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Original Article

Supplementation of Aloe Vera to Formulate the Fortified Bread by Managing its Nutritional Profile

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ABSTRACT

Aloe vera is the medicinal plant rich in bioactive compounds. It has a potential to cure many health related problems. The challenge for the food sector is to create novel food products with health promising properties. **Objective:** To fortify the bread with two different varieties of Aloe vera in different treatments in varying proportion (0, 10, 20, 30%). **Methods:** The raw material was analyzed and compositional results of proximate analysis of Aloe vera gel indicated the moisture content (98 ± 0.081, 98.5 ± 0.816), crude protein (0.39 ± 0.008, 0.34 ± 0.0081), crude fat (0.22 ± 0.081, 0.23 ± 0.016) crude ash (0.4 ± 0.081, 0.4 ± 0.081), crude fiber (0.59 ± 0.081, 0.31± 0.0816), and NFE (0.4 ± 0.163, 0.01 ± 0.0081) % for *Aloe barbedensis miller* and *A. barbedensis marlothi* respectively. The product was developed using different proportions of both varieties of Aloe vera. The proximate, mineral, color, texture and sensory properties of the Aloe vera fortified bread samples were evaluated. **Results:** Results of the proximate analysis showed that addition of 20% of *marlothi* variety of Aloe vera enhance the texture of bread. However, the sensory evaluation indicated that the 20% Aloe vera fortified bread was not significantly different from the bread produced from 100% wheat flour in terms of quality attributes evaluated in this study. **Conclusions:** Fortification of bread with Aloe vera gel increased the fiber content and macronutrient of conventional bread. In addition, the Aloe vera fortified bread was rich in minerals and also acceptable to consumers at 20% fortification level.

INTRODUCTION

Aloe vera is a drought-resistant, lance-shaped, pointy, juicy plant that can survive for more than 7 years without water [1]. There are more than 300 types of aloevera, but the most popular and well-known is *Aloe barbadensis miller* [2]. The Aloe vera plant grows best in hot, dry regions like those found in deserts [3]. Aloe vera possesses more than 200 bioactive chemicals, giving it the name wonder plant [4]. The *liliaceae* family includes plants including *A. Barbadensis miller, A. Chinensis baker, A. Ferox miller, A. Perryi baker* and *A. Indica royle* [5]. Aloe vera gel has a low calorie count because of its high water content. In fact, given that a serving of 200 mL only provides 5 kcal [6]. As a result of its ability to eradicate or restrain viruses, bacteria, fungi and mold, Aloe vera compounds are known as antiseptics. A wide range of immune system illnesses and disorders are helped under control [7]. It has a lot of health benefits i.e., helps in digestion, builds immunity, detoxify and cleanse the digestive system, reduce inflammation,

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heal wound [8], cure heart disease, psoriasis [9], cell proliferation [10], and intestinal absorption [11]. Bread is the most popular non-indigenous cuisine and one of the most significant basic foods. There are numerous ways to serve bread, and it can be consumed as a snack or as an ingredient in other dishes. A baked product made using wheat flour is bread. There are numerous types of bread depending on its size, shape, weight, crust hardness, crumb cell structure, softness, and color [12]. Daily consumption of bakery items is high, and they play a crucial part in human nutrition [13]. Aloe vera also has been used to enhance the postharvest quality of fruit like sweet cherry [14], table grapes [15], blueberries [16] and kiwifruit slices [17]. According to the results of Vieira *et al.*, Aloe vera is the best coating for maintaining product's sensory qualities during storage as well as extending the post-harvest shelf life [17]. There are various kind of food products that have been fortified with Aloe vera gel and powder in which meat burgers [18], cakes [19], jelly [20], chocolates [21], marmalade [22], yoghurt [23], buttermilk [24], smoothies and Aloe vera tea [25] are included. Thus, fortification of wheat flour with Aloe vera gel could, therefore, significantly improve the nutritional quality of Aloe vera bread.

The objective of this study was to determine the effect of both varieties (*A. barbadensis miller and A. barbadensis marlothi*) of Aloe vera gel fortification on the nutritional profiling and sensory attribute. So, different treatments of 10, 20 and 30% from both varieties were performed with the bread.

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METHODS

Procurement of Raw Material

Both varieties of Aloe vera were purchased from the local nursery of Multan. All the chemicals and reagents used in this study were provided in the value addition lab of the Department of Food Science and Technology and Nutrient Analytical Laboratory, Central Lab System (CLS) of MNS-University of Agriculture Multan. For bread preparations, all raw materials were procured from local supermarket of Multan. This study was performed from March to June in 2023.

Proximate Analysis of Aloe Vera Gel

Aloe vera was processed by the steps, as hown in figure 1, for thorough analysis. The moisture percentage was determined by hot air oven method no. 44-15A, crude protein by kjeldhal apparatus method no. (46-10), crude fat content was determined by Soxhlet apparatus by method no. 30-25 [26]. Ash content was determined by Muffle furnace method no 08-01 and crude fiber was also measured by Muffle furnace method no. 32-10 . NFE and other analysis were done following the method of Shahrezaee *et al.*

$$\textbf{Moisture\%} = \frac{original\ sample\ weight\ (g) - sample\ dry\ weight\ (g)}{original\ sample\ weight\ (g)} \times 100$$

$$\textbf{Nitrogen\%} = \frac{\text{volume of 0.1N H2SO4 used} \times 0.0014 \times \text{vol.of dilution(250)}}{\text{weight of sample} \times 10} \times 100$$

$$\textbf{Protein\%} = \textbf{Nitrogen percentage} \times 6.25$$

$$\textbf{Fat\%} = \frac{(sample\ weight + thimble) - weight\ of\ fat - free\ sample\ after\ drying}{weight\ of\ sample} \times 100$$

$$\textbf{Fiber\%} = \frac{oven - dried\ weight\ of\ sample\ (g) - sample\ weight\ after\ ashing(g)}{weight\ of\ sample(g)} \times 100$$

$$\textbf{Ash\%} = \frac{ash\ weight\ (g)}{measure\ of\ sample\ size\ (g)} \times 100$$

NFE = 100- (Percentage of moisture + Percentage of Crude Fat + Percentage of Ash + Percentage of crude Fiber + Percentage of Crude Protein).

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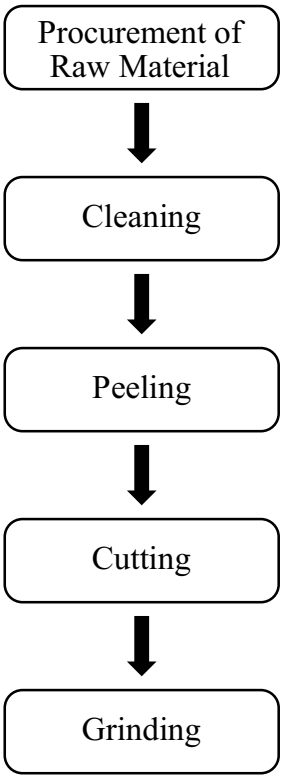

Figure 1: Flow Diagram of Aloe Vera Processing

Total Soluble Solids (TSS)

A digital refractometer was used to determine the total soluble solids in the gel.

Total Phenolic Content (TPC)

Total phenolic contents of gel was determined by a standard method.

2, 2-Diphenyl-1-picrylhydrazyl (DPPH) Assay

The antioxidant activity of aloevera was determined according to the procedure explained.

Minerals

The mineral profile of bread was measured using an atomic absorption spectrophotometer and flame photometer according to the Shahrezaee *et al.* Zinc, copper and magnesium were analyzed in both varieties of Aloe vera gel.

Preparation of Bread

Bread were prepared according to standard recipe by adding Aloe vera gel of both varieties according to the given treatment plan (Table 1).

Table 1: Treatment Plan for Bread Fortification

Treatment (g)	Wheat flour (g)	Eggs (g)	Oil (g)	Yeast (g)	*A. Barbadensis miller* (g)	*A. Barbadensis marlothi* (g)
T_0	100	50	10	2	0	0
T_1	90	50	-	2	10	0
T_2	80	50	-	2	20	0
T_3	70	50	-	2	30	0
T_4	90	50	-	2	0	10
T_5	80	50	-	2	0	20
T_6	70	50	-	2	0	30

Figure 2 shows the process of bread preparation.

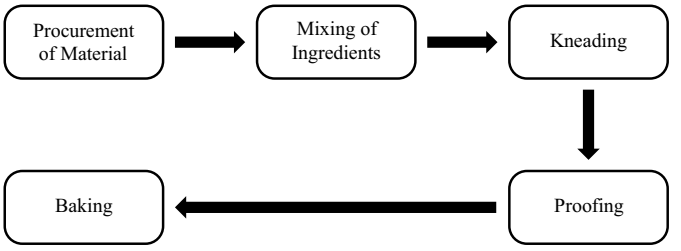

Figure 2: Bread Preparation Process

Data Interpretation

The data were collected as triplicate and subjected to statistical analysis using ANOVA under CRD. Beyond ANOVA, LSD was applied to check the significant difference between the treatments.

RESULTS

The results of chemical analysis are in table 2. The moisture content in *A. Barbadensis marlothi* was slightly higher than *A. Barbadensis miller*. Crude protein in *A. Barbadensis miller* was 0.39% and in *A. Barbadensis marlothi*, it was 0.34% showing slight difference. Ash content was almost same in both varieties of Aloe vera. There was no significant difference in fiber content in both varieties. The results indicated that total soluble solids were higher in *A. Barbadensis marlothi* as compared to *A. Barbadensis miller*. *A. Barbadensis miller* variety had higher amount of antioxidants than *A. Barbadensis marlothi* i.e., TPC was 0.204mg/L and DPPH decolorizing concentration was 3.778mg/L. Mineral analysis indicated the presence of high amount of beneficial mineral elements such as zinc, copper and magnesium.

Table 2: Chemical Analysis of Aloe Vera Varieties

Treatment	*A. Barbadensis miller* (g)	*A. Barbadensis marlothi* (g)
Moisture	98 ± 0.081^a	98.5 ± 0.816^a
Crude Protein	0.39 ± 0.008^a	0.34 ± 0.0081^b
Fat	0.22 ± 0.081^a	0.23 ± 0.016^b
Ash	0.4 ± 0.081^a	0.4 ± 0.081^a
Fiber	0.59 ± 0.081^a	0.59 ± 0.081^b
NFE	0.4 ± 0.163^a	0.01 ± 0.0081^b
TSS	0.4 ± 0.081^a	0.6 ± 0.081^a
TPC	0.204 ± 0.081^a	0.202 ± 0.008^a
DPPH	3.778 ± 0.008^a	1.645 ± 0.049^b
Zinc	0.7915 ± 0.081^a	0.7292 ± 0.0816^b
Copper	0.1646 ± 0.0085^a	0.0415 ± 0.002^b
Magnesium	0.358 ± 0.0085^a	0.407 ± 0.0012^b

Different letters with values (Mean ± SD) represent a statistically significant difference (p=0.05)

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Proximate Composition of Aloe vera Fortified Bread

The results for proximate composition of Aloe vera fortified bread are shown in table 3. Slight change in moisture content was due to the addition of Aloe vera gel. Aloe vera gel also contain elasticity property. This component was rich in fiber and gel had capability to bind the ingredients. The moisture percentage in T_0, T_1, T_2, T_3, T_4, T_5 and T_6 was 27.5 ± 0.081, 24.9 ± 0.92, 26.4 ± 0.081, 25.1 ± 0.163, 27.1 ± 0.124, 24.9 ± 0.124 and 24.4 ± 0.355 respectively. The highest value of moisture was observed in treatment T0 which is 27.5 ± 0.081. The fat contents of bread decreases as the concentration of Aloe vera increases. The fat content percentage in T_0, T_1, T_2, T_3, T_4, T_5 and T_6 was 7.18 ± 0.009, 6.94 ± 0.054, 7.17 ± 0.024, 7.84 ± 0.139, 7.1 ± 0.328, 7.51 ± 0.128 and 6.75 ± 0.171 respectively. Highest value of fat was observed in T_0 and T_1 respectively. Protein is an important macronutrient essential for various bodily functions. Value of protein for T_0, T_1, T_2, T_3, T_4, T_5 and T_6 was 5.76 ± 0.287, 5.81 ± 0.12, 6.07 ± 0.049, 6.1 ± 0.063, 6.14 ± 0.0309, 6.24 ± 0.0047 and 6.29 ± 0.047 respectively. The highest value of protein was found in T6 that is 6.29 ± 0.047. Ash content represents the inorganic mineral content of a sample after complete combustion. The ash content of Aloe vera can provide insights into its mineral composition. Value of protein for T_0, T_1, T_2, T_3, T_4, T_5 and T_6 was 2.21 ± 0.004, 2.1 ± 0.05, 2.12 ± 0.028, 2.24 ± 0.009, 2.31 ± 0.0047, 2.33 ± 0.020, 2.42 ± 0.008. Highest value of ash was found in T_5 that is 2.42%. The fiber contents of bread gradually increased with increase in Aloe vera gel concentration. There is a minimum variation between the treatments of Aloe vera bread. Value of fiber for T_0, T_1, T_2, T_3, T_4, T_5 and T_6 was 0.48 ± 0.008, 0.47 ± 0.008, 0.51 ± 0.016, 0.51 ± 0.004, 0.46 ± 0.012, 0.47 ± 0.012 and 0.58 ± 0.016 respectively. Highest value of fiber is in T_6.

Table 3: Proximate Analysis of Aloe Vera Fortified Bread

Treatment	Moisture	Fat	Protein	Ash	Fiber
T_0	27.5 ± 0.081^{a}	$7.18 \pm 0.009b^{c}$	5.76 ± 0.287^{b}	2.21 ± 0.004^{c}	0.48 ± 0.008^{bcd}
T_1	24.9 ± 0.92^{c}	6.94 ± 0.054^{c}	5.81 ± 0.12^{b}	2.1 ± 0.05^{cd}	0.47 ± 0.008^{cd}
T_2	26.4 ± 0.081^{ab}	7.17 ± 0.024^{bc}	6.07 ± 0.049^{ab}	2.12 ± 0.028^{d}	0.51 ± 0.016^{bc}
T_3	25.1 ± 0.163^{bc}	$7.8\ 4\pm 0.139^{a}$	6.1 ± 0.063^{ab}	2.24 ± 0.009^{bc}	0.51 ± 0.004^{b}
T_4	27.1 ± 0.124^{a}	7.1 ± 0.328^{bc}	6.14 ± 0.0309^{ab}	2.31 ± 0.0047^{b}	0.46 ± 0.012^{d}
T_5	24.9 ± 0.124^{c}	7.51 ± 0.128^{ab}	6.24 ± 0.0047^{a}	2.33 ± 0.020^{ab}	0.47 ± 0.012^{cd}
T_6	24.4 ± 0.355^{c}	6.75 ± 0.171^{c}	6.29 ± 0.047^{a}	2.42 ± 0.008^{a}	0.58 ± 0.016^{a}

Different letters with values (Mean ± SD) represent a statistically significant difference (P=0.05)

Sensory Analysis

The Aloe vera bread samples were coded and served to twenty judges. Members of the panel were MNS University of Agriculture Multan students, faculty and staff from the department of Food Science and Technology. The judges were asked to score the bread samples for color, shape, texture, sweetness, flavor, mouth feel and overall acceptability, using a nine (9) point's hedonic scale, where 1 to 9 represented dislike extremely and like extremely, respectively. Bread produced from 100% wheat flour (conventional bread) was used as control. The results are displayed in table 4.

Table 4: Mean Values for Sensory Attributes of Aloe Vera Bread

Treatment	Color	Appearance	Texture	Aroma	Flavor	Taste	Overall Acceptability
T_0	5.2 ± 2.0655^{a}	4.15 ± 0.87^{f}	4.17 ± 1.25^{e}	5.3 ± 1.309^{e}	5.4 ± 0.74^{f}	6.23 ± 0.77^{c}	6.23 ± 0.72^{d}
T_1	5.2 ± 1.7511^{a}	4.76 ± 0.88^{e}	5.3 ± 0.73^{d}	5.46 ± 0.71^{d}	4.46 ± 0.805^{g}	5.84 ± 0.74^{f}	5.3 ± 1.04^{g}
T_2	5.2 ± 1.43^{a}	5.23 ± 1.083^{d}	5.84 ± 0.83^{d}	5.46 ± 0.78^{d}	5.69 ± 0.92^{d}	5.53 ± 0.81^{e}	5.53 ± 0.71^{f}
T_3	5.2 ± 1.51^{a}	5.76 ± 0.952^{c}	6 ± 1.203^{d}	6.23 ± 1.62^{c}	5.61 ± 0.97^{e}	5.69 ± 0.95^{d}	5.92 ± 0.71^{e}
T_4	5.2 ± 0.67^{a}	7.3 ± 0.57^{b}	7.07 ± 0.85^{c}	7.15 ± 1.6^{b}	7.3 ± 0.85^{b}	7.46 ± 0.85^{a}	6.76 ± 0.61^{c}
T_5	5.2 ± 0.979^{a}	7.5 ± 1.14^{a}	7.61 ± 1.12^{b}	8.2 ± 0.77^{a}	8.07 ± 0.61^{a}	7.23 ± 1.02^{b}	7.61 ± 0.72^{a}
T_6	5.2 ± 0.83^{a}	7.7 ± 0.77^{a}	8 ± 0.44^{a}	7.2 ± 0.95^{b}	7.07 ± 0.92^{c}	7.23 ± 0.61^{b}	7 ± 0.88^{b}

Different letters with values (Mean ± SD) represent a statistically significant difference (P=0.05)

Color and Texture Analysis

The table 5 and 6 have depicted the results of crust and crumb color of all treatments recipes where no significant difference was found for the crust color for all L, A and B values, however crumb color was significantly different among treatments particularly with regard to L value.

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Table 5: Color Measurements of Aloe Vera Bread (Crumbs)

Treatment	L*	A*	B*
T_0	70.88 ± 0.00471^{c}	5.06 ± 0.016^{b}	24.25 ± 0.016^{bc}
T_1	68.97 ± 0.0081^{b}	5.31 ± 0.0047^{b}	23.79± 0.081^{a}
T_2	70.43 ± 0.081^{a}	5.53± 0.081^{b}	24.22 ± 0.0081^{ab}
T_3	68.75 ± 0.020^{a}	5.66 ± 0.089^{a}	24.43 ± 0.081^{c}
T_4	68.97 ± 0.0471^{a}	5.31± 0.081^{b}	23.79 ± 0.0081^{a}
T_5	70.88 ± 0.089^{c}	5.53 ± 0.046^{b}	24.25 ± 0.0094^{b}
T_6	70.43 ± 0.047^{b}	5.06 ± 0.016^{a}	24.22 ± 0.0047^{a}

Different letters with values (Mean ± SD) represent a statistically significant difference (P=0.05)

Table 6: Color Measurements of Aloe Vera Bread (Crust)

Treatment	L*	A*	B*
T_0	55.9 ± 0.416^{a}	11.73 ± 0.005^{c}	24.1 ± 0.081^{b}
T_1	57.33 ± 0.008^{c}	11.64 ± 0.008^{b}	24.33 ± 0.0081^{c}
T_2	60.61 ± 0.004^{b}	11.64 ± 0.008^{a}	24.21 ± 0.0081^{b}
T_3	60.75 ± 0.008^{d}	11.96 ± 0.008^{a}	24.03 ± 0.0047^{a}
T_4	60.61 ± 0.008^{c}	11.73 ± 0.081^{b}	24.33 ± 0.0047^{c}
T_5	55.9 ± 0.058^{a}	11.64 ± 0.0081^{a}	24.1 ± 0.057^{b}
T_6	57.33 ± 0.008^{b}	11.96 ± 0.0081^{c}	24.33 ± 0.081^{b}

Different letters with values (Mean ± SD) represent a statistically significant difference (P=0.05)

When investigated for texture (Table 7), T4 and T1 had lower values 6.53 ± 0.12 and 6.32 ± 0.18 respectively showing less brittleness in the bread.

Table 7: Texture Measurements of Aloe Vera Bread

Treatment	Texture
T_0	7.26 ± 0.34
T_1	6.32 ± 0.18
T_2	6.82 ± 0.16
T_3	7.18 ± 0.017
T_4	6.53 ± 0.12
T_5	7.17 ± 0.037
T_6	7.26 ± 0.34

The values are represented as Mean ± SD.

DISCUSSION

The results of moisture content of this study ranged 24.4 ± 0.355 – 27.1 ± 0.124 % and fat percentage ranged from 6.75 ±0.171 to 7.84 ± 0.139, that are in line with the findings of Soltanizadeh and Ghiasi-Esfahani, and Sedman *et al* [19, 27]. These results are significant. The fiber content of Aloe vera bread ranges from 0.46 ± 0.012 to 0.58 ± 0.016and protein content varied from 5.76 ± 0.287 to 6.29 ± 0.047similar to the findings of Palve *et al.*, and Bolarinwa *et al* [21, 28]. The ash content of Aloe vera bread are similar to the findings of Soltanizadeh and Ghiasi-Esfahani, and Bolarinwa *et al* [19, 28]. The results of fiber and ash are non-significant and for protein, results are significant compared to both studies. Any food's color can serve as a sign of doneness and is linked to variations in food color and flavor. Consumers use color as a key indicator of food quality. Present study was in line with the study of Manoharan *et al*, who found L* value ranged from 69.76 to 74.02, a* value ranged from 2.99 to 3.09 and color b* value is 23.03 to 24.35 in bread [29]. The L* value is non-significant while a* and b* color values are significant. The enzymatic browning reaction occurs when baking time and temperature are increased, causes l* and b* values of baked products to fall first, while a* value initially rise. The major causes of changes in the color of baked products are caramelization and the milliard reaction. Ingredients utilized, baking and chemical reactions that occur during baking can all affect the color of a bakery product. Color, along with texture, fragrance and flavor, is a major feature of bakery items that influences consumer choice. Present result resembled with the result of Manoharan *et al.*, who found l* value ranged from 60.39 to 61.09, a* value ranged from 11.45 to 12.19 and b* value ranged from 23.67 to 24.21 in Aloe vera bread [29]. Variation might be due to difference in variety, environmental conditions and treatments. The texture analysis of bread calculated in this study is similar to that reported by Manoharan *et al.*, and Shin *et al* whose findings ranged from 6.17 to 7.47 [29, 30]. The first sense that the customer experiences and uses to accept or reject food is the look of the meal, which is mostly controlled by surface appearance. Results of current study was in line with the study of Manoharan, and Jayabalan and Karthikeyan, who found 4.5 to 6.0 appearance and aroma value 6.6 to 7.5 in bread [22, 29]. Flavor is a sensory experience that combines the senses of taste, odor, heat and cold, as well as texture or "mouth feel." Food's look is significant, but its quality and acceptance are ultimately determined by its flavor. Current study was resembled with the study of Manoharan *et al.*, and found 6.5 to 7.1 flavor value in bread [29]. Foods might be smooth or lumpy, soft or firm, mushy or crunchy. Food texture is essential for food satisfaction and acceptance. Results of current study agreed with the study of other researchers Manoharan *et al.*, who found 7.7 to 8.1 texture of bread [29]. Color can be the most important sensory quality in the food industry. Food color gives buyers an almost instantaneous sense of the quality, flavor, and freshness of a food. This affects a customer's decision to purchase that product rather than another that they perceive to be more appealing. Present study was similar with the study of the Manoharan *et al.*, and Jayabalan and Karthikeyan, who found 6.5 to 8.0 color value in bread [22, 29]. The degree to which the food is accepted by a consumer at any particular time is closely correlated with that degree. Since consumers choose foods with certain sensory qualities, these characteristics have the most role in deciding how well-liked the food is. Present study was similar with the study of Manoharan *et al.*, and Jafari and Hosseini Ghaboos, who found overall acceptability score 6.1 to 8.1 in bread [29, 31].

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CONCLUSIONS

The bread preparation involved using different ratios of Aloe vera gel as fortifying agent may be utilized as nutritious, practical foods to improve nutritional value and health. Statistical analysis of the obtained data showed significant results for color, texture, flavor and aroma. Overall, the study elaborated the potential of Aloe vera as a medicinal plant rich in bioactive compounds. The development of Aloe vera fortified bread using different ratios of the two Aloe vera species provided insights into their effects on the nutritional composition and sensory characteristics of the bread. So we can conclude that the inclusion of Aloe vera enhanced the bread's texture and increased consumer acceptance. The addition of 20% Aloe vera variety i.e., *A. Barbadensis marlothi* bread was preferred by the judges than the other variety. Addition of Aloe vera gel did not affect the acceptability of bread

Authors Contribution

Conceptualization: NR, MS, BS
Methodology: NR, NB, AN
Formal analysis: AH, AN, MS, MK, MK
Writing- review and editing: NR, SM, MK, AM, BS

All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest

The authors declare no conflict of interest.

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