

Research Article

Extraction, Purification, Optimization, and Application of Galactomannan-Based Edible Coating Formulations for Guava Using Response Surface Methodology

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Galactomannan from fenugreek and guar seeds were extracted, purified, and used in edible coatings, optimized via response surface methodology. The results showed that the emulsifying capacity and stability of fenugreek galactomannan (FG) and guar galactomannan (GG) increased with increase in the concentration of galactomannan up to 0.5–1%. The average optimized values of FG and GG in edible coatings were predicted to be 1.71% and 2.11% for weight loss, 0.72% and 2.14% for firmness, 1.02% and 1.44% for TSS, 0.83% and 1.36% for pH, 1.03% and 1.44% for acidity, respectively. Significant decrease in weight loss and maximum retention of firmness was observed in coated guava. The TSS increased up to a certain storage period in all treatments and decreased as the storage period progressed, whereas pH exhibited an increasing trend while a significant decrease in acidity was observed. The findings revealed that the shelf life of guava could considerably be improved by incorporating 1.24% galactomannan from GG and 1.01% from FG in the edible coating.

1. Introduction

Guava (*Psidium guajava* L.) is a well-renowned fruit in Pakistan [1]. Guava quickly softens and thus has poor shelf stability that is susceptible to shipping and storage damage [2–4]. The guava's shelf life varies with the ambient temperature from three to five days [5]. Between harvest and consumption, significant losses are rendered in the quality and quantity of fruits and vegetables [6]. Food packaging innovations can help alleviate changing market demands such as consumer choice for safe and high-quality food items, and the decrease in environmental adverse effects of food packaging [7]. The need for a replacement for synthetic packaging with bio-based polymers is important.

Edible coatings can significantly increase shelf life and enhance food quality by the reduction in weight and

moisture loss, creating barrier properties against oil, gas, aroma, and flavor. The preservation of mechanical, rheological attributes, food color, and appearance of food are also improved. It is an environmentally friendly invention [6, 8]. Edible coatings generate a protective environment around the fruits and vegetables [9].

Galactomannans, water-soluble heterogeneous polysaccharides found in many leguminous seeds and mainly consists of mannose and galactose. They differ from each other by the mannose/galactose ratio. Fenugreek, guar gum, locust bean gum, and alfalfa are the most common sources of galactomannans [10]. It finds applications in different sectors (pharmaceutical, textile, cosmetics, biomedical, and food) due to its versatile characteristics and nontoxic effect. In the food industry, galactomannans are extensively used in film/coating formulation, dietary,

powdered and baby foods, soups, seasoning, sauces, meat, bakery, and dairy products [11].

Extraction and utilization of polysaccharides from plant and animal sources have gained significant attention in recent times because of their stability, biodegradability, and ecologically friendly. Guar gum is a natural polysaccharide [12]. Guar gum is extracted from the endosperm of guar seeds belongs to the family Leguminosae and is commonly known as guar, guaran, or cluster bean [13]. The chief suppliers of guar throughout the world are Pakistan and India, where 80% of the total guar is produced [14]. Guar gum is expensive and forms a viscous gel in cold water, so it is extensively used in different food products and industries as a stabilizer as well as an emulsifying and thickening agent [15].

Fenugreek (*Trigonella foenum-graecum*) is a popular spice consumed globally and commonly known as “Methi” [16]. It is widely grown in Pakistan, Turkey, China, Egypt, India, and the Mediterranean [17]. The seeds of fenugreek are rich in gum, fiber, alkaloids, flavonoids, saponins, and volatile compounds [18]. The plant seeds are a good source of polysaccharides [19]. In the past, fenugreek use in the manufacture of food has been increased by its emulsifying, thickening, and stabilizing properties in many food products [20].

The ripening process continues in guava even after detaching from trees and respire at a higher rate due to climacteric in nature. Due to short shelf life, marketing and subsequent storage of guava are difficult. Therefore, some treatments that can extend shelf life and conserve the quality of guava fruits are required (Anjum et al.) [21]. The study of postharvest physiology is of immense importance. The shelf life of fresh fruit is short at room temperature without any pre or posttreatment during storage. Reduction in losses and waste of fresh guava is essential for the fact that this fruit provides vital nutrients. Keeping in view the above-mentioned facts, current research work was carried out to improve the quality characteristics and extend the shelf life of local variety (Gola) of guava during the storage period by optimization of polysaccharides levels in edible coatings through the response surface methodology (RSM). Thus optimum galactomannan-based coating formulation was the main objective of the study and to study the effect of coating on quality attributes as well as shelf stability of guava.

2. Materials and Methods

2.1. Raw Materials. Fenugreek and guar seeds were purchased from the local market of Sargodha, Punjab, Pakistan. Freshly picked guava cv gola, for coating purposes, was purchased from guava orchard at Sargodha district, Punjab province (Figure 1). The ripe fruits were selected along with the stem, uniform size, color, free of any physical damage and fungal infestation, further followed by manual sorting and grading in the laboratory. Olive oil (extra light) and glycerin were purchased from the market at Sargodha. Chemicals including ethanol, isopropanol, and NaCl were purchased from Sigma-Aldrich, Germany available in the local market.

2.2. Extraction and Purification of Polysaccharide from Guar and Fenugreek Seeds. The seeds of guar and fenugreek were soaked separately in water overnight at room temperature. The polysaccharide extraction and purification were carried out according to the method described by [19]. Cleaned whole seeds were crushed and immersed for 24 hrs at 50°C in a 5% salt solution having 3 pH adjusted by acetic acid. The polysaccharides were extracted separately by muslin cloth. Crude gums were purified by the addition of IPA spirit (a blend of 10% isopropanol and 90% ethanol) in a ratio of 3:1 by volume with continuous stirring followed by centrifugation at 6000 rpm for 7 min. The white precipitate obtained from both gums was filtered by muslin cloth. The pomace of seeds was again immersed in a 5% salt solution of 3 pH until the maximum gum was extracted following the purification procedure. The purified polysaccharides were dried in an oven at 50°C for 24 hrs. The purified dried polysaccharide was weighed and stored in airtight jars at a cool dry place for further utilization and analysis (supplementary Figures 1–6).

2.2.1. Emulsifying Properties. The emulsifying capacity (EC) and stability of the polysaccharides were determined according to [19] with some minor modifications. Suspension (60 ml) of each gum was prepared with an increase in the galactomannan concentration (0.5, 0.75, 1.0% w/v), at the same commercial olive oil level (6 ml) homogenized at 10,000 rpm for 1 min. The suspensions were then centrifuged at 1300 rpm for 5 min. EC was calculated as

$$\text{emulsion capacity (\%)} = \frac{\text{emulsion volume}}{\text{total volume}} \times 100. \quad (1)$$

Emulsion stability (ES) against high temperatures were determined in the emulsions that were heated in a water bath at 80°C for 30 min and centrifuged at 1300 rpm for 5 min. The ES was calculated as

$$\text{emulsion stability (\%)} = \frac{\text{final emulsion volume}}{\text{initial volume}} \times 100. \quad (2)$$

2.3. Edible Coating Formulation. The coatings were prepared following the protocol of Vishwasrao and Ananthanarayan [2] with minor modifications. Purified polysaccharides extracted from fenugreek and guar seeds were used in the formulations of coating as a source of thickening, gelling, and stabilizing agent, glycerol as a plasticizer, olive oil as hydrophobic phase, and distilled water to make the edible coating. Measured amounts of guar galactomannan (GG) and fenugreek galactomannan (FG) powder were dissolved in 100 ml distilled water according to the treatment plan created by the central composite design and stirred by hot plate magnetic stirrers to form a gel, and then emulsifying agent (glycerol) was added. After the addition of oil, the mixture was homogenized for 5 min to get physically and chemically stable, inert, and nongreasy emulsion (Table 1). Emulsions prepared were cooled at room temperature and were kept in clean, dried, and airtight glass bottles for further application for one week.



FIGURE 1: The geographic location of procurement of guava fruits.

TABLE 1: Edible coating formulations from fenugreek galactomannan and guar galactomannan through central composite design.

| Treatments | Fenugreek galactomannan (g/100 ml) | Guar galactomannan (g/100 ml) | Olive oil (ml/100 ml) | Glycerin (ml/100 ml) | Distilled water (ml/100 ml) |
|------------------|------------------------------------|-------------------------------|-----------------------|----------------------|-----------------------------|
| EC ₀ | | | Control | | |
| EC ₁ | 1.00 | 1.5 | 5 | 0.3 | 92.2 |
| EC ₂ | 1.00 | 0.80 | 5 | 0.3 | 92.9 |
| EC ₃ | 0.30 | 1.50 | 5 | 0.3 | 92.9 |
| EC ₄ | 1.00 | 1.50 | 5 | 0.3 | 92.2 |
| EC ₅ | 1.00 | 2.20 | 5 | 0.3 | 91.5 |
| EC ₆ | 1.71 | 1.50 | 5 | 0.3 | 91.49 |
| EC ₇ | 1.00 | 1.50 | 5 | 0.3 | 92.2 |
| EC ₈ | 0.50 | 1.00 | 5 | 0.3 | 91.2 |
| EC ₉ | 1.00 | 1.50 | 5 | 0.3 | 92.2 |
| EC ₁₀ | 1.00 | 1.50 | 5 | 0.3 | 92.2 |
| EC ₁₁ | 1.50 | 1.00 | 5 | 0.3 | 92.2 |
| EC ₁₂ | 0.50 | 2.00 | 5 | 0.3 | 92.5 |
| EC ₁₃ | 1.00 | 1.50 | 5 | 0.3 | 92.5 |
| EC ₁₄ | 1.5 | 2.00 | 5 | 0.3 | 91.2 |

2.4. Preparation of Fruit Samples. Selected guavas were washed, rinsed, dried, and dipped in chlorinated water (chlorine level up to 150 ppm). After that guavas were divided into ten lots, each carrying an equal number (15 No) of fruits, and were coated by emulsion, prepared according to the treatment formulation except EC₀ (control), via the dipping method. Each fruit was dipped for one minute, twisted, and placed in baskets, then allowed to dry for 20 minutes at room temperature. Before dipping guava in the solution, the mixture was homogenized to get a uniformly dispersed emulsion. The coated guavas were placed in baskets at the storage temperature of $24 \pm 5^\circ\text{C}$ and stored for about 20 days.

2.5. Physico-Chemical Analysis. The weight of each coated and uncoated fruit was recorded by the electronic weighing balance, on 1st day of the experiment and after every 5 days interval for 20 days and the weight loss percentage were calculated. The fruit firmness was measured by using a penetrometer (model GY-2, Walfront, USA) and expressed as kg/cm^2 . A refractometer (RHB-32 ATC model, China)

was used for the determination of total soluble solids (TSS) of the guava fruit juice and expressed as Brix. acidity (acetic acid) was determined by titrating the juice against 0.1 M sodium hydroxide solution and six drops of phenolphthalein until the color changed to faint pink color. The volume of NaOH used was noted to get the value of acid (in grams per 100 mL). A digital calibrated pH meter (PH-8414 model) was used to measure the pH of juice according to the standard method [22]. The experiment was performed in triplicate; three fruits were taken each time after every five days.

2.6. Statistical Analysis. Multiple regressions to fit second-order polynomial equation through response surface methodology was applied to estimate the responses of fenugreek and guar gum used as independent variables during storage. Central composite design was used to optimize the levels of independent variables using Minitab 16 statistical software. Response surface graphs were generated to see the effects of independent variables on dependent variables. The data were analyzed by Nadeem et al. [23].

3. Results and Discussion

3.1. Emulsifying Properties. Due to high surface energy on the contact surface of oil and water molecules, thermodynamic emulsions are unstable and can result in the complete separation of two immiscible layers by coalescence; therefore, the stability of emulsions was examined. Emulsions prepared with high concentrations of galactomannan have been found to be more stable and have higher emulsion capacity [19].

Emulsions containing various concentrations of galactomannan (0.5, 0.75, 1.0% w/v) showed pronounced emulsifying property which increased with increasing galactomannan concentration up to 1.0% but the FG showed slightly lower than GG. The emulsion stability (ES) and emulsion capacity (EC) of guar galactomannan and fenugreek galactomannan are shown in Table 2. GG provided an excellent emulsion stabilization effect during two weeks of storage period at 25°C as compared to FG. Moreover, emulsions with 0.5% of GG and FG could even maintain the emulsification stability up to 94% and up to 97% over two weeks, respectively.

As the concentration of polysaccharides increases, the globules become more strongly enclosed in polymeric conformation. Higher polymer concentrations provide good viscosity to the aqueous phase, which further prevents the mobility of the globules and slows their amalgamation. The emulsifying properties of the hydrocolloids are either due to their interface behavior or their viscosity modifying properties that further inhibit the contact of the globules which also allows more time for the polymer to be absorbed in the interface [24].

3.2. Physico-Chemical Properties. Polysaccharides from guar galactomannan and fenugreek galactomannan were used in different levels to increase the shelf-life of guava. The effect of fenugreek and guar galactomannan on the physicochemical properties of guava fruit during storage (at 0, 5, 10, 15, and 20 days) was assessed. The RSM was applied to estimate the responses of fenugreek and guar galactomannans used as independent variables during storage.

3.3. Weight Loss. Weight loss in guava during ripening is due to both transpiration and respiration. Weight loss results in textural changes and surface shrinkage that adversely affects the shelf life of climacteric fruits and vegetables. The models were developed for weight loss of guava fruit as affected by independent variables during 20 days storage. The results depicted in Table 3 and Figure 2 revealed the significant effect of fenugreek galactomannan and guar galactomannan on weight loss in guava fruits during 20 days. The linear terms of FG and GG have a statistically significant ($p < 0.05$) effect for weight loss during the storage period. The FG^2 and GG^2 quadratic terms are found significant at 5, 10, 15, and 20 days storage intervals. The coefficients of determination (R^2) were studied as above 90% at most of the storage intervals, therefore it could be assured that models are well fitted and the coefficients of polynomial equations were calculated by

the equations given at the top of each graph (Figure 1). Optimized average values of fenugreek and guar galactomannan in the edible coating were found to be 1.70% and 2.10%, respectively. A decrease in weight loss was observed in edible-coated guava fruits during storage intervals. These results were found parallel to the findings of previously reported studies [25, 26]. Edible coatings reduced the respiration rates, water loss, and oxidation reaction rates by making a semipermeable barrier against oxygen (O_2), carbon dioxide (CO_2), moisture, and solute movement. The cumulative loss in weight increased gradually in all the treatments by advancement in storage duration. Coatings act as a barrier to desiccation and lead to maintained fresh weight of fruits (Khaliq et al.) [27]. Increased concentrations of polysaccharides allowed the formation of a thick layer around the fruit surface which retains firmness by reducing the loss of moisture and gas permeability. In this work, physiological weight loss was probably lower in coated fruits due to inhibited desiccation.

3.4. Firmness. Firmness is associated with water content and metabolic changes that occur in fruits and it is an important parameter that affects consumer acceptability [28]. The models were developed for the firmness of guava fruit as affected by fenugreek galactomannan and guar galactomannan during 20 days storage (Figure 3). The statistical analysis by applying the analysis of variance technique to the full regression of model (Table 4) shows a significant effect of FG and guar galactomannan. However, linear and quadratic terms of FG and GG are observed to positively change the firmness of fruits at 5, 10, 15, and 20th day of storage intervals, whereas at 1st day of storage have no effect. When the interaction of these two terms ($FG * GG$) was studied, it was found negative for all storage intervals. The coefficients of determination (R^2) were more than 85%, indicating well-fitted response models. The data showed that FG and guar galactomannan contributed toward firmness in guava fruit at 5 to 20 days storage intervals (Figure 2). For good firmness, optimized average values of fenugreek galactomannan and guar galactomannan in the edible coating were predicted to be 0.72% and 2.14%, respectively. The results showed that the optimized formulation was effective in maintaining the firmness of the guava fruit. The above-given results of firmness were similar to previous study results [29]. Another research reported a few variations in texture changes during the storage period [30]. Moreover, the edible coating is helpful in maintaining firmness [31].

3.5. TSS. The regression coefficients of variables in models showed that fenugreek galactomannan and guar galactomannan did not contribute toward change in TSS in guava fruits at the start of storage, but after that contributed significantly from 5th to 20 days storage intervals. The effect of linear terms of FG and GG are statistically significant ($p < 0.05$) for TSS at all days of storage intervals except the start of the study (Figure 4 and supplementary Table 1). The X^2 quadratic terms are found significant at 5, 10, 15, and 20 days storage intervals, whereas, the quadratic terms for FG^2

TABLE 2: Emulsion capacity and stability of fenugreek galactomannan and guar galactomannan.

| Galactomannan | Concentration (g) (% w/v) | Emulsion capacity (%) | Emulsion stability (%) |
|---------------|---------------------------|-----------------------|------------------------|
| Fenugreek | 1.0 | 98.23 ± 0.15 | 97.17 ± 0.11 |
| | 0.75 | 96.73 ± 0.15 | 95.87 ± 0.15 |
| | 0.5 | 94.8 ± 0.1 | 94.57 ± 0.35 |
| Guar | 1.0 | 98.83 ± 0.15 | 99.06 ± 0.06 |
| | 0.75 | 97.13 ± 0.06 | 98.83 ± 0.15 |
| | 0.5 | 95.95 ± 0.02 | 97.5 ± 0.1 |

TABLE 3: Analysis of variance (sum of the square) for response surface model of weight loss of guava fruits during storage.

| SOV | Df | Days | | | |
|-------------------|----|----------------------|----------------------|----------------------|----------------------|
| | | 5 | 10 | 15 | 20 |
| Model | 5 | 22.8021* | 8.4648* | 22.5618* | 13.5515* |
| Linear | 2 | 1.6024* | 0.1901* | 0.0951* | 1.1588* |
| FG | 1 | 1.1915* | 0.1321* | 0.0882* | 1.1581* |
| GG | 1 | 0.4109* | 0.0580* | 0.0069* | 0.0006* |
| Square | 2 | 21.1907* | 8.2195* | 19.5255* | 8.5903* |
| FG * FG | 1 | 20.5353* | 0.3314* | 10.9630* | 1.1501* |
| GG * GG | 1 | 12.3077* | 6.4152* | 19.0467* | 7.7648* |
| 2-way interaction | 1 | 0.0090 ^{ns} | 0.0552 ^{ns} | 2.9412 ^{ns} | 3.8025 ^{ns} |
| FG * GG | 1 | 0.0090 ^{ns} | 0.0552 ^{ns} | 2.9412 ^{ns} | 3.8025 ^{ns} |
| Error | 3 | 0.6967 | 3.8930 | 11.6652 | 6.3300 |
| Total | 8 | 23.4988 | 12.3578 | 34.2270 | 19.8815 |
| R-square (%) | | 94.04 | 88.50 | 95.92 | 88.16 |

If $p > 0.05$, ^{ns}non-significant ($p > 0.05$); $p > 0.05$, *significant; and $p > 0.01$, **highly significant fenugreek galactomannan (FG), guar galactomannan (GG).

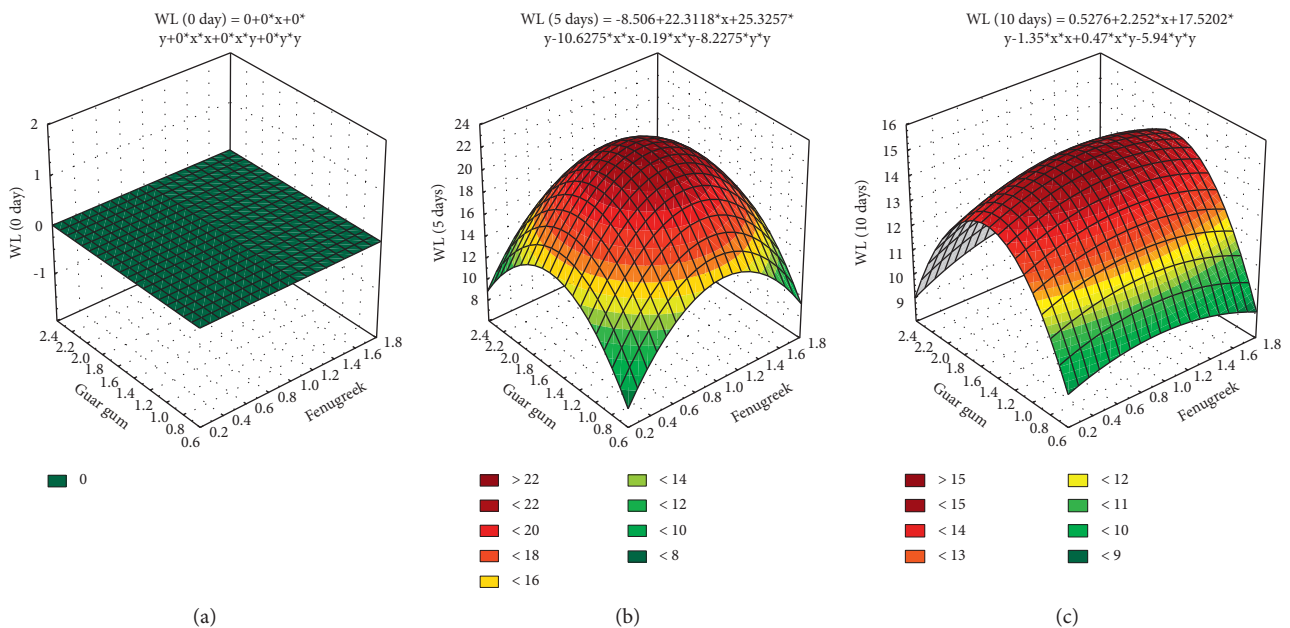


FIGURE 2: Continued.

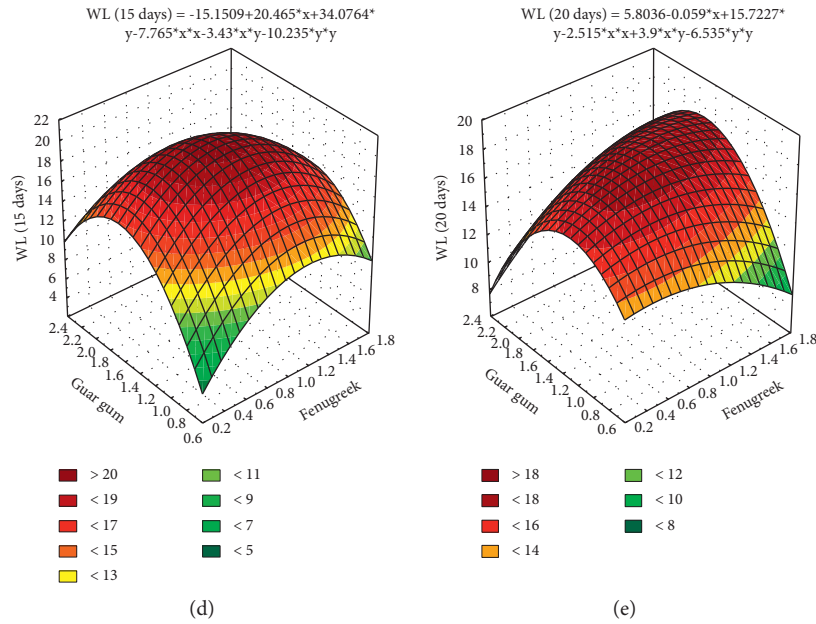


FIGURE 2: Response surface plot showing the impact of guar galactomannan and fenugreek galactomannan on weight loss of guava fruit during storage. (a) $WL(0 \text{ days}) = 0 + 0 * x + 0 * y + 0 * x * x + 0 * x * y + 0 * y * y$. (b) $WL(5 \text{ days}) = -8.506 + 22.3118 * x + 25.3257 * y - 10.6275 * x * x - 0.19 * x * y - 8.2275 * y * y$. (c) $WL(10 \text{ days}) = 0.5276 + 2.252 * x + 17.5202 * y - 1.35 * x * x + 0.47 * x * y - 5.94 * y * y$. (d) $WL(15 \text{ days}) = -15.1509 + 20.465 * x + 34.0764 * y - 7.765 * x * x - 3.43 * x * y - 10.235 * y * y$. (e) $WL(20 \text{ days}) = 5.8036 - 0.059 * x + 15.7227 * y - 2.515 * x * x + 3.9 * x * y - 6.535 * y * y$.

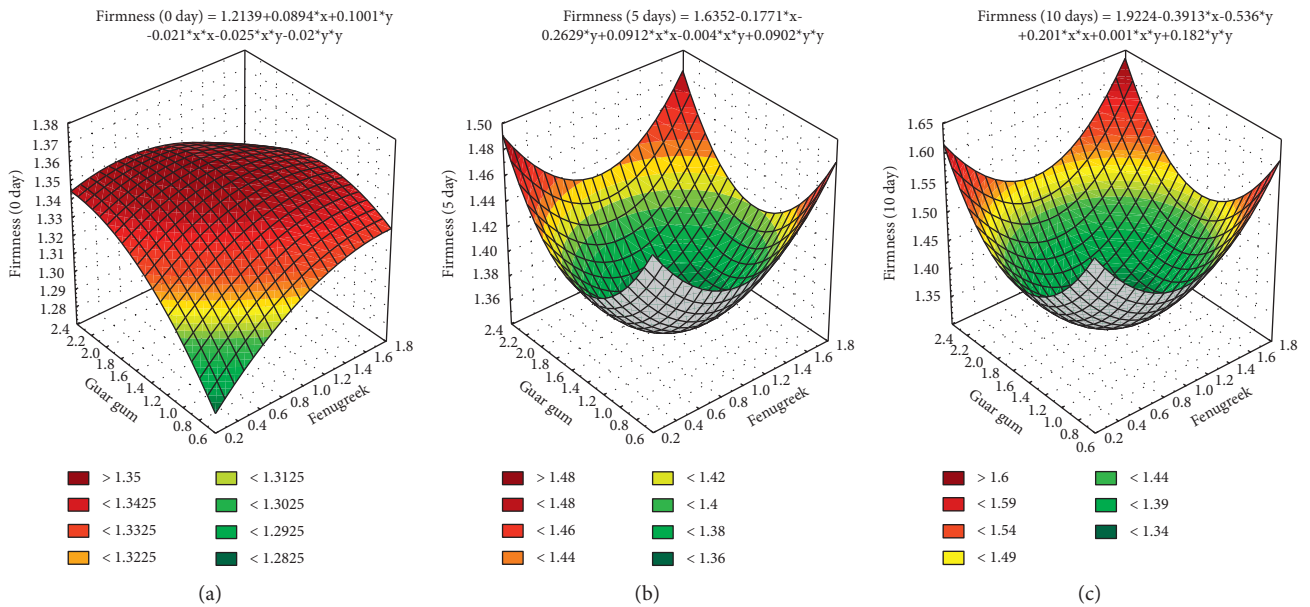


FIGURE 3: Continued.

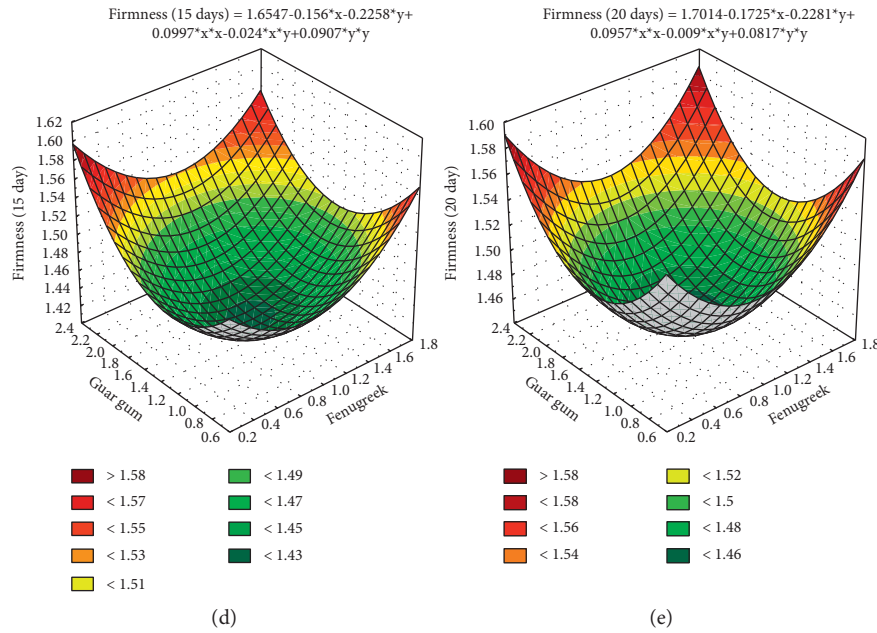


FIGURE 3: Response surface plot showing the impact of guar galactomannan and fenugreek galactomannan on the firmness of guava fruit during storage. (a) Firmness (0 days) = 1.2139 + 0.0894 * x + 0.1001 * y - 0.021 * x * x - 0.025 * x * y - 0.02 * y * y. (b) Firmness (5 days) = 1.6352 - 0.1771 * x - 0.2629 * y + 0.0912 * x * x - 0.004 * x * y + 0.0902 * y * y. (c) Firmness (10 days) = 1.9224 - 0.3913 * x - 0.536 * y + 0.201 * x * x + 0.001 * x * y + 0.182 * y * y. (d) Firmness (15 days) = 1.6547 - 0.156 * x - 0.2258 * y + 0.0997 * x * x - 0.024 * x * y + 0.0907 * y * y. (e) Firmness (20 days) = 1.7014 - 0.1725 * x - 0.2281 * y + 0.0957 * x * x - 0.009 * x * y + 0.0817 * y * y.

TABLE 4: Analysis of variance (sum of the square) for response surface model of fruit firmness during storage.

| SOV | Df | Days | | | | |
|-------------------|----|------------------------|------------------------|------------------------|------------------------|------------------------|
| | | 0 | 5 | 10 | 15 | 20 |
| Model | 5 | 0.000902 ^{ns} | 0.001865* | 0.008777* | 0.003303* | 0.002014* |
| Linear | 2 | 0.000652 ^{ns} | 0.000031* | 0.000537 ^{ns} | 0.001122* | 0.000194 ^{ns} |
| Fenugreek (FG) | 1 | 0.000197 ^{ns} | 0.000001* | 0.000297 ^{ns} | 0.000114* | 0.000060 ^{ns} |
| Guar gum (GG) | 1 | 0.000455 ^{ns} | 0.000030 ^{ns} | 0.000240 ^{ns} | 0.001009* | 0.000134 ^{ns} |
| Square | 2 | 0.000094 ^{ns} | 0.001830* | 0.008240 ^{ns} | 0.002036 ^{ns} | 0.001799* |
| FG * FG | 1 | 0.000080 ^{ns} | 0.001514* | 0.007346* | 0.001809* | 0.001667* |
| GG * GG | 1 | 0.000073 ^{ns} | 0.001481* | 0.006023 ^{ns} | 0.001497 ^{ns} | 0.001215* |
| 2-way interaction | 1 | 0.000156 ^{ns} | 0.000004 ^{ns} | 0.000000 ^{ns} | 0.000144* | 0.000020 ^{ns} |
| FG * GG | 1 | 0.000156 ^{ns} | 0.000004 ^{ns} | 0.000000 ^{ns} | 0.000144* | 0.000020 ^{ns} |
| Error | 3 | 0.002181 | 0.000239 | 0.002070 | 0.000497 | 0.001094 |
| Total | 8 | 0.003083 | 0.002104 | 0.010847 | 0.003800 | 0.003108 |
| R-square (%) | | 29.26 | 88.64 | 90.91 | 96.92 | 94.79 |

If $p > 0.05$, ^{ns} non-significant ($p > 0.05$); $p < 0.05$, * significant; and $p < 0.01$, ** highly significant fenugreek galactomannan (FG), guar galactomannan (GG).

and GG² are found significant at the 5th and 10th days. The interaction of two variables (XY) was non-significant at the start of the study and then significant effect on the TSS of guava fruits at various storage intervals. The coefficient of determination (R^2) was low on the 1st day and then was above 80% after the 5th day, indicating that models are well fitted (Figure 3). The average optimized levels of fenugreek and guar galactomannan in the edible coating were found to be 1.02 and 1.44%, respectively. The effect of edible coating on Brix of guava described that the total soluble solid increased up to a certain storage period in all the treatments and thereafter reduced as the storage period progressed. These results are consistent with previous findings [26]. The initial increase in TSS during storage was mainly due to the

conversion of starch into soluble forms of sugars and the subsequent decrease in TSS was due to rapid utilization of reducing sugar and other organic metabolites [5].

3.6. pH. The regression coefficients of variables showed that FG and GG significantly affected the change in pH of guava fruits during storage. The effect of linear terms of fenugreek galactomannan and guar galactomannan are statistically significant ($p < 0.05$) for pH at all days of storage intervals (supplementary Figure 7 and supplementary Table 2). The X² quadratic terms (FG² and GG²) are found significant at 5, 10, 15, and 20 days' storage intervals. The interaction of two variables (FG * GG) shows a significant effect on the pH of

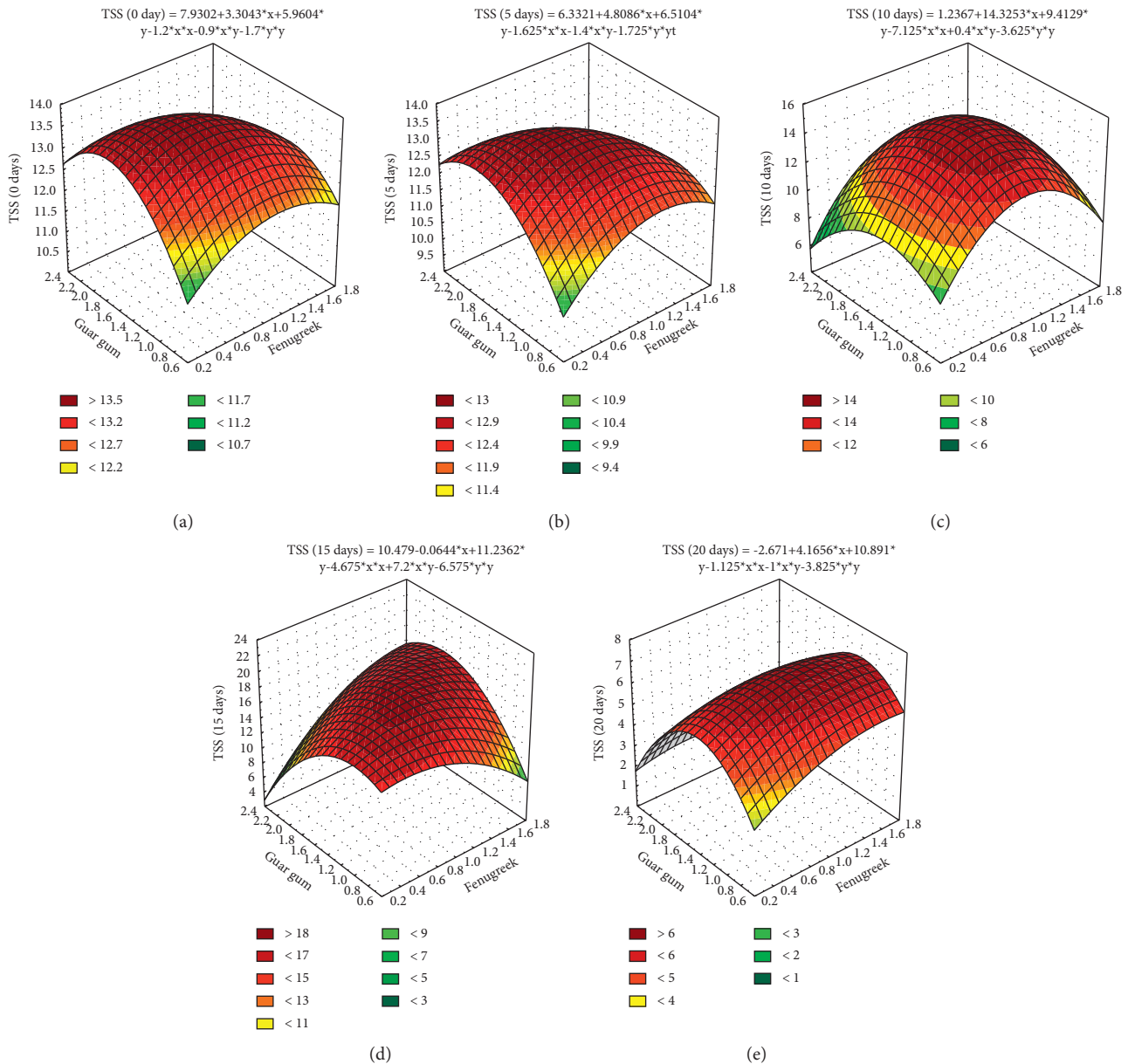


FIGURE 4: Response surface plot showing the impact of guar galactomannan and fenugreek galactomannan on TSS of guava fruit during storage. (a) TSS (0 days) = $7.9302 + 3.3043 * x + 5.9604 * y - 1.2 * x * x - 0.9 * x * y - 1.7 * y * y$. (b) TSS (5 days) = $6.3321 + 4.8086 * x + 6.5104 * y - 1.625 * x * x - 1.4 * x * y - 1.725 * y * y$. (c) TSS (10 days) = $1.2367 + 14.3253 * x + 9.4129 * y - 7.125 * x * x + 0.4 * x * y - 3.625 * y * y$. (d) TSS (15 days) = $10.479 - 0.0644 * x + 11.2362 * y - 4.675 * x * x + 7.2 * x * y - 6.575 * y * y$. (e) TSS (20 days) = $-2.671 + 4.1656 * x + 10.891 * y - 1.125 * x * x - 1 * x * y - 3.825 * y * y$.

guava fruits at all storage intervals. The coefficient of determination (R^2) was 99.10% on the 1st day, 96.78% on the 5th day, 75.58% on the 10th day, 88.48% on the 15th day, and 82.13% on the 20th day, indicating that models are well fitted (supplementary Figure 7).

A similar increasing trend in pH values of the galactomannan coated guavas throughout the storage period has been observed in the previously reported study [32]. Moreover, another study reported a minor raise in pH values after eight days of storage time [30]. That variation was occurred owing to the decrease in the amount of malic acid, because of the rise in respiration rate during cutting and

peeling. The average optimized levels of fenugreek galactomannan and guar galactomannan in the edible coating were 0.83% and 1.36%, respectively. The increased pH value was due to the development of organic acids through maturity or storage period [27].

3.7. Acidity. The regression coefficients of variables in models showed that the FG and GG did not contribute toward change in acidity in guava fruits at the start of storage but after that contributed significantly from 5th to 20 days storage intervals. The effect of linear terms of fenugreek galactomannan and

guar galactomannan are statistically significant ($p < 0.05$) for acidity at all days of storage intervals except the start of the study (supplementary Figure 8 and supplementary Table 3). The X^2 quadratic terms are found significant at 5, 10, 15, and 20 days storage intervals, whereas the quadratic terms for FG^2 and GG^2 are found significant at 5th and 10th days. The interaction of two variables (XY) shows a nonsignificant effect at the start and a significant effect on the acidity of guava fruits after 5 days storage intervals. The coefficients of determination (R^2) were well enough for well-fitted models (supplementary Figure 8). A significant decrease in acidity was observed at 5, 10, and 20 days of storage. The increase in pH and decrease in acidity in coated fruits was due to the reduction in respiration rate of fruits and consequently limit the over consumption of organic acids in respiration reactions [33]. The average optimized levels of fenugreek galactomannan and guar galactomannan in edible coating were 1.03% and 1.44%, respectively. The reduction of organic acids leads to decline of TA that subsequently results in increased juice pH of guava fruits [34]. The influence of treatments and storage times was significant on titratable acidity (TA) of guava fruits. Overall, the TA was progressively decreased but the reduction was significantly higher in control than the treated guava fruits.

4. Conclusion

According to the present study, 1.24 g guar galactomannan and 1.01 g fenugreek galactomannan in edible coating appeared to be the optimum coating formulation for improving the post-harvest quality of guava fruit. The response surface methodology was observed as an effective statistical tool to discriminate the interactive effects of independent variables. The FG and GG-based edible coating significantly reduced the weight loss and TSS. Moreover, the coated fruit was fresh, firmer, and low in TA during storage. Thus, the RSM could be effectively used to optimize edible coating formulations leading to the overall enhancement of the quality and shelf-life of guava fruit.

Abbreviations

CO₂: Carbon dioxide
 CCD: Central composite design
 EC: Emulsifying capacity
 ES: Emulsion stability
 FG: Fenugreek galactomannan
 GG: Guar galactomannan
 RSM: Response surface methodology
 TSS: Total soluble solids
 O₂: Oxygen.

Data Availability

The dataset supporting the conclusions of this article is included in the manuscript.

Additional Points

Practical Application. Galactomannan-based edible coating can be served as environmentally friendly packaging by

replacing the use of chemicals and reducing packaging waste in the food industry.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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Supplementary Materials

Supplementary Figure 1: extraction and purification of guar galactomannan. Supplementary Figure 2: extraction and purification of fenugreek galactomannan. Supplementary Figure 3: XRD pattern of guar galactomannan. Supplementary Figure 4: XRD pattern of fenugreek galactomannan. Supplementary Figure 5: FTIR spectra of guar galactomannan. Supplementary Figure 6: FTIR spectra of fenugreek galactomannan. Supplementary Figure 7: response surface plot showing the impact of guar galactomannan and fenugreek galactomannan on pH of guava fruit during storage. Supplementary Figure 8: response surface plot showing the impact of guar galactomannan and fenugreek galactomannan on the acidity of guava fruit during storage. Supplementary Table 1: analysis of variance (sum of the square) for response surface model of fruit firmness during storage. Supplementary Table 2: analysis of variance (sum of the square) for response surface model of fruit TSS during storage. Supplementary Table 3: analysis of variance (sum of the square) for response surface model of fruit pH during storage. Supplementary Table 4: analysis of variance (sum of the square) for response surface model of fruit acidity during storage. (*Supplementary Materials*)

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