DEVELOPMENT AND CHARACTERIZATION OF LOW-FAT MAYONNAISE INCORPORATING SOYBEAN PROTEIN AS A FAT REPLACER

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Abstract
The development of plant-based mayonnaise represents a significant innovation in the food industry, driven by increasing consumer demand for healthier, egg-free alternatives. This study focused on the formulation and optimization of soybean-based mayonnaise, evaluating its nutritional composition, sensory attributes, and shelf life. Soybean oil bodies (SOB) were used to replace egg yolk at varying levels (25–100%), resulting in reduced fat content and improved moisture retention. Proximate analysis showed that protein and fat decreased with increasing SOB, whereas carbohydrate content increased (p<0.05). Color analysis indicated higher lightness and reduced redness and yellowness in SOB-based treatments. Physicochemical properties and sensory evaluation confirmed the product's stability and acceptability during storage. The findings highlight soybean-based mayonnaise as a nutritious and sustainable alternative to conventional products.

1. INTRODUCTION

Soybean, also known as *Glycine max*, has been cultivated for over 5000 years, originally in China. It later spread to Japan, Korea, Southeast Asia, and by the 18th century, reached Europe and North America. Evolving from its wild ancestor *Glycine soja*, soybean became a major global crop due to its high-quality protein and vegetable oil yields. Today, Brazil leads in soybean production with 169 million metric tons, followed by the United States with 118.84 million metric tons, and Argentina with 48.21 million metric tons. China and India contribute significantly as well, though China remains the largest importer of soybeans to meet its rising domestic demand, while India uses most of its production for soy meal and oil (Dukariya *et al.*, 2020).

Soybeans are valued for their balanced nutritional composition. They contain around 40% protein, 20% fat, 30% carbohydrates, 17% fiber, and 5% ash. This makes them highly suitable for a wide range of food applications. The high protein content provides essential properties such as emulsification, gelling, and water-binding, which are particularly useful in food products like mayonnaise. The unsaturated fats present in soybeans contribute to heart health while also enhancing texture and mouthfeel. Their carbohydrate content helps control viscosity and

texture, and the fiber aids digestion and increases the feeling of fullness. Essential minerals such as calcium, iron, potassium, and magnesium enhance both nutrition and food stability. Soybeans are also rich in isoflavones, plant-based phytoestrogens that have been linked to health benefits like hormone balance, reduced menopausal symptoms, and a lower risk of certain types of cancers (Tamangwa *et al.,* 2023).

Soybeans are a complete protein source, containing all the essential amino acids, making them a valuable part of plant-based diets. They are low in saturated fat, cholesterol-free, and contain beneficial omega-3 fatty acids, supporting cardiovascular health. The high fiber content aids digestion and stabilizes blood sugar levels. However, soybeans also contain anti-nutritional compounds such as phytates, oxalates, gums, saponins, and trypsin inhibitors, which can hinder the absorption of minerals if not properly processed (Fang and Kong, 2022).

The uses of soybeans extend beyond food. Compounds like isoflavones and lecithin are used in cosmetics and pharmaceuticals, often in anti-aging products and cholesterol-lowering supplements. Soy protein hydrolysates are being explored in sports nutrition due to their fast digestibility and support for muscle recovery. As global awareness of sustainability and health grows, soy protein is being used as a foundation for plant-based meat and dairy alternatives. Research also highlights the therapeutic benefits of soy-derived bioactive peptides with anti-inflammatory, antioxidant, and antihypertensive effects (Voora *et al.,* 2020).

Mayonnaise, a widely consumed condiment, is a key example of a food product where soy-based ingredients can make a significant impact. Condiments are used globally in everyday meals, and mayonnaise stands out for its rich flavor and emulsified texture. Traditionally, mayonnaise is made from refined vegetable oils, egg-based ingredients, dairy components, sugar, starches, spices, and preservatives. Though flavorful and popular, conventional mayonnaise contains 70–80% oil, mostly saturated fat, and has been linked to health concerns such as obesity, high

cholesterol, and cardiovascular diseases (Van Eck *et al.,* 2020).

These concerns have led to increased demand for low-fat and vegan mayonnaise options. As consumers shift toward healthier diets, food manufacturers are replacing eggs and saturated fats with plant-based alternatives like soy proteins, pea proteins, and hydrocolloids. These ingredients help maintain texture, stability, and flavor while lowering calorie content. Innovations include using soy milk, canola, wheat germ, and white lupin to create egg-free, low-fat mayonnaise. Soy milk has shown particular promise in replacing egg yolk due to its emulsifying properties and nutritional benefits (Huang *et al.,* 2022).

Mayonnaise has several functional characteristics that make it versatile in food preparation. Its emulsifying agents create stable oil-water mixtures with a smooth consistency. It has high water-holding capacity, retains moisture, and maintains oxidative stability, helping to prevent rancidity. The product's viscosity ensures good thickness and spreadability, while its ability to carry fat-soluble flavors ensures even flavor distribution. The smooth mouthfeel and adhesion to food surfaces enhance sensory appeal. The acidic components lower the pH, increasing microbial safety and shelf life. In low-fat versions, emulsifiers like soy lecithin and soy proteins play a vital role in replicating these properties by stabilizing oil droplets and improving texture and mouthfeel. Additional ingredients like hydrocolloids and starches are used to compensate for the fat reduction, preserving creaminess and consistency (Wang *et al.,* 2022).

One promising innovation in low-fat mayonnaise production is the use of soy oil body, which is naturally rich in unsaturated fat and can replace egg yolk to create a cholesterol-free product. Soy oil body also contains antioxidants like tocopherol and phytosterols, which improve shelf life and product stability. However, relying solely on soy oil can lead to issues like product instability or quality deterioration. To address this, ingredients like xanthan gum, modified starch, and pectin are used to promote flocculation and prevent the movement of oil droplets. These hydrocolloids

stabilize the emulsion, enhance texture, and prevent separation. Hydrophobic hydroxypropyl distarch can also be incorporated into oil-in-water emulsions to further improve structural integrity and quality. This combination of ingredients allows the creation of low-fat mayonnaise products that closely match traditional formulations in both texture and sensory appeal (Mirzanajafi-Zanjani *et al.,* 2019).

In summary, soybeans stand out not only for its nutritional value but also for its functional versatility in food systems. Its protein, oil, and bioactive compounds provide numerous benefits in health, nutrition, cosmetics, pharmaceuticals, and food innovation. With increasing consumer demand for sustainable and health-conscious products, soy-based alternatives are driving the development of vegan, low-fat, and egg-free mayonnaise options that maintain the taste, texture, and stability of conventional products. This study aims to develop mayonnaise with varying concentrations of soybean oil. Subsequently, assess the effect of soybean oil on the chemical composition, physicochemical, and sensory analysis of mayonnaise.

2.   MATERIALS AND METHODS

2.1. Procurement of Raw Material
All the raw materials (soybeans, soybean oil, eggs, salt, sugar, and vinegar) were purchased from the local market of Faisalabad. Chemicals and reagents were used of analytical grade (Sigma Aldrich, Germany).

2.2. Extraction of Soybean Oil
Extraction of soybean oil (SO) was carried out according to the method described by Zhou *et al.* (2022) with minor modifications. In this method, clean soybeans were soaked in ionized water at 20% (w/v) for 12 hours. Swollen soybeans were homogenized by a tissue grinder in ice-cold deionized water, and the solution was filtered through degreased gauze to obtain the filtrate. Sucrose was added to the filtrate and diluted to 20% (w/v). The solution was centrifuged at 10,000 rpm for 20 minutes to obtain the upper suspended paste, and the process was repeated twice. The paste obtained was suspended again in deionized water and centrifuged at the same conditions to obtain the SO. Finally, the prepared SO was heated at 100°C for 10 minutes to inactivate the enzymes.

2.3. Preparation of Mayonnaise Samples
Mayonnaise was prepared at different soybean oil concentrations (Table 1) according to the reported procedure (Wang *et al.,* 2022).

Table 1. Development of mayonnaise using varying concentrations of soybean oil		
Treatments	Egg Yolk (%)	Soybean Oil Body (%)
T_0	100	-
T_1	75	25
T_2	50	50
T_3	25	75
T_4	-	100

2.4. Chemical Composition
The chemical composition of the supplemented mayonnaise was measured by using the suggested procedure (AOAC, 2023).

2.5. pH Analysis
For pH determination, the AOAC (2023) official method was followed using a digital pH meter. Before analysis, the pH meter was calibrated with standard buffer solutions of pH 4.0 and pH 7.0 at room temperature to ensure measurement

accuracy. Approximately 10 g of the mayonnaise sample was homogenized with 10 mL of distilled water to create a uniform slurry, allowing for consistent electrode contact and accurate readings. The homogenized sample was placed in a clean beaker, and the electrode was carefully immersed, ensuring it was fully submerged without touching the container's sides or bottom. The reading was allowed to stabilize for about 1 to 2 minutes before recording the pH value. All measurements were conducted at a controlled temperature of 25°C, as temperature variations can affect pH readings.

2.6. Water Activity

Water activity (aw) was determined by following the AOAC (2023) method. Water activity (aw) of soy-based mayonnaise was measured by using a calibrated water activity meter. Before measurement, the instrument was calibrated at room temperature using standard saturated salt solutions with known aw values to ensure accuracy. Approximately 5 g of mayonnaise was placed into the sample cup, ensuring the surface was smooth and free of air pockets to avoid measurement errors. The cup was then sealed in the measurement chamber to prevent moisture exchange with the environment during analysis. The device was allowed to equilibrate until a stable reading was obtained, typically within 5 to 10 minutes. Measurements were conducted at 25°C, as temperature fluctuations can significantly affect aw values.

2.7. Color analysis

The color of mayonnaise was assessed using a CIE Lab SPACE system (Color Tec-PCM, NY, USA), following established methods (Khan *et al.*, 2021). The parameters L*, a*, and b* were measured in triplicate.

2.8. Texture Analysis

The texture of mayonnaise samples was analyzed using a texture analyzer, following the method of Sun *et al.* (2018). The consistency of the samples was measured with a Texture Analyzer fitted with a 5000 g load cell. A back extrusion cell with a 35 mm diameter compression disc was used for testing. The mayonnaise samples were carefully transferred into Petri dishes, reaching a depth of

55 mm. A single cycle was executed at a steady velocity of 1 mm/s until a depth of 40 mm was reached. A force vs. time curve was recorded, displaying values on the monitor for texture properties such as firmness and adhesiveness. Firmness was identified as the highest force applied when the test cell penetrated the mayonnaise. The area under the opposing force curve, representing the energy needed to detach the compressing nozzle from the sample, was termed adhesiveness. Texture analyzer, compression method, Texture Analyzer with a 5000 g load cell, back extrusion cell, and transferring samples into Petri dishes belong to one set. Compression disc, sample depth, steady velocity cycle, force vs. time curve, and texture properties belong to another set.

2.9. Total Bacterial Count

The total bacterial count of soybean mayonnaise was analyzed using the AOAC (2023) method. The mayonnaise samples were first homogenized, and serial dilutions were prepared with sterile saline as the diluent. A 10-fold serial dilution was performed, and 100 µL of the diluted sample was transferred to plate count nutrient agar medium using the pour plate method. The plates were gently swirled to distribute the sample evenly on the agar and left to solidify. Incubation was carried out at 37°C for 24–48 hours. The total bacterial count was determined by multiplying the colony count by the corresponding dilution factor, providing an estimate of bacterial load in the mayonnaise samples.

2.10. Sensory Analysis

Organoleptic properties of the sample were evaluated by a panel of 20 semi-trained judges. Before evaluation, the judges were provided with a description of the 9-point hedonic scale and instructed to score the sample relative to the control sample. The judges evaluated attributes such as flavor, color, texture, mouthfeel, spreadability, and general acceptability using the 9-point scale. The exact composition of the mayonnaise was not disclosed to ensure unbiased judgment. Samples were served on white polystyrene plates at room temperature and labeled with 3-digit codes. Judges were seated in

separate booths and instructed to rinse their mouths with water between evaluations to neutralize their taste buds (Meilgaard's, 2023).

2.11. Statistical Analysis

All experiments were carried out in triplicate, and results are presented as mean values along with standard deviations. The Tukey HSD test (α = 0.05) was used to assess significant differences among treatments using Statistix 8.1.

3. RESULTS AND DISCUSSION

3.1 Chemical Composition of Soybean Mayonnaise

The effect of varying concentrations of soy oil body (SOB) on the proximate composition of soybean-based mayonnaise formulations was analyzed, revealing statistically significant differences across all treatments (T_0–T_4) for moisture, crude protein, crude fat, ash, and carbohydrate content. Moisture content exhibited a positive correlation with increased SOB incorporation, ranging from 30.12% in the control (T_0) to 34.86% in T_4. The increase in moisture across treatments T_1 to T_4 can be attributed to the presence of moisture-binding components such as soy proteins and hydrocolloids, which enhance the water-holding capacity and emulsion stability. These findings are consistent with previous studies that demonstrated improved moisture retention in emulsions with plant and animal gums or hydrocolloids such as chitosan and guar gum (Al-Aubadi et al., 2021; Kumar et al., 2021). Crude protein content decreased progressively as the proportion of SOB increased, with the highest value recorded in T_0 (3.35%) and the lowest in T_4 (2.05%). The decline was associated with the replacement of egg yolk, which is rich in protein, with SOB, which contains relatively less protein. Despite this reduction, all formulations maintained structural stability, suggesting that a balanced protein level can still be achieved using plant-based emulsifiers. Similar trends were reported in formulations using soymilk as an emulsifier (Obatolu et al., 2018) and studies highlighting challenges in protein retention in low-fat emulsions (El-Deab et al., 2018; Ihsan et al., 2021).

A significant reduction in crude fat content was observed across all treatments, indicating the successful reduction of lipid concentration in the formulations. T_0 showed the highest fat content (53.28%), consistent with conventional mayonnaise formulations, while T_4 recorded the lowest (30.15%). The trend highlights the effectiveness of SOB in producing a lower-fat product without compromising emulsion texture and mouthfeel. These results are supported by previous work evaluating the use of medium-chain triglyceride blends and soy-based emulsifiers in low-fat mayonnaise development (Hsu et al., 2020; Odep et al., 2024). Ash content, representing the mineral fraction of the formulations, showed a consistent increase with rising levels of SOB. The ash content ranged from 1.32% in T_0 to 1.93% in T_4. The enhancement in ash content indicates an improved mineral profile in soy-based mayonnaise, possibly due to the inherent mineral richness of SOB and modifications during processing that favor mineral retention. This observation aligns with findings by Wiguna et al. (2023) and Heikal et al. (2023), who reported similar improvements in ash content and product integrity in low-fat soy-based emulsions. Carbohydrate content increased substantially with SOB concentration, with the control T_0 recording the lowest value (24.75%) and T_4 reaching a peak of 84.92%. This rise was due to a compositional shift in which fat and protein were replaced with carbohydrate-rich plant materials and stabilizers. The results confirm that SOB incorporation contributes not only to fat reduction but also to carbohydrate enhancement. Comparable patterns were observed in vegan mayonnaise formulations using defatted soybean flour or blended oils, as reported by Sung Gil et al. (2024) and Liu et al. (2024).

Table 2. Chemical composition of soybean mayonnaise (%)					
Treatments	Moisture	Crude Fat	Crude Protein	Ash	Carbohydrates
T_0	30.32 ± 2.12^e	53.28 ± 3.72^a	3.35 ± 0.23^a	1.32 ± 0.09^d	24.75 ± 1.73^e
T_1	31.30 ± 2.19^d	48.42 ± 3.38^b	3.24 ± 0.22^b	1.54 ± 0.10^c	47.36 ± 3.31^d
T_2	32.49 ± 2.27^c	45.05 ± 3.15^c	2.19 ± 0.15^c	1.76 ± 0.12^b	53.84 ± 3.76^c
T_3	33.44 ± 2.34^b	43.39 ± 3.03^d	2.12 ± 0.14^d	1.83 ± 0.12^b	68.04 ± 4.76^b
T_4	34.86 ± 2.44^a	30.15 ± 2.11^e	2.05 ± 0.14^d	1.93 ± 0.13^a	84.92 ± 5.94^a

3.2. pH Analysis

The pH of soybean oil mayonnaise is crucial to its stability, texture, and microbial safety. Mean squares for pH of various treatments were statistically analyzed as depicted in table 4.6. The mean values of all five treatments during storage of 12 days and evaluation at every 4-day interval is shown in table 4.6. indicated that the results obtained were highly significant. The parameter should range from (3.6 to 4.2) for proper emulsification as well as preventing the growth of microbes. At 0 days, the treatments T_3 and T_4 yielded the highest values of pH (4.13), with T_2 giving the lowest value of pH (4.02). As the 12-day storage day elapsed, increases in pH for treatments T_0, T_1, and T_2 were noted, with T_0 recording the highest pH (4.35) on the last day. Treatments T_3 and T_4 were noted to decline, with the lowest pH by T_4 on day 12 (3.41). The mean pH between the various treatments followed the same trend, with T_0 recording the highest mean pH (4.19) and T_4

recording the lowest mean pH (3.54). So, T3 and T4 with 75 and 100% SOB formulation was found to be safer than all other treatments. The results showed conformity with McWhorter *et al.* (2021), who conducted research on acidification and extended storage at room temperature of mayonnaise reduce Salmonella Typhimurium virulence and viability; the present study results were also aligned with it in relation to the pH variability in treatments. Significant differences in pH values across treatments and storage days were observed by Hakimian et al. (2022) in their evaluation of the microbial and physicochemical properties of mayonnaise containing zinc oxide nanoparticles. Low pH enhances shelf stability by preventing spoilage, whereas higher pH compromises product safety. Therefore, an optimal pH is needed to balance acidity, flavor, and overall quality.

Table 3. Effect of storage days and treatment on pH in soybean mayonnaise				
Treatments	pH			
	0 Day	4 Days	8 Days	12 Days
T_0	4.02 ± 0.28^h	4.13 ± 0.28^f	4.56 ± 0.31^{cd}	4.35 ± 0.30^a
T_1	4.07 ± 0.28^h	4.11 ± 0.28^{fg}	4.50 ± 0.31^d	4.28 ± 0.29^{ab}
T_2	4.10 ± 0.028^h	4.08 ± 0.28^g	4.40 ± 0.30^e	4.10 ± 0.28^{bc}
T_3	4.15 ± 0.29^f	3.92 ± 0.27^i	3.39 ± 0.23^j	3.74 ± 0.26^k
T_4	4.30 ± 0.30^k	3.58 ± 0.25^l	3.28 ± 0.22^m	3.41 ± 0.23^n

3.3. Water Activity

Water activity is a significant parameter in determining the shelf life, stability, and microbial growth potential of mayonnaise. Mean squares for water activity of various treatments were statistically analyzed as depicted in table 4.7. The mean values of all five treatments during storage of 12 days and evaluation at every 4-day interval providing the temperature 25^0C. is shown in table 4.7. indicated that the results obtained were non-significant. In the current study, the effect of various treatments and storage durations on the water activity of soy-based mayonnaise was determined. Results found were in constant ranges from (0.90 to 0.95) on all days. The analysis of variance showed that the treatment or the storage duration had no highly significant effect on water activity all of which were comparatively low. These results showed that the water activity variations were not significantly influenced by the treatment or the storage duration *i.e.* the formulations were stable in the sense of their water activity levels during storage. The water activity values of each treatment during storage showed little variations. For the control treatment T_0,

water activity was comparatively constant for the entire 12-day storage duration, ranging from (0.90) on day 0 to (0.95) on day 12, with a gradual increase with time. This gradual increase may be due to the absorption of moisture from the atmosphere or the slight variations in the emulsion attributes with time. The T_1 and T_2 treatments also showed a similar trend, with slight increases in water activity with time, *i.e.*, their water content was stable on storage. Nonetheless, treatment T_3 had a remarkable fluctuation, where water activity dropped on day 4 (0.81), only to return on subsequent days. This could be due to formulation adjustments that resulted in temporary changes in the emulsion stability or product moisture content. T_4 had the same water activity for the duration of the storage period. Similar results were also found by (Huang *et al.,* 2022) who studied freeze-thaw stability of mayonnaise stabilized solely by a heated soy protein isolate. Formulations were stored at 25^0C, and water activity values were found to be from (0.92 to 0.96) across all the treatments over storage time. The average water activity readings for all treatments and storage durations had a slight rise.

Table 4. Effect of storage days and treatment on water activity in soybean mayonnaise				
Treatments	**Water Activity**			
	0 Day	**4 Days**	**8 Days**	**12 Days**
T_0	0.90 ± 0.06^a	0.91 ± 0.06^a	0.91 ± 0.06^a	0.92 ± 0.06^a
T_1	0.92 ± 0.06^a	0.92 ± 0.06^a	0.92 ± 0.06^a	0.92 ± 0.06^a
T_2	0.93 ± 0.06^a	0.93 ± 0.06^a	0.93 ± 0.06^a	0.94 ± 0.06^a
T_3	0.94 ± 0.06^a	0.81 ± 0.05^b	0.94 ± 0.06^a	0.95 ± 0.06^a
T_4	0.95 ± 0.06^a	0.95 ± 0.06^a	0.95 ± 0.06^a	0.95 ± 0.06^a

3.4. Texture Analysis

The texture analysis of soy-based mayonnaise formulations demonstrated statistically significant variations across treatments in terms of hardness, adhesiveness, cohesiveness, and springiness. These parameters are crucial in determining product

quality and consumer acceptability. Hardness decreased progressively with increasing soy oil body (SOB) concentration. The maximum hardness was observed in the control sample T_0 (9.06 N), while the lowest was in T_4 (4.49 N),

indicating that increased moisture from SOB contributed to softer consistency. Treatments T_1, T_2, and T_3 showed intermediate values 7.80 N, 6.59 N, and 6.59 N, respectively. The observed decline is in agreement with findings by Jing *et al.* (2021) and Mahapatra *et al.* (2019), where higher moisture levels due to soy peptides and amaranth protein incorporation resulted in reduced hardness. Adhesiveness also had an inverse, proportionate relationship with increasing SOB content. The highest adhesiveness was observed at T0 (84.05%) and the lowest at T4 (47.29%), with a gradual decrease across the intermediate treatments. This trend suggests that while SOB contributes to moisture retention, it may weaken the structural bonding essential for stickiness. Similar reductions were observed in formulations using herbal extracts and wheat gluten as egg yolk substitutes (Parzhanova *et al.*, 2021; Liu *et al.*, 2024). Cohesiveness values ranged from 0.260 in T_1 to 0.166 in T_4, indicating reduced internal bonding in the emulsions with higher SOB content. The decrease may be linked to reduced fat or emulsifier levels in treatments with SOB

substitution. This is consistent with findings by Wang *et al.* (2022) and Ibrahim *et al.* (2023), who demonstrated the weakening of internal structure due to lower fat or altered oil blends.

Springiness followed a similar declining trend, with T_0 having the highest value (0.26) and T_4 the lowest (0.17). Higher springiness in T_0 and T_1 indicated more elastic emulsions, while reduced springiness in T_4 reflected weakened emulsion structure due to lower emulsifier efficiency or imbalance in composition. These results align with studies of Heikal *et al.* (2023) and El-Waseif *et al.* (2022), which emphasized the role of soy proteins and stabilizers in preserving elastic texture and recovery. Overall, increasing SOB concentration led to significant declines in all texture parameters analyzed. While the use of SOB as a fat replacer improves nutritional composition, it may compromise textural integrity unless balanced with adequate stabilizers or emulsifiers. These findings underscore the importance of optimizing formulation components to achieve desirable sensory and structural properties in low-fat, plant-based mayonnaise.

Table 3.4. Means for the Texture of soybean mayonnaise (%)

Treatments	Hardness	Adhesiveness	Cohesiveness	Springiness
T_0	9.06 ± 0.63[a]	84.05 ± 5.88[a]	0.26 ± 0.01[a]	0.26 ± 0.01[a]
T_1	8.10 ± 0.56[ab]	75.12 ± 5.25[b]	0.24 ± 0.01[ab]	0.25 ± 0.01[a]
T_2	7.15 ± 0.50[b]	70.10 ± 4.90[bc]	0.23 ± 0.01[ab]	0.24 ± 0.01[a]
T_3	6.10 ± 0.42[c]	60.25 ± 4.21[c]	0.20 ± 0.01[b]	0.21 ± 0.01[b]
T_4	4.80 ± 0.33[d]	50.30 ± 3.52[d]	0.17 ± 0.01[c]	0.18 ± 0.01[b]

3.5. Color Analysis

The color of mayonnaise, typically ranging from off-white to creamy yellow when prepared with egg yolk, is known to shift towards a lighter, whitish appearance when egg yolk is replaced in the formulation. The primary objective of this study was to develop a low-fat mayonnaise by replacing egg yolk with soy oil body (SOB), which led to significant changes in the color parameters, particularly L*, a*, and b* values. The observed increase in whiteness of the samples was also reported to enhance visual appeal and consumer acceptability. Food technologists have clarified

that reduction in oil ratio contributes to a whiter hue, a desirable trait in fat-reduced emulsions. L* values, representing the lightness of the product, showed a statistically significant upward trend across treatments. This increase indicated formulation changes that enhanced the brightness of the mayonnaise samples. As shown in the data, L* values ranged from 94.73 in the control sample (T_0) to 98.78 in T_4. The control sample, containing 100% egg yolk, exhibited the lowest lightness, corresponding to its characteristic yellow appearance. A gradual rise in L* values was

observed with the progressive substitution of egg yolk with SOB: T_1 (95.63), T_2 (96.34), T_3 (97.43 ± 0.02), and T_4 (98.78), clearly indicating a consistent increase in brightness. These findings suggest that SOB, being inherently white, significantly influenced the lightness of the formulation. Similar result was reported by Heikal *et al.* (2023), who observed increased lightness in soy-based mayonnaise enriched with virgin palm oil, with values ranging from 74.50 to 79.43%. Additionally, Cerro *et al.* (2022) found comparable increases in L* values ranging from 80.24 to 92.06% in vegan dressings, supporting the trend that lighter appearance is generally more attractive to consumers. The a* values, which measure the red to green spectrum of color, revealed a steady and statistically significant decline across treatments. A reduction in a* value denotes a shift away from redness towards a greener or more neutral appearance. The control treatment (T_0) showed the highest a* value (1.56), indicating the strongest red tone. With increased substitution of egg yolk with SOB, the redness diminished: T_1 (1.27), T_2 (0.64), T_3 (0.45), and T_4 (0.25). This gradual reduction can be attributed to the removal of egg yolk, which imparts yellow-red tones, and its replacement with the pale SOB emulsion. These findings are aligned with Yang *et al.* (2020), who observed a decline in a* values (1.35 to 0.62%) while developing low-fat emulsions using sodium alginate and konjac glucomannan. Furthermore, Ihsan *et al.* (2021) reported a decreasing trend in a* values (1.15 to 0.46%) in mayonnaise made with non-traditional

ingredients, supporting the conclusion that removing egg yolk contributes to reduced redness. Regarding the b* values, which quantify yellowness (positive values) or blueness (negative values), significant reductions were recorded across treatments. The control sample (T_0) had the highest b* value (28.28), indicating a strong yellow color due to the presence of egg yolk. As egg yolk was replaced with SOB, the yellowness of the samples progressively diminished: T_1 (25.43), T_2 (21.50), T_3 (18.60), and T_4 (14.29). These results confirmed that T_4, which had 100% SOB substitution, was the least yellow among all treatments. The decline in b* values demonstrate the effect of ingredient modifications on the visual appearance of mayonnaise, likely influenced by changes in emulsifier type and oil content. These findings are consistent with those of Filho *et al.* (2023), who reported b* values ranging from 20.76 to 17.65% in vegan mayonnaise made using aquafaba. The lighter yellow tone was attributed to differences in emulsifier selection and blending processes. Similar observations were made by Dukariya *et al.* (2020) in a study on mayonnaise formulated with roselle seed oil, where b* values ranged from 25.47 to 14.48%.

In summary, the substitution of egg yolk with soy oil body led to a noticeable enhancement in the visual characteristics of mayonnaise, including increased lightness and decreased redness and yellowness. These color modifications not only contributed to a more appealing product but also aligned with consumer expectations for low-fat and plant-based alternatives.

Table 5. Effect of varying concentration of soybean oil in mayonnaise (%)			
Treatments	**L***	**a***	**b***
T_0	94.73±6.63[e]	1.56±0.10[a]	28.28±1.97[a]
T_1	95.63±6.69[d]	1.27±0.08[b]	25.43±1.78[b]
T_2	96.34±6.74[c]	0.64±0.04[c]	21.50±1.50[c]
T_3	97.43±6.82[b]	0.45±0.03[d]	18.60±1.30[d]
T_4	98.78±6.91[a]	0.25±0.01[e]	14.29±1.00[e]

3.6. Total bacterial count (TBC)

TBC in mayonnaise is the number of bacteria in a particular sample. Mean squares for total bacterial count of various treatments were statistically analyzed as depicted in table 4.9. The mean values of all five treatments during storage of 12 days and evaluation at every 4-day interval is shown in table 4.5. Results found for CFU/g were in the range (8.94 to 7.72) on 0 day and (30.79 to 24.44) on 12th day. The data showed a clear decreasing trend in the TBC across treatments and storage durations. At 0 days, the bacterial count was lowest for treatment T_4 (7.72) and highest for T_0 (8.94). Over time, the TBC increased for all treatments, with the highest counts recorded at 12 days, ranging from T_4 (18.647) to T_0 (30.79). The mean TBC across storage durations followed a similar trend, with values increasing from (8.43) at 0 days to (24.44) at 12 days. The results indicated that treatment T_4 at (0% egg yolk) consistently exhibited the lowest TBC across all storage durations, suggesting its superior microbial stability compared to other treatments. Because eggs are from highly perishable food commodities so when completely eliminated, mayonnaise safety got prolonged. This could be attributed to specific formulation changes, such as the inclusion of antimicrobial agents, preservatives or adjustments that inhibited bacterial growth. In contrast, T_0 had the highest TBC, reflecting the absence of such modifications.

Similar trends were described in the research conducted on essential oil and its Nano emulsion on oxidative stability and microbial growth in mayonnaise during storage by Moradi *et al.* (2023). Time intervals of storage were 10 days with total of 4 to 25 days. CFU/g on 4th day was (4.87 to 3.57), on day 14th it was (6.44 to 6.03) and on 25th day the range was recorded as (8.73 to 7.49). El Wesif *et al.* (2022) depicted similar results when developed mayonnaise enriched with flaxseed oil omega-3 fatty acids content, sensory quality and stability during the storage. Storage duration was also 12 days and evaluation of samples was done at 25^0C. Study trialed different blending ratio of flaxseed oil and soybean oil and found TBC decline in similar pattern as highest for T_0 and lowest for T_4 (7.68 to 5.89) on 0 day and on 12 day (34.25 to 28.67). So, the T_0 (100% egg yolk) is less safe in microbial terms as compared to T_4 (0% egg yolk).
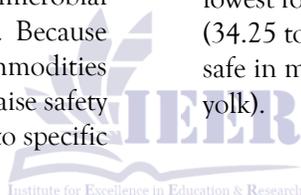
Table 3.6.

Table 6. Effect of treatments and storage on total bacterial count of soybean mayonnaise				
Treatments	**TBC (CFU/g)**			
	0 day	**4 days**	**8 days**	**12 days**
T_0	8.7±0.3[a]	10±2.1[b]	20±2.2[c]	20±2.4[cd]
T_1	8.5±0.2[a]	10±3.1[b]	20±2.5[c]	20±2.2[cd]
T_2	8.3±0.4[a]	10±1.9[b]	20±2.0[c]	20±2.3[cd]
T_3	8.0±0.3[ab]	9.5±1.2[b]	10±1.1[d]	10±1.5[b]
T_4	7.8±0.2[b]	9.0±1.1[b]	10±1.3[d]	10±1.6[b]

3.7. Sensory Analysis

Sensory evaluation plays a pivotal role in assessing the quality and consumer acceptance of food products, particularly in the development of innovative formulations such as soy-based mayonnaise. Using a 9-point hedonic scale, trained panelists evaluated various sensory attributes such as color, texture, flavor, mouthfeel, spreadability, and overall acceptability across treatments with incremental substitution of egg yolk by soy oil body (SOB). Color scores varied significantly among treatments (Table 4.4), ranging from 5.21 to 8.13. The highest score was observed in T_4 (8.13), containing 100% SOB, followed by T_3 (7.42), suggesting that panelists found SOB-substituted samples visually appealing,

though not intensely colored. The declining color intensity with higher SOB content may be due to differences in pigment and emulsion properties compared to egg yolk, aligning with findings of Gaikwad *et al.* (2020) and Parzhanova *et al.* (2021), who reported similar effects of formulation modifications on color perception in mayonnaise. Texture, a critical determinant of product quality, also exhibited significant variation. Scores ranged from 8.32 (T_0, control) to 5.22 (T_4), indicating a clear reduction in firmness as the egg yolk content decreased. Texture remained within acceptable limits up to T_2, beyond which a noticeable decline was observed. This trend confirms previous findings of Wang *et al.* (2022) and He *et al.* (2021), which showed that changes in emulsifier or stabilizer composition can weaken structural integrity in low-fat emulsions. The results underscore the importance of optimizing fat and emulsifier ratios to preserve desired textural properties, as emphasized by Kadian *et al.* (2021). Flavor scores demonstrated a significant downward trend from 8.35 (T_0) to 6.84 (T_4). While T_1 (7.52) and T_2 (7.32) remained within acceptable sensory limits, higher levels of SOB substitution negatively affected flavor, particularly in T_3 and T_4. These results corroborate earlier studies of Mangiapelo *et al.* (2023) and Onwuzuruike *et al.* (2022), who reported flavor deterioration in soy-based emulsions due to the presence of beany notes and reduced lipid-based aroma compounds. Mouthfeel, influenced by the product's creaminess and smoothness, followed a similar pattern, with scores ranging from 8.44 (T_0) to 6.41 (T_4).

Reduced fat content and altered emulsification likely contributed to the diminished mouthfeel. However, T_1 (7.67) and T_2 (7.49) remained acceptable, indicating that partial substitution can maintain favorable sensory texture. These findings are aligned with Mohammadai *et al.* (2024) and Jing *et al.* (2023), who noted decreased creaminess in low-fat mayonnaise formulations, reinforcing the relationship between fat content and perceived mouthfeel. Spreadability improved notably with SOB incorporation. The score increased from 4.33 (T_0) to 7.77 (T_4), reflecting enhanced emulsion stability and oil-moisture interaction in SOB-substituted formulations. A consistent upward trend from T_1 (5.29) through T_3 (6.72) suggests better application properties in higher SOB samples. Similar improvements were reported by Kadian *et al.* (2021) and Lee *et al.* (2024), emphasizing the role of processing and formulation in improving spreadability in vegan mayonnaise.

Overall acceptability, integrating all sensory parameters, revealed a gradual decline with increasing SOB substitution. The control sample T_0 achieved the highest acceptance score (8.73), while T_1 (8.51) and T_2 (7.32) remained within the acceptable range but T_3 (6.46) and T_4 (6.12), get lower score, not rejected outright, suggesting partial substitution is more favorable for consumer satisfaction. These findings align with observations of Cai *et al.* (2021) and Chen *et al.* (2022), who noted reduced overall liking in soy-based products due to formulation-related sensory shifts.

Table 7. Sensory analysis of soybean mayonnaise (%)						
Treatments	Color	Texture	Flavor	Mouth feel	Spread ability	Overall Acceptability
T_0	5.21±0.36[e]	8.32±0.58[a]	8.35±0.58[a]	8.44±0.59[a]	4.33±0.30[e]	8.73±0.61[a]
T_1	6.42±0.45[d]	7.45±0.52[b]	7.52±0.52[b]	7.67±0.53[b]	5.29±0.37[d]	8.51±0.59[b]
T_2	7.24±0.50[c]	6.73±00.47[c]	7.32±0.51[c]	7.49±0.52[c]	6.48±0.45[c]	7.32±0.51[c]
T_3	7.42±0.51[b]	5.75±0.40[d]	7.24±0.50[d]	7.31±0.51[d]	6.72±0.47[b]	6.46±0.45[d]
T_4	8.13±0.56[a]	5.22±0.36[e]	6.84±0.47[e]	6.41±0.44[e]	7.77±0.54[a]	6.12±0.42[e]

4. CONCLUSION

Commercial mayonnaise is a widely consumed condiment primarily composed of oil, egg yolk, vinegar, and salt. It is categorized as an oil-in-water emulsion with a fat content ranging from 65% to 80%, where egg yolk serves as the principal emulsifier, stabilizing the mixture and imparting a smooth texture. Consumers are looking for healthier solutions that are lower in fat without sacrificing flavor or texture. In order to satisfy vegan and allergy-conscious consumers, egg yolks must also be substituted; hence, soy protein's emulsifying qualities make it a perfect option for formulation. Egg yolk was substituted with SOBs at varied percentages to create five distinct formulations: 25%, 50%, 75%, and 100%. In all these treatments the T_4 is good in comparison to the T_0 due to its nutritional profile and sensory acceptability. The study concluded that, considering all aspects, soybean-based mayonnaise is a good substitute for traditional mayonnaise, providing a plant-based, lower-fat alternative without appreciably sacrificing stability or flavor. The results back up the possibility of commercialization and fit in with the growing movement toward more sustainably produced and healthier food options.

Conflict of interest

The authors declare that they have no known conflict of interest.

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