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Physicochemical properties of surimi made from edible insects using washing and pH shift methods

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A R T I C L E I N F O

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A B S T R A C T

Edible insects, characterized by their eco-friendly nature and high nutrient value, are promising protein sources. Therefore, we aimed to assess the suitability of insects as source ingredients for surimi, a widely-used, intermediate food material. Mealworm (*Tenebrio molitor* L.) and two-spotted cricket (*Gryllus bimaculatus* L.) surimi were prepared, and their physicochemical and rheological properties were examined. Myofibrillar protein-rich fractions were obtained using the washing and pH shift methods. For the pH shift method, the myofibrillar proteins were extracted at acid (pH 2) or alkaline (pH 11) conditions, and surimi gel was prepared by heating myofibrillar protein-rich fractions. The pH shift method resulted in a higher surimi yield from edible insects than the washing method, whereas the washing method resulted in a higher surimi yield from tilapia (*Oreochromis niloticus*) and chicken breast (*Gallus gallus domesticus*). After acid treatment, lipid oxidation increased in all samples; however, edible insect surimi exhibited lower oxidation levels than tilapia and chicken breast surimi. Insect proteins, except for acid-treated mealworm proteins, successfully formed gel structures upon heating, resulting in softer gels than those obtained from tilapia and chicken breast. Consequently, the pH shift method resulted in elevated insect surimi yield, and the alkaline treatment was more appropriate for producing fine-quality edible insect surimi. Our study demonstrates the usefulness of edible insects as surimi ingredients, particularly for soft-gel food production. These findings emphasize the innovative application of edible insects in the food industry, suggesting the possibility of expanding their use as alternative protein food ingredients.

1. Introduction

The global population, which is continuously increasing, is projected to reach approximately 9.7 billion by 2050 (UN. United Nations Department of Economic and Social Affairs, 2022). This entails a consequent rise in the demand for food production; however, the decrease in the availability of land resources will exacerbate food scarcity (Premalatha et al., 2011). Edible insects serve as an alternative protein resource and are associated with several advantages, including low greenhouse gas emissions and small land area requirements (Oonincx and De Boer, 2012). Insects also have a higher feed conversion efficiency than traditional livestock, i.e., they convert feed into body mass more effectively, further highlighting their eco-friendly attribute (Nino et al., 2021). Edible insects are rich in proteins, and their consumption is associated with various potential health benefits, including antioxidant, antimicrobial, and anti-inflammatory effects. Despite these advantages, the use of insects as a food resource has been limited by psychological aversion in consumers (Megido et al., 2016). Therefore, insect processing, via methods such as pulverization and specific substance extraction, has gained research attention as it improves consumer acceptance (Borges et al., 2022; Queiroz et al., 2023). However, edible insect-based protein extraction, as well as the development of protein-based food materials from the extracted protein, has not been sufficiently explored.

Surimi, a protein-rich food material primarily obtained from fish fillets, is an important food product, with approximately 822,000 metric tons produced worldwide in 2018 (Jaziri et al., 2023). Repeated washing of homogenized fish fillets produces concentrated myofibrillar proteins, which are the primary components of surimi (Yoshie-Stark et al., 2009). Myosin, a representative myofibrillar protein in muscle tissues, predominantly contributes to the gelling and water-binding properties of surimi (Jin et al., 2007). Upon heating, the abundant

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myofibrillar proteins in surimi form three-dimensional gel network structures, making surimi an excellent intermediate ingredient for seafood-based gel production (Sun et al., 2021).

Conventionally, surimi is produced using washing methods; however, the pH shift extraction method can be used to increase myofibrillar protein yield (Undeland et al., 2002). The pH shift method involves adjusting the pH (acidic, <3.5; or basic, >10.5) to a pH at which myofibrillar proteins are soluble in water, followed by the precipitation of myofibrillar protein-enriched fractions at the isoelectric point (Y. Zhou et al., 2021). As compared to traditional washing methods, the pH shift method is associated with a higher myofibrillar protein yield and the production of protein of higher purity as insoluble and unwanted residues are removed during processing (Kristinsson and Liang, 2006). Therefore, washing and pH shift methods have been evaluated as novel methods of surimi processing using non-seafood resources such as beef, pork, and chicken (Kenney et al., 1992; Park et al., 1996; Srinivasan and Xiong, 1996). In recent years, overexploitation of fish resources has decreased the biodiversity of marine fisheries and aquatic ecosystems (Trindade-Santos et al., 2020). Fish farms have been able to meet up with fish the resource requirements for the nourishment of the world's populations; however, aquatic farming is aggravating ocean pollution, especially through the release of plastic debris from equipment (Krüger et al., 2020). Thus, replacement of fish fillets with edible insects for surimi production can help resolve this global environmental issue; however, to date, approaches involving the use of edible insects as ingredients for surimi, with the substitution of traditional ingredients, have been lacking.

Therefore, in this study, we aimed to elucidate the characteristics of surimi produced from two popular edible insect species, mealworm larvae (*Tenebrio molitor* L.) and the two-spotted cricket (*Gryllus bimaculatus* L.). Both the washing and pH shift methods were evaluated for insect surimi production, and various physicochemical properties of surimi sol and gel were analyzed to determine the optimal conditions for their production. Through this study, we hope to expand the industrial use of edible insects and describe the basic physicochemical characteristics of insect proteins.

2. Materials and methods

2.1. Sample materials

All chemicals used in this study were procured from Sigma-Aldrich (St. Louis, MO, USA). Live mealworms (M) and two-spotted crickets (C) were sourced from a domestic farm in Gyeonggi-do, South Korea. The tilapia (T) (*Oreochromis niloticus*) used in this study was sourced from Indonesia and sold by Costco (Seoul, South Korea), and chicken breast (B) (*Gallus gallus domesticus*) was purchased from Harim (Seoul, South Korea).

2.2. Determination of raw material protein characteristics

2.2.1. Amino acid composition

Following the acid hydrolysis of the lyophilized raw materials, their amino acid composition was analyzed as previously described by Jang et al. (2009). Each analysis was performed in triplicate.

2.2.2. Preparation of the myofibrillar protein solution

The raw materials were finely minced using a mortar and pestle, and the minced samples were homogenized in phosphate buffer (0.02 mmol/L, pH 7.0) at a 1:10 (w/v) ratio for 2 min using the T 25 digital ULTRA-TURRAX® device (IKA, Staufen, Germany). To obtain sarcoplasmic proteins, the homogenate was centrifuged at 8000 rpm for 20 min at 4 °C (2236R; Labogene, Lillerød, Denmark) (Dara et al., 2021). The sarcoplasmic proteins were extracted twice using a 0.02-mmol/L phosphate buffer (pH 7) containing 0.6 mol/L KCl to obtain a myofibrillar protein solution. The extract obtained was centrifuged at 8000 rpm for 20 min at 4 °C, and the supernatant was filtered using a 0.45-μm syringe filter.

2.2.3. Surface hydrophobicity, and total and active sulfhydryl (SH) content

The surface hydrophobicity of the myofibrillar protein solution was determined as previously described by Hu et al. (2022); in addition, the total and active SH content of the myofibrillar protein solution was determined using 5,5′-dithiobis (2-nitrobenzoic acid) (DTNB), as described by Gao et al. (2019).

2.3. Preparation of surimi sol

2.3.1. Washing method

Traditional surimi is produced by concentrating fish protein through four washing cycles, resulting in substantial water wastage (Moosavi-Nasab et al., 2005). In addition, repeated washing cycles reduce myofibrillar protein yield. To address this, Jin et al. (2007) proposed using two washing cycles instead of four and found that this did not reduce surimi quality. Therefore, we adopted the two washing cycle method in this study.

Raw materials were homogenized with distilled water (1:4, w/v) for 3 min and filtered through an 18-mesh test sieve (CG-20341-18; Chung Gye, Seoul, South Korea). To remove exoskeleton residue, the mealworm and cricket homogenates were further filtered through a 35-mesh test sieve (CG-20341-35; Chung Gye). The resulting filtrate was centrifuged at 10,000 rpm for 25 min at 4 °C, and the precipitate obtained was collected. Then, the precipitate was re-homogenized with distilled water and centrifuged again at 10,000 rpm for 25 min at 4 °C to obtain the final precipitate.

2.3.2. pH shift method

The raw materials were homogenized with water (1:9, w/v) for 3 min. Next, the homogenate obtained was filtered through an 18-mesh test sieve, and then further filtered through a 35-mesh test sieve to remove exoskeleton residues. The pH of the filtrate was adjusted to 2 or 11 using 1 N HCl or NaOH solution and verified using a pH meter (pHi 510, Beckman Coulter, Brea, USA). After pH adjustment, the mixture was centrifuged at 10,000 rpm for 25 min at 4 °C. Then, the aqueous layer was collected, and the pH was adjusted to 5 for protein precipitation. After 30 min, the precipitate was collected through centrifugation at 10,000 rpm for 25 min at 4 °C.

2.3.3. Surimi sol preparation

Myofibrillar-rich protein fractions were extracted from four materials (mealworm, cricket, tilapia, and chicken breast) using three methods i.e., the washing method (W), the acid-pH shift method with extraction at pH 2 (2), and the alkaline-pH shift method with extraction at pH 11 (11). To prepare surimi sol (paste), the pH and moisture content of the myofibrillar-rich protein fractions were adjusted to 7 and 85%, respectively.

2.4. Differential scanning calorimetry (DSC)

The thermal properties of the surimi sol were analyzed using a differential scanning calorimeter (Discovery SDT 650, TA Instruments, New Castle, DE, USA). Each sample (10 mg) was sealed in an aluminum pan, and a temperature sweep test was conducted from 25 to 95 °C at a scan rate of 10 °C/min.

2.5. Determination of the physicochemical and rheological characteristics of surimi gel

2.5.1. Surimi gel preparation

Surimi gel was prepared following the method described by Jin et al. (2007). Surimi sol was packed into polyvinylidene chloride casings (18-mm diameter) and cooked in a water bath at 90 °C for 60 min. After cooking, the gels obtained were cooled and stored in a refrigerator (4 °C)

for 1 day before use.

2.5.2. Sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS–PAGE)

SDS–PAGE was conducted following the method described by Pan et al. (2018). The raw materials and cooked surimi gel samples were homogenized in a 5% SDS solution (1:9, w/v) and centrifuged at 5000 rpm for 10 min. The supernatant was mixed with SDS-PAGE loading buffer and heated in a water bath at 95 °C for 3 min before electrophoresis. In total, 20 μg of protein was loaded onto polyacrylamide gels consisting of a 5% stacking gel and a 10% running gel, set up in a Mini-PROTEAN 3 electrophoresis module (Bio-Rad Laboratories Inc., Hercules, CA, USA). A protein marker (Xpert Prestained Protein Marker, Katy, TX, USA) was loaded into the first lane, and 20 μg of sample protein was loaded into the other lanes. The samples were electrophoresed at 60 V for 30 min and then at 100 V for 80 min. Coomassie brilliant blue R-250 solution (0.25 g/100 mL, dissolved in 50% methanol, 10% acetic acid, and 40% deionized water) was used for band staining.

2.5.3. Proximate compositions

Proximate composition analyses were conducted according to the standard methods proposed by the American Association of Cereal Chemists (AACC, 2000). Moisture (Method 44-15A), ash (Method 08-01), fat (Method 30-25), and crude protein (Method 46-13) content were measured, and the carbohydrate content was determined by subtracting the total content of the other components from 100%.

2.5.4. Yield, cooking loss, and water activity (a_w)

Surimi gel yield was calculated as a ratio of the weight of surimi gel to that of the raw materials. Cooking loss was calculated as the percentage weight loss from the surimi sol (uncooked) to the surimi gel (cooked) (Jin et al., 2007). The a_w of the surimi gel was measured at 25 °C using an electronic dew point water activity meter (WA-160A, AMITTARI, Guangzhou, China). Measurements were conducted in triplicate.

2.5.5. Water-holding capacity (WHC)

WHC was measured following the method described by Jin et al. (2007), with slight modifications. The surimi gel was cut into 5-mm-thick slices, placed in centrifuge tubes, and centrifuged at 2000×*g* for 15 min. After centrifugation, the weight of the gel slice was measured again. WHC was determined based on the water loss of samples after centrifugation (n = 4).

2.5.6. Thiobarbituric acid reactive substances (TBARS)

TBARS values were determined following the method described by Chaijan et al. (2021), with slight modifications. Surimi (1 g) was homogenized with distilled water (1:4, w/v), and the supernatant was collected after centrifugation. Then, the supernatant (450 μL) was mixed with 900 μL of TBARS reagent (containing 0.375% TBA, 15% TCA, and 0.25 N HCl) and heated in a water bath at 95 °C for 10 min until a pink color appeared. After cooling on ice, the solution was centrifuged at 3600 rpm for 20 min at 25 °C. The absorbance of the supernatant was measured at 532 nm. A standard curve was prepared using malonaldehyde (0–10 ppm), and the TBARS value was represented as mg of malonaldehyde equivalent per kg of sample (n = 4).

2.5.7. Measurement of surimi sample color values

The color of each surimi sample was determined using the CIE L*a*b* system. Color values were measured five times for each sample using a colorimeter (CM-36dG; KONICA MINOLTA, Tokyo, Japan).

2.5.8. Field emission scanning electron microscopy (FE–SEM)

Surimi surface images were analyzed using FE–SEM (ZEISS GeminiSEM 560; ZEISS, Oberkocchen, Germany) (Pyo et al., 2024). The surimi samples were fixed in 2.5% glutaraldehyde dissolved in 0.2 M phosphate buffer (v/v, pH 7.2). Then, the samples were rinsed in distilled water for 1 h and dehydrated using graded ethanol (50%, 70%, 80%, 90%, and 100% ethanol). After dehydration, the samples were mounted on stubs and coated with platinum using a sputter coater (BAL-TEC/SCD 005 sputter coater; BALTEC AG, Pfäffikon, Zürich, Switzerland). Surface images were observed at an accelerating voltage of 5 kV and sample images were captured at a magnification of × 3000.

2.5.9. Texture profile analysis (TPA)

The surimi was cut into cubes (1 × 1 × 1 cm^3) (Hou et al., 2016) and subjected to TPA at the Chung-Ang University BT Research Facility Center using a texture analyzer (TA.XTplus, Stable Micro Systems Ltd., Godalming, UK). A 36-mm P/36R probe was used to conduct TPA. The pre-test and post-test speeds were 5 and 1 mm/s, respectively, at a compression rate of 50%. The interval between the first and second compression was 5 s, and the trigger force was 10 g (n = 6).

2.5.10. Dynamic rheological properties

The dynamic oscillatory test for surimi was conducted using an ARES-RFS III rheometer (TA Instruments, New Castle, DE, USA). Under the frequency sweep mode, the elastic and viscous properties of gels were described using the storage modulus (G') and loss modulus (G''), respectively, as previously described (Ji et al., 2017). The test was performed at 4 °C, at a frequency range of 1–100 rad/s. To ensure that all dynamic measurements were conducted within the linear viscoelastic region, the amplitude strain was set to 5%. Dynamic parameters were measured as a function of each frequency (ω) (Huang et al., 2019).

2.6. Statistical analysis

For comparison of the mean values of all analyzed parameters, one-way analysis of variance was performed using the IBM SPSS Statistics software version 28 (IBM, Armonk, NY, USA). Duncan's test was used to determine significant differences between multiple groups. P-values less than 0.05 were considered statistically significant.

3. Results and discussion

3.1. Raw material protein physicochemical properties

3.1.1. Amino acid composition of raw materials

The amino acid compositions of mealworm, cricket, tilapia, and chicken breast are presented in Table 1. We found a similar edible insect amino acid content to that reported by Ghosh et al. (2017), with glutamic acid being the most abundant amino acid in all materials, followed by aspartic acid, leucine, lysine, alanine, and arginine. Overall, total amino acid content was higher in tilapia and chicken breast than in edible insects. Edible insects exhibited lower content for lysine, an essential amino acid; this is similar to the findings of Oliveira et al. (2024) who reported that lysine is a limiting amino acid in mealworm and cricket. Comparatively, tyrosine content was higher in edible insects. Not only does the amino acid composition of raw materials affect their nutritional value, it can also affect the physicochemical properties of surimi. For example, each amino acid has a unique pK_a and isoelectric point that can affect protein solubility. In gel foods, ionic strength, hydrophobicity, and disulfide bonds arbitrate protein crosslinking (Li et al., 2019), and the ratio of the contents of hydrophobic amino acids, such as glycine, alanine, valine, phenylalanine, isoleucine, leucine, and proline, was slightly higher in the two insects than tilapia and chicken breast. Furthermore, we also focused on the unusually high proline content of mealworms as Y. Zhou and Yang (2020) found that proline can break myosin disulfide bonds in solution.

Table 1
Amino acid composition of raw materials (mg/g dry basis).

	Mealworm	Cricket	Tilapia	Chicken breast
Aspartic acid	35.55 ± 0.34^{c} [a]	45.60 ± 1.83^{b}	87.11 ± 0.59^{a}	84.80 ± 0.29^{a}
Glutamic acid	59.05 ± 1.12^{c}	70.78 ± 0.98^{b}	146.31 ± 0.65^{a}	148.75 ± 0.93^{a}
Serine	20.21 ± 0.12^{d}	22.57 ± 0.52^{c}	32.44 ± 0.38^{b}	34.33 ± 0.00^{a}
Histidine	13.83 ± 0.01^{c}	11.90 ± 0.46^{d}	21.16 ± 0.09^{b}	28.66 ± 0.75^{a}
Glycine	22.10 ± 0.02^{d}	28.46 ± 0.94^{c}	43.73 ± 1.33^{a}	37.40 ± 0.49^{b}
Threonine	19.64 ± 0.30^{c}	22.83 ± 0.00^{b}	37.99 ± 0.25^{a}	40.63 ± 1.32^{a}
Arginine	26.87 ± 1.15^{c}	39.87 ± 0.76^{b}	55.09 ± 0.54^{a}	52.77 ± 0.50^{a}
Alanine	32.95 ± 0.09^{c}	45.95 ± 1.39^{b}	53.98 ± 0.59^{a}	52.60 ± 0.23^{a}
Tyrosine	32.62 ± 0.51^{a}	32.28 ± 0.94^{a}	29.24 ± 0.02^{b}	31.77 ± 0.25^{a}
Valine	28.67 ± 0.34^{d}	30.31 ± 0.21^{c}	43.34 ± 0.15^{b}	45.90 ± 0.41^{a}
Phenylalanine	17.71 ± 0.23^{c}	21.23 ± 0.33^{b}	38.86 ± 0.30^{a}	37.70 ± 0.34^{a}
Isoleucine	21.29 ± 0.16^{d}	22.79 ± 0.07^{c}	43.81 ± 0.01^{b}	46.39 ± 0.40^{a}
Leucine	34.42 ± 0.46^{d}	39.23 ± 0.68^{c}	72.19 ± 0.23^{b}	74.86 ± 0.29^{a}
Lysine	20.44 ± 0.33^{c}	26.41 ± 0.54^{b}	69.08 ± 1.68^{a}	71.67 ± 0.38^{a}
Proline	35.80 ± 0.57^{a}	20.89 ± 0.36^{b}	24.97 ± 3.12^{b}	27.65 ± 3.23^{ab}

Data are presented as mean ± standard deviation.
[a] Different superscripts within rows represent significant differences at $p < 0.05$.

3.1.2. *Surface hydrophobicity and total/active SH content of myofibrillar proteins*

The highest surface hydrophobicity value (50.27 μg) was observed in tilapia (Fig. 1A), indicating an abundant unfolding of myofibrillar proteins and surface exposure of non-polar amino acid residues (Dara et al., 2021). The surface hydrophobicity value (39.70 μg) in crickets was approximately twice as high as that in mealworms. Surface hydrophobicity reflects the number of hydrophobic groups distributed on the protein surface, making it a suitable parameter for estimating changes in protein exposure and aggregation (Xu et al., 2019). The tertiary structure of a protein is characterized by hydrophobic interactions, and these play a crucial role in determining myofibrillar protein structural and functional properties, including stability, solubility, and gelation (Dara et al., 2021).

Total SH groups represent all the SH groups in a protein structure, whereas reactive SH groups are chemically-reactive groups that are exposed under non-denaturing conditions. Of all the myofibrillar proteins evaluated, those obtained from mealworms exhibited the lowest total and reactive SH content at 0.91×10^{-5} mol/g and 0.34×10^{-5} mol/g, respectively, whereas chicken breast exhibited the highest total and reactive SH content at 3.75×10^{-5} mol/g and 3.50×10^{-5} mol/g, respectively (Fig. 1B and C). The most significant difference between total and reactive SH content was observed in mealworm myofibrillar proteins; a significant difference between total and reactive SH values suggests that many thiol groups are not exposed on the protein surface but are buried within its structure, indicating a potentially well-folded protein. SH groups are associated with weak secondary bonds that stabilize the tertiary structure of the protein (Wang et al., 2018). Moreover, exposed -SH groups cross-link to form more SS bonds, aggregating proteins into larger particles, making the protein gel denser and increasing gel strength (Ko et al., 2007). Collectively, the low surface hydrophobicity and total/active SH content observed in mealworm myofibrillar proteins were speculated to result in weak surimi gelation power.

3.2. *Thermodynamic properties of surimi sol*

Using DSC, the denaturation transition temperature range and thermal properties of surimi sol during irreversible transformation into surimi gel were determined (Fig. 2). Peak temperature and enthalpy changes vary depending on the stability, composition, and structure of source proteins as the characteristics of heat-induced protein unfolding and denaturation differ (Wu et al., 2020). Quinn et al. (1980) demonstrated that myofibrillar proteins such as myosin, sarcoplasmic proteins, and actin denature at approximately 60 °C, 66 °C, and 83 °C, respectively. As concerns TW and BW samples, TW showed peaks between 55–57 °C and 67–71 °C, while BW showed peaks at 61 °C and 79–81 °C, corresponding to myosin and actin. Although TW exhibited lower peak temperatures, it exhibited similar patterns to BW. In contrast, MW and CW exhibited different denaturation patterns to BW and TW, and this may stem from disparate myofibrillar protein structures in edible insects. Denaturation transition temperatures for T2, T11, B2, and B11 samples were not as distinct as those for TW and BW, suggesting that the proteins unfolded following acid and alkali treatment during the extraction process, altering their thermal denaturation properties. This finding is similar to that reported by Tadpitchayangkoon et al. (2010) for fish sarcoplasmic proteins. MW and M11 exhibited similar patterns, with peak temperatures of 59 °C and 69 °C, and 63 °C and 69 °C, respectively, indicating different thermal properties to those of M2. Similarly, as compared to C2, CW and C11 samples exhibited distinct peaks, indicating that acid treatment may induce unfolding and destruction of the protein structure during the process.

3.3. *Physicochemical and rheological characteristics of surimi gels*

3.3.1. *SDS–PAGE*

The separated protein bands of raw materials and surimi gels prepared under various conditions are presented in Fig. 3. There were no remarkable differences in the protein patterns between T, TW, T2, and T11. In addition, no substantial differences in band intensity were observed and the protein bands showed patterns similar to those of the major components of cod proteins, such as myosin heavy chain (190 kDa), actin (45 kDa), tropomyosin (35 kDa), and myosin light chain (18–25 kDa), as reported by Thorarinsdottir et al. (2002). Chicken breast samples also showed no significant differences in band composition, and

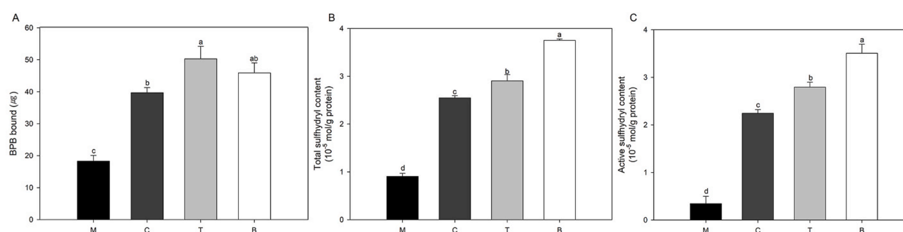

Fig. 1. Hydrophobicity (A) and total (B) and active (C) sulfhydryl content of myofibrillar protein-rich fractions. M: Mealworm; C: cricket; T: tilapia; B: chicken breast.

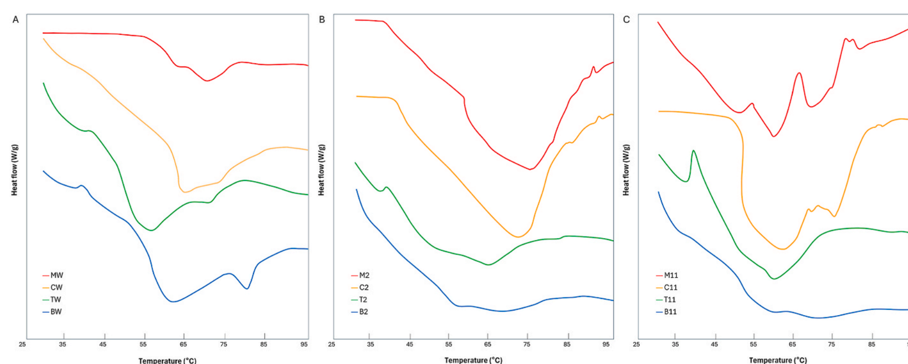

Fig. 2. Differential scanning calorimetric thermograms of surimi sols prepared using the washing method (A), the acid-pH shift method (B), and the alkaline-pH shift method (C). M: Mealworm; C: cricket; T: tilapia; B: chicken breast; W: washing method; 2: acid-pH shift method (pH 2); 11: alkaline-pH shift method (pH 11).

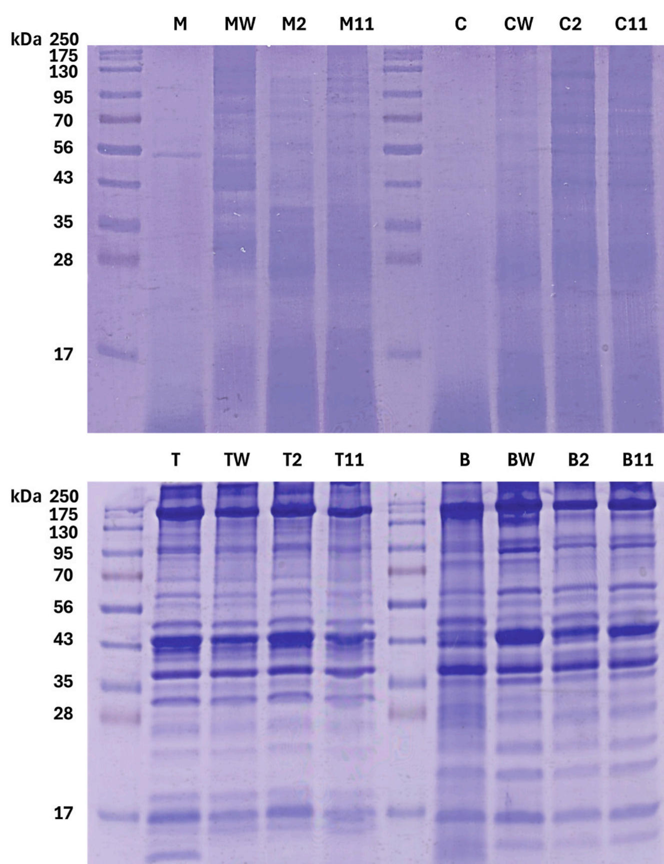

Fig. 3. Sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS-PAGE) images. M: Mealworm; C: cricket; T: tilapia; B: chicken breast; W: washing method; 2: acid-pH shift method (pH 2); 11: alkaline-pH shift method (pH 11).

the thickest actin band (45 kDa) was observed in BW. Raw mealworms were rich in proteins with a molecular weight of 55 kDa and below 15 kDa; raw crickets were also abundant in proteins with molecular weight below 15 kDa. We could not observe clear bands for typical myofibrillar proteins in raw insect samples, and this may indicate low myofibrillar protein content. Due to the lack of myofibrillar proteins, we speculated that insect-based surimi gel would not have a firm texture. Although myofibrillar proteins were not abundant in raw insect materials, the density of high kDa proteins increased in insect surimi gels. This observation indicates that the concentration of macromolecular proteins increased with heat-induced protein denaturation. Of the mealworm-based surimi gel samples, MW exhibited a higher abundance of high molecular weight proteins involved in gel formation than M2 and M11. Particularly, the high molecular weight protein content was lower for M2 samples than for MW and M11, indicating a weak gel formation ability. As opposed to mealworm surimi gels, for cricket surimi gels (C2 and C11), the pH shift method resulted in greater high molecular weight protein content than the washing method. These results indicate that gelling ability and technological properties are dependent on the edible insect species.

3.3.2. Proximate composition of surimi gels

The proximate composition of surimi gel is presented in Supplementary Table 1. Except for M2, surimi gels prepared using the pH shift method exhibited higher protein content than surimi gels prepared using the washing method. Particularly, the alkaline pH shift method resulted in higher protein content, possibly because alkaline conditions break down protein disulfide bonds, resulting in higher protein extraction yields (Yu et al., 2023). Of the surimi gels prepared using the pH shift method, edible insect surimi gels exhibited a substantially higher lipid content. This is because edible insects contain more lipids than other materials (Adeniyi et al., 2012; Koh and Yu, 2015; Zielińska et al., 2015). Unlike the washing method, which involves repeated washing steps to remove impurities, the pH shift method does not include washing steps, resulting in higher lipid, carbohydrate, and ash content. Lipids can negatively affect surimi product quality as lipids interfere with gel formation and cause rancidity; in addition, oxidized lipids interact with proteins, causing denaturation, polymerization, and changes in functional properties (Jin et al., 2007; Somjid et al., 2021). Therefore, additional processes to remove excessive lipids from edible insect surimi may enhance the physicochemical quality of products.

3.3.3. Yield, cooking loss, and physicochemical properties of surimi gels

The yields, cooking loss, moisture content, a_w, and WHC of surimi gels are presented in Table 2. Edible insect surimi gels exhibited lower yields than tilapia and chicken breast surimi gels. This is possibly because a substantial amount of exoskeleton and chitin was removed from the edible insects during the preparation process. Edible insect surimi gels exhibited higher yields when prepared using the pH shift method, while tilapia and chicken breast surimi gels exhibited higher yields when prepared using the washing method. Cooking loss values for tilapia and chicken breast were similar to those reported by Jin et al. (2007) for surimi gel prepared from pollock and chicken breast using the washing method. During the heat-induced gelation process, proteins bind with water molecules and trap other components to form a gel network structure (Guo et al., 2019). However, continued heating can cause the breakdown of the protein gel network and weaken protein–-water interactions, resulting in cooking loss due to the expulsion of water molecules (X. Zhou et al., 2020). Of all the sample groups, MW exhibited the lowest cooking loss, indicating significant water retention during heating and resulting in the formation of surimi gel with a high moisture content.

Table 2
Yield, cooking loss, moisture content, water activity, water-holding capacity, and TBARS values for surimi gels.

Sample	Yield (%)	Cooking loss (%)	Moisture content (%)	Water Activity	Water holding capacity (%)	TBARS (mg MDA eq/kg)
MW [a]	14.00 ± 2.33^{f} [b]	26.47 ± 1.03^{d}	84.91 ± 0.41^{a}	0.93 ± 0.01^{def}	80.51 ± 0.88^{bc}	0.52 ± 0.03^{g}
M2	19.13 ± 1.78^{f}	42.19 ± 1.67^{ab}	70.12 ± 1.24^{c}	0.92 ± 0.00^{fg}	- [c]	0.80 ± 0.09^{g}
M11	16.00 ± 2.25^{ef}	42.87 ± 1.96^{a}	74.88 ± 0.03^{b}	0.94 ± 0.00^{abc}	85.65 ± 4.68^{b}	0.76 ± 0.02^{g}
CW	13.73 ± 1.27^{f}	33.73 ± 1.73^{c}	84.79 ± 0.79^{a}	0.90 ± 0.00^{h}	75.80 ± 6.15^{c}	1.35 ± 0.06^{f}
C2	28.79 ± 0.22^{d}	33.64 ± 1.46^{c}	70.34 ± 0.21^{c}	0.92 ± 0.00^{ef}	81.21 ± 4.03^{b}	1.37 ± 0.21^{f}
C11	20.30 ± 0.17^{e}	37.91 ± 1.09^{bc}	74.21 ± 0.89^{b}	0.92 ± 0.00^{ef}	84.90 ± 4.03^{b}	1.23 ± 0.09^{f}
TW	54.87 ± 0.20^{a}	37.63 ± 2.11^{c}	83.72 ± 0.21^{a}	0.91 ± 0.00^{g}	94.67 ± 1.03^{a}	2.75 ± 0.41^{c}
T2	37.69 ± 1.81^{c}	37.29 ± 1.55^{c}	62.19 ± 0.84^{d}	0.93 ± 0.00^{cdef}	98.60 ± 0.46^{a}	6.78 ± 0.09^{a}
T11	31.70 ± 1.70^{d}	42.09 ± 0.06^{ab}	59.44 ± 0.96^{a}	0.94 ± 0.00^{ab}	99.11 ± 0.19^{a}	1.89 ± 0.09^{e}
BW	45.13 ± 1.80^{b}	36.43 ± 0.40^{c}	71.27 ± 0.40^{c}	0.93 ± 0.00^{bcdf}	95.97 ± 2.53^{a}	2.43 ± 0.37^{cd}
B2	42.03 ± 1.74^{bc}	43.20 ± 0.91^{a}	62.69 ± 1.20^{d}	0.94 ± 0.00^{a}	98.49 ± 0.81^{a}	3.12 ± 0.27^{b}
B11	38.70 ± 2.50^{c}	37.46 ± 0.90^{c}	62.02 ± 1.43^{de}	0.93 ± 0.00^{abcd}	98.68 ± 0.54^{a}	2.29 ± 0.15^{d}

[a] M: mealworm; C: cricket; T: tilapia; B: chicken breast; W: washing method; 2: acid-pH shift method (pH 2); 11: alkaline-pH shift method (pH 11).
[b] Data are presented as mean ± standard deviation. Different superscripts within columns represent significant differences at $p < 0.05$.
[c] We could not analyze M2 because it did not form a gel structure.

As compared to the washing method, the pH shift method reduced moisture content in surimi gels made from all the evaluated materials. The pH shift process denatures and coagulates proteins; when pH is out of the optimal range, proteins unfold and aggregate, exposing previously hidden hydrophobic regions within their structure (Yu et al., 2023). This leads to stronger protein–protein binding than protein–water binding, resulting in greater moisture loss. T11 exhibited the lowest moisture content (59.44 ± 0.96%), and this may be associated with its firm gel strength.

A_w measures the amount of water molecules available in food for microbial growth and chemical reactions (Aberoumand, 2010). Therefore, determining the a_w of food is crucial for assessing sensitivity to quality degradation, predicting shelf life, and identifying storage conditions. When a_w is lower than 0.93, the growth of most bacteria is significantly retarded (Park, 2015). In this study, most of the surimi gel samples satisfied this criterion, implying that they exhibited a more prolonged shelf-life than raw materials.

WHC, which is a function of protein–water interactions, gel structure, and water distribution, refers to the ability of a gel to bind water (X. Zhou et al., 2020). Additionally, WHC is a crucial characteristic of heat-induced protein gels and impacts meat product quality. Andrés-Bello et al. (2013) reported that pH greatly influences physical properties such as WHC, meat tenderness, and color. Generally, high pH, high protein content, and low moisture content are closely related to high WHC and shear force in meat (Jin et al., 2007). This possibly explains why alkaline-treated surimi gels made from cricket, tilapia, and chicken breast exhibited the highest WHC values. After heating, M2 formed a sol rather than a gel; thus, WHC measurement was not possible. The failure of M2 to form insect surimi gel may be due to its particularly high proline content as proline can inhibit the formation of disulfide bonds in myosin (Zhang et al., 2024; Y. Zhou et al., 2020). Moreover, the pK_a of the carboxyl acid side chain in proline is 1.99, which is almost the same as the pH for acid treatment; Ibrahimi et al. (2020) reported that protonated proline can induce changes in its interfacial interactions with other molecules. However, further studies are required to elucidate the interactions between protonated proline and myofibrillar proteins during mealworm protein extraction.

TBARS is widely used to evaluate the extent of lipid oxidation in food samples (Zhao et al., 2019). Surimi gels prepared through the acid treatment method exhibited the highest TBARS values for all materials, which is consistent with the findings of Kristinsson and Liang (2006). The lower TBARS values associated with the washing method were likely due to the removal of a substantial portion of secondary oxidation products through repeated washing. As compared to alkaline treatments, acidic treatments promote lipid hydrolysis, thereby increasing the solubility of free fatty acids that are prone to oxidation; moreover, protein denaturation under acidic conditions exposes lipids to oxidative environments (Kim et al., 2016). In this study, among the acid-treated samples, M2 surimi gel exhibited the lowest TBARS value (0.80 mg MDA eq/kg, $p < 0.05$), while T2 exhibited the highest TBARS value (6.78 mg MDA eq/kg) among the acid-treated samples. The low TBARS value for edible insect surimi is possibly due to the presence of antioxidant compounds in the edible insects. Cho and Ryu (2021) found that the TBARS value for meat analog products decreased with increase in mealworm larvae content, and this may be related to the presence of antioxidant compounds in mealworms, as indicated by their high DPPH activity. These compounds can prevent lipid oxidation during the production of edible insect-based foods (Rumpold and Schlüter, 2015). Although insect surimi exhibited a lower initial TBARS than tilapia and chicken breast surimi, the abundance of lipids in insect materials poses a latent threat to the shelf-life of the product. Thus, there is need to identify or develop appropriate additives that will impede lipid oxidation in insect surimi.

3.3.4. Color values of surimi gels

Table 3 shows the color values of surimi gels. The lightest (L*) surimi gel was observed with TW and BW, and samples were slightly darkened when the pH shift method was used. Cricket surimi gel samples exhibited high redness (a*), while chicken breast surimi gels exhibited

Table 3
Color values of surimi gels.

	L* [b]	a*	b*
MW [a]	45.43 ± 0.42^{i} [c]	1.82 ± 0.17^{c}	4.94 ± 0.23^{e}
M2	50.55 ± 0.06^{g}	0.98 ± 0.01^{d}	9.15 ± 0.02^{d}
M11	47.46 ± 0.14^{h}	0.73 ± 0.04^{e}	5.54 ± 0.10^{e}
CW	54.81 ± 0.03^{f}	2.99 ± 0.02^{b}	−1.33 ± 0.02^{i}
C2	57.68 ± 0.10^{e}	3.73 ± 0.01^{a}	−0.42 ± 0.04^{h}
C11	58.40 ± 0.11^{e}	3.11 ± 0.02^{b}	0.35 ± 0.05^{c}
TW	70.01 ± 0.12^{b}	−2.29 ± 0.04^{i}	−0.07 ± 0.16^{gh}
T2	68.51 ± 0.92^{c}	−2.42 ± 0.16^{i}	5.04 ± 0.25^{e}
T11	69.42 ± 1.33^{bc}	−2.30 ± 0.17^{i}	4.17 ± 0.99^{f}
BW	72.19 ± 1.44^{a}	−0.77 ± 0.10^{f}	11.02 ± 0.67^{b}
B2	65.11 ± 1.69^{d}	−1.55 ± 0.28^{h}	9.79 ± 0.49^{c}
B11	69.06 ± 0.89^{bc}	−1.10 ± 0.18^{g}	11.65 ± 0.41^{a}

[a] M: mealworm; C: cricket; T: tilapia; B: chicken breast; W: washing method; 2: acid-pH shift method (pH 2); 11: alkaline-pH shift method (pH 11).
[b] L*: lightness; a*: redness; b*: yellowness.
[c] Data are presented as mean ± standard deviation. Different superscripts within columns represent significant differences at $p < 0.05$.

high yellowness (b*). For tilapia surimi samples, T2 and T11 were significantly more yellow (b*) than TW ($p < 0.05$). Surimi samples made from edible insects were darker than those made from tilapia and chicken breast. This darkening is likely due to enzymatic browning reactions that occur during protein extraction and processing following exposure of insect components to air. These reactions are caused by some enzymes, including tyrosinase, which has been identified as the main cause of browning in mealworm larvae during grinding (Janssen et al., 2017). Crickets exhibited higher L* values than mealworms, possibly due to their lower enzyme content; this leads to a greater enzymatic browning reaction extent in mealworms. To improve the color traits of insect surimi, antioxidative agents such as glutathione (Wu, 2013) could be superinduced in surimi production. pH adjustments could be a countermeasure against the enzyme reaction in insect materials. Mealworms exhibit the highest browning reaction extent at pH 6 (Janssen et al., 2017). Likewise, in this study, insect surimi manufactured using the pH shift method was lighter in color than that manufactured using the washing method. Color characteristics, especially whiteness, are important factors that drive consumer preferences for surimi and surimi products (C. Liu et al., 2021a). Therefore, further research is needed to develop methods of inhibiting browning enzymes in edible insects for the production of lighter-colored insect surimi.

3.3.5. *Surface microstructures of surimi gels*

An FE–SEM analysis was performed to compare surimi gel surface microstructures (Fig. 4). All surimi gel samples exhibited a network structure, indicating the inherent elasticity of the gels (Somjid et al., 2021). Surimi made from edible insects, as well as that made from tilapia and chicken breast, using the washing method exhibited a porous network, with fine gaps consisting of very small spherical and fibrous forms dispersed throughout the gel structure. In contrast, T2, T11, B2, and B11 samples exhibited larger coagulation areas and, in some regions, larger and deeper localized holes, suggesting greater WHC (Somjid et al., 2021). According to Bertram et al. (2008), larger and deeper pores result from water trapped within the cooked gel network. Additionally, T11 and B11 samples exhibited a unique fibrous bundle structure, indicating that myofibrillar proteins were more efficiently aggregated, leading to protein cross-linking and the formation of a denser gel network (Alvarez et al., 1999).

3.3.6. *Texture profiles of surimi gels*

The texture profiles of surimi gels are shown in Table 4. Hardness, defined as the force required to achieve deformation (Y. Liu et al., 2021b), varied significantly between samples. The lowest hardness value was observed for CW (94.06 g), while the highest was observed for T2 (7444.48 g). The elasticity of surimi gel and its products primarily depends on the characteristics of the gel network (Chandra and Shamasundar, 2015), in addition, Gabriele et al. (2001) reported that springiness is an important indicator of surimi gel elasticity. Among the samples evaluated, T11 and T2 exhibited higher springiness values. Cohesiveness, which is a measure of the ability of a material to withstand internal bonds until deformation (Szczesniak, 2002), was also significantly higher in T11 and T2. The cohesiveness value for TW (0.43) was similar to that previously reported for Golden Pompano surimi prepared using the washing method (Y. Liu et al., 2021b). Additionally, T2 and T11 exhibited lower adhesiveness values than the other edible insect surimi samples. Chewiness, defined as the energy required to masticate food to a swallowable state, was approximately 2000-fold higher for T11 (11,725.29 g) than for C2 (5.69 g). Collectively, tilapia and chicken breast surimi gels prepared using the pH shift method exhibited significantly firmer structures than those prepared using the washing method. This finding suggests that pH changes increase the hardness of tilapia and chicken breast surimi gel, which is likely associated with lower moisture content. Although a stronger gel is desirable for fish products, excessively hard or tough food requiring a high mastication force may cause discomfort to the consumer (Yang et al., 2024). Therefore, appropriate gel texture characteristics should be selected, and edible insects could form softer-textured surimi gels than chicken breast and tilapia.

3.3.7. *Dynamic rheological properties of surimi gels*

The viscoelastic measurements, G' and G'', are parameters used to characterize myosin proteins and are indicative of gel strength. G' measures the stored energy and characterizes the elasticity, while G'' measures the energy dissipated as heat and characterizes the viscosity (Cao et al., 2012). Dynamic frequency sweeps were conducted to determine the frequency dependence of the elastic and viscous moduli, and the G' and G'' profiles of surimi gels prepared under the different conditions are depicted in Fig. 5. In MW and C11, the mid-frequency crossover indicated elastic dominance, with G' surpassing G'', while in CW and BW, the mid-frequency crossover indicated viscous dominance, with G'' surpassing G'. The crossover at mid-frequency signifies a transition from elasticity to viscosity or vice versa and can be used to identify the gelation point of the material, indicating structural changes in the protein gel (Gazo Hanna et al., 2023). Aside from the four samples that passed the crossover point, the remaining surimi gel samples exhibited a higher slope for G', characteristic of materials like gels or solid-like substances. Aside from the four aforementioned samples and MW, the

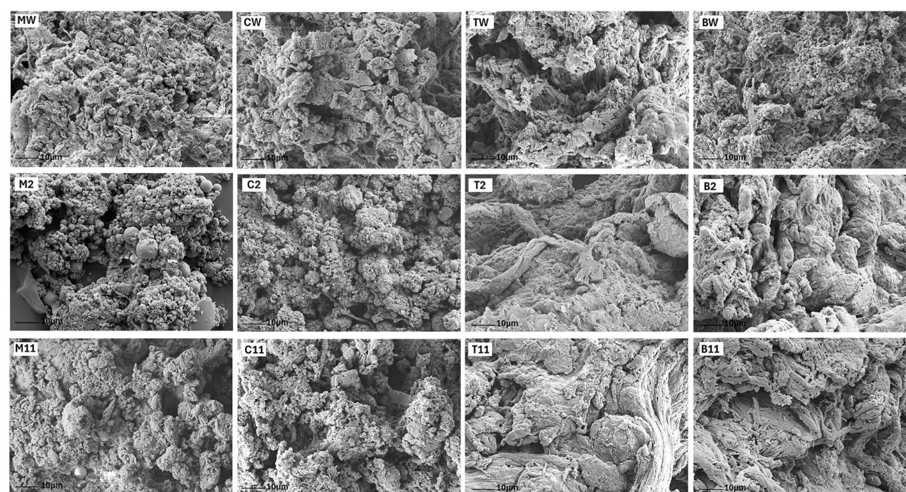

Fig. 4. Field emission scanning electron microscopic images of surimi gels. Images were captured at × 3,000. M: Mealworm; C: cricket; T: tilapia; B: chicken breast; W: washing method; 2: acid-pH shift method (pH 2); 11: alkaline-pH shift method (pH 11).

Table 4
Texture profiles of surimi gels.

	Hardness (g)	Springiness	Cohesiveness	Gumminess (g)	Chewiness (g)	Adhesiveness (g)	Resilience
MW [a]	171.80 ± 29.47^{e} [b]	0.87 ± 0.08^{d}	0.41 ± 0.12^{de}	69.64 ± 22.17^{d}	61.26 ± 23.55^{c}	−11.32 ± 3.99^{b}	0.10 ± 0.03^{e}
M2	- [c]	–	–	–	–	–	–
M11	104.58 ± 13.59^{e}	0.54 ± 0.15^{d}	0.22 ± 0.03^{f}	23.68 ± 6.03^{d}	13.34 ± 6.89^{c}	−18.63 ± 6.73^{c}	0.06 ± 0.01^{fg}
CW	94.05 ± 23.17^{e}	0.85 ± 0.09^{d}	0.35 ± 0.02^{e}	33.04 ± 7.89^{d}	27.58 ± 5.04^{c}	−9.48 ± 2.57^{b}	0.09 ± 0.01^{ef}
C2	119.64 ± 45.79^{e}	0.29 ± 0.04^{d}	0.17 ± 0.01^{f}	19.78 ± 6.52^{d}	5.69 ± 1.43^{c}	−18.02 ± 6.66^{c}	0.04 ± 0.01^{g}
C11	215.40 ± 47.30^{e}	0.52 ± 0.06^{d}	0.22 ± 0.05^{f}	48.56 ± 18.40^{d}	25.53 ± 11.14^{c}	−13.62 ± 4.61^{bc}	0.08 ± 0.02^{ef}
TW	484.01 ± 62.12^{e}	0.94 ± 0.01^{d}	0.43 ± 0.03^{de}	211.21 ± 38.67^{d}	198.70 ± 37.00^{c}	−14.13 ± 1.38^{bc}	0.16 ± 0.02^{d}
T2	7444.48 ± 793.16^{a}	2.80 ± 0.83^{b}	1.26 ± 0.04^{b}	5633.38 ± 765.45^{a}	9240.96 ± 2273.09^{b}	−2.78 ± 4.27^{a}	0.63 ± 0.04^{b}
T11	6803.18 ± 933.29^{ab}	3.84 ± 1.03^{a}	1.27 ± 0.02^{b}	5191.96 ± 720.64^{ab}	11,725.29 ± 2734.81^{a}	−2.75 ± 4.44^{a}	0.66 ± 0.04^{b}
BW	2624.49 ± 434.11^{d}	1.60 ± 0.05^{c}	0.67 ± 0.06^{c}	1058.83 ± 255.60^{c}	1018.27 ± 245.68^{c}	−1.41 ± 0.79^{a}	0.26 ± 0.02^{c}
B2	6490.54 ± 841.37^{bc}	2.74 ± 0.86^{b}	1.41 ± 0.06^{a}	5486.34 ± 597.64^{a}	9061.38 ± 3066.45^{b}	−0.83 ± 1.12^{a}	0.75 ± 0.04^{a}
B11	5863.48 ± 1707.09^{c}	2.99 ± 0.92^{b}	1.37 ± 0.05^{a}	4809.19 ± 1372.42^{b}	8122.00 ± 1520.44^{b}	−0.19 ± 0.12^{a}	0.76 ± 0.05^{a}

[a] M: mealworm; C: cricket; T: tilapia; B: chicken breast; W: washing method; 2: acid-pH shift method (pH 2); 11: alkaline-pH shift method (pH 11).
[b] Data are presented as mean ± standard deviation. Different superscripts within columns represent significant differences at $p < 0.05$.
[c] We could not analyze M2 because it did not form a gel structure.

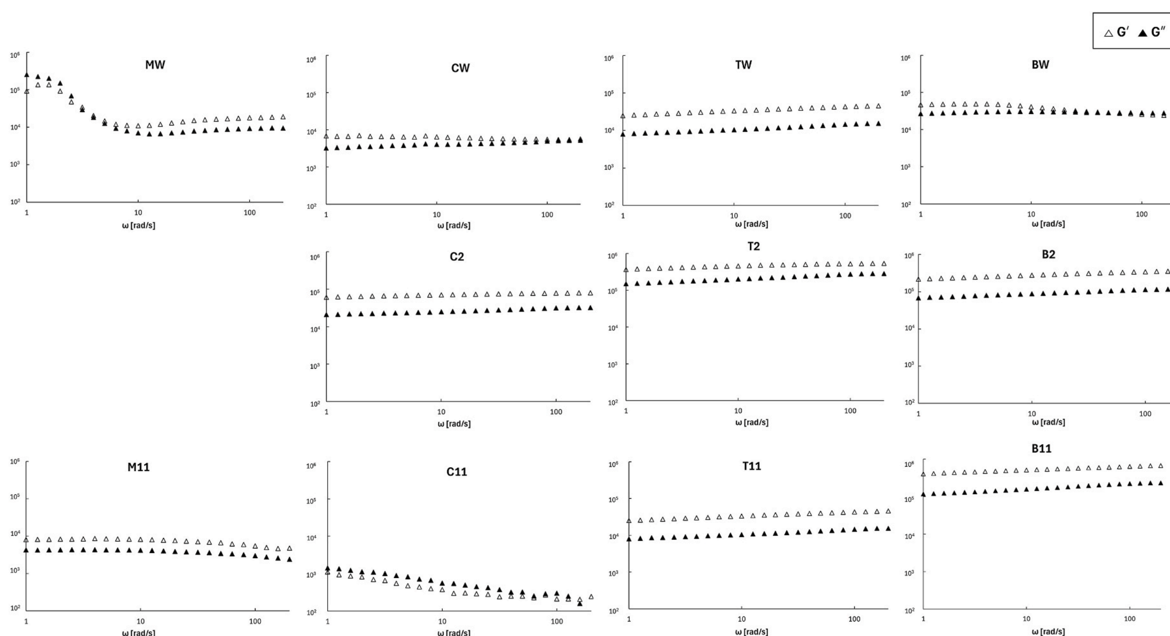

Fig. 5. Storage (G′) and loss (G″) moduli for surimi gels. M: Mealworm; C: cricket; T: tilapia; B: chicken breast; W: washing method; 2: acid-pH shift method (pH 2); 11: alkaline-pH shift method (pH 11). We could not analyze M2 because it did not form a gel structure.

other surimi samples exhibited frequency-dependent behavior, indicating an increase in elasticity with increased frequency. In contrast, CW and BW exhibited an increase in G″ values with increased frequency, indicating an increase in viscosity. T2 and B11 exhibited the highest G′ and G″ magnitudes.

4. Conclusion

We aimed to demonstrate the potential of edible insects as source ingredients for surimi by evaluating their physicochemical characteristics. Myofibrillar protein-rich fractions of the two insects were prepared using both the washing and pH shift methods. The pH shift method resulted in higher edible insect surimi yields. In the DSC analysis, acid-treated surimi sols exhibited simpler graph structures and fewer peak temperatures, indicating lower protein thermal stability; moreover, acid treatment markedly increased TBARS values in all samples. Furthermore, the acid treatment method could not produce an appropriate surimi gel structure for mealworm upon heating, emphasizing that alkaline treatment is the most appropriate method for insect surimi production. Hardness values for edible insect surimi gels were lower than those for tilapia and chicken breast surimi gels. Although tilapia and chicken breast exhibited superior gel formation abilities than edible insects, the adoption of the pH shift method resulted in the formation of overly-firm tilapia and chicken breast surimi gels. In contrast, edible insects formed softer surimi gels than tilapia and chicken breast. Consequently, our findings demonstrate that edible insects are novel source ingredients for surimi production, particularly for soft-gel food production. However, additional studies are required to develop methods of inhibiting browning reactions during the edible insect protein extraction process and to evaluate the effects of additives on surimi production.

CRediT authorship contribution statement

Chae-Ryun Moon: Investigation, Validation, Formal analysis, Methodology, Data curation, Writing – original draft, Visualization. **Young-Woong Ju:** Investigation, Formal analysis, Methodology. **Su-Hyeon Pyo:** Investigation, Formal analysis, Methodology. **So-Won Park:** Investigation, Formal analysis, Methodology. **Seul Lee:** Investigation, Formal analysis, Methodology. **Mzia Benashvili:** Investigation, Formal analysis, Methodology. **Yang-Ju Son:** Project administration, Investigation, Conceptualization, Methodology, Data curation, Writing –

original draft, Funding acquisition, Supervision.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.crfs.2024.100952.

Data availability

Data will be made available on request.

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