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Combined Application of Stevia and Amylase Enzyme in Wheat Flour Buns

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Abstract

In the modern food industry, sugar reduction strategies in bakery products are highly relevant due to growing consumer demand for low-glycemic alternatives. However, the multifaceted technological functions of sucrose pose significant challenges when substituting it in yeast-leavened formulations. This study evaluates the combined effect of *Stevia rebaudiana*, as a natural high-intensity sweetener, and alpha-amylase, acting as an enzymatic modifier, in wheat flour bun production. The feasibility of complete sucrose substitution using stevia alongside varying doses of alpha-amylase was investigated. The rheological properties of the dough, fermentation kinetics, physicochemical characteristics, and the sensory profile of the final baked goods were systematically evaluated. The results demonstrated that the incorporation of alpha-amylase significantly mitigates the adverse quality effects induced by stevia, markedly improving loaf volume, crumb porosity, and textural attributes. The optimal formulation was achieved by replacing 100% of sucrose with stevia extract supplemented with 40 ppm of alpha-amylase, which successfully restored the dough stability and gas production capacity to levels comparable to the control.

Keywords: *Stevia rebaudiana*; amylase; Sugar substitution; Dough rheology; Fermentation kinetics; Functional baking.

1. Introduction

Excessive dietary sugar consumption is closely linked to global metabolic disorders, including obesity, cardiovascular diseases, and type 2 diabetes. Consequently, the development of low-calorie and low-glycemic food alternatives has become a priority for food technologists. In baking technology, sucrose serves not merely as a flavor modifier but also as a critical structural, hygroscopic, and biochemical component. It acts as the primary substrate for yeast (*Saccharomyces cerevisiae*) metabolism and participates in thermal browning via the Maillard reaction and caramelization, which define the crust color and aroma of the finished product [3, 5].

Stevia rebaudiana Bertoni is widely recognized as a safe, natural, zero-calorie sweetener containing steviol glycosides, which possess intense sweetness (200–300 times sweeter than sucrose). Beyond its sweetening capacity, stevia exhibits therapeutic potential, including anti-hyperglycemic, anti-hypertensive, and antioxidant activities [7]. However, the total substitution of sucrose with stevia in yeast-leavened doughs presents severe technological drawbacks. The absence of fermentable bulk sugars leads to a dramatic suppression of yeast activity, reduced gas production, impaired dough rheology, and a diminished volume, often accompanied by an undesirable bitter aftertaste [8].

To overcome these structural and fermentative limitations, enzymatic modification offers a promising clean-label approach. The introduction of exogenous alpha-amylase (EC 3.2.1.1) facilitates the endohydrolysis of alpha-(1,4)-glucosidic linkages in amylose and amylopectin. This enzymatic breakdown generates fermentable sugars (mainly maltose and maltotriose) directly from the flour starch matrix, thereby compensating for the lack of added sucrose and restoring yeast metabolic pathways [6].

While individual effects of sweeteners and enzymes have been documented, the synergistic application of a stevia-alpha-amylase binary system remains insufficiently explored in sweet bun formulations. Therefore, this study aims to investigate the combined effect of stevia extract and varying concentrations of alpha-amylase on dough proofing dynamics, farinographic characteristics, gas retention, and the overall quality attributes of the final product.

2. Materials and Methods

Premium-quality commercial wheat flour (moisture 14.0%, ash 0.55%, wet gluten 28.0%), compressed baker's yeast (*Saccharomyces cerevisiae*), commercial *Stevia rebaudiana* extract (high-purity steviol glycosides), industrial-grade fungal alpha-amylase (enzyme activity: 20,000 U/g), sodium chloride, commercial sucrose, vegetable oil, and distilled water were used in the formulations.

The dough was prepared using a straight-dough baking method. The control formulation consisted of wheat flour (100%), sucrose (10%), compressed yeast (3%), sodium chloride (1.5%), vegetable oil (4%), and water (determined by water absorption capacity). In the experimental formulations, sucrose was completely replaced by stevia extract (scaled to match the sweetness equivalence of the control). Fungal alpha-amylase was incorporated into the stevia formulations at concentrations of 20, 40, and 50 ppm (mg/kg of flour).

The parameters recorded included Water Absorption Capacity (WAC, %), Dough Development Time (DDT, min), Dough Stability (DS, min), and Mechanical Weakening/Degree of Softening .

Dough proofing dynamics and cumulative carbon dioxide ({CO}_) production were monitored using a standard rheofermentometer-type setup over a 90-minute fermentation period at

30circ\text and 85% relative humidity. Total gas production (mL) and gas retention capacity (%) were calculated.

Buns were baked at 210circ\text for 18 minutes. After cooling for 2 hours, the moisture content was determined by drying at 105circ\text to a constant weight (AACC Method 44-15A). Crumb porosity (%) was measured by the volumetric displacement method. Descriptive sensory analysis was performed by a trained panel using a 10-point hedonic scale scoring aroma, taste, and crust appearance.

3. Results and Discussion

The impact of complete sucrose replacement with stevia extract on dough proofing volume expansion was monitored over a 90-minute period (Table 1).

Table 1: Dough proofing dynamics under sucrose-to-stevia substitution conditions

Proofing Time (min)	Control Sample .%	Experimental Sample .%
30	45	20
60	85	42
90	120	58

As indicated by the data in Table 1, the total substitution of sucrose with stevia severely hindered the dough proofing process, resulting in a dramatic reduction of the final volume expansion from 120% (control) down to 58%. This inhibition is primarily driven by the metabolic starvation of *Saccharomyces cerevisiae*, as steviol glycosides are non-fermentable by standard baker's yeast [8]. This deficiency in fermentable mono- and disaccharides suppresses the glycolytic pathway of the yeast, limiting gas generation and dough expansion. To resolve this technological bottleneck, exogenous alpha-amylase was introduced as an enzymatic corrective agent in the subsequent phase.

The effects of combining stevia with various concentrations of alpha-amylase on the structural-mechanical and farinographic properties of the dough were investigated to establish optimal proportions (Table 2).

Table 2. Structural-mechanical characteristics of dough with alternative sweeteners and enzymatic modification

Sample Designation	Dough Development Time (min)	Water Absorption Capacity (%)	Dough Stability (min)	Mechanical Weakening (FU)
Control (100% Sugar)	2.5	58.0	8.0	60
Stevia + Amylase 20 ppm	2.9	59.5	7.2	75
Stevia + Amylase 40 ppm	2.8	59.2	7.5	70
Stevia + Amylase 50 ppm	3.4	60.8	6.9	90

Experimental data showed that the combined use of stevia and α-amylase slightly altered the water absorption capacity (WAC) of the flour, increasing it from 58.0% to 60.8% at higher enzyme levels. This is attributed to the intense hygroscopicity of stevia extracts and the structural alterations in starch granules [4].

The formulation containing stevia combined with 40 ppm of amylase exhibited the most balanced rheological profile, maintaining high mechanical stability (7.5 min) and a moderate softening degree (70). However, increasing the alpha-amylase dose to 50 ppm led to a prominent increase in mechanical weakening (90) and a drop in stability (6.9 min). Excessive endohydrolysis of starch by alpha-amylase over-produces dextrins, which dilutes and destabilizes the gluten matrix, resulting in a sticky, technologically unmanageable dough [2, 9]. Thus, 40 ppm was established as the critical threshold for maintaining structural integrity.

Since replacing sugar with stevia alters the nutrient medium for yeast, it was essential to determine how the selected enzyme concentrations affect carbon dioxide ({CO} generation and retention within the dough matrix (Table 3).

Table 3: Influence of stevia and amylase on dough gas production and gas retention capacity

Sample Designation	CO2 Production (mL)	Gas Retention (%)	Fermentation Rate
Control	1450	92	High
Stevia + Amylase 20 ppm	1300	88	Moderate
Stevia + Amylase 40 ppm	1380	90	High
Stevia + Amylase 50 ppm	1200	82	Moderate

The data outline that sugar reduction leads to a decline in yeast activity, directly diminishing \text{CO}_2 generation due to the lack of fermentable sugars. However, the addition of alpha-amylase effectively compensates for this by hydrolyzing damaged starch into maltose, providing an alternative nutrient substrate for yeast fermentation [6].

The sample with 40 ppm amylase closely approached the control values, achieving a total gas production of 1380 mL and 90% gas retention. Interestingly, the 50 ppm sample showed decreased gas retention (82%), which correlates with the farinographic findings: the over-degradation of starch weakens the gas-retaining gluten-starch matrix, allowing gas to escape prematurely [2].

To determine how these fermentation improvements translate into the final baked goods, the organoleptic and physicochemical parameters of the buns were evaluated (Table 4)

Table 4. Organoleptic and qualitative parameters of the finished baked products

Sample Designation	Moisture Content (%)	Porosity (%)	Aroma Score (Max 10)	Taste Score (Max 10)	Crust Color
Control	28.5	72	9.5	9.5	Light, bright golden
Stevia + Amylase 20 ppm	29.5	68	8.5	8.5	Intensive dark golden
Stevia + Amylase 40 ppm	28.7	70	9.0	9.0	Uniform golden
Stevia + Amylase 50 ppm	28.7	70	9.0	9.0	Golden

Based on these results, the formulation supplemented with 40 ppm of -amylase yielded the best technological performance. It successfully achieved a high porosity level (70%) and excellent sensory scores (9.0 for both taste and aroma). The enzymatic accumulation of reducing sugars via amylase action provided the necessary precursors for the Maillard reaction, resulting in a desirable uniform golden crust color, effectively eliminating the pale, unappealing crust typical of pure stevia-substituted bakery formulations [1, 5].

4. Conclusion

The total substitution of sucrose with *Stevia rebaudiana* extract negatively affects dough fermentation kinetics, farinographic stability, and the organoleptic properties of baked goods due to substrate starvation in yeast cells. The incorporation of exogenous fungal amylase effectively compensates for sugar deficiencies by continuously liberating fermentable maltose from the starch matrix.An optimal concentration of 40 ppm of -amylase combined with stevia extract establishes a balanced binary system, ensuring high gas retention, optimal crumb porosity, and desired crust coloration via the Maillard reaction.This established formulation model presents a highly viable approach for the commercial production of low-calorie, low-glycemic, and functional bakery products without sacrificing traditional quality attributes.

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სტევიისა და ფერმენტ ამილაზას კომბინირებული გამოყენება ხორბლის ფქვილისგან დამზადებულ ფუნთუშებში

ეკატერინე გობრონიძე

ტექნიკის მეცნიერებათა აკადემიური დოქტორი, ა(ა)იპ საქართველოს აგრარული უნივერსიტეტი, ჩაის სუბტროპიკული კულტურების და ჩაის მრეწველობის ინსტიტუტი

ანოტაცია: თანამედროვე კვების ინდუსტრიაში შაქრის შემცირების სტრატეგიები აქტუალურია, თუმცა მისი ტექნოლოგიური ფუნქციების მრავალფეროვნება მნიშვნელოვან სირთულეს ქმნის საცხობ პროდუქტებში ჩანაცვლებისას. წინამდებარე კვლევის მიზანია, სტევიის, როგორც ბუნებრივი მაღალი ინტესივობის დამატკბობლის და ფერმენტ ამილაზას, როგორც ფერმენტული მოდიფიკატორის კომბინირებული გავლენის შეფასება ხორბლის ფქვილისაგან დამზადებულ ფუნთუშებში. შესწავლილ იქნა შაქრის ჩანაცვლების შესაძლებლობა სტევიათი და ფერმენტ ამილაზათი. შეფასდა ცომის რეოლოგიური თვისებები, ფერმენტაციის კინეტიკა, მზა პროდუქტის ფიზიკო-ქიმიური მახასიათებლები. შედეგებმა აჩვენა, რომ ამილაზას დამატება მნიშვნელოვნად ამცირებს სტევიის გამოყენებით გამოწვეულ უარყოფით ეფექტებს, აუმჯობესებს მოცულობას, ფორიანობასა და ტექსტურას. ოპტიმალური ფორმულაცია მიღწეული იქნა შაქრის სტევიით ჩანაცვლებით და ფერმენტ ამილაზის გამოყენებით.

საკვანძო სიტყვები: სტევია, ამილაზა, შაქრის ჩანაცვლება, ფერმენტული კორექცია