Original Paper

# Efficacy of biosurfactant as a coating material for post-harvest preservation of jamun fruit (*Syzygium cumini*)

# Jyoti Solanki\*, Sayali More, Kaushik Jodhani, Dhaval Patel, M. Nataraj

Post Graduate Department of Bioscience, Sardar Patel University, Satellite campus, Bakrol-Vadtal Road, Bakrol 388 315, Anand, Gujarat, India

\*Corresponding author, E-mail: jytsolanky269@gmail.com

### Abstract

*Syzygium cumini* L., Indian blackberry, popularly known as 'jamun', is an extremely perishable fruit with high antioxidant, nutritive and medicinal values. The present study investigated the efficacy of biosurfactant alone and/or in combination with gum ghatti and xanthan gum as edible coating to enhance shelf life and quality of jamun fruit. Coating treatments included 3% gum ghatti (T1), 0.1% xanthan gum (T2), 2% biosurfactant (T3), 3% gum ghatti and 2% biosurfactant (T4), and 0.1% xanthan gum and 2% biosurfactant (T5). Fruit quality was checked at regular intervals by evaluating physiochemical parameters and enzyme assays. The coating treatments reduced weight loss and decay percentage of jamun compared to the control. The increase of total soluble solid concentration was less in fruits treated with gum ghatti ans well as xanthan gum and biosurfactant than in the control. Moreover, sugar (glucose, fructose and sucrose) concentration of phenolic substances increased in fruit treated with biosurfactant alone and in combination with agum. Concentration of ascorbic acid remained unchanged in fruit coated with biosurfactant. Activity of cell wall degrading enzymes was reduced in biosurfactant-treated fruit throughout the storage period. Conclusively, biosurfactant can be used as a promising edible coating, as it delays metabolic changes, maintains firmness and quality of jamun, and therefore increases the shelf life of jamun during postharvest storage.

**Key words:** biosurfactant, edible coating, glycoprotein, jamun, post-harvest preservation. **Abbreviations:** BS, biosurfactant; CAT, catalase; CEL, cellulase; GG, gum ghatti; PG, polygalacturonase; PME, pectin methyl esterase; POD, peroxidase; SOD, superoxide dismutase; ROS, reactive oxygen species; TA, titratable acidity; TSS, total soluble solid; XG, xanthan gum.

### Introduction

Indian blackberry (Syzygium cumini L.), a subtropical under-utilized fruit tree of the Indian subcontinent, belongs to the family Myrtaceae. Some common names of the Indian blackberry fruit are 'jamun', 'jambulan', 'black plum' and 'java plum' (Khaliq et al. 2020). Sweet, tasteful and juicy jamun fruit are commercially available for a short period i.e., from late June to middle of the August, which increases demand in the market (Vandana et al. 2015). The wide popularity of jamun fruit is due to its health beneficiary constituents such as phenolic substances anthocyanins, tannins, flavonoids and other biologically active compounds (Archana et al. 2019). Jamuns are extremely perishable after harvest and deteriorate rapidly within two to three days at ambient temperature due to their high moisture and sugar content (Patil et al. 2012). Moreover, local vendors sell fruit unhygienically on roadsides, leading to rapid deterioration (Vandana et al. 2019). In recent years, public awareness about the health

benefits of jamun make them admired. However, its short shelf life after harvest is a major hindrance for sufficient supply, which results in a high price.

Various types of synthetic compounds, elicitors and growth regulators have been used solely or in combination to delay ripening and to reduce the post-harvest loss of fruit (Salunkhe et al. 1991). The excessive use of chemicals in packaging and preservation results in bioaccumulation and biomagnification, possibly leading to health hazards (Adetunji et al. 2018). Therefore, there is a need for safe and environment-friendly alternatives that extend postharvest shelf life and maintain fruit quality. Several studies have been documented so far on preservation of jamun fruit, which include modification of atmosphere packaging (Rai et al., 2011), coating with calcium chloride (Vandana et al. 2015), zein based coating (Baraiya et al. 2015), polysaccharide based coating (Gol et al. 2015), coating of putrescine (Archana et al. 2019), Aloe vera gel coating (Vandana et al. 2019), chitosan coating (Saurabh et al. 2019b) and guggul gum coating (Khaliq et al. 2020).



Environmental and



Each technique has its own pros and cons. For instance, during storage in a modified or controlled atmosphere, CO<sub>2</sub> accumulates, which can cause off-flavour due to anaerobic respiration (Bender et al. 2000). Protein coating exhibits good gas permeability and mechanical properties, but it suffers from low moisture permeability (Umaraw et al. 2020). Moreover, there is relatively high probability of causing allergenic reactions and thus limiting their use (Bourtoom, 2008). Lipid based coatings are not easily applied to the surface of fruits and vegetables because of their greasiness and thickness. In addition, lipid-based coatings poorly adhere to food products with hydrophilic surfaces (Dhall 2013). Thick coating can limit O<sub>2</sub> exchange and cause off-flavour development (Smock 1940). Some of these edible coatings are hygroscopic in nature, which results in increased chances of higher microbial growth.

Edible films and coatings can carry various active agents, such as emulsifiers, antioxidants, antimicrobials, nutraceuticals, flavours, and colorants to enhance food quality and safety (Han 2002). Emulsifiers or biosurfactants are surface-active agents of amphiphilic nature that are capable of reducing the surface tension at the airwater interfaces and the interfacial tension at oil-water interfaces (Marchant, Banat 2012). They modify surface energy to control the adhesion and wettability of the film surface (Krochta 2002). They have good moisture barrier properties and are important for preventing physiological deterioration of food products (Umaraw et al. 2020). Exploration of biosurfactants as edible coating materials to extend the shelf life of jamun fruit has not been studied previously. Biosurfactants are driving much attention in food processing industries due to their unique properties like low or non-toxicity, biodegradability, biocompatible, antimicrobial and/or antifungal activities (Nitschke and Costa 2007).

The present study aimed to check the efficacy of biosurfactant as a coating material on jamun, a perishable fruit, alone and in a combination of gum ghatti and xanthan gum, to extend their shelf life.

### **Materials and methods**

### Bacterial culture

*Dyadobacter fermentans* JDP9 (GenBank accession number: MZ266577.1) was used as a producer of glycoprotein biosurfactant. JDP9 was isolated from automobile workshop soil (Anand, Gujarat, India N 22°54' and E 72°95') and was stored in 40% (v/v) glycerol at -80 °C and a working stock was maintained on trypticase soya agar slants at 4 °C.

# *Production and purification of biosurfactant from* D. fermentans *JDP9*

For inoculum preparation, a well grown isolated colony of JDP9 was inoculated in trypticase soya broth, and incubated at 30 °C till optical density ( $OD_{600}$ ) reached 1.00.

Later, 1% (v/v) of the inoculum was added in Bushnell-Haas medium supplemented with 1% sucrose (pH 7.0) for biosurfactant production. The fermentation process was carried out in an incubator shaker at  $3 \times g$  and 30 °C for 4 days. After completion of fermentation, cell-free broth was obtained by centrifugation at 4427 × g for 10 min. The cell free broth was acidified with 6 N HCl and kept at 4 °C till visual precipitates appeared. Precipitates were collected by centrifugation at 12 298 × g for 10 min at 4 °C in a cooling centrifuge (Eppendorf Centrifuge 5430 R). The pellets were re-suspended in distilled water. Biosurfactant was further purified by gel permeation chromatography. The glass column (300 × 10 mm) contained Sephadex G-50 as a stationary phase. Phosphate buffer (pH 7.0) was used as a mobile phase at 1 mL min<sup>-1</sup> flow rate.

### Coat forming capacity of biosurfactant

Various concentrations of biosurfactant (0.2, 0.5, 1.0, 2.0 and 5.0% w/v) were prepared by dispersing biosurfactant precipitates in 10 mL of distilled water and stirring for 15 min using a magnetic stirrer (Genei, India). The coat forming capacity was checked by pouring on an inverted Petri dish. Solutions were allowed to dry at ambient temperature for 30 min. The plates were observed for thin layer formation of biosurfactant.

### Edible coating preparation

For preparation of edible coating, biosurfactant (BS), gum ghatti (GG) and xanthan gum (XG) were used alone as well as in combination. Biosurfactant (2%) was prepared by dissolution of 2 g of biosurfactant in 100 mL deionized water followed by stirring on a magnetic stirrer (2 MLH, Remi Equipment, India) for 30 min. Gum ghatti solution (3%) was prepared by adding 3 g gum ghatti powder in 100 mL of deionized water. Similarly, 0.1% xanthan gum was prepared by mixing 0.1 g xanthan gum in 100 mL deionized water until the solution became clear. Other mixtures were prepared by adding a similar amount of biosurfactant and gum ghatti as well as biosurfactant and xanthan gum. The resultant mixture was then filtered through muslin cloth to remove impurities and undissolved solid particles. Glycerol at concentration 0.75% was added to improve plasticity in all edible coatings.

### Collection of fruit

Jamuns were collected at physiological maturity from an orchard near Vallabh Vidyanagar, Anand, Gujarat, India. Freshly harvested fruit were transported to the research laboratory within an hour.

# Edible coating treatments and experimental design

The harvested fruit were washed with tap water to remove dust and unwanted debris, followed by 2.0% sodium hypochlorite treatment for 2 min. Then, the jamun fruit were sorted for their use in this experiment. The selected fruits had similar physical size, appearance, colour and were free from injury. The fruits were washed three times with distilled water to remove the residues of sodium hypochlorite and allowed to air dry at room temperature. The fruit (100 g each) were weighed in 18 plastic containers and divided equally in six groups. Each group was treated with one of the following coating solutions: 3% GG (T1), 0.1% XG (T2), 2% BS (T3), 3% GG + 2% BS (T4), 0.1% XG + 2% BS (T5) and control without coating (C). The experiment for each treated group was performed in triplicate. Application of edible coating treatment was done by dipping jamun fruit in the respective coating solution for 5 min. The treated fruit were kept at room temperature for 30 min for drying and then placed in a storage cabinet at  $10 \pm 2$  °C with 70% relative humidity. The stored fruit were analyzed for weight loss, decay, pH, acidity, soluble solids, sugars, ascorbic acids, phenolic substances, 2,2-diphenyl-1-picrylhydrazyl-scavenging antioxidant activity. anthocyanins, peroxidase activity, superoxide dismutase, polygalacturonase, cellulase and pectin methyl esterase at 4, 8, 12 and 16 days of storage. One group of fruit was analysed immediately after application of the coating and considered as a control (Day 0).

### Weight loss and decay percentage

The weight loss percentages were determined as described by the Association of Official Analytical Chemists (AOAC 1994) using the following formula:

Weight loss (%) = Initial weiht (g) – Final weight (g) / Initial weight (g) × 100.

Decay percentage was determined by the method of El-Anny et al. (2009) using the following formula:

Decay (%)= Weight of decayed fruits (g) / Weight of fruits stored (g)  $\times$  100.

# Measurement of pH, total soluble solids and titratable acidity

For determining the pH, total soluble solids (TSS) and titratable acidity (TA), 1 g fruit tissue was homogenized in 10 mL of distilled water. The pH was measured by a digital pH meter (Systronic Pvt. Ltd., India; Model 335). TSS were estimated by a digital refractometer (Atago Co., Tokyo, Japan). TA was measured by the method suggested by Mazumdar and Majumder (2003). TA was expressed as equivalent to citric acid. The following formula was used to calculate acidity:

*Titratable acidity* (%) =  $d \times 0.064 \times C / a \times b \times 100$ , where *a* is weight of sample, *b* is volume of the aliquot, *C* is make up volume with deionized water, *d* is average burette reading.

### Estimation of sugars using ion exchange chromatography

One gram of jamun fresh tissue was homogenized in 10 mL of ultrapure water followed by centrifugation at 4427  $\times$  *g* for 15 min at 4 °C in a cooling centrifuge to obtain clear supernatant. The supernatant was then filtered through 0.45 µm filter and used for sugar estimation using ion

exchange chromatography. Briefly, monosaccharide and disaccharide concentration was analyzed using a Metrohm anion exchange chromatography system (940 Professional IC Vario, Switzerland) embedded with a Hamilton RCX-30 column (250  $\times$  4.1 mm filled with styrene divinylbenzene copolymer of 7 µm particle size). The column was washed with 200 mM NaOH (GR grade, Sigma Aldrich) as an eluent at a flow rate of 1 mL min<sup>-1</sup> to obtain a linear baseline. A standard sugar mixture of glucose, fructose and sucrose solution (1 mg L<sup>-1</sup> each) was first injected in to system for the reference and retention time as well as peak areas were recorded. Later, samples were passed through a Dosino dosing unit attached with the instrument to remove air bubbles and to obtain continuous flow of the sample though column for analysis. The mobile phase (10 mM sodium acetate in 100 mM NaOH) was injected in the column at 1 mL min<sup>-1</sup> flow rate and at less than 2.14 MPa pressure. Later, 10 µL of filtered sample was injected in the column with the mobile phase for 20 min at 1 mL min<sup>-1</sup> (8.61 MPa pressure and 32 °C temperature) as per the optimized process of standard. The retention time and concentration of sugars present in the sample were determined in the form of a chromatogram automatically generated by the system embedded with a 'Amp Detector-2' (954 Professional Detector Vario 1).

### Concentration of phenolic substances

Briefly, 1 g of jamun tissue was homogenized in 10 mL of 80% methanol and centrifuged at  $3075 \times g$  for 20 min to obtain clear supernatant. Concentration of total phenolic substances was estimated using the method proposed by McDonald et al. (2001). The concentration of phenolic substances was calculated against a standard curve of catechol and expressed as mg g<sup>-1</sup>.

# Determination of an antioxidant activity by 2,2-diphenyl-1-picrylhydrazyl

Antioxidant activity was determined by the method suggested by Ghasemnezhad et al. (2011). Briefly, 1 g of jamun tissue was extracted in 5 mL of 80% methanol followed by centrifugation at 4427 × g for 15 min at 4 °C. The reaction mixture consisted of 100  $\mu$ L supernatant and 3 mL of 0.1 mM 2,2-diphenyl-1-picrylhydrazyl, which was incubated in dark for 30 min and absorbance was measured at 515 nm using a UV-Vis spectrophotometer against methanol as a blank. The scavenging activity was calculated using the following formula and results were expressed as a percentage:

*Percentage inhibition = (Absorbance of blank – Absorbance of test) / Absorbance of blank* ×100.

### Concentration of anthocyanins

One gram of jamun tissue was ground in a mixture of 95% ethanol and 1.5 N hydrochloric acid (85:15, v/v) according to Lees and Francis (1972). The solution was stored at 4 °C overnight. The solution was centrifuged for 10 min at

5600×g and absorbance of the supernatant was measured at 535 nm. The total anthocyanin concentration was expressed as  $\mu g g^{-1}$ .

### Concentration of ascorbic acid

One gram of jamun tissue was blended with 10 mL of 5% meta-phosphoric acid and 2 M glacial acetic acid followed by centrifugation at  $4427 \times g$  for 15 min. Ascorbic acid was estimated from the supernatant using a method provided by Roe (1954). A standard graph was prepared from 1.0% L- ascorbic acid and expressed as  $\mu g g^{-1}$ .

# Extraction and assay of polygalacturonase, cellulase and pectin methyl esterase activity

For extraction, 2 g of fruit tissue was homogenized in 10 mL of 20 mM sodium phosphate buffer (pH 7.0) containing 20 mM cysteine-HCl, 20 mM EDTA and 0.05% Triton X 100. The homogenate was centrifuged at 27  $670 \times g$  for 30 min at 4 °C. The clear supernatant was used to assay activity of polygalacturonase (PG), cellulase and pectin methyl esterase (PME) according to the method suggested by Srivastava and Dwivedi (2000). The total protein estimation was carried out by the method described by Lowry et al. (1951).

For PG activity, reaction mixture consisted 0.2 mL sodium acetate buffer (200 mM, pH 4.5), 0.1 mL sodium chloride (200 mM), 0.3 mL 1% polygalacturonic acid (pH 4.5), and 0.1 mL crude enzyme in a final volume 1.0 mL made up with distilled water, which was incubated at 37 °C for 1 h. Then, 3 mL 3,5-dinitrosalicylic acid was added into the mixture and placed in a boiling water bath for 5 min. In a control tube, substrate polygalactouronic acid was added after heat treatment to avoid any reaction. After incubation in the boiling water-bath, the absorbance was measured at 540 nm. Enzyme activity was calculated against a standard graph prepared from 1% galacturonic acid. One unit of enzyme activity was defined as the amount of enzyme required to liberate 1 nmol of galacturonic acid per min per g of the fresh weight. The enzyme activity was recorded in units mg<sup>-1</sup> of protein.

For cellulase activity, reaction mixture consisted 0.25 mL of 100 mM sodium acetate buffer (pH 5.0), 0.5 mL 1.5% carboxy methyl cellulose and 0.1 mL enzyme extract in a total volume of 1.0 mL made up with distilled water. The reaction mixture was then incubated at 37 °C for 16 h followed by addition of 3 mL 3,5-dinitrosalicylic acid reagent, which was placed in a boiling water bath for 5 min. In a control tube, the substrate was added after incubation to avoid any reaction with the enzyme. The absorbance was measured at 540 nm. One unit of enzyme activity was defined as the amount of enzyme that catalyzed the formation of 1 µmol reducing group per min per g of the original fresh weight. The enzyme activity was recorded in units mg<sup>-1</sup> of protein.

The reaction mixture for PME assay consisted 1 mL of 0.01% (w/v) citrus pectin (pH 7.5), 0.2 mL 0.15 M NaCl,

0.1 mL 0.01% bromothymol blue, 0.2 mL distilled water and 0.1 mL crude enzyme extract. Absorbance was measured immediately at 620 nm after addition of crude enzyme extract, which was considered as 0 s. Later, the absorbance of sample was recorded constantly for 180 s at regular intervals of 30 s. The difference in the absorbance between 0 s and 180 s was considered for further calculation. One unit of enzyme activity was defined as the amount of enzyme required for liberating 1 mmol of methyl ester per minute.

# Extraction and estimation of superoxide dismutase and peroxidase

The antioxidant enzymes superoxide dismutase (SOD) and peroxidase (POD) were extracted to assay their activities using the procedure described by Xing et al. (2011). To determine SOD activity, 3 mL reaction mixture containing 50 mM sodium phosphate buffer (pH 7.8), 75 µM nitro blue tetrazolium salt, 13 mM methionine, 10 µM EDTA, 2 µM riboflavin and 0.1 mL fruit extract was kept under constant light for 20 min and the colour development was measured at 560 nm. An identical mixture placed in the dark considered as a blank. One unit of enzyme was defined as the amount of enzyme that corresponded to 50% inhibition of nitro blue tetrazolium reduction. For assaying the POD activity, 500 µL of crude extract was mixed with 2 mL 100 mM sodium phosphate buffer (pH 7.8) containing 20 mM guaiacol. The reaction mixture was incubated at 30 °C for 5 min. Later, 1 mL 20 mM H<sub>2</sub>O<sub>2</sub> was added to the mixture as a substrate. Immediately after the addition of substrate, the increase in absorbance was measured at 460 nm for 180 s at intervals of 30 s. Specific activity of POD was expressed as units mg<sup>-1</sup> protein.

# Statistical analysis

All of the experiments were performed in triplicate and the results were expressed as mean  $\pm$  standard deviation. The statistical significance of differences was calculated using one-way ANOVA in IBM<sup>®</sup> SPSS statistical software version 22. Multiple comparisons between the treatments were conducted using the least significant difference at  $P \le 0.05$  level. The Duncan's multiple range test was used to compare the mean values in different storage intervals (Bico et al. 2009).

# Results

# Fruit weight loss

The weight loss percentage of jamun fruit in both control and treated fruit increased gradually during their storage (Table 1). Weight loss percentage on zero day was 0% in all sets of stored fruit, which gradually increased in the control (i.e. 20.67% on Day 4, 57.33% on Day 8 and 82.33% on Day 12). Among all the treatments, T3 showed least weight loss percentage throughout the storage period (i.e. 3.33% on Day 4, 12.33% on Day 8, 23.33% on Day 12 and 28.67% on Day 16). **Table 1.** Relative weight loss and decay of jamun fruit in different coating treatments. C, uncoated control; T1, 3% gum ghatti; T2, 0.1% xanthan gum; T3, 2% biosurfactant; T4, 3% gum ghatti +2% biosurfactant; T5, 0.1% xanthan gum + 2% biosurfactant. Data presented as mean  $\pm$  standard error of three replicates. Different letters over the bars represent significant differences between the treatments on the same storage day at  $P \le 0.05$  according to Duncan's Multiple Range Test

Treatment	Day 0	Day 4	Day 8	Day 12	Day 16	
Weight loss (%)						
С	0	$20.67 \pm 3.06 \text{ d}$	57.33 ± 3.06 b	82.33 ± 2.52 a	0	
T1		5.33 ± 2.08 e	18.67 ± 1.15 d	31.00 ± 3.61 cd	35.33 ± 4.04 cd	
T2		6.00 ±2.00 e	$16.33 \pm 4.73d$	35.00 ± 1.73 c	44.67 ± 4.73 b	
Т3		$3.33\pm0.58~{\rm f}$	12.33 ± 2.52 de	23.33 ± 1.15 d	28.67 ± 3.51 cd	
T4		9.67 ± 1.53 e	21.33 ± 2.52 d	$38.00 \pm 1.00 \text{ c}$	$44.00\pm2.00~\mathrm{b}$	
T5		$10.00 \pm 1e$	$28.00 \pm 2.00 \text{ d}$	$43.00 \pm 2.00 \text{ b}$	$54.67 \pm 4.04 \text{ b}$	
		Decay	v (%)			
С	0	22.22 ± 4.81 cd	55.56 ± 4.81 b	97.22 ± 4.81 a	0	
T1		0	$8.33 \pm 0.00 \text{ d}$	22.22 ± 4.81 cd	38.89 ± 4.81 c	
T2		0	2.78 ± 4.81 d	38.89 ± 4.81 c	50.00 ± 8.33 bc	
Т3		0	$2.78 \pm 4.81 \text{ d}$	19.44 ± 4.81 cd	36.11 ± 4.81 c	
T4		0	$8.33 \pm 0.00 \text{ d}$	27.78 ± 4.81 c	52.78 ± 4.81 b	
T5		0	11.11 ± 4.81 d	36.11 ± 4.81 c	$47.22 \pm 4.81 \text{ bc}$	

### Decay evaluation

Decay percentage in all the treatments increased with storage time (Table 1). On the fourth day of storage, greater visible decay was observed in control fruit compared to other treatments. The treated fruit retained their quality and remain fresh up to Day 16 of storage period. Fruit treated with BS showed the least decay percentage (36.11%) on Day 16 of storage, while control fruit were completely decayed on Day 12 of the storage period (97.22%). Hence, the control fruit were observed for 12 days, while treated fruit were analyzed for their quality for up to 16 days of storage.

**Table 2.** Changes of pH, acidity and TSS of jamun fruit in different coating treatments. C, uncoated control; T1, 3% gum ghatti; T2, 0.1% xanthan gum; T3, 2% biosurfactant; T4, 3% gum ghatti +2% biosurfactant; T5, 0.1% xanthan gum + 2% biosurfactant. Data presented as mean  $\pm$  standard error of three replicates. Different letters over the bars represent significant differences between the treatments on the same storage day at *P* ≤ 0.05 according to Duncan's Multiple Range Test

Treatment	Day 0	Day 4	Day 8	Day 12	Day 16	
pH (units)						
С		$4.08\pm~0.02~b$	$4.15 \pm 0.05$ a	$4.16 \pm 0.05$ a	0	
T1		3.79 ± 0.12 cd	$3.94 \pm 0.01 \text{ bc}$	$4.08\pm0.02~\mathrm{b}$	4.15 ± 0.03 a	
T2	3.58 ± 0.05 e	$3.74 \pm 0.05 \text{ cd}$	$3.75 \pm 0.04 \text{ cd}$	$3.87\pm0.04~c$	$3.85 \pm 0.03 \text{ c}$	
Т3	5.58 ± 0.05 e	$3.85 \pm 0.02 \text{ c}$	$3.86 \pm 0.03 \text{ c}$	$3.86 \pm 0.12$ c	$3.89 \pm 0.07 \text{ bc}$	
T4		$3.78 \pm 0.03 \text{ cd}$	$4.06\pm0.04~ab$	$4.10 \pm 0.03 \text{ ab}$	$4.15 \pm 0.14 \text{ ab}$	
T5		$3.94 \pm 0.06 \text{ b}$	$3.86 \pm 0.04 \text{ bc}$	$4.12 \pm 0.04$ a	$4.15 \pm 0.09$ a	
Acidity (units)						
С		$0.43 \pm 0.01$ a	$0.39\pm0.02~\mathrm{b}$	$0.36 \pm 0.02 \text{ c}$	0	
T1		$0.38\pm0.03~b$	$0.43 \pm 0.02$ a	$0.41 \pm 0.02 \text{ ab}$	$0.41 \pm 0.01$ ab	
T2	$0.45 \pm 0.22$ a	$0.41\pm0.05~ab$	$0.41 \pm 0.01 \text{ ab}$	$0.36 \pm 0.02 \text{ c}$	$0.33 \pm 0.03 d$	
T3	$0.43 \pm 0.22$ a	$0.43 \pm 0.03$ a	$0.39\pm0.03~\mathrm{b}$	$0.38\pm0.03~b$	$0.38\pm0.02~b$	
T4		$0.38\pm0.06~b$	$0.41 \pm 0.07 \text{ ab}$	$0.38\pm0.04~b$	$0.33 \pm 0.02 \text{ d}$	
T5		$0.37 \pm 0.03c$	$0.40\pm0.04~ab$	$0.31 \pm 0.04 \text{ d}$	$0.28\pm0.02~\mathrm{e}$	
TSS (%)						
С		$10.67 \pm 0.10 \text{ c}$	$14.67 \pm 0.29$ a	$16.00\pm0.00~d$	0	
T1		$9.00 \pm 0.15 \text{ e}$	11.33 ± 0.22 c	7.33 ± 0.58 ef	$8.47 \pm 0.01 \text{ e}$	
T2	$8.00 \pm 0.00 e$	$10.00 \pm 0.00 \text{ d}$	$10.67 \pm 0.22 \text{ c}$	$7.00 \pm 1.00 \text{ ef}$	$7.37\pm0.01~{\rm f}$	
Т3	0.00 ± 0.00 e	$11.00 \pm 0.32$ c	$12.00 \pm 0.00 \text{ b}$	$12.00 \pm 0.41 \text{ b}$	$12.40\pm0.06~\mathrm{b}$	
T4		$11.33 \pm 0.10 \text{ c}$	$13.33 \pm 0.44$ a	$10.67 \pm 1.51 \text{ bc}$	11.33 ± 0.09 cd	
T5		$10.00 \pm 0.09 \text{ d}$	8.33 ± 0.11 e	6.33 ± 1.53 ef	$7.57\pm0.02~{\rm f}$	

### pH, TSS and acidity

The pH level was maintained in T2 and T3 treated fruit i.e. from 3.58 at Day 0 to 3.85 and 3.89 after 16 days of storage (Table 2). The pH value of control fruit increased to 4.16 at Day 12. Titratable acidity of jamun fruit decreased continuously in control fruit during the storage period (Table 2). Fruit treated with T3 showed least change in acidity percentage 0.38% at Day 12 of storage. Total soluble solid concentration increased continuously in control fruit, and this rise was delayed in all other treated fruit (Table 2). The initial TSS concentration was 8% and was increased to 16% in control fruit at Day 12 of storage, while TSS concentration was 12% at the end of storage period in fruit treated with T3.

### Glucose, fructose and sucrose concentration

The results of sugar estimated by ion exchange chromatography are shown in Table 3. In freshly harvested fruit at Day 0, sucrose, fructose and glucose concentration was 203, 101 763 and 45 951 mg kg<sup>-1</sup> of fresh mass, respectively. On Day 4 of storage, glucose (37 086 mg kg<sup>-1</sup>) and fructose (81 323 mg kg<sup>-1</sup>) concentration was lower in T3 than control fruit (64384 mg kg<sup>-1</sup> glucose and 148 966 mg kg<sup>-1</sup> fructose). Sucrose concentration was higher (697 mg kg<sup>-1</sup>) in T3 as compared to the control (518 mg kg<sup>-1</sup> of fresh) fruit, which showed lower conversion of complex sugars to simple sugars. On Day 8 glucose (46 085 mg kg<sup>-1</sup>) and fructose (114 556 mg kg<sup>-1</sup>) concentration in T3 has higher. Gradually, the conversion rate of sugars was

increased or remained constant in all treatments till the end of the storage period.

### Total phenolic concentration and antioxidant activity

The level of phenolic substances was observed to increase during the initial days of storage in both treated as well as control fruit, and started to decline thereafter. Phenolic concentration in control fruit remained unchanged (0.565 mg g<sup>-1</sup>) till Day 12 of storage (Fig. 1A). Jamun treated with 2% BS had a higher total phenolic concentration (i.e. 1.653 mg g<sup>-1</sup> on Day 12 and 1.361 mg g<sup>-1</sup> on Day 16 of storage) compared to control fruit. On Day 0 the total antioxidant activity was 41.41% and it increased in all treatments (Fig. 1B). On Day 12 the maximum activity was obtained in T3 (94.03%), which was higher than for the control fruit (80.23%).

### Anthocyanin concentration

Anthocyanin concentration increased during the entire storage period in both control and treated jamun (Fig. 2). For BS-treated fruit, a longer time was necessary for anthocyanin concentration to reach a maximum than for the other treatments and control fruit. Anthocyanin concentration on Day 0 was 4.37  $\mu$ g g<sup>-1</sup> and maximum (22.52  $\mu$ g g<sup>-1</sup>) was reached on Day 12 in control fruit. Later, the control fruit had decayed. BS treatment was effective in delaying anthocyanin accumulation and its concentration was 14.59  $\mu$ g g<sup>-1</sup> at Day 16 of storage.

		Day 4	Day 8	Day 12	Day 16
		Glucos	e (mg kg <sup>-1</sup> FM)		
С	45 951	64 384	19 391	18 793	0
T1		46 386	28 806	27 386	18 191
T2		70 667	62 314	30 304	0
Т3		37 086	46 038	31 288	13 943
T4		43 472	48 850	18 984	32 087
T5		59 306	29 446	38 376	40 760
		Fructos	e (mg kg <sup>-1</sup> FM)		
С	101 763	148 466	44 224	40 895	0
T1		100 917	63 046	51 546	41 326
T2		164 909	139 285	67 663	0
Т3		81 323	114 556	68 349	31 446
T4		94 664	108 124	41 623	73 880
T5		127 062	64897	86 101	96 511
		Sucros	e (mg kg <sup>-1</sup> FM)		
С	203	518	55	164	0
T1		1253	425	81	92
T2		67	0	76	0
Т3		697	0	83	92
T4		452	0	12	64
T5		53	56	72	58

**Table 3.** Changes of sugar concentration in jamun fruit for different coating treatments. C, uncoated control; T1, 3% gum ghatti; T2, 0.1% xanthan gum; T3, 2% biosurfactant; T4, 3% gum ghatti +2% biosurfactant; T5, 0.1% xanthan gum + 2% biosurfactant

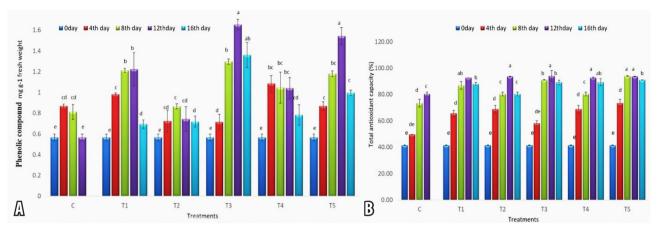


Fig. 1. Effect of coating treatments on concentration of phenolic substances (A) and antioxidative activity (B) in jamun fruit.

#### Ascorbic acid concentration

Concentration of ascorbic acid decreased gradually from 43.83  $\mu$ g g<sup>-1</sup> (Day 0) to 24.52  $\mu$ g g<sup>-1</sup> in T3 followed by T4 (23.77  $\mu$ g g<sup>-1</sup>) and T5 (21.84  $\mu$ g g<sup>-1</sup>) at Day 16 of storage (Fig. 3). In treatment T3, the reduction in ascorbic acid content was observed to be delayed (i.e. 29.58  $\mu$ g g<sup>-1</sup>) whereas it had decreased rapidly in the control to 16.69  $\mu$ g g<sup>-1</sup> on Day 12 of storage.

#### Activity of cell wall-degrading, softening-related enzymes

Table 4 shows activity of the cell wall degrading enzymes examined in the study i.e. PG, cellulase and PME. Higher enzyme activity occurred in parallel with jamun fruit softening in both control and treated groups. A slower rate of increase in PG activity was seen in treated fruit, and more rapid increase in activity was observed in control fruit. PG activity in jamun fruit at Day 0 was 0.0014 U mg<sup>-1</sup> protein and the activity increased to 0.00712 U mg<sup>-1</sup> in control fruit at Day 12 of storage. PG activity was found to be lowest in T4 (0.00103 U mg<sup>-1</sup> at Day 12 and 0.00314 U mg<sup>-1</sup> on Day 16) followed by T3 (0.00308 U mg<sup>-1</sup> at Day 12 and 0.00391 U mg<sup>-1</sup> on Day 16 of storage).

Similarly, cellulase activity in both control as well as treated jamun fruit increased gradually till Day 12 of storage when it reached a maximum and then slowly started to decrease (Table 4). Initially, cellulase activity was 0.0018 U mg<sup>-1</sup> protein, gradually increasing to 0.0388 U mg<sup>-1</sup> in control fruit. Cellulase activity in T3 was 0.0120 U mg<sup>-1</sup> at the end of the storage period, which was lower than the activity found in control fruit.

PME activity was lower (0.0055 U mg<sup>-1</sup> of protein) at the beginning of storage of freshly harvested jamun fruit, but as the storage period increased, activity of PME also gradually increased (Table 4). On the Day 12, activity of PME in control fruit was higher (0.0205 U mg<sup>-1</sup>) than in the T3 treatment (0.0069 U mg<sup>-1</sup>).

### Activity of antioxidant enzymes

BS-coated fruit had higher SOD and POD activity than in the control fruit (Fig. 4) The activity of SOD was 2.765501 U mg<sup>-1</sup> protein on Day 0), and gradually increased in the T3 treatment (3.280715 U mg<sup>-1</sup> of protein on Day 4, 5.13339 U mg<sup>-1</sup> of protein on Day 8 and 7.071487 U mg<sup>-1</sup> of protein on Day 2). The other treatments showed a gradual decrease during the storage. Similarly, activity of POD was higher in T3 treatment compared to other treatments and control fruit. Activity of POD was 0.001516 U mg<sup>-1</sup> protein in treated and control jamun fruit on day zero. Treatment T3 showed the highest POD activity (0.029188 U mg<sup>-1</sup> of protein) on Day 12. The corresponding value in the control was 0.009567 U mg<sup>-1</sup> of protein.

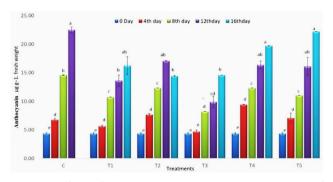


Fig. 2. Effect of coating treatments on concentration of anthocyanins in jamun fruit.

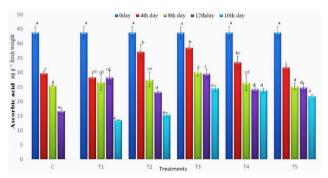


Fig. 3. Effect of coating treatments on concentration of ascorbic acid in jamun fruit.

**Table 4.** Cell wall degrading enzyme activity of jamun fruit in different coating treatments. C, uncoated control; T1, 3% gum ghatti; T2, 0.1% xanthan gum; T3, 2% biosurfactant; T4, 3% gum ghatti +2% biosurfactant; T5, 0.1% xanthan gum + 2% biosurfactant. Data presented as mean  $\pm$  standard error of three replicates. Different letters over the bars represent significant differences between the treatments on the same storage day at  $P \le 0.05$  according to Duncan's Multiple Range Test

Treatment	Day 0	Day 4	Day 8	Day 12	Day 16	
Activity of polygalacturonase (U mg <sup>-1</sup> protein)						
С	$0.0014 \pm 0.0006$ e	$0.00298 \pm 0.0003 c$	$0.00579 \pm 0.0009$ a	$0.00712 \pm 0.0005$ a	$0.0000 \pm 0.0000$	
T1		0.00221 ± 0.0003 d	$0.00152 \pm 0.0004 \text{ e}$	$0.00483 \pm 0.0009 \text{ b}$	$0.0028 \pm 0.0003 \text{ c}$	
T2		$0.00143 \pm 0.0003 \text{ e}$	$0.00114 \pm 0.0004 \text{ e}$	$0.00338 \pm 0.0004 \text{ bc}$	$0.00441 \pm 0.0006 \ b$	
T3	$0.0014 \pm 0.0000 e$	$0.0014 \pm 0.0009 \text{ e}$	0.00360 ± 0.0005 bc	$0.00308 \pm 0.0007 \text{ bc}$	$0.00391 \pm 0.0003 \text{ c}$	
T4		$0.00178 \pm 0.0004 \text{ d}$	$0.00103 \pm 0.0006 \text{ e}$	$0.00103 \pm 0.0008 \text{ e}$	$0.00314 \pm 0.0007 \text{ c}$	
T5		$0.002226 \pm 0.0010 \text{ d}$	$0.00066 \pm 0.0007$ a	$0.00338 \pm 0.0009 \text{ c}$	$0.00529 \pm 0.0009 \text{ b}$	
Activity of cellulase (U mg <sup>-1</sup> protein)						
С		$0.0035 \pm 0.0003 \text{ e}$	$0.0240 \pm 0.0017 \text{ b}$	$0.0388 \pm 0.0048$ a	$0.0000 \pm 0.0000$	
T1		$0.0120 \pm 0.0001 \text{ c}$	$0.0124 \pm 0.0002 \text{ c}$	$0.0202 \pm 0.0015 \text{ b}$	0.0240 ±0 .0017 bc	
T2	0.0018 ± 0.0006 e	$0.0027 \pm 0.0003 \text{ e}$	0.0063 ± 0.0001 d	$0.0239 \pm 0.0007 \text{ b}$	$0.0124 \pm 0.0006 \text{ c}$	
Т3	$0.0018 \pm 0.0006 e$	$0.0070 \pm 0.0006 \text{ d}$	$0.0150 \pm 0.0014 \text{ c}$	$0.0244 \pm 0.0022 \text{ b}$	$0.0120 \pm 0.0001 \text{ c}$	
T4		$0.0202 \pm 0.0015 \text{ e}$	$0.0240 \pm 0.0017 \text{ b}$	$0.0342 \pm 0.0014 \text{ b}$	$0.0202 \pm 0.0003$ bc	
T5		$0.0049 \pm 0.0002 \text{ e}$	$0.0155 \pm 0.0007 \text{ c}$	$0.0275 \pm 0.0018 \text{ b}$	$0.0101 \pm 0.0002 \text{ cd}$	
Activity of pectin methyl esterase (U mg <sup>-1</sup> protein)						
С		$0.0127 \pm 0.0021 \text{ b}$	$0.0137 \pm 0.0033 \text{ b}$	$0.0205 \pm 0.0023$ a	$0.0000 \pm 0.0000$	
T1		$0.0084 \pm 0.0001 \text{ c}$	$0.0090 \pm 0.0023 \text{ c}$	$0.0131 \pm 0.0020 \text{ b}$	$0.0084 \pm 0.0030$ a	
T2	0.0055 ± 0.0010 e	$0.0069 \pm 0.0013 \text{ d}$	$0.0054 \pm 0.0009 \text{ e}$	$0.0089 \pm 0.0019 \text{ d}$	0.0057 ± 0.0016 e	
Т3		$0.0067 \pm 0.0030 \text{ d}$	$0.0047 \pm 0.0020 \text{ e}$	0.0069 ± 0.0013 d	0.0055 ± 0.0010 e	
T4		0.0052 ± 0.0035 e	0.0073 ± 0.0011 cd	0.0137 ± 0.0023 b	0.0093 ± 0.0015 c	
				$0.0115 \pm 0.0024$ bc	$0.0078 \pm 0.0008$ cd	

### Discussion

*Syzygium cumini*, also known as 'jamun', is an important under-utilized perishable fruit with excellent medicinal properties. Therefore, many attempts have been made to enhance its shelf life, including alteration in storage conditions and coating with chemicals or plant-based materials (Gol et al. 2015; Bender et al. 2000; Khaliq et al. 2020). As consumers demand for organic vegetables and fruit with minimum or no chemical treatment is increasing, there is a need for alternatives that are both biocompatible and environment-friendly. The search of such components leads researchers to test bioactive surface-active compounds produced by microorganisms ('biosurfactants'), which represent a structurally diverse group of amphiphilic molecules e.g., rhamnolipids produced from *Pseudomonas aeruginosa* (Terziyski et al. 2014), surfactin from *Bacillus subtilis* (Amani Keshtkar 2013), emulsan from *Acinetobacter calcoaceticus* (Goldman el atl. 1982) and sophorolipids from *Candida bombicola* (Price et al. 2012). They can be produced from agro-industrial waste or byproducts, which also solve the problem of waste disposal (Solanki et al. 2020). This group of compounds includes glycolipids, phospholipids, glycoproteins, lipoproteins and lipopolysachharides. Among them, glycoproteins are less explored as a coating material (Patel et al. 2022).

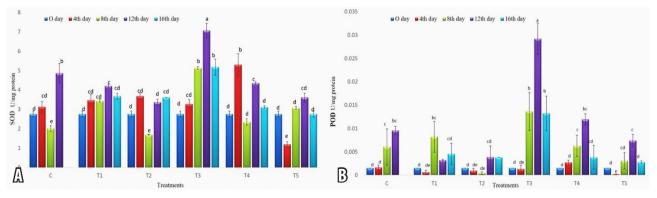


Fig. 4. Effect of coating treatments on superoxide dismutase (SOD) activity (A) and peroxidase (POD) activity (B) in jamun fruit.

Preliminary laboratory trials performed in the present study showed that 2.0% biosurfactant (BS) was the most effective among the concentrations tested to form a uniform layer. Therefore, it was used to improve shelf life of Jamun fruit. Higher concentration of biosurfactant solution (> 2%) led to shrinkage of fruit surface, possibly due to anaerobic respiration. A similar study on use of BS-based coating was also performed with orange fruits (Adetunji et al. 2018). It was reported that a 2% rhamnolipid coating was effective for biocontrol against Penicillium digitatum for up to 8 weeks at 25 °C (Adetunji et al. 2019). A rhamnolipid coating on lemon, potato and tomato (1 mg mL-1; Sharma et al. 2018), on maize (50 mg  $L^{-1}$ ; Borah et al. 2016) and on chilli (500 mg L<sup>-1</sup>; Lahkar et al. 2018) were also reported to have antifungal activity. A sophorolipid coating on citrus fruit, peach, apricot, tomato, cucumber and citrus was effective against phytopathogens (Chen, Yuan 2012; Joshi-Navare et al. 2013). Therefore, BS could be an alternative for a chemical coating for post-harvest protection of fruit and vegetables.

Fruit starts to depreciate soon after harvesting, as it is biological material. Reduction in weight of the fruit is generally the resultant effect of respiration and metabolic activities (Baraiya et al. 2015). The present investigation suggested that treatment with 2% BS (T3) was the most effective in reducing weight loss during the storage period. BS may act as a physical barrier against oxygen, carbon dioxide and moisture, preventing water loss and shrinkage of the coated fruit (Adetunji et al. 2018). Our results are comparable with the results reported by Oluwaseun et al. (2018), who used rhamnolipid with chitosan to minimize weight loss in coated orange. Similarly, reduction in weight loss percentage of jamun fruit was reported by using 1.5% CaCl, (Vandana et al. (2015). Later, 1.5% chitosan + salicylic acid (Saurabh et al. 2019b), Aloe vera gel (Vandana et al. 2019), putrescine (Archana et al. 2019) and guggul gum + basil essential oil (Khaliq et al. 2020) have been used as a coating material for jamun fruit.

Food decay is an un-avoidable step of fruit ripening and it creates a serious problem in shipping as well as in storage. It also influences the nutritional value of fruit. As shown in Table 1, the lowest decay percentage was found in fruit treated with BS coating. BS coating can provide protection to the fruit and delay senescence that reduces cell or tissue integrity. Similar results were found with rhamnolipid + chitosan coating to extend the shelf life of sweet orange for 8 weeks (Oluwaseun et al. 2018). The rhamnolipid coating on lemon, potato and tomato maximized shelf-life up to 15 days (Sharma et al 2018). BS produced by *Nocardia vaccinii* IMV B-7405 was used for coating on vegetables such as tomato, cucumber and squash to extend their shelf life (Pirog et al. 2019).

The degree of ripening of fruit is indicated by its sweetness. Biochemically it is denoted as a decrease of acidity, and increase in pH and total soluble solid concentration. In the present study, minimum pH change was observed, which can be due to a maintained level of organic acids and lower metabolic activity (Jodhani, Nataraj 2019). Results are quite comparable to those of Gol et al. (2015), who tested 1.5% chitosan + CMC-coated jamun. Total soluble solid of fruit has direct association with their texture and composition. The TSS level was higher in control fruit than in treatment T3 and T4 (both containing BS) indicates the good preservation of fruit texture and quality. The TSS level was also associated with pH and total tritable acidity. The hydrolysis of starch into sugar leads to an increase in the amount of TSS during ripening. A previous report on TSS concentration of jamun treated with 10% Aloe vera gel and 1% CaCl, documented a lower TSS at 15 days of storage (Vandana et al. 2019; Vandana et al. 2015). The lower TSS content observed in 2% rhamnolipid + 2% chitosan-treated oranges was due to a decrease in respiration and metabolic activity, which delayed the ripening process (Oluwaseun et al. 2019). Similar results were reported for guggul gum + basil essential oil-treated jamun, where the TSS level increased from 8.73 to 10.67, compared to the highest level of TSS 21.96 on 12 day for the controls (Khaliq et al. 2020). Similarly, titratable acidity is an important chemical characteristic of the fruit, which can affect the taste and choice of consumer. Reduction in the acidity during ripening stage makes the fruit sweeter due to the conversion of organic acids into sugars (Pattar et al. 2021). The notable change in titratable acidity in control fruit indicated the high metabolic activity, resulting in the utilization of organic acids as respiratory substrates during storage. However, minimum change in fruit treated with BS and gum ghatti indicated that organic acids were preserved during storage and ultimately delaying ripening stages. Similarly, it was reported that total acidity was lower in 2% rhamnolipid + 2% chitosan-coated sweet orange (Oluwaseun et al. 2018). Our data are comparable with other studies on jamun, which tested coating of 1.5% chitosan (Gol et al. 2015), 1% CaCl, (Vandana et al. 2015), Aloe vera gel (Vandana et al. 2019) and guggul gum + basil essential oil (Khaliq et al. 2020).

Sweetness in ripened fruit is due to the presence of simple sugars that are made from starch by hydrolytic enzymes. Jamun is a non-climacteric fruit (Koley et al. 2011). The mechanism of sugar accumulation in nonclimacteric fruit is less explored (Davies, Robinson 1996). Sugars are produced through the photosynthetic pathway and transported to fruit through phloem (Swanson, Elshishiny 1958). Sucrose is converted into glucose and fructose by invertase enzyme activity. Glucose and fructose are accumulated in fruit throughout their ripening period (Prasanna et al. 2007). In the present study, it was noted that in control fruit, sucrose, fructose and glucose concentration was highest on Day 4, and then gradually decreased. The glucose and fructose concentration was highest in the treatment with BS coating on Day 8 of storage, and then remained constant or slightly decreased during storage days. This indicated that conversion of sucrose to glucose and fructose was delayed in treated fruit compare to control fruit.

Phenolics in fruit and vegetables are human dietary compounds. They also contribute to fruit quality and nutritive value by changing colour, taste, aroma and flavour, and also provides beneficial health effects (Khaliq et al. 2020). Phenolic compounds have a direct relation with the antioxidant activity. People are interested in small fruits like jamun with outstanding nutraceutical properties due to high polyphenolic content and natural antioxidant activities. The antioxidant property of phenolic compounds makes them act as natural reducing agents, hydrogen donors, oxygen quenchers and metal chelators (Hasan et al. 2008). Control fruit showed no change in phenolic content, associated with minimum metabolic activity or conversion of sugar. BS-coated fruit preserved high phenolic concentration till Day 16 of storage, which indicated good quality, steady conversion of sugar and increased shelf life. Decline of phenol concentration in control fruit indicated accumulation of sugars, which resulted in the loss of astringency (Pattar et al. 2021) or the breakdown of cell structure at the senescence stage (Silva et al. 2011). In the present study, antioxidant activity in all treated fruit was higher than in control fruit, indicating good health and quality of fruit. There is a positive correlation between antioxidant capacity and total phenolic content. These results are in agreement with the earlier reported observations on jamun fruit coated with zein + ascorbic acid (Baraiya et al. 2015), chitosan + carboxy methyl cellulose (Gol et al. 2015), chitosan + salicylic acid (Saurabh et al. 2019), and guggul gum + basil essential oil (Khaliq et al. 2020).

The colour of the fruit changes during ripening due to degradation of chlorophyll and synthesis of different types of pigments like anthocyanin, along with their accumulation in vacuoles (Lizada et al. 1993). As the fruit ripens, accumulation of anthocyanin takes place due to oxidation of phenolic compounds. Higher anthocyanin concentration is an indicator of a ripe fruit. Moreover, it makes fruit attractive and also can benefit health of consumers (Belwal et al. 2019). In this investigation, BScoated fruit showed higher anthocyanin concentration than in control fruit, indicating that it was acceptable for human consumption and provide a health benefit for a longer time. The coating may act as a gas barrier, thus modifying the internal atmosphere in the fruit and this in turn may retard the biochemical reactions leading to anthocyanin synthesis (Baraiya et al. 2015). Rapid anthocyanin degradation in control fruit was due to moisture loss, causing plasmolysis, membrane breakdown, thereby leading to spillage of anthocyanins from vacuoles, and accelerating its degradation by polyphenol oxidase and peroxidase enzymes (Saurabh et al. 2019).

Ascorbic acid is an important antioxidant compound present naturally in fruit and it improves the nutritional value of fresh produce (Khaliq et al. 2020). Apart from antioxidant activity, it is also a co-factor for redox reactions. Ascorbic acid prevents fruit from browning. It is also known that as ripening advances, the level of ascorbic acid decreases (Lee, Kader 2000). The fruit treated with BS coating efficiently preserved the ascorbic acid content with minimum loss compare to the control fruit. Similarly, 2% rhamnolipid + chitosan treatment showed lower ascorbic acid reduction in orange (Oluwaseun et al. 2018). Other reports also indicated coating with guggul gum + basil essential oil (Khaliq et al. 2020), chitosan + CMC (Gol et al. 2015) and chitosan + salicylic acid (Saurabh et al. 2019) preserved amount of ascorbic acid efficiently in jamun fruit. Coating serves as a protective layer and controls the permeability barrier for O<sub>2</sub> and CO<sub>2</sub>, thus decreasing the autoxidation of ascorbic acid (Shahkoomahally et al. 2014). Thus, we can say that the maintained ascorbic acid content in treated fruit with BS improves the antioxidant capacity of the fruit and contributes to the nutritional quality of fruit.

Edible coating forming a thin layer on a fruit surface can modify exchange of gases and thus helps to retain firmness of fruit by reducing the activities of cell wall degrading enzymes such as PG, CEL and PME (Khaliq et al. 2017). The cell wall structure is composed of pectin, cellulose and hemicellulose. The modification in polysaccharide and the breakdown of cell wall structure is closely associated with increased activity of cell wall degrading enzymes, which lead to polymer break down and softening of fruit (Spadonia et al. 2014). CEL activity was higher in control fruit compared to the BS-coated fruits. Lower levels of CEL activity favoured the retention of brittleness and firmness of fruit (Gol et al. 2015). Increase in PME activity causes complete loss of tissue integrity and fruit firmness during ripening (Brummell, Harpster 2001). PME hydrolyses pectin and PG disintegrates pectic acid chains and glycoside bonds of galacturonic acid, resulting in the dissolution and depolymerisation of pectin (Hodges et al. 2004). In the present investigation, BS-coated fruit showed lower activity of PME compared to control fruit, and also showed that the fruit shelf life was extended till Day 16 of storage. Similar results were observed in jamun fruit treated with chitosan + CMC (Gol et al. 2015), zein + ascorbic acid (Baraiya et al. 2015) and guggul gum + basil essential oil (Khaliq et al. 2020), where textural changes were inhibited.

Deterioration of fruit quality is due to accumulation of reactive oxygen species (ROS) such as hydrogen peroxide, singlet oxygen, hydroxyl and superoxide radicals. This reduces the quality and consumer acceptance (Mittler 2002). Fruit limits its ROS damage through the production of antioxidant enzymes like SOD and POD (Jodhani, Nataraj 2019). SOD is responsible for the conversion of ROS into hydrogen peroxide, which is further processed into water with the help of POD enzyme in order to protect the cell from oxidative stress (Yan et al. 2014). Fruit in the ripen stage has lower activity of SOD and POD. The results indicated that the BS coating effectively reduced ROS by enhancing the antioxidant activity of SOD and POD to maintain fruit quality and indirectly by inducing disease resistance. Therefore, treated fruit had good condition up to Day 16 compare to the control fruit which were decayed on Day 12. Rhamnolipid-treated cherry tomato showed higher antioxidant enzymes activity that was maintained during storage. Rhamnolipid + chitosan coating on an orange increased the activity of SOD and POD by reducing the generation of superoxide free radicals and also stimulated a defense mechanism to control post-harvest disease (Oluwaseun et al. 2018).

### Conclusions

The glycoprotein BS produced by Dyadobacter fermentans JDP9 was efficient to extend shelf life of jamun fruit. Fruit treated with BS maintained acceptable quality up to the 16th day, longer than control fruit. Among physical parameters, the biosurfactant-treated fruit showed less weight loss and decay percentage compared to the control. Physiological parameters were also improved in BS-coated fruit. The pH and total soluble solids were observed lower in BS treated fruit compared to control. Total acidity, and phenolic and ascorbic acid concentration did not decrease in BS-treated fruit for 16 days. Similarly, SOD and POD activity were higher, indicating good antioxidant activity of fruit, which is beneficial for human health. BS-treated fruit showed lower concentration of anthocyanins and low sugar conversion rate. Moreover, activity of PG, CEL and PME were lower than in control fruit. These parameters contributed to maintenance of tissue integrity in BScoated fruit in comparison to the control. Therefore, it is concluded the BS could be a promising coating material in post-harvest technology.

### Acknowledgements

The authors are grateful to the Head of P. G. Department of Biosciences, Sardar Patel University, Gujarat, India for providing the necessary facilities for conducting this work. This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors. All authors declare that they have no conflict of interest. The data sets supporting the conclusions of this article are included in the article. The conclusions are based on the data generated from the current study. The author can be contacted for any additional supporting data required by the journal..

#### References

- AOAC. 1994. Official Methods of Analysis. 16th Ed. Association of Official Analytical Chemists, Washington.
- Adetunji C.O., Adejumo I.O., Afolabi I.S., Adetunji J.B., Ajisejiri E.S. 2018. Prolonging the shelf life of 'Agege Sweet' orange with

chitosan-rhamnolipid coating. *Hortic. Environ. Biotechnol.* 59: 687–697.

- Adetunji C.O., Oloke J.K., Osemwegie O.O. 2018. Environmental fate and effects of granular pesta formulation from strains of *Pseudomonas aeruginosa* C1501 and *Lasiodiplodia pseudotheobromae* C1136 on soil activity and weeds. *Chemosphere* 195: 98–107.
- Adetunji C.O., Afolabi I.S., Adetunji J.B. 2019. Effect of rhamnolipid-*Aloe vera* gel edible coating on post-harvest control of rot and quality parameters of 'Agege Sweet' orange. *Agric. Nat. Resour.* 53: 364–372.
- Amani H., Haghighi M., Keshtkar M.J. 2013. Production and optimization of microbial surfactin by *Bacillus subtilis* for *ex situ* enhanced oil recovery. *Petrol. Sci. Technol.* 31: 1249–1258.
- Archana T.J., Suresha G.J., Vandana A.K., Swamy G.S.K. 2019. Effect of exogenous application of putrescine on storage behaviour of jamun (*Syzygium cumini* Skeels) fruits. *Acta Hortic.* 1241: 577–582.
- Baraiya N.S., Rao T.V.R., Thakkar V.R. 2015. Improvement of postharvest quality and storability of jamun fruit (*Syzygium cumini* L. Var. Paras) by zein coating enriched with antioxidants. *Food Bioproc. Technol.* 8: 2225–2234.
- Belwal T., Pandey A., Bhatt I.D., Rawal R.S., Luo Z. 2019. Trends of polyphenolics and anthocyanins accumulation along ripening stages of wild edible fruits of Indian Himalayan region. Sci. Rep. 9: 5894.
- Bender R.J., Brecht J.K., Sargent S.A., Huber D.J. 2000. Mango tolerance to reduced oxygen levels in controlled atmosphere storage. J. Amer. Soc. Hortic. Sci. 125: 707–713.
- Bico S.L.S., Raposo M.F.J., Morais R.M.S.C., Morais A.M.M.B. 2009. Combined effects of chemical dip and/or carrageenan coating and/or controlled atmosphere on quality of fresh cut banana. *Food Control* 20: 508–514.
- Borah S.N., Deka S., Sarma H.K. 2016. First report of *Fusarium verticillioides* causing stalk rot of maize in Assam, India. *Plant Dis.* 100: 15501.
- Bourtoom T. 2008. Edible films and coatings: characteristics and properties. *Int. Food Res.* 15: 237–248.
- Brandão T.S.O., de Sena A.R., Teshima E., David J.M., Assis S.A. 2011. Changes in enzymes, phenolic compounds, tannins, and vitamin C in various stages of jambolan (*Syzygium cumini* Lamark ) development. *Ciênc. Tecnol. Aliment.* 31: 849–855. ŠIS IR KĀ [[[Silva et al.]]]
- Brummell D.A., Harpster M.H. 2001. Cell wall metabolism in fruit softening and quality and its manipulation in transgenic plants. *Plant Mol. Biol.* 47: 311–340.
- Chen J., Yuan B. 2012. Sophorolipid fruit preservative and use thereof in fruit preservation. Chinese Patent CN 101886047 B.
- Davies C., Robinson S.P. 1996. Sugar accumulation in grape berries: cloning of two putative vacuolar invertase cDNAs and their expression in grapevine tissues. *Plant Physiol.* 111: 275–283.
- Dhall R.K. 2013. Advances in edible coatings for fresh fruits and vegetables: a review. *Crit. Rev. Food Sci. Nutr.* 53: 435–450.
- El-Anany A.M., Hassan G.F.A., Rehab Ali F.M. 2009. Effects of edible coatings on the shelf-life and quality of Anna apple (*Malus domestica* Borkh) during cold storage. *J. Food Technol.* 7: 5–11.
- Ghasemnezhad M., Sherafati M., Payvast A. 2011. Variation in phenolic compounds, ascorbic acid and antioxidant activity of five coloured bell pepper (*Capsicum annum*) fruits at two different harvest times. *J. Funct. Foods* 3: 44–49.

- Gol N.B., Vyas P.B., Ramana Rao T.V. 2015. Evaluation of polysaccharide-based edible coatings for their ability to preserve the postharvest quality of Indian blackberry (*Syzygium cumini* L.). *Int. J. Fruit Sci.* 15: 198–222.
- Goldman S., Shabtai Y., Rubinovitz C., Rosenberg E., Gutnick D.L. 1982. Emulsan in Acinetobacter calcoaceticus RAG-1: distribution of cell-free and cell-associated cross-reacting material. Appl. Environ. Microbiol. 44: 165–170.
- Hasan S.M., Hossain M.M., Faruque A., Mazumder M.E.H., Rana M.S., Akter R., Alam M.A. 2008. Comparison of antioxidant potential of different fractions of *Commelina benghalensis* Linn. *Bangladesh J. Life Sci.* 20: 9–16
- Hodges D.M., Lester G.E., Munro K.D., Toivonen P.M. 2004. Oxidative stress: importance for postharvest quality. *Hortic. Sci.* 39: 924–929.
- Jodhani K.A., Nataraj M. 2019. Edible coatings from plant-derived gums and clove essential oil improve postharvest strawberry (*Fragaria* × *ananassa*) shelf life and quality. *Environ. Exp. Biol.* 17: 123–135.
- Joshi-Navare K., Singh P.K., Prabhune A.A. 2014. New yeast isolate *Pichia caribbica* synthesizes xylolipid biosurfactant with enhanced functionality. *Eur. J. Lipid Sci. Technol.* 116: 1070–1079.
- Khaliq G., Saleh A., Ali G., Rehman K. 2020. Guggul gum incorporated with basil essential oil improves quality and modulates cell wall-degrading enzymes of jamun fruit during storage. *Sci. Hortic.* 273: 109608.
- Kim J.Y., Han M.R., Chang M.J., Kim B.Y., Kim M.H. 2002. Study on the extending storage life of grape by applying edible coating materials. *Appl. Biol. Chem.* 45: 207–211.
- Koley T.K., Barman K., Asrey R. 2011. Nutraceutical properties of jamun (*Syzygium cumini* L.) and its processed products. *Indian Food Industry* 30: 43–46.
- Krochta J.M. 2002. Proteins as raw materials for films and coatings: definitions, current status, and opportunities. In: Gennadios A. (Ed.) *Protein-Based Films and Coatings*. Taylor & Francis, pp. 1–40.
- Lahkar J., Goswami D., Deka S., Ahmed G. 2018. Novel approaches for application of biosurfactant produced by *Pseudomonas aeruginosa* for biocontrol of *Colletotrichum capsici* responsible for anthracnose disease in chilli. *Eur. J. Plant Pathol.* 150: 57–71.
- Lee S.K., Kader A.A. 2000. Preharvest and postharvest factors influencing vitamin C content of horticultural crops. *Postharv. Biol. Technol.* 20: 207–220
- Lees D.H., Francis F.J. 1972. Standardization of pigment analysis in cranberries. *Hort. Sci.* 7: 83–84.
- Lizada C. 1993. Mango. In: Seymour G.B., Taylor J.E., Tucker G.A. (Eds.) *Biochemistry of Fruit Ripening*. Chapman and Hall, London, pp. 255–271.
- Lohani S., Trivedi P.K., Nath P. 2004. Changes in activities of cell wall hydrolases during ethylene-induced ripening in banana: effect of 1-MCP, ABA and IAA. *Postharv. Biol. Technol.* 31: 119–126.
- Lowry O.H., Roserbrough N.J., Farr A.L., Randall R.J. 1951. Protein measurement with Folin-phenol reagent. *J. Biol. Chem.* 193: 262–267.
- Marchant R., Banat I.M. 2012. Biosurfactants: a sustainable replacement for chemical surfactants. *Biotechnol. Lett.* 34: 1597–605.
- Mazumdar B.C., Majumder K. 2003. Methods on Physico-Chemical Analysis of Fruit. Daya Publishing House, Delhi, pp. 112–115.

- McDonald S., Prenzler, P.D., Antolovich, M., Robards, K. 2001. Phenolic content and antioxidant activity of olive extracts. *Food Chem.* 73: 73–84.
- Mittler R. 2002. Oxidative stress: antioxidants and stress tolerance. *Trends Plant Sci.* 7: 405–410.
- Nitschke M., Costa S.G.V.A.O. 2007. Biosurfactants in food industry. *Trends Food Sci. Technol.* 18: 252–259.
- Patel D., Solanki J., Ingale S., Nataraj M. 2022. Optimization of production parameter and application of glycoprotein produced from *Sphingobacterium thalpophilum* DP9. *Vegetos* 35: 26–37.
- Patil S.S., Thorat R.M., Rajasekaran P. 2012. Utilization of jamun fruit (*Syzygium cumini*) for production of red wine. *J. Adv. Lab. Res. Biol.* 3: 200–203.
- Pattar A., Kukanoor L., Naik K.R., Naik N., Jholgikar P., Koulagi S., Naika B.M. 2021. Influence of post-harvest treatments on quality and shelf life of jamun (*Syzygium cuminii* Skeels.) fruits under ambient storage. *Pharma Innov.* 10: 523–528.
- Pirog T., Beregova K., Geichenko B. 2019. Application of surfaceactive substances produced by *Nocardia vaccinii* IMB B-7405 for the treatment of vegetables. *Ukrainian Food J.* 8: 99–109.
- Prasanna V., Prabha T.N., Tharanathan R.N. 2007. Fruit ripening phenomena An overview. *Crit. Rev. Food Sci. Nutr.* 47: 1–19.
- Price N.P.J., Ray K.J., Vermillion K.E., Dunlap C., Kurtzman C.P. 2012. Structural characterization of novel sophorolipid biosurfactants from a newly identified species of *Candida* yeast. *Carbohydr. Res.* 348: 33–41.
- Rai D.R., Chadha S., Kaur M.P., Jaiswal P., Patil T.R. 2011. Biochemical , microbiological and physiological changes in Jamun (*Syzyium cumini* L.) kept for long term storage under modified atmosphere packaging. *J. Food Sci. Technol.* 48: 357– 365.
- Roe J.H. 1954. Chemical determination of ascorbic, dehydroascorbic and diketoglutonic acids. In: Glick D. (Ed.) *Methods of Biochemical Analysis.* Wiley, pp. 115–139.
- Salunkhe D.K., Boun H.R., Reddy N.R. 1991. Storage Processing and Nutritional Quality of Fruits and Vegetables: Fresh Fruits and Vegetables. CRC Press, Boston.
- Sambanthamoorthy K., Feng X., Patel R., Patel S., Paranavitana C. 2014. Antimicrobial and antibiofilm potential of biosurfactants isolated from *Lactobacilli* against multi-drugresistant pathogens. *BMC Microbiol.* 14: 197.
- Saurabh V., Barman K., Singh A.K. 2019. Synergistic effect of salicylic acid and chitosan on postharvest life and quality attributes of jamun (*Syzygium cumini* Skeels) fruit. Acta Physiol. Plant. 41: 89.
- Sharma V., Garg M., Talukdar D., Thakur P., Henkel M., Sharma D., Kumar G. 2018. Preservation of microbial spoilage of food by biosurfactant-based coating. *Asian J. Pharmac. Clin. Res.* 11: 98–101.
- Smock R.M. 1940. Some additional effects of waxing apples. *Proc. Amer. Soc. Horticult. Sci.* 1939: 448–452.
- Solanki J., Patel D., Ingale S., Nataraj M. 2020. Screening and optimization of agro-industrial wastes for glycoprotein biosurfactant production from *Sphingobacterium thalpophilum* DP9. /preprint/ https://doi.org/10.21203/ rs.3.rs-32779/v1
- Spadoni A., Guidarelli M., Sanzani S.M., Ippolito A., Mari M. 2014. Influence of hot water treatment on brown rot of peach and rapid fruit response to heat stress. *Postharv. Biol. Technol.* 94: 66–73.
- Srivastava M.K., Dwivedi U.N. 2000. Delayed ripening of banana

fruit by salicylic acid. Plant Sci. 158: 87-96.

- Swanson C.A., El-Shishiny E.D.H. 1958. Translocation of sugars in the concord grape. *Plant Physiol.* 33: 33–37.
- Terziyski I., Alexandrova L., Stoineva I., Christova N., Todorov R., Cohen R. 2014. Foam and wetting films from rhamnolipids produced by *Pseudomonas aeruginosa* BN10. *Coll. Surf. A Physicochem. Eng. Asp.* 460: 299–305.
- Umaraw P., Munekata P.E., Verma A.K., Barba F.J., Singh V.P., Kumar P., Lorenzo J.M. 2020. Edible films/coating with tailored properties for active packaging of meat, fish and derived products. *Trends Food Sci. Technol.* 98: 10–24.
- Vandana A.K., Suresha G.J., Archana T.J., Swamy G.S.K. 2019. Influence of environment friendly bio-preservative *Aloe vera* gel coating on quality parameters of jamun fruits. *Acta Hortic*. 1241: 699–704.

- Vandana A.K., Suresha G.J., Swamy G.S.K. 2015. Impact of calcium chloride pre storage treatment on jamun (Syzygium cumini Skeels) fruits under cold storage. Bioscan 10: 199–202.
- Xing Y., Li X., Xu Q., Yun J., Lu Y., Tang Y. 2011. Effects of chitosan coating enriched with cinnamon oil on qualitative properties of sweet pepper (*Capsicum annuum* L.). *Food Chem.* 124: 1443–1450.
- Yaman O., Bayoindirli L. 2002. Effects of an edible coating and cold storage on shelf-life and quality of cherries. *LWT Food Sci. Technol.* 35: 146–150.
- Yan F., Xu S., Guo J., Chen Q., Meng Q., Zheng X. 2015. Biocontrol of postharvest *Alternaria alternata* decay of cherry tomatoes with rhamnolipids and possible mechanisms of action. *J. Sci. Food Agric*, 95: 1469–1474.