



NUTRITIONAL, ANTIOXIDANT AND SHELF LIFE OF STEVIA BASED MIXED FRUIT BAR

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Abstract: The aim of the study was to develop stevia based mixed fruit bar and to assess nutritional, antioxidant and storage stability of the product. Fruit bar was prepared from guava and ber at different ratio viz. (25:25, 60:10, 50:20, 40:30) with stevia at 2%-6%. Firstly, prepared fruit bar subjected to sensory analysis to evaluate the suitability for consumption. Then, nutritional composition and antioxidant activity of best fruit bar were analyzed. Nine point hedonic score system was used for sensory evaluation of prepared mixed fruit bar and treatment T₂ found to be the best (6% sugar+ 4% stevia+ 50% guava+ 20% ber+ 10% orange juice+ 8% lemon juice + 2% gelatine). Organoleptically best treatment was stored in Low polyethylene (LDPE) at refrigerator for 30 days. The stored sample was drawn at the time interval of 15 days for microbial analysis. The nutritional composition of best treatment T₂ and control treatment T₀ were analyzed and found all the nutrients were significantly higher in T₂ except calories. The storage study indicated that the mixed fruit bar was significantly safe for consumption at refrigerator temperature for 1 month. The TPC count of treatment T₂ (278.22mgGAE/g) was 42% higher than control treatment T₀ (195.7mgGAE/g). In the same line, DPPH activity of stevia based mixed fruit bar T₂ (68.85) was higher than control treatment T₀ (52.3).

Key words: Antioxidant Analysis, Ber, Guava, Fruit Bar, Nutritional Composition, Shelf life, Stevia.

1. INTRODUCTION

Bars are high in calorific value, good in taste, easy to pack and consume, which make them a convenient option for a busy families and children on the go. They are suitable for school lunches, after-school activities, and travel. However, the high sugar contents in bars is harmful for children and as well as for adults. High-sugar, high-fat, and calorie-dense snacks have been linked to the rising rates of obesity and related chronic diseases, such as diabetes and heart disease (De Ciccio, *et.al.*, 2016). A healthier bar can contribute to the fight against these health issues by providing a nutritious and less calorie snack option. There is a growing awareness and concern of parents for healthier bars with reduced sugar content. In this context the extensive study have been perform for the formulation of bar such as cereal bar, fruit bar, natural sugar substitute bar.

Recently, scientific interest arose for development of fruit base candy, bars and other confectionary fruit product due to its appealing color, taste and nutritional value. Fruits are rich in valuable nutritional competent includes sugar, fiber, vitamins, minerals and bioactive compounds that have therapeutic values. Although, they perishable

and many of them are seasonal which affect its availability throughout the year. Therefore fruit based processed fruit product need to develop several products. Fruit bar is a confectionary product prepared by drying fruit pulp and mixing it with sugar and pectin. Now a day's fruit bar are more commonly known as fruit leathers. Ber and Guava are tropical and seasonal fruit and have good nutritional value. Guava, a tropical fruit with a distinctive flavor and aroma, boasts a range of nutritional and functional properties that make it a valuable addition to a healthy diet (Leite *et.al.*, 2006). Ber, also known as Indian jujube or *Ziziphus mauritiana*, is a small, sweet and tangy fruit renowned for its exceptional nutritional and functional properties. In China is known as "Chinese dates", "Tsao", or red dates. Once dried, it is known as "Sedra" in Arabic countries and the edible fruit is called "beg" or "Ennab"; and in India and Iran, it is known as "ber" (Rashwan *et al.*, 2020).

Nutritionally, Ber and Guava both are rich source of essential vitamins and minerals, particularly vitamin C and vitamin A. A mere handful of these fruits can provide a significant portion of the daily recommended intake of vitamin C, bolstering the immune system and promoting healthy skin. The presence of vitamin A contributes to good vision, supports immune function, and maintains healthy skin and mucous membranes (Seth *et.al.*, 2001). Furthermore, both are an excellent source of dietary fiber, which is vital for digestive health. This fiber aids in regulating bowel movements, preventing constipation, and supporting a healthy gut micro biome. There fiber content can also help manage weight and control blood sugar levels, making it a valuable addition to the diet of individuals with diabetes or those looking to maintain a healthy weight.

Beyond its nutritional benefits, both fruits are renowned for its functional properties. It possesses potent antioxidant compounds, including flavonoids and carotenoids, which combat oxidative stress and inflammation, potentially reducing the risk of chronic diseases. The fruit's high content of dietary fiber aids in cholesterol management and supports heart health by reducing the risk of cardiovascular conditions. The phytochemicals present in fruit also exhibit anti-inflammatory properties, contributing to overall well-being. Additionally, the fruit's low glycemic index makes it suitable for individuals seeking to control blood sugar levels.

High calorie and sugar with negligible vitamins, mineral and bioactive compounds make it unhealthier choice for children and adults. Presently, low calories sweeteners or natural sweeteners have been used in a wide variety of foods and drinks such as dairy products, jams, pickles, sauces, fruit preserves, chewing gum, cakes, ice- creams, puddings, chocolates,. (Cook *et.al.*, 2000). Stevia is a natural, calorie-free sweetener, which reduces the overall calorie content of the mixed fruit bar. Stevia is a natural sweet herbal plant from the *Chrysanthemum* family. It has a long history of safe use. Stevia-based bars are a great choice for individuals who need to control their sugar consumption, such as diabetes, over-weight and health conscious people (Singh *et.al.*, 2007). Therefore, present study undertaken to formulate stevia incorporated mixed fruit bar and to assess its nutritional and antioxidant activity.

2. MATERIALS AND METHODS

The present study was conducted in the Department of Food Nutrition and Public Health, Ethelind College of Community Sciences, Sam Higginbottom University of Agriculture, Technology & Sciences (SHUATS), Prayagraj.

2.1 PROCUREMENT OF RAW MATERIALS

The raw materials for the preparation of bar like guava, ber, orange, lemon were purchased from the local market of Naini, Mahewa area, Prayagraj then gelatin,sugar were purchased from Ahar mart in Mahewa, and Stevia leaf powder were taken from Bioved Research Institute, Allahabad.

2.2 PREPARATION OF MIXED FRUIT BAR

The fruits like guava and ber was washed and peeled. Then, the fruits were chopped into small pieces and were kept for steaming. After steaming, fruits were kept aside to cool down into room temperature then the fruits were taken to grind into smooth pulp. And for the extraction of the juice of lemon and orange: first cut lemon and orange then squeeze the fruit to take the entire juice using strainer so that the pulp particles can be separated. Concentrated sugar syrup was prepared with the addition of water. Then lemon and orange juice was added and was cooked for sometime then the pulp of the fruits was added. After cooking the pulp for sometime gelatin was added and was stirred continuously. Once the pulp is thick and concentrated it was taken out in a greased sheet (butter paper) then was cooled in a room temperature then it was cut into rectangular shape and stored.

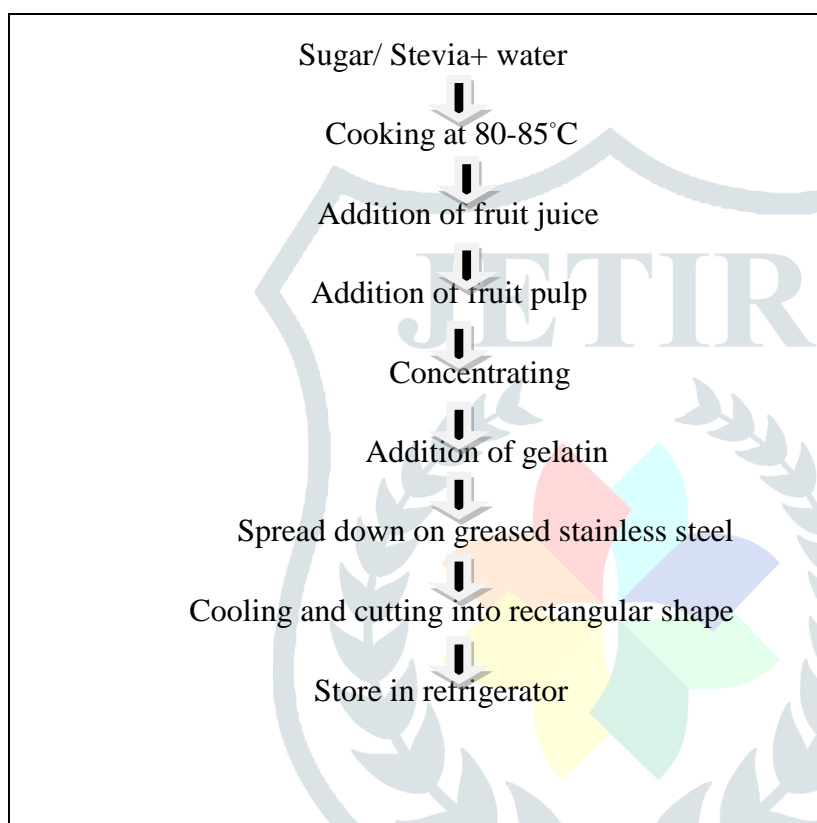


Figure 1: Flow Chart of Fruit Bar

Table 1: Treatments of the Mixed Fruit Bar

Ingredients	Treatments			
	T ₀	T ₁	T ₂	T ₃
SUGAR	30	8	6	4
STEVIA	-	2	4	6
GUAVA	25	60	50	40
BER	25	10	20	30
ORANGE	10	10	10	10
LEMON	8	8	8	8
GELATIN	2	2	2	2
TOTAL:-	100gms	100gms	100gms	100gms

2.3 SENSORY ANALYSIS OF THE DEVELOPED PRODUCT

Sensory evaluation of the prepared fruit bar was done according to the method of nine point hedonic scale by panel members of 10 judges. Sensory parameters for evaluation of the samples includes appearance, colour, aroma, taste, texture and overall acceptability (Lakshmi *et al.*, 2014).

2.4 NUTRITIONAL ANALYSIS

The nutritional composition includes moisture, ash, protein, iron, dietary fiber, fat, carbohydrates and calories of the mixed fruit bar were done by the following procedure of AOAC (2010), whereas Calcium and beta carotene was done by the procedure of Ranganna (1986).

2.5 ANTIOXIDANT ANALYSIS

The total phenolic compounds in each extract were assessed using Folin-Ciocalteu reagent following the method of Jayashree and Londonkar, (2014) and DPPH analysis of the mixed fruit bar was done by the following procedure of Krings and Berger, (2001).

2.6 SHELF LIFE OF MIXED FRUIT BAR

Organoleptically best treatment was stored in Low polyethylene (LDPE) pouches at refrigerator temperature (5°C). The samples were drawn periodically for 15-15 days and examine for microbial stability. The observation was taken in intervals of 15 days for 1 month.

2.7 STATISTICAL ANALYSIS

The data was statistically analyzed by using Analysis of variance (Two-way ANOVA or Two-way classification) technique. A significant difference between the treatments was determined by using CD (Critical Difference) test (Appendix). 'T' Test was performed for comparing the difference in the nutritional content between control and best treatment of the developed food products (Chandel *et al.*, 2006).

3. RESULTS AND DISCUSSION

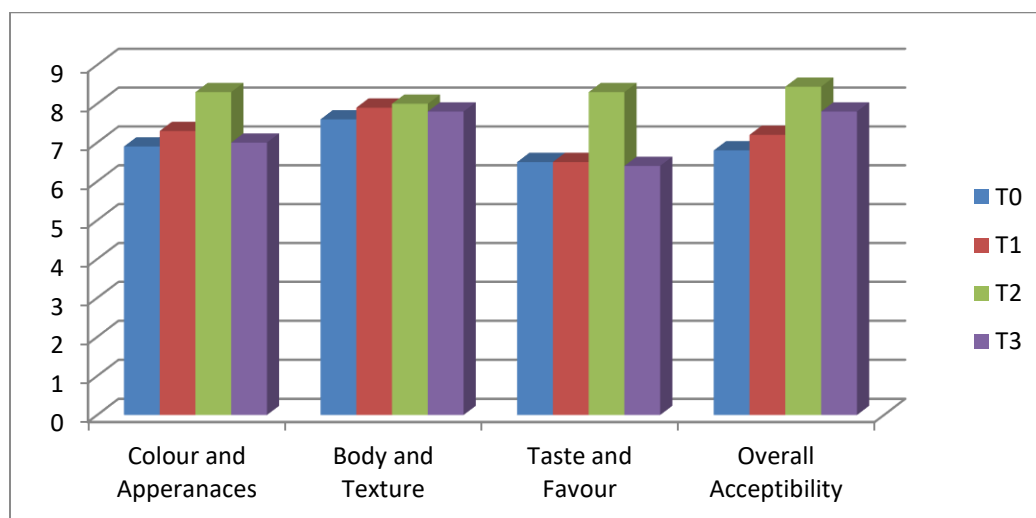
The mean sensory scores for colour and appearance shows that T₂ (8.3) scored highest in colour and appearance, followed by T₁ (7.3) and T₃ (7) whereas control treatment T₀ has least colour score of 6.9. It indicates that the treatment T₂ was most liked by the panel members, whereas other treatments were moderately liked regarding the colour and appearance of the mixed fruit bar (Sonuet *et al.*, 2022). Furthermore, increase in pulp of guava might be responsible for improvement of colour and appearance of the mixed fruit bar as it provide the light pink colour in the mixed fruit bar.

The mean sensory scores obtained for the mixed fruit bar in relation to body and texture. T₂ has the highest scores (8), followed by T₁ (7.9); T₃ (7.8) and T₀ (7.6). It indicates that T₂ was liked very much by the panel members, whereas T₀ was liked moderately regarding the body and texture of mixed fruit bar. Our finding reported that guava pulp has more roughness than ber pulp and increasing the ber pulp from 10% to 20 % and decreasing the guava pulp from 50%-40% improves the body and texture of mixed fruit bar. Although, further decreasing the guava pulp produce a detrimental effect of body as some roughness is required for texture of bar (Prabha *et al.*, 2022).

3.1 SENSORY ANALYSIS

The result of taste and flavor sensory scores revealed that T₂ has the highest scores (8.3), followed by T₀ (6.5); T₁ (6.5) and T₃ (6.4). It indicates that the taste of T₂ was liked very much by the panel members, whereas T₀ and T₁ were liked moderately. This might be because T₂ has an increasing amount of fruit pulp of guava and ber, which have enhance the flavour and taste of bar. The treatment T₂ attributed to the best combination of fruit pulp and it was also found that stevia have positive effect of taste of mixed fruit bar up to 10% (Prasad *et al.*, 2022).

Figure 2: Sensory Score of Mixed Fruit Bar



Our finding reported that T₂ has the highest overall acceptability with 8.44 scores followed by T₃ (7.8); T₂ (7.2) and T₀ (6.8). It indicates that T₂ (Guava: 50gms + ber: 20gms + sugar: 6gms+ stevia: 4 gms + lemon juice: 8gms + orange juice 10 gms + gelatin: 2gms) was liked very much by the panel members, and it was the best treatment among all the treatments (Bahadur *et. al.*, 2022).

3.2 NUTRITIONAL ANALYSIS

Proximate Analysis

Table 2 shows the moisture content of fruit bar was 13.39 (T₀) and 15 (T₂). Finding reported that moisture content of best rated mixed fruit bar was slightly higher than control T₀; although the difference was non significant at $P < 0.05$. The moisture content increased in the mixed fruit was due to the increase amount of guava pulp and ber pulp (T₀ < T₂). Less moisture content indicates the less effect of microbial activities. As the moisture content was increased with increase of fruit pulp of guava, the storage of the mixed fruit bar may also be less in the T₀ followed by T₂. Higher moisture content in the fruit bars increases the growth of undesirable microorganisms (Fontana, *et. al.*, 2000). Less moisture content in the fruit bar increases the shelf life of food (Figiel *et al.* 2006). It was also reported that the moisture content adversely affects the texture of fruit leather/bar (Huang *et. al.*, 2005). The ash content was found highest in T₂ (2.32) followed by T₀ (1.65). Our finding reported that the ash content in the mixed fruit bar increases with increasing the guava pulp and ber pulp (T₀ < T₂). The result attributed to the high content of minerals in guava. In this support, (Munhoz *et al.* 2010) found that average ash content (2.69 g.100 g⁻¹) of guava was higher than control. As shown in Table 2, there was slightly a significant difference in protein content of control and best rated treatment and found that protein higher in T₂ (1.7g/100g) than T₀ (1.5g/100g). The results obtained demonstrate that the fruit bar made with the largest concentrations of fruit pulp had the greatest protein levels, which is directly related to the high protein content of the fruit bar (Uchoa *et al.*, 2008). The nutritional values of the mixed fruit bar in which dietary fiber content values was T₀ (5.07g/100g) and T₂ (6.67g/100g). Dietary fiber was found highest in T₀ (Guava: 25gms + ber: 25gms + sugar: 30gms + lemon juice 8gms + orange juice: 10gms + gelatin: 2gms) followed by T₂(Guava:50gms + ber: 20gms + sugar: 6gms+ stevia: 4gm + lemon juice: 8gms + orange juice 10 gms + gelatin: 2gms). The dietary fiber content increased in the mixed fruit bar was due to the increase amount of guava pulp. Fruit used in making bar is rich in dietary fiber (Souza *et al.* 2008). The nutritional values of the mixed fruit bar reported that treatment T₂ (0.5gm/100g) have slightly higher fat content than control T₀ (0.3gm/100g). This attributed to less fat content in fruits and stevia. The carbohydrate content was T₀ (33.6g/100g) and T₂ (24.2g/100g) in control and best rated prepared mixed fruit bar. The carbohydrate content increased in the mixed fruit was due to the increase amount of guava and ber pulp (T₂ < T₀).

Carbohydrate content ranged from 24.2-33.6% among the fruit bar. High carbohydrate content of the fruit bar may be attributed to the addition of large amount of sugar in the preparation of fruit bar. Similarly, observed 77% total sugars in ber candy (Kaikadi *et al.* 2006). The calories were found highest in control T₀ (156.4 kcal/100kcal) than T₂ (106.4 kcal/100kcal). The calories content decreased in the prepared stevia based mixed fruit bar (T₂<T₀) was due to the increase amount of sugar.

Table 2: Nutritional Analysis of Prepared Mixed Fruit Bar

Nutrient	T ₀	T ₂	(Difference) T ₂ -T ₀	t.cal.	t.tab.	Results
Moisture (%)	13.39	15	1.2	2.79	2.447	NS
Ash (%)	1.65	2.32	0.67	3.1	2.447	S
Protein (g)	1.5	1.7	0.2	2.81	2.447	S
Iron (mg)	2.65	4.21	1.56	3.95	2.447	S
Dietary Fiber (g)	5.07	6.67	1.6	3.83	2.447	S
Fat (%)	0.3	0.5	0.2	1.22	2.447	NS
Carbohydrate (g)	24.2	33.6	9.4	4.38	2.447	S
Calories (Kcal)	156.4	106.4	50	46.15	2.447	S
Calcium (mg)	17.1	18	0.9	2.68	2.447	S
Beta Carotene (mcg)	120.14	157.24	37.1	339.52	2.447	S

Vitamin and Minerals

The mineral values of the mixed fruit bar reported that iron content was (2.65mg/100g) and (4.21mg/100g) in control treatment T₀ and best rated treatment T₂ respectively. The iron content increased in the mixed fruit was due to the increase amount of guava pulp. The fruits used in making fruit bar is rich in iron. The calcium content values was T₀ (17.1mg/100mg) and T₂ (18mg/100mg). Calcium was found highest in T₂ followed by T₀. The calcium content increased in the mixed fruit was due to the increase amount of fruit pulp. Guava used in making fruit bar is rich in calcium. (T₀<T₂). Beta carotene was found highest in T₂ (157.24 mcg/100g) followed by T₀ (120.14mcg/100g). Beta carotene content increased in the mixed fruit was due to the increase amount of fruits which are rich in beta carotene such as orange, lemon, guava and are used in making fruit bar. (T₀<T₂). The differences in the levels of lycopene and β-carotene may be partially explained by problems with bioavailability of lycopene from different food sources, as some types of fibers found in foods, such as pectin, can reduce the bioavailability of lycopene (Maguer *et. al.*, 2000).

3.3 ANTIOXIDANT ANALYSIS

The TPC of stevia based fruit bar treatment T₂ was found (278.22mg GAE/g) while control treatment T₀ (195.7 mg GAE/g). The higher TPC in best treatment might be attributed to high content of guava and stevia. Stevia is a medicinal plant rich in polyphenols (Kim *et. at.*, 2011) and in support (Bender *et. al.*, 2018) reported the total

phenolic compounds in extract of stevia leaf. Furthermore, fruit such as guava and ber rich in polyphenol and might be responsible for its higher total phenolic compounds.

Figure 3: Total Phenolic Content of Mixed Fruit Bar:

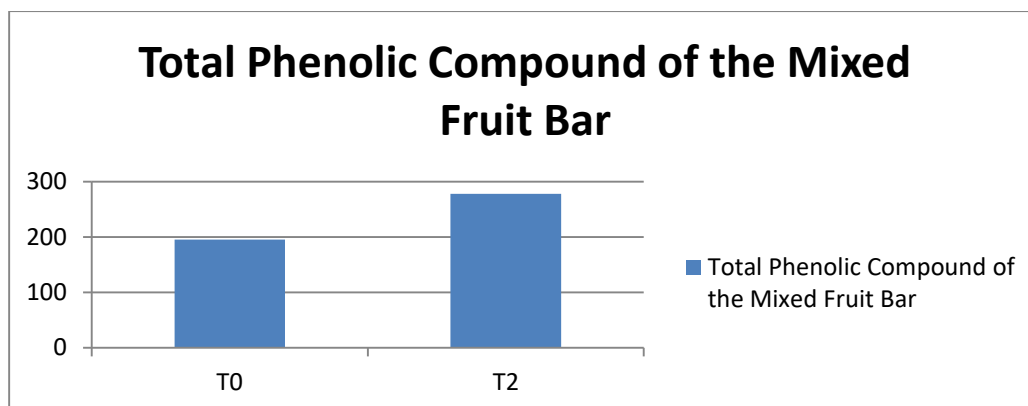


Figure 4: DPPH content of the Control and Best Treatment in Mixed Fruit Bar

The mean value of DPPH radical scavenging activity of fruit bars were presented in figure 5. The DPPH activity of stevia based mixed fruit bar treatment T₂ (68.85) was higher than the control treatment T₀ (52.3). This might be due to the higher DPPH radical scavenging activity of fruits and stevia. Our results were in accordance with the finding of (Shin *et al.*, 2021). Similar results were found by (Srivastava *et al.*, 2019) in orange-guava fruit bar. It was also reported that the increased antioxidant activity might be due to the presence of increased levels of total phenolic, flavanoids content in fruits and stevia (Yen *et al.*, 2022).

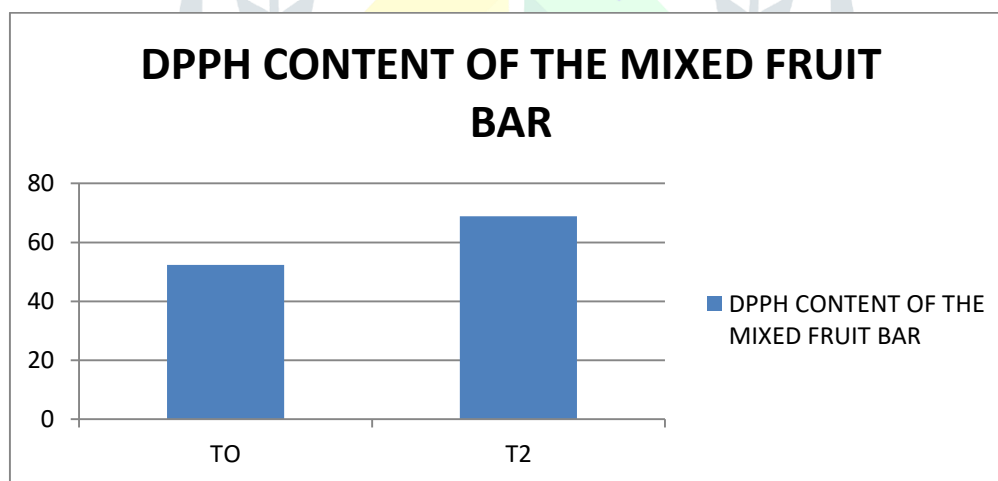


Figure 5: Analysis of Shelf life of Prepared Mixed Fruit Bar

3.4 SHELF LIFE OF MIXED FRUIT BAR

Our finding reported that there was no growth of microbes at zero days of storage. With increasing the storage period the total plate count (TPC) and Yeast and mold (Y/M) count increases. Although, there was no growth of coliforms were observed during one month of storage. This implies that the products are safe to consume and hygienically prepared. Less microbial growth was observed in the prepared fruit bar samples as compared to control stored in refrigerator. Our study reported that in 15 days, the TPC of the fruit bar was (4.7×10^1 CFU/g) in T₀ and (2.7×10^1 CFU/g) in T₂. Whereas, on the storage of 30 days T₀ was observed (2.3×10^2 CFU/g) and T₂ was

(1.5×10^2 CFU/g). It was observed that Y/M count in the control treatment (T_0) and best treatment (T_2) was nil in 15 days and the 30 days of storage it was reported (4.8×10^1 CFU/g) in T_0 and (2.5×10^1 CFU/g) in T_2 . It may be due to the high amount of stevia present in the fruit bar as it has antimicrobial properties which may have controlled the growth of microbes in the fruit bar (Shaikh *et al.*, 2015).

	T₀ (DAY 15)	T₂ (DAY 15)	T₀ (DAY 30)	T₂ (DAY30)
Total plate count (TPC) - CFU/g	4.7×10^1	2.7×10^1	2.3×10^2	1.5×10^2
Yeast / Mould (CFU/g)	ND	ND	4.8×10^1	2.5×10^1
Coliform	0	0	0	0

In consistent to the present study, (Kourany *et al.*, 2017) reported that molds and yeasts were not detected in their fruit bar while the total bacterial counts showed gradual negligible increases during two months of storage period stored in refrigerator. However, the acceptable amount of microbes (yeast and mould) was observed at the end of 15 and 30 days of storage, which were negligible in number and safe to consume according to US Food & Drug Administration. As per (FDA 2001) guidelines, the total microbial count should be less than 1×10^4 cfu/g and the total yeast/mold count is 10^2 cfu/g. Therefore, the fruit bar prepared with different blended ratios of guava and ber pulp was highly stable and safe from consumption point of view.

4. CONCLUSION

Our finding reported that pulp of guava and ber could be utilized to prepare the bar. Further, we found that stevia can replace the processed sugar and minimize the use of sugar from 30% to 4%. Among the different treatment, treatment T_2 (Guava: 50gms + ber: 20gms + Sugar: 6gms+ stevia: 4gm + lemon juice: 8gms + orange juice 10 gms + gelatin: 2gms); was most liked on the basis of sensory attributes. Nutritional analysis of fruit bar reported that best rated treatment T_2 have higher content of ash, protein, iron, dietary fiber, carbohydrate, calcium and no significant difference were found in moisture content and fat. The TPC and the DPPH activity of stevia based mixed fruit bar treatment T_2 was higher than the control treatment T_0 . The study reported that there was no growth of microbes at zero days of storage. With increasing the storage period, the total plate count (TPC) and Yeast and mold (Y/M) count increases. However, the product was safe to consume for one month of storage period. There was no growth of coliforms were observed during one month of storage. This implies that the products are safe to consume and hygienically prepared.

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