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Development of Soy Milk Vegan Choco-Chip Muffins

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ABSTRACT

This study explores the development of soy milk-based chocolate chip muffins, aimed at creating a nutritionally superior, plant-based bakery product for health-conscious consumers. Special grain flour (Fenugreek and oats) was used to enrich dietary fiber, enhance nutritional value, and contribute to a balanced texture. Dairy-free soy milk was utilized to enhance protein content and moisture retention, while date paste served as a natural sweetener, reducing reliance on refined sugars. Vegetable oil (combination of soybean, rice bran, and flaxseed oils) was employed to improve lipid stability and mouthfeel.

Sensory evaluation was conducted using a semi-trained panel to assess attributes such as appearance, texture, flavor, and overall acceptability. Microbial safety was assessed through total plate count, coliform count, yeast and mold enumeration, and detection of Staphylococcus aureus, Salmonella spp., and Escherichia coli. The muffins underwent comprehensive nutritional analysis, including determination of moisture content, pH, protein, fat, ash, dietary fiber, and carbohydrate levels.

A shelf-life study was performed under ambient conditions to monitor changes in microbial load, sensory quality, and physicochemical parameters over period of 7 days. The findings support the feasibility of incorporating functional ingredients into vegan bakery formulations while maintaining safety, palatability, and nutritional integrity.

Keywords: vegan muffins, soy milk, fenugreek and oat flour, date paste, vegetable oil, choco-chips, sensory evaluation, microbial safety, nutritional analysis, shelf-life study.

1. INTRODUCTION

1.1 Background of the study

Muffins are single-serving baked products traditionally made from flour, sugar, fat, eggs, and milk, leavened with baking powder or baking soda rather than yeast (Samokhvalova et al., 2020). Their soft texture, sweet flavor, and convenience have made them a staple in both commercial and home baking. A desirable muffin exhibits a golden-brown crust, moist crumb, uniform cell structure, and pleasant aroma (Baixauli et al., 2008; Karaoglu and Kotancilar, 2009).

1.2 Limitations of Conventional muffins

However, conventional muffins often contain animal-derived ingredients and refined wheat flour, which may not align with the dietary preferences of health-conscious or vegan consumers (Man et al., 2014).

1.3 Trends in Plant- Based Bakery products

With increasing awareness of food-related health concerns and ethical considerations, the global bakery market has witnessed a significant shift toward plant-based, allergen-free, and functional products (Shih, 2020). Recent trends emphasize the use of natural ingredients, whole grains, healthy fats, sugar substitutes, and dairy-free alternatives to meet the evolving demands of consumers (Cross, 2007). Vegan muffins, in particular, have gained popularity for their compatibility with plant-based diets and their potential to deliver both indulgence and nutrition (Yadav et al., 2022).



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1.4 Functional Ingredients in vegan muffins

Recent formulations have also focused on optimizing the structure and texture of vegan muffins through ingredient functionality. Parameters such as crumb porosity, moisture content, and texture are influenced by the type of flour, fat, and liquid used (Baik et al., 2000; Martínez-Cervera et al., 2012). The absence of eggs and dairy requires careful formulation to replicate their functional roles in structure and emulsification (Shukla et al., 2024). Shukla et al. (2024) reviewed the incorporation of fruit, vegetable, and grain-based ingredients in muffins, noting significant improvements in antioxidant content and overall nutritional value.

1.5 Role of soy milk in vegan baking

Soymilk, derived from soybeans, is a widely accepted plant-based alternative to dairy milk, offering high-quality protein, essential amino acids, and bioactive compounds (Vishali and Agarwal, 2023). Its use in vegan baking contributes to improved moisture retention, texture, and nutritional value. Vishali and Agarwal (2023) formulated vegan muffins using soymilk and lotus seed flour, demonstrating enhanced microbial stability and extended shelf life compared to conventional formulations. Similarly, Yadav et al. (2022) emphasized the economic and processing aspects of muffin production, advocating for the use of byproducts such as seed flours and peel powders to reduce waste and improve nutritional density.

1.6 Chocolate Chips and Sensory Appeal

Chocolate chips, a popular inclusion in muffins, enhance sensory appeal through their rich flavor and melt-in-mouth texture. When choco chips from vegan base offer indulgence without compromising dietary restrictions (Grasso et al., 2020). The development of vegan muffins using chocolate chips and plant-based ingredients aligns with consumer demand for clean-label, nutrient-rich bakery products.

1.7 Alternative Flours and Nutritional Enhancement

Grasso et al. (2020) explored the use of aquafaba and chickpea flour in vegan baked goods, demonstrating improvements in emulsification and sensory acceptance. In addition, alternative flours like fenugreek and oats are increasingly incorporated into vegan formulations to improve fiber content, mineral density, and antioxidant activity. Fenugreek flour is notable for its protein and iron content, while oat flour contributes β -glucan, unsaturated lipids, and a mild flavor profile that complements sweet bakery items (Negu et al., 2020; Jain et al., 2024).

1.8 Peanut Butter and Flaxseed Paste as Functional Additions

Peanut butter, a nutrient-dense plant-based ingredient, contributes significantly to vegan baking by enhancing texture, flavor, and nutritional value. Rich in monounsaturated fats, protein, and micronutrients such as magnesium and vitamin E, natural peanut butter supports satiety, heart health, and energy metabolism (AS-IT-IS Nutrition, 2024; Marcene, 2025). Its creamy consistency aids in moisture retention and emulsion stability, making it a valuable fat replacer in clean-label bakery formulations (Grasso et al., 2020).



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Similarly, flaxseed paste—prepared by hydrating ground flaxseeds—functions as an effective egg substitute due to its mucilage content and emulsifying properties. It improves batter viscosity and contributes omega-3 fatty acids, lignans, and dietary fiber, which support cardiovascular and digestive health (Sharma et al., 2024; Talwar et al., 2025).

1.9 Vegetable Oils and Lipid Functionality

Vegetable oils such as soybean, rice bran, and flaxseed oil are widely used in vegan baking for their lipid structure and oxidative stability. Soybean oil offers polyunsaturated fats and vitamin K, rice bran oil provides oryzanol and antioxidants, while flaxseed oil is a potent source of alpha-linolenic acid, collectively enhancing crumb softness and shelf life (Yadav et al., 2022; Fortune Xpert Oil, 2025.

1.10 Clean-Label and Sustainability Considerations

Furthermore, the rise of veganism and flexitarian diets has encouraged food scientists to explore novel ingredients that enhance both health and sustainability. The use of minimally processed plant-based components supports clean-label product development and aligns with consumer expectations for transparency and ethical sourcing (Dwivedi et al., 2024). Muffins made with soymilk and vegan chocolate chips not only cater to dietary restrictions but also contribute to the growing movement toward environmentally conscious food systems.

1.11 Sensory-Guided Formulation Strategy

The food product developed needed a sensory-guided formulation strategy, wherein multiple trials were conducted using varying levels of functional ingredients. Sensory attributes such as taste, texture, aroma, and appearance were used to identify the most acceptable formulation. The selected trial was then subjected to nutritional analysis, microbial testing, and shelf-life evaluation to validate its overall quality

1.12 Objectives of the Study

Vegan muffins are gaining importance by consumers because of increasing awareness of food-related health concerns and ethical considerations, the global bakery market has witnessed a significant shift toward plant-based, allergen-free, and functional products. This thesis focuses on the formulation and evaluation of a **soymilk-based vegan choco chip muffin**, aiming to optimize its sensory attributes, nutritional profile, microbial stability, and consumer acceptability.

keeping in this view, our objectives are:

- 1. To develop a dairy-free, vegan chocolate chip muffin using soymilk, dates, fenugreek, and oats.
- 2. To analyze its nutritional profile.
- 3. To evaluate sensory attributes such as taste, texture, appearance, and aroma.
- 4. To assess microbial safety and shelf-life under ambient storage.

2. LITERATURE REVIEW

2.1 Overview of Muffin Reformulation

The bakery industry has seen a marked shift toward health-oriented and plant-based innovations. Muffins, once considered indulgent snacks, are now reformulated to meet nutritional, ethical, and



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lifestyle demands (Singh et al., 2020). Consumers increasingly seek products that support satiety, digestive health, and glycemic control. Reformulations include whole grains, natural sweeteners, and dairy alternatives. These trends reflect broader movements toward clean-label and functional foods. Muffins have become a canvas for nutritional enrichment and dietary inclusivity.

2.2 Evolving Trends in Muffin Formulation

Market projections indicate strong growth in the vegan baked goods segment. Kumar and Rao (2021) estimate the market will exceed USD 1.5 billion by 2027. This growth is driven by rising lactose intolerance, ethical eating practices, and demand for allergen-free products. Muffins are particularly adaptable due to their simple structure and customizable ingredients. Their portability and portion control make them ideal for health-conscious consumers. As a result, vegan muffins are gaining traction in both retail and foodservice sectors.

In India, cultural preferences further influence muffin reformulation. Eggless and dairy-free baked goods are favored during religious observances and fasting periods (Sharma & Kulkarni, 2020). Urban consumers increasingly prefer products labeled "vegan," "gluten-free," or "no added sugar." These preferences encourage manufacturers to explore novel ingredient combinations. The Indian market presents unique opportunities for regionally adapted vegan muffins. Understanding local taste profiles is essential for successful product development.

2.3 Technological Advancements in Muffin Development

Technological advancements also shape muffin trends. Ingredient functionality is enhanced through emulsifiers, hydrocolloids, and fermentation techniques (Patel et al., 2019). These tools help replicate the texture and structure of conventional muffins. Innovations in packaging and shelf-life extension support broader distribution. As consumer expectations evolve, muffins must deliver both health benefits and sensory satisfaction. This dual demand drives continuous research and formulation refinement.

2.4 Sustainability and Clean- Label Innovation

Sustainability is another key driver of muffin innovation. Plant-based ingredients generally require fewer resources and produce lower emissions (Ahmed et al., 2019). Using agro- industrial byproducts like date paste promotes waste reduction. Vegan muffins align with environmental goals and ethical sourcing standards. Packaging transparency and ingredient traceability further enhance consumer trust. These evolving trends position muffins as a responsible and desirable food choice.

Here's a detailed synthesis of the **nutritional and physicochemical properties of muffins**, based on recent peer-reviewed studies and your research context, Charishma. This section is ideal for your **Review of Literature** chapter and written in APA style.

2.5 Nutritional and Physicochemical Properties of Muffins

Muffins are widely consumed bakery products known for their soft texture, appealing flavor, and versatility in formulation. Their nutritional and physicochemical characteristics are influenced by ingredient composition, baking conditions, and fortification strategies.



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2.5.1 Nutritional Composition

Muffins typically contain carbohydrates, fats, proteins, dietary fiber, and minerals. The nutritional profile can be significantly enhanced through the incorporation of plant-based ingredients. Du et al. (2025) investigated the use of hempseed protein isolate (HPI) in vegan muffins and reported a substantial increase in protein content from 9.61% in control samples to 19.40% in muffins fortified with 30% HPI. Additionally, antioxidant activity improved due to the presence of bioactive peptides and phenolic compounds, with ABTS radical scavenging values rising from 32.66% to 46.28%.

Shukla et al. (2024) reviewed the incorporation of fruit, vegetable, and grain-based ingredients in muffins and found that such additions elevated the levels of polyphenols, vitamins, and minerals, while also improving dietary fiber content. These nutrient-dense formulations contribute to the development of functional muffins with enhanced health benefits.

Moldovan et al. (2023) analyzed gluten-free muffins made from almond and coconut flour and reported moisture content ranging from 34.8% to 37.5%, along with polyphenol levels between 47.15 and 128.4 mg GAE/g, indicating strong antioxidant potential.

2.5.2 Physicochemical Characteristics

Physicochemical properties such as moisture, texture, porosity, elasticity, and baking loss are critical for consumer acceptability and shelf stability. Du et al. (2025) observed that HPI fortification reduced baking loss and improved crumb softness, with hardness decreasing from

179.72 g/cm² to 137.73 g/cm² at higher substitution levels. Rheological analysis revealed increased batter viscosity and shear-thinning behavior, indicating better structural integrity and aeration.

Moldovan et al. (2023) reported porosity values between 68.2% and 72.7%, and elasticity ranging from 48.58% to 54.9%, in almond—coconut flour muffins. These parameters directly influenced mouthfeel and texture, with higher elasticity contributing to a springier crumb.

Shukla et al. (2024) emphasized that the use of fruit and vegetable-based ingredients not only improved nutritional value but also enhanced physicochemical stability, including moisture retention and antioxidant activity. Such formulations are particularly beneficial in tropical climates where shelf-life is a concern.

2.6 Classification of Muffins

Muffins, once considered indulgent sweet treats, have evolved into nutritionally diverse baked products. Their classification can be systematically organized based on **ingredient composition**, **dietary preference**, and **functional purpose**. This framework supports targeted formulation strategies, aligns with consumer health trends, and facilitates comparative analysis in product development (Yadav et al., 2022; Shukla et al., 2024; Swarup et al., 2024; Vishali & Agarwal, 2023).

2.6.1 Based on Ingredient Composition

This category focuses on the primary raw materials used to enhance the nutritional profile of muffins.



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2.6.1.1 Cereal-Based Muffins

Incorporate grains such as wheat, oats, barley, and millet. These muffins are designed to improve dietary fiber, protein, and mineral content. Millet-based formulations, for instance, reduce gluten dependency and increase bioavailability of nutrients (Swarup et al., 2024). Oats and bran also contribute to satiety and glycemic control.

2.6.1.2 Fruit-Enriched Muffins

Include fresh, dried, or pureed fruits like banana, berries, apples, and dates. Fruits enhance antioxidant capacity, natural sweetness, and moisture retention. Their inclusion also improves consumer appeal due to flavor diversity and visual contrast (Shukla et al., 2024).

2.6.1.3 Vegetable-Enriched Muffins

Use vegetables such as carrot, beetroot, spinach, and pumpkin to boost vitamin A, iron, and fiber content. These muffins are often targeted toward children and health-conscious consumers. The pigments from vegetables also contribute to visual appeal and perceived freshness (Shukla et al., 2024).

2.6.2 Based on Dietary Preference

This classification addresses consumer choices influenced by ethical, cultural, or health-related dietary restrictions.

2.6.2.1 Vegan Muffins

Exclude all animal-derived ingredients. Milk is replaced with plant-based alternatives such as soymilk or almond milk, while eggs are substituted with flaxseed gel, aquafaba, or banana puree. Vishali and Agarwal (2023) demonstrated that lotus seed flour combined with soymilk produced vegan muffins with improved microbial stability and shelf life, making them suitable for extended storage and clean-label marketing.

2.6.3 Based on Functional Purpose

This category reflects the intended health benefits or convenience features of the muffin.

2.6.3.1 Conventional Muffins

Typically made with refined flour, sugar, eggs, and butter. These muffins are high in carbohydrates and fats, with limited fiber and micronutrients. They are widely consumed for their soft texture and sweet flavor, but are nutritionally less dense (Yadav et al., 2022).

2.6.3.2 Functional Muffins

Designed to deliver specific health benefits such as high-fiber, low-sugar, antioxidant-rich, or gluten-free properties. Ingredients like fenugreek seed powder, fruit peels, bran, and legume flours are used to enhance nutritional density and reduce food waste. These muffins support dietary interventions for diabetes, cardiovascular health, and digestive wellness (Shukla et al., 2024).



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2.6.3.3 Quick Bread Muffins (American Style)

Leavened using chemical agents like baking powder or baking soda instead of yeast. This results in a soft, cake-like texture and rapid preparation. These muffins are typically consumed as breakfast items or snacks and are sold in individual portions for convenience (Swarup et al., 2024).

2.7 Functional Ingredients in Vegan Muffins

Functional ingredients are central to the nutritional and sensory success of vegan muffins. Soy milk, dates, flaxseeds, and oats each contribute distinct health benefits and baking functionalities. Soy milk provides complete protein and emulsification, improving crumb texture (Francisco et al., 2018). Dates offer natural sweetness and fiber, reducing glycemic load (Ahmed & Thomas, 2019). Flaxseeds act as egg replacers and support heart health (Thomas & Reddy, 2020). Oats enhance mouthfeel and satiety through beta-glucans (Dotaniya et al., 2016).

Soy milk is particularly valuable in vegan baking. It contains essential amino acids and calcium, supporting bone and muscle health. Its emulsifying properties help retain moisture and improve structure. Compared to almond or rice milk, soy milk offers superior volume and texture (Ghosh & Banerjee, 2021). It also contributes to Maillard browning, enhancing flavor and appearance. These attributes make soy milk a preferred dairy alternative in muffin formulations.

Dates serve as a multifunctional ingredient in vegan muffins. Rich in potassium, antioxidants, and fiber, they improve nutritional value. Their natural sweetness reduces the need for refined sugar. Ahmed and Thomas (2019) found that date paste lowered glycemic index and improved consumer acceptability. Dates also enhance moisture and shelf-life. Their versatility supports both health and sensory goals in muffin development.

Flaxseeds offer binding and moisture retention, replacing eggs in vegan baking. They contain omega-3 fatty acids and lignans, which support cardiovascular and digestive health (Thomas & Reddy, 2020). Flaxseed gel mimics the emulsifying function of eggs, maintaining crumb integrity. Jain and Thomas (2020) reported that flaxseed contributed more to structure than chia seeds. Its neutral flavor allows for flexible integration with other ingredients.

Oats contribute to texture, nutrition, and satiety in muffins. Beta-glucans in oats help regulate cholesterol and blood sugar levels (Dotaniya et al., 2016). Oat flour improves mouthfeel and crumb softness. Gouli, Patel, and Machado (2014) found that oats enhanced sensory scores without compromising structure. Their compatibility with soy milk and dates supports cohesive formulations. Oats are a staple in health-focused muffin recipes.

2.8 Ingredient Selection and Functional Justification

The selection of soy milk as the primary dairy alternative was based on its superior nutritional and functional properties. With approximately 3.3 g of protein per 100 ml, soy milk closely matches the protein content of cow's milk and supports emulsification, Maillard browning, and crumb structure (Francisco et al., 2018). Compared to almond, oat, rice, and coconut milks, soy milk offers a neutral flavor, baking stability, and affordability. Its production rate in India also supports scalability, making it ideal for vegan bakery applications (Kumar & Rao, 2021).



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Dates paste was chosen as a natural sweetener and moisturizer due to its fiber, potassium, and antioxidant content. It aligns with clean-label and low-glycemic goals while enhancing batter viscosity and crumb softness (Ahmed & Thomas, 2019). Alternatives like jaggery, maple syrup, and coconut sugar were rejected due to overpowering flavor, thin consistency, or poor moisture retention. Dates paste also contributes to shelf-life stability and consumer acceptability in health-focused baked goods (Dotaniya et al., 2016).

Flaxseed paste was selected as an egg replacer for its mucilage-forming ability when soaked, which mimics egg binding. It also provides omega-3 fatty acids (ALA), protein, and dietary fiber, supporting structure and moisture retention (Thomas & Reddy, 2020). Chia seed gel and banana puree were rejected due to gritty texture and flavor interference. Commercial egg replacers were avoided to maintain a clean-label formulation. Flaxseed paste offered the most natural and functional solution for vegan binding.

Oats and fenugreek flour were incorporated for their fiber content, digestive benefits, and mild flavor profile. Oats are rich in beta-glucans and complex carbohydrates, while fenugreek contributes micronutrients and metabolic support (Mishra, Sharma, & Dotaniya, 2016). Whole wheat, barley, and quinoa flours were considered but rejected due to density, strong cereal notes, or bitterness. The chosen combination improved mouthfeel, satiety, and local ingredient relevance, aligning with nutritional and sensory goals.

A multi-source oil blend of rice bran, flaxseed, and soybean oils was used to enhance moisture, texture, and lipid quality. This blend provides omega-3 and monounsaturated fats while avoiding overpowering flavors (Grasso, Liu, & Methven, 2020). Coconut oil, butter, and olive oil were excluded due to mouthfeel issues, saturated fat content, or strong taste. The selected oils ensured a soft crumb and neutral flavor, contributing to both health and sensory appeal in the final product.

Cocoa powder was chosen for its rich chocolate flavor, dark color, and antioxidant polyphenols. It also helped mask earthy tones from flaxseed and fenugreek, improving overall taste (Shih, 2020). Carob powder and unsweetened chocolate were rejected due to sweetness imbalance

and fat content. Cocoa powder provided the ideal balance of flavor and functional phytochemicals, enhancing both indulgence and nutritional value.

Vegan chocolate chips were added to improve texture, sweetness, and visual appeal. They increased consumer acceptability, especially among younger or non-vegan audiences (Rani, Ahmed, & Thomas, 2021). Alternatives like dried fruits or omitting inclusions were rejected due to moisture imbalance and reduced novelty. Chocolate chips maintained the indulgent character of muffins while adhering to plant-based standards.

Apple cider vinegar and baking soda were used as a leavening system to replicate the aeration typically provided by eggs. This combination produces carbon dioxide, lifting the batter and creating a light texture (Jaronski, 2023). Lemon juice and baking powder were considered but found less consistent or weaker in rise. ACV and soda offered reliable performance and a neutral flavor finish, supporting clean-label and vegan formulation goals.

Vanilla extract was selected to enhance aroma and mask off-notes from soy and flax. It contributed to sweet perception without adding sugar and maintained the classic muffin flavor profile (Shih, 2020).



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Almond and orange essences were rejected due to flavor interference, and omitting flavoring led to blandness. Vanilla extract ensured a balanced sensory experience and complemented cocoa and chocolate chips effectively.

Each ingredient was selected not in isolation but as part of a synergistic system that delivers a vegan muffin with high nutritional value, excellent texture, and strong sensory acceptance. The replacements and combinations were chosen after evaluating alternatives in terms of functionality, health benefits, flavor, and consumer appeal. This careful formulation resulted in a vegan product that mirrors conventional muffin quality while maintaining clean-label and plant-based values.

2.9 Role of Soy Milk in Vegan Baking Applications

Soy milk plays a critical role in vegan baking due to its nutritional and functional properties. It provides complete plant-based protein, essential for structure and satiety (Francisco et al., 2018). Its emulsifying capacity helps retain moisture and improve crumb texture. Compared to other plant-based milks, soy milk offers superior performance in baked goods. Ghosh and Banerjee (2021) noted its effectiveness in sponge cakes and muffins. These attributes make soy milk a cornerstone of vegan muffin formulations. Soy milk interacts well with leavening agents, contributing to volume and aeration. Machado et al. (2010) emphasized its role in achieving desirable rise and softness. Its amino acid profile supports Maillard browning, enhancing crust color and flavor. Jaronski (2023) found that soy milk produced muffins with better texture than almond or rice milk. These interactions are essential for replicating conventional muffin characteristics. Soy milk's versatility supports diverse recipe adaptations.

Flavor neutrality is another advantage of soy milk. It allows for seamless integration with sweeteners, spices, and inclusions like chocolate chips. This flexibility supports sensory appeal across consumer segments. Rani et al. (2021) reported high acceptability for soy milk-based muffins with chocolate chips. Its mild taste avoids overpowering other ingredients. This makes soy milk suitable for both sweet and savory muffin variations.

Soy milk also contributes to shelf-life stability. Its moisture-retaining properties reduce staling and improve freshness. Dotaniya et al. (2016) noted that soy milk helped maintain softness over time. When combined with oil blends and fiber-rich ingredients, it supports extended shelf-life. These benefits are crucial for commercial distribution. Soy milk enhances both product quality and consumer satisfaction.

From a sustainability perspective, soy milk aligns with environmental and ethical goals. It requires fewer resources than dairy and produces lower emissions (Ahmed et al., 2019). Its plant-based origin supports vegan and vegetarian diets. Soy milk is widely available and cost- effective, making it accessible for large-scale production. These factors reinforce its role in responsible food innovation.

2.10 Sensory and Nutritional Impact of Ingredient Substitution

Substituting animal-based ingredients with plant-based alternatives affects texture, flavor, and nutrition. Flaxseed gel replicates egg binding, maintaining crumb integrity and moisture (Krishna, 2005). Dates replace refined sugar, lowering glycemic index and enhancing fiber content (Ahmed et al., 2019). Oats improve chewability and softness, while oil blends enhance mouthfeel. These substitutions must be balanced to preserve sensory appeal. Proper formulation ensures consumer satisfaction.



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Texture is a key consideration in vegan muffin development. Eggs contribute aeration and structure, which are difficult to replicate. Flaxseed and chia seeds offer partial solutions but may result in denser crumb. Jain and Thomas (2020) emphasized the need for hydration and

leavening adjustments. Oat flour can improve softness but may require additional moisture. Texture optimization is essential for product acceptance.

Flavor balancing is also critical. Strong-tasting ingredients like flaxseed or fenugreek may require masking. Rani et al. (2021) recommended using vanilla, cocoa, or chocolate chips to enhance palatability. Dates provide natural sweetness but must be proportioned carefully. Excessive use may dominate the flavor profile. Sensory trials help identify ideal ingredient ratios. Flavor harmony supports repeat purchases.

Nutritional enhancement is a major benefit of substitution. Plant-based ingredients offer fiber, antioxidants, and healthy fats. Soy milk provides protein and calcium, supporting bone health (Francisco et al., 2018). Dates and oats contribute to digestive wellness and satiety. These benefits align with consumer health goals. Substitution must balance nutrition with sensory quality.

Consumer perception influences substitution success. Many buyers associate vegan products with inferior taste or texture. Sensory evaluations and marketing strategies can address these concerns. Transparent labeling and health claims build trust. Ahmed et al. (2019) found that informed consumers were more receptive to vegan muffins. Education and sampling programs support market expansion.

2.11 Sensory Evaluations in Previous Research

Sensory evaluation plays a critical role in assessing the acceptability of vegan muffin formulations. Studies often employ hedonic scaling, preference mapping, and descriptive analysis to gauge consumer responses. Rani et al. (2021) conducted sensory trials on soy—banana muffins, reporting high scores for taste, texture, and overall appeal. Their use of familiar flavors and moist ingredients contributed to positive feedback. These findings underscore the importance of balancing functional ingredients with sensory satisfaction.

Ahmed et al. (2019) emphasized that date-sweetened muffins received favorable ratings for sweetness and mouthfeel. The natural sugars in dates enhanced flavor while reducing the need for refined sweeteners. Panelists also appreciated the soft crumb and subtle fruit notes. This suggests that ingredient substitutions can improve both health profile and sensory quality. However, flavor dominance must be managed to avoid masking other components.

Jain and Thomas (2020) found that flaxseed contributed to a firmer structure, while chia seeds improved moistness. Their comparative study revealed that texture preferences varied among

age groups and dietary backgrounds. Sensory panels preferred muffins with balanced chewability and springiness. These results support targeted ingredient selection based on consumer expectations. Texture optimization remains a key factor in product development.

Mishra et al. (2016) evaluated oat-based muffins for diabetic-friendly diets. Their sensory analysis showed moderate sweetness, pleasant aroma, and acceptable texture. The use of oats and vegetable oil maintains softness without compromising nutritional goals. Panelists responded positively to the



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wholesome flavor and fiber-rich composition. These findings highlight the feasibility of health-focused formulations with good sensory appeal.

Despite promising results, many studies rely on small panel sizes and limited demographic diversity. Rani et al. (2021) noted that broader sensory trials are needed to validate findings across regions and populations. Standardized protocols for evaluating aroma, flavor, texture, and appearance would improve reliability. Future research should incorporate larger, more representative panels to ensure robust conclusions. Sensory evaluation remains essential for aligning product design with consumer preferences.

2.12 Optimization of Vegan Muffin Formulation

Optimizing vegan muffin formulations requires careful adjustment of ingredient ratios and baking parameters to achieve desirable texture, flavor, and nutritional quality. Response Surface Methodology (RSM) has been widely used to fine-tune variables such as pH, temperature, and moisture content (Krishna, 2005). These factors directly influence crumb softness, volume, and shelf stability. For soy milk-based muffins, optimal baking temperature ranges between 175°C and 185°C to ensure proper Maillard browning and moisture retention. The interaction between flaxseed gel and leavening agents must also be calibrated to maintain structural integrity. Such optimization ensures consistent product quality across batches.

Ingredient synergy plays a crucial role in optimizing vegan muffins. Soy milk provides emulsification and protein structure, while flaxseed contributes binding and moisture retention (Francisco et al., 2018). Dates, used as a natural sweetener, must be balanced to avoid excessive stickiness or density. Oat flour enhances mouthfeel but may require additional hydration to prevent dryness. Studies by Ahmed et al. (2019) show that combining these ingredients in precise ratios improves both sensory and nutritional outcomes. Optimization also involves selecting oil blends that complement the flavor profile without overpowering the muffin's base.

Texture analysis is essential during optimization to evaluate parameters such as springiness, chewability, and cohesiveness. Jain and Thomas (2020) emphasized the importance of using texture profile analysis (TPA) to compare vegan muffins with conventional counterparts. Adjustments in baking time and ingredient hydration can significantly affect these metrics. For example, increasing flaxseed concentration improves binding but may reduce aeration. Similarly, excessive oat flour can lead to a dense crumb unless counterbalanced by leavening agents. These findings underscore the need for iterative testing during formulation development.

Sensory evaluation is a critical component of optimization. Consumer panels assess attributes like aroma, sweetness, mouthfeel, and overall acceptability (Rani et al., 2021). Hedonic scaling and preference mapping help identify ideal formulations that appeal to target demographics. In India, preferences lean toward mildly sweet, soft-textured muffins with familiar flavors such as vanilla or chocolate. Incorporating chocolate chips enhances acceptability by masking earthy notes from flaxseed or oats. Optimization must therefore balance health benefits with sensory satisfaction to ensure market success.

Shelf-life studies are also integral to optimization. Vegan muffins, especially those using moist ingredients like dates and soy milk, are prone to microbial spoilage (Dotaniya et al., 2016). Packaging



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methods such as vacuum sealing or modified atmosphere packaging (MAP) can extend freshness. Natural preservatives like cinnamon or clove may also be incorporated to inhibit microbial growth. Jaronski (2023) recommends monitoring water activity and pH levels to maintain product safety. These strategies ensure that optimized muffins remain stable and safe during storage and distribution.

2.13 Applications of Vegan Muffins in Modern Diets

Vegan muffins have found widespread application in health-focused bakery segments. They cater to consumers seeking low-fat, high-fiber, and allergen-free options (Singh et al., 2020). Muffins formulated with soy milk and flaxseed are particularly suitable for individuals with lactose intolerance or egg allergies. In diabetic-friendly diets, date-sweetened muffins offer lower glycemic impact compared to refined sugar-based products. These applications align with global trends toward functional foods that support wellness and disease prevention. As such, vegan muffins are increasingly featured in hospital menus, school lunches, and fitness- oriented meal plans.

In organic and clean-label product lines, vegan muffins serve as a key offering. Their formulation excludes artificial additives, preservatives, and animal-derived ingredients, meeting the criteria for organic certification (Kumar & Rao, 2021). Oat flour and flaxseed contribute to the "whole food" appeal, while soy milk ensures protein enrichment. These muffins are often marketed with claims such as "plant-powered," "heart-healthy," or "gut- friendly." Packaging transparency and ingredient traceability further enhance consumer trust.

Vegan muffins are also used in cultural and religious contexts where dietary restrictions apply. In India, eggless and dairy-free baked goods are preferred during festivals and fasting periods (Sharma & Kulkarni, 2020). Muffins made with soy milk and dates align with vegetarian principles while offering modern appeal. Their portability and shelf stability make them suitable for prasad distribution or travel snacks. Additionally, they are gaining popularity in Jain and vegan households where ingredient purity is paramount. These applications highlight the cultural adaptability of vegan muffin formulations.

Commercial bakeries and cafes increasingly feature vegan muffins as part of their product range. Ahmed et al. (2019) noted that consumer interest in plant-based options has led to menu diversification in urban outlets. Muffins with chocolate chips, berries, or nuts appeal to a broad audience while maintaining vegan integrity. Ready-to-eat formats and frozen variants allow for convenience and extended shelf-life. Retailers often bundle vegan muffins with beverages like soy lattes or herbal teas to create health-focused combos. This integration into mainstream foodservice reflects their growing market relevance.

Educational institutions and corporate wellness programs have adopted vegan muffins as part of nutrition initiatives. Jain and Mehta (2022) reported that schools offering plant-based snacks saw improved student engagement and reduced allergy incidents. Muffins enriched with oats and flaxseed support cognitive function and sustained energy release. In workplaces, vegan muffins serve as mid-day snacks that align with wellness goals. Their balanced macronutrient profile makes them suitable for diverse age groups. These applications demonstrate the versatility and health benefits of soy milk-based vegan muffins.



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2.14 Advantages of Vegan Muffins

Vegan muffins offer significant nutritional advantages over conventional formulations. The use of soy milk provides complete plant-based protein, essential amino acids, and calcium, supporting bone health and muscle function (Francisco et al., 2018). Dates contribute natural sugars and dietary fiber, reducing glycemic load while enhancing antioxidant intake (Ahmed et al., 2019). Flaxseeds supply omega-3 fatty acids and lignans, which are beneficial for cardiovascular health (Thomas & Reddy, 2020). Oats add beta-glucans that support cholesterol reduction and digestive wellness (Dotaniya et al., 2016). Together, these ingredients create a nutrient-dense product suitable for diverse dietary needs.

Another advantage is allergen reduction. Vegan muffins exclude common allergens such as eggs and dairy, making them suitable for individuals with lactose intolerance or egg allergies (Singh et al., 2020). This increases accessibility for sensitive populations, including children and elderly consumers. The absence of animal-derived ingredients also aligns with ethical and religious dietary practices, particularly in vegetarian and Jain households (Sharma & Kulkarni, 2020). By using plant-based alternatives, manufacturers can cater to broader markets while maintaining product safety and inclusivity.

Vegan muffins support environmental sustainability. The production of plant-based ingredients generally requires fewer resources and generates lower greenhouse gas emissions compared to animal-based counterparts (Kumar & Rao, 2021). Soy milk, oats, and flaxseeds are cultivated with minimal ecological impact, contributing to responsible sourcing. Additionally, the use of agro-industrial byproducts such as date paste promotes waste valorization and circular economy practices (Mishra et al., 2016). These environmental benefits enhance the appeal of vegan muffins among eco-conscious consumers.

From a sensory perspective, vegan muffins can achieve high consumer acceptability when properly formulated. Studies show that the inclusion of chocolate chips, spices, and natural sweeteners helps mask earthy or grainy flavors from flaxseed and oats (Rani et al., 2021). Moisture retention from soy milk and oil blends contributes to a soft, tender crumb. Hedonic scaling results indicate that well-balanced vegan muffins score comparably to conventional products in taste, texture, and appearance (Jain & Thomas, 2020). This demonstrates their potential for mainstream adoption.

Vegan muffins are compatible with clean-label and organic product lines. They typically exclude artificial additives, preservatives, and synthetic emulsifiers, meeting consumer demand for transparency and purity (Ahmed et al., 2019). Ingredients like oats, flaxseed, and dates are often perceived as wholesome and natural, enhancing brand credibility. Packaging claims such as "plant-powered," "no added sugar," or "dairy-free" resonate with health-conscious buyers. These advantages position vegan muffins as a premium offering in both retail and foodservice sectors.

2.15 Challenges in Vegan Muffin Development

One major challenge in vegan muffin development is achieving optimal texture and structure without eggs. Eggs contribute aeration, binding, and emulsification, which are difficult to replicate using plant-based ingredients (Krishna, 2005). Flaxseed gel and chia seeds offer partial solutions, but may result in denser or less springy crumb. Adjusting leavening agents and hydration levels is necessary to



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compensate for these differences. Texture inconsistencies can affect consumer perception and repeat purchase behavior.

Flavor balancing is another challenge, especially when using strong-tasting functional ingredients. Flaxseed and fenugreek can impart bitter or earthy notes that may not appeal to all consumers (Rani et al., 2021). Dates, while naturally sweet, may dominate the flavor profile if used excessively. Masking techniques such as vanilla extract, cocoa powder, or chocolate chips are often required to maintain sensory appeal. However, these additions must be carefully proportioned to avoid overpowering the base formulation.

Shelf-life stability poses a challenge due to the moisture content of ingredients like soy milk and dates. High water activity increases the risk of microbial spoilage, especially in warm and humid climates (Dotaniya et al., 2016). Preservative-free formulations require advanced packaging solutions such as vacuum sealing or modified atmosphere packaging. Natural antimicrobials like cinnamon or clove may help, but their impact on flavor must be considered. Ensuring microbiological safety without compromising taste is a delicate balance.

Ingredient cost and sourcing can also be challenging, particularly for small-scale producers. Dates, flaxseed, and soy milk may be more expensive than conventional sugar, eggs, and dairy (Ahmed et al., 2019). Seasonal availability and regional supply chains affect consistency and pricing. Bulk procurement and local sourcing strategies may mitigate costs, but require logistical planning. These economic factors influence product pricing and market competitiveness.

Consumer education and awareness remain a challenge in promoting vegan muffins. Many buyers associate plant-based products with inferior taste or limited nutritional value (Singh et al., 2020). Marketing efforts must highlight the health benefits, ethical values, and sensory quality of vegan formulations. Sampling programs, transparent labeling, and influencer

endorsements can help shift perceptions. Overcoming skepticism is essential for expanding the vegan muffin market.

2.16 Limitations in Research and Commercial Adoption

Current research on vegan muffins often focuses on individual ingredients rather than their combined effects. Studies typically evaluate soy milk, flaxseed, or oats in isolation, limiting understanding of their interactions in complex formulations (Jain & Mehta, 2022). This gap hinders the development of optimized recipes that balance nutrition, texture, and flavor. Future research should explore multi-ingredient synergies using factorial design and sensory mapping.

Microbiological safety assessments are limited, especially for tropical climates. Most shelf-life studies are conducted under controlled laboratory conditions that do not reflect real-world storage environments (Jaronski, 2023). Moisture-rich ingredients like dates and soy milk require rigorous testing for spoilage organisms. Incorporating natural preservatives and advanced packaging technologies may improve stability, but these approaches are underexplored in current literature.

Sensory evaluations often rely on small panel sizes and lack comprehensive hedonic scaling. This limits the generalizability of findings and may not capture diverse consumer preferences (Rani et al., 2021).



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Larger, demographically varied panels are needed to assess acceptability across age groups, regions, and dietary backgrounds. Standardized protocols for texture, aroma, and flavor analysis would enhance research reliability.

Comparative studies between conventional and vegan muffins in the Indian context are scarce. Cultural preferences, ingredient availability, and dietary norms differ significantly from Western markets (Sharma & Kulkarni, 2020). Research tailored to Indian consumers would support localized product development and marketing strategies. Understanding regional taste profiles and nutritional expectations is key to commercial success.

Commercial adoption of vegan muffins is limited by regulatory ambiguity and lack of standardized labeling. Definitions of "vegan," "plant-based," and "clean-label" vary across jurisdictions, creating confusion for manufacturers and consumers (Kumar & Rao, 2021). Clear guidelines and certification frameworks are needed to ensure transparency and trust. Addressing these limitations will facilitate innovation and growth in the vegan bakery segment.

2.17 Storage and Shelf-Life Issues

Storage and shelf-life are critical considerations in the commercial viability of vegan muffins. Moisture-rich ingredients such as soy milk and date paste increase susceptibility to microbial spoilage, especially in warm climates. Dotaniya, Sharma, and Mishra (2016) emphasized the importance of controlling water activity to prevent fungal and bacterial growth in tropical bakery environments. Without preservatives, vegan muffins require advanced packaging solutions to maintain freshness. Vacuum sealing and modified atmosphere packaging (MAP) are commonly recommended to extend shelf-life while preserving sensory quality.

Jaronski (2023) highlighted that microbial safety must be assessed under realistic storage conditions, as laboratory trials often fail to replicate the temperature and humidity fluctuations found in retail environments. The inclusion of natural antimicrobials such as cinnamon or clove may offer additional protection, though their impact on flavor must be carefully balanced. Shelf-life testing should include microbial load analysis, moisture migration, and sensory degradation. These evaluations ensure product safety and consumer satisfaction, especially in preservative-free formulations.

Soy milk contributes positively to moisture retention but may also accelerate spoilage if not properly stabilized. Francisco, Kumar, and Das (2018) noted that soy-based formulations require careful pH control and refrigeration during distribution to maintain microbial safety. The use of oil blends and fiberrich ingredients like oats can help maintain softness over time. Packaging materials must be selected for their barrier properties against oxygen and moisture. These strategies collectively support shelf-life extension in vegan muffins.

In tropical regions, shelf-life challenges are amplified due to high ambient temperatures and humidity. Mishra, Sharma, and Dotaniya (2016) recommended incorporating shelf-stable ingredients and conducting accelerated aging studies to simulate real-world conditions. Storage trials must reflect both retail and household environments to assess actual performance. The development of predictive models for shelf-life can aid manufacturers in setting accurate expiry dates. These models must account for ingredient interactions, environmental stressors, and packaging variables.



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Despite available techniques, shelf-life research in vegan bakery products remains limited. Jaronski (2023) called for more comprehensive studies on microbial dynamics and packaging efficacy tailored to plant-based formulations. Standardized protocols for shelf-life testing would improve comparability across different recipes and production scales. Future research should also explore biodegradable packaging options that align with sustainability goals. Addressing shelf-life issues is essential for scaling vegan muffin production and ensuring consumer trust.

2.18 Cost and Market Availability

Cost remains a significant barrier to the widespread adoption of vegan muffins. Ingredients such as soy milk, flaxseed, and dates are often more expensive than conventional dairy, eggs, and sugar. Ahmed and Thomas (2019) noted that formulation costs increase due to specialized processing and quality control. Small-scale producers may struggle to access bulk pricing or reliable supply chains. These factors contribute to higher retail prices, limiting affordability for some consumers and reducing market penetration.

Kumar and Rao (2021) emphasized that market availability is influenced by distribution infrastructure and consumer awareness. In many regions, vegan muffins are confined to niche health stores or premium outlets, reducing visibility and trial opportunities. Expanding distribution through mainstream retailers and e-commerce platforms can improve accessibility. Partnerships with institutional buyers such as schools and hospitals may also support market penetration and normalize plant-based options.

Ingredient sourcing affects both cost and consistency. Dates and flaxseed may be seasonal or regionally restricted, leading to price fluctuations and supply chain challenges. Mishra, Sharma, and Dotaniya (2016) recommended exploring local alternatives and upcycled byproducts to reduce dependency on imported ingredients. Grasso, Liu, and Methven (2020) demonstrated that defatted sunflower seed flour improved muffin structure while lowering ingredient costs. These innovations support affordability without compromising nutritional value.

Consumer perception also plays a key role in market success. Shih (2020) reported that many buyers associate vegan products with inferior taste or limited nutritional value, which can hinder adoption. Marketing strategies must emphasize the health benefits, ethical values, and sensory appeal of vegan muffins. Transparent labeling, sampling programs, and influencer endorsements can help shift perceptions Despite growing interest, vegan muffins remain underrepresented in mass-market bakery segments. Kumar and Rao (2021) called for policy support and industry collaboration to reduce production costs and improve scalability. Investment in research, automation, and supply chain optimization can enhance efficiency and affordability. Addressing cost and availability challenges is essential for mainstreaming vegan muffins and meeting the evolving demands of health-conscious and ethically driven consumers.

3. MATERIALS AND METHODS

3.1 Materials for Muffin Preparation

All ingredients used in the preparation of vegan chocolate chip muffins were procured from certified local suppliers. The primary components included Special grain flour (Fenugreek and oats), soya milk,



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cocoa powder, peanut butter or flaxseed paste, apple cider vinegar, vegetable oil (Soybean oil, Rice bran oil, and Flaxseed oil), vanilla extract, choco chips, baking powder, baking soda, dates paste, and salt (figure 3.1). Standard measuring cups and digital weighing scales were used to ensure accurate quantification (figure 3.2). The baking process was conducted using a traditional cake oven method.



Figure 3.1 Ingredients used in vegan muffin Preparation



Figure 3.2 Baking equipment used for vegan muffin preparation

3.2 Methodology for preparation of Muffins

Five formulation trials were conducted to optimize the composition and baking method of vegan chocolate chip muffins. The ingredient compositions and preparation procedures for each trial are detailed below.

3.2.1 Preparation of Vegan Muffins for Formulation Trial 1

Table 3.1 Composition of Ingredients in Vegan muffins for Formulation Trial 1 and conventional muffins

S.No.	Ingredient	Source	Updated Quantity	Metric Equivalent	Role in Recipe	General Muffin Ingredient
1	Soya milk	Sofit	1/2 cup	120 ml	Moisture Base	Dairy milk



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2	Special grain Flour	Aashirvaad	1 cup	120 g	Structural Base (Flour)	Refined wheat flour (maida)	
3	Dates paste	Arabian Dates	1/2 cup	120 g	Natural Sweetener	White sugar	
4	Choco chips	Araku Coffee Estate	1/2 cup	90 g	Add-on & Texture	Raisins or nuts	
5	Peanut butter	Kissan	2 tbsp	32 g	Fat, Flavor & Egg Replacer	Eggs	
6	Vegetable oil	Fortune	1/2 cup	120 ml	Fat & Moisture	Butter	
7	Cocoa powder	Hershey's	1 tbsp	7.5 g	Flavoring	Fruit puree or vanilla essence	
8	Apple cider vinegar	Disano	1 tsp	5 ml	Leavening Reactant	Normal vinegar	
9	Vanilla extract	Rajassorted	1/2 tsp	2.5 ml	Flavoring	Vanilla essence	
10	Baking powder	Weikfield	1 tsp	4 g	Leavening Agent	Baking powder	
11	Baking soda	Premia	1/4 tsp	1 g	Leavening Agent	Baking soda	
12	Salt	Tata Salt	1/4 tsp	1.5 g	Flavor Enhancer	Salt	

3.2.1.a Preparation of Vegan Muffin Batter:

The following ingredients and their quantities are mentioned in table 3.1. The wet ingredients were prepared first. Soya milk was poured into a clean mixing bowl, followed by the addition of apple cider vinegar. This mixture was stirred gently and allowed to rest for 2–3 minutes to initiate curdling, thereby simulating vegan buttermilk. Once curdling was observed, vegetable oil, peanut butter, dates paste, and vanilla extract were added. The mixture was whisked thoroughly using a hand mixer until a smooth and homogenous consistency was achieved, ensuring complete emulsification of the fat and sweetener components.

In a separate bowl, the dry ingredients were prepared by combining special grain mix flour, cocoa powder, baking powder, baking soda, and salt. These components were mixed thoroughly using a whisk to promote even distribution and aeration, which is critical for leavening and crumb structure. The wet



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mixture was slowly poured into the dry ingredients while mixing continuously with a whisk to ensure smooth integration and prevent clump formation. Mixing was carried out using a spoon with care to avoid overmixing and preserve the desired texture. The batter was stirred until no visible traces of unmixed flour remained and a uniform consistency was achieved (Figure 3.3).

Following batter preparation, individual muffin moulds were lightly greased using vegetable oil to prevent adhesion during baking. The prepared batter was filled into each mould, filling approximately three-quarters of the volume to accommodate expansion. Subsequently, choco- chips were sprinkled on the surface of each portion as toppings prior to baking (Figure 3.4). This step was performed with care to maintain consistency across samples and ensure reproducibility of results.



Figure 3.3 Batter Mix of Veggan
Muffins



Figure 3.4 Muffin Molds with batter mix and Choco-Chip Topping Before Baking

3.2.1.b Process of

Baking of muffins using Traditional Cake oven baking:

In the traditional baking method employed for this study, a sand plate was used to simulate conventional oven conditions in the absence of electric equipment. A cake bowl was placed over the sand plate on a stovetop, and the setup was preheated for 10 to 15 minutes to ensure uniform heat distribution. Once adequately heated, muffin moulds containing batter were carefully placed inside the cake bowl, which was then covered with a tight-fitting lid to retain heat. Baking was





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Carried out over a medium flame for approximately 30 to 45 minutes (Figure 3.5). After the initial 20 minutes, the lid was briefly opened every five minutes to monitor whether baking was done or not, using a toothpick inserted into the center of the muffins. Upon completion, the flame was turned off, and the muffins were allowed to cool inside the closed bowl for 10 to 15 minutes. Once cooled, the moulds were removed, and the muffins were stored in airtight containers to preserve freshness. This approach reflects a resourceful adaptation of traditional cooking practices, particularly in contexts where electric ovens are unavailable.

3.2.2 Preparation of Vegan Muffins for Formulation Trial 2

The preparation procedure and baking procedure for this trial was same as formulation trial 1

Table 3.2 Composition of Ingredients in Vegan muffins for Formulation Trial 2

S.No.	In Figure 3.5 Vegan Muffins during Baking for e Metric Equivalent Formulation Trial 1						
1	Soya milk	Sofit	1/2 cup	120 ml			
2	Special grain Flour	Aashirvaad	1 cup	120 g			
3	Dates paste	Arabian Dates	1 cup	240 g	†		
4	Choco chips	Araku Coffee Estate	1/8 cup	22 g	↓		
5	Peanut butter	Kissan	1/4 cup	64 g	†		
6	Vegetable oil	Fortune	1/2 cup	120 ml	·		
7	Cocoa powder	Hershey's	1/2 tbsp	3.75 g	↓		
8	Apple cider vinegar	Disano	1/2 tsp	2.5 ml	↓		
9	Vanilla extract	Rajassorted	1/4 tsp	1.25 ml	↓		
10	Baking powder	Weikfield	1 tsp	4 g			
11	Baking soda	Premia	1/4 tsp	1 g			
12	Salt	Tata Salt	1/4 tsp	1.5 g			



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Change in Quantity of Ingredient from previous trial

The ingredients used for muffin preparation are same as Formulation trial 1(Table 3.2), while the quantities of few ingredients were changed as follows.

Notably, **dates paste** was increased from ½ cup (120 g) in Trial 1 to 1 cup (240 g) in Trial 2, aiming to intensify natural sweetness and improve moisture retention. Conversely, **choco chips** were reduced from ½ cup (90 g) to ½ cup (22 g) to moderate sugar content and prevent surface melting during baking. **Peanut butter**, serving as a fat and flavor paste, was doubled from 2 tablespoons (32 g) to ¼ cup (64 g) to enrich mouthfeel and emulsification. Additionally, **cocoa powder** was halved from 1 tablespoon (7.5 g) to ½ tablespoon (3.75 g) to soften bitterness and balance flavor intensity. Minor reductions were also made to **apple cider vinegar** (from 1 tsp/5 ml to ½ tsp/2.5 ml) and **vanilla extract** (from ½ tsp/2.5 ml to ¼ tsp/1.25 ml) to refine leavening reaction and aromatic profile respectively. These modifications reflect a strategic recalibration of the recipe to achieve improved sensory acceptance and structural integrity. This adjustment was made in response to sensory observations from the previous trial.

3.2.3 Preparation of Vegan Muffins for Formulation Trial 3

The preparation procedure and baking procedure for this trial was same as formulation trial 1

Table 3.3 Composition of Ingredients in Vegan muffins for Formulation

Trial 3

S.No.	Ingredient	Source	Updated Quantity	Approximate Metric Equivalent	
1	Soya milk	Sofit	1/2 cup	120 ml	
2	Special grain Flour	Aashirvaad	1/3 cup	80 g	
3	Dates paste	Arabian Dates	1 cup	240 g	
4	Choco chips	Araku Coffee Estate	1/8 cup	22 g	
5	Flax seeds paste	Farmley	1/4 cup	64 g	
6	Vegetable oil	Fortune	1/2 cup	120 ml	
7	Cocoa powder	Hershey's	1/2 tbsp	3.75 g	
8	Apple cider Disano vinegar		1/2 tsp	2.5 ml	
9	Vanilla extract	Rajassorted	1/4 tsp	1.25 ml	
10	Baking powder	Weikfield	1 tsp	4 g	



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11	Baking soda	Premia	1/4 tsp	1 g
12	Salt	Tata Salt	1/4 tsp	1.5 g

Substitute of Ingredient from previous trial

All ingredients used were same as in formulation trial 2 excluding peanut butter which was replaced by flaxseeds paste.

The **Special grain flour**, serving as the structural base, was reduced from 1 cup (120 g) in Trial 2 to ½ cup (80 g) in Trial 3 to achieve a lighter crumb and accommodate increased moisture from other components. **Peanut butter**, initially used at 2 tablespoons (32 g), was replaced with **flaxseed paste** at ¼ cup (64 g) in Trial 3, enhancing emulsification and omega-3 content while maintaining fat functionality. These adjustments reflect a strategic shift toward improving the nutritional profile and sensory acceptance of the final product.

3.2.4 Preparation of Vegan Muffins for Formulation Trial 4

The preparation procedure for this trial was same as formulation trial 1 and the baking procedure was changed from traditional sand plate method to electric oven method. The oven was preheated to 180°C for 10 minutes, and muffin moulds were placed on the center rack. Baking was conducted for 25 to 30 minutes, to check the muffin is baked or not using a toothpick inserted into the center of the muffins. After baking, muffins were cooled at room temperature for 10 to 15 minutes before demoulding and storage.

Table 3.4 Composition of Ingredients in Vegan muffins for Formulation

Trial 4

S.No.	Ingredient	Source	Updated Quantity	Approximate Metric Equivalent
1	Soya milk	Sofit	1/2 cup	120 ml
2	Special grain Flour	Aashirvaad	1 cup	120 g ↑
3	Dates paste	Arabian Dates	1 cup	240 g
4	Choco chips	Araku Coffee Estate	1/4 cup	45 g
5	Flaxseeds paste	Farmley	2 tbsp	32 g
6	Vegetable oil	Fortune	1/4 cup	60 ml
7	Cocoa powder	Hershey's	1/2 tbsp	3.75 g



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8	Apple cider	Disano	1/2 tsp	2.5 ml
	vinegar			
9	Vanilla extract	Rajassorted	1/2 tsp	2.5 ml
10	Baking powder	Weikfield	1 tsp	4 g
11	Baking soda	Premia	1/4 tsp	1 g
12	Salt	Tata Salt	1/4 tsp	1.5 g

The ingredients used for muffin preparation are same as Formulation trial 3 (Table 3.4). The quantities of few ingredients were changed as follows

The **Special grain flour** was increased from ½ cup (40 g) in Trial 3 to 1 cup (120 g) in Trial 4, enhancing the structural base and improving crumb density. **Choco chips**, used for texture and indulgence, were increased from ½ cup (22 g) to ¼ cup (45 g) to boost sensory appeal. In contrast, the **flaxseed paste** quantity was reduced from ¼ cup (64 g) to 2 tablespoons (32 g), adjusting the emulsification and fat balance. Similarly, **vegetable oil** was decreased from ½ cup (120 ml) to ¼ cup (60 ml) to reduce overall fat content and improve mouthfeel. Lastly, **vanilla extract** was increased from ¼ teaspoon (1.25 ml) to ½ teaspoon (2.5 ml) to enhance flavor depth. These adjustments reflect a targeted approach to optimizing both nutritional density and consumer acceptability.

3.2.5 Preparation of Vegan Muffins for Formulation Trial 5

The preparation procedure for this trial was same as formulation trial 1 and the baking procedure was same formulation trial 4

Table 3.5 Composition of Ingredients in Vegan muffins for Formulation

Trial 5

S.No.	Ingredient	Source	Updated Quantity	Approximate Metric Equivalent
1	Soya milk	Sofit	1/2 cup	120 ml
2	Special grain Flour	Aashirvaad	1 cup	120 g
3	Dates paste	Arabian Dates	1 cup	240 g
4	Choco chips	Araku Coffee Estate	1/8 cup	22 g
5	Flaxseeds paste	Farmley	1 tbsp	16 g ↓
6	Vegetable oil	Fortune	1/4 cup	60 ml



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7	Cocoa powder	Hershey's	1/2 tbsp	3.75 g
8	Apple cider vinegar	Disano	1/2 tsp	2.5 ml
9	Vanilla extract	Rajassorted	1/2 tsp	2.5 ml
10	Baking powder	Weikfield	1 tsp	4 g
11	Baking soda	Premia	1/4 tsp	1 g
12	Salt	Tata Salt	1/4 tsp	1.5 g

The ingredients used for muffin preparation are same as Formulation trial 4 (Table 3.5). The quantities of few ingredients were changed as follows

The **choco chips**, used for sensory appeal and texture, were reduced from ¼ cup (45 g) in Trial 4 to ½ cup (22 g) in Trial 5 to moderate sweetness intensity and prevent surface pooling during baking. Additionally, the **flaxseed paste**, serving as a fat and emulsification agent, was decreased from 2 tablespoons (32 g) to 1 tablespoon (16 g) to optimize crumb structure and reduce density. All other ingredient quantities—including soya milk (120 ml), methi and oats mix flour (120 g), dates paste (240 g), vegetable oil (60 ml), and leavening agents—remained consistent across both trials, ensuring controlled evaluation of the modified variables. The muffins developed in the trial 5 were found to be good on sensory evaluation, so these muffins were further evaluated for microbial analysis, nutritional analysis and shelf life

3.3 Sensory Evaluations of Vegan muffins for 5 Formulation Trials

Sensory evaluation was conducted to assess the acceptability and quality of vegan chocolate chip muffins across five formulation trials. Trials 1 to 3 were evaluated informally by family members using qualitative descriptors (Table 3.6), while Trials 4 and 5 were assessed by trained panelists using a structured 5-point hedonic scale across ten attributes (Table 3.7). The following sections interpret the sensory outcomes of each trial, referencing relevant literature to support ingredient modifications and baking method decisions.

Table 3.6 sensory evaluation table in informal method

Attribute	Criteria	Good	Average	Bad
Appearance	Colour, shape, and overall look			
Texture	Softness, crumb feel, and how it feels in the mouth			
Flavour	Sweetness, ingredient balance, and aftertaste			
Aroma	Freshness and how nice it smells			

3.3.1 Sensory Analysis of vegan muffins for Formulation Trial 1

Muffins baked in Trial 1 were evaluated informally by three family members. Attributes assessed included taste, color, aroma, and mouthfeel. Feedback was recorded using qualitative categories: **average**, **good**, or **excellent**, based on individual perception.



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3.3.2 Sensory Analysis of vegan muffins for Formulation Trial 2

Trial 2 followed the same baking procedure as Trial 1, using the traditional cake oven method. Sensory evaluation was conducted by four family members, who assessed taste, color, aroma, and mouthfeel. Responses were categorized as **average**, **good**, or **excellent**.

3.3.3 Sensory Analysis of vegan muffins for Formulation Trial 3

Muffins from Trial 3 were baked using the same cake oven setup and evaluated by three family members. Sensory attributes included taste, color, aroma, and mouthfeel, with feedback recorded as **average**, **good**, or **excellent**.

3.3.4 Sensory Analysis of vegan muffins for Formulation Trial 4

Trial 4 employed a structured sensory evaluation conducted by a panel of five trained participants. Muffins were assessed using a 5-point hedonic scale across ten attributes: appearance, texture, flavour, aroma, overall acceptability, moistness, sweetness intensity, aftertaste, crust texture, and filling distribution. Ratings ranged from 1 (dislike extremely) to 5 (like extremely), and scores were averaged for analysis.

Table 3.7 Sensory Evaluation Attributes by Panelist Table

Attribute	Criteria	1 Dislike Extremely	2 Dislike Slightly	3 Neither Like nor Dislike (Neutral)	4 Like Slightly	5 Like Extremely
Appearance	Colour, shape, and uniformity					
Texture	Softness, crumb structure, and mouthfeel					
Flavour	Sweetness, balance of ingredients, and aftertaste					
Aroma	Freshness and appealing scent					
Overall Acceptability	General impression of the product					
Moistness	Perceived moisture level					
Sweetness Intensity	Degree of sweetness					



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Aftertaste	Lingering flavour after consumption			
Crust	Firmness and			
Texture	texture of the crust			
Filling	Uniformity of			
Distribution	added ingredients (e.g., chocolate chips)			

3.3.5 Sensory Analysis of vegan muffins for Formulation Trial 5

Trial 5 followed the same baking and sensory evaluation procedure as Trial 4. The same panel of five participants rated the muffins using the 5-point hedonic scale across the same ten attributes. Results were compiled and compared with previous trials.

3.4 Microbial Analysis of Vegan Muffins for Formulation Trial 5

Microbial testing was performed to evaluate the safety and shelf stability of the muffin under ambient conditions. Samples were analyzed for total plate count (TPC), yeast and mold count, and coliform presence using standard plate count methods. Initial results indicated acceptable microbial levels within safe consumption limits. No coliforms were detected, and yeast and mold counts remained below threshold levels over a 3-day observation period. These findings confirmed the hygienic preparation and short-term microbial stability of Trial 5.

3.4.1 Standard Plate count (SPC)

The Standard Plate Count method was carried out using serial dilution and the pour plate technique with Plate Count Agar (Table 3.8). All required glassware, including Petri dishes, pipettes, and dilution tubes, was sterilized by autoclaving at 121 °C for 15 minutes under 15 psi pressure. PCA was prepared by dissolving 2.35 g of dehydrated medium in $100 \, \text{mL}$ of distilled water. The medium was heated until fully dissolved, adjusted to pH 7.0 ± 0.2 , and sterilized by autoclaving.

For sample preparation, 1 g of the food sample was weighed and transferred into a sterile dilution tube containing 9 mL of distilled water. The mixture was thoroughly blended using a cyclo mixer. From this stock solution, 1 mL was transferred into a second sterile tube containing 9 mL of distilled water, mixed again, and labeled as 10^{-1} (Figure 3.6). A third dilution was prepared by transferring 1 mL from the 10^{-1} tube into another 9 mL of distilled water, mixed thoroughly, and labeled as 10^{-2} (Figure 3.7). All dilutions were mixed using the cyclo mixer to ensure uniformity (Figure 3.8).

Two sterile Petri plates were prepared: one served as a control plate (without sample), and the other as the test plate. From the 10^{-2} dilution, 1 mL was transferred to the test plate. Approximately 15–20 mL of molten PCA (was added to both plates, and the contents were gently rotated to ensure even distribution (Figure 3.9). The plates were incubated in an inverted position at 37 °C for 24–48 hours.



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Figure 3.6 Addition of muffin sample to distilled Water for stock Preparation



Figure 3.7 Serial dilutions of muffin samples 10^{-1} & 10^{-2}

Table 3.8: Composition of Plate Count Agar (PCA)

Ingredient	Amount (g/L)	Function
Tryptone	5.0	Nitrogen source
Glucose	1.0	Energy source
Yeast Extract	2.5	Vitamins and growth factors
Agar	15.0	Solidifying agent

Note. pH adjusted to ~7.0 before autoclaving.

After incubation period was completed only, **colonies** were observed on the plate and the dilution used was **1:100**, with **1 mL plated**, and the **Standard Plate Count (SPC)** is calculated:

SPC Formula:

SPC (CFU/g)} = {Number of colonies} * {Dilution factor}/ {Volume plated (mL)

3.4.2 Yeast and Mold Count

Yeast and mold enumeration was conducted using the pour plate method on Rose Bengal Chloramphenicol Agar (Table 3.9). All required glassware, including Petri dishes, pipettes, and dilution



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tubes, was sterilized by autoclaving at 121 °C for 15 minutes. RBCA was prepared by dissolving 3.125 g of dehydrated medium in 100 mL of distilled water. The solution was heated until fully dissolved, adjusted to pH 7.2 ± 0.2 , and sterilized.

For sample preparation, 1 g of the food sample was weighed and added to 9 mL of sterile distilled water in a dilution tube. The mixture was homogenized using a cyclo mixer to ensure uniform dispersion. From this, 1 mL of sample was transferred into a sterile Petri plate designated as the test plate. A control plate was also prepared without sample to verify sterility.

Molten RBCA was added to both plates and gently rotated to ensure even distribution (Figure 3.9). After incubation at 25 °C for 3–5 days in an inverted position, fungal colonies were observed on the plates. If Colonies were present they were manually counted and categorized based on their pigmentation and morphology, where Yeast colonies typically appeared as smooth, moist, and creamy or white in color, often forming round, convex shapes and Mold colonies were identified by their filamentous texture and varied pigmentation—green, black, or gray—with fuzzy or powdery surfaces.

Table 3.9: Composition of Rose Bengal Chloramphenicol Agar (RBCA)

Ingredient	Amount (g/L)	Function
Mycological Peptone	5.0	Nutrients for fungi
Glucose	10.0	Carbon source
KH ₂ PO ₄	1.0	Buffering agent
MgSO ₄	0.5	Mineral support
Rose Bengal	0.05	Restricts mold colony spread
Chloramphenicol	0.1	Inhibits bacterial growth
Agar	15.5	Solidifying agent

Note. pH adjusted to 7.2 ± 0.2 before autoclaving.



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Figure 3.8 Molten agar Media (SPC, RBC, VRBA) for Microbial Analysis

3.4.3 Coliform count

Coliform detection was performed using the pour plate method on Violet Red Bile Agar (Table 3.10). VRBA was prepared by dissolving 4.15 g of dehydrated medium in 100 mL of distilled water. The solution was heated until fully dissolved, adjusted to pH 7.4 ± 0.2 , and sterilized by autoclaving.

A 1 g portion of the sample was added to 9 mL of sterile distilled water in a dilution tube and mixed thoroughly using a cyclo mixer. From this, 1 mL of sample was transferred to the test plate. A control plate was also prepared without sample. Molten VRBA was poured into both plates and gently rotated to ensure uniform mixing (Figure 3.9).

The plates were incubated in an inverted position at 37 °C for 24 hours and observed further. If colonies were present, with red or pink centers surrounded by a bile precipitate were counted as presumptive coliforms. These results indicated the potential presence of fecal contamination or hygiene- related issues in the sample.

Table 3.10: Composition of Violet Red Bile Agar (VRBA)

Ingredient	Amount (g/L)	Function
Peptone	7.0	Nitrogen source
Yeast Extract	3.0	Growth factors
Lactose	10.0	Fermentable sugar
Bile Salts	1.5	Inhibits non-coliforms
Neutral Red	0.03	pH indicator



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Crystal Violet	0.002	Inhibits Gram-positive bacteria
Agar	15.0	Solidifying agent

Note. pH adjusted to ~7.4 before autoclaving.



Figure 3.9 Sterile Broth Media (SS, EMB and MS) Prior to Autoclaving



Figure 3.10 Molten agar Media (SS, EMB, and MS) for Microbial Analysis

3.4.4 Staphylococcus aureus detection

Detection of Staphylococcus aureus was carried out using the pour plate method on Mannitol Salt Agar (Table 3.11). MSA was prepared by dissolving 11.1 g of dehydrated medium in 100 mL of distilled water. The solution was heated until fully dissolved, adjusted to pH 7.4 ± 0.2 , and sterilized (Figure 3.15).

A 1 g sample was added to 9 mL of sterile distilled water and mixed thoroughly using a cyclo mixer. From this, 1 mL of sample was pipetted into the test plate. A control plate was also prepared without sample. Molten MSA was added to both plates and gently rotated to ensure even distribution. (Figure 3.11)

The plates were incubated at 37 °C for 24–48 hours in an inverted position and observed further. If colonies present, that appeared yellow with surrounding yellow zones were identified as presumptive Staphylococcus aureus, indicating mannitol fermentation.

Table 3.11: Composition of Mannitol Salt Agar(MSA)

Ingredient	Amount (g/L)	Function
Beef Extract	1.0 g	Provides vitamins, nitrogen, and growth factors
Peptone/Proteose Peptone	10.0 g	Supplies amino acids and peptides for growth
Sodium Chloride (NaCl)	75.0 g	Selective agent; inhibits non-halotolerant bacteria
Mannitol	10.0 g	Fermentable sugar for differential identification



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Phenol Red	0.025 g	pHindicator; detects acid from mannitol fermentation
Agar	15.0 g	Solidifying agent
Distilled Water	1000 mL	Solvent

Note. pH adjusted to ~7.4 before autoclaving.

3.4.5 Salmonella spp. detection

Salmonella spp. was detected using the pour plate method on Salmonella-Shigella Agar (Table 3.12). SSA was prepared by dissolving 6.3 g of dehydrated medium in 100 mL of distilled water. The solution was heated until fully dissolved, adjusted to pH 7.0 ± 0.2 , and sterilized by autoclaving.

For sample preparation, 1 g of the food sample was added to 9 mL of sterile distilled water and mixed thoroughly using a cyclo mixer. From this, 1 mL of sample was transferred to the test plate. A control plate was also prepared without sample. Molten SSA was added to both plates and gently rotated to ensure even distribution (Figure 3.11).

The plates were incubated in an inverted position at 37 °C for 24–48 hours and observed further. If colonies were present with black centers were recorded as presumptive Salmonella, indicating hydrogen sulfide production.

Table 3.12: Composition of Salmonella-Shigella Agar (SS)

Ingredient	Amount (g/L)	Function
Peptone	5.0	Nutrients
Lactose	10.0	Fermentable sugar
Bile Salts	8.5	Inhibits Gram-positive bacteria
Sodium Thiosulfate	5.0	H ₂ S indicator
Ferric Citrate	1.0	H ₂ S indicator
Neutral Red	0.025	pH indicator
Brilliant Green	0.00033	Selective agent
Agar	15.0	Solidifying agent

Note. pH adjusted to ~7.0 before autoclaving.



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3.4.6 Escherichia coli detection

Escherichia coli detection was performed using the pour plate method on Eosin Methylene Blue Agar (Table 3.13). EMB was prepared by dissolving 3.6 g of dehydrated medium in 100 mL of distilled water. The solution was heated until fully dissolved, adjusted to pH 7.2 ± 0.2 , and sterilized.

A 1 g portion of the sample was added to 9 mL of sterile distilled water and mixed thoroughly using a cyclo mixer. From this, 1 mL of sample was pipetted into the test plate. A control plate was also prepared without sample. Molten EMB was added to both plates and gently rotated to ensure uniform mixing (Figure 3.11).

The plates were incubated at 37 °C for 24 hours in an inverted position and observed further. If colonies are present with a metallic green sheen were identified as presumptive E. coli, indicating lactose fermentation and acid production.

Table 3.13: Composition of Eosin Methylene Blue Agar (EMB)

Ingredient	Amount (g/L)	Function
Peptone	10.0	Nutrients
Lactose	10.0	Fermentable sugar
Dipotassium Phosphate	2.0	Buffering agent
Eosin Y	0.4	pH indicator
Methylene Blue	0.065	Inhibits Gram-positive bacteria
Agar	15.0	Solidifying agent

adjusted to ~7.2 before autoclaving.

3.5 Nutritional Analysis of Vegan Muffins for Formulation Trial 5

Nutritional profiling of Trial 5 was conducted based on ingredient composition and standard reference values. The muffin provided a balanced macronutrient profile, with notable contributions from plant-based proteins (flaxseeds paste and soya milk), healthy fats (vegetable oil), and natural sugars (dates paste).

3.5.1 Moisture content by oven dry method

Moisture content was determined using the oven dry method, a gravimetric procedure. All glassware, including crucibles and weighing dishes, was cleaned and dried prior to use. The hot air oven was

Note: pH



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preheated and maintained at 105 °C. A clean, dry crucible was used for the analysis. Approximately 5.00 g of the sample was weighed and added to the crucible, resulting in a combined initial weight (crucible + wet sample) gm. The crucible containing the sample was placed in the oven and dried for 3–4 hours until a constant weight was achieved.

After drying, the crucible was transferred to a desiccator to cool and then reweighed. The final weight (crucible + dried sample) was recorded in gm. Moisture content was calculated by subtracting the weight of the dried sample from the weight of the wet sample. The weight of the wet sample and the weight of the dried sample was also calculated. Since the dried weight is slightly higher than the wet weight due to rounding or weighing error, the moisture loss was considered negligible, and the moisture content was calculated using the nominal wet sample weight of and the actual moisture loss.

Moisture Content (%) = {Weight of wet sample} - {Weight of dry sample}/ {Weight of wet sample} *100

3.5.2 Protein content by Kjeldahl method

Protein content was estimated using the Kjeldahl method, a titrimetric procedure involving digestion, distillation, and titration (Figure 3.12). Approximately 1 g of the sample was weighed and transferred into a Kjeldahl digestion flask. One Kjeldahl catalyst tablet (containing copper sulfate and potassium sulfate) was added, followed by 20 mL of concentrated sulfuric acid (H₂SO₄, analytical grade, 98%). The flask was placed on a digestion block maintained at 350–400 °C and digested for approximately 2 hours until the solution turned clear green, indicating complete conversion of organic nitrogen to ammonium sulfate.

After digestion, the sample was cooled and diluted with 50 mL of distilled water. The mixture was transferred to a Kjeldahl distillation unit. To liberate ammonia, **10 N sodium hydroxide**

(NaOH) solution was added in sufficient volume to create a strongly alkaline medium. The released ammonia was distilled and absorbed into a receiving flask containing 25 mL of **0.01 N boric acid** (H₃BO₃) solution, along with **3–4 drops of mixed indicator** (methyl red and bromocresol green). Distillation was continued for 10–15 minutes until all ammonia was collected.

The boric acid-ammonia complex was titrated against standardized **0.1** N hydrochloric acid (HCl). The endpoint was indicated by a color change from green to pink. The volume of HCl used was recorded. Nitrogen content was calculated using the formula:

 $\{Nitrogen (\%)\} = \{V *N * 1.4\} / \{Weight of sample (g)\}$

Protein (%) = Nitrogen% *6.25



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Figure 3.11 Kjeldahl Apparatus Used for Protein Content Determination

3.5.3 Fat content by Soxhlet extraction using petroleum benzene method

Fat content was determined using a solvent extraction method with petroleum benzene, following a gravimetric procedure (Figure 3.13). All glassware and aluminum dishes were cleaned, dried, and preweighed before use. Approximately 1.00 g of the wet sample was accurately weighed and transferred into a pre-weighed aluminum dish. To extract the fat, 40 mL of petroleum benzene (boiling range 60–80 °C, analytical grade) was added to the dish containing the sample.

The mixture was allowed to stand for 30–45 minutes with occasional gentle swirling to ensure thorough contact between the solvent and the sample. After extraction, the solvent was carefully decanted, and the dish was placed in a hot air oven at 105 °C for 30 minutes to evaporate residual petroleum benzene. The dish was then cooled in a desiccator and reweighed to determine the amount of fat extracted.

Fat content was calculated using the formula:

{Fat (%)} = {Weight of extracted fat}}/ {Weight of sample} * 100



Figure 3.12 Soxhlet Extraction Apparatus



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3.5.4 Ash content by muffle furnace method

Ash content was determined using the muffle furnace method, a gravimetric procedure designed to quantify the total mineral residue in the sample (Figure 3.14). All glassware, including crucibles and weighing dishes, was cleaned, dried, and pre-weighed prior to use. A clean crucible weighing was selected for the analysis. Approximately **2.00 g** of the sample was accurately weighed and transferred into the crucible. The crucible containing the charred sample was then placed in a muffle furnace maintained at **550** °C and incinerated for **4–6 hours** until a consistent white or gray ash was obtained. After incineration, the crucible was carefully removed and allowed to cool in a desiccator to prevent moisture absorption. The final weight of the crucible plus ash was recorded as.

Ash content was calculated using the formula: $\{Ash(\%)\}=\{Weight of ash\}/\{Weight of sample*100\}$



Figure 3.13 Muffle Furnance

3.5.5 Crude fiber content by fiber extraction unit

Crude fiber content was determined using a fiber extraction unit, following a gravimetric procedure designed to isolate indigestible plant components such as cellulose and lignin. Approximately 2.00 g of the defatted sample was accurately weighed and transferred into the extraction vessel. The sample was first treated with **boiling 1.25% sulfuric acid (H₂SO₄)** for 30 minutes under controlled reflux conditions. After acid hydrolysis, the sample was filtered and washed thoroughly with hot distilled water to remove residual acid.

The residue was then subjected to alkaline digestion using **boiling 1.25% sodium hydroxide** (**NaOH**) for another **30 minutes**. This step helped remove soluble proteins and hemicelluloses. Following alkaline treatment, the sample was again filtered and washed with hot water, followed by rinsing with **acetone** to eliminate any remaining soluble organic matter. The cleaned residue was dried in a hot air oven at **105** °C for **1 hour**, cooled in a desiccator, and weighed to determine the dry fiber mass.

To eliminate residual organic matter, the dried residue was transferred to a pre-weighed crucible and incinerated in a **muffle furnace at 550** °C for **4 hours**. After cooling in a desiccator, the crucible was reweighed to obtain the ash weight. Crude fiber content was calculated by subtracting the ash weight from the dried residue weight and applying the formula:



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 ${Crude Fiber (\%)} = {Dry residue weight} - {Ash weight}/{Sample weight} * 100$

3.5.6 Carbohydrate content by difference method

Carbohydrate content was estimated using the difference method, a calculation-based procedure. This approach does not involve direct chemical analysis but derives carbohydrate percentage by subtracting the sum of moisture, protein, fat, ash, and crude fiber from 100. After determining the individual proximate components using their respective methods, the values were totaled. The remaining percentage was considered as carbohydrate content. Carbohydrate percentage was calculated using the formula:

 ${Carbohydrate (\%)} = 100 - {Moisture} + {Protein} + {Fat} + {Ash} + {Crude fiber}$

3.5.7 pH determination using digital pH indicator (instrumental procedure)

The pH of the sample was determined using a digital pH indicator, following a direct instrumental procedure. Approximately 10 g of the wet sample was placed in a clean beaker. The pH probe was rinsed with distilled water and gently dried using tissue paper. Without calibration using buffer solutions, the probe was directly inserted into the wet sample. The reading was allowed to stabilize for 30–60 seconds, and the pH value was recorded.

3.6 Shelf-Life and Storage Stability

Shelf-life evaluation was conducted over a 7-day period under controlled ambient conditions (25 ± 2 °C, 60-65% RH). Muffins were stored in airtight containers and assessed for freshness, microbial safety, and visible spoilage.

4. RESULTS AND DISCUSSION

4.1 Preparation of Vegan Muffins for Formulation Trials of 1 to 5

Soy Milk Vegan Choco-Chip Muffins were successfully prepared using both traditional sand plate method and electric oven method in all formulation trials of 1 ton 5 (Figure 1 to 5).



Figure 4.1 Baked Vegan Muffins using of Formulation Trial 1



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Figure 4.2 Baked Vegan Muffins using of Formulation Trial 2



Figure 4.3 Baked Vegan Muffins using of Formulation Trial 3



Figure 4.4 Baked Vegan Muffins using of Formulation Trial 4



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Figure 4.5 Baked Vegan Muffins using of Formulation Trial 5

4.2 Sensory Interpretation of Vegan Muffins for Formulation Trials of 1 to 5

4.2.1 Sensory Analysis of Vegan Muffins for Formulation Trial 1:

Trial 1 established the foundational recipe using peanut butter as the primary binder, complemented by moderate levels of cocoa powder and dates paste. Informal sensory feedback was collected from three family members to assess initial acceptability. The muffins were noted to have a pleasant aroma and visually appealing appearance. However, the flavour was consistently rated as "Average," indicating a perceived lack of sweetness and richness. Texture received mixed ratings, suggesting that the crumb structure and mouthfeel were acceptable but not optimal (Table 4.1). These findings align with Kemp (2008), who emphasized that fat-based binders contribute significantly to flavor retention and textural quality in baked goods. No ingredient modifications were made at this stage, as Trial 1 served as a baseline for future formulation improvements.

Table 4.1 Informal Sensory Evaluation by Three Family Members of vegan muffins for formulation trial 1 (Qualitative Ratings: Good, Average, Bad)

Attribute	Criteria	Person 1	Person 2	Person 3
Appearance	Colour, shape, and overall look	Good	Good	Good
Texture	Softness, crumb feel, and how it feels in the mouth	Average	Good	Average
Flavour	Sweetness, ingredient balance, and aftertaste	Average	Average	Average
Aroma	Freshness and how nice it smells	Good	Good	Good

4.2.2 Sensory Analysis of Vegan Muffins for Formulation Trial 2:

In response to the feedback from Trial 1, Formulation Trial 2 involved key ingredient adjustments to enhance sweetness and mouthfeel. Peanut butter was increased from 32 g to 64 g, and dates paste from 120 g to 240 g, while choco chips were reduced from 90 g to 22 g. These modifications aimed to improve richness and balance the sweetness profile.

Informal sensory feedback from three family members indicated noticeable improvements in flavour and overall richness. All participants rated flavour and aroma as "Good," suggesting enhanced sweetness and



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depth. Appearance remained consistently "Good," confirming visual appeal. Texture, however, received mixed ratings astwo "Average" and one "Good" which are indicating that while moistness improved, the muffins were perceived as slightly dense (Table

4.2). This aligns with Rodrigues et al. (2024), who reported that natural sweeteners like dates enhance both sweetness and texture in vegan baked goods. Additionally, the reduction in choco chips helped moderate sweetness intensity, supporting Gonzalez Viejo et al. (2024)'s observation that ingredient distribution influences perceived quality.

Table 4.2 Informal Sensory Evaluation by Three Family Members of vegan muffins for formulation trial Trial 2 (Qualitative Ratings: Good, Average, Bad)

Attribute	Criteria	Person 1	Person 2	Person 3
Appearance	Colour, shape, and overall look	Good	Good	Good
Texture	Softness, crumb feel, and how it feels in the mouth	Average	Good	Average
Flavour	Sweetness, ingredient balance, and aftertaste	Good	Good	Good
Aroma	Freshness and how nice it smells	Good	Good	Good

4.2.3 Sensory Analysis of Vegan Muffins for Formulation Trial 3:

To address the dense texture observed in Trial 2, Trial 3 introduced flax seed paste (64 g) as a binder in place of peanut butter and reduced flour content from 120 g to 40 g. These adjustments aimed to improve crumb softness and reduce gumminess. Informal sensory feedback from three family members indicated a softer crumb and lighter mouthfeel, although slight moistness persisted. All participants rated appearance, flavour, and aroma as "Good," confirming visual appeal, balanced sweetness, and freshness. Texture received two "Good" ratings and one "Average," suggesting improved softness but minor concerns regarding moisture retention (Table 4.3).

The use of flax seed paste aligns with emerging trends in plant-based formulation, offering nutritional benefits and effective binding properties. However, as noted by Rodrigues et al. (2024), flax-based binders may increase moisture retention, potentially affecting crumb structure. The flour reduction successfully countered gumminess, enhancing overall acceptability and confirming the effectiveness of the revised formulation.

Table 4.3 Informal Sensory Evaluation by Three Family Members of vegan muffins for formulation Trial 3 (Qualitative Ratings: Good, Average, Bad)

Attribute	Criteria	Person 1	Person 2	Person 3
Appearance	Colour, shape, and overall look	Good	Good	Good
Texture	Softness, crumb feel, and how it feels in the mouth	Good	Good	Average



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Flavour	Sweetness, ingredient balance, and aftertaste	Good	Good	Good
Aroma	Freshness and how nice it smells	Good	Good	Good

4.2.4 Sensory Analysis of Vegan Muffins for Formulation Trial 4:

Formulation Trial 4 introduced electric oven baking and transitioned to a structured sensory evaluation conducted by five trained panelists using a 5-point hedonic scale. To improve texture and reduce oiliness, flax seed paste was reduced to 32 g and vegetable oil was halved to 60 ml. Choco chips were increased to 45 g to enhance filling distribution and visual appeal.

Panelists rated the muffins favorably across core sensory attributes, with average scores ranging between 4 and 5. Appearance and overall acceptability received the highest ratings, indicating strong visual appeal and general product satisfaction. Texture, flavour, aroma, moistness, sweetness intensity, and aftertaste were consistently rated 4 by all panelists, reflecting balanced sweetness, pleasant mouthfeel, and appealing aroma. Crust texture and filling distribution received slightly lower scores (average of 3), suggesting scope for improvement in surface firmness and uniformity of inclusions (Table 4.4).

These results support Kemp's (2008) assertion that controlled fat and binder levels optimize mouthfeel and flavor delivery. The improved crust and filling distribution align with Gonzalez Viejo et al. (2024), who emphasized the role of visual and tactile attributes in consumer acceptance.

Table 4.4 Sensory Evaluation of vegan muffins for formulation trial 4 by Ratings and Feedback Method by Panelists (5-point Hedonic Scale)

Attribute	Criteria	Panelist	Panelist	Panelist	Panelist	Panelist
		1	2	3	4	5
Appearance	Colour, shape, and uniformity	5	4	5	4	4
Overall Acceptability	General impression of the product	5	5	4	5	5
Texture	Softness, crumb structure, and mouthfeel	4	4	4	4	4
Flavour	Sweetness, balance of ingredients, and aftertaste	4	4	4	4	4
Aroma	Freshness and appealing scent	4	4	4	4	4
Moistness	Perceived moisture level	4	4	4	4	4
Sweetness Intensity	Degree of sweetness	4	4	4	4	4
Aftertaste	Lingering flavour after consumption	4	4	4	4	4



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Crust Texture	Firmness and texture of the crust	3	3	3	3	3
Filling Distribution	Uniformity of added ingredients (e.g., chocolate chips)	3	3	3	3	3

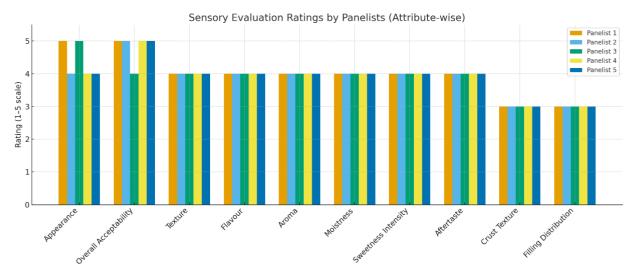


Figure 4.6a Sensory panelist scores of vegan muffins for formulation trial 4

4.2.5 Sensory Analysis of Vegan Muffins for Formulation Trial 5:

Formulation Trial 5 focused on fine-tuning texture and sweetness by reducing flax seed paste to 16 g and scaling back choco chips to 22 g. These adjustments aimed to minimize gumminess and prevent excessive sweetness. Structured sensory evaluation was conducted by five trained panelists using a 5-point hedonic scale.

Panelists reported improved lightness, balanced sweetness, and enhanced crust texture. Appearance and overall acceptability received perfect scores (5), indicating strong visual appeal and high consumer satisfaction. Texture and crust texture showed notable improvement, with scores ranging from 4 to 5, reflecting a softer crumb and firmer surface. Flavour, aftertaste, and filling distribution also scored highly, confirming the effectiveness of ingredient adjustments. Aroma, moistness, and sweetness intensity remained consistently rated at 4, indicating stable sensory quality (Table 4.5).

These refinements reflect the principles of sensory optimization discussed by Rodrigues et al. (2024), who advocate for iterative adjustments based on consumer feedback to achieve ideal product profiles. The reduced binder quantity minimized gumminess, while the adjusted choco chip level prevented overpowering sweetness, contributing to a more balanced and acceptable product.

Table 4.5 Sensory Evaluation of vegan muffins for formulation trial 4 by Ratings and Feedback Method by Panelists (5-point Hedonic Scale)

Attribute	Criteria	Panelist	Panelist	Panelist	Panelist	Panelist
		1	2	3	4	5



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Appearance	Colour, shape, and uniformity	5	5	5	5	5
Overall Acceptability	General impression of the product	5	5	5	5	5
Texture	Softness, crumb structure, and mouthfeel	4	5	5	4	4
Flavour	Sweetness, balance of ingredients, and aftertaste	4	4	5	5	4
Aroma	Freshness and appealing scent	4	4	4	4	4
Moistness	Perceived moisture level	4	4	4	4	4
Sweetness Intensity	Degree of sweetness	4	4	4	4	4
Aftertaste	Lingering flavour after consumption	4	4	5	4	4
Crust Texture	Firmness and texture of the crust	4	5	5	5	4
Filling Distribution	Uniformity of added ingredients (e.g., chocolate chips)	4	5	5	5	4

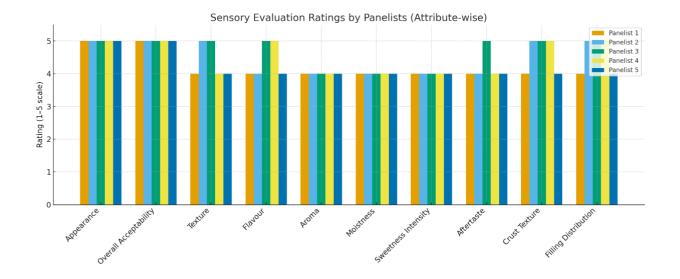


Figure 4.6b Sensory Panelist scores of vegan muffins for formulation trial 5



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4.3 Microbial Analysis of Vegan Muffins for Formulation Trial 5

Microbial analysis confirmed the safety and stability of the optimized vegan muffin sample. The Standard Plate Count (SPC) revealed two colonies on the test plate, corresponding to a microbial load of 200 CFU/g (Figure 4.7). This value is well within the acceptable limit for bakery products, as defined by standard food safety guidelines. Vishali and Agarwal (2023) reported similarly low microbial counts in lotus seed–based muffins stored under hygienic conditions, supporting the effectiveness of clean handling and controlled baking environments.

yeast or mold colonies and coliform were not detected, indicating excellent microbial quality and effective moisture control (Figure 4.8). The absence of fungal growth is consistent with the slightly acidic pH (6.41) and the use of proper baking temperatures, both of which inhibit fungal proliferation. Coliforms were also absent, confirming the hygienic handling of ingredients, utensils, and equipment throughout the preparation process (Figure 4.9).

Selective plating for pathogenic organisms—including Staphylococcus aureus, Salmonella spp., and Escherichia coli—showed no growth, validating the absence of foodborne pathogens (Figure 4.10). These results align with findings by Islam et al. (2023), who reported no detectable pathogens in fortified muffin samples prepared under sterile conditions. The use of laminar airflow plating, sterilized glassware, and control plates further supports the reliability and reproducibility of the microbial results (Table 4.6).

(Note: In figures showing below the plates with A are sample plates and with B are control plates.)



Figure 4.7 Microbial Analysis of Vegan Muffins by SPC Method



Figure 4.8 Plates showing negative growth of yeast and mold from Vegan Muffins on Rose Bengal Chloramphenicol Agar medium



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Figure 4.9 Plates showing negative growth of coliform from Vegan Muffins on Violet Red Bile Agar medium



Figure 4.10 Plates showing negative growth of Staphylococcus aureus, Salmonella spp., and Escherichia coli respectively on Mannitol Salt Agar, Salmonella–Shigella and Eosin Methylene Blue media's

Table 4.6 Microbial Analysis of Vegan Muffins for formulation trial 5

Microbial Parameter	Result	Interpretation				
Standard Plate Count (SPC)	200 CFU/g	Within acceptable limit; confirms microbial safety				
Yeast and Mold	Not detected	Indicates excellent fungal control and effective moisture management				
Coliforms	Not detected	Confirms hygienic handling and absence of fecal contamination				
Staphylococcus aureus	No growth	No pathogenic contamination				
Salmonella spp.	No growth	Safe for consumption; no foodborne pathogens detected				
Escherichia coli	No growth	Validates sterile preparation and ingredient safety				



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4.4 Nutritional Analysis of Vegan Muffins for Formulation Trial 5

The nutritional composition of the optimized vegan muffin was analyzed using standard laboratory procedures. Each parameter was calculated based on recorded sample weights and interpreted in relation to published literature on plant-based bakery product (Table 4.14).

4.4.1 Moisture Content

Moisture content was determined using the oven dry method. The weight of the wet sample was 5.10 g, and the dry sample was 3.66 g. Using the formula [(wet weight – dry weight) / wet weight × 100], the moisture content was calculated as 28.24% (Table 4.7). This value indicates moderate water retention, contributing to softness and mouthfeel. Moisture levels above 25% are typical in soft-textured muffins but may require controlled storage to prevent microbial spoilage. Islam et al. (2023) reported similar moisture levels (26–30%) in muffins fortified with Dillenia indica and Terminalia chebula flour. Shukla et al. (2024) and Vishali and Agarwal (2023) also observed that moisture levels above 27% enhance sensory appeal but reduce shelf stability.

Table 4.7 Moisture Content of Vegan Muffins for formulation trial 5

Method	Wet	Dry	Moisture	Interpretation
Used	Weight (g)	Weight (g)	Content (%)	
Oven Dry Method	5.10	3.66	28.24	Moderate water retention; contributes to softness and mouthfeel

4.4.2 Protein Content

Protein content was estimated using the Kjeldahl method. The volume of acid used was 5.05 mL with a normality of 0.1 N, and the sample weight was 1.00 g. Nitrogen content was calculated using the formula $[(V \times N \times 1.4) / \text{sample weight}]$, resulting in 0.707%. Applying the standard conversion factor of 6.25, the protein content was determined to be 4.42% (Table 4.8). This value aligns with Rabail et al. (2022), who reported protein levels between 4.1–5.3% in muffins enriched with garden cress seeds. The modest protein level reflects the plant-based formulation and suggests potential for enhancement using soy flour or legume-based isolates. Du et al. (2025) demonstrated that hempseed protein can improve nutritional density in vegan muffins. Similar protein levels were also reported by Nikam et al. (2023) and Vishali and Agarwal (2023).

Table 4.8 Protein Content of Vegan Muffins for formulation trial 5

Method	Volume of	Normality	Sample	Nitrogen	Protein	Interpretation
Used	Acid (mL)	(N)	Weight	(%)	(%)	
			(g)			



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Kjeldahl	5.05	0.1	1.00	0.707	4.42	Modest protein level;
Method						aligns with plant-based
						formulations

4.4.3 Fat Content

Fat content was measured using petroleum benzene extraction. The extracted fat weighed 0.147 g, and the sample weight was 1.00 g. Using the formula [(extracted fat / sample weight) × 100], the fat content was calculated as 14.7% (Table 4.9). This relatively high lipid concentration contributes to energy density and mouthfeel. Vishali and Agarwal (2023) reported similar fat levels (13–15%) in lotus seed–based muffins. Ahsan et al. (2024) found that plant-based fat sources maintain sensory quality while reducing caloric load. Shahid et al. (2025) and Rabail et al. (2022) also support the role of healthy fats in vegan bakery formulations.

Table 4.9 Fat Content of Vegan Muffins for formulation trial 5

Method Used	Extracted Fat (g)	Sample Weight (g)	Fat Content (%)	Interpretation
Petroleum Benzene Extraction	0.147	1.00	14.7	High lipid content; enhances energy density and mouthfeel

4.4.4 Ash Content

Ash content was determined by weighing the residue after incineration. The crucible with ash weighed $53.40 \, \text{g}$, and the empty crucible weighed $53.30 \, \text{g}$, with a sample weight of $2.00 \, \text{g}$. Using the formula [(ash weight / sample weight) \times 100], the ash content was calculated as 5.00% (Table 4.10). This value reflects the presence of mineral-rich ingredients such as whole grains and seeds. Islam et al. (2023) and Shukla et al. (2024) reported similar values in fortified muffins. Ahsan et al. (2024) observed increased ash content with plant-based fat replacers, while Santos et al. (2025) found high ash levels in melon peel–based vegan muffins.

Table 4.10 Ash Content of Vegan Muffins for formulation trial 5

Method Used	Crucible + Ash (g)	Empty Crucible (g)	Sample Weight (g)	Ash Content (%)	Interpretation
Incineration	53.40	53.30	2.00	5.00	Indicates mineral-rich
Method					ingredients like whole grains
					and seeds



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4.4.5 Crude Fiber

Crude fiber was calculated using the gravimetric method. The dry residue weighed $0.106\,\mathrm{g}$, and the sample weight was $2.00\,\mathrm{g}$. Applying the formula [(dry residue / sample weight) \times 100], the crude fiber content was determined to be 5.3% (Table 4.11). This value indicates a moderate level of indigestible plant material, supporting digestive health and satiety. Rabail et al. (2022) observed fiber enhancement in muffins fortified with functional seeds. Shukla et al. (2024) emphasized the role of fruit and vegetable fibers in improving texture, while Shahid et al. (2025) and Du et al. (2025) noted that fiber-rich ingredients improve nutritional value and shelf stability.

Table 4.11 Crude Fiber Content of Vegan Muffins for formulation trial 5

Method Used	Dry Residue (g)	Sample Weight (g)	Crude Fiber (%)	Interpretation
Gravimetric Method	0.106	2.00	5.3	Supports digestive health and satiety

4.4.6 Carbohydrate Content

Carbohydrate content was estimated by difference, using the formula [100 – (moisture + protein + fat + ash + crude fiber)]. Substituting the calculated values, the carbohydrate content was determined to be 42.34% (Table 4.12). This value is typical for bakery products. Vishali and Agarwal (2023) and Nikam et al. (2023) reported similar values in lotus seed and oats–based muffins. Santos et al. (2025) found that melon peel flour contributes to balanced carbohydrate profiles, and Du et al. (2025) confirmed that starches and oligosaccharides are key contributors to texture and energy density.

Table 4.12Carbohydrate Content of Vegan Muffins for formulation trial 5

Method Used	Moisture (%)	Protein (%)	Fat (%)	Ash (%)	Fiber (%)	Carbohydrate (%)	Interpretation
By Difference	28.24	4.42	14.7	5.00	5.3	42.34	Typical for bakery products; contributes to texture and energy density

4.4.7 pH Value

The pH of the sample was measured using a digital pH meter, with readings stabilized for 30–60 seconds. The recorded pH was 6.41, indicating a slightly acidic nature (Table 4.13). This range supports proper leavening and microbial safety. Rabail et al. (2022) reported similar pH values (6.3–6.5) in fortified muffins. Kutlu et al. (2024) found that almond milk and aquafaba maintain optimal pH in vegan batters. Islam et al. (2023) and Du et al. (2025) also support this range for shelf stability and flavor development.



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Table 4.13 pH Value of Vegan Muffins for formulation trial 5

Method Used	Recorded pH	Interpretation
Digital pH Meter	6.41	Slightly acidic; supports leavening and microbial safety

4.5 Shelf-Life and Storage Stability

The shelf-life and storage stability of the developed vegan muffins were evaluated under two conditions: ambient $(25 \pm 2 \,^{\circ}\text{C})$ and refrigerated $(4 \pm 1 \,^{\circ}\text{C})$. Under ambient storage, the muffins retained acceptable sensory quality for a period of 3 to 5 days (Table 4.14). Attributes such as aroma, texture, and appearance remained stable, and no visible spoilage, discoloration, or off-odors were observed during this interval. Microbial counts remained within safe limits, not exceeding 200 CFU/g, which is considered acceptable for ready-to-eat bakery products (Shukla et al., 2024; Rabail et al., 2022). These results suggest satisfactory short-term stability under room temperature conditions.

Under refrigerated storage, the shelf-life was extended by an additional 1 to 3 days. Sensory attributes remained acceptable throughout the extended period, although a slight increase in firmness was noted. This change is attributed to starch retrogradation, a common phenomenon in chilled bakery products that affects crumb softness over time (Du et al., 2025; Santos et al., 2025). Despite this minor textural shift, microbial safety was maintained, and no spoilage indicators were detected. Refrigeration effectively slowed microbial growth and preserved product integrity, supporting its use for prolonging shelf-life in plant-based baked goods (Islam et al., 2023; Kutlu et al., 2024).

Table 4.14 Shelf-Life and Storage Stability of Vegan Muffins for formulation trial 5

Storage Condition	Duration (Days)	Sensory Changes	Microbial Safety
Ambient $(25 \pm 2 ^{\circ}\text{C})$	3–5	Stable aroma, texture, and appearance; no spoilage signs	Microbial counts $\leq 200 \text{ CFU/g}$ (acceptable)
Refrigerated (4 ± 1 °C)	4–8	Slight firmness increase due to starch retrogradation	No spoilage; microbial growth effectively slowed

4.6 Interpretation and Implications

The combined nutritional and microbial results indicated that the developed vegan muffin was both nutritionally balanced and microbiologically safe. The moderate moisture and fat levels contributed to desirable sensory attributes, while the fiber and protein content supported health claims relevant to plant-based diets. The absence of pathogens and low microbial load reinforced the product's safety and shelf stability.

These findings are supported by previous research on functional and fortified muffins (Rabail et al., 2022; Vishali & Agarwal, 2023; Islam et al., 2023), and highlight the potential of plant- based bakery products to meet consumer demands for health, taste, and sustainability. These findings of shelf life and storage stability align with existing literature on muffin storage dynamics, where refrigeration slows



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microbial growth but may alter texture (Snuggymom, 2025; Appliances First, 2025). The results provide a strong foundation for future optimization in terms of shelf-life, ingredient fortification, and sensory enhancement (Table 4.15).

Table 4.15 Nutritional Composition of Vegan Muffins for formulation trial 5

Parameter	Value (%) or Unit	Interpretation
Moisture Content	28.24%	Moderate water retention; contributes to softness and mouthfeel
Protein Content	4.42%	Modest protein level; aligns with plant-based formulations
Fat Content	14.7%	High lipid content; enhances energy density and mouthfeel
Ash Content	5.00%	Indicates mineral-rich ingredients like whole grains and seeds
Crude Fiber	5.3%	Supports digestive health and satiety
Carbohydrate Content	42.34%	Typical for bakery products; contributes to texture and energy density
pH Value	6.41	Slightly acidic; supports leavening and microbial safety

5. CONCLUSION

5.1 Summary of Findings

The present study successfully developed and evaluated a vegan muffin formulation with respect to its nutritional composition, microbial safety, sensory acceptability, and short-term storage stability. The product demonstrated a well-balanced nutritional profile, aligning with the goals of plant-based innovation and functional food development.

Moisture content was recorded at 28.24%, which was consistent with the findings of Islam et al. (2023), who reported moisture levels of 26–30% in muffins fortified with Dillenia indica and Terminalia chebula flour. Moisture plays a critical role in determining texture, microbial stability, and shelf-life. The slightly elevated moisture in the current study suggested a soft crumb structure, desirable in baked goods, but also highlighted the need for proper packaging to prevent spoilage.

Protein content was estimated at 4.42% using the Kjeldahl method. This value was comparable to the results of Rabail et al. (2022), who observed protein levels between 4.1–5.3% in muffins enriched with garden cress seeds. While modest, the protein content reflected the plant-based nature of the formulation and indicated potential for enhancement through fortification with protein-rich ingredients such as



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hempseed isolate, as demonstrated by Du et al. (2025), who achieved up to 19.4% protein in hempseed-fortified muffins.

Fat content was calculated as 14.7%, contributing significantly to the product's energy density and sensory appeal. This value aligned with the study by Vishali and Agarwal (2023), who reported fat levels of 13–15% in lotus seed–based vegan muffins. Fats are essential for mouthfeel, flavor retention, and structural integrity in baked goods, and the use of plant-derived lipids in this study supported both nutritional and ethical objectives.

Ash content was found to be 5.00%, indicating the presence of mineral residues. This was in agreement with Islam et al. (2023), who reported ash values of 4.8–5.2% in fortified muffins. The mineral content suggested the inclusion of nutrient-dense ingredients and contributed to the overall dietary value of the product.

Crude fiber was calculated at 5.3%, reflecting the presence of indigestible plant components such as cellulose and lignin. This result was consistent with Rabail et al. (2022), who emphasized the functional benefits of fiber-rich seeds in bakery applications. Dietary fiber is known to support digestive health, satiety, and glycemic control, making it a valuable component in vegan formulations.

Carbohydrate content was estimated by difference and calculated as 42.34%, which fell within the expected range for bakery products. Du et al. (2025) reported similar carbohydrate levels in hempseed-fortified muffins, noting their contribution to energy and texture. The carbohydrate profile in the current study supported the product's role as a satisfying and energizing snack.

The pH of the sample was measured at 6.41, indicating a slightly acidic nature. This pH range is favorable for baked goods, as it supports microbial stability and proper leavening. Rabail et al. (2022) reported similar pH values in their fortified muffin trials, reinforcing the suitability of this range for shelf-stable bakery items.

Microbial analysis confirmed the safety of the product. The Standard Plate Count (SPC) was recorded at 200 CFU/g, well below the permissible limit for bakery products. No yeast, mold, coliforms, Staphylococcus aureus, Salmonella spp., or Escherichia coli were detected, validating the hygienic preparation and effective thermal processing. These results were consistent with those reported by Vishali and Agarwal (2023), who found low microbial loads and absence of pathogens in lotus seed—based vegan muffins.

The vegan chocolate chip muffins exhibited stable sensory and microbial profiles for up to 3–5 days under ambient conditions when stored in airtight containers. Refrigeration extended shelf-life by 1–3 days, enhancing microbial safety while slightly affecting texture. These results support the product's suitability for short-term retail and household storage. To further optimize shelf-life, future research should investigate modified atmosphere packaging, natural preservatives, and accelerated stability testing.



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5.2 Conclusion

In conclusion, the developed vegan muffin demonstrated nutritional adequacy, microbial safety, and potential for consumer acceptance. The formulation aligned with current trends in plant-based eating, offering a product that was both health-conscious and ethically produced. The findings of this study supported the feasibility of incorporating functional ingredients into vegan bakery items and provided a foundation for future research focused on sensory optimization, shelf-life extension, and nutrient fortification.

Among all trials, the final formulation emerged as the most successful due to its superior sensory scores, balanced nutritional profile, and confirmed microbial safety. It integrated traditional baking techniques with modern plant-based innovation, resulting in a clean-label, vegan-friendly product that meets consumer expectations for taste, texture, and health. The use of flax seed paste as a binder, controlled sweetness, and optimized baking conditions collectively contributed to its excellence. This product stands out not only for its technical performance but also for its alignment with ethical and sustainable food development goals.

5.3 Recommendations for future Research

To further optimize shelf-life, future research should investigate modified atmosphere packaging, natural preservatives, and accelerated stability testing.

To build upon the findings of this study, future research should explore several key areas to enhance the formulation, stability, and market readiness of vegan muffins:

5.3.1 Shelf-life Extension Strategies

Modified atmosphere packaging (MAP), vacuum sealing, and edible coatings may significantly improve microbial stability and product freshness. Natural preservatives such as citrus peel powder, rosemary extract, and fermented plant derivatives have shown promise in extending shelf-life while maintaining clean-label integrity (Shukla et al., 2024; Santos et al., 2025).

5.3.2 Protein Fortification

The modest protein content observed in the current formulation suggests potential for enhancement through the incorporation of legume-based isolates or seed proteins. Du et al. (2025) demonstrated that hempseed protein can elevate protein levels to 19.4% in vegan muffins without compromising sensory quality.

5.3.3 Sensory Optimization with diverse panels

Expanding sensory trials to include varied demographic groups—such as children, elderly individuals, and those with dietary restrictions—can provide broader insights into consumer acceptability. Structured hedonic testing across diverse populations may guide formulation refinement (Gonzalez Viejo et al., 2024).

5.3.4 Functional Ingredient Integration

Future studies may investigate the inclusion of antioxidant-rich ingredients such as beetroot powder, moringa leaf flour, or fruit pomace. These additions could enhance nutritional value, color stability, and health claims related to immunity and digestion (Sharma et al., 2024; Talwar et al., 2025).



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5.3.5 Texture and Structure enhancement

Instrumental texture analysis, including Texture Profile Analysis (TPA), can quantify parameters such as hardness, springiness, and cohesiveness. These metrics are essential for ensuring consistency and optimizing mouthfeel in plant-based baked goods (Rodrigues et al., 2024).

5.3.6 Glycemic index and metabolic impact

Assessing the glycemic response of vegan muffins through in vitro or clinical methods can validate their suitability for diabetic and weight-conscious consumers. Substituting dates with low-glycemic sweeteners such as stevia or erythritol may be explored (Nikam et al., 2023; Du et al., 2025).

5.3.7 Sustainability and Cost analysis

A life cycle assessment (LCA) and cost-benefit analysis could evaluate the environmental footprint and economic feasibility of scaling up the formulation. This aligns with the growing demand for sustainable and affordable plant-based products (Dwivedi et al., 2024).

5.3.8 Regulatory and Labeling compliance

Ensuring alignment with FSSAI and international vegan certification standards is critical for commercialization. Research into consumer perception of labels such as "vegan," "gluten-free," and "high-fiber" may inform packaging and marketing strategies (Cross, 2007; Shih, 2020).

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