

# 1. Introduction

Strawberries are one of the most popular and widely grown fruits worldwide, because of their sweet juicy taste and health benefits. They are very rich in vitamin C, manganese, folate and phenolic substances, which is mostly antioxidant substances, which benefit heart health and blood sugar control [1]. Most countries have their own variety of strawberries that are grown on different climate, altitude, day length, production type [2]. Strawberries are produced for both immediate consumption and for processing as frozen, preserved berries. Given they are very perishable in nature, they are grown near the center consumption and processing area so that the loss of phytochemical compounds is minimized [2]. In this regard, post-harvest practices must be adapted to preserve the quality and composition during storage; for example, use of edible coating to prevent these changes and to increase the shelf life.

Edible coatings are an edible layer formed to coat the surface of food products. This layer is composed of biopolymers and acts as a barrier between food products and the atmosphere [3]. This barrier prevents transportation of gas and moisture with the atmosphere and delays the ripening process, slowing down fruit respiration [3]. This decrease in moisture loss prevents weight loss, and the decrease in respiration slows down the loss of polyphenols and antioxidants [4].

Several different biopolymers, such as starch, pectin, carrageenan, alginate, chitosan, and xanthan gum are extensively used as coating materials [5]. Application of edible coating seen in industries and already being used for many fruits such as apple, grapes, mangoes, strawberries etc. However, the problems still exist for choosing best biopolymers and optimal concentrations to be used; since each polymer adhere different properties for protection, adhesiveness that can affect food matrices [6].

Alginate is a polysaccharide made from brown seaweed; it is a nontoxic, biodegradable, low-cost. alternative that has potential to be used as coating material [7]. However, it can form a porous structure with poor barrier properties that can fasten the ripening process. To avoid this. alginate is often used with other polysaccharides such as guar gum, pectin, galactomannans etc [7].

Guar gum is a galactomannan rich flour and water-soluble polysaccharide which is obtained from the leguminous Indian cluster bean *Cyamopsis tetragonoloba* (L.) Taub [8]. It is largely used in the form of guar gum powder as an additive in food, pharmaceuticals, paper, textile, explosive, oil well drilling and cosmetics industry. It is often used as stabilizer, emulsifier and thickener in various food products and contributes to soluble dietary fiber (SDF) portion of seed total dietary fiber (TDF). However, studies that use guar gum for its functional properties with alginate in nonexistent [9].

Pectin is a class of complex water-soluble polysaccharides used to form coatings. It is a purified carbohydrate product obtained by aqueous extraction of some edible plant material, usually citrus fruits or apples [10]. Under certain circumstances, pectin forms gels; this property has made them a very important

additive in jellies, jams, marmalades and confectionaries, as well as edible coatings. Pectin is a high-volume and potentially important food ingredient available in high percentages in agricultural wastes. In addition, its nutritional benefits for human health and its pharmaceutical activities make it interesting to use in a variety of food products [11].

Simplex Centroid mixture design is to determine combinations of different polymers with food gelatin to produce edible coatings and evaluate their effect on the physicochemical and bioactive properties of coated strawberries.

This study used different combinations of Alginate, Guar Gum, and Pectin as coating materials.

## 2. Materials and Methods

### 2.1 Raw Materials Collection

Strawberry was collected from local market of Sylhet, Bangladesh. Alginate, Guar gum and Pectin was purchased from Loba Chemie Pvt.Ltd, India respectively. Gelatin was purchased from Loba Chemie Pvt.Ltd, India. All chemicals were stored at room temperature on the laboratory.

### 2.2 Simplex-Centroid Mixture Design

The optimal mixture of different biopolymers to be used as a coating on Strawberry was evaluated using a simplex-centroid mixture design. Four treatments were performed, with the components (alginate, Guar Gum, Pectin) evaluated at 3 concentrations: 3%, 1.5%, 0%.

**Table 1:** Matrix of the simplex-centroid mixture design and the biopolymer combinations (coded and real values) used to produce the edible coatings.

Runs	Alginate	Guar Gum	Pectin
Treatment 1	3%	3%	0%
Treatment 2	3%	0%	3%
Treatment 3	3%	1.5%	1.5%

### 2.3 Preparation and Application of Edible Coating

Polymer samples were dissolved in distilled water at 70 °C and stirred in magnetic stirrer until complete dissolution, at concentrations defined by simplex centroid method. Food gelation (2%) was added to all filmogenic solutions to facilitate adherence of polymers in the surface of the strawberry. Strawberries were dispersed into filmogenic solution and drained in sterilized stainless-steel sieve. All samples were disposed

in the sterilized stainless-steel sieve at 25-30 °C for 6 days, then submitted to physicochemical and bioactive analysis. For control assay, the strawberries were submerged into 2% gelatin solution for 3 minutes and then stored at same conditions as for the treatment assays. For analysis, whole strawberries were blended with juice blender to make puree. The puree was used for all analysis, which was carried out at 0, 2, 4, 6 days of storage (except for weight loss, which was carried out daily).

## **2.4 Physicochemical Analysis**

### **2.4.1 Weight Loss**

Pre weighted polypropylene trays containing samples were weighted on daily basis using a semi analytical balance. Sample weight loss was calculated according to the equation

$$\text{Weight Loss (\%)} = \left( \frac{\text{Initial Weight} - \text{Final Weight}}{\text{Initial weight}} \right) \times 100$$

### **2.4.2 Total Soluble Solids**

Total soluble solid was measured using analog refractometer at a scale between 0 to 35° Brix directly on the juice sample.

### **2.4.3 pH**

The pH was measured using a digital pH meter. In between each take the meter was washed with distilled water. Then submerged into buffer solution then washed with distilled waster again before taking measurements again.

## **2.5 Bioactive Analysis**

### **2.5.1 Total Phenoloc Content**

The total Phenolic Content was determined using Folin-Ciocalteu method [12]. Aliquots of 60 µL from each sample were mixed with 300 µL of Folin-Ciocalteu solution and mixed well for 8 minutes. Then 900 µL of Sodium Carbonate solution were added to the mixture. The test tubes containing mixtures were incubated at 37 ° temperature for 40 minutes in the dark. After the incubation period the absorbance of the mixture was measured at 765nm wavelength using a spectrophotometer (Shimadzu, Japan). The blank solution was prepared by the exact same procedure except for the sample proportion of water added instead of sample. The standard curve was prepared using different concentrations of gallic acid solutions.

### **2.5.1 DPPH Assay**

The antioxidant activity of the sample was evaluated through DPPH radical scavenging and performed using a spectrophotometer at 517nm wavelength, a method described by Brand-Williams et al. (1995), with slight modifications [13].

Strawberry extract aliquots of 100  $\mu$ L were mixed with 1.4ml of DPPH solutions. Then the mixtures were incubated for 30 minutes in dark condition before measuring absorbance. For blank solution the extract was replaced by water.

### 3. Results and Discussion

#### 3.1 Characterization of fresh Grapes

The fresh strawberry sample had a mean value of 5.25° Brix for total soluble solids, 3.33pH. The total phenolic content was 520.55 mg/100g and the antioxidant activity determined 0.43  $\mu$ mol TE/g.

Table 3.1: Physicochemical and Bioactive properties of Raw Strawberries

Parameters	Fresh Strawberry
<b>Physicochemical Profile</b>	
Total Soluble Solid (°Brix)	5.25 ° brix
pH	3.33
<b>Bioactive Properties</b>	
Total Phenolic Content (mg/g)	399.25 mg/g
DPPH	0.42 $\mu$ mol TE/g

#### 3.2 Weight Loss

Percentage weight loss of coated strawberries is shown in fig1. All assays showed significant difference in weight loss by the end of 12 days period. According to [14] weight loss is associated with moisture evaporation and increases at high temperature during storage.

The control assay showed the highest weight loss compared to others, with a significant increase from 5.58% at day 3 to 9.93% on day 5. The high weight loss is due to the high storage temperature (30 ° Celsius), which promoted rapid evaporation. Also, the control assay was only composed of gelatin (2%) which didn't provide any significant protection against evaporation. On the other hand, the coated samples showed less weight loss indicating effective barrier properties.

Samples in treatment 1, 2 and 3 showed 9.52%, 7.02%, 5.81% weight loss by the end of 6 days period. Amongst them treatment 3 (3% alginate + 1.5% guar gum + 1.5% pectin) had the lowest weight loss. It indicates that, incorporation of guar gum and pectin with alginate in gelatin matrix forms a protective barrier against temperature, able to decrease weight loss in strawberries.

The use of guar gum and pectin together with alginate as coating on strawberries have not yet been reported by any literature, with this being the first. On the other hand, some authors have reported their use of guar gum on different fruits and promoted lower loss (15%) during 20 days of storage [15]. These studies prove usage of guar gum as potential coating alternatives.

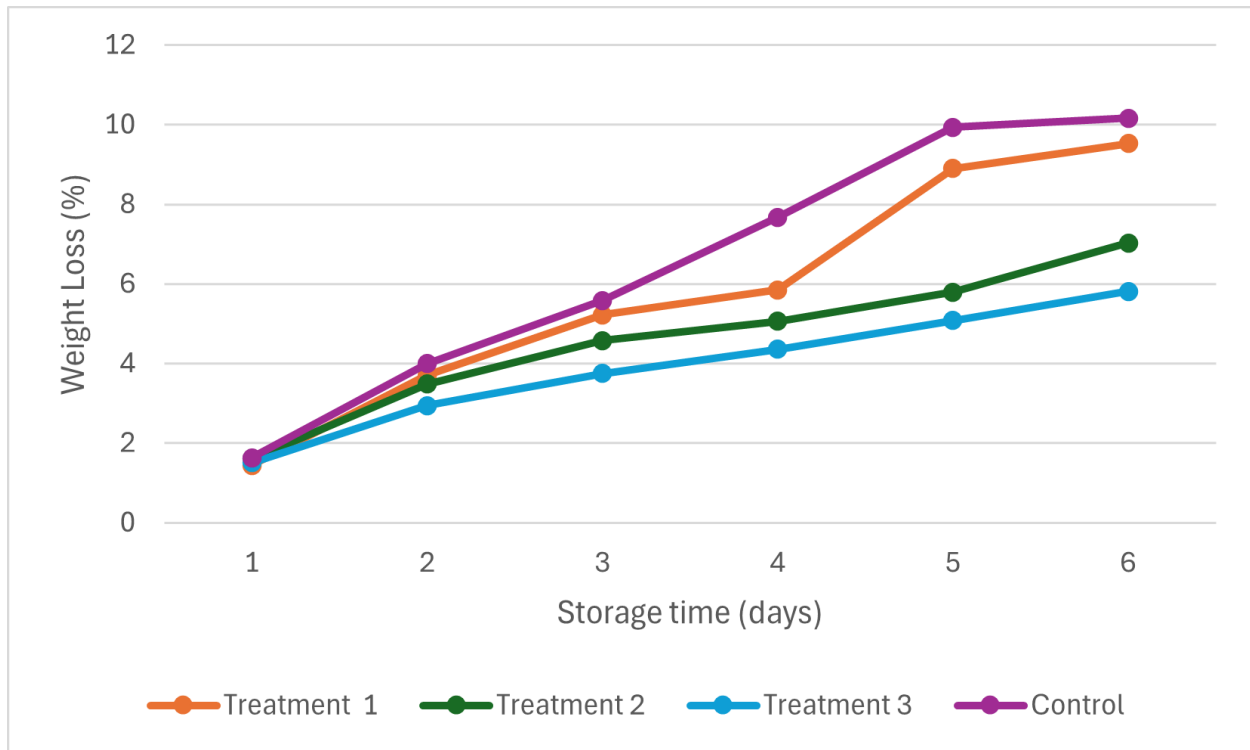


Fig 4.1: Weight loss data

### 3.3 Total Soluble Solids (TSS), pH

The results of physicochemical analyses are shown in fig 4.2. A statistical difference ( $P < 0.05$ ) was observed for TSS in all treatments, indicating that that parameter was affected by the edible coatings during storage time. Edible coatings affect the TSS content because they impair respiratory and metabolic processes of fruits and delay ripening [16].

Total soluble solid of treatment 1 and 2 increase from day 2 to day 3 and then reduced at day 6. Treatment 1 shows the highest loss in tss content during the storage time. It indicates that treatment 1 couldn't stop respiration and ripening. For control the total soluble solid content dropped continuously from day 2 (5.5°

brix) today 6 (4.4° brix). The highest retain in soluble content is found from treatment 2. it fell from (5.2° brix) to (4.8° brix) during the storage time.

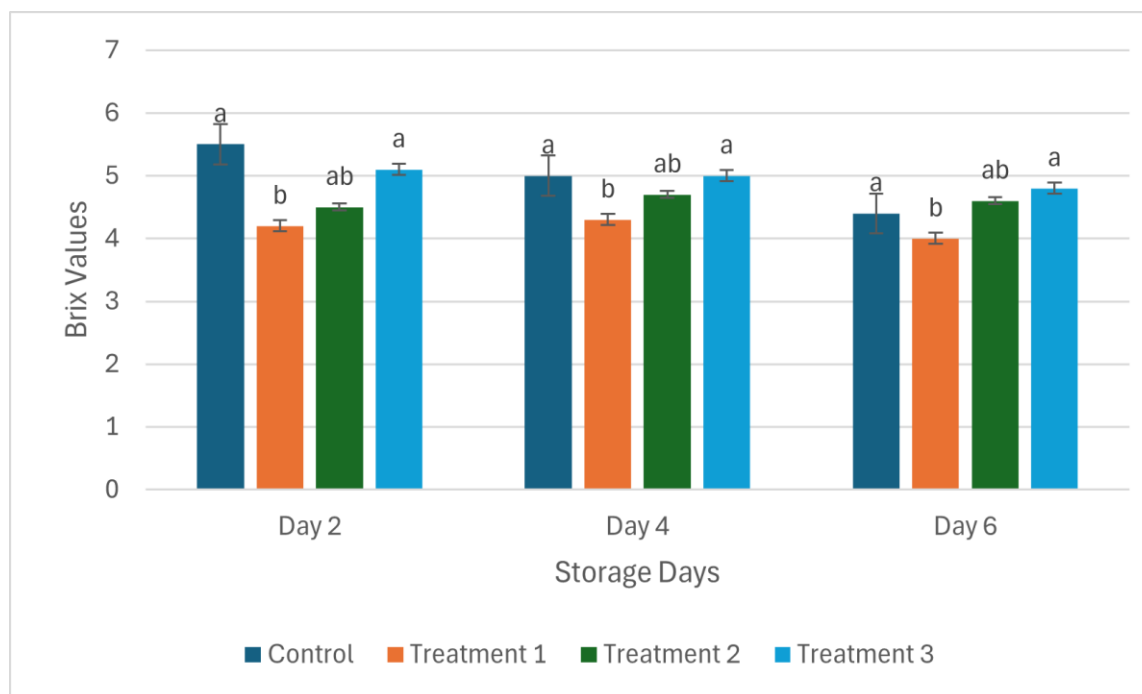


Fig 4.2: Total Soluble Solid content of different samples (Value with the same letter in each storage days were not significantly different  $P > 0.05$ )

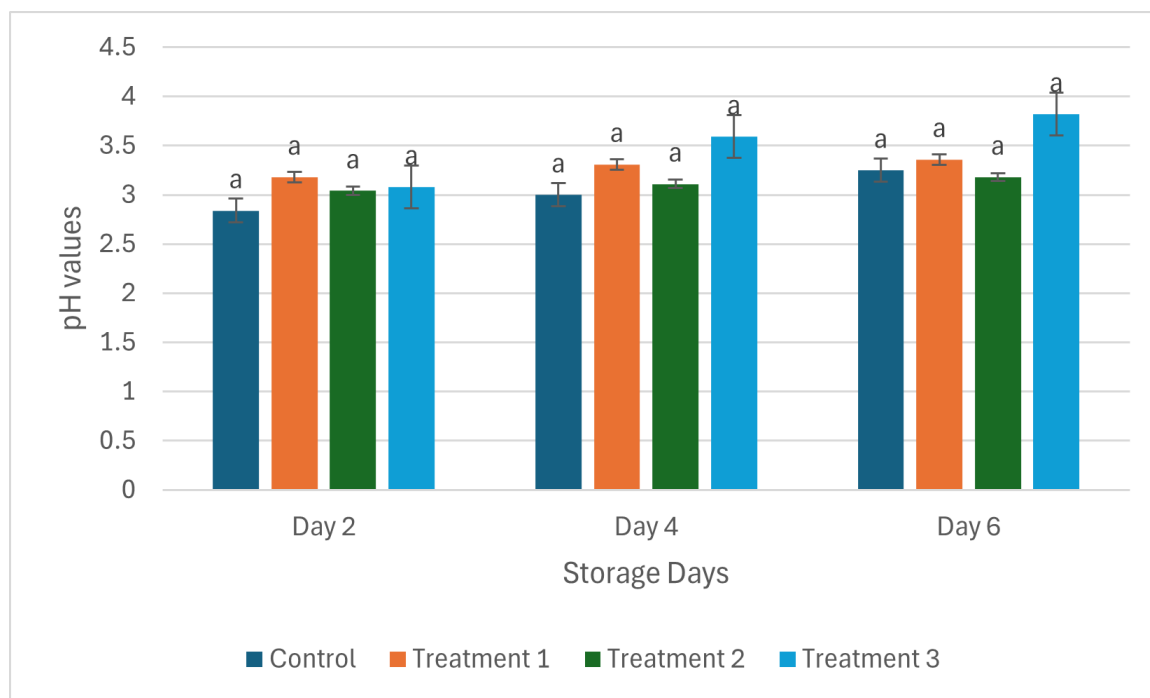


Fig 4.3: pH values of different samples (Values with the same letter in each storage days were not significantly different  $P>0.05$ )

### 3.4 Bioactive Properties: TPC, DPPH

Result for total phenolics is shown in (table). Phenolic compounds are positively correlated with the antioxidant activity of grapes. These compounds are distributed in different parts of the grape, such as skin, pulp, pedicle. They can be affected by many parameters such as variety, level of fertilization, or date of planting [17].

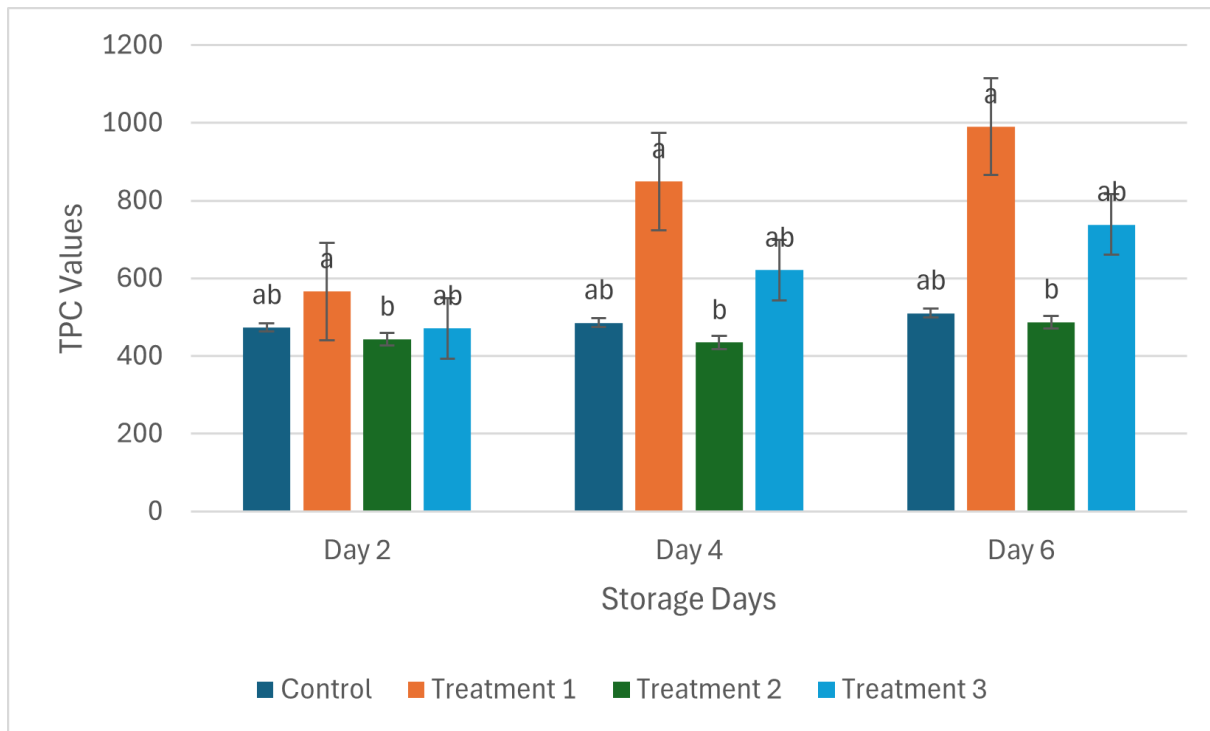


Fig 4.4: Total phenolic content (TPC) value of different samples (Values with the same letter in each storage days were not significantly different  $P>0.05$ )

The phenolic content varied during storage in all treatments. The phenolic content in treatment 2, 3, and control gradually increased during storage, presenting a significant difference ( $p<0.05$ ). On the other hand, for treatment 1 the phenolic content decreased at day 4 and increased at day 6. This behavior was also observed by other scientists. The author reported that these changes are due to the inability of some coating to control ripening process and the activity of polyphenol oxidase and peroxidase. After day 6 days of storage, the sample in treatment 1 showed the highest level of total phenolic content (989.65 mg/100g). The increase in phenolic content can be associated with the loss of cell membrane integrity in the fruits; leading to cellular structure disruption and releasing enzymes involved in phenolic substrates. The high loss of total soluble solids indicates the release of polyphenolic compounds. Fruits present low or no energy source

(starch), because necessary, the use of the sugars present in the fruit as an energy source for respiration, which results in a reduction of the fruits SS content [18]. These results indicate that this treatment did not control ripening process and, therefore, not advised for coating strawberries. The control and Treatment 1 showed highest loss of DPPH value from 0.38 to 0.27  $\mu\text{mol TE/g}$ .

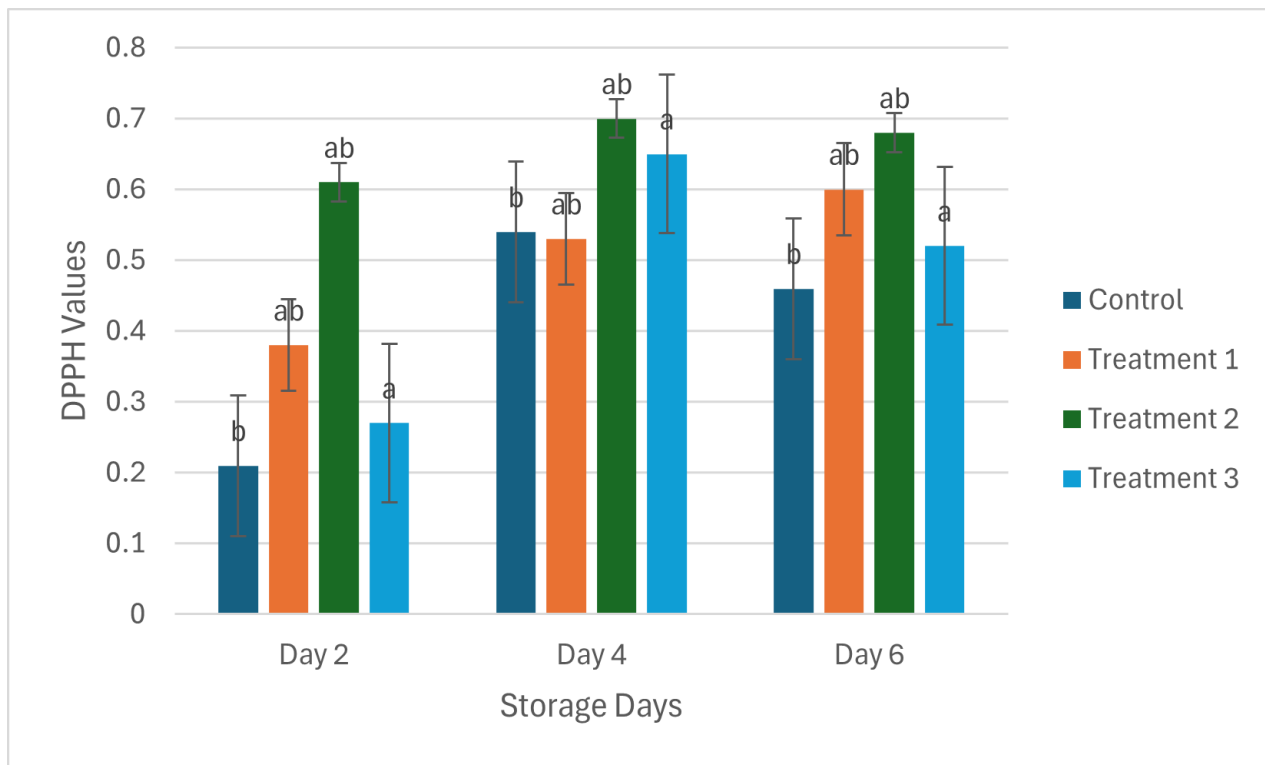


Fig 4.5: DPPH value of different samples (Values with the same letter in each storage days were not significantly different  $P>0.05$ )

Treatment 3 showed the highest level of polyphenolic content (989.65 g/100g) at the end of the storage time and high antioxidant capacity, DPPH radicals (.81  $\mu\text{mol TE/g}$ ). This coating is composed of alginate (3%), guar gum (1.5%), pectin (1.5%) and gelatin (2%). It demonstrated that use of alginate combined with natural gum and pectin may improve the antioxidant potential of strawberries.

## 4. Conclusion

The investigations into different chemicals and gums, such as guar gum, pectin along with alginate indicated that they can be effectively used for edible coating preparation. Also, they are low cost, abundant and easy to source. They also slow down the maturation process of strawberries, as well as control physicochemical and bioactive properties.



The use of a mixture design to define the polymer concentrations was an excellent tool to optimize the most adequate combinations to provide a protective effect on coated strawberry. The combination of alginate (3%), Guar Gum (1.5%), Pectin (1.5%), Gelatin (2%) was the best formulation; controlled weight loss, firmness loss, maintained good pH levels, improved phenolic activities (TPC) and promoted antioxidant potential.

The coating failed to provide any antimicrobial resistenty. Furthur study can be done to combine biocides with the presented coatings to increase the functionality.

Based on the promising result reported in this work the use of these polymers in edible coating preparation is encouraged, and maybe used in other fruits.

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