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Effect of seaweed (*Eucheuma cottonii*) addition on the physical, chemical and hedonic properties of peas-based meatballs in comparison with conventional meatball

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ABSTRACT

Vegetable proteins, such as those derived from peas (*Pisum sativum* L.), lack a naturally fibrous structure, resulting in a texture that differs significantly from conventional meat when processed into meat analogues like meatballs. Seaweed, known for its structural complexity and dietary fiber content, was explored in this study as a functional ingredient to enhance the texture and overall quality of pea-based meatballs. The objective was to evaluate the effects of seaweed flour addition on the physical, chemical, sensory, and preference properties of these plant-based products. Seaweed was added at concentrations of 0, 1, 1.5, and 2% (w/w) into a mixture of meatball (pea paste, tapioca, and spices). Conventional beef meatballs were used as the control. The samples were evaluated for moisture, protein, fat content, water holding capacity, cooking loss, textural attributes (hardness, cohesiveness, springiness, adhesiveness), crude fiber, yield, sensory quality, and consumer preference. Data were analyzed using one-way ANOVA and principal component analysis (PCA). Results showed that seaweed flour addition up to 2% significantly increased the moisture and protein contents, WHC, texture parameters, yield, and fiber content, while reducing cooking loss compared to the control pea-based meatballs, with no significant effect on fat content ($p > 0.05$). Compared to beef meatballs, pea-based meatballs had lower values in most quality parameters but showed higher hardness, springiness, adhesiveness, and fiber. PCA confirmed distinct characteristics between pea- and beef-based meatballs, in agreement with the ANOVA findings.

Introduction

Meatballs are a processed meat product that, in addition to being a nutritious food source, are also palatable, widely accepted by various age groups, and suitable for consumption on diverse occasions. They serve as a popular means of consuming protein-rich foods. However, the accessibility of meat remains limited for some segments of the population due to its relatively high cost. In contrast, plant-based protein sources, such as legumes particularly peas, are more readily available. Thus, the development of pea-based meatballs offers a promising alternative protein source. This type of product is classified as plant-based meatballs. Processing legumes into meatballs has been shown to reduce the typical beany odor, thereby enhancing consumer acceptance and supporting increased consumption of plant-derived protein (Van Der Meer *et al*., 2022).

Pea (*Pisum sativum* L.) is a leguminous plant widely utilized for its pods due to their high protein content, which ranges from 21.2% to 32.9% (Windsor *et al*., 2024). In addition to protein, Peas possess a diverse nutritional composition, featuring protein alongside essential amino acids, dietary fiber, carbohydrates, and a range of micronutrients, including vitamins and minerals (Kumari and Deka, 2021). The chewy texture of conventional meatballs is largely attributed to the fibrous structure of meat proteins (Beniwal *et al*., 2021). Proteins play a crucial role in meatball formulation, primarily functioning as a binder during the cooking process, which contributes to the formation of a cohesive and compact texture (Sun *et al*., 2024). However, globular proteins, such as those found in peas, tend to result in a softer texture when used as the primary protein source (Martins *et al*., 2018). Hence, it becomes essential to identify functional ingredients that can replicate the structural role of fibrous meat proteins in plant-based formulations.

Seaweed (*Eucheuma cottonii*) contains approximately 4.3% protein, 2.1% fat, 90.9% carbohydrates, 83.2% total dietary fiber, 2.7% ash, and 65.7% carrageenan (Hidayah *et al*., 2022). Carrageenan is a polysaccharide fiber known for its functional properties as a stabilizer, thickening agent, gelling agent, and emulsifier. In meatball applications, carrageenan contributes to improved texture and helps prevent fat loss during processing (Kumar and Sharma, 2004). Therefore, this study aimed to evaluate the effects of seaweed flour fortification on the physical, chemical, sensory, and hedonic properties of pea-based meatballs.

Materials and methods

Preparation of seaweed flour

Seaweed flour was prepared following the method described with slight modifications (Agusman *et al*., 2020). *Eucheuma cottonii* seaweed was first soaked in freshwater to remove impurities, rinsed under running water, and then drained. The cleaned seaweed was subsequently soaked in a 5% lime solution for 5 hours, washed again, and drained. It was then dried using a drying apparatus for 24 hours. Once dried, the seaweed was ground into flour and sieved using an 80-mesh screen.

Preparation of pea paste

Pea paste was prepared according to the method described by Genisa *et al*. (2015) with modifications. The peas were soaked for 12 hours, after which their skins were removed. The dehulled peas were then boiled in water for 30 minutes. Following boiling, the peas were cooled and blended until smooth and homogeneous. The resulting pea paste was ready for use.

Preparation of pea-based meatballs with seaweed flour

Pea-based meatballs with seaweed flour were prepared based on the method described by Utafiyani *et al*. (2018) with modifications. The

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meatball production involved mixing pea paste with additional ingredients such as tapioca flour, seasoning, and ice water. Seaweed flour was then incorporated according to the treatment (0, 1, 1.5, and 2% (w/w)). The mixture was stirred until homogeneous and formed into balls (1.5 cm in diameter). The formed meatballs were then placed into boiling water and cooked for approximately 5–10 minutes at a temperature of 80°C.

Chemical, physical, and hedonic evaluation

Moisture content

Moisture content was determined following the method of AOAC (1990). A porcelain crucible was pre-dried in an oven at 105°C for 1 h, then removed and placed in a desiccator for 15 minutes. The dried crucible was weighed and recorded as weight A. A 2 g sample was weighed (recorded as weight B) and placed into the crucible, then dried in the oven at 105°C for 5 hours. After drying, the crucible was cooled in a desiccator for 15 minutes and reweighed to obtain weight C. Moisture content was calculated using Equation 1.

Moisture Content=(B-(C -A))/(B)×100% [Equation 1]

Protein content

Protein content was determined using the Kjeldahl method as described by AOAC (1990). A 0.5 g sample was mixed with 0.5 g selenium catalyst and 10 mL of concentrated sulfuric acid (H_2SO_4). The mixture was digested until the solution turned clear green. The digested solution was then distilled by adding 100 mL of distilled water and 40 mL of 45% NaOH, along with 5 mL of 4% boric acid (H_3BO_3) containing two drops each of mixed indicators methyl red (MR) and methylene blue (MB). The distillate was then titrated with 0.1 N HCl until the color changed to purplish. The nitrogen content was calculated using Equation 2, where Sample weight is the weight of the sample (g), V sample is volume of titrant used for the sample (mL),V blank is volume of titrant used for the blank (mL), N H_2SO_4 is normality of sulfuric acid, N is nitrogen equivalent weight (14.008).

%N=((V sampel-V blank)×N H_2SO_4×bst N×100)/(sampel weight (g)×1000)

%N=(V sampel - V blank)×0.05 N×14.008×100)/(sampel weight (g)×1000)

Protein=% N×6.25 [Equation 2]

Water holding capacity

Water holding capacity (WHC) was measured based on the method (Wilhelm *et al*., 2010). A 0.3 g sample was placed on filter paper and compressed between two plates under a 35 kg load for 5 minutes. The area covered by the sample and the surrounding wet area were then marked and measured using a planimeter. The difference between the two was recorded as the wet area. Water holding capacity was calculated using Equation 3.

MgH_2O=(wet area)/0,0948

MgH_2O=(stained area -sampel area)/0,0948 [Equation 3]

Cooking loss

Cooking loss was measured following the method (Öztürk and Turhan, 2020). It was calculated as the percentage difference between the sample weight before and after cooking, relative to the initial weight prior to cooking. Cooking loss was determined using Equation 4, where W1 is weight of the sample before cooking (g), W2 is weight of the sample after cooking (g).

Cooking loss (%)=(W_1-W_2)/W_2 ×100% [Equation 4]

Texture

Texture analysis was conducted using a texture analyzer (Brookfield CT3) by applying a defined force to the sample in order to measure its textural profile. The texture parameters evaluated included hardness, springiness, adhesiveness, and cohesiveness. Springiness refers to the extent to which a food product returns to its original shape after being compressed (Akbar *et al*., 2019). Hardness is defined as the maximum force recorded during the first compression cycle. Springiness is calculated as the degree to which a deformed sample returns to its undeformed condition once the deforming force is removed. Cohesiveness is measured as the ratio of the area under the second compression curve to that under the first (Monsalve-Atencio *et al*., 2021). Texture measurements were carried out at the Integrated Laboratory, Diponegoro University.

Cooking yield

Yield was measured based on the method (Hannachi *et al*., 2019). The weight of the meatball mixture (raw batter) was recorded prior to cooking, and the weight of the cooked meatballs was measured afterward. The yield was then calculated by comparing the weight of the cooked product to the initial batter weight, multiplied by 100%. Yield was determined using Equation 5.

Yield (%)=(Weight of Cooked Meatballs)/(Weight of Batter)×100% [Equation 5]

Fat content

Fat content was determined according to the method described by AOAC (1990). Filter paper was first dried in an oven at 100°C for one hour. The sample was macerated, weighed (2 g), and recorded as weight A. It was then wrapped in filter paper and dried in an oven for 4 hours at 100°C. The dried sample was cooled in a desiccator for 15 minutes and weighed, recorded as weight B. The sample was then placed in a Soxhlet extractor, and extraction was performed using ether for 6 hours. After extraction, the sample was left to stand for 30 minutes, dried again in an oven at 100°C for one hour, and weighed as weight C. Fat content was calculated using Equation 6, where A is the initial weight of the sample before drying and extraction (g), B is the weight after drying, before fat extraction (g), and C is the weight after fat extraction and final drying (g).

Fat Content (%)=(B-C)/A×100% [Equation 6]

Crude fiber content

Crude fiber content was determined according to the method described by (AOAC, 1990). A sample was weighed to obtain an initial weight of y grams and placed into a fritted crucible. The crucible was mounted onto the Fibertec apparatus and secured using the locking brush. Then, 100 mL of 1.25% sulfuric acid solution was poured into each fritted crucible on the Fibertec unit. The small valve was set to the closed position, the power button was turned on, and the thermostat dial was set to level 6. The heater cover was placed over the heating unit. Once the solution reached boiling, the thermostat was reduced to level 4 and maintained for 30 minutes. The solution was then filtered using the built-in filter tap. The valve was then set to the vacuum position, and each crucible was rinsed with 50 mL of hot distilled water. Next, 100 mL of 1.25% sodium hydroxide solution was added to each crucible in the closed position, using the same method as for the acid solution. After completion, the main valve was released to unlock the crucibles, which were then removed one by one. Each crucible was rinsed again in the Fibertec unit using acetone, following the same procedure. The crucibles were then placed in an oven at 105°C for 4 hours. After drying, they were cooled in a desiccator for 15 minutes and weighed to obtain x. The crucibles were then burnt in a furnace at 600°C, turned off, and allowed to cool to 100°C. The samples were again transferred to a desiccator for 15–19 minutes and then weighed to obtain z. Crude fiber content was calculated using Equation 7, where x is weight of the fritted crucible with sample residue after drying (g), z is weight of the fritted crucible with the sample residue after furnacing (g), y is weight of the initial sample (g).

Crude Fiber (%)=(z-x)/y×100% [Equation 7]

Hedonic test

Hedonic evaluation was carried out following the method described by Triyannanto *et al.* (2022). A total of 25 semi-trained panelists, consisting of university students aged 20–25 years, participated in the test. The cooked pea-based meatball samples were served on plates and evaluated based on color, aroma, taste, and texture. The meatball was heated for 2 minutes on 70°C. Each attribute was rated using a 10-point hedonic scale, where 1 indicated "dislike extremely" and 10 indicated "like extremely." Panelists were asked to assign scores based on their level of liking for each attribute.

Data analysis

The data of moisture content, protein content, water holding capacity, cooking loss, and texture were analyzed using Analysis of Variance (ANOVA) at a 95% confidence level, followed by Duncan's Multiple Range Test (DMRT) using IBM SPSS Statistics 26.0 for Windows. Additionally, the data were analyzed using Multivariate Data Analysis (MVDA) to examine the similarities and differences among treatments through Principal Component Analysis (PCA), conducted with the assistance of Chemoface 1.61 for Windows.

Results

Physical characteristics

The physical characteristics evaluated in this study included yield, water holding capacity (WHC), and cooking loss, as well as texture parameters such as hardness, cohesiveness, springiness, and adhesiveness. Results for yield, WHC, and cooking loss are presented in Table 1, while texture profile data are shown in Table 2. These parameters are essential for assessing the functional properties and quality of meat and plant-based meatball formulations.

Table 1. Physical characteristics of pea-based meatballs with and without seaweed flour, and beef meatballs.

Treatment	Yield (%)	WHC (%)	Cooking Loss (%)
BF	2.8±0.12^{c}	56.98±2.17^{d}	15.72±0.65^{a}
PP	0.54±0.07^{a}	24.09±0.54^{a}	15.01±0.29^{a}
PS1	1.00±0.20^{b}	26.24±1.20^{a}	13.63±0.47^{b}
PS2	1.39±1.05^{c}	30.17±1.39^{b}	12.38±0.47^{a}
PS3	1.74±0.25^{d}	33.65±1.63^{c}	11.79±0.63^{a}

BF: beef meatballs; PP: plain pea-based meatballs without seaweed flour; PS1: pea-based meatballs enriched with 1% seaweed flour (w/w); PS2: pea-based meatballs enriched with 1.5% seaweed flour (w/w); PS3: pea-based meatballs enriched with 2% seaweed flour (w/w). Data are presented as Mean±Standard Deviation (n = 4). Values in the same column with different superscript letters indicate significant differences according to Duncan's Multiple Range Test (DMRT) at $p < 0.05$.

The yield of the sample shown in Table 1. It was found that the BF sample had a significantly higher value ($p < 0.05$), compared to other samples, followed by PS1 and PS2. Beef meatballs exhibited a higher yield compared to both plain pea-based meatballs and pea-based meatballs fortified with seaweed flour. Water holding capacity of BF sample has a similar phenomenon as yield, WHC exhibited a significantly higher value ($p < 0.05$) compared to other samples, followed by PS3. This was highly associated with yield suggests that the superior protein matrix structure in beef meatballs enhances water retention, contributing to their higher yield during processing.

Cooking loss of the samples showed no significant difference ($p > 0.05$) among BF, PP, PS2, and PS3. While the lowest value of cooking loss was exhibited by PS1. The value of cooking loss is also related to the value of water holding capacity. However, those two parameters showed different trends. Regarding the hardness, PP sample had a significantly higher value ($p < 0.05$), followed by PS1, PS2, and PS3. Furthermore, BF samples showed the lowest value among samples. In contrast, cohesiveness was significantly higher ($p < 0.05$) in BF sample, followed by PS3, PS2, and PS1. This trend also can be seen in springiness and adhesiveness; BF exhibited a significantly higher value compared to other sample.

Table 2. Texture characteristics of pea-based meatballs with and without seaweed flour and beef meatballs.

Treatment	*Hardness (g)*	*Cohesiveness*	*Springiness (mm)*	*Adhesion (mJ)*
BF	206.87±16.66^{a}	0.59±0.02^{c}	7.17±0.59^{a}	0.91±0.25^{a}
PP	426.00±38.66^{e}	0.36±0.04^{a}	7.10±0.53^{a}	6.16±2.24^{c}
PS1	369.50±22.06^{d}	0.43±0.04^{b}	8.15±0.85^{b}	4.32±0.60^{b}
PS2	279.87±6.20^{c}	0.49±0.07^{b}	8.82±0.35^{b}	2.39±0.88^{a}
PS3	244.00±18.67^{b}	0.50±0.03^{b}	9.02±0.68^{b}	1.95±0.97^{a}

BF: beef meatballs; PP: plain pea-based meatballs without seaweed flour; PS1: pea-based meatballs enriched with 1% seaweed flour (w/w); PS2: pea-based meatballs enriched with 1.5% seaweed flour (w/w); PS3: pea-based meatballs enriched with 2% seaweed flour (w/w). Data are presented as Mean±Standard Deviation (n = 4). Values in the same column with different superscript letters indicate significant differences according to Duncan's Multiple Range Test (DMRT) at $p < 0.05$.

Chemical characteristics

The chemical characteristics of pea-based meatballs with seaweed flour fortification, as well as beef meatballs, are presented in Table 3. The chemical characteristics of pea-based meatballs and beef meatballs, including moisture, protein, fat, and crude fiber contents. The result of moisture content and protein content showed similar trend, BF sample exhibited a significant higher value ($p < 0.05$) compared to other samples. This value was followed by PS3, PS2, and PS1. Meanwhile, fat content had no significant value among samples. In contrast, crude fiber showed a significantly higher value ($p < 0.05$) in PS3, followed by PS2 and PS2, while BF exhibited the lowest value.

Table 3. Chemical characteristics (%) of pea-based meatballs with and without seaweed flour and beef meatballs.

Treatment	Moisture Content	Protein Content	Fat Content	Crude Fiber
BF	75.68±1.18^{c}	10.31±0.09^{c}	2.69±0.24^{a}	1.49±0.27^{a}
PP	51.78±1.49^{a}	7.50±015^{a}	1.97±0.22^{a}	3.99±0.58^{b}
PS1	52.50±1.54^{a}	8.04±0.13^{b}	1.91±0.48^{a}	4.14±0.69^{b}
PS2	54.43±1.01^{b}	8.18±0.16^{b}	1.76±0.36^{a}	4.67±0.87^{b}
PS3	54.63±1.42^{b}	8.22±0.19^{b}	1.55±0.13^{a}	5.90±1.28^{c}

BF: beef meatballs; PP: plain pea-based meatballs without seaweed flour; PS1: pea-based meatballs enriched with 1% seaweed flour (w/w); PS2: pea-based meatballs enriched with 1.5% seaweed flour (w/w); PS3: pea-based meatballs enriched with 2% seaweed flour (w/w). Data are presented as mean±standard deviation (n = 4). Values in the same column with different superscript letters indicate significant differences according to Duncan's Multiple Range Test (DMRT) at $p < 0.05$.

Hedonic characteristics

The sensory attributes and hedonic quality of pea-based meatballs with seaweed flour fortification, as well as beef meatballs, are presented in Table 4. The sensory attributes and hedonic quality of pea-based meatballs and beef meatballs, including color, aroma, texture, and taste.

For color, the BF sample exhibited the lowest score (3,25±0,50), which means it had darker appearance compared to other samples. In contrast, pea-based meatballs, plain and fortified with seaweed flour (PS1, PS2, PS3), showed significantly higher scores. Regarding the aroma, PP sample recorded the highest score (6.00±0.00), followed by pea-based samples PS1 (5.50±0.57), PS2 (5.00±0.00), PS3 (5.00±0.00). For the texture, BF sample demonstrated the highest value (PS1 (8.00±0.00), significantly higher than all pea-based formulations. Among the plant-based formulations, PS2 and PS3 samples scored slightly better than PP and PS1, with texture scores ranging between 4.00±0.00 and 5.00±0.05. Regarding the taste, there were no significant differences (p > 0.05), between BF, PS2, and PS3, but showed significantly higher than PS1 and PP.

Table 4. Sensory and hedonic characteristics of pea-based meatballs with and without seaweed flour and beef meatballs.

Treatment	Color	Aroma	Texture	Taste
BF	3.25±0.50^{a}	1.75±0.95^{a}	8.00±0.00^{e}	6.75±0.05^{c}
PP	7.00±0.00^{b}	6.00±0.00^{c}	5.00±0.00^{d}	4.00±0.00^{b}
PS1	7.00±0.00^{b}	5.50±0.57^{b}	3.00±0.05^{a}	3.25±0.05^{a}
PS2	8.00±0.00^{c}	5.00±0.00^{b}	5.00±0.05^{c}	4.00±0.00^{b}
PS3	7.00±0.00^{b}	5.00±0.00^{b}	4.00±0.00^{b}	6.00±0.00^{c}

BF: beef meatballs; PP: plain pea-based meatballs without seaweed flour; PS1: pea-based meatballs enriched with 1% seaweed flour (w/w); PS2: pea-based meatballs enriched with 1.5% seaweed flour (w/w); PS3: pea-based meatballs enriched with 2% seaweed flour (w/w). Data are presented as mean±standard deviation (n = 4). Values in the same column with different superscript letters indicate significant differences according to Duncan's Multiple Range Test (DMRT) at p < 0.05.

Principal component analysis

The results of ANOVA and post hoc tests are often difficult to interpret clearly; therefore, Principal Component Analysis (PCA) was also conducted to facilitate pattern recognition among treatments. The outcomes of this analysis are presented in Fig 1.

According to the PCA biplot, the beef meatball sample (BF) was closely associated with moisture content and water holding capacity (WHC), indicating the highest values for both variables. Compared to beef, pea paste contains less protein, resulting in reduced water-binding ability. BF and PP formed distinct clusters, indicating that these samples exhibited different characteristics compared to the seaweed-fortified pea-based meatballs (PS1, PS2, PS3).

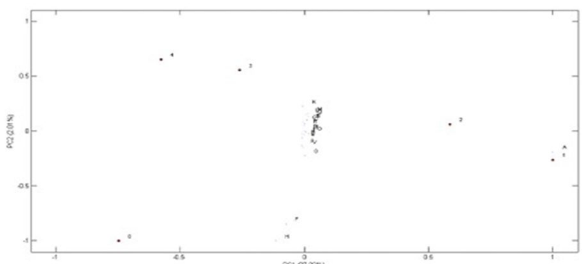

Fig. 1. PCA biplot of meatball samples and variables. Sample codes: 0 = beef meatball (animal-based control); 1 = meatball made from pea paste without seaweed flour; 2 = pea-paste meatball with 1% seaweed flour; 3 = with 1.5% seaweed flour; 4 = with 2% seaweed flour. Variable codes: (A) hardness, (B) cohesiveness, (C) springiness, (D) adhesiveness, (E) protein content, (F) moisture content, (G) cooking loss, (H) water holding capacity, (I) fat content, (J) yield, (K) crude fiber, (L) grayish-green color, (M) nutty aroma, (N) beany odor, (O) seasoning aroma, (P) chewy texture, (Q) hard texture, (R) coarse mouthfeel, (S) bitter taste, (T) seasoning taste, (U) raw ingredient taste, (V) aftertaste, and (W) overall hedonic score.

Discussion

The high yield of BF had the highest value since beef meatballs are primarily attributed to the presence of animal protein in beef. According to Ockerman (1985), animal proteins can retain more water, which contributes to the higher yield observed in beef meatballs. The elevated yield is due to the protein's ability to bind water content. The greater water retention in beef meatballs results in reduced water loss during cooking, thereby increasing the overall yield (Li *et al.*, 2025). In contrast, the yield of pea-based meatballs increased progressively with the addition of seaweed flour. The carrageenan content in seaweed flour may act as a stabilizer and binder, helping to integrate other components and minimize losses during the production of plant-based meatballs (Agusman *et al.*, 2014).

Highest value of WHC in BF sample is associated with the amount of protein in beef meatball, meat protein content will be followed by water holding capacity, this is also related to the damage of protein, when the oxidation of protein occurs, WHC of meat will reduce (Zhu *et al.*, 2019). Pea meatballs experience an increase in water holding capacity with the addition of seaweed flour. This finding is consistent with the report which states that the higher the concentration of seaweed used, the higher the water holding capacity because seaweed contains carrageenan which has the ability to bind water (Sari *et al.*, 2021). The water holding capacity value is higher because hydrocolloids bind water and increase the density around the protein matrix (Chen *et al.*, 2020).

Cooking loss is closely related to pH levels. Low pH causes weak protein bonds, resulting in more water loss during boiling, resulting in higher cooking losses (Osen *et al.*, 2015). In pea-based meatballs, cooking loss decreased with the addition of seaweed flour. This reduction is attributed to the presence of hydrocolloid compounds such as carrageenan in seaweed, which possesses strong water-binding capacity, thereby reducing water release during the cooking process. Hydrocolloids have the characteristic of absorbing and binding water in the manufacture of food products and can be used as additives for stabilizers, texturizers, and water absorbers (Salehi, 2019).

The lower hardness value in beef meatballs is influenced by the amount of extracted protein, the water-binding capacity, and gel formation by protein and starch (Zhang *et al.*, 2022; Aslinah *et al.*, 2018; Li *et al.*, 2022; Jiang *et al.*, 2025). In addition, the high moisture content in beef meatballs also contributes to the reduction in hardness (Utama *et al.*, 2020). In contrast, the hardness of pea-based meatballs decreased with the addition of seaweed flour. Carrageenan, a hydrocolloid compound present in seaweed, improved the texture and stability of food products (Kalsi *et al.*, 2025). Carrageenan contains higher levels of insoluble dietary fiber, it can bind water and trap it in the matrix during carrageenan gel formation (Lévesque *et al.*, 2014). The higher the amount of water trapped in the matrix, the softer and more hydrated gel, reducing its resistance to compression. This is consistent with reported that the highly hygroscopic nature of carrageenan results in a strong molecular structure that binds to water (Cheng *et al.*, 2022). Therefore, the higher the seaweed concentration, the greater the water entrapment in the product, which in turn leads to a decrease in hardness.

Regarding the cohesiveness value of sample, actin and myosin proteins stabilize the emulsion between fat and water in meat products, causing to a more compact and elastic structure (Agustini *et al.*, 2015). Texture is also influenced by the presence of filler ingredients such as starch; during cooking, as meat proteins contract, starch molecules occupy the resulting gaps, thereby enhancing the compactness of the texture (Carballo *et al.*, 1995). In pea-based meatballs, cohesiveness increased with the addition of seaweed flour. This is likely due to the ability of carrageenan to strengthen gel networks, making the meatballs more cohesive and firmer. Carrageenan interacts with charged macromolecules such as proteins, which can affect viscosity, gel formation, precipitation, and stabilization (Agustini *et al.*, 2015).

Myofibrillar proteins have the ability to bind water and fat, playing a crucial role in gel formation, coagulation, and improving the elasticity of processed meat products (Acton *et al.*, 1983). Increasing protein content will increase the springiness of beef meatballs. The greater water content in meatballs results in a juicier and more elastic texture (Wi *et al.*, 2020). Furthermore, the high amylopectin content in tapioca flour enhances springiness, as it contributes to the formation of a chewy gel matrix (Wang e al., 2020). In pea-based meatballs, springiness increased with the addition of seaweed flour. This may be due to the increase in moisture and fiber content provided by seaweed, which is rich in dietary fiber. As a result, the texture becomes softer and more elastic.

Beef meatballs showed lower adhesiveness values compared to both plain pea-based meatballs and those fortified with seaweed flour. Higher amylose content is associated with reduced stickiness in meatballs. This is because the presence of amylose can increase the strength of intermolecular bonds that can form crystalline clusters that can inhibit amylose leaching and reduce adhesiveness (Zhang *et al*., 2017). Pea meatballs with the addition of seaweed flour reduced the adhesion value. This is because the protein contained in seaweed plays a role in the gel formation process by increasing water binding capacity, thereby reducing stickiness in the meatballs (Mamat *et al*., 2013). Therefore, the higher the concentration of seaweed flour, the lower the adhesive value.

Beef meatballs (BF) showed higher moisture content than pea meatballs and pea meatballs fortified with seaweed flour. The filler used in beef meatballs was tapioca flour, which is derived from cassava, containing amylose approximately at 21.43% (Shariffa *et al*., 2009). The high amylose content in tapioca flour causes a rapid increase in water content in meatballs because amylose is able to bind water easily. In pea-based meatballs, moisture content increased with the addition of seaweed flour. This is due to the hydrocolloid properties of seaweed, which enable it to absorb and retain water efficiently (Lomartire and Gonçalves, 2023). Seaweed contains relatively high moisture content, about 27.8% on a dry basis (Arnyke, 2014). Based on the Indonesian National Standard (SNI 3818-2014), the maximum allowable moisture content in meatballs is 70% (BSN, 2014). Therefore, the moisture observed in this study remains within the standard according to national standards.

Beef meatballs (BF) exhibited higher protein content than pea meatballs and pea meatballs fortified with seaweed flour. An increased proportion of beef in the formulation tends to elevate the overall protein content of the final product. Beef contains approximately 18.80 g of protein per 100 g, while peas contain around 12.4 g per 100 g (Purwanto *et al*., 2015). Meanwhile, seaweed flour contains about 7.91% protein on a dry basis (Tapotubun, 2018). In the pea-based meatballs, protein content increased with the addition of seaweed flour. This increase might be due to the presence of carrageenan in seaweed, which can bind water and reduce protein loss during the cooking process. Carrageenan has the ability to bind and trap water within a gel matrix, thereby minimizing the leaching of water-soluble proteins during boiling (Gao *et al*., 2015). Based on the Indonesian National Standard (SNI 3818-2014), the minimum protein content requirement for meatballs is 11%. The protein content observed in this study did not meet this requirement. One of the causes of the low protein content in beef meatballs is due to the high carbohydrate content caused by the starch content exceeding 50% of the meat content in meatballs, this is intended to reduce production costs by meatball producers. The low protein content in pea meatballs and seaweed flour fortified pea meatballs is due to the low protein content in peas and seaweed.

Beef meatballs (BF) exhibited higher fat content compared to both plain pea-based meatballs and those fortified with seaweed flour. The meat used may contain muscle fibers rich in intramuscular fat. Furthermore, beef contains approximately 3.67% fat. The result of the study showed that the fat content of pea-based meatballs decreased with the addition of seaweed flour. This reduction is attributed to the use of raw materials with inherently low-fat content. Peas contain only 1–2% fat, while seaweed flour also has a low fat content of approximately 0.58% (Pugalenthi *et al*., 2004).

PS3 exhibited significantly highest value in crude fiber, while BF showed the lowest value. This is likely due to the type of feed consumed by the cattle, which can influence the quality and fiber composition of the resulting meat. The crude fiber content in beef meatballs directly affects their overall quality (Apriyanto *et al*., 2018). In contrast, the crude fiber content in pea-based meatballs increased with the addition of seaweed flour. Seaweed contains cellulose, a component of crude fiber known for its ability to absorb water (Ariyani and Ayustaningwarno, 2013).

The color of beef meatballs is influenced by the myoglobin content in the meat; the higher the myoglobin level, the redder the meat appears. During cooking, the red color undergoes a transformation to a grayish-brown hue due to oxidation processes. Pea-based meatballs, including those fortified with seaweed flour, exhibited a grayish-green color. This might be due to browning reactions occurring between proteins and heat during cooking, resulting in a color similar to conventional meatballs, which tend to have a slightly brownish tone and are generally preferred by panelists. The slight grayish-green color of the product was still well accepted, possibly due to protein-related reactions. Non-enzymatic browning of proteins, such as Maillard reactions, can occur during thermal processing, contributing to desirable color development in the final product (Ames, 1992).

Beef meatballs did not exhibit any undesirable beany odor. The use of fresh beef contributes to the characteristic aroma of cooked meat products (Berutu *et al*., 2010). In contrast, both plain and seaweed-fortified pea-based meatballs had a slight beany aroma. This off flavor is attributed to the activity of lipoxygenase enzymes, which hydrolyze or break down the lipids in legumes, leading to the formation of volatile compounds responsible for the beany odor (Pangastuti *et al*., 2013).

The springiness of beef meatballs is related to the strength of the gel formed by heating. Gelatinization in meatballs consists of starch gelatinization and protein gelatinization, but starch gelatinization is more dominant in influencing the springy texture of meatballs. The gelatinization process involves the embrittlement of water by a network formed by chains of starch or protein molecules (Han and Hamaker, 2002). Pea meatballs fortified with seaweed flour have a less chewy texture. Seaweed flour has the property of binding water so that the texture of meatballs with the addition of seaweed flour becomes hard (Huang *et al*., 2005). The increase in firmness is also attributed to the higher carbohydrate content in the formulation, as a greater amount of starch can lead to increased hardness in meatball products.

The savory flavor of beef meatballs can be attributed to the incorporation of fresh beef. Meatballs crafted from pre-rigor meat typically showcase superior flavor due to the proteins remaining fresh and functional, a higher pH level, and the absence of lipid oxidation. This combination aids in preserving more umami compounds, resulting in a cleaner and fresher taste. Pea-based meatballs fortified with seaweed flour retained a slight flavor characteristic of the pea ingredient. The use of flour as a filler may also influence flavor, as amylose in starch can form inclusion complexes with flavor compounds such as salt and seasonings, thereby altering the perception of taste (Wang *et al*., 2020).

Moisture content in meatballs is influenced by the protein content of the raw materials. While, WHC is primarily affected by myofibrillar proteins, which are abundant in beef. These proteins, along with stromal proteins (collagen, elastin, reticulin) and sarcoplasmic proteins, contribute to beef's superior water retention capacity (Zurriyati, 2011). As a result, beef meatballs tend to retain more water during cooking, leading to lower cooking loss (Irawati *et al*., 2015). In contrast, the variable hardness was positioned closer to the pea-based meatball without seaweed fortification (PP), and farther from BF and the seaweed-fortified samples (PS1, PS2, PS3). This indicates that the PP sample had a firmer texture than the others. Hardness may be due to a higher proportion of tapioca flour, which functions as both a filler and binder (Pramuditya & Yuwono, 2014). The high amylopectin content in tapioca enables water absorption and dense gel formation, resulting in a firmer, less elastic texture.

The BF sample was closely associated with the variables of chewy texture, raw ingredient flavor, and aftertaste, suggesting that BF had the highest scores for these sensory attributes. Beef contains relatively high levels of protein, which contributes to its chewy texture (Lawrie, 1995). The strong raw ingredient flavor observed in BF may be attributed to beef's high water-holding capacity and pH, which enhance flavor retention. The PP sample was positioned near the variables hard texture and nutty aroma, indicating that the pea-based meatball exhibited a distinct legume aroma. This characteristic aroma is likely due to the use of pea paste as the main ingredient. The firmer texture in PP may have resulted from insufficient grinding of the peas, leading to a coarser particle size and less refined texture. Meanwhile, the seaweed-fortified pea-based samples (PS1–PS3) were located near the variables seasoning aroma and seasoning taste, suggesting that the addition of seaweed and spices improved the overall flavor profile. The use of seasonings in plant-based meatball formulations enhances palatability and helps mask off-flavors commonly found in legumes (Suradi, 2007).

Conclusion

The addition of seaweed flour to pea-based meatballs improves several quality parameters, particularly texture. Seaweed flour enhance the firmness of the product, addressing the typically soft texture of plant-based meatballs. The inclusion of seaweed flours up to 2% (w/w) has no significant effect on cooking loss or fat content, and contributes to increase the moisture content, protein content, water holding capacity, hardness, cohesiveness, springiness, adhesiveness, yield, and crude fiber. The findings indicate that while pea-based formulations offer certain textural and nutritional advantages, further optimization is needed to match the overall quality and consumer acceptance of conventional beef meatballs.

Conflict of interest

The authors have no conflict of interest to declare.

References

Acton, J.C., Ziegler, G.R., Froning, G.W., Burge, D.L., 1983. Functionality of muscle constituents in the processing of comminuted meat products. Crit. Rev. Food Sci. Nutr. 18, 99–121.

Agusman, A., Apriani, S.N.K., Murdinah, M., 2014. Penggunaan tepung rumput laut (*Eucheuma cottonii*) pada pembuatan beras analog dari tepung modified cassava flour (Mocaf). J. Pascapanen Bioteknol. Kelaut. Perikan. 9, 1–10.

Agustini, T.W., Putra, D.A.P., Wijayanto, I., 2015. Pengaruh penambahan karagenan sebagai stabilizer terhadap karakteristik otak-otak ikan kurisi (Nemipterus nematophorus). J. Pengolahan Bioteknol. Has. Perikan. 4, 1–10.

Akbar, A., Sadiq, M.B., Ali, I., Anwar, M., Niaz, M., *et al*., 2019. Lactococcus lactis subsp. lactis isolated from fermented milk products and its antimicrobial potential. J. Food 17, 214–220.

Ames, J.M., 1992. The Maillard reaction. In: Labuza, T.P., Reineccius, G.A., Monnier, V.M., O'Brien, J., Baynes, J. (Eds.), Maillard Reactions in Chemistry, Food and Health. Springer, Boston, pp. 99–153.

AOAC, 1990. Official methods of analysis of the Association of Official Analytical Chemists. 15th ed. AOAC, Washington DC.

Apriyanto, E., Sudjatmiko, S., Susatya, A., Nugoho, P.B., Aulia, E., 2018. The potency of oil palm fruit fiber as growth media for ketapang (*Terminalia catappa*) seedling. Int. J. Agric. For. Plantation 7, 73–78.

Ariyani, M., Ayustaningwarno, F., 2013. Pengaruh penambahan tepung duri ikan lele dumbo (*Clarias gariepinus*) dan bubur rumput laut (*Eucheuma cottonii*) terhadap kadar kalsium, kadar serat kasar dan kesukaan kerupuk. J. Nutr. Coll. 2, 223–231.

Arnyke, E.V., Rosyidi, D., Radiati, L.E., 2014. Peningkatan potensi pangan fungsional naget daging kelinci dengan substitusi wheat bran, pollard dan rumput laut. J. Ilmu-Ilmu Peternakan 24, 56–71.

Aslinah, L.N.F., Ismail-Fitry, M.R., Mat Yusoff, M., 2018. Simultaneous use of adzuki beans (*Vigna angularis*) flour as meat extender and fat replacer in reduced-fat beef meatballs (bebola daging). J. Food Sci. Technol. 55, 3241–3248.

BSN (Badan Standardisasi Nasional), 2014. Bakso daging. Badan Standardisasi Nasional, Jakarta.

Beniwal, A.S., Hardacre, A., Singh, H., Kaur, L., Singh, J., 2021. Meat analogs: Protein restructuring during thermomechanical processing. Compr. Rev. Food Sci. Food Saf. 20, 1221–1249.

Berutu, K.M., Suryanto, E., Utomo, R., 2010. Kualitas bakso daging sapi peranakan ongole yang diberi pakan basal tongkol jagung dan undergraded protein dalam complete feed. J. Peternakan 34, 103–113.

Carballo, J., Barreto, G., Colmenero, F.J., 1995. Starch and egg white influence on properties of Bologna sausage as related to fat content. J. Food Sci. 60, 673–677.

Chen, J., Li, J., Wang, C., Yi, S., Deng, T., Mi, H., Li, X., 2020. Effect of hydrocolloids on gel properties and protein secondary structure of silver carp surimi. J. Sci. Food Agric. 100, 2252–2260.

Cheng, C., Chen, S., Su, J., Zhu, M., Zhou, M., Chen, T., Han, Y., 2022. Recent advances in carrageenan-based films for food packaging applications. Front. Nutr. 9, 1004588.

Gao, X.Q., Kang, Z.L., Li, Y.P., Zhang, W.G., Zhou, G.H., 2015. Combination of κ-carrageenan and soy protein isolate effects on functional properties of chopped low-fat pork batters during heat-induced gelation. Food Bioprocess Technol. 8, 1524–1531.

Genisa, J., Sukendar, N.K., Langkong, J., Abdullah, N., 2015. Analog bakso sehat dari protein kacang merah (*Phaseolus vulgaris* L.). J. Agri Techno. 8, 1–9.

Han, X.Z., Hamaker, B.R., 2002. Partial leaching of granule-associated proteins from rice starch during alkaline extraction and subsequent gelatinization. Starch/Stärke 54, 454–460.

Hannachi, Y., Hafidh, A., Ayed, S., 2019. Effectiveness of novel xerogels adsorbents for cadmium uptake from aqueous solution in batch and column modes: Synthesis, characterization, equilibrium, and mechanism analysis. Chem. Eng. Res. Des. 143, 11–23.

Hidayah, N., Maulina, N.W., Noviandi, C.T., Astuti, A., Dono, N.D., Widyastuti, T., 2022. Chemical composition of brown and red algae from Kelapa Beach, Tuban, East Java and their potential as ruminant feed. IOP Conf. Ser. Earth Environ. Sci. 1114, 012003.

Huang, S.C., Shiau, C.Y., Liu, T.E., Chu, C.L., Hwang, D.F., 2005. Effects of rice bran on sensory and physico-chemical properties of emulsified pork meatballs. Meat Sci. 70, 613–619.

Irawati, A., Warnoto, Kususiah, 2015. Pengaruh pemberian jamur tiram putih (*Pleurotus ostreatus*) terhadap pH, DMA, susut masak dan uji organoleptik sosis daging ayam broiler. J. Sain Peternakan Indones. 10, 125–135.

Jiang, T., Zhao, Y., Huang, M., Zhang, Z., Mao, Y., Zuo, H., 2025. Influence of interactions between drawing soy protein and myofibrillar proteins on gel properties. Foods 14, 1064.

Kalsi, G., Hazarika, U., Baruah, L.D., Bordoloi, P.L., Gogoi, M., 2025. Comprehensive review of carrageenan's multifaceted role in health and food systems. Discov. Food 5, 405.

Kumar, M., Sharma, B.D., 2004. The storage stability and textural, physico-chemical and sensory quality of low-fat ground pork patties with carrageenan as fat replacer. Int. J. Food Sci. Technol. 39, 31–42.

Kumari, T., Deka, S.C., 2021. Potential health benefits of garden pea seeds and pods: A review. Legume Sci. 3, e82.

Lawrie, R.A., 1995. Ilmu daging. Parakkasi, A. (Ed.). Universitas Indonesia Press, Jakarta.

Lévesque, B., Gervais, M.C., Chevalier, P., Gauvin, D., Anassour-Laouan-Sidi, E., Gingras, S., Fortin, N., Brisson, G., Greer, C., Bird, D., Gauvin, D., 2014. Prospective study of acute health effects in relation to exposure to cyanobacteria. Sci. Total Environ. 2014, 466–467, 397–403.

Li, N., Zhang, M., Li, P., Wang, D., 2025. Effects of the trehalose and star anise extract on the water retention of pork meatballs and the functional structure of myofibrillar protein. J. Sci. Food Agric.

Li, S., Bao, Z., Sun, N., He, X., Jiang, P., Lin, S., 2022. Contribution of κ-/ι-carrageenan on the gelling properties of shrimp myofibrillar protein and their interaction mechanism exploration. J. Sci. Food Agric. 103, 524–533.

Lomartire, S., Gonçalves, A.M.M., 2023. Algal phycocolloids: Bioactivities and pharmaceutical applications. Mar. Drugs 21, 384.

Mamat, H., Matanjun, P., Ibrahim, S., Abdul Hamid, M., Md Amin, S.F., Rameli, A.S., 2013. The effect of seaweed composite flour on the textural properties of dough and bread. J. Appl. Phycol. 26, 1057–1062.

Martins, J.T., Vicente, A.A., Fasolin, L.H., Bourbon, A.I., Pinheiro, A.C., 2018. Protein-based structures for food applications: From macro to nanoscale. Front. Sustain. Food Syst. 2, 77.

Monsalve-Atencio, R., Sanchez, K., Chica, J., Camaño, J., Saldarriaga, J., Quintero-Quiroz, J., 2021. Effect of the drying method and texture improvers on reconstitution behavior of yoghurt powder: physical and microbiological properties. J. Vitae 28, 1–14.

Ockerman, H.W. 1985. Quality control of Post-Mortem Muscle Tissue Dept of Animal Sciences. The Ohio State University; Columbus, OH, USA.Osen, R., Eisner, P., Toelstede, S., Schweiggert-Weisz, U., 2015. Effect of high moisture extrusion cooking on protein–protein interactions of pea (*Pisum sativum* L.) protein isolates. Int. J. Food Sci. Technol. 50, 1390–1396.

Öztürk, T., Turhan, S., 2020. Physicochemical properties of pumpkin (*Cucurbita pepo* L.) seed kernel flour and its utilization in beef meatballs as a fat replacer and functional ingredient. J. Food Process. Preserv. 44, e14695.

Pangastuti, H.A., Affandi, D.R., Ishartani, D., 2013. Karakterisasi sifat fisik dan kimia tepung kacang merah (*Phaseolus vulgaris* L.) dengan beberapa perlakuan pendahuluan. J. Teknosains Pangan 2, 1–8.

Pramuditya, G., Yuwono, S.S., 2014. Penentuan atribut mutu tekstur bakso sebagai syarat tambahan dalam SNI dan pengaruh lama pemanasan terhadap tekstur bakso. J. Pangan Agroindustri 2, 200–209.

Pugalenthi, M., Vadivel, V., Gurumoorthi, P., Janardhanan, K., 2004. Comparative nutritional evaluation of little known legumes, Tamarindus indica, Erythrina indica and Sesbania bispinosa. Trop. Subtrop. Agroecosyst. 4, 107–123.

Purwanto, A.P., Ali, A.A., Herawati, N.H., 2015. Kajian mutu gizi bakso berbasis daging sapi dan jamur merang (*Volvariella volvaceae*). J. Sagu 14, 1–8.

Salehi, F., 2019. Effect of common and new gums on the quality, physical, and textural properties of bakery products: A review. J. Texture Stud. 51, 361–370.

Sari, W.K., Sari, N.I., Leksono, T., 2021. Pengaruh penambahan tepung rumput laut (Eucheuma sp.) terhadap mutu dan karakteristik amplang ikan tongkol (*Euthynnus affinis*). J. Teknol. Ind. Pertan. Indones. 13, 9–15.

Shariffa, Y.N., Karim, A.A., Fazilah, A., Zaidul, I.S.M., 2009. Enzymatic hydrolysis of granular native and mildly heat-treated tapioca and sweet potato starches at sub-gelatinization temperature. Food Hydrocoll. 23, 434–440.

Sun, L., Ye, X., Zeng, X., Li, H., Liu, S., Cao, J., Liu, X., Xu, Q., 2024. Research progress on plant-based glue in meat substitutes: main components, formation mechanisms, challenges, and development. Int. J. Food Sci. Technol. 59, 8656–8667.

Suradi, K., 2007. Tingkat kesukaan bakso dari berbagai jenis daging melalui beberapa pendekatan statistik. J. Ilmu Ternak 7, 110–123

Tapotubun, A.M., 2018. Komposisi kimia rumput laut (*Caulerpa lentillifera*) dari perairan Kei Maluku dengan metode pengeringan berbeda. J. Pengolah. Has. Perikan. Indones. 21, 13–23.

Triyannanto, E., Febrisiantosa, A., Kusumaningrum, A., Amri, A.F., Fauziah, S., Sulistyono, E.P., *et al*., 2022. The quality characteristics of ready-to-eat empal gentong affected by meat pre-cooking. Food Sci. Anim. Resour. 42, 557–565.

Utafiyani, Y., Yusasrini, N.L.A., Ekawati, I.G.A., 2018. Pengaruh perbandingan tepung kacang hijau (*Vigna radiata*) dan terigu terhadap karakteristik bakso analog. J. Ilmu Teknol. Pangan 7, 12–22.

Utama, C.S., Sulistiyanto, B., Rahmawati, R.D., 2020. Kualitas fisik organoleptis, hardness dan kadar air pada berbagai pakan ternak bentuk pellet. J. Litbang Provinsi Jawa Tengah 18, 43–53.

Van Der Meer, M., Fischer, A.R.H., Onwezen, M.C., 2022. Same strategies – different categories: An explorative card-sort study of plant-based proteins comparing omnivores, flexitarians, vegetarians and vegans. Appetite 180, 106315.

Wang, Y., Zheng, D., Ji, H., Li, X., Svensson, B., Bai, Y., Jin, Z., 2020. Thermophilic 4-α-glucanotransferase from Thermoproteus uzoniensis retards the long-term retrogradation but maintains the short-term gelation strength of tapioca starch. J. Agric. Food Chem. 68, 5658–5667.

Wi, G., Bae, J., Kim, H., Cho, Y., Choi, M.J., 2020. Evaluation of the physicochemical and structural properties and the sensory characteristics of meat analogues prepared with various non-animal based liquid additives. Foods 9, 461.

Windsor, N., Rubiales, D., Bridges, W., Boyles, R., Thavarajah, D., Boatwright, L., 2024. Characterizing dry pea (*Pisum sativum* L.) for improved nutritional traits and the potential for biofortification. Legume Science 6, 250.

Zhang, X., Xue, J., Yanniotis, S., Guo, D., Mandala, I., 2017. The effect of salt concentration on swelling power, rheological properties and saltiness perception of waxy, normal and high amylose maize starch. Food Funct. 8, 3792–3802.

Zhang, Y., Zhang, J., Chen, Q., He, N., Wang, Q., 2022. High-moisture extrusion of mixed proteins from soy and surimi: Effect of protein gelling properties on the product quality. Foods 11, 1397.

Zhu, W., Bu, Y., Li, X., Sun, X., Shiuan, D., Huan, H., Li, J., 2019. Effects of hydroxyl radical induced oxidation on water holding capacity and protein structure of jumbo squid (*Dosidicus gigas*) mantle. Int. J. Food Sci. Technol. 54, 2159–2168.

Zurriyati, Y., 2011. Palatabilitas bakso dan sosis sapi asal daging segar, daging beku, dan produk komersial. J. Peternakan 8, 49–57.