na, Mg, K, Ca, P) and 7 trace elements (Fe, Mn, Zn, Cd, As, Se, Cu).

Notably, "Jianghaida No. 1" showcased significantly higher Na and K contents, with Na being the highest at 10.73 $\text{mg}\cdot\text{g}^{-1}$ and 7.83 $\text{mg}\cdot\text{g}^{-1}$, respectively. Lower sodium content in food can contribute to preventing osteoporosis and high blood pressure (Czech et al., 2018). Potassium aids in blood pressure regulation, maintaining nervous system function, and supporting muscle function (Haddy et al., 2006). Conversely, "Sutong No. 1" demonstrated notably higher Ca and Mg levels compared to "Jianghaida No. 1". These minerals play vital roles in bone and teeth strength, blood clotting, energy metabolism, nerve transmission, fatigue alleviation, and protein synthesis (MacKenzie et al., 2008; Winter & Harris, 2020). Among trace elements, Fe and Zn exhibited the highest content, with "Sutong No. 1" displaying significantly higher levels than "Jianghaida No. 1". Zinc aids in preventing oxidative stress, enhancing metabolism, DNA, and protein synthesis, and bolstering immunity (Aubourg et al., 2021). Iron contributes to hemoglobin synthesis, redox processes, immunity, cognitive functions, and alleviating fatigue (Abbaspour et al., 2014). Although the manganese (Mn) content showed no significant difference between the two strains, it plays a crucial role in connective tissue formation and cellular protection against oxidative stress (Mehri, 2020). Both strains of P. yezoensis exhibit rich mineral elements, meeting human body needs and possessing high edible value.

5. Conclusions and future perspectives

In summary, "Jianghaida No. 1", in comparison to the "Sutong No. 1" strain of *P. yezoensis*, stands out as a high-quality strain characterized by high protein content, low fat, and abundant minerals. The findings lay the groundwork for improving the nutritional value of *P. yezoensis* products and meeting the dynamic demands of consumers, thereby

paving the way for future large-scale promotion and cultivation efforts of this new strain of *P. yezoensis*.

While this study successfully screened for the nutrient-rich new strain "Jianghaida No. 1" from two locally cultivated Porphyra varieties, there are inherent limitations that should be acknowledged. First, "Jianghaida No. 1" displayed promising growth performance and nutritional value only under the specific environmental conditions present in its breeding location of Lianyungang, Jiangsu Province. Its suitability for large-scale nationwide cultivation remains to be validated. Additionally, the assessment of nutritional value was not comprehensive, as vitamin content and functional bioactive compounds were not analyzed. Future research expanding cultivation trials of "Jianghaida No. 1" to sites across China while incorporating a wider array of nutritional parameters will be crucial for substantiating its potential for nationwide proliferation and consumer marketability.

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CRediT authorship contribution statement

Bin Guan: Writing – review & editing, Writing – original draft, Data curation. Yuyan Sun: Writing – review & editing, Writing – original draft, Data curation. Xuxiao Liu: Visualization, Validation, Investigation. Chongyu Zhong: Visualization, Validation, Investigation. Desheng Li: Visualization, Validation, Investigation. Desheng Li: Visualization, Validation, Investigation. Xin Shan: Visualization, Validation, Investigation. Xingxing Hui: Visualization, Validation, Investigation. Chaofa Lu: Visualization, Validation, Investigation. Yujia Huo: Visualization, Validation, Investigation. Runkai Sun: Visualization, Validation, Investigation. Min Wei: Conceptualization. Wei Zheng: Conceptualization.

Declaration of competing interest

The authors declare that they have no conflicts of interest in the submission of this manuscript and approve the manuscript for publication.

Data availability

Data will be made available on request.

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