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Production and Characterization of Edible Beads Using Natural Calcium Extracted from Catfish Bone Powder through Direct and Reverse Gelation Techniques

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Abstract

The significant number of catfish heads generated during processing represents an underutilized resource rich in calcium and proteins. This study aimed to valorize this waste by extracting natural calcium from catfish bone powder and using it to produce edible alginate beads through direct and reverse gelation techniques. Natural calcium was extracted from 0.4% catfish bone powder using microwave digestion at 300 W for 60 seconds with 0.1 M citric acid. In the direct gelation process, alginate solutions at concentrations of 0.4, 0.5, 0.6, 0.8, and 1.0% were dropped into the extracted calcium solution. The lowest concentration to form a gel was 0.5%, but 1.0% was selected for firmer gel formation. Reverse gelation involved dropping calcium solutions with 0, 10, 20, 30, and 40% gelatin into a 0.5% alginate solution, with the 20% gelatin formulation exhibiting the highest hardness and thus was chosen. The characteristics and stability of edible beads produced by both gelation methods were comparatively evaluated. Chemical composition analysis showed higher protein and calcium levels in reverse gelation beads than in direct gelation beads. Additionally, beads from reverse gelation demonstrated superior textural properties and greater acceptability compared to direct gelation beads. Both bead types showed limited storage stability when kept in water, as measured by changes in diameter. However, storage in 0.1 M citric acid solution significantly improved their stability.

In conclusion, natural fish bone calcium effectively induces alginate gelation by both direct and reverse methods. Reverse gelation produced nutritionally enhanced, acid-stable edible beads, presenting a promising approach for fish waste valorization with potential applications in acidic beverages. This method offers both economic and environmental benefits by transforming fish processing by-products into functional ingredients.

Keywords: *fish bone powder; alginate bead; direct bead formation; reverse bead formation; gelatin; natural calcium*

1. Introduction

Fish processing industries generate substantial amounts of waste, including fish bones, which are often discarded despite being rich in valuable nutrients such as calcium. Repurposing fish bone waste as a natural calcium source not only enhances the value of this by-product but also promotes sustainable and environmentally friendly practices. One innovative application of this calcium-rich material lies in the formation of edible beads, which are commonly produced through the immediate gelation of polysaccharides when introduced into a solution. Among these polysaccharides, sodium alginate is frequently used due to its ability to rapidly gel in the presence of divalent ions, particularly calcium ions (Ca^{2+}). This process, known as “direct bead formation,” allows for the efficient creation of gel beads and has been widely studied (Liu et al., 2025; Wang et al., 2023).

Direct bead gelation is based on the principle of ionotropic gelation, in which a gelling agent (alginate) is dropped into an inducing solution containing calcium ions, resulting in the immediate formation of spherical gel beads. Innovative creation for this kind of product can be enhanced by adding several additives, e.g., colors and flavors (Dey, & Nagababu, 2022). Adding fruit juice is used to produce beads with fruity flavors, while combining broth flavors is commonly used to produce imitation fish roe. Currently, several products are available commercially, produced using this technique (Lee, & Mooney, 2012). Commercial edible beads are commonly induced by calcium ions derived from food grade chemicals. However, this method has certain limitations, such as affecting taste and lacking essential nutrition. The use of natural Ca^{2+} derived from fish bone powder to induce alginate gelation has been developed (Chanshotikul et al., 2024). Digestion of fish bone powder using citric acid solution under microwave heating has been documented to release the available calcium ion (Sriuttha et al., 2024). This strategy results in higher calcium content in the edible beads. In addition, natural calcium intake is likely more effective for physiological functions than chemical calcium ions (Malde et al., 2010). This provides an alternative way to utilize fishery by-products for food innovation. However, calcium ion uptake into the edible bead is still limited when produced by direct bead formation. Therefore, application of reverse bead gelation might overcome this limitation and could increase the protein and calcium content in the beads.

Reverse bead gelation could be achieved by dropping a calcium solution into a sodium alginate solution in order to obtain the gelling beads. Calcium solution would likely to be embedded into the bead resulted in more calcium uptake by this technique. However, applying this technique can be challenging due to the limited ability of the calcium solution to effectively penetrate the alginate solution. This limitation is primarily attributed to the difference in densities between the two solutions, with the alginate solution typically having a higher density than the calcium solution. Therefore, it is necessary to adjust the density of the calcium solution to ensure successful bead formation.

A promising strategy to overcome this limitation involves incorporating gelatin to increase the density of the calcium solution. The success of reverse bead formation depends on achieving an appropriate density of the dropping solution so that it sinks into the gelling medium. If the alginate solution is used in the gelling medium, higher density calcium solution must be obtained. Modifying the density of the calcium solution by adding food-based ingredients is, especially commercial gelatin, a practical alternative. Gelatin is a protein-based powder derived from denatured collagen through thermal or acid treatments (Obas et al., 2024; Sinthusamran et al., 2014). It is widely used in food applications as thickening or gelling agent, depending on its concentration (Chen et al., 2025; Wang, & Hartel, 2022; Zou et al., 2024). Incorporating gelatin at suitable concentration into the calcium solution would enhance its density, allowing the formation of alginate beads via reverse bead gelation (Sagiri et al., 2015). Thus, using gelatin to modify fish bone-derived natural calcium solution likely offers a promising approach for producing edible beads via the reverse gelation technique.

2. Objectives

The objectives of this research were to investigate the feasibility of using natural calcium derived from catfish bone powder to induce gelation of alginate solutions for edible bead production, employing both direct and reverse gelation methods. Comparative characterization of the nutritional composition, physical attributes, textural properties, and sensory acceptability of the produced beads was performed. Finally, the storage stability of the edible beads prepared by both gelation techniques in either water or citric acid was evaluated.

3. Materials and Methods

3.1 Materials

Catfish heads, which were by-products from a processing industry, were kindly provided by a catfish farm in Nong Khai Province, Thailand. Fish bone powder was produced from catfish heads following the method described by Hemung (2013). Sodium alginate powder was purchased from Bangkok Chemical Co., Ltd. (Bangkok, Thailand), and gelatin powder was obtained from Union Science Co., Ltd. (Khon Kaen, Thailand). Citric acid was sourced from R&B Food Supply Co., Ltd. (Ayutthaya, Thailand). Red grape juice and sparkling green tea with KYOHO flavor were purchased from a department store.

3.2 Extraction of Calcium Solution from Fish Bone Powder

A calcium solution was prepared according to the method of Sriuttha et al. (2024) using fish bone powder. Fish bone powder (0.40 g) and 0.1 M citric acid were dissolved in 100 mL of DI water, heated in a microwave at 300 W for 60 s to extract calcium, and then allowed to cool.

3.3 Production of Edible Beads by Direct Bead Gelation Technique

3.3.1 Effect of Alginate Concentration on Hardness of Bead Formed by Direct Bead Gelation Technique

Sodium alginate solutions were prepared at concentrations of 0.4, 0.5, 0.6, 0.8, and 1.0% by dissolving the powder in DI water and heating using a microwave at 300 W for 60 seconds according to previous protocol (Chanshotikul et al., 2024). The obtained solution was dropped into the calcium solution with the dropping rate of about 20 drops/min and left for 10 min before recovering the bead. Edible beads were stored in extracted calcium solution at 4°C for 12 h for further determination of hardness as described previously (Wang et al., 2024). The hardness of the samples was measured using the texture analyzer (TA XT Plus, Godalming, UK). Samples (diameter approximately 4.00 ± 0.20 mm) were compressed to 50% of their height using a P/50 probe. The average values were reported based on 10 measurements.

3.3.2 Production of Edible Bead by Direct Bead Gelation

The natural calcium solution was prepared by digesting catfish bone powder (0.4 g) in 0.1 M citric acid using microwave power 300 W for 60 seconds. Then, the alginate solution (1.0%) was prepared according to the optimal concentration reported previously (Chanshotikul et al., 2024). This solution was dropped into the natural calcium solution. Edible beads were stored in the calcium solution at 4°C for 12 h for further characterization

3.4 Production of Edible Beads by Reverse Bead Gelation Technique

3.4.1 Effect of Gelatin Concentration on Reverse Bead Gelation

Gelatin powder at varying concentrations (0, 10, 20, 30, and 40%) was added to the calcium solution extracted using the conditions described in section 3.3.2. The resulting calcium/gelatin solutions were then cooled to room temperature prior to measuring the density by weighing the solution for 50 mL using balance (Mettler ME204, Greifensee, Switzerland). The obtained calcium/gelatin solutions were individually dropped into the alginate solution (0.5%) in order to form edible beads by reverse bead gelation. The obtained beads were stored in calcium solution at 4°C for 12 h prior to determining the hardness as mentioned previously in section 3.3.1.

3.4.2 Production of Edible Bead by Reverse Bead Technique

The calcium solution extracted using the same protocol in section 3.3.2 was mixed with gelatin to obtain a final concentration of 20%. This solution was dropped into the sodium alginate solution (0.5%) at a dropping rate of about 20 drops/min. Edible beads were stored in calcium solution at 4°C for 12 h for further characterization.

3.5 Characterization of Edible Beads

3.5.1 Water Content

The surface moisture of the prepared edible beads, after storage at 4 °C for 12 h, was removed using filter paper. Subsequently, the beads without surface water were weighed. The water content was calculated using the following equation:

$$\text{Moisture content (\%)} = ((M_0 - M_1)/M_0) \times 100$$

where M_0 is the original weight of the edible bead, and M_1 is the dry gel bead weight obtained after freeze-drying.

3.5.2 Protein Content

The protein content of the edible beads was analyzed by Central Laboratory (Thailand) Co., Ltd., Khon Kaen branch, following the AOAC (2019) standard method 981.10.

3.5.3 Calcium Content

The calcium content of edible beads was determined by Central Laboratory (Thailand) Co., Ltd., Khon Kaen branch, using the method described in *Analyst*, August 1994, Vol. 119, pp. 1683-1686.

3.5.4 Size and Sphericity Factor

A vernier caliper was used to measure the average size of the edible beads ($n = 10$). The sphericity factor of edible beads was calculated by the following formula (Lee et al., 2013):

$$\text{Sphericity factor} = (D_{Max} - D_{Min})/(D_{Max} + D_{Min})$$

where D_{Max} is the maximum diameter of the edible bead (mm), and D_{Min} is the minimum diameter of the edible bead (mm).

3.5.5 Texture Analysis

Texture profile analysis (TPA) of the uniform-sized beads was performed using a texture analyzer (TA XT Plus, Godalming, UK). Each sample was compressed to 50% of its original height using a P/50 cylindrical probe. The pre-test, test, and post-test speeds were all set at 1 mm/s, with slight modifications to the method previously described by Geng et al., (2024). Textural parameters, including hardness, springiness, cohesiveness, gumminess, and chewiness, were reported as averages from 10 measurements.

3.5.6 Swelling Ratio

The beads with an exact weight (W_0) were soaked in 20 mL of DI water at 4°C for 24 h. Thereafter, the beads were removed from the solution and wiped with filter paper to remove surface water before weighing (W). The swelling ratio of the beads over time was computed using the following equation:

$$\text{Swelling ratio\%} = (W/W_0) \times 100$$

3.5.7 Stability of Edible Bead

Edible beads (10 samples per treatment) were soaked in DI water with and without citric acid (0.1 M) and stored at 4°C for 7 days. Changes in bead diameter and appearance were monitored. Average diameters were recorded based on 3 measurements.

3.5.8 Sensory Analysis

Panelists were selected through individual interviews to ensure they had no allergies to the product or its ingredients, were not pregnant, did not smoke, and voluntarily agreed to participate in the study. Additionally, all participants were undergraduate students in the Food Technology and Innovation program at Khon Kaen University, representing the consumer group.

Sensory evaluation was performed using 30 panelists (semi-trained judges) to assess appearance, aroma, taste, and flavor, texture, elasticity, consistency, aftertaste, and overall acceptance using a 9-point hedonic scale (Sahin et al., 2023). To minimize bias during the evaluation, the edible beads were prepared in sparkling green tea, and three-digit random codes were assigned to the product presentation to the panelists.

Ethical considerations: This study involved human participants for sensory evaluation and was submitted to the Khon Kaen University Ethics Committee for Human Research, Panel 2 (HE672159). The committee reviewed the submission and confirmed that it met the criteria for exemption in accordance with their guidelines. As per institutional policy, a memorandum of acknowledgment was issued in lieu of a certificate of approval, serving as official confirmation that the study qualifies as exempt research.

3.6 Statistical Analysis

Data were expressed as mean ± standard deviation (SD) from at least three independent replicates, unless otherwise specified. Statistical analyses were performed using one-way analysis of variance (ANOVA) and independent sample t-tests via SPSS software (SPSS Inc., Chicago, IL, USA). Duncan's multiple range test was applied to identify significant differences among means at a confidence level of $p < 0.05$.

4. Results and Discussion

4.1 Edible Beads Produced by Direct Bead Gelation Technique

4.1.1 Effect of Alginate Solution on Bead Formation

The ability of bead formation induced by calcium solution was tested at different alginate concentration in order to identify the minimum concentration required for direct bead formation. The results showed that the alginate solution at 0.4% was unable to form a gel (Table 1). In contrast, increasing the alginate concentration to 0.5% enabled gel formation, establishing this as the minimum threshold for alginate gelation. It was observed that forming beads at higher alginate concentrations resulted in firmer gel beads. This was supported by the increase in hardness of alginate bead from 0.132 to 1.224 N. A higher concentration of alginate could form thicker and stronger alginate gels, and a similar observation was reported previously (Geng et al., 2024). Therefore, the minimum alginate concentration of 0.5% was subsequently chosen to produce reverse beads in the next experiment.

Table 1 Effect of alginate concentrations on edible bead formation (direct bead) and their hardness

Alginate (%)	Ability to form bead	Hardness (N)
0.4	Unable to form bead	-
0.5	Able to form bead	0.132 ± 0.008^{d}
0.6	Able to form bead	0.312 ± 0.032^{c}
0.8	Able to form bead	0.562 ± 0.052^{b}
1.0	Able to form bead	1.224 ± 0.091^{a}

Different superscript letters (a–d) within the same column indicate statistically significant differences at $p < 0.05$.

Table 2 Effect of gelatin concentration in fish bone suspension on gel formation (reverse bead) and their hardness

Gelatin (%)	Density (kg/L)	Ability to form bead	Hardness (N)
0	1.05 ± 0.01^{e}	Unable to form	-
10	1.08 ± 0.00^{d}	Unable to form	-
20	1.11 ± 0.00^{c}	Able to form	0.021 ± 0.003^{a}
30	1.12 ± 0.01^{b}	Able to form	0.011 ± 0.003^{b}
40	1.15 ± 0.01^{a}	Able to form	0.012 ± 0.001^{b}

Different superscript letters (a–d) within the same column indicate statistically significant differences at $p < 0.05$.

4.2 Edible Beads Produced by Reverse Bead Gelation Technique

4.2.1 Effect of Gelatin in Calcium Solution on Reverse Bead Formation

To produce reverse gel beads, the calcium solution must be dropped into the alginate solution. However, directly dropping the digested fish bone powder into the 0.5% alginate solution was not successful in forming gel beads. Instead, a sheet-like gel formed on the surface of the sodium alginate solution rather than spherical beads. This may have occurred due to the lower density of the digested fish bone solution (1.05 kg/L) compared with the 0.5% alginate solution (1.06 kg/L). Thus, the droplets of fish bone powder solution could not sink into the alginate solution. Only a thin gel layer formed on the top of the solution. To overcome this, the density of the fish bone powder solution was increased by adding gelatin, allowing the calcium solution to sink into the alginate solution. As a result, reverse beads were successfully formed by adding more than 20% gelatin to the fish bone powder solution (Table 2). Rapid bead formation and immediate sinking were observed when the gelatin concentration increased to 30 and 40%. This indicates that a higher density of dropping phase facilitated reverse bead formation. Using this technique, the entire fish bone suspension was likely trapped within the spheric bead. This technique also allows for the incorporation of proteins from both gelatin and fish bone powder, as well as natural calcium, into the beads. Thus, the nutritional value of these beads was ultimately enhanced. It was observed that the hardness of the reverse beads decreased as the gelatin concentration increased (Table 1). This may be attributed to the higher gelatin concentrations forming gels, which retard the migration of calcium ions needed to interact with alginate at the outer shell of the beads. The critical concentration for gelatin gelation has been reported to be above 14% (Qiao et al., 2013). In addition, the gelling state occurs during the cooling of the gelatin solution, as applied in reverse bead gelation in this study. It has been reported that high protein concentrations affect the ability of sodium alginate to form gels (Munialo et al., 2014).

4.3 Appearance of Edible Beads Formed by Direct and Reverse Bead Formation

Edible beads formed by both techniques exhibited a spherical shape, as shown in Figure 1. The spherical shape of the direct beads was formed through the immediate gelation of the alginate solution when dropped into the calcium solution derived from digested fish bone powder in the presence of 0.1 M citric acid. This suggests that the calcium concentration in the solution was sufficient to induce alginate gelation. A spherical shape was also observed in edible beads formed using the reverse bead technique. In addition, larger beads appeared to be obtained with this method (Figure 1b). Therefore, the difference in bead diameter formed by the two techniques should be measured to confirm this observation.

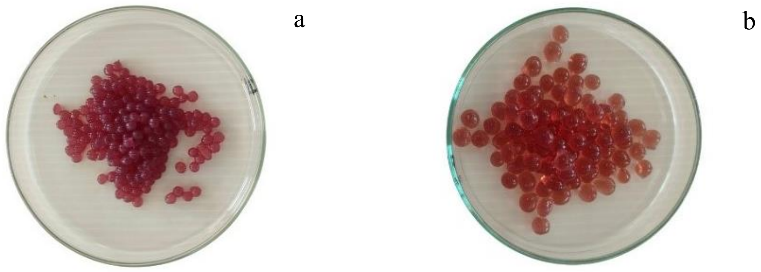

Figure 1 Appearance of edible beads formed using direct bead gelation (a) and reverse bead gelation (b)

Table 3 Physical properties of edible beads

Bead type	Size (mm)			Sphericity factor (SF)	Swelling ratio (%)
	Minimum	Maximum	Mean		
Direct bead	3.91±0.22[b]	4.05±0.24[b]	3.98±0.23[b]	0.017±0.012[a]	133.60±6.30[a]
Reverse bead	6.02±0.31[a]	6.24±0.34[a]	6.13±0.32[a]	0.019±0.012[a]	123.88±9.27[b]

Different superscript letters (a–b) within the same column indicate statistically significant differences at $p < 0.05$.

4.4 Characterization of Edible Beads Formed by Direct and Reverse Bead Formation

4.4.1 Physical Properties

It can be clearly seen that the size of reverse beads was larger than that of direct beads. This was evidenced by the larger diameter as shown in Table 3. Direct gelation produced beads with an average diameter of 3.98 mm, whereas reverse gelation resulted in significantly larger beads, approximately 1.5 times bigger (6.13 mm). This result confirmed the hypothesis, which was previously based on visual observation of the beads with the naked eye. This expansion was likely due to the thicker alginate layer formed during reverse gelation compared with the calcium layer in the direct bead gelation.

Normally, the spherical shape of bead is observed when the sphericity factor (SF) is lower than 0.05 (Lee et al., 2013). The data in Table 3 clearly showed that the obtained bead had a spherical shape, as indicated by the SF values of approximately 0.02. In addition, similar SF values were found regardless of the type of beads. This result suggested that the SF value corresponded well with the spherical shape observed in the beads' appearance.

The swelling behavior of beads provides information regarding water uptake during storage. It is important for evaluating the stability of beads in desirable solution. It can be seen that direct beads absorbed more water than reverse beads. This was supported by the higher swelling ratio, approximately 34%, as shown in Table 3. It can be hypothesized that direct beads could absorb more flavors or dissolved compounds from the solution than the reverse beads. The water uptake ability may mainly result from the water absorption capacity of the alginate gel. This is because the entire direct bead is composed of alginate gel, whereas in the reverse beads, alginate gel is present only in the outer shell. Therefore, the ability of the reverse beads to absorb water was lower than that of the direct beads.

4.4.2 Chemical Properties

The edible beads formed by reverse bead formation showed lower moisture content than that of direct beads, resulting in higher total solids (Table 4). This solid content was primarily attributed to gelatin and fish bone suspension. This was expected to carry a higher content of both protein and calcium.

The reverse beads contained approximately 4.1% protein, which was significantly higher than that of the direct beads (Table 4). Specifically, the protein content of the reverse beads was 51.25-fold higher. This increase in protein may be attributed to the incorporation of gelatin and fish bone suspension. Protein content was still observed in the fish bone powder even though alkaline extraction was used for its removal (Hemung, 2013). Thus, the addition of gelatin not only increased the density of the calcium solution but also increased protein content in the reverse bead.

Table 4 Chemical composition of edible beads

Bead type	Moisture content (%)	Protein content (%)	Calcium content (mg/kg sample)
Direct bead	86.80±0.39^{a}	0.08±0.01^{b}	0.67±0.01^{b}
Reverse bead	83.85±0.26^{b}	4.10±0.06^{a}	4.76±0.01^{a}

Different superscript letters (a–b) within the same column indicate statistically significant differences at $p < 0.05$.

Table 5 Textural properties of edible beads

Bead type	Hardness (N)	Springiness	Cohesiveness	Gumminess (N)	Chewiness (N)
Direct bead	0.36±0.05^{b}	0.42±0.05^{a}	0.26±0.03^{b}	0.09±0.02^{b}	0.04±0.01^{b}
Reverse bead	0.70±0.11^{a}	0.43±0.04^{a}	0.38±0.04^{a}	0.27±0.06^{a}	0.12±0.03^{a}

Different superscript letters (a–b) within the same column indicate statistically significant differences at $p < 0.05$.

The calcium content in reverse beads also increased by approximately 7.1-fold through reverse bead formation. This emphasized the effectiveness of calcium enrichment by reverse bead technique. The role of gelatin in improving the nutritional quality of gel-based food products was also reported previously (Mada Hatsa et al., 2024). The increase in either protein or calcium in the reverse beads would be beneficial for biological functions when this technology is applied to innovative food products. The bioavailability of natural calcium from fish bone powder has been tested based on enzymatic reactions (Hemung, 2013).

4.4.3 Textural Properties

The edible beads formed by the reverse gelation technique showed a significant increase in textural properties for almost all attributes ($p < 0.05$). The hardness, cohesiveness, gumminess, and chewiness of the reverse beads were higher compared to those of the direct beads (Table 5). However, springiness, which refers to the ability of the gel to recover its shape after compression by the probe, remained unaffected by the gelation technique. The increase in the textural properties of the reverse beads may be due to the higher solid content, particularly gelatin, which provides double gelation during bead formation. The core of the bead is composed of gelatin gel, while the outer shell is made of alginate gel. This was consistent with a previous report (Geng et al., 2024).

4.4.4 Stability

The stability of edible beads stored in DI water and 0.1 M citric acid solution at 4°C for 7 days was evaluated based on changes in diameter. The results, as shown in Table 6, revealed that the storage medium significantly affected the structural integrity of the beads formed by different gelation techniques. The direct beads exhibited a gradual increase in diameter when stored in water, reaching a maximum of 5.07 mm after 6 days. This expansion likely resulted from continuous water absorption caused by the difference in osmotic pressure between the internal gel matrix and the surrounding environment. However, the beads stored in citric acid solution shrank slightly over time, suggesting improved structural stability. This could be attributed to the ionic interactions between alginate and hydrogen ions in the acidic environment, which tightened the gel network, as previously reported (Nordin et al., 2018). In contrast, reverse beads exhibited even greater swelling when stored in water and broke apart after 3 days. This instability may be due to the less compact structure formed by the dual network of gelatin and alginate. When reverse beads were stored in citric acid solution, their structural integrity was maintained throughout the storage period. Only a slight increase in diameter was observed after 2 days, which corresponded to limited water penetration into the outer alginate layer a behavior consistent with the findings of Lai et al., (2020), who demonstrated enhanced hydrogel stability in acidic solutions.

These findings confirmed that gelation techniques and storage media play a crucial role in the structural stability of edible beads. Although reverse bead formation improved nutritional value, its water stability was limited. Nevertheless, this limitation could be addressed by switching the storage medium to an edible acid, such as citric acid, to enhance bead stability and shelf life. This also indicated that the beads formed in this study may be suitable for application in acidic beverages.

Table 6 Diameter (mm) of edible beads after storge in DI water and citric acid (0.1 M) at 4°C for 7 days

Bead type	Storage solution	Storage time (day)						
		1	2	3	4	5	6	7
Direct bead	DI water	$4.16{\pm}0.20^{aC}$	$4.75{\pm}0.32^{aB}$	$5.01{\pm}0.34^{aA}$	$5.05{\pm}0.32^{aA}$	$4.85{\pm}0.23^{aAB}$	$5.07{\pm}0.25^{aA}$	$5.03{\pm}0.19^{aA}$
	Citric acid	$4.07{\pm}0.21^{aA}$	$3.75{\pm}0.20^{bB}$	$3.48{\pm}0.09^{bC}$	$3.80{\pm}0.56^{bB}$	$3.49{\pm}0.14^{bC}$	$3.52{\pm}0.19^{bC}$	$3.46{\pm}0.11^{bC}$
Reverse bead	DI water	$5.90{\pm}0.36^{aC}$	$8.21{\pm}0.19^{aB}$	$9.93{\pm}0.13^{aA}$	-	-	-	-
	Citric acid	$5.96{\pm}0.52^{aB}$	$7.36{\pm}0.51^{bA}$	$7.80{\pm}0.51^{bA}$	$7.77{\pm}0.64^{A}$	$7.79{\pm}0.68^{A}$	$7.96{\pm}1.05^{A}$	$7.74{\pm}0.77^{A}$

Different superscript letters (a–b) within the same column in each bead type indicate statistically significant differences at $p < 0.05$.
Different superscript letters (A–C) within the same row indicate statistically significant differences at $p < 0.05$.

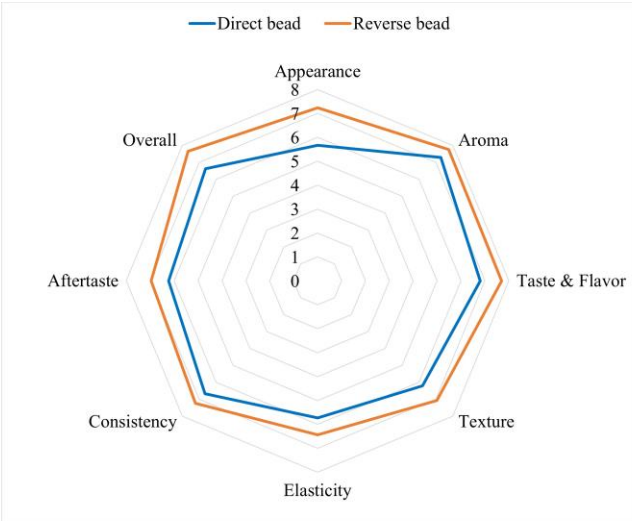

Figure 2 Sensory evaluation of edible beads prepared using different gelation techniques

4.4.5 Sensory Evaluation

The sensory evaluation of edible beads produced using different gelation techniques was conducted after topping them onto sparkling green tea. This assessment helped determine the compatibility of the beads for further application as fruity beads. Consumer preference showed significantly higher favorability for the reverse bead over the direct beads, particularly in terms of appearance ($p < 0.05$). Furthermore, the overall acceptance of the reverse beads was significantly higher than that of the direct beads (Figure 2), likely influenced by its larger size, as shown in Table 3. This suggests that a larger bead size may enhance consumer perception. Preferences regarding aroma, taste, flavor, and aftertaste were also significantly higher for the reverse beads ($p < 0.05$), possibly due to more effective absorption of volatile compounds and flavors from the sparkling green tea. In addition, preference scores for texture, elasticity, and consistency were higher for the reverse beads, correlating with the higher textural properties observed in TPA measurements.

These results indicate that the reverse beads exhibited superior sensory quality over the direct beads in all evaluated attributes ($p < 0.05$), highlighting the effectiveness of the reverse gelation technique in enhancing hedonic qualities.

5. Conclusion

This study established the feasibility of using catfish bone-derived calcium to induce sodium alginate via direct gelation and a modified reverse gelation technique. Modification of reverse gelation by adding 20% gelatin to the calcium solution produced beads with enhanced nutritional (higher protein and calcium), texture, and sensory properties. While water stability was initially lower, citric acid storage effectively prolonged the shelf life of these edible beads. This research offers an innovative and sustainable approach for valorizing fishery by-products into nutrient-rich gels with significant potential in functional food applications.

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7. CRediT Statement

Worawut Thammawong: Investigation, Methodology, Formal analysis, Writing – original draft.
Bung-Orn Hemung: Project Administration, Funding acquisition, Supervision.
Nachayut Chanshotikul: Conceptualization, Methodology, Validation, Resources, Data Curation, Writing – Review & Editing.

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