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RESEARCH ARTICLE

Edible Coatings of Calcium Chloride and Carrageenan Mitigate Chilling Injury and Maintain Quality of Red-Fleshed Pitaya (*Hylocereus polyrhizus*) during Cold Storage

Nuratika Tamimi Sheikh Mohamed[1]*, Laily Pang Fui Meiy[1], Shiamala Devi Ramaiya[1], Noorasmah Saupi[1], Hanisah Kamilah[1,2], Khairunnasrin Hamzah[3], Nurul Hidayah Mohd Khairlani[4], Shahidah Md. Nor[5] , Siti Fairuz Yusoff[6] and Lirong Yu Abit[7]

[1]Department of Crop Science, Faculty of Agriculture and Forestry Sciences, Universiti Putra Malaysia Sarawak, 97008 Bintulu, Sarawak, Malaysia
[2]Halal Products Research Institute, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor Darul Ehsan, Malaysia
[3]Faculty of Applied Sciences, Universiti Teknologi MARA, Sabah Branch, Kota Kinabalu Campus, 88997 Kota Kinabalu, Malaysia
[4]Faculty of Plantation and Agrotechnology, Universiti Teknologi MARA Sarawak Branch, Mukah Campus, 96400 Mukah, Sarawak, Malaysia
[5]Department of Technology and Natural Resources, Faculty of Applied Sciences and Technology, Universiti Tun Hussein Onn, UTHM-Campus (Pagoh Branch), Pagoh Education Hub, KM 1, Jalan Panchor, 84600 Muar, Johor, Malaysia
[6]Agricultural Science Department, Faculty of Technical and Vocational, Universiti Pendidikan Sultan Idris, 35900, Tanjong Malim, Perak, Malaysia
[7] Curtin Aquaculture Research Laboratories (CARL), Faculty of Engineering and Applied Sciences (FOES), Curtin University Malaysia, CDT 250, Kuala Baram, 98009 Miri, Sarawak, Malaysia

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☒ * **CORRESPONDING AUTHOR**
Dr. Nuratika Tamimi Sheikh Mohamed
Department of Crop Science, Faculty of Agriculture and Forestry Sciences, Universiti Putra Malaysia Sarawak, 97008 Bintulu, Sarawak, Malaysia
Email: tamimi@upm.edu.my

ABSTRACT

Red-fleshed pitaya (*Hylocereus polyrhizus*), is a non-climacteric fruit esteemed due to its vivid red-purple colouration and high antioxidant content. This fruit is prone to rapid deterioration and chilling injury during low-temperature storage. Thus, the study evaluated the efficacy of three edible coating treatments: calcium chloride ($CaCl_2$, 3% w/v), carrageenan (0.5% w/v) and a combined formulation consisting of 1% $CaCl_2$ + 0.5% carrageenan on the visual quality and phytochemical attributes of *H. polyrhizus* stored at 10°C and 85-90% relative humidity for 20 days. Fruits were assessed at five-day intervals for visual quality, total betacyanin content, total phenolic content (TPC) and antioxidant activity using 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) assays. The combined $CaCl_2$-carrageenan coating most effectively maintained peel integrity and reduced chilling injury exhibited through minimal scale browning and dehydration up to day 20 in comparison with the control. In contrast, fruits coated with 3% calcium chloride ($CaCl_2$) retained significantly higher ($P \leq 0.05$) betacyanin levels (0.72-0.85 mg/g) during mid-storage than those coated with carrageenan or the combined formulation (0.47-0.54 mg/g), indicating the more prominent role of calcium in pigment stabilization. TPC was also better preserved in coated fruits (0.013-0.016 mg GAE/g), whereas uncoated fruits exhibited a significant decline (0.009-0.010 mg GAE/g) by days 5 and 10, demonstrating rapid phenolic degradation without protective coatings. Antioxidant activity (DPPH) was highest ($P \leq 0.05$) in fruits coated with carrageenan (53.47%) and $CaCl_2$ (48.95%), while carrageenan also produced greater ABTS inhibition (16.58%) when compared with uncoated fruits (13.92%). However, the combined coating showed reduced antioxidant retention, this was possibly due to antagonistic interactions in the composite coating. Overall, single component coatings ($CaCl_2$ or carrageenan) were more effective for preserving antioxidant properties, whereas the combined formulations better maintained external visual quality and chilling-injury resistance during prolonged cold storage. Edible coatings thus represent a sustainable postharvest strategy for extending the shelf life and nutritional quality of pitaya during cold chain distribution.

1 INTRODUCTION

Pitaya (*Hylocereus polyrhizus*), commonly referred to as red-fleshed dragon fruit, is a non-climacteric tropical fruit distinguished by its vivid red-purple peel, consumable seeds, and elevated antioxidant content. Originally from Central and South America, its cultivation has proliferated throughout Asia, particularly in Malaysia, where more than 560 hectares are cultivated in response to rising market demand (Chen et al., 2024; Belbase and Bhaskar, 2025). The term 'pitaya' derives from the squamous exterior of the fruit, which gives it the characteristic appearance of a 'scaly fruit'. Cho and Ding (2021) highlighted the aesthetic attributes of this plant, including its night-blooming flowers, which are approximately 25 cm in diameter and creamy white in colour. There are three

primary species of dragon fruit distinguished by their peel and flesh colouration: H. polyrhizus (pink peel with red flesh), H. megalanthus (yellow peel with white flesh) and H. undatus (pink peel with white flesh). Despite their morphological differences, these species share similar physiological characteristics and demonstrate strong adaptability to tropical and subtropical climates, indicating their potential for cultivation in water-scarce regions due to their efficient-water use physiology.

Being a non-climacteric fruit, pitaya does not exhibit significant ripening changes after harvest. Thus, harvesting at full maturity is essential to ensure optimal fruit quality. However, the fruit is sensitive to chilling injury between 5°C and 10°C (Zee et al. 2004; Sheng et al., 2020). The postharvest quality and shelf life of pitaya are affected by its maturity at harvest. Imchen et al. (2025) noted that overripe fruits are more susceptible to senescence which shortens shelf life and reduces quality. Whereas immature fruits may develop cell wall ruptures and physiological disorders due to incomplete tissue development.

To address postharvest challenges, coating technologies have been widely employed to enhance the preservation and shelf life of fruits and vegetables. Edible coatings can enhance aesthetic appeal, suppress ripening and shrinkage, reduce water loss and increase resistance to decay (Razali et al., 2021). These coatings typically function by regulating the diffusion of respiratory gases and optimizing water vapor permeability, thereby creating a modified atmosphere that decreases oxygen levels and increases carbon dioxide concentration (Tokatlı and Demirdöven, 2020). Razali et al. (2021) demonstrated that the application of a submicron chitosan coating (1.0%) maintained the quality of pitaya for up to 28 days at 10°C. Conversely, the administration of a composite coating containing chitosan and oleic acid was found to be effective in impeding fungal deterioration, reducing skin wrinkling, and minimizing water loss (Espinal-Hernández et al., 2021). Pu et al. (2023) reported that propolis coatings, known for their adhesive properties, slowed ripening and enhanced the production of beneficial components in pitaya. These emulsified coatings maintain stability at ambient temperatures and preserved both flavor and texture without any significant alterations.

Calcium chloride CaCl2 has been extensively applied to fruits such as strawberries, passion fruit and litchi to delay senescence, strengthen cell walls and reduce postharvest decay through the formation of calcium-pectin cross-links that enhance tissue firmness (Wei et al., 2024; Eroğul et al., 2024; Guo et al., 2023). When incorporated into edible coatings, $CaCl_2$ further contributes to maintaining fruit firmness and minimizing physiological disorders during storage (Ahmad et al., 2023). Concurrently, carrageenan, a natural polysaccharide derived from red seaweed has also gained attention as an eco-friendly coating material capable of reducing water loss, maintaining pigment stability and delaying ripening in fruits such as bananas (Pamungkas et al., 2023). Despite extensive studies on the individual effects of $CaCl_2$ and carrageenan, their combined application as a dual-component edible coating remains largely unexplored. In particular, limited information exists regarding their synergistic role in preserving betacyanin stability, antioxidant capacity and overall postharvest quality of red-fleshed dragon fruit (*H. polyrhizus*) under cold storage conditions. Addressing this gap could offer a promising strategy to enhance the storage life and visual quality of this highly perishable tropical fruit.

Reducing respiration rates, delaying the initial phase of decay and minimizing water loss are three ways in which low-temperature storage extends the shelf life of freshly harvested produce. However, the ideal storage temperature for pitaya is also dependent on its particular genetic composition, cultivation environment and stage of maturation (Sheng et al., 2020). Storing dragon fruit at temperatures between 5°C and 10°C may cause chilling damage. Indicators of chilling injury in pitaya include peel translucency, darkening of scales, fruit softening, fruit shrinkage and loss of flavor (Sheng et al. 2020). Symptoms of chilling injury worsen with prolonged storage. Consequently, it is advisable to store pitaya at minimum postharvest temperatures according to its stage of ripeness and to further extend the storage period by applying storage temperatures from 10°C to a minimum of 5°C (To et al. 2002; Hoa et al. 2006; Jiang et al., 2020). Compared to white-fleshed pitaya, the red-fleshed variety has a higher antioxidant content due to the presence of the red pigment betalain (Attar et al., 2022). While temperature is a critical factor in betalain stability, other factors such as pH, oxygen, light and moisture also influence betalain degradation (Calva-Estrada et al., 2022). Despite numerous studies investigating the use of betalains as natural food colourants (Nabi et al., 2023), limited research has delved into the aspect of long-term preservation of these phytochemicals in dragon fruit under cold storage conditions, particularly when combined with postharvest coating treatments.

While postharvest coating technologies have been extensively explored for white-fleshed dragon fruit (*H. undatus*), there is limited research addressing their effects on the red-fleshed species (*H. polyrhizus*), particularly regarding the preservation of betacyanin and antioxidant activity under cold storage conditions. Given the commercial and nutritional importance of betacyanin-rich pitaya, this study addresses a critical knowledge gap in the postharvest handling of this high-value fruit. Therefore, the primary objective of this research

was to evaluate the effects of postharvest coatings on the phytochemical stability of red-fleshed dragon fruit during cold storage. Specifically, this research aimed to assess the effectiveness of calcium chloride and carrageenan coatings in preserving betacyanin content in *H. polyrhizus* over prolonged low-temperature storage.

2 MATERIALS AND METHODS

2.1 Plant materials

A total of 60 uniform, mature *Hylocereus polyrhizus* (index 5) fruits were obtained from Koperasi Petani Bintulu Sarawak Berhad. To eliminate surface contaminants, the fruits were washed with a 1% (v/v) sodium hypochlorite solution and subsequently rinsed three times using distilled water. Thereafter, the fruits were air-dried at ambient room temperature. The fruits were randomly divided into four treatment groups (control, calcium chloride (3% w/v), carrageenan (0.5% w/v) and 1% calcium chloride + 0.5% carrageenan) each comprising 30 fruits. The treatment concentrations were selected based on preliminary studies that demonstrated superior coating uniformity, adherence and drying performance on red-fleshed pitaya fruits. Analyses were conducted at five-day intervals over a 20-day storage period (day 0, 5, 10, 15 and 20).

2.2 Coating treatments and storage conditions

A 3% (w/v) calcium chloride ($CaCl_2$) solution was prepared following the protocol of Gemeda (2021) by dissolving the salt ($CaCl_2$ anhydrous, Supelco, USA) in distilled water and agitating the mixture for 30 min using a magnetic stirrer. Tween 20 (Tween 20, Sigma-Aldrich, USA) was incorporated to improve coating plasticity and wettability. For the carrageenan coating, 0.5% (w/v) carrageenan (κ-carrageenan, Sigma-Aldrich, USA) was dispersed in distilled water pre-heated to 80°C and continuously stirred until complete solubilization. Carboxymethyl cellulose (CMC, Sigma-Aldrich, USA) and glycerol (Supelco, USA) were then added as plasticizers and the mixture was stirred for an additional 30 min at 80°C before being cooled to 50°C.

A combined coating formulation was prepared according to the method of Bico et al. (2009). Briefly, a 1% (w/v) calcium chloride solution was mixed with a 0.5% (w/v) carrageenan solution at a 1:1 ratio (v/v) and stirred for 30 min to ensure homogeneity. Fruits were submerged for 5 min in their respective coating solutions: calcium chloride (3% w/v), carrageenan (0.5% w/v) and 1% calcium chloride + 0.5% carrageenan. Meanwhile, the fruits in the control group were dipped in 100 mL of distilled water. All fruits were then air-dried at room temperature for 1 hour and followed by storage continously at 10±1°C for a period of 20 days. Temperature and relative humidity (85-90% RH) were monitored daily to ensure stability. Sampling and analyses were conducted every five days, with three replicates per treatment.

2.3 Visual Quality Assessment

Visual assessments were performed at five-day intervals (days 0, 5, 10, 15 and 20) to monitor external appearance and detect symptoms of chilling injury. At each interval, five fruits per treatment were evaluated for peel colour uniformity, scale wilting, surface dehydration, and specific chilling injury symptoms such as peel translucency and scale browning. Observations were documented photographically (Figure 1) to illustrate progressive visual changes throughout the storage period. These visual assessments were used to evaluate the efficacy of each coating treatment in prolonging shelf life and improving resistance to chilling injury.

2.4 Sample extraction

The extraction of pitaya fruit pulp for phytochemical and antioxidant assays was conducted according to the protocol of Shian et al. (2012) with minor modifications. Twenty grams of pitaya pulp were ground using a pestle and mortar. Five grams of pulp were extracted with 70% ethanol under dim lighting and an orbital shaker set to 180 rpm for one hour. The homogenate was then centrifuged at 2000 rpm for 10 minutes and the supernatant collected for further analysis.

2.5 Total betacyanin content

Total betacyanin content was determined using a spectrophotometer (S1200, Spectrowave spectrophotometer, Cambridge, England), following the method described by Phebe et al. (2009) with minor modifications. McIlvaine buffer with pH 6.5 was used for the analysis. Each sample was prepared by diluting 0.1 mL extract with 2.9 mL of McIlvaine buffer in a quartz cuvette. A blank was prepared using 3.0 mL of McIlvaine buffer. Absorbance was measured at 538 nm. Betacyanin content was calculated using the formula:

$$\text{Betacyanin (mg/g)} = \frac{\text{A538} \times \text{MW} \times \text{V} \times \text{DF}}{\varepsilon \times \text{B} \times \text{W}}$$

Where,

A_{538} = absorbance at 538 nm

MW = molecule weight of betanin

V = total extract volume (mL)

DF = dilution factor

E = molar absorptivity for betanin (6.5×104 cm^{-1} mol^{-1} L)

B = path length (1.0 cm)

W = fresh weight of extracting materials (g)

2.6 Total Phenolic Content (TPC)

TPC was determined by mixing a 0.2 mL extract, 0.6 mL distilled water and 0.2 mL of Folin-Ciocalteu's phenol reagent (Supelco, USA) following (Shian et al., 2012). After 5 min, 1 mL of 8% (w/v) sodium carbonate (HmbG Chemicals, Germany) was added, and the total volume was adjusted to 3 mL with distilled water. The reaction was incubated in the dark for 30 min, centrifuged and absorbance was measured at 765 nm. TPC was expressed as mg gallic acid equivalent (GAE) per g fresh weight using a gallic acid standard curve.

2.7 DPPH Radical Scavenging Activity

The radical-scavenging activity against 1,1-diphenyl-2-picrylhydrazyl (DPPH) was measured according to Brand-Williams et al. (1995). A total of 0.1 mL extract was mixed with 3.9 mL of 1 mM DPPH (Sigma-Aldrich, USA) dissolved in ethanol. The mixture was vortexed and incubated in the dark for 30 min. Absorbance was measured at 517 nm. The percentage of DPPH inhibition was calculated as:

$$\%\text{DPPH inhibition} = \frac{(A0 - A1)}{A0} \times 100$$

Where,

A_0 = absorbance of the control

A_1 = absorbance of the sample

2.8 ABTS radical scavenging activity

The ABTS radical cation decolourization assay was performed as described by Re et al. (1999). $ABTS^{\cdot+}$ radicals were generated by mixing 7 mM ABTS reagent (Sigma-Aldrich, USA) with 2.45 mM potassium persulfate in a 1:1 ratio and incubating the mixture in the dark at room temperature for 12 to 16 h. This solution was diluted with 70% ethanol to an absorbance of 0.700 ± 0.020 at 734 nm. For the assay, 3 mL of the diluted ABTS solution was mixed with 100 µL of the extract, vortexed and incubated for 10 minutes at room temperature. Absorbance was measured at 734 nm. Results were expressed as percentage inhibition of ABTS, calculated using:

$$\%\text{ABTS inhibition} = \frac{(A0 - A1)}{A0} \times 100$$

Where,

A_0 = absorbance of the control

A_1 = absorbance of sample

2.9 Experimental design and statistical analysis

The experiment utilized a completely randomized design (CRD) with a factorial arrangement (four coating treatments × five storage durations). Each treatment was triplicated. Data was analyzed using a one-way analysis of variance (ANOVA) and mean comparisons were performed using Duncan's Multiple Range Test (DMRT) at a significance level of $P \leq 0.05$. All statistical analyses were performed using SAS software version 9.4 (SAS Institute Inc., Cary, NC, USA).

3 RESULT AND DISCUSSION

3.1 Visual quality evaluation of pitaya fruit under different postharvest coatings

The visual appearance of pitaya fruit during storage at 10°C and 85-90% relative humidity was significantly affected by the type of postharvest coating applied. As illustrated in Figure 1, fruits were assessed at five-day intervals over a 20-day period (D0-D20) under four treatments: T1 (uncoated control), T2 (3% $CaCl_2$), T3 (0.5% carrageenan) and T4 (1% $CaCl_2$ + 0.5% carrageenan).

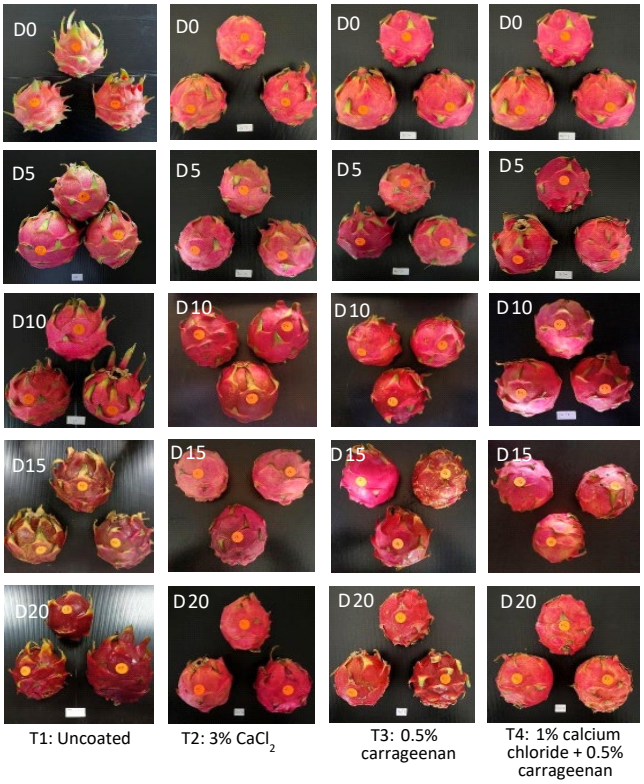

Figure 1: Effect of different coating treatment on pitaya fruit over a 20-days storage period at 10°C and 85-90% relative humidity. The visual appearance of pitaya fruit after day 0 (D0), 5 (D5), 10 (D10), 15 (D15) and 20 (D20) of storage period under four different postharvest coating treatments: T1: control, T2: 3% calcium chloride, T3: 0.5% carrageenan and T4: 1% calcium chloride + 0.5% carrageenan.

At D0, all pitaya fruits appeared fresh, firm and vibrant in colour with no visible defects, indicating uniform initial quality across treatments. However, by D10 the uncoated fruits began exhibiting typical chilling injury (CI) symptoms, including scale wilting, surface dullness, and

increasing translucency. By D15, these fruits exhibited advanced senescence and had entirely lost their commercial appeal. In contrast, T4-treated fruits consistently retained superior visual quality throughout the storage period. These fruits exhibited minimal surface dehydration and maintained structural integrity up to D20. The improved performance is attributed to the synergistic effect of calcium and carrageenan, whereby calcium reinforces tissue firmness through cross-linking with pectic substances (Schumann et al., 2022; Zhang et al., 2024), while carrageenan forms a semi-permeable film that reduces water vapor loss from the fruits (Cheng et al., 2022). The reduced chilling injury in coated fruits may also be linked to lower membrane lipid peroxidation and suppression of lipoxygenase (LOX) activity, which is commonly associated with cold-induced oxidative stress in tropical fruits.

The presence of Ca^{2+} stabilizes membrane phospholipids, thereby minimizing electrolyte leakage and reactive oxygen species (ROS) accumulation (Feng et al., 2023). Carrageenan coatings may possibly enhance antioxidant enzyme defense systems such as superoxide dismutase (SOD) and catalase (CAT) which mitigate ROS-induced damage during prolonged cold exposure (Cruz-Monterrosa et al., 2023). T3-treated fruits initially showed delayed visual degradation but began to manifest chilling injury by D20 which was characterized by watery and translucent peel. This suggests that although carrageenan provides an effective moisture barrier, it lacks the structural reinforcement needed to prevent cold-induced tissue breakdown for extended periods. Increased LOX activity and peroxidative damage at late storage stages may have accelerated membrane deterioration in carrageenan-only fruits, as carrageenan coatings primarily act as moisture barriers without reinforcing cell wall structure.

This physiological response is consistent with previous findings in cold-stored guava and mango, where elevated LOX activity and lipid peroxidation were directly linked to membrane damage and chilling injury (Bhardwaj et al., 2022; Fan et al., 2022). In contrast, the T2 treatment (3% calcium chloride alone) preserved fruit structure better than T3 (0.5% carrageenan alone) up to D20, with no significant signs of chilling injury This highlights the crucial role of calcium in maintaining cell wall integrity even in the absence of a strong moisture barrier. These findings are consistent with previous research in litchi fruit where the calcium ions stabilized the fruit cell walls by forming calcium-pectate complexes, thereby reducing cell wall degradation and delaying senescence (Guo et al., 2023). In contrast, carrageenan, a hydrophilic polysaccharide that forms gel-like films does not inherently strengthen cell wall structure causing fruit to be more susceptible to chilling injury under cold storage stress conditions.

The observed differences in chilling injury intensity among treatments may be further explained by differential ROS accumulation and antioxidant responses. Coated fruits likely maintained a lower malondialdehyde (MDA) level, a biomarker of lipid oxidation thereby preserving cell membrane integrity under cold stress (Gull et al., 2024). The application of postharvest coating treatments on pitaya fruits has been shown to significantly preserve visual quality, minimize chilling injury and delay senescence compared to untreated fruits. In the present study, both 3% $CaCl_2$ and a combined treatment of 1% $CaCl_2$ with 0.5% carrageenan effectively maintained visual appearance and extended the shelf life of pitaya under cold storage. In contrast, 0.5% carrageenan alone was insufficient to preserve tissue structure as evidenced by the appearance of chilling injury symptoms observed by D20. These findings align with previous research by Razali et al. (2021) who reported that dragon fruits coated with chitosan retained quality for up to 28 days at 10°C without signs of decay or off-flavors. Additionally, Pu et al. (2023) found that propolis coatings derived from honeybees effectively slowed the ripening process of dragon fruit and enhanced the accumulation of beneficial bioactive compounds.

3.2 Effect of coating treatments on total betacyanin content

Betacyanins, a class of water-soluble pigments are responsible for the vivid colouration observed in many fruits and flowers (Fernández-López et al., 2020). Among these, isobetanidin and betanidin are recognized as two of the most prominent betacyanins (Kumorkiewicz-Jamro et al., 2021). In this study, betacyanin concentrations in pitaya fruit was significantly (P≤0.05) influenced by the interaction between coating treatments and storage duration (Table 1).

Table 1: Main and interaction effects of different coating treatments and storage duration on betacyanin content of red-fleshed pitaya fruit stored under cold storage at 10°C and 85-90% relative humidity.

Factors	Total betacyanin content (mg g^{-1})
Coating treatments (CT)	
Uncoated	0.83 a[z]
3% calcium chloride	0.77 ab
0.5% carrageenan	0.68 b
1% $CaCl_2$ + 0.5% carrageenan	0.70 b
Storage duration (SD)	
0	0.74 bc
5	0.89 a
10	0.78 ab
15	0.69 bc
20	0.63 c
Interactions	
CT x SD	**

[z]Means followed by the same letter in the same column within factors are not significantly different at P≤0.05 according to DMRT. **Significant difference at P≤0.01.

The concentration peaked on D5 (0.89 mg g^{-1}) and subsequently declined to 0.63 mg g^{-1} by the end of the 20-day storage period. This decline is primarily attributed to the inherent instability of betacyanins to environmental stress, particularly at low temperatures. Their degradation is accelerated by other factors such as light exposure, oxygen, pH fluctuations and changes in water activity (Herbach et al., 2004; Nurhadi et al., 2024). The reduction in betacyanin content by D20 reflects the cumulative effects of these stressors which are further exacerbated by the natural senescence process of the fruit.

Betacyanin degradation during chilling may also involve oxidative stress-related enzymatic activity, where low-temperature stress induces reactive oxygen species (ROS) accumulation leading to pigment oxidation and cell membrane injury. Recent studies showed that cold-stored pitaya and beetroot experienced betacyanin decline linked to increased peroxidase (POD) and polyphenol oxidase (PPO) activities, which catalyze pigment breakdown (Mai et al., 2022). The $CaCl_2$-based coating could have mitigated this by reducing cell membranes and reducing ROS generation while carrageenan may limit oxygen diffusion, thereby reducing enzymatic oxidation (Guo et al., 2023).

Uncoated fruits retained higher betacyanin levels than coated fruits after 10 days of storage (Figure 2). However, by D15, fruits coated with 3% $CaCl_2$ exhibited a significantly higher (P≤0.05) betacyanin content (0.85 mg g^{-1}) compared to those treated with a combined solution of 1% $CaCl_2$ and 0.5% carrageenan (0.54 mg g^{-1}). By D20, no significant differences were observed among treatments, suggesting that pigment degradation had progressed in all samples.

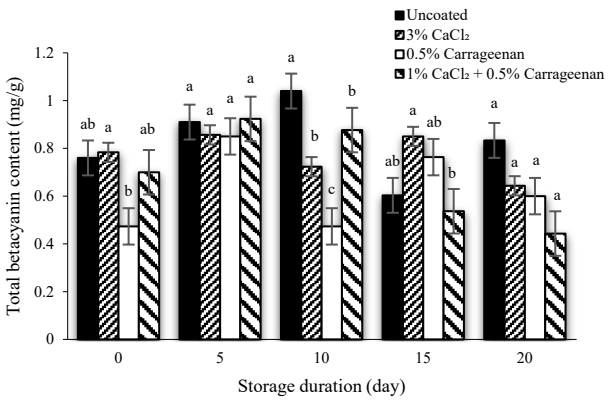

Figure 2: Effect of different edible coatings on total betacyanin content of pitaya fruit stored at 10ºC/85% relative humidity. Different letters in each storage duration indicate significant difference at P≤0.05 according to Duncan's multiple range test. Vertical bars indicate S.E. of means for the three replicates.

These findings indicate that 3% $CaCl_2$ was the most effective treatment in preserving betacyanin content during mid-storage, whereas carrageenan alone provided limited protection under cold stress conditions. The observed pattern also suggests LOX-mediated lipid peroxidation could accelerate pigment degradation as membrane leakage exposes betacyanins to oxidative enzymes (Martinez et al., 2024). Edible coatings that suppress LOX activity can therefore indirectly maintain pigment stability under chilling conditions.

The lack of enhanced in pigment retention in the combined treatment may be due to a suboptimal calcium concentrations and limited synergistic interaction between $CaCl_2$ and carrageenan. Enhancing betacyanin stability using protective matrices through microencapsulation technologies (such as with pectin, gum arabic and maltodextrin) has been shown to improve pigment retention during storage and processing of pitaya fruits (Rahayuningsih et al., 2020). This suggests that more advanced encapsulation or delivery systems may offer better protection of sensitive pigments such as betacyanins under cold storage conditions. Similar results were reported in papaya, where slices treated with hydrothermal calcium chloride and chitosan coatings showed improved colour retention and higher levels of β-carotene and lycopene in comparison to untreated controls, indicating effective preservation of pigment compounds (Ayón-Reyna et al., 2015).

3.3 Effect of coating treatments on total phenolic content

Phenolic compounds are well known for their potent antioxidant properties with some exhibiting greater antioxidant activity than ascorbic acid and α-tocopherol (Kruk et al., 2022). In the present study, a significant (P≤0.05) interaction was observed between postharvest edible coating treatments and storage duration on the total phenolic content (TPC) of red-fleshed dragon fruit (Table 2). As shown in Table 2 and Figure 3, TPC levels in all fruits showed a fluctuating trend during 20 days of cold storage at 10°C 85-90% RH. An initial decline in phenolic content was observed up to D10 which was followed by a stabilization until D20. This trend is consistent with prior studies by Mustafa et al. (2016), which suggest that cold storage can induce phenolic compound accumulation as part of fruit defense response.

The transient decline and later stabilization of TPC may be linked to an early burst of ROS and activation of phenylpropanoid pathway enzymes such as phenylalanine ammonia-lyase (PAL), which is often induced under chilling stress to synthesize protective phenolic (Sogvar et al., 2020). Calcium ions may enhance PAL activity and cell wall cross-

linking, while carrageenan coatings restrict gas exchange and reduce oxidative enzymatic degradation of phenols (Li et al., 2021; Schumann et al., 2022).

Table 2: Main and interaction effects of different coating treatments and storage duration on total phenolic content, DPPH and ABTS radical scavenging activity in red-fleshed pitaya fruit stored under cold storage at 10°C/85% relative humidity

Factors	Total phenolic content (mg GAE/g fresh weight)	DPPH inhibition (%)	ABTS inhibition (%)
Coating treatments (CT)			
Uncoated	0.013 a[z]	42.36 c	15.91 ab
3% calcium chloride	0.015 a	48.95 ab	18.34 a
0.5% carrageenan	0.014 a	53.47 a	14.96 b
1% $CaCl_2$ + 0.5% carrageenan	0.014 a	35.72 c	16.46 ab
Storage duration (SD)			
0	0.014 a	43.20 a	19.96 a
5	0.014 a	44.04 a	14.57 c
10	0.013 b	50.49 a	14.42 c
15	0.015 a	42.17 a	17.95 ab
20	0.014 a	45.72 a	15.18 bc
Interactions			
CT x SD	***	ns	*

[z]Means followed by the same letter in the same column within factors are not significantly different at P≤0.05 according to DMRT.
ns, *, ***No significant difference at P≥0.05 or significant difference at P≤0.05 and P≤0.001, respectively.

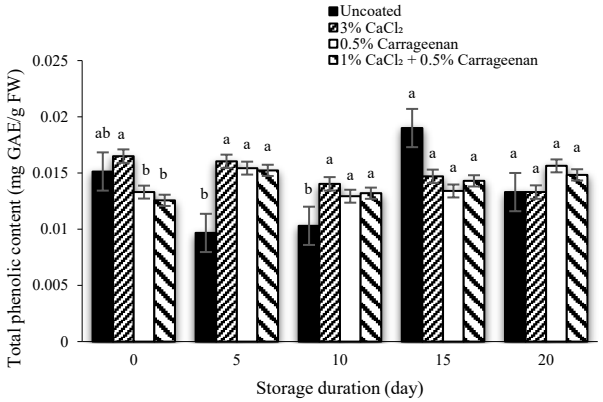

Figure 3: Effect of different edible coatings on total phenolic content of pitaya fruit stored at 10ºC/85% relative humidity. Different letters in each storage duration indicate significant difference at P≤0.05 according to Duncan's multiple range test. Vertical bars indicate S.E. of means for three replicates.

The uncoated fruits exhibited significantly lower (P≤0.05) TPC values, ranging from 0.013 to 0.016 mg GAE/g fresh weight, compared with the coated fruits on days 5 and 10, which recorded values between 0.009 and 0.010 mg GAE/g fresh weight. This trend indicates a more rapid degradation of phenolic compounds in fruits without protective coatings (Figure 3). At these early storage stages, coatings containing 3% $CaCl_2$, 0.5% carrageenan and particularly their combination (1% $CaCl_2$ + 0.5% carrageenan) consistently maintained higher phenolic levels. However, by D15 and 20, differences between treatments were no longer statistically significant, suggesting the diminishing effect of coatings over an extended storage period. This observation is attributable to the stabilization of enzymatic activity and oxidative processes as fruit ripening and senescence progressed.

In cold-stored fruits, the balance between ROS generation and antioxidant metabolism determines phenolic preservation. Edible coatings that suppress ROS accumulation or maintain higher superoxide dismutase (SOD) and catalase (CAT) activities tend to retain TPC longer (Pan et al., 2024; Cruz-Monterrosa et al., 2023). This mechanism likely contributed to the improved phenolic retention observed in coated pitaya fruits.

These findings demonstrated that calcium chloride and carrageenan-based coatings can effectively delay the degradation of phenolic compounds, potentially preserving the nutritional and antioxidant quality of pitaya fruit for up to 10 days. The efficacy of carrageenan-based coatings in maintaining phenolic content may be linked to their polysaccharide structure. As indicated by Rajamani and Khora (2025), carrageenan is a sulfated polysaccharide derived from red seaweed which forms a semi-permeable film that reduces oxidative stress by limiting gas exchange and moisture loss. Additionally, Feng et al, (2024) reported that polysaccharide-based coatings in strawberries reduced free radical damage and suppressed excessive reactive oxygen species (ROS) production, primarily by stabilizing antioxidant enzyme activity. A similar mechanism may explain the enhanced phenolic retention observed in pitaya treated with carrageenan and calcium chloride coatings during the present study.

3.4 Effect of coating treatments on antioxidant activities

DPPH (1,1-Diphenyl-2-picrylhydrazyl) is a stable organic free radical widely employed to evaluate antioxidant activity due to its sensitivity to antioxidants that donate electrons or hydrogen atoms. In the present study, no statistically significant interaction was observed between coating treatments and storage duration in influencing DPPH radical scavenging activity (Table 2).

Among the treatments, fruits coated with 0.5% carrageenan and 3% calcium chloride demonstrated the highest DPPH inhibition percentages at 53.47% and 48.95%, respectively. These values were significantly greater (P≤0.05) than those recorded for uncoated fruits (42.36%) and fruits treated with the combined formulation of 1% $CaCl_2$ + 0.5% carrageenan (35.72%). This suggests that single-component coatings were more effective in enhancing and maintaining antioxidant activity compared to the combined treatment. Throughout the 20-day storage period at 10°C, DPPH

inhibition remained relatively stable in all coating treatments, with no significant differences observed throughout the storage duration (Table 2). This stability implies that coatings could moderate oxidative metabolism by reducing ROS generation and maintaining the activities of antioxidant enzymes like SOD, CAT and ascorbate peroxidase (APX). Continuous low-temperature storage can cause oxidative imbalance and coatings that form semi-permeable barriers help sustain intracellular redox homeostasis (Cice et al., 2024; Pan et al., 2024).

The increased DPPH inhibition observed in coated fruits is possibly associated with the retention and potential enhancement of phenolic compounds which are known to contribute significantly to antioxidant activity. Phenolic compounds act as free radical scavengers which disrupting free radical chain reactions and delaying lipid oxidation (Kruk et al., 2022). Ahmed et al. (2021) demonstrated a positive correlation between total phenolic content and antioxidant capacity in *Syzygium claviflorum*, highlighting the contribution of compounds like gallic acid to DPPH scavenging activity. Similarly, Maisuthisakul et al. (2007) observed that increases in free radical scavenging activity closely mirrored elevated concentrations of phenolic phytochemicals. The effectiveness of coatings in maintaining antioxidant capacity could be attributed to reduced LOX-mediated lipid peroxidation, thereby preserving membrane integrity and preventing oxidative chain reactions common under chilling stress (Djian et al., 2022; Magri et al., 2023).

A carrageenan-based edible coating (1.5% w/v) has been shown to reduce weight loss and oxidative stress as indicated by lower malondialdehyde, superoxide anion and hydrogen peroxide levels while preserving higher concentrations of phenolics, flavonoids and ascorbic acid in grapefruit during storage (Ali et al., 2025). This supports its role as a semi-permeable barrier that limits oxygen and moisture exchange thereby preserving antioxidants. In contrast, the lower DPPH inhibition observed in the combined $CaCl_2$ and carrageenan treatment suggests the possibility of antagonistic interactions between the components, which may negatively impact the bioavailability and activity of antioxidant compounds.

The ABTS radical scavenging activity of red-fleshed dragon fruit was significantly affected by both coating treatments and storage duration ($P \leq 0.05$) (Table 2). As illustrated in Figure 4, antioxidant activity measured as ABTS inhibition (%) varied considerably during the 20-day cold storage period at 10°C and 85-90% relative humidity.

At D0, freshly harvested fruits whether coated or uncoated exhibited the initial ABTS activities values ranging from 12.11 to 28.04%. By D5 and D10, uncoated fruits showed marked decline in scavenging activity with inhibition values of 9.32 and 9.85, respectively. In contrast, fruits coated with carrageenan and $CaCl_2$ whether applied individually or in combination, maintained significantly higher ABTS activity. This indicates that these edible coatings effectively delayed the degradation of antioxidant compounds during the early stages of storage. The coated fruits demonstrated stronger antioxidant potential due to their enhanced ability to scavenge free radicals and reduce oxidative stress. This improvement in ABTS activity likely corresponds to suppression of ROS accumulation and reduced lipid peroxidation, which are major contributors to chilling injury. The coatings may also maintain higher levels of ascorbate and glutathione thus supporting the non-enzymatic antioxidant defense network (Cice et al., 2024).

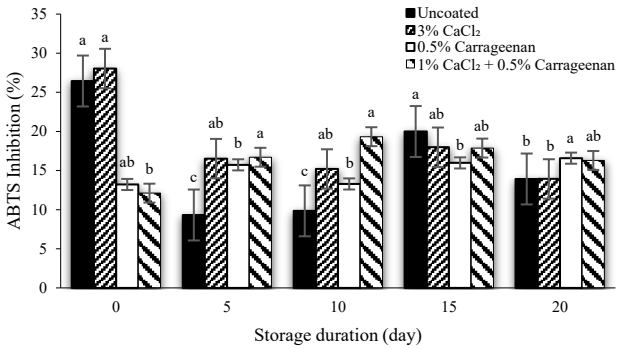

Figure 4: Effect of different edible coatings on ABTS radical scavenging activity of pitaya fruit stored at 10ºC/85% relative humidity. Different letters in each storage duration indicate significant difference at $P \leq 0.05$ according to Duncan's multiple range test. Vertical bars indicate S.E. of means for three replicates.

This protective effect is consistent with previous findings by Cruz-Monterrosa et al. (2023), who reported that polysaccharide-based coatings can modulate oxidative metabolism by limiting exposure to external oxidative stressors. By the end of the storage period (D20), fruits coated with 3% carrageenan exhibited the highest ABTS activity compared to uncoated fruits. However, the difference was not statistically significant when compared to the other coating treatments. This suggests that while carrageenan and calcium chloride-based coatings are effective in preserving antioxidant activity during early to mid-storage, their protective effects may diminish over time as fruit senescence advances and oxidative processes become dominant.

In addition to the present findings, it should be noted that fluctuating storage temperatures could alter the

performance of the coatings and potentially exacerbate chilling injury by disrupting membrane stability and coating integrity. The coatings may also influence fruit aroma and volatile profiles by altering respiration and gas exchange, highlighting the need for further sensory and volatile analyses to ensure the maintenance of quality attributes. Additionally, $CaCl_2$ and carrageenan-based coatings present a promising strategy for long-distance transport of red-fleshed pitaya as they can potentially reduce chilling injury and complement existing packaging technologies to extend fruit shelf life for premium markets. Future work may incorporate nano or microencapsulation technologies to stabilize betacyanins under export-chain conditions, since nanostructured coatings or Ca^{2+}-polysaccharide complexes could provide stronger protection against oxidation, moisture loss and pigment degradation during extended cold storage periods.

4 CONCLUSION

Postharvest coatings significantly improved the storage quality of red-fleshed pitaya with effects dependent on formulation and concentration. Single-component coatings particularly 3% $CaCl_2$ or 0.05% carrageenan were more effective in preserving antioxidant activity during early to mid-storage, whereas combined treatments provided superior structural protection and visual quality by minimizing chilling injury. Future research should focus on optimizing single-coating formulations and concentrations to maximize both bioactive preservation and fruit quality for extended storage and export.

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