

Article

Optimization of Gum Arabic and Starch-Based Edible Coatings with Lemongrass Oil Using Response Surface Methodology for Improving Postharvest Quality of Whole “Wonderful” Pomegranate Fruit

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Abstract: The effects of edible coatings based on gum arabic (GA) (0.5–1.5%), maize starch (MS) (0.5–1.5%), lemongrass oil (LO) (2–4%), and glycerol (GC) (0.5–1%) developed using response surface methodology (RSM) on “Wonderful” pomegranate fruit were studied. After 42 days of storage (5 ± 1 °C, $95 \pm 2\%$ RH) and 5 days at ambient temperature (20 ± 0.2 °C and $60 \pm 10\%$ RH), whole fruit were evaluated for weight loss (%) and pomegranate juice (PJ) for total soluble solids (°Brix), titratable acidity (% Citric acid), and antioxidant capacity. The optimization procedure was done using RSM and the response variables were mainly influenced by the concentrations of MS and GA. The optimized coating consisted of GA (0.5%), MS (0.5%), LO (3%), and GC (1.5%) with desirability of 0.614 (0—minimum and 1—maximum). The predicted values of response variables, for the coating were weight loss (%) = 5.51, TSS (°Brix) = 16.45, TA (% Citric acid) = 1.50, and antioxidant capacity (RSA = 58.13 mM AAE/mL PJ and FRAP = 40.03 mM TE/mL PJ). Therefore, the optimized coating formulation is a potential postharvest treatment for “Wonderful” pomegranate to inhibit weight loss and maintain overall quality during storage and shelf-life.



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Keywords: glycerol; antioxidant; formulation; temperature

1. Introduction

In recent years, the application of natural biopolymer-based packaging materials to reduce postharvest losses in fresh fruit and vegetables has grown into prominence [1,2]. This can be explained by high consumer demand for nutritious food of low toxicity and increased environmental waste problems derived from the disposal of non-biodegradable petrochemical-based plastic packaging materials [3,4]. Studies have highlighted several benefits associated with the use of natural biopolymer-based packaging materials from renewable resources for food preservation, which include reduced waste disposal [5,6], reduction of product quality losses [7,8], enhanced nutrition [9], and individual packaging of particulate foods [10,11].

Several biodegradable biopolymers exist, including polysaccharides, proteins, lipids, and their composites/combination [8,12,13]. Environmentally friendly edible coatings are thin materials developed from natural biopolymer materials applied in liquid form on food products to serve as surface barriers to moisture and gas movement [14]. Edible coatings act as potential substitutes for synthetic polymers and petrochemicals in food packaging by offering physical protection and creating proper physicochemical conditions

on food products [15,16]. Gum arabic (GA) is an example of a complex branched, anionic, and hydrophilic heteropolysaccharide [17,18]. The polysaccharide is derived from gummy exudation of *Acacia senegal* and *Acacia seyal* tree branches which belong to the *Leguminosae* family [17,18]. Hard nodules formed from the sticky substance are processed into a highly soluble hydrocolloid with broad application in the industrial sector as a food additive, in bakery products, pharmaceuticals, and cosmetics [19]. Gum arabic has the highest toxicology safety, according to the Joint Food and Agriculture Organization/World Health Organization (FAO/WHO) Expert Committee on Food Additive [17,20]. Components such as galactose, rhamnose, arabinose, and glucuronic acid give the polysaccharide an excellent film-forming ability of low viscosity and high emulsification [21,22]. Studies show that GA-based edible coatings are applied to extend postharvest shelf-life and maintain the quality of pomegranate arils [23], tomato [24], and banana [19], including other food products.

Maize starch (MS) is another commonly used agricultural raw material for edible coatings [25]. The main component, amylose, exhibits good film-forming properties and determines compatibility with solvents and plasticizers for the best physical and/or mechanical properties [26]. Starch-based coatings exhibit excellent oxygen barrier properties under low moisture conditions [27]. Earlier studies found that starch-based coatings decreased weight loss and extended storage life in strawberries [28,29], tomato [30], and lemon [31], amongst many others. However, studies show that starch coatings exhibit poor water vapor barrier properties, which is often compensated by adding plasticizers and emulsifiers. For instance, glycerol (GC), a plasticizer, is used to produce starch and lipid-based biodegradable coatings with great flexibility and low hydrophilic properties for fresh food preservation [32]. Glycerol improves the interaction and mobility between polymer macromolecules which promotes flexibility of the final coating solution when applied on fresh food products [33]. Moreover, the incorporation of lipids into hydrophilic polysaccharide coatings improves water vapor barrier properties to produce a hydrophobic composite coating with low permeability to CO₂ and O₂ [4,34].

A wide range of antimicrobial agents such as essential oils, bacterions, and enzymes can be integrated into edible coatings to reduce microbial proliferation in food products [10,23,35]. For example, studies have found that flax-gum, alginate, and chitosan-based coatings enriched with lemongrass essential oil (LO) improved the quality and extended the shelf life of pomegranate arils, fresh-cut pineapples, and strawberries [36–38]. Lemongrass essential oil is a natural mixture of compounds such as terpenes, alcohols, aldehydes, ketones, carboxylic acids, and others found to exhibit antimicrobial activity against different types of microorganisms [39]. Even though reports show successful application of edible coatings on fresh products, other applications may result in unfavorable responses. Edible coatings modify the internal atmosphere and limit water movement, which may result in off-flavors and physiological disorders related to high CO₂ or low O₂ concentrations [40]. For that purpose, optimization of the different components of the edible coatings is important to ensure the best results upon application on fresh food products.

Response surface methodology (RSM) is a common multivariate statistical tool used to optimize concentration of individual components of composite edible coatings [41–43]. The method is mainly employed to determine the best conditions from multiple experimental runs deduced from several variables to describe experimental data with polynomial equations and draw statistical conclusions [44]. The RSM has been applied to optimize edible coating solutions to extend storage and shelf life of papaya [42], guava [43], pear [45], fresh-cut apples [41], and longan fruit [46], amongst many others. “Wonderful” pomegranate (*Punica granatum* L.) is a fruit of high commercial value that is highly susceptible to excessive weight loss and develops undesirable physiological and pathological disorders during cold storage, leading to major postharvest losses [47,48]. These characteristics reduce the storage and shelf life of the fruit during transportation and on the retail market. Considering this, developing postharvest treatments for preserving “Wonderful” pomegranate is a high priority. Several authors have reported edible coatings as a promising treatment for pomegranate whole fruit and fractions to minimize postharvest losses during storage and

shelf life [23,36,49–51]. However, limited studies derive edible coating formulations with optimum concentrations of ingredients to potentially extend pomegranate storage and shelf-life. Therefore, this study intended to optimize the concentration of gum arabic, maize starch, lemongrass oil, and glycerol in coating formulations for postharvest preservation of “Wonderful” pomegranate.

2. Materials and Methods

2.1. Materials

Pomegranate (cv. Wonderful) fruit harvested at commercial maturity were procured from Sonlia Pack House, Wellington, Western Cape (33°38'23" S, 19°00'40" E), South Africa. Fruit without defects were sanitized by dipping in 0.02% NaClO for 5 min and allowed to dry before cold storage (5 ± 1 °C, $95 \pm 2\%$ RH). Gum arabic (Sigma–Aldrich Co., Johannesburg, South Africa) and maize starch (Chem Lab Suppliers Co., Johannesburg, South Africa) were used as the edible coating components and glycerol (Sigma–Aldrich Co., Johannesburg, South Africa) as the plasticizer. Lemongrass essential oil was purchased from Umuthi Botanicals Co. South Africa and stored (4 °C) before use.

2.2. Preparation of Coating Solutions

A central composite design with three factors, namely GA, MS, LO, and GC was applied to determine the concentration of each component of the coating solutions [45] (Table 1). For each factor, there were three different points, namely factorial, central, and axial. Center points indicate runs in the experimental domain equal to the median of each factor [44]. Axial points are experimental runs for points located on the axes of the coordinate system with respect to the central point except for one factor, with the values below and above the median of the two factorial levels [44]. The range of each factor was selected based on previous work with slight modifications [41,52,53]. The coating solutions (Table 1) were prepared in 1000 mL warm milli-Q water (25 °C) under magnetic stirring for 45 min. The coating solutions were homogenized at 2500 rpm for 30 min in an overhead stirrer (Scientech Co., Indore, India).

Table 1. The central composite design employed for formation of edible coating composition.

Point Type	A: Gum Arabic (%)	B: Maize Starch (%)	C: Lemongrass Oil (%)	D: Glycerol (%)
Factorial	0.5	0.5	2	0.5
Factorial	1.5	0.5	2	0.5
Factorial	0.5	1.5	2	0.5
Factorial	1.5	1.5	2	0.5
Factorial	0.5	0.5	4	0.5
Factorial	1.5	0.5	4	0.5
Factorial	0.5	1.5	4	0.5
Factorial	1.5	1.5	4	0.5
Factorial	0.5	0.5	2	1.5
Factorial	1.5	0.5	2	1.5
Factorial	0.5	1.5	2	1.5
Factorial	1.5	1.5	2	1.5
Factorial	0.5	0.5	4	1.5
Factorial	1.5	0.5	4	1.5
Factorial	0.5	1.5	4	1.5
Factorial	1.5	1.5	4	1.5
Axial	0	1	3	1
Axial	2	1	3	1
Axial	1	0	3	1
Axial	1	2	3	1
Axial	1	1	1	1
Axial	1	1	5	1
Axial	1	1	3	0
Axial	1	1	3	2

Table 1. Cont.

Point Type	A: Gum Arabic (%)	B: Maize Starch (%)	C: Lemongrass Oil (%)	D: Glycerol (%)
Center	1	1	3	1
Center	1	1	3	1
Center	1	1	3	1
Center	1	1	3	1
Center	1	1	3	1
Center	1	1	3	1

2.3. Coating Application and Storage

Fruit were submerged into coating treatments for 1 min as designed using RSM [44] (Table 1) and allowed to dry at 20 ± 0.2 °C. “Wonderful” pomegranate fruit were packaged into standard open top ventilated cartons (dimensions: 0.40 m long, 0.30 m wide, and 0.12 m high) (10 fruit per carton) containing Xtend® liners and stored (5 ± 1 °C, $95 \pm 2\%$ RH) for 42 days (maximal sea freight duration of “Wonderful” pomegranate fruits from South Africa to Europe across the Atlantic Ocean) and 5 days at ambient temperature (20 ± 0.2 °C and $60 \pm 10\%$ RH). Measurements were done after 5 days at ambient temperature for weight loss, respiration rate, total soluble solids, titratable acidity, and antioxidant capacity.

2.4. Weight Loss

An electronic weighing balance (ML3002.E, Mettler Toledo, Switzerland) was used to determine weight from ten randomly selected fruit per treatment [54]. Weight loss was calculated using the following equation:

$$W_L = (W_O - W_t) / W_O \times 100 \quad (1)$$

where W_L is weight loss (%), W_O is the initial weight (g) of fruit, and W_t is the fruit weight (g) at time of analysis.

2.5. Respiration Rate

Respiration rate was determined from CO₂ production and fresh fruit weight in a closed system using a gas analyzer (Checkmate 3, PBI Dansensor, Ringstead, Denmark) [55]. Briefly, in triplicates per coating treatment, fruit were pre-conditioned for 2 h in a sealed glass jar (volume = 3 L) with a lid containing a rubber septum in the middle. Following that, CO₂ produced by fruit within the glass jar was measured from the headspace through the rubber septum. To ensure accurate measurements, the device was auto calibrated with the atmospheric gas composition prior to taking each measurement and the results were expressed as a percentage of CO₂ gas.

2.6. Total Soluble Solids and Titratable Acidity

Total soluble solids (TSS) and titratable acidity (TA) were measured from pomegranate juice (PJ) extracted from ten randomly selected fruit per treatment. To collect the PJ, fruit were carefully cut at the equatorial zone with sharpened knives (Sigma-Aldrich, Johannesburg, South Africa) and peels and arils were manually separated [54]. The PJ was extracted from all the collected arils using a blender (Mellerware, South Africa). Titratable acidity was evaluated using a titrosampler (Metrohm 862, Herisau, Switzerland) from diluted PJ (2 mL diluted with 70 mL milli-Q water) titrated against 0.1 N NaOH to an endpoint of pH = 8.2. The results were expressed as percentage of citric acid (% CA). The TSS was determined using a digital refractometer (Atago, Tokyo, Japan).

2.7. Antioxidant Activity

2.7.1. Radical-Scavenging Activity

Radical scavenging activity (RSA) of PJ was determined spectrophotometrically following a method reported by Kawhena et al. [23]. Briefly, in triplicates, 15 µL of PJ was

mixed with 735 μL methanol and added to 750 μL of 2,2-Diphenyl-1-picryl-hidrazil (DPPH) solution (0.1 mM). Absorbance was measured at 517 nm and RSA of PJ was expressed as ascorbic acid (mMol) equivalent per 100 mL pomegranate juice (mM AAE/100 mL PJ).

2.7.2. Ferric Ion Reducing Antioxidant Power

To determine ferric ion reducing antioxidant power (FRAP), the method reported by Kawhena et al. [23] modified according to Fawole et al. [56] was followed. The FRAP solution was prepared from 25 mL acetate buffer (300 mM acetate buffer, pH = 3.6), 2.5 mL of 2,4,6-tripyridyl-s-triazine (TPTZ) solution (10 mM), and 2.5 mL of FeCl_3 solution (20 mM). In triplicates, 1 mL of PJ was diluted with 10 mL aqueous methanol (50%) and centrifuged at 4000 rpm for 10 min. The solutions were mixed with 2850 μL FRAP solution and absorbance was measured at spectrophotometrically at 593 nm. The results were expressed as Trolox (mMol) equivalents (mM TE) per 100 mL of pomegranate juice (mM TE/100 mL PJ).

2.8. Experimental Design and Statistical Analysis

Design Expert software (version 12, Statease Inc., Minneapolis, MN, USA) was used to generate experimental designs and perform regression analysis of experimental data. Independent variables were expressed at 3 levels (−1, 0, and +1) (Table 2). For the central composite design, six replicates of the center point were chosen in random order for a stable variance of the predicted response and to calculate the repeatability of the method [57]. Coefficient of determination (R^2) was used to determine quality of fit of polynomial mathematical models. Optimization was carried out using numerical optimization tools involving four independent variables: the concentrations of GA, MS, LO, and the plasticizer GC. The following relationship between the responses and the coded form of the independent variables were developed according to Equation (2):

$$Y = \beta_0 + \sum \beta_i X_i + \sum \beta_{ii}(X_i)^2 + \sum \beta_{ij} X_i X_j, \quad (2)$$

where Y is the response variables, β_0 is the constant; β_i , β_{ii} , β_{ij} are linear, quadratic, and interaction coefficients, respectively.

Table 2. Independent variables and the three levels of each factor used in the central composite design (CCD).

Independent Variable	Levels		
	Low (−1)	Center (0)	High (+1)
A-Gum arabic (% w/v)	0.5	1	1.5
B-Maize starch (% w/v)	0.5	1	0.5
C-Lemongrass oil (% w/v)	2	3	4
D-Glycerol (% w/v)	0.5	1	1.5

3. Results

3.1. Weight Loss

The regression coefficients for weight loss are shown in Table 3 and experimental data in Table 4. The negative coefficient values (−0.6816, −0.5087) for weight loss indicate that when GA and MS concentrations increased, weight loss values decreased. This observation is desirable as it suggests that the coatings minimized weight loss in fruit during storage. The model F-value was significant ($p < 0.0001$) with an F-value of 20.85 (0.01% chance of noise occurring at this F-value). Model terms A, B, AB, AD, A^2 , B^2 , and C^2 were all significant ($p < 0.05$) and thus, weight loss was mainly controlled by the concentration of GA, MS, and their interaction with LO and GC, and their quadratic effects. The “Lack of Fit” F-value of 0.51 implied that the “Lack of Fit” was not significant relative to the pure error (85.75% chance of noise at this F-value). The predicted R^2 value of 0.80 agreed with the adjusted R^2 of 0.86 (difference < 0.2). The Adeq Precision ratio of 13.26 indicated that an adequate signal for navigating the design space. The 3D-surface image (Figure 1) showed

that MS concentration changes had a greater effect on weight loss than GA. An increase in MS concentration was inversely related to weight loss. These results suggest that both MS and GA significantly minimized water loss when applied as surface coatings. The predictive model in terms of actual factors for weight loss is indicated in Equation (3):

$$\text{Weight loss (\%)} = 3.58189 - 1.98279A - 3.50795B + 1.02017C - 1.14851D - 1.70309AB + 0.862239AD - 1.25261A^2 - 1.41111B^2 - 0.173606C^2. \quad (3)$$

Table 3. The regression coefficients, mean, R² and F-values for dependent variables of “Wonderful” pomegranate fruit coated with different concentrations of gum arabic, maize starch, lemongrass oil, and glycerol (WL—weight loss, RR—respiration rate, TSS—total soluble solids, TA—titratable acidity, RSA—radical scavenging activity, FRAP—ferric ion reducing antioxidant power).

Regression Coefficients	WL (%)	RR (CO ₂ mg·kg ⁻¹ ·h ⁻¹)	TSS (°Brix)	TA (% CA)	RSA (mMAAE/ mL PJ)	FRAP (mMTE/mL PJ)
A-Gum arabic	-0.6816 *	-3.55 *	-1.15 *	-0.1861 *	-801.18 *	-497.25 *
B-Starch	-0.5087 *	-3.81 *	-0.9222 *	-0.1586 *	-631.83 *	-420.19 *
C-Lemongrass oil	-0.0215	-	-0.0722	-0.0031	7.14	-32.75
D-Glycerol	-0.1431	-	-0.2556	-0.0150	-68.61	-17.18
AB	-0.4258 *	-2.81 *	-0.2958	-0.1104 *	-502.59 *	-362.16 *
AC	0.1024	-	0.4750	0.0521	189.43 *	139.82 *
AD	0.2156 *	-	0.1792	0.0300	172.56 *	76.00
BC	-0.1627	-	-0.3833	-0.0325	-84.29	-40.83
BD	0.1407	-	-0.2292	-0.0104	-0.1777	-18.99
CD	-0.0857	-	0.0833	0.0012	-47.85	24.31
A ²	-0.3164 *	-2.44 *	-0.3882 *	-0.0754 *	-276.08 *	-206.33 *
B ²	-0.3560 *	-	-0.5757 *	-0.0887 *	-340.54 *	-245.99 *
C ²	-0.1768 *	-	-0.3632 *	-0.0437 *	-154.82 *	-119.53 *
D ²	-0.0225	-	0.0243	-0.0029	0.9567	-22.92
Mean	5.25	32.92	14.27	1.26	5122.93	3443.71
R ²	0.8603	0.7011	0.7284	0.9095	0.9490	0.9506
Model F value	20.85	14.66	12.11	37.43	35.37	50.50

* Significant at $p < 0.05$.

Table 4. The central composite design and experimental data obtained for starch and gum arabic based edible coatings applied on “Wonderful” pomegranate fruit during cold storage (GA—gum arabic, MS—maize starch, LO—lemongrass oil, GC—glycerol).

Point Type	A: GA (%)	B: MS (%)	C: LO (%)	D: GC (%)	WL (%)	RR (CO ₂ mg·kg ⁻¹ ·h ⁻¹)	TSS (°Brix)	TA (%)	RSA (mMAAE/ mL PJ)	FRAP (mMTE/ mL PJ)
-	(%)	(%)	(%)	(%)	(%)	(CO ₂ mg·kg ⁻¹ ·h ⁻¹)	(°Brix)	(%)	(mMAAE/ mL PJ)	(mMTE/ mL PJ)
Factorial	0.5	0.5	2	0.5	6.22	37.83	16.13	1.48	61.58	40.44
Factorial	1.5	0.5	2	0.5	5.28	32.55	13.60	1.24	49.35	34.70
Factorial	0.5	1.5	2	0.5	6.36	39.81	16.60	1.54	62.19	42.14
Factorial	1.5	1.5	2	0.5	3.10	18.44	12.63	0.72	27.38	19.03
Factorial	0.5	0.5	4	0.5	6.39	37.90	15.90	1.46	59.77	38.16
Factorial	1.5	0.5	4	0.5	5.62	35.22	14.90	1.38	58.12	37.59
Factorial	0.5	1.5	4	0.5	5.46	35.20	14.20	1.31	57.66	35.76
Factorial	1.5	1.5	4	0.5	3.49	17.14	11.37	0.70	26.48	18.96
Factorial	0.5	0.5	2	1.5	5.22	38.24	16.30	1.52	61.61	41.33
Factorial	1.5	0.5	2	1.5	5.13	32.18	13.37	1.22	50.93	34.84
Factorial	0.5	1.5	2	1.5	5.73	34.81	14.47	1.33	54.51	36.44
Factorial	1.5	1.5	2	1.5	3.98	18.90	11.20	0.76	31.02	20.45
Factorial	0.5	0.5	4	1.5	5.23	36.22	14.90	1.39	55.66	36.65
Factorial	1.5	0.5	4	1.5	5.22	30.20	14.83	1.37	54.37	38.28

Table 4. Cont.

Point Type	A: GA	B: MS	C: LO	D: GC	WL	RR	TSS	TA	RSA	FRAP
-	(%)	(%)	(%)	(%)	(%)	(CO ₂ mg·kg ⁻¹ ·h ⁻¹)	(°Brix)	(%)	(mMAAE/mL PJ)	(mMTE/mL PJ)
Factorial	0.5	1.5	4	1.5	4.94	30.20	12.77	1.17	49.34	34.70
Factorial	1.5	1.5	4	1.5	3.29	20.47	11.57	0.78	32.56	21.48
Axial	0	1	3	1	6.22	38.57	16.17	1.51	61.71	40.83
Axial	2	1	3	1	3.26	38.57	11.23	0.79	31.62	21.30
Axial	1	0	3	1	5.64	34.56	14.70	1.36	54.43	36.44
Axial	1	2	3	1	3.51	21.54	11.20	0.83	33.74	22.53
Axial	1	1	1	1	5.08	31.47	13.27	1.23	49.93	34.56
Axial	1	1	5	1	5.51	33.83	14.33	1.32	53.09	34.52
Axial	1	1	3	0	5.97	36.23	15.40	1.46	58.73	38.78
Axial	1	1	3	2	5.85	36.22	15.30	1.42	56.76	38.03
Center	1	1	3	1	5.19	31.54	15.50	1.27	50.53	34.52
Center	1	1	3	1	6.65	42.41	15.27	1.61	63.51	43.53
Center	1	1	3	1	5.93	36.49	13.40	1.42	55.24	39.73
Center	1	1	3	1	5.75	35.46	17.33	1.37	56.89	38.00
Center	1	1	3	1	6.13	38.04	15.47	1.47	60.09	39.34
Center	1	1	3	1	6.02	37.39	14.93	1.44	58.10	40.05

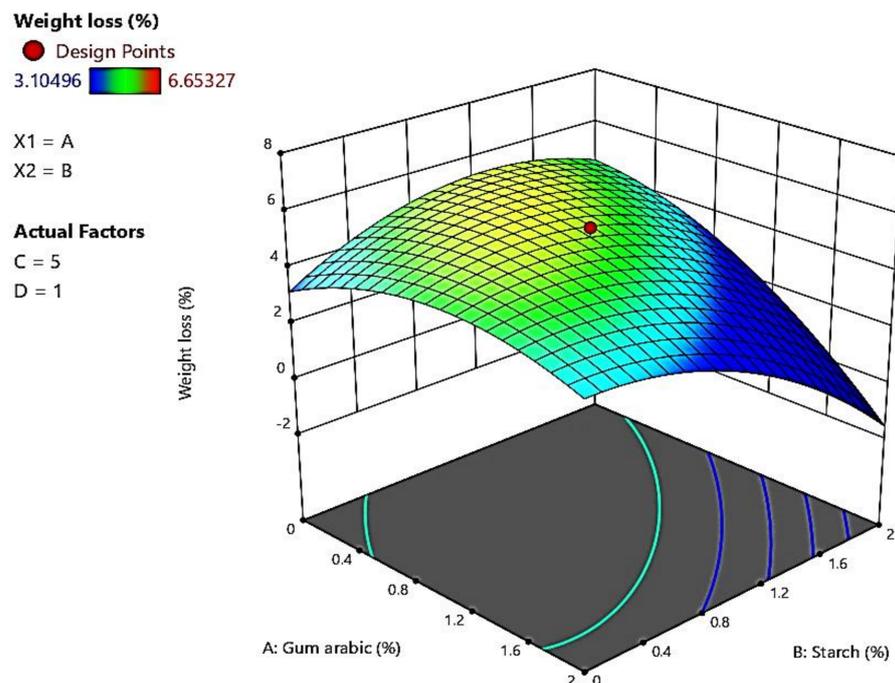


Figure 1. Response surface plot for weight loss (%) of coated “Wonderful” pomegranate as a function of gum arabic and maize starch concentration.

3.2. Respiration Rate

Tables 3 and 4 show regression coefficient values and experimental data obtained for respiration rate, respectively. There were negative regression coefficient values (−3.55, −3.81, −2.81, and −2.44) recorded for respiration rate. This observation suggests that when GA and MS concentrations increased, respiration rate decreased. From the four independent variables (GA, MS, LO, and GC) measured, respiration rate was mainly influenced by changes in concentration of GA and MS, and the quadratic effect of GA. A significant model F-value of 14.66 was obtained and model terms, namely A, B, AB, A² were significant ($p < 0.05$). The calculated “Lack of Fit” F-value of 1.36 indicated that the

“Lack of Fit” was not significant relative to the pure error. The predicted R^2 value of 0.5151 agreed with the adjusted R^2 value of 0.6533 (difference < 0.2). The Adeq Precision ratio was 14.912, showing an adequate signal to navigate the design space. The 3D surface image (Figure 2) showed that increases in GA and MS concentration reduced respiration rate of whole fruit. The predictive model in terms of actual factors for respiration rate is indicated in Equation (4):

$$\text{Respiration rate (CO}_2 \text{ mg}\cdot\text{kg}^{-1}\cdot\text{h}^{-1}) = 28.55278 + 4.16312A + 23.1952B - 11.25532AB - 9.77935B^2. \tag{4}$$

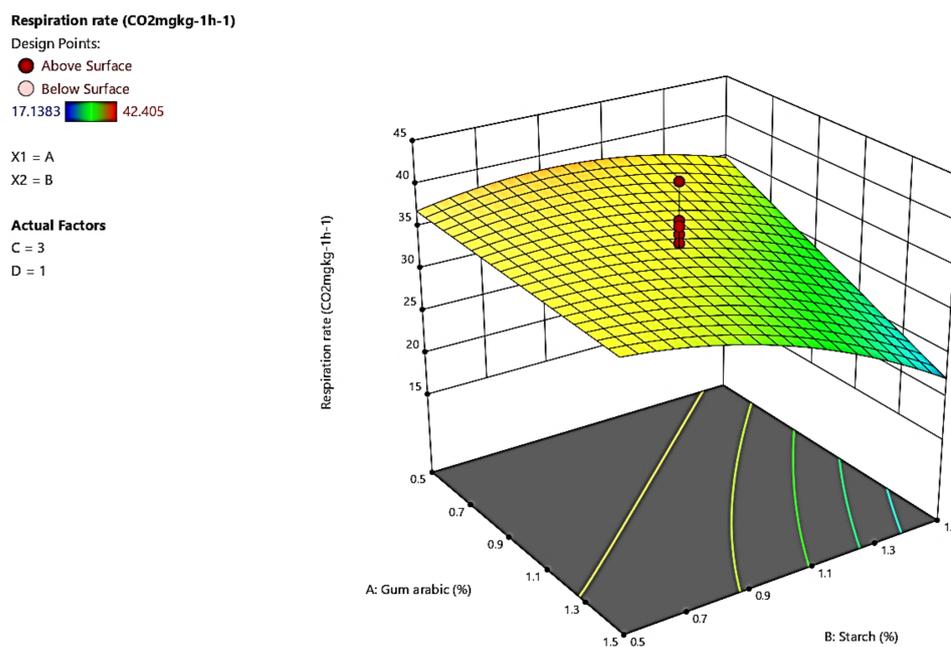


Figure 2. Response surface plot for respiration rate (CO₂ mg·kg⁻¹·h⁻¹) of coated “Wonderful” pomegranate as a function of gum arabic and maize starch and concentration.

3.3. Total Soluble Solids

Table 3 shows the regression coefficients table for RSM analysis of TSS of PJ as the response variable and its corresponding values of average Mean and R^2 value. The experimental data obtained for all response variables including TSS are shown in Table 4. The negative coefficient values (−1.15, −0.9222) indicated that when GA and MS concentrations increased, the TSS component values decreased. The Model F-value of 12.11 implied that the model was significant ($p < 0.05$) and model terms A, B, AC, A², B², and C² were all significant ($p < 0.05$). The “Lack of Fit” F-value of 0.38 was not significant ($p < 0.05$) relative to the pure error. The predicted $R^2 = 0.65$ agreed with the adjusted R^2 of 0.73 (difference < 0.2). The “Adeq Precision” was 11.59, indicated an adequate signal of the model to navigate the design space. Observation of 3D surface image (Figure 3) shows that an increase in MS and GA concentrations reduced the TSS in PJ. The predictive model in terms of actual factors for TSS is indicated in Equation (5):

$$\text{Total soluble solids (°Brix)} = 15.37778 - 2.02222A + 2.78889B + 1.17778C + 0.95AC - 1.56667A^2 - 2.31667B^2 - 0.36667C^2. \tag{5}$$

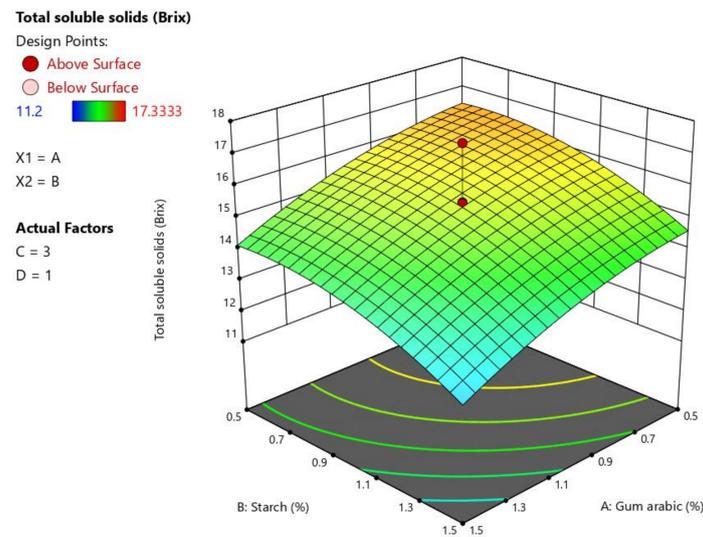


Figure 3. Response surface plot for total soluble solids (°Brix) of coated “Wonderful” pomegranate as a function of gum arabic and maize starch and concentration.

3.4. Titratable Acidity

Table 3 and Table 4 show the regression coefficients from RSM analysis and experimental data for TA, respectively. The Model F-value of 37.43 specifies the significance ($p < 0.05$) of the model with a 0.01% chance for noise at the F-value. Model terms A, B, AB, AC, A^2 , B^2 , and C^2 were all significant ($p < 0.05$) and the “Lack of Fit” (F-value = 0.38) was not significant relative to the pure error. The Predicted R^2 of 0.8694 was reasonably agreed with the adjusted R^2 of 0.9095 (difference < 0.2). The “Adeq Precision” was 17.611, indicating an adequate signal and eligibility to navigate the design space. As shown in Figure 4, an increase in both GA and MS concentrations resulted in decreased TA of aril juice. The 3D surface plot (Figure 4), showed a steeper slope for changes related to MS than GA concentration. The predictive model in terms of actual factors for weight loss is indicated in Equation (6):

$$\text{Titratable acidity (\% CA)} = 0.954444 + 0.356944A + 0.831111B + 0.152778C - 0.441667AB + 0.104167AC - 0.30A^2 - 0.353333B^2 - 0.0433333C^2. \quad (6)$$

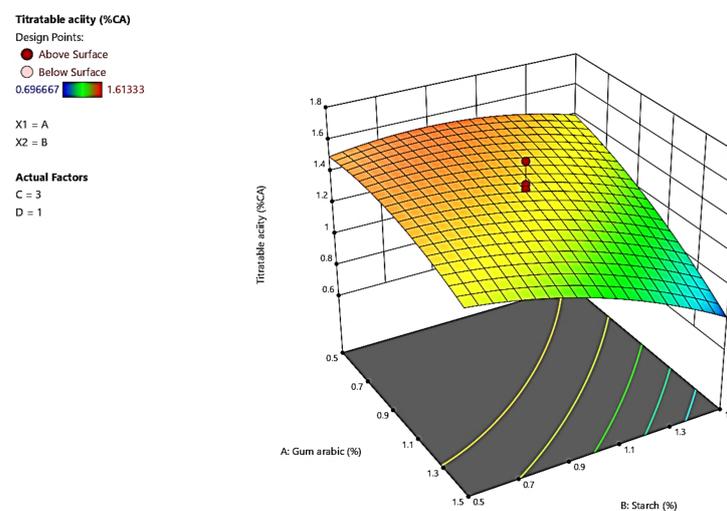


Figure 4. Response surface plot for titratable acidity (% CA) of coated “Wonderful” pomegranate as a function of gum arabic and maize starch and concentration.

3.5. Radical Scavenging Activity

The regression coefficients for the RSM analysis of RSA of PJ are shown in Table 3. Table 4 shows the experimental data obtained for all response variables including RSA. The Model F-value of 35.37 denotes that the model was significant ($p < 0.05$). Model terms A, B, AB, AC, AD, A^2 , B^2 , and C^2 were significant ($p < 0.05$). The “Lack of Fit” F-value of 0.34 showed that the “Lack of Fit” was not significant relative to the pure error. The Predicted R^2 of 0.8866 agreed with the Adjusted R^2 of 0.9222 (difference < 0.2). The Adeq Precision was 18.791, implying an adequate signal for the model to navigate the design space. As shown in Figure 5, MS concentration exerted a greater effect on RSA of PJ than GA, with RSA decreasing as MS concentration was elevated. The predictive model in terms of actual factors for RSA is indicated in Equation (7):

$$\begin{aligned} \text{Radical scavenging activity (mMAAE/mL PJ)} = & 46.76500 + 7.90907A + \\ & 34.72130B + 5.58030C - 8.27475D - 20.10375AB + 3.78861AC + 6.90255AD - \\ & 11.04867A^2 - 13.62708B^2 - 1.54958C^2. \end{aligned} \quad (7)$$

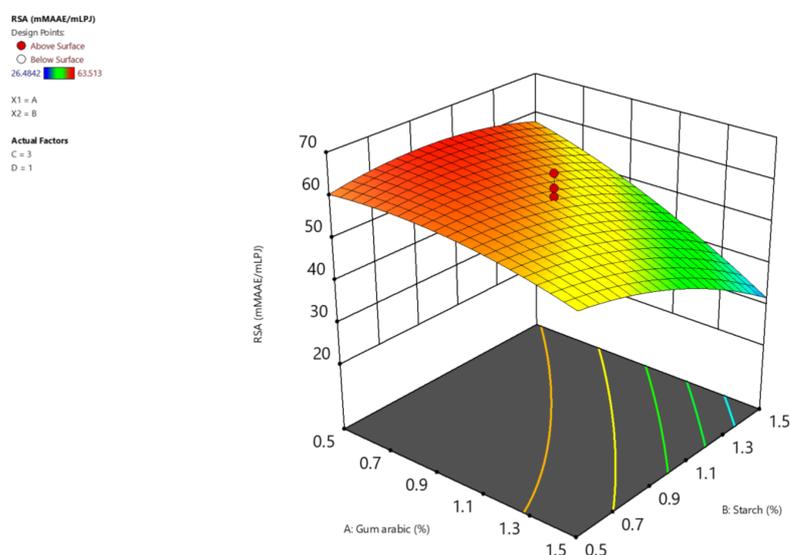


Figure 5. Response surface plot for radical scavenging activity (mMAAE/mL PJ) of coated “Wonderful” pomegranate as a function of gum arabic and maize starch and concentration.

3.6. Ferric Reducing Antioxidant Power

Table 3 and Table 4 show the regression coefficients from RSM analysis and experimental data for response variables including the FRAP of PJ, respectively. The Model F-value was 50.50, implying that the model was significant ($p < 0.05$) and all model terms (A, B, AB, AC, A^2 , B^2 , C^2) were significant ($p < 0.05$) with values less than 0.1. The “Lack of Fit” F-value of 0.26 denoted that the “Lack of Fit” for the model was not significant relative to the pure error. The Predicted R^2 of 0.9095 agreed with the Adjusted R^2 of 0.9318 (difference < 0.2). The ‘Adeq Precision’ for the model was 20.034, indicating an adequate signal for the model to navigate the design space. Similar to Figure 4, the 3D surface plot (Figure 6) showed that change in MS concentration imposed a greater effect on FRAP of PJ compared to GA. The results showed that MS concentration was negatively related to FRAP values like observation for RSA. The mathematical equation in terms of actual factors for FRAP is indicated in Equation (8):

$$\begin{aligned} \text{Ferric ion reducing antioxidant power (mMAAE/mL PJ)} = & 23.87305 + \\ & 12.39678A + 25.50037B + 3.85163C - 14.48650AB + 2.79638AC - 8.12226A^2 - \\ & 9.70879B^2 - 1.16259C^2. \end{aligned} \quad (8)$$

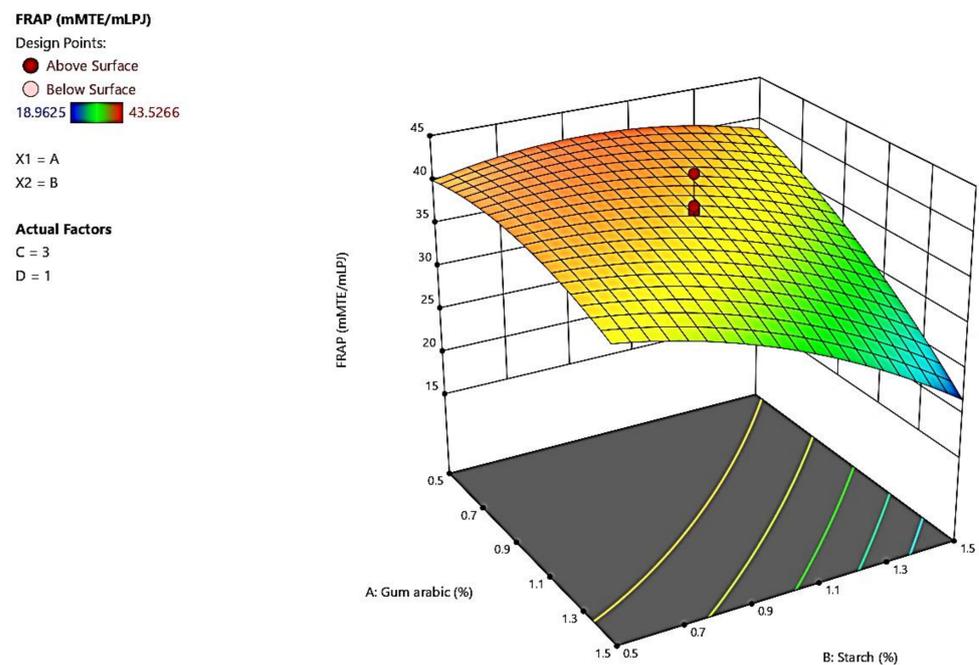


Figure 6. Response surface plot for ferric reducing antioxidant power (mMTE/mL PJ) of coated “Wonderful” pomegranate as a function of gum arabic and maize starch and concentration.

3.7. Optimization

The optimization procedure followed was (i) minimizing weight loss, respiration rate, and concentration of MS and GA; (ii) maximizing antioxidant capacity (RSA and FRAP); (iii) targeting 3% (*v/v*) of lemongrass oil; (iv) limiting glycerol concentration (range of 0.5 to 1.5% *v/v*), total soluble solids (range of 11.2 to 17.3), titratable acidity (range of 0.697 to 1.613). The best coating formulation with the highest desirability of 0.614 was given as GA (0.5%), MS (0.5%), LO (3%), and GC (1.5%). The predicted values of response variables, for this edible coating as calculated from formulas were, weight loss (%) = 5.66, TSS (°Brix) = 16.45, TA (% CA) = 1.50, RSA (mMAAE/mL PJ) = 58.13, and FRAP (mMTE/mL PJ) = 40.03. The final mathematical models were tested for adequacy using one sample t-test to compare experimental with predicted values. The results showed no significant ($p < 0.05$) differences between the experimental and predicted values (Table 5). There was corroboration between the two values and verified the mathematical models’ adequacy fitted by RSM.

Table 5. The predicted and experimental values of responses of “Wonderful” pomegranate coated with optimum formulation of gum arabic, maize starch, lemongrass oil, and glycerol.

Response Variable	Predicted Value ^a	Experimental Value ^a
TSS (°Brix)	11.88	11.37 ± 0.67
TA (% CA)	0.78	0.70 ± 0.06
Weight loss (%)	3.25	3.49 ± 0.27
RSA (mMAAE/mL PJ)	31.26	26.48 ± 1.98
FRAP (mMAAE/mL PJ)	21.59	18.96 ± 1.04

^a No significant ($p < 0.05$) difference between predicted and experimental values.

4. Discussion

4.1. Weight Loss

The results suggest that the concentration of both MS and GA significantly influenced reduced weight loss when applied as surface coatings on “Wonderful” pomegranate fruit. Several studies have shown that GA and MS-based coatings inhibit migration of moisture from the fruit surface into the environment, thus reducing weight loss [23,58–60].

4.2. Respiration Rate

As surface barriers, edible coatings reduce gas interchange, resulting in low oxygen availability for respiration rate in fruit [61,62]. Gum arabic and MS coatings both exhibit good gas barrier properties and the observed decrease in respiration rate as concentration of GA and MS increased corroborates with results reported by Meighani et al. [49] for coated “Malase Torshe Saveh” pomegranates. Similarly, the results were consistent with Varasteh et al. [56], who found that prestorage application of chitosan (1 and 2%) coatings reduced respiration rate of “Rabbab-e-Neyriz” pomegranate after 90 days of cold storage (2 ± 0.5 and 5 ± 0.5 °C).

4.3. Total Soluble Solids

Consumption of soluble solids as substrates of respiratory metabolism or their transformation into sugars is often associated with their gradual decline during ripening and storage [63–65]. These results suggest that both MS and GA significantly reduced the decline of soluble solids when applied on fruit. Studies have proved that application of GA and MS-based coatings inhibits changes in TSS during storage for mangoes [66], cucumber [67], and other fruit types [68–70]. The reduction in respiration rate for coated fruit slows down the conversion of carbohydrates to sugar, reducing synthesis and depletion of metabolites [71].

4.4. Titratable Acidity

Contrary to the findings from this study, several studies report that the application of edible coatings reduces the respiration rate of the fruits and delay the consumption of organic acids [45,65,72,73]. Hernández-Guerrero et al. [74] reported that mango “Ataulfo” fruit coated with starch-based edible coatings recorded the highest acidity values during storage. However, TA values decreased across all coated treatments as storage duration increased. This may suggest that the surface coatings did slow down biological processes associated with the use of organic acids in pomegranate, leading to a decrease in TA.

4.5. Radical Scavenging Activity

The results suggest that, despite reducing the loss of antioxidant capacity, application of edible coatings only limited processes associated with decrease in antioxidant capacity which may include depletion of phenolic compounds and accumulation of reactive oxygen species under cold storage [62,75]. In agreement with the results, Chiabrando and Giacalone [76,77] found that antioxidant activity of coated and un-coated “Berkeley” and “O’Neal” blueberries decreased continuously during 45 days of cold storage (0 °C). Similarly, Ghasemnezhad et al. [3] observed a decrease in antioxidant capacity of PJ from pomegranate arils treated with chitosan coatings during cold storage, with the decline being lower in coated arils. In contrast, Dávila-Aviña et al. [7] reported that carnauba and mineral oil application presented higher antioxidant capacity values in tomatoes because of high ascorbic acid accumulation during 28-day storage at 10 °C.

4.6. Ferric Reducing Antioxidant Power

Like RSA, the results suggest that the application of coatings only limited FRAP loss but did not halt the decrease in antioxidant capacity, which is related to loss of phenolic compounds and accumulation of reactive oxygen stress species under stress conditions of cold storage [5,78].

5. Conclusions

In this study, various composite edible coating formulations were developed to improve postharvest quality of “Wonderful” pomegranate from gum arabic, maize starch, lemongrass oil, and glycerol using response surface methodology. Mathematical models were developed with response variables including weight loss, respiration rate, total soluble solids, titratable acidity, and antioxidant capacity. Change in concentration of gum

arabic and maize starch mainly reduced weight loss, increased total soluble solids, titratable acidity, and reduced loss of antioxidant capacity. The study showed that GA (0.5%), MS (0.5%), LO (3%), and GC (1.5%) as the optimum concentration for the best coating formulation. Response surface methodology has a potential for optimizing composite edible coatings as a postharvest treatment. However, future studies are required to determine the effects of the optimized coating on physiological responses, physicochemical properties, and antioxidant capacity of pomegranate during storage and shelf life.

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