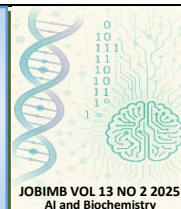


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Development of Plant-Based Ball from Shiitake Mushroom and Chickpea: Physicochemical Properties and Sensory of Selected Formulation

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Abstract

Shiitake mushrooms and chickpeas, valued for their nutritional benefits, functional properties, and environmental sustainability, are promising ingredients for alternative food development, yet limited research has explored their combined use in plant-based products. This study aims to transform shiitake mushrooms into valuable food products —specifically plant-based balls —by combining them with chickpea flour, thereby enhancing their value and promoting sustainability. Shiitake mushrooms and chickpea flour were combined in three ratios (15:35, 25:25, and 35:15) to formulate the mushroom balls. The research assessed the sensory characteristics of plant-based balls using a 9-point hedonic scale. The evaluation focused on chewiness, flavour intensity, umami, colour, saltiness, aftertaste, and overall acceptability. The research also includes physicochemical properties and calorie content of plant-based balls for nutritional labelling. Additionally, it examines the effect of cooking time (7, 10, and 13 min) on the texture and colour of the most selected plant-based ball. Sensory analysis revealed that the 35:15 ratio received the highest preference levels for overall acceptability. Consequently, it was chosen for further chemical and physical analyses. The chemical composition of the preferred plant-based ball showed moisture, protein, fat, ash, and carbohydrate levels of 53.36%, 13.48%, 1.29%, 5.25%, and 26.62%, respectively. The calorie value was 172 kcal per 100g. Hardness and chewiness decreased with longer cooking time, with 10 min being optimal. Lightness (L^*) decreased from 37.44 to 13.06 as cooking time increased, with 10 min indicating medium lightness. Combining shiitake mushrooms with chickpea flour (35:15) creates a promising formulation with moderate protein content and favorable consumer preference.

INTRODUCTION

Meatballs, known for their distinct flavour, are a popular Indonesian dish and a form of processed meat product. The rise of plant-based meatballs stems from a growing trend of using plant-based ingredients as an alternative to traditional meat. In addition to being a valuable source of protein, plant-based meat closely mimics the flavour, colour, nutritional value, and texture of particular meats [1]. According to Mazumder et al. [9], the rapid development of plant-based meat substitutes has been fueled by several factors, including the growing number of vegans, heightened awareness of the benefits of substitutes over traditional meat products, and increased investment by the food industry. Analogues refer to products that closely resemble the structure of meat in terms of taste, texture, appearance, and occasionally nutritional composition, but are made from plant-based ingredients rather than animal meat [2]. These ingredients

boast essential amino acids, vitamins, minerals, fibre, and a certain protein content while offering lower calorie content [1]. Shiitake mushrooms are highly favoured in vegan cooking primarily for their umami flavour, which imparts a rich savoury taste similar to beef, meaty texture. Although there are thousands of mushroom varieties worldwide, only approximately 25 are typically consumed as food [3].

Shiitake mushrooms (*Lentinula edodes*) are the second most favoured edible mushroom around the world, contributing to approximately 25% of total mushroom production. In particular, shiitake mushrooms are not only appreciated for their organoleptic properties but also for their bioactive compounds, including β glucans, ergosterol, and phenolic compounds that act as antioxidants Morales[4]. Chickpea (*Cicer arietinum L.*) presents itself as a promising substitute for soy protein. Chickpeas contain approximately 17-22% protein, along with

fiber and complex carbohydrates [5]. Their neutral flavour, nutritional composition, and functional properties make them ideal for formulating plant-based meat analogues.

This study explores the synergy between shiitake mushrooms and chickpeas in plant-based ball formulations. Firstly, the study aims to determine the sensory characteristics of three different formulations of plant-based balls with varying shiitake mushroom to chickpea ratios (15:35, 25:25, and 35:15) using a 9-point hedonic scale. Secondly, to evaluate the physicochemical properties and calorie content of the selected formulation to support nutritional labelling, and lastly, the study investigates the effect of different cooking times (7, 10, and 13 minutes) on the texture and colour of the selected plant-based ball to determine optimal preparation conditions.

MATERIALS AND METHODS

Raw materials

Shiitake mushrooms (*Lentinula edodes*) were obtained from a local market in Selangor, Malaysia in 2024. Chickpea (*Cicer arietinum*) flour, tapioca starch, garlic powder and onion powder were purchased from a supermarket in Selangor, Malaysia.

Materials

Shiitake mushrooms were washed thoroughly until clean to remove any dirt and mud. Extra water was removed by keeping the mushrooms in a strainer for 5 minutes. The cleaned mushrooms were then blended for subsequent use.

Preparation of plant-based balls

The mushroom ball was prepared using different ratios of shiitake mushroom and chickpea flour (15:35, 25:25, and 35:15 w/w). The recipe for plant-based protein mushroom balls included shiitake mushrooms, chickpea flour, salt (0.70%), spices (0.97%), modified starch (13.91%), pepper (0.97%), and water (13.91%). All ingredients were mixed thoroughly, portioned, and shaped into round balls weighing approximately 10 g each. The prepared plant-based balls were stored at -18°C until further use.

Sensory evaluation

Approval for the sensory evaluation protocol was granted by the Ethics Committee, Universiti Teknologi MARA. Plant-based balls with different formulations (15:35, 25:25, and 35:15) were evaluated for sensory attributes by 30 panellists from the Faculty of Applied Sciences, UiTM Shah Alam, Selangor, Malaysia. The samples were served in random order. Sensory attributes assessed included chewiness, flavour intensity, umami, colour, saltiness, aftertaste, and overall acceptability using a 9-point hedonic scale (9 = like extremely; 0 = dislike extremely). Each attribute was evaluated to draw meaningful conclusions regarding consumer acceptability.

Proximate analysis

Proximate composition analysis was carried out to determine the nutritional quality of the plant-based balls. All analyses were performed in triplicate, and results were expressed as mean \pm standard deviation (SD) on a wet-weight basis unless otherwise stated. Standard analytical procedures described by the Association of Official Analytical Chemists (AOAC, 2000) were followed.

Moisture content

Moisture content was determined using the oven-drying method according to AOAC (2000) [6]. Approximately 2 g of homogenized sample was placed in a pre-weighed aluminium dish and dried in a hot air oven at 105°C until a constant weight was obtained. The percentage of moisture content was calculated based on the weight loss of the sample after drying.

Protein content

The total protein content was determined using the Kjeldahl method as described by AOAC (2000) [6]. Samples were digested in concentrated sulfuric acid with a catalyst mixture, followed by distillation and titration of liberated ammonia. The nitrogen content was converted to protein using a conversion factor of 6.25.

Fat content

Crude fat was extracted using the Soxhlet extraction method [6]. Approximately 3 g of the dried sample was placed in a thimble and continuously extracted with petroleum ether (boiling range $40\text{--}60^{\circ}\text{C}$) for 6 hours. The extracted solvent was evaporated, and the flask containing the residual oil was dried and weighed to determine the fat percentage.

Ash content

Ash content was determined by incinerating about 2 g of the sample in a muffle furnace at 525°C for 6 hours until a grey or white residue was obtained [1]. The percentage of ash was calculated as the ratio of the weight of the residue to the initial dry weight of the sample.

Carbohydrate content

Total carbohydrate content was estimated by difference, subtracting the sum of the percentages of moisture, protein, fat, and ash from 100%. The result was expressed as a percentage of the total sample composition.

Texture analysis

Textural parameters such as hardness and chewiness were determined using a texture analyser. Cylindrical samples of uniform size (approximately 10 g each) were equilibrated to room temperature before testing. The compression test was performed with a 50 mm cylindrical probe under the following conditions: pre-test speed 5.0 mm/s, test speed 10.0 mm/s, post-test speed 10.0 mm/s, and penetration distance 10.0 mm [7]. The peak force during compression represented hardness, while the product of hardness, cohesiveness, and springiness was taken as chewiness.

Colour measurement

The colour of the plant-based balls was measured using a chromameter (Konica Minolta Sensing, Japan), which was calibrated with a standard white calibration plate prior to analysis. Colour parameters were recorded in the CIE $L^*a^*b^*$ system, where L^* indicates lightness, a^* represents the red–green axis, and b^* represents the yellow–blue axis. Three readings were taken from different surface areas of each sample, and the average values were reported.

Statistical analysis

All experimental data were analysed using one-way analysis of variance (ANOVA) at a 95% confidence level to determine significant differences among sample means. When a significant difference ($p < 0.05$) was detected, Tukey's Honest Significant

Difference (HSD) post hoc test was performed to identify which groups differed significantly. Results were expressed as mean \pm

standard deviation (SD) based on triplicate measurements. Statistical analyses were carried out using IBM SPSS Statistics software.

RESULTS AND DISCUSSION

The results show significant results across different samples on different parameters. **Table 1** shows the results of the sensory evaluations of the plant-based ball.

Table 1. Sensory evaluation of plant-based ball.

Formulation	F1	F2	F3
Chewiness	4.83 ± 2.05 ^b	5.53 ± 2.43 ^{ab}	6.47 ± 2.52 ^a
Flavour Intensity	4.83 ± 2.28 ^b	5.77 ± 2.05 ^{ab}	6.83 ± 2.01 ^a
Umami	4.83 ± 2.15 ^b	5.43 ± 2.10 ^{ab}	6.27 ± 2.16 ^a
Colour	5.37 ± 1.79 ^b	5.77 ± 1.81 ^{ab}	6.50 ± 1.76 ^a
Saltiness	5.07 ± 2.16 ^a	5.37 ± 2.46 ^a	6.17 ± 2.21 ^a
After taste	5.17 ± 2.15 ^b	5.53 ± 2.19 ^b	6.77 ± 2.10 ^a
Overall acceptability	4.80 ± 2.28 ^b	5.53 ± 2.40 ^b	6.83 ± 2.25 ^a

Note: The three varieties of the plant-based ball formulations were F1 with a 15:35 shiitake mushroom to chickpea ratio, F2 with an equal 25:25 ratios and F3 with a greater ratio of 35:15. Values are expressed as mean ± standard deviation. Means values with different superscript letters were significantly different ($p < 0.05$) in the same row.

As shown in **Table 1**, F2 exhibit no significant difference from F1 across all attributes and was also comparable to F3 in terms of chewiness, flavor intensity, umami, colour, and saltiness. In contrast, a comparison between F3 and F1 revealed substantial differences in chewiness, flavor intensity, umami, colour, and aftertaste. F3 emerged as the most preferred formulation due to its superior aftertaste and overall sensory attributes. Consequently, it was selected for further evaluation, including proximate and physical analyses. **Table 2** shows a proximate analysis and a nutrition information of the plant-based ball (F3) per 100 g and per serving of 30 g. The ash content of the plant-based ball (F3) was 5.25%, indicating the total mineral content in the sample. For a plant-based product, this is comparatively high, indicating that the ingredients, like shiitake mushrooms and chickpea are rich in minerals [2]. While the moisture content of the plant-based ball (F3) was 53.36%.

Moisture content is an important factor in determining the overall quality and freshness of the plant-based ball. It is an important aspect in determining the texture, flavor, and shelf life of plant-based balls. The protein content of 13.48% is a reasonable value for a plant-based product such as this ball, providing a moderate protein source for those looking for plant-based alternatives to animal protein. The result was supported by a study that shows similar findings regarding protein content for meatball with edible mushroom as a fat replacer, which is 13.66% [8]. In contrast to beef meatballs, which had a protein content ranging from 7.39 to 12.51% in previous research, the selected plant-based ball (F3) had a protein content of 13.48%, which is higher than beef meatballs [9].

The combination of edible mushrooms and chickpeas in a plant-based ball (F3) creates a protein-rich product. Additionally, the plant-based ball (F3) contains a relatively low fat content of 1.29% making it a healthier choice for individuals aiming to reduce fat intake. The protein content of *Lentinula edodes* mushrooms ranged from 20% to 23%, meanwhile chickpeas boast a protein content ranging from 17% to 22% in their composition [5]. This low-fat content is attributed to ingredients such as shiitake mushrooms and chickpea flour, which are naturally low in fat.

The result was supported by a study, which showed similar findings for meatballs made with edible mushroom as a fat replacer had a 1.53% fat content [10]. These results align with previous studies that found shiitake mushrooms to have a low fat content, indicating that they are a low-calorie dietary food source [11]. The fat content of beef meatballs ranged from 7.05% - 9.25% [9]. Fat content in plant-based balls (F3) was lower than beef meatballs due to the plant-based balls are typically made from low-fat ingredients such as mushrooms and chickpeas, while beef meatballs contain animal fat, which is naturally higher in saturated fat content. The plant-based ball (F3) with less fat is a healthier alternative due to it satisfies the criteria for “a low-fat food”. As shown in **Table 2**, the carbohydrate content of the plant-based ball (F3) was 26.62%.

The results were obtained by calculating the difference, which involves subtracting the percentage of protein, ash, fat, and moisture content from 100%. The plant-based ball (F3) has higher carbohydrate content due to the content mainly comes from chickpea and shiitake mushrooms in the formulation, which are both rich in complex carbohydrates and fiber. The total caloric content (kcal) per 100 g of the samples was calculated using the Atwater values for fat (9 kcal/g), protein (4.02 kcal/g), and alternative values for carbohydrates (3.87 kcal/g). The resulting value was 172 kcal per 100 g of product (wet basis), which includes 53% moisture. This is notably lower than the typical energy content of conventional beef meatballs, which range between 200-270 kcal per 100g.

Table 2. Nutrition information of the plant-based ball per 100 g and per serving (30 g).

Nutrition Information		
Serving size: 100g		
Serving per package: 3		
	Per 100 g	Per serving (30 g)
Energy (kcal)	172	51.6
(kJ)	720	216
Carbohydrate (g)	26.62	7.99
Protein (g)	13.48	4.05
Fat (g)	1.29	0.39

Table 3 shows the texture and colour analysis of the plant-based ball (F3) at different cooking times. Values are expressed as mean ± SD of triplicate measurements. Superscripts with different letters are significantly different at $p < 0.05$ in the same row. The data in **Table 3** illustrates how cooking time affects the texture and colour of the plant-based ball at different times (7, 10, and 13 min). Hardness, springiness, cohesiveness, and chewiness are the parameters essential for understanding the textural quality and consumer acceptability of the product. There was a significant difference in hardness and chewiness properties treated with different cooking times. At 7 min, the hardness was significantly higher at 3583.52 N as compared to 13 min at 1673.05 N, showing that there was a significant difference and decrease in hardness as cooking time increased. This is due to the protein in the plant-based ball (F3) undergoing structural changes and softening as a result of prolonged heating. Extended cooking duration causes protein denaturation and myofibrillar degradation, resulting in diminished chewiness and hardness of plant-based ball [12]. Next, there were no significant differences in springiness and cohesiveness across the cooking times (7, 10, and 13 min). This suggests that cooking time did not significantly affect the springiness or cohesiveness of the plant-based ball (F3).

According to previous findings, meatballs with 10% shiitake and 74% meat maintain stable springiness (0.92 to 0.94) and cohesiveness, proving that plant-based ingredients can replicate meat-like texture [13]. The colour characteristics of the plant-based ball (F3) were assessed based on the L* values. The L* value represents the lightness of the mushroom ball. The colour results were displayed in **Fig. 1** and **Table 3**. The lightness (L*) values of the plant-based ball (F3) at different cooking times showed a significant decrease as the cooking time increased. At 7 min, the lightness (L*) values were 37.44, which was significantly higher as compared to 13 min at 13.06.

According to Liu et al. [14], found that darker colour is due to the heating, which triggers the Millard reaction, caramelization, and melanin pigments in the shiitake mushrooms. At 10 min, the lightness value showed a moderate lightness. This result was supported by previous research, which stated that Lightness (L*) values of meatballs with shiitake mushrooms decreased slightly as cooking time increased [13]. The reddish-brown of the mushroom may be influenced by the iron content in the shiitake mushrooms, particularly after heat treatment or cooking. Excessive heat can cause cell tissue to shrink, which concentrates the browning ingredients and makes the product appear dark brown [13].

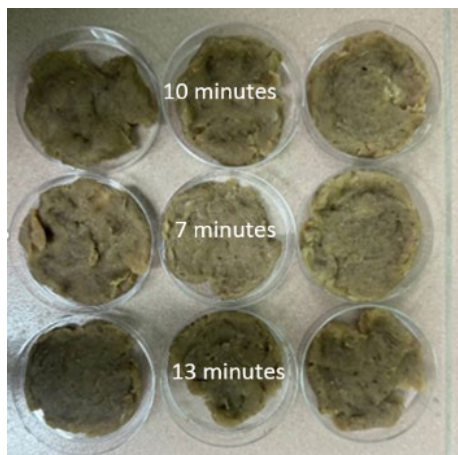


Fig. 1. Colour analysis of the plant-based ball with different cooking times.

Table 3. Texture and colour analysis of plant-based ball (F3) at different cooking times.

Time (min)	Properties	Treatment
7	Hardness (N)	3583.52 ± 944.17 ^a
10		3142.82 ± 710.98 ^{ab}
13		1673.05 ± 676.65 ^b
7	Springiness (Dimensionless)	0.90 ± 0.02 ^a
10		0.92 ± 0.06 ^a
13		0.94 ± 0.03 ^a
7	Cohesiveness (Dimensionless)	0.77 ± 0.04 ^a
10		0.76 ± 0.03 ^a
13		0.80 ± 0.01 ^a
7	Chewiness (g.mm)	2480.45 ± 449.99 ^a
10		2179.50 ± 316.08 ^a
13		1259.76 ± 528.00 ^b
Colour		
7	Lightness (L*)	37.44 ± 0.74 ^a
10		28.73 ± 1.25 ^b
13		13.06 ± 2.01 ^c

Note: Values are expressed as mean ± standard deviation. Means values with different letters were significantly different ($p < 0.05$) between the cooking time of the same properties.

This finding aligns with previous research by Asyryl et al. (2023) [13], which reported that the Lightness (L) values of meatballs containing shiitake mushrooms slightly decreased as cooking time increased. Excessive heat exposure can cause cell tissue to shrink, concentrating browning compounds and resulting in a darker brown appearance.

CONCLUSION

The study successfully evaluated the physical, chemical, and sensory characteristics of plant-based balls, demonstrating their potential as a nutritious and appealing alternative to conventional meat-based products. Among the formulations tested, F3 was the most favoured, likely due to the optimal combination of shiitake mushrooms and chickpeas, which enhanced both texture and flavor. Proximate analysis of F3 revealed a nutritional profile of 172 kcal per 100 g with 53.36% moisture, 13.48% protein, 1.29% fat, 5.25% ash and 26.62% carbohydrates, making it a suitable option for plant-based diets. Its health benefits are underscored by its high protein and complex carbohydrate content, along with a low fat content. Physical testing showed that cooking time significantly impacted the texture of F3, with 10 min being the optimal duration to maintain ideal hardness and chewiness. However, springiness and cohesiveness were not greatly affected by cooking time. Additionally, cooking time influenced the colour characteristics, with a medium lightness at 10 min of cooking being determined to be the most desirable.

CONFLICT OF INTEREST

The authors have declared that no conflict of interest exists.

ETHICS STATEMENT

Ethical approval for the involvement of human subjects in this study was granted by FERC FSG University Research Ethics Committee, Reference number FERC/FSG/24/077(UG)

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