

Journal of Meat Science Year 2025 (June), Volume-20, Issue-1



Effect of Different Ingredients Loading Sequences During Mixing on the Physicochemical and Sensorial Properties of Buffalo Patties

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ARTICLE INFO

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doi10.48165/jms.2025.20.01.10

ABSTRACT

Some burgers are just better; it is all in the techniques – the right ingredients loading sequence makes a better-emulsified batter. This present work aims to evaluate the influence of the sequences of ingredients loading during mixing on the physicochemical (shrinkage percentage, cooking yield, water holding capacity (WHC), moisture content, pH, colour, rheological properties, texture) and sensory properties of buffalo patties. Additionally, this work aims to determine the best loading sequence among six tested variations using similar ingredients: T1 (meat + salt \rightarrow fat \rightarrow other non-meat ingredients), T2 (meat + sodium tripolyphosphates \rightarrow fat \rightarrow other non-meat ingredients), T3 (meat + hydrated corn starch \rightarrow fat \rightarrow other non-meat ingredients), T4 (meat + salt \rightarrow other non-meat ingredients \rightarrow pre-emulsified fat), T5 (meat + fat \rightarrow other non-meat ingredients) and T6 (all ingredients). Samples that follow the T4 sequence exhibit the highest cooking yield (75.42%), greatest WHC (53.74%), and minimal shrinkage percentage (7.51%). Interestingly, there was no significant difference in meat texture profile regardless of the sequence used. Although T4 did not receive the highest hedonic score for overall acceptability in sensory evaluations, the score obtained was statistically comparable to the highest-rated sample (T5). It is worth noting that the T4 sequence resulted in buffalo patties with reduced product loss and superior quality, making it a preferable choice for food manufacturers. As a result, it is recommended that food manufacturers employ the T4 loading sequence of ingredients when preparing buffalo patties.

Keywords: Buffalo patties, loading sequences, physicochemical properties, sensory evaluation, gel strength.

INTRODUCTION

Buffalo meat serves as an excellent substitute for beef, as it exhibits a near-identical composition, quality, and organoleptic profile (*Naveena and Kiran*, 2014). In response to its cost-effectiveness and enhanced nutritional value, fast

food manufacturers have transitioned from beef to buffalo meat as a primary raw material choice. In comparison to beef, buffalo meat boasts lower levels of fat, cholesterol, and calories (Anjaneyulu et al. 2007, Maheswarappa et al. 2022). Among fast foods, the burger reigns as one of the most beloved choices, cherished for its distinctive flavour and

texture. The quality of the burger is significantly influenced by the meticulous meat emulsion process during ingredient mixing. Meat emulsion, in essence, involves the creation of an oil-in-water emulsion, encompassing water, protein, and fat, wherein fat functions as the dispersed phase while the aqueous medium, laden with salt and protein, serves as the continuous phase (Rezaee and Aider, 2023).

Meat patty formulations typically encompass a combination of meat components (such as meat and fat) and non-meat ingredients (including starch, emulsifiers like lecithin and Tween 80, salt, sugar, garlic, black pepper, sodium tripolyphosphates (STPP), and water) (Mariana et al. 2019, Ismail et al. 2021, Ran et al. 2020). Each of these ingredients plays a pivotal role in upholding the stability of the meat emulsion while contributing to the formation of the desired texture and flavour of the patties (Meng et al. 2022). Notably, sodium chloride (NaCl), commonly known as salt, not only enhances flavour but also functions as a preservative in meat products (Kim et al. 2021). It additionally acts as a binding agent by extracting proteins, facilitating the adhesion of meat particles, thus enhancing the overall texture (Zhu et al. 2018). NaCl's presence elevates the viscosity of the meat mixture, aiding in the creation of a robust meat batter. Furthermore, it activates proteins, thereby improving hydration and the meat's water-binding capacity. Phosphates are another crucial ingredient, serving as quality enhancers that interact with NaCl to amplify the solubility of myofibrillar proteins, subsequently enhancing both water and oil retention within the minced meat system (Meng et al. 2022). Various flours, such as corn flour and tapioca flour, are employed as fillers and binding agents, imparting stability to the emulsion, augmenting water absorption capabilities, increasing product weight, and reducing production costs (Joly and Anderstein, 2009; Mariana et al. 2019).

Food emulsifiers like sodium caseinate and soy protein isolate play a role in stabilizing fat within the meat emulsion (Joly and Anderstein, 2009). Pre-emulsion represents an innovative component, employed for pre-emulsifying fat and enabling the utilization of lower-quality fats (up to 20%). This pre-emulsion typically comprises one part emulsifier, coupled with five to six parts fat and five to six parts water (Joly and Anderstein, 2009). Notably, pâté, a particular type of meat product created using pre-emulsified pork fat, exhibited reduced fat content and elevated moisture and protein levels (Domínguez et al. 2016). Consequently, within the scope of this study, a pre-emulsion was incorporated as one of the ingredients within the tested loading sequences. The appropriate sequence of ingredient loading during the mixing process plays a pivotal role in achieving a highquality meat emulsion, while concurrently offering the potential to reduce the overall manufacturing time for meat products (Meng et al. 2022). An effective emulsion process facilitates the extraction of myofibrillar proteins, which are instrumental in shaping the texture and enhancing the waterholding capacity (WHC) of meat. Shortening the processing time is a sought-after objective for food manufacturers, as it can lead to cost savings. In the context of this study, the sequences were devised based on insights gleaned from previous scientific investigations into the formulation of diverse meat products, including beef patties (Skog et al. 1992; Berry, 1998; Puolanne et al. 2001; Tobin et al. 2012), buffalo patties (Suman and Sharma, 2003), pork patties (Rodríguez-Carpena et al. 2012), chicken patties (Wan Rosli et al. 2011), pork sausages (Puolanne et al. 2001), beef sausages (Puolanne et al., 2001), beef bologna (Bishop et al., 1993) pork nuggets (Berry, 1994), and meatballs (Hsu and Chung, 2001). While these literature sources did not specifically explore the impact of ingredient loading sequences on product quality, they provided valuable guidelines for designing the experimental framework of the present study. To the best of the author's knowledge, the investigation into the influence of ingredient loading sequences on the quality and sensory attributes of buffalo meat patties has not been previously undertaken by other researchers. This study thus introduces a novel dimension by examining six distinct loading sequences.

MATERIALS AND METHODS

Buffalo patties preparation

Frozen buffalo topside meat was procured from a local supermarket (Shahrul Fresh and Frozen, Cheras, Selangor) to serve as the primary raw material. The preparation of buffalo patties was executed following the method outlined by Ibrahim et al. (2018) with slight modifications. The buffalo patties were formulated with 70% buffalo meat, 10% fat, 3.5% corn starch, 3% lecithin, 1.2% salt, 1% sugar, 0.5% garlic, 0.5% black peppers, 0.3% sodium tripolyphosphate (STPP), and 10% ice water. Initially, the visible fat portions were meticulously trimmed from the meat. Subsequently, the meat and fat were independently ground using a mincing machine (Mincer Hobart 4822, USA). The resulting ground meat and fat were stored at -18°C until further processing. To create the meat batters, a bowl cutter (K3 Model-BenchType, Taiwan) was employed, with the ingredients introduced in varying loading sequences. Table 1 delineates the six distinct treatments, with each treatment representing a unique loading sequence, involving a 3-step mixing process. The selection of these treatments was guided by previous literature that employed meat products as a reference. The loading sequence adopted for meat product preparation in this study was adapted from these sources. Maintaining consistency, the mixing speed and duration remained constant, with each mixing step taking 5 min and accumulating to a total of 15 min for each treatment. Each patty, weighing 80 grams, was meticulously shaped into a round form using a burger mould (Manual Burger Mould, BT10) featuring a 100 mm diameter.

To prevent adhesion between patties, a layer of plastic was interposed. Subsequently, the raw patties were frozen and stored in a freezer at -18°C until the subsequent analytical procedures were performed. A visual representation of the buffalo patty preparation process is depicted in Figure 1.

Cooking procedure

The buffalo patties underwent a cooking process employing

a griddling method, a technique adapted from Dreeling et al. (2002). Before cooking, the raw buffalo patties were thawed, and the cooking procedure was executed using an MSM Countertop Electric Griddle (Model HP-6000, Malaysia). In this method, the griddle's surface was preheated and lightly coated with a thin layer of palm oil. The griddling duration was precisely set at approximately 10 min, with the patties being flipped every 2 min during the initial 8 min, and subsequently, every 1 minute for the remaining cooking time.

Table 1. Treatments of buffalo patties

Treatment	First step	Second step	Third step				
	(First batch)	(Second batch)	(Third batch)				
T1	Meat + salt	Fat	Other ingredients (corn starch, lecithin, sugar, garlic, black peppers, STPP, water)				
T2	Meat + STPP	Fat	Other ingredients (corn starch, lecithin, salt, sugar, garlic, black peppers, water)				
Т3	Meat + hydrated corn- starch (corn starch hydrated with 100 g of water)	Fat	Other ingredients (lecithin, salt, sugar, garlic, black peppers, STPP)				
T4	Meat + salt	Other ingredients (cornstarch, sugar, garlic, black peppers, STPP)	Pre-emulsion (fat was emulsified with lecithin and 100 g of hot water in a blender (Panasonic Blender MX-GM1011H, Malaysia))				
T5	Meat + fat		Other ingredients (cornstarch, lecithin, salt, sugar, garlic, black peppers, STPP, water				
Т6	All ingredients						
	(meat, fat, corn starch, lecithin, salt, sugar, garlic, black peppers, STPP, water						

Shrinkage percentage

The assessment of shrinkage percentage represents a crucial aspect in evaluating the physical changes that occur during the cooking process of buffalo patties (Ismail et al. 2021). This method entails measuring the diameter (in millimetres, mm) and thickness (in millimetres, mm) of both the raw and cooked buffalo patties using a Vernier calliper (Mitutoyo, Japan). The shrinkage percentage of the buffalo patties was quantified utilizing the following formula (Equation 1):

 $\frac{\text{Shrinkage (\%)} = \text{Equation (1)}}{\text{diameter of raw sample}} + \frac{\text{diameter of cooked sample}}{\text{diameter of raw sample}} \times 100\%$

Cooking Yield

Cooking yield is a fundamental parameter in the assessment of meat products, serving as an indicator of the product's weight retention during the cooking process (Lim et al. 2023). This measurement plays a crucial role in evaluating the efficiency of meat preparation methods and understanding the physical changes that occur in buffalo patties as they transition from their raw to cooked states. The cooking yield of buffalo patties was determined by comparing the weights of the raw and cooked patties. This evaluation utilized a formula (Equation 2) adopted from Wan Rosli et al. (2011) to quantify the cooking yield.

Cooking yield (%) = Weight of cooked buffalo meatballs $\times 100\%$ Weight of uncooked buffalo meatballs

pH analysis

To initiate the analysis, 5 grams of both raw and cooked buffalo patties underwent manual homogenization in 40 millilitres of distilled water, utilizing a mortar and pestle. Subsequently, the pH values of these raw and cooked patties were meticulously measured employing a pH meter (Jenway Model 3505 pH meter, UK). Each sample was subjected to triplicate measurements to ensure accuracy and consistency

in the obtained data.

Water holding capacity

The assessment of water holding capacity (WHC) in buffalo patties followed the methodology outlined by Lim et al. (2023), with modification. This analysis involved homogenizing 5 grams of pre-weighed buffalo patties in 32 millilitres of distilled water for one minute, utilizing a homogenizer (Heidolph Homogenizer DIAX 900). Following homogenization, the mixture was allowed to stand undisturbed for 10 min before undergoing centrifugation at 2900 g for 25 min, utilizing a centrifuge (Micro Refrigerated Centrifuge Model 3740, Japan). Subsequently, the supernatant was carefully discarded, and the residual sample was subjected to drying at a 10 to 20° inclination downward for 20 min at 50°C. The dried samples were then meticulously weighed and the weights were recorded for further analysis. The WHC was computed utilizing the following formula (Equation 3): Equation (3)

WHC (%) =
$$\frac{(y-x) - (z-x)}{(z-x)} \times 100$$

Where,

x = weight of empty centrifuge (g)

y = weight of centrifuge with supernatant (g)

z =weight of dried centrifuge (g)

Moisture content

The moisture content of both raw and cooked buffalo patties was assessed following the standardized procedures established by the Association of Official Analytical Chemists (AOAC). This evaluation involved the utilization of a hot air oven (BINDER, United States). The procedure commenced with the drying of empty crucibles within the oven at a temperature of 105°C for three hours. Subsequently, these crucibles were transferred to a desiccator to facilitate cooling, following which their weights were accurately recorded. For the analysis, 3 grams of both raw and cooked patties were meticulously weighed and then transferred into the prepared crucibles. These crucibles, containing the respective samples, were once again subjected to drying in the oven, maintaining the temperature at 105°C for three hours. Post-drying, the crucibles were relocated to the desiccator to undergo cooling, after which their weights were meticulously recorded. The moisture content percentage of both raw and cooked buffalo patties was calculated using Equation 4: Equation (4)

Moisture content (%) =
$$\frac{(W1 - W2)}{W1}$$
 x 100

Where.

W1 = weight of the sample before drying (g)

W2 = weight of the sample after drying (g)

Colour measurements

Colour measurements for both the raw and cooked samples were conducted using a colorimeter (CR-40 Konica Minolta Camera Co., Japan), following the method delineated by Ismail et al., (2021) with certain modifications. Before analysis, the raw samples were allowed to thaw at room temperature for a duration ranging from 30 min to 1 hour. Similarly, the cooked patties were left at room temperature for a period ranging from 30 min to 1 hour prior to the commencement of the colour analysis. The evaluation encompassed the determination of L* (lightness), a* (redness), and b* (yellowness) values for each sample. To ensure robustness and reliability, all measurements were performed in triplicate for each sample.

Texture Profile Analysis

Texture profile analysis (TPA) was conducted on cooked buffalo patties representing six distinct treatments, following the protocol outlined by Faridah et al. (2023). The evaluation was performed using a Texture Analyser TA-XT2 (Stable Micro Systems, Surrey, UK). To facilitate the analysis, the cooked buffalo patties were uniformly cut into dimensions measuring 3 x 3 x 1 cm and subsequently subjected to the texture profile analyser. During testing, the samples were compressed to 75% of their original height utilizing a Warner Bratzler probe set at p/75. The testing protocol incorporated a pre-test speed of 1.00 mm/s, a test speed of 3.00 mm/s, and a post-test speed of 5.00 mm/s to ensure standardized conditions. Various texture parameters were assessed, including hardness, adhesiveness, springiness, gumminess, and chewiness, collectively offering a comprehensive understanding of the textural attributes of the cooked buffalo patties across the different treatments.

Gel strength analysis

The procedure for evaluating gel strength closely adhered to the method outlined by Asyrul-Izhar et al. (2021) with certain adaptations to suit the experimental conditions. Gel strength assessment was conducted using a Heavy Duty Texture Analyser, specifically the TA-XT2 model from Stable Micro Systems, Surrey, UK. This device was equipped with a 10-mm spherical plunger, and the crosshead speed was meticulously set at 0.83 mm/s to ensure consistency in the testing procedure. To prepare the samples for testing, the cooked patties were allowed to equilibrate at room temperature for 30 min before being uniformly cut into dimensions measuring 3 cm x 3 cm x 1 cm. Subsequently, the cut patties were subjected to axial compression until rupture occurred. Gel strength was quantified by multiplying the breaking strength (in kilograms) by the deformation (in centimetres). It is noteworthy that all measurements for each sample were conducted in triplicate to guarantee the

precision and reproducibility of the results.

Sensory evaluation

To assess the sensory properties of the cooked buffalo patties, a preference test was conducted, guided by the methodology presented by Lim et al. (2023). This evaluation employed a 9-point hedonic scale to gauge various attributes, including appearance, taste, juiciness, texture, and overall acceptability. On the hedonic scale, each sensory attribute was rated as follows: 1 = "dislike extremely", 2 = "dislike very much", 3 = "dislike moderately", 4 = "dislike slightly", 5 = "neither like nor dislike", 6 = "like slightly", 7 = "like moderately", 8 = "like very much", and 9 = "like extremely". A panel of 30 untrained individuals, whose ages ranged from 20 to 30 years and included students and staff from the Faculty of Food Science and Technology at Universiti Putra Malaysia, participated in the sensory evaluation. This assessment was conducted in a single session within a standard sensory laboratory situated in the Department of Food Technology, Faculty of Food Science and Technology, Universiti Putra Malaysia. During the evaluation, each sample comprised cooked buffalo patties, discreetly labelled with a unique 3-digit random code. To prevent flavour carryover between samples, plain water was provided to the panellists to rinse their mouths after tasting each sample. This meticulous setup ensured the reliability and accuracy of the sensory assessment.

Statistical analysis

All analyses were conducted in triplicate for each sample, encompassing the six distinct treatments. The amassed data were subjected to comprehensive statistical analysis, employing MiniTab Statistical Software Version 19 (MiniTab Inc., USA). To discern significant differences among the six samples, a one-way analysis of variance (ANOVA) was executed for all the analyses, which included assessments of shrinkage percentage, cooking yield, water holding capacity, moisture content, pH, colour, texture profiles, gel strength, and sensory evaluation. Subsequently, Tukey's test was applied for pairwise comparisons, with a significance level set at 95% (p=0.05). This stringent statistical approach allowed for robust and precise comparisons between the different treatments.

RESULTS AND DISCUSSION

Cooking yield, shrinkage percentage and pH value

Table 2 presents the data on the cooking yield and shrinkage percentage of the buffalo patties. It is noteworthy that while there were no significant differences (p>0.05) observed

in cooking yield among the various treatments, notable disparities were evident in the shrinkage percentage (p<0.05). The cooking yield of the buffalo patties exhibited a range from 68.96% to 75.42%. Specifically, samples from treatment T5 displayed the lowest cooking yield at 67.59%, while those from treatment T4 showcased the highest cooking yield, reaching 75.42%. In contrast, the shrinkage percentage exhibited diverse values, with samples from treatment T5 registering the highest value at 15.45%, while treatment T4 exhibited the lowest shrinkage percentage of 7.51%. A prior study conducted by Ramadhan et al. (2012) reported the shrinkage of Malaysian commercial burger patties to range from approximately 2% to 10%. As discerned from Table 2, the shrinkage percentages of samples T1, T3, T5, and T6 were notably less favourable. Generally, within the realm of meat-processed products, a high cooking yield coupled with a low shrinkage percentage is the desired outcome. The phenomenon of shrinkage in meat products during the cooking process can be attributed to the denaturation of proteins and the consequent loss of fat and water (Ramadhan et al., 2012). It is plausible that the higher shrinkage percentages observed in samples T3 and T5, as compared to other treatments, may be attributed to the mixing of meat and fat/corn starch before the addition of salt and phosphates. This mixing sequence could potentially disrupt the ability of fat globules and starch molecules to effectively retain water, resulting in a heightened loss of water during the cooking process (Aktaş and Gençcelep, 2006; García-García and Totosaus, 2008).

The pH, recognized as a fundamental and key parameter in determining meat quality, holds a central position due to its intricate interplay with various meat processing factors, including emulsifying capacity and water-holding capacity (Kandeepan et al., 2013). The pH measurements for buffalo patties formulated using distinct loading sequences are detailed in Table 2. Notably, no significant difference (p<0.05) was discerned in the pH values of both raw and cooked patties. Nevertheless, it is pertinent to observe that the pH values of the cooked patties exhibited a slightly elevated profile compared to their raw counterparts. This phenomenon aligns with findings by López-Botea and Calvo (2023), who expounded that pH values in cooked meat products tend to rise due to the reduction of free acidic groups as meat temperature escalates during the cooking process. Specifically, the pH values for raw samples were recorded within the range of 5.61 to 5.78, while the pH values for cooked samples ranged from 5.72 to 5.89. Though no statistically significant differences were evident, it is noteworthy that samples T1, T2, and T4 exhibited higher pH values compared to the remaining samples. A plausible explanation for this observation could be attributed to the initial mixing of meat with salt or phosphates during the first batch, resulting in an augmentation of negative charges on the proteins responsible for the subsequent increase in pH value (Zhu et al., 2018).

Table 2. Cooking yield, shrinkage percentage, and pH of buffalo patties formulated with different treatments.

Treat- ments	Variables							
	Cooking yield (%)	Shrinkage (%)	pH raw	pH cooked				
T1	69.92 ± 3.21^{a}	10.94 ± 2.58^{ab}	$5.78\pm0.02^{\mathrm{a}}$	$5.89 \pm 0.03^{\mathrm{a}}$				
T2	$69.25\pm5.63^{\mathrm{a}}$	9.69 ± 3.30^{ab}	$5.73\pm0.02^{\rm a}$	$5.86\pm0.03^{\rm a}$				
T3	$68.96\pm3.63^{\mathrm{a}}$	12.99 ± 2.19^{ab}	5.61 ± 0.18^{a}	$5.75\pm0.13^{\mathrm{a}}$				
T4	$75.42 \pm 0.4^{\circ}a$	$7.51 \pm 1.9^{8}b$	$5.71\pm0.02^{\rm a}$	$5.82\pm0.02^{\rm a}$				
T5	$67.59\pm4.02^{\mathrm{a}}$	$15.45\pm1.99^{\mathrm{a}}$	5.61 ± 0.20^{a}	$5.72\pm0.12^{\mathrm{a}}$				
T6	$71.54\pm2.44^{\mathrm{a}}$	10.18 ± 1.26^{ab}	5.73 ± 0.01^{a}	$5.81\pm0.02^{\rm a}$				

Means SD with different superscripts in a column within the same analysis is significantly different (p<0.05)

Water holding capacity (WHC) and moisture content

Water holding capacity (WHC) for meat patties refers to the ability of the meat to retain water or moisture during processing, cooking, and storage (Rezaee and Aider, 2023). It is an essential quality parameter for meat products, as it influences various attributes such as juiciness, texture, and overall eating experience (Lim et al. 2023). A higher WHC indicates that the meat patty can retain more moisture, resulting in juicier and more succulent patties. On the other hand, lower WHC can lead to dry and less appealing meat products. As indicated in Table 3, there are significant differences (p<0.05) discernible in the water holding capacity (WHC) of buffalo patties formulated using different treatments (loading sequences). The WHC values exhibited a range spanning from 46.61% to 53.74%. Notably, sample T4 achieved the highest WHC, while sample T1 recorded the lowest WHC among the tested loading sequences. In Treatment T1, the sequencing involved the introduction of fat in the second step, followed by the addition of other ingredients in the third step. Conversely, Treatment T4 adopted a different approach, utilizing pre-emulsion fat in the third step and introducing other ingredients in the second step. It is worth noting that both of these treatments commenced with the inclusion of meat and salt in the first step of the formulation process. The utilization of preemulsified fat has enhanced water-holding capacity (WHC), which is corroborated by previous findings (Domínguez et al. 2016). Specifically, research has indicated that meat products crafted with pre-emulsified fat exhibit a reduction in fat content while experiencing increased moisture and protein levels (Domínguez et al., 2016). This underscores the role of pre-emulsified fat in improving WHC.

Moisture content, defined as the ratio of water mass to sample mass (*Li et al.*, 2018), plays a significant role in shaping the textural attributes of meat products, alongside fat content (Chen and Rosenthal, 2015). Table 3 presents the results of moisture content in both raw and cooked buffalo

patties formulated using various treatments. Interestingly, no statistically significant differences (p<0.05) were observed in either the raw or cooked samples. For raw patties, moisture content ranged from 61.63% to 63.14%, with sample T4 recording the highest moisture content, while sample T3 displayed the lowest. In contrast, for the cooked patties, moisture content ranged from 50.12% to 53.07%, with sample T6 exhibiting the highest moisture content and sample T3 the lowest. It is worth noting that, consistently across all samples, raw patties showed higher moisture content compared to their cooked counterparts. This decrease in moisture content during cooking aligns with findings by Shen et al. (2010), who attributed it to the loss of water during the heating and cooking processes. It is established that moisture content exhibits an inverse relationship with product hardness (Chin et al. 2004). This assertion finds support in the hardness values presented in Table 5, where it is evident that sample T3 recorded the highest hardness value, while sample T6 displayed one of the lowest hardness values.

Table 3. Water holding capacity and moisture content of raw and cooked buffalo patties with different treatments.

Treat- ments	Variables							
	WHC (%)	Moisture con-	Moisture content					
Inches		tent raw (%)	cooked (%)					
T1	46.61 ± 4.04^{b}	63.06 ± 1.80^{a}	52.13 ± 1.36^{a}					
T2	51.63 ± 1.63^{ab}	62.12 ± 0.50^{a}	51.65 ± 1.63^{a}					
T3	50.12 ± 0.63^{ab}	61.63 ± 0.57^{a}	50.12 ± 0.63^{a}					
T4	$53.74 \pm 3.5^{9}a$	63.14 ± 1.17^{a}	50.41 ± 2.58^{a}					
T5	50.54 ± 0.09^{ab}	61.69 ± 2.85^{a}	50.54 ± 0.09^{a}					
T6	53.07 ± 0.90^{a}	62.76 ± 0.81^{a}	53.07 ± 0.90^{a}					

Means SD with different superscripts in a column within the same analysis is significantly different (p<0.05)

Colour measurements

Colour plays a significant role in influencing consumer perception and acceptability of products (Suman et al. 2023). In this regard, the results of colour measurements for buffalo patties formulated using various treatments are presented in Table 4. Table 4 shows significant differences (p<0.05) in the optical intensity properties—lightness (L*), redness (a*), and yellowness (b*)—of both raw and cooked buffalo patties, except for the a* values of cooked patties, where no significant differences were observed (p>0.05).

For raw patties, the L* values ranged from 52.29 to 56.60, while the b* values varied between 13.13 and 15.01. Notably, sample T1 demonstrated the highest L* and b* values, whereas samples T6 and T5 exhibited the lowest L* and b* values, respectively. The L* (lightness) and b* (yellowness) values of the products have a known association with fat content (Ismail et al. 2009). An increase in fat content typically leads to an increase in product lightness, attributable to the whitish

appearance of fat, which elevates the L value. Nevertheless, it is essential to emphasize that the fat content in this study was maintained at a constant level, accounting for 10% of the total buffalo patty formulation. This constancy in fat content implies that it might not have played a role in generating variations in lightness and yellowness among the samples. A plausible explanation for the observed statistical differences in L* and b* values among the samples could be related to the even distribution of fat within the meat batter. Samples with higher lightness values may have achieved a more uniform distribution of fat, contributing to their enhanced lightness compared to samples with lower lightness values.

In terms of a* values (representing redness), samples T3 showed the highest values in both their raw and cooked forms, recording 9.27 and 7.46, respectively. This observation aligns with findings by Ibrahim et al., (2018), who reported

that consumers tend to associate the redness of raw meat products with freshness. Conversely, when it comes to cooked products, the perception is often linked to the meat's darker (brown) hue, which is typically regarded as more palatable. The significance of redness in both raw and cooked meat products cannot be overstated. In the case of raw meat, the presence of redness is frequently linked to freshness, thereby influencing consumer preferences. On the other hand, in the context of cooked meat, the level of redness or its transformation into a desirable brown colour plays a crucial role in determining the product's overall palatability and acceptance, as evidenced by the sensory evaluation results (Table 6), where sample T3, characterized by its redness, garnered the highest hedonic score, signifying its preference among panellists.

Table 4. Colour analysis of raw and cooked buffalo patties with different treatments.

	Variables					
Treatments	Raw			Cooked		
	L*	a*	b*	L*	a*	b*
T1	56.60 ± 0.83^{a}	$8.27\pm0.30^{\mathrm{b}}$	15.01 ± 0.49^{a}	34.77 ± 1.64^{ab}	7.13 ± 1.03^{a}	8.34 ± 1.57^{ab}
T2	$54.83\pm0.77^{\mathrm{ab}}$	9.03 ± 0.18^{ab}	14.36 ± 0.69^{ab}	36.81 ± 2.12^{ab}	6.75 ± 0.44^{a}	8.77 ± 0.67^{ab}
Т3	55.70 ± 2.39^{ab}	9.27 ± 0.24^{a}	14.35 ± 0.72^{ab}	36.08 ± 0.38^{ab}	7.46 ± 1.40^{a}	8.20 ± 2.97^{ab}
T4	53.39 ± 1.26^{ab}	9.06 ± 0.28^{ab}	13.17 ± 0.73^{b}	$37.51\pm0.65^{\mathrm{a}}$	7.29 ± 0.24^{a}	11.19 ± 1.50^{a}
T5	52.63 ± 0.25^{b}	8.77 ± 0.53^{ab}	13.13 ± 0.15^{b}	33.06 ± 1.05^{b}	6.74 ± 0.15^{a}	$6.84\pm0.33^{\text{b}}$
T6	52.29 ± 1.13^{b}	9.03 ± 0.18^{ab}	13.18 ± 0.41^{b}	34.41 ± 2.01^{ab}	6.30 ± 0.40^{a}	6.07 ± 0.49^{b}

Means \pm SD with different superscripts in a column within the same analysis is significantly different (p<0.05).

Texture profile analysis (TPA) and gel strength

The results of the texture profile analysis (TPA) conducted on buffalo patties formulated using different treatments are summarized in Table 5. Remarkably, the treatments applied in the production of all six samples under study did not exert a significant difference (p>0.05) on any of the TPA parameters examined. These parameters encompass hardness, adhesiveness, springiness, cohesiveness, and chewiness. However, it's noteworthy that T6 samples exhibited a marginally lower score in terms of springiness and chewiness compared to the other samples, despite the absence of significant mean differences (p>0.05).

A positive correlation between hardness and overall acceptance, as demonstrated by Ahmad et al. (2022) in their restructured buffalo meat fillets study, suggests that consumers generally favour meat products with a firmer texture. In the case of hardness, the values ranged from 22,041.26 to 26,948.07, with T1 samples displaying the lowest hardness score and T3 samples registering the highest hardness value. Although this disparity did not reach statistical significance (p>0.05), the formulation utilized for T3 samples yielded buffalo patties with the firmest texture, potentially attributed to the absorption of water by starch

granules before complete protein extraction.

Huda et al. (2009) have recommended that sensory attribute evaluation, particularly texture, should be conducted concurrently with textural analysis to identify the optimal range of attributes preferred by consumers. In this context, the sensory assessment of texture attributes was conducted, as detailed in Table 6. Notably, the hedonic scores assigned to the texture of the patties did not show any statistically significant differences (p>0.05) among the samples. Importantly, these findings are in alignment with the texture analysis results (Table 5), which consistently indicated an absence of significant variations in texture attributes (including hardness, adhesiveness, springiness, cohesiveness, and chewiness) across all the samples. Nevertheless, it is noteworthy that in the sensory evaluation (specifically assessing texture attributes as shown in Table 6), T4 samples received the highest scores. This preference aligns with the fact that T4 samples exhibited a hardness value of 24,747.53 g, making them the preferred choice for texture hardness among the panellists.

As suggested by Serdaroğlu et al. (2005), the textural properties of comminuted meat products are influenced by factors such as stromal protein content, the type and level of non-meat ingredients, the extent of comminution, and the degree of extracted myofibrillar proteins. However, the

findings from the Texture Profile Analysis (TPA) in this study demonstrate that the utilization of various loading sequences in the production of buffalo patties did not yield significant variations in the textural properties of the patties.

Gel strength analysis is a critical parameter that plays a crucial role in ensuring the quality, consistency, and consumer appeal of buffalo patties. It is an important aspect of product development and quality control in the food industry (Gao et al., 2016). The results of gel strength analysis for buffalo patties formulated with different treatments are presented in Table 5. Furthermore, gel strength is essential for ensuring the safe storage and transportation of fish-meat gels (Gao et al. 2016). This aspect of gel strength is equally applicable to meat batter.

Table 5 reveals that no significant difference (p>0.05) was observed in the gel strength of buffalo patties among the various treatments. Despite the lack of significant differences (p>0.05), it's noteworthy that sample T3 shows the highest gel strength value, measuring 633.7 g, while sample T1 displayed the lowest gel strength at 401.1 g. In principle, higher gel strength values are indicative of firmer products (Ahhmed et al. 2007). This observation aligns with the hardness results obtained from the Texture Profile Analysis (TPA), as

presented in Table 6, where samples T3 demonstrated the highest hardness values, whereas T1 samples exhibited the lowest hardness.

The enhanced gel strength in sample T3 may be attributed to their initial mixing with hydrated corn starch. The starch granules likely absorbed the water associated with the meat, resulting in increased gel strength (García-García and Totosaus, 2008; Jamilah et al. 2009). Another plausible explanation is that when protein molecules became entrapped within the starch granules, the salt and phosphates couldn't effectively function to extract myofibrillar proteins (actin and myosin) from the meat. Multiple studies have demonstrated that myosin plays a key role in forming excellent gels (Fretheim et al.1986; Brewer et al. 2005; Sun and Holley, 2011).

According to Sun and Holley (2011), several factors can influence gelation properties, including the type of muscle used, meat species, myofibrillar protein fraction employed, and processing conditions (i.e., ionic strength, pH, heating rate, pressure, and temperature). In the present study, the different treatments, represented by distinct ingredient loading sequences used to formulate the buffalo patties, did not significantly affect the gel strength of the patties.

Table 5. Texture profile analysis (TPA) of buffalo patties formulated with different treatments.

Treat-	Texture Parameters							
ments	Hardness (g)	Adhesiveness	Springiness(mm)	Cohesiveness	Chewiness (g x mm)	Gel strength (g.cm)		
T1	22041.26 ± 5084.74 ^a	-2.07 ± 0.75^{a}	0.70 ± 0.08^{a}	0.41 ± 0.02^{a}	6287.73 ± 1928.73 ^a	401.1 ± 157.3 ^a		
T2	$26339.54 \pm 973.5^{9}a$	-2.20 ± 0.72^{a}	0.75 ± 0.06^{a}	0.41 ± 0.03^{a}	8242.83 ± 1576.80^{a}	509.0 ± 186^{a}		
T3	26948.07 ± 1572.97^{a}	-2.68 ± 0.12^{a}	0.65 ± 0.08^{a}	0.43 ± 0.02^{a}	7392.56 ± 1122.56^{a}	633.7 ± 81.3^{a}		
T4	24747.53 ± 3203.01^{a}	-1.49 ± 0.38^{a}	0.76 ± 0.06^{a}	$0.39\pm0.05^{\rm a}$	7310.40 ± 1784.67^{a}	619.6 ± 143.6^{a}		
T5	22431.91 ± 3353.24^a	-0.67 ± 0.54^{a}	0.68 ± 0.21^{a}	0.68 ± 0.21^{a}	6146.10 ± 2691.10^{a}	416.3 ± 172.9^{a}		
T6	22428.03 ± 3474.26^{a}	-1.51 ± 1.18^{a}	0.61 ± 0.05^{a}	0.61 ± 0.05^{a}	5234.91 ± 1777.38^{a}	417.9 ± 95.5 ^a		

Means \pm SD with different superscripts in a column within the same analysis is significantly different (p<0.05).

Sensory evaluation

The results of the sensory evaluation for buffalo patties prepared using various treatments are summarized in Table 6. The sensory attributes assessed included appearance, texture, juiciness, flavour, colour, aroma, and overall acceptability. Consumers' preferences for the palatability of food products are strongly influenced by these attributes (Troy and Kerry, 2010). In a study conducted by Resurreccion (2003), attributes such as juiciness, tenderness, flavour, and appearance were identified as key factors influencing consumer acceptability of meat and meat products.

Table 6 reveals that no statistically significant differences (p>0.05) were observed among the six samples in terms of all attributes except for flavour and overall acceptability. Consequently, flavour emerged as the critical attribute influencing consumer acceptance of buffalo patties. The hedonic overall acceptability scores further support this observation, with T5 samples receiving the highest score of

6.57 and T2 samples ranking second with a score of 6.50. Conversely, T1 and T6 samples were the least preferred, with hedonic scores of 5.37 and 5.33, respectively. While the flavour results did not exhibit an identical pattern, T2 and T5 samples also garnered the highest and second-highest scores, while T6 and T1 samples were the least favoured.

Juiciness represents another pivotal sensory attribute in the evaluation of meat products. In a study by Yi et al. (2012) on the physicochemical and organoleptic characteristics of seasoned beef patties enriched with glutinous rice flour, they found that samples rated highest for juiciness in sensory evaluations were the most preferred. This observation was attributed to the enhanced water retention capacity of the patties. In this study, samples from T4 exhibited the highest values for moisture content and water holding capacity (WHC) (Table 4). Therefore, it can be inferred that juiciness is contingent upon the moisture content and WHC of meat products.

Table 6. Sensory evaluation of buffalo patties formulated with different treatments.

Componer Attnibutos	Treatments						
Sensory Attributes	T1	T2	Т3	T4	T5	T6	
Appearance	6.07 ± 1.53^{a}	6.40 ± 1.22^{a}	6.47 ± 1.38^{a}	6.10 ± 1.73^{a}	6.43 ± 1.38^{a}	5.97 ± 1.69^{a}	
Texture	5.67 ± 1.75^{a}	6.07 ± 1.46^{a}	5.77 ± 1.43^{a}	6.40 ± 1.70^{a}	6.33 ± 1.58^{a}	5.60 ± 1.80^{a}	
Juiciness	5.53 ± 1.85^{a}	5.83 ± 1.60^{a}	5.37 ± 1.85^{a}	6.13 ± 1.48^{a}	6.00 ± 1.86^{a}	5.20 ± 1.86^{a}	
Flavour	4.83 ± 2.12^{b}	6.43 ± 1.61^{a}	5.93 ± 2.02^{ab}	5.83 ± 2.04^{ab}	6.17 ± 1.66^{ab}	5.50 ± 2.22^{ab}	
Colour	6.23 ± 1.43^{a}	6.27 ± 1.17^{a}	6.60 ± 1.19^{a}	6.03 ± 1.97^{a}	6.37 ± 1.45^{a}	5.97 ± 1.97^{a}	
Aroma	5.83 ± 1.66^{a}	6.30 ± 1.66^{a}	6.27 ± 1.48^{a}	6.00 ± 1.76^{a}	6.27 ± 1.70^{a}	5.47 ± 1.80^{a}	
Overall Acceptability	5.37 ± 1.71^{ab}	6.50 ± 1.36^{ab}	6.06 ± 1.52^{ab}	6.27 ± 1.57^{ab}	6.57 ± 1.61 ^a	5.33 ± 1.86^{b}	

Means \pm SD with different superscripts in a row within the same analysis is significantly different (p<0.05)

CONCLUSION

In summary, the influence of ingredient loading sequences on buffalo patty quality attributes was investigated. Cooking yield remained consistent, while shrinkage percentages varied among samples. Loading sequences significantly influenced water holding capacity, but moisture content showed no significant difference. While pH values remained stable during processing, they increased when the patties were cooked. Colour attributes showed differences and gel strength analysis showed no significant variations. However, sensory evaluation highlighted flavour as a critical factor for overall acceptability, with specific loading sequences yielding more favourable profiles. Juiciness was linked to water holding capacity. Notably, sample T4's loading sequence excelled in several aspects, making it the most effective for buffalo patties, and offering valuable insights for product development and quality control. Future research should explore modifications and various meat species to enhance emulsification and product quality.

ACKNOWLEDGMENT

The authors would like to acknowledge the financial support provided by Universiti Putra Malaysia (9774300) Putra IPS grant.

AUTHOR CONTRIBUTION

MRF: Conceptualization, Methodology, Data Curation, and Writing of the Original Draft. NIK: Data Curation and Writing of the Original Draft. MRI, PV, & AQS: Conceptualization, Supervision, Project Administration, and Reviewing the Final Manuscript.

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