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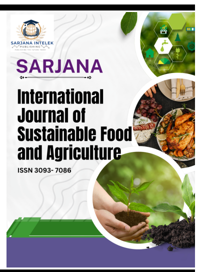

Screening Physicochemical Properties of Rabbit Nuggets Enriched with Chickpea Flour: A Plackett-Burman Design Approach

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ABSTRACT

Rabbit meat is valued for its outstanding nutritional profile, characterized by high protein and low-fat content. However, price constraints and restricted processing alternatives have hindered its widespread usage. The addition of plant-based extenders, such as chickpea flour, has possibilities for boosting the texture and economic feasibility of rabbit meat products. This study aims to screen significant ingredients affecting the physicochemical parameters of rabbit nuggets enriched with chickpea flour using Plackett-Burman Design (PBD). Minitab software (version 21.1) was utilized to generate 12 experimental formulations. The study investigated the influence of ingredient composition on four key responses: pH, fat content, moisture content, and hardness. Pareto analysis was employed to identify statistically significant factors ($p<0.05$), followed by a response optimizer technique to obtain the optimal ingredient values. Results demonstrated that chickpea flour and textured vegetable protein (TVP) significantly influenced product pH ($p<0.05$), while ice content was a significant factor for hardness ($p<0.05$). Notably, no factors showed a statistically significant effect on fat and moisture content within the tested ranges. The multi response optimizer determined the optimal formulation composition (g/100 g) to be: rabbit meat (65.00), shortening (4.90), TVP (5.00), ice (4.30), tapioca starch (3.21), and chickpea flour (2.73) with a composite desirability (D) of 0.83. These findings demonstrate the possibility of developing quality-enriched rabbit nuggets using a systematic screening design.

1. Introduction

Nowadays, the global demand for processed chicken products is expanding. This is because these products are easily available on the market, quick and ready to eat. However, traditional chicken-based nuggets are generally linked with high fat content and promote a shift towards healthier eating alternatives [1]. Consumer confidence in rabbit meat is increasing every year due to its nutritional content which is good for health. This meat contains high protein and is low in fat and cholesterol

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compared to meat from other animals[2]. Despite these advantages, the commercial consumption of rabbit meat is still limited by high production costs, limited market availability and consumer unfamiliarity with its preparation.

Nuggets are a very popular fast snack around the world and are loved for their ease of preparation and taste [3]. Reformulating nuggets with rabbit meat provides a healthier alternative to chicken nuggets which a low cholesterol and high protein alternative to nuggets [4]. However, lowering the quantity of fat in processed meat-based products might have a negative impact on texture resulting to a rougher texture or a dry taste. Therefore, the problem of producing rabbit nuggets involves on maintaining the right texture while taking advantage of the healthy benefits of rabbit meat [5].

To minimize the economic cost and improve texture of rabbit-based products, the introduction of plant-based substances has gained attention. Chickpea flour, which is rich in protein, dietary fiber and minerals, has the potential to give considerable benefits as a binder and improver for meat-based products [4]. The ability of this plant to increase water holding capacity and improve the textural quality of the product makes it an excellent option for increasing the quality of meat products produced while reducing production costs [6]. However, studies on the development of nuggets other than chicken or beef requires a thorough investigation of multiple factors such as meat content and other ingredient. On the other hand, products for chickpea enriched rabbit nuggets are still limited. The study needs to be performed carefully because each ingredient has a role and can significantly influence the texture and nutrition of the final product.

Traditional one factor at a time study methodologies are time consuming and imprecise for investigating the effect of interactions within a process or product formulation. Therefore, statistical screening methods such as Plackett-Burman Design (PBD) are significant as one method to systematically screen the most critical variables from a wide variety of potential factors before optimizing using more complex models such as Box-Behnken design (BBD) or Central Composite design (CCD) [7]. Since the RSM approach, notably PBD, has been widely employed especially in product screening, it was adopted in the present study. This study aims to screen the critical ingredients that affect the physicochemical properties of rabbit nuggets enhanced with chickpea flour using PBD. Then, using the multi-response optimizer function, the optimal formulation composition of rabbit nuggets was determined.

2. Methodology

2.1 Overview

The experimental procedure for this study was split into three different phases, as shown in Figure 1. The procedure began with pre-treatment, which included the preparation of raw materials and manual deboning of rabbit meat. This was followed by a screening phase, where PBD was used to study the influence of formulation ingredients on four essential quality attributes: pH, fat content, moisture content and hardness. Finally, the study ended with a response optimization phase, which was utilized to find the ideal formulation for chickpea-enriched rabbit nuggets.

PRE - TREATMENT
Deboner and raw material preparation

⇩

SCREENING: PLACKETT-BURMAN DESIGN (PBD)
Factor: Rabbit meat, shortening, TVP, ice, ISP, tapioca starch and chickpea flour
Responses: pH, fat, moisture, and hardness

⇩

RESPONSE OPTIMIZER
Validation and produce best formulation of the model

Fig. 1. Overview workflow of research study

Experimental design and statistical analysis were performed using Minitab® 21.1 software (Minitab Inc., State College, PA, USA). A PBD was applied to construct 12 experimental formulations [8], [9]. The study investigated the effect of formulation variables on four critical responses: pH, fat content, moisture content, and hardness. Analysis of variance (ANOVA) was used to analyze the statistical significance of the terms at a significance level of $p<0.05$. Pareto analysis was performed to screen for statistically significant factors ($p<0.05$), followed by the implementation of the response optimizer technique to obtain the appropriate formulation composition (g/100 g).

2.2 Screening with Plackett-Burman Designs (PBD)

The present study utilized a PBD to screen for significant formulation factors. Seven independent factors were evaluated: rabbit meat, shortening, TVP, ice, ISP, tapioca starch, and chickpea flour. The significant influence of each factor was determined by comparing the mean response values between the low (-1) and high (+1) coded levels. Table 1 outlines the experimental factors and the corresponding amounts.

Table 1
Levels for factors in Plackett-Burman design

Factor	Low (-1)	High (+1)
A: rabbit meat	65	70
B: shortening	3	5
C: TVP	3	5
D: Ice	4	8
E: ISP	0	2
F: Tapioca Starch	3	6
G: Chickpea Flour	2	8

The most relevant factors of the major ingredient that influences rabbit nugget formulation was generated utilizing a 12-experiment produced design as indicated in Table 2. The lowest levels for rabbit meat, shortening, TVP, ice, ISP, tapioca starch and chickpea flour were 65, 3, 3, 4, 0, 3 and 2, respectively. The highest levels of rabbit meat, shortening, TVP, ice, ISP, tapioca starch and chickpea flour were 70, 5, 5, 8, 2, 6 and 8, respectively. All analyses for response (pH, fat, moisture, and hardness) were performed at room temperature. The Pareto chart constructed was used to determine the factors that significantly affect the rabbit nugget formulation.

Table 2
Levels for factors in Plackett-Burman design

Run	A Rabbit Meat	B Shortening	C TVP	D Ice	E ISP	F Tapioca Starch	G Chickpea Flour
1	70	3	3	4	2	6	8
2	70	5	3	8	0	3	2
3	70	5	5	4	2	6	2
4	65	5	5	4	2	3	2
5	70	3	5	8	0	6	2
6	70	5	3	8	2	3	8
7	65	5	3	4	0	6	8
8	65	3	3	8	2	6	2
9	65	5	5	8	0	6	8
10	70	3	5	4	0	3	8
11	65	3	5	8	2	3	8
12	65	3	3	4	0	3	2

*Each factor in the formulation (A-G) was weighed in grammes and added premix to a total of 100 g

2.3 Response Optimizer

To optimize all responses with various targets, a response optimizer technique based on the desirability function and a graphical optimization strategy based on the overlay plot can be used [10]. A composite desirability (D) with values ranging from 0 (unacceptable response value) to 1 (desirable value) was used to optimize seven responses at the same time.

3. Results

3.1 Plackett-Burman Design (PBD) for Screening of Rabbit Nugget

This PBD design analyzes the most important effects based on the results of tests conducted in accordance with a matrix that examines low and high levels of factor. Using the PBD, it was efficiently applied to develop 12 various rabbit nugget formulations. This experimental design technique allows excellent screening of varied factors and their effects on the response variable of interest [9]. By utilizing this method, each formulation can identify the percentage of each element used in the formulation of rabbit nugget. As a result, the 12 formulas produce rabbit nugget results with a good appearance, which is a key quality characteristic.

The Pareto chart as shown in Figure 2 displays the significant effects of multiple factors on the response variable pH, using a significant difference of $p<0.05$. Each bar indicates a factor and its length indicates how much it effects the pH value. Factors with a standardized effect greater than the red reference line (2.776) are statistically significant at the ($p<0.05$) level. Chickpea flour demonstrated the strongest standardized effect drastically effect the pH value. It was the most important factor in this study. The pH of raw rabbit meat normally ranges from 5.60 to 5.80, depending on the breed and muscle type [11], [12]. This low pH is attributed to the formation of lactic acid after 24 hours post-slaughter [12]. The addition of chickpea flour resulted in a significant increase in the pH of the rabbit nuggets ($p<0.05$). This can be explained to the natural pH of chickpea flour, which has been observed to range from 6.38 to 6.52 [13], effectively buffering the lower acidity of the rabbit meat. The major effect of chickpea flour on pH can be related to the buffering ability of legume proteins, which may neutralize the acidity of the meat matrix, hence increasing the water holding capacity and textural tenderness of the rabbit nuggets [14].

The results of the PBD showed that TVP had the second most significant positive effect (p<0.05) on the pH of rabbit nuggets. This increase in pH can be explained to the intrinsic alkalinity of soy-based proteins, which typically have a pH between 6.4 and 6.8, substantially higher than the pH of rabbit meat (about 5.6–5.8) [11]. The addition of TVP supplied buffering capability to the meat matrix, neutralizing the lactic acid found in muscle tissue. This finding is in accordance with previous studies on meat extenders, where the integration of soy protein was found to improve the pH of the final product, hence enhancing water holding capacity [11]. While other factor such as rabbit meat, shortening, ice, ISP and tapioca starch did not show a substantial variation in the pH levels.

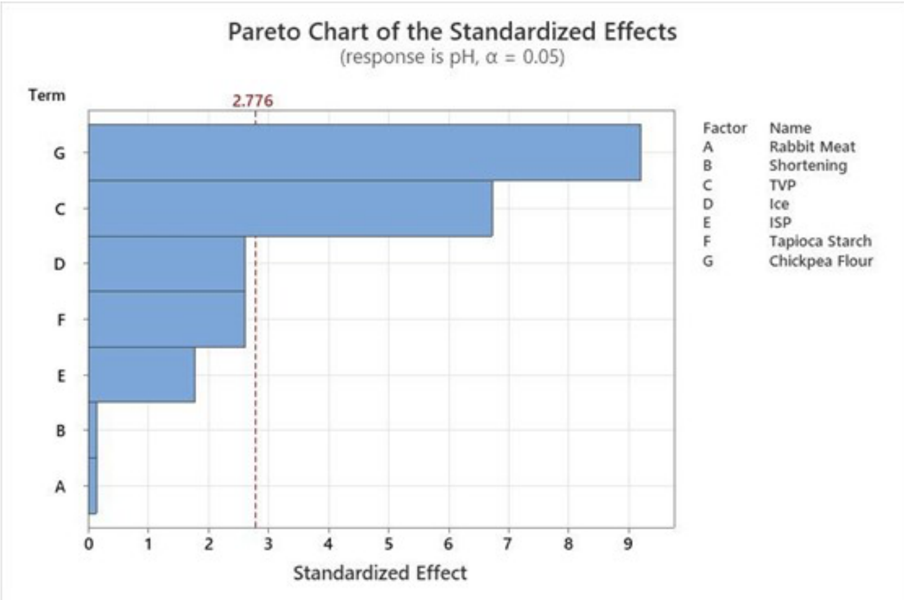

Fig. 2. Pareto chart of pH analysis

Figure 3 shows a Pareto chart that illustrates the standardized effects of seven factors on the response variable of fat content. The red dashed line at the standardized effect of 2.776 functions as the reference line for the graphic at the significance level. Any effect over this threshold is deemed statistically significant at the 0.05 level. The study findings indicate that all components remain below the threshold value of 2.776. This indicates that none of the parameters exert a statistically significant influence on fat content at the significance threshold of P<0.05 within the boundaries of this experiment. This finding demonstrates that, within the experimental parameters established for this PBD, no changes in ingredients exerted a statistically significant effect on the end result fat content of the nugget. Despite rabbit meat and shortening factor had the greatest effect magnitude among the factors, they failed to achieve statistical significance. This is due to rabbit meat's significantly lower fat content compared to other animals [15]. This study demonstrates that these ingredients can be combined to enhance texture or yield while maintaining the low-fat characteristics of rabbit nuggets. This "nutritional stability" is a valuable attribute for industrial production, indicating that the formulation is stable and maintains a uniform fat profile despite minor variations in ingredient composition [16].

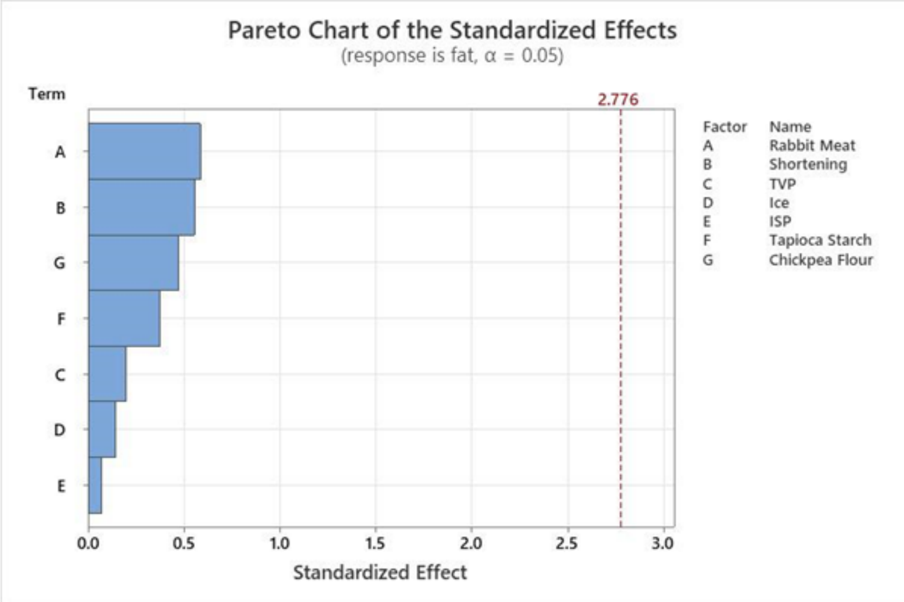

Fig. 3. Pareto chart of fat analysis

Figure 4 shows the Pareto chart illustrating the standardized effects of the seven factors on the moisture content response parameter. The red dashed line at the standardized effect of 2.776 acts as the reference line for the graphic at the significance level. Any effect exceeding this threshold is deemed statistically significant at the $p<0.05$ level. The research indicates that no component exceeds the threshold value of 2.776. This indicates that none of the variables in this experiment exert a statistically significant influence on moisture content at the $p<0.05$ significance level. Although shortening and ice showed the largest standard effects among the factors studied, their influence remained not statistically significant within the constraints of the experimental design. This lack of change suggests that the nugget formulation has a high level of physicochemical stability. Moisture retention in the product is principally controlled by the creation of a stable protein-water-fat matrix (emulsion stability). The observed consistent moisture content can be due to the functional qualities of the non-meat constituents, particularly chickpea flour and tapioca starch. These ingredients are noted for their strong water holding capacity due to their carbohydrate and protein composition. The presence of these hydrocolloids probably buffers the system, absorbing the extra water introduced by fluctuations in ice content and reducing moisture loss during thermal processing [17]. As a result, the rabbit nugget formulation demonstrates desired stability, preserving juiciness and consistent production regardless of product composition variations.

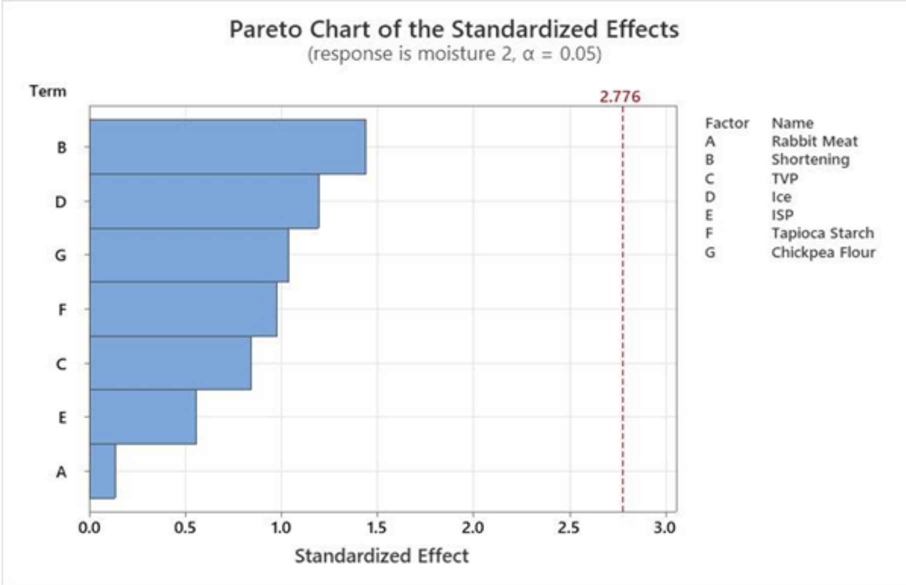

Fig. 4. Pareto chart of moisture analysis

The Pareto chart in figure 5 illustrates the standardized effects of seven factors on the hardness response variable. The red dashed line at the standardized effect of 2.776 acts as the chart reference line at the p<0.05 significance level. Any effect greater than this threshold is considered statistically significant at the p<0.05 level. Only one factor showed significance above the critical value of 2.776, indicating that it had a statistically significant effect on texture at the p<0.05 level. Other ingredients such as rabbit meat, shortening, TVP, ISP, tapioca starch and chickpea flour, revealed no significant differences. Further research could adjust the levels of important elements or introduce new elements to better understand their influence on texture. The strong influence of ice content on nuggets hardness can be related to its two roles in the creation of meat emulsions. First, ice functions as a solvent for myofibrillar proteins, which are necessary for creating the gel matrix that gives the nuggets their structure [18]. Second, the ratio of water to protein directly effects the density of this structure. Higher water additions frequently serve to enhance the gap between protein chains and provide a smoother texture, whereas lower water levels make a denser, harder texture [19].

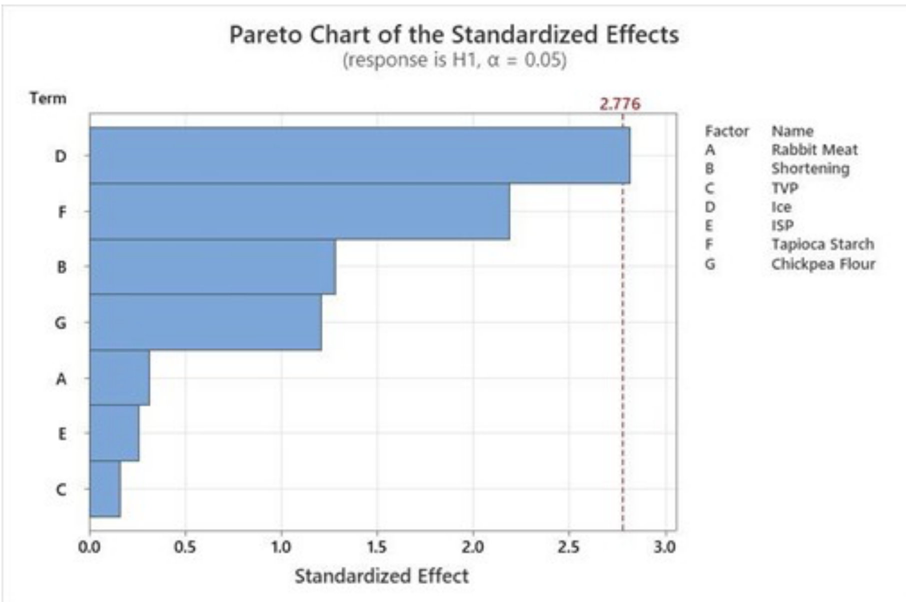

Fig. 5. Pareto chart of hardness analysis

Table 3 shows the response optimization of the rabbit nugget. In the study, the best formulation among 12 formulations had a pH, fat and moisture and hardness value of 6.30, 15.01, 176.71 and 246.04, respectively. This suggests a decreased fat content and a visible pH, moisture and hardness in the texture of the rabbit nugget. On the other hand, the rabbit nugget formulation produced uses chickpea flour and other binders to improve product qualities. The use of chickpea flour in meat products is known to improve the texture and act as a binder, improving water and fat binding qualities. The multi response optimization analysis showed a composite desirability (D) of 0.83, indicating that the generated formulation satisfied the combined desired characteristics for pH, fat, moisture, and hardness. The comparison shows diverse methods to fat content and texture optimization in meat products, highlighting the many strategies employed in product development.

Table 3
Response optimization of the rabbit nugget

Solution	*Rabbit Meat*	*Shortening*	*TVP*	*Ice*	*ISP*	*Tapioca Starch*	*Chickpea Flour*	*pH Fit*
1	*65*	*4.89899*	*5*	*4.30057*	*0.0000015*	*3.21212*	*2.73294*	*6.30001*

Solution	*Fat Fit*	*Moisture Fit*	*Hardness Fit*	*Composite Desirability*
1	*15.0089*	*176.701*	*246.037*	*0.832818*

4. Conclusions

In conclusion, the multi response optimizer determined the optimal formulation composition (g/100 g) to be: rabbit meat (65.00), shortening (4.90), TVP (5.00), ice (4.30), tapioca starch (3.21), and chickpea flour (2.73) with a composite desirability (D) of 0.83. Chickpea flour and TVP significantly influenced the pH value of the rabbit nugget ($p<0.05$). While only ice significantly affected the hardness of the rabbit nugget ($p<0.05$). PBD has been effectively implemented in rabbit nugget formulation and indicated that three parameters (chickpea flour, TVP and ice) have significant impacts on two responses (pH and hardness). This study has revealed the possibility and possible advantages of adding chickpea flour into rabbit meat to improve texture. Chickpea flour has been demonstrated to be effective combine with rabbit meat while having a good influence on its textural features. Future research paths should target further optimization of chickpea flour inclusion levels and processing procedures to attain the desired low-fat content without compromising texture. Exploring interactions between chickpea flour and other nugget ingredients, combined with innovative processing processes, can provide greater insights into texture improvement. Other than that, a comparative analysis of commercial chicken nuggets and the formulated rabbit nuggets can understand their market feasibility. This research contributes to developing a nutritionally balanced product, with an attractive and consumer acceptable texture with rabbit meat and enrich with chickpea flour.

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