DR-2052
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RESEARCH ARTICLE — Asian Journal of Dairy and Food Research

Enhancement of Shelf Life of Guava Fruits by Application of Chitosan based Nanoemulsion

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10.18805/ajdfr.DR-2052

ABSTRACT

Background: Edible coating is an effective and widely used approach for preservation of quality of fruits. The physiological changes remain intact with edible coating that leads to maintain the nutritional quality. The current study is an endeavor to develop an appropriate edible coating material that may be applied over fruits as freshness indicators.

Methods: Chitosan nanoemulsion was prepared with ultrasonication and guava samples were dipped in prepared nanoemulsion. Quality evaluation of guava samples was evaluated during 15 day storage in triplicates.

Result: Guava fruits were coated with chitosan based emulsion to preserve its nutritional qualities. Weight loss, TSS, acidity (%), ascorbic acid and 2,2 diphenyl-1-picrylhydrazyl (DPPH) inhibition activity were measured in coated and control guava samples. Samples coated with guava were found to preserve overall quality. TSS and acidity content, which is ripening indicator, was found to be not affected significantly in coated sample whereas in control samples TSS increased and acidity decreased during storage. The changes in weight loss were significantly more in control samples than coated samples. Ascorbic acid and antioxidant activity variation was observed more in control samples.

Key words: Acidity, Antioxidant, Coating, Nutritional, TSS.

INTRODUCTION

Guava (*Psidium guajava* L.) belongs to family Myrtaceae and considered as the commercial fruit crops of India. The guava fruit has major portion of edible part and represents excellent source of vitamin C and rich source of minerals like calcium, phosphorus, iron as well as the source of vitamin A, pantothenic acid, riboflavin, thiamin and niacin. Further, the polyphenols and carotenoids content provide its significantly high nutritional value (Thaipong *et al*., 2006). However, the fruits tend to rapid perish ability due to high respiration rate and consequently high metabolic activities being a climacteric nature that ripens after harvesting (Singh and Pal, 2008). It is susceptible to chilling injury when stored below 10°C, therefore require constant attention to prevent spoilage loss during transportation and marketing (Reyes and Paull, 1995).

Edible coatings have been conceptualized widely now days to enhance the shelf life of whole as well as fresh-cut fruits remarkably. In fact, the physiological processes such as respiration, degradation of cell wall, transpiration and deterioration by microbial action improves by the appropriate coating method, thereby maintaining the quality of the fruit and vegetable (Ali *et al*., 2011). They could be good alternative in place of synthetic preservatives. Moreover, edible coatings can be used as antimicrobial agents which helps in preventing the microbial spoilage and consequently extending the shelf life of the fruits (Raybaudi-Massilia *et al*., 2008). Generally, the materials polysaccharides, proteins and lipids for fabrication of edible coating can be used to extend the shelf life of various horticultural commodities (Chen *et al*., 2016; Gardesh *et al*., 2016). Pectin was used successfully as coating material to preserve colour and quality of dehydrated pineapple during storage (Saini and Sharma, 2016). Carboxy methyl cellulose was found suitable for edible coating due to its film solubity and elongation ability (Dipanwita and Kandeepan, 2017). Whey protein is extensively used for edible coating to enhance the shelf life of foods (Ranganathan *et al*., 2019).

Among all these coating materials chitosan (polysaccharide) based edible coatings are gaining popularity in recent times. Chitosan is obtained by N-deacetylation of chitin in an alkaline environment (Butola 2019). Chitosan is a non-toxic, non-allergenic, biodegradable, biocompatible and film-forming material with beneficial biological effects (antifungal, antibacterial, antitumour) (Chaudhary *et al*., 2020). Chitosan films are ecofriendly, biodegradable and having good mechanical properties that selectively allow gas permeability for CO_2 and O_2. The respiration rate of fruits decreases as the edible chitosan coating applied to the surface (Haaji *et al*., 2018). However, chitosan coating

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How to cite this article: Kumar, R., Sharma, P., Dipsikha, Sharma, T. and Ranaut, S. (2023). Enhancement of Shelf Life of Guava Fruits by Application of Chitosan based Nanoemulsion. Asian Journal of Dairy and Food Research. doi:10.18805/ajdfr.DR-2052.

Submitted: 02-12-2022 **Accepted:** 06-06-2023 **Online:** 04-07-2023

possesses high water vapour permeability thereby its application gets restricted in humid environment (Candir *et al*., 2018). Water vapour permeability signifies the organoleptic characteristics of stored fruit for its ability to fight against dehydration or rehydration of the film; Chitosan as an edible coating has low water solubility therefore acquires rigid crystal structure and not used directly. Chitosan and apple peel extract were used as edible coating for preserving nutritional qualities of strawberries. Chitosan (1, 2, or 3%) were used as edible coating for mangoes to maintain firmness, reduce water loss by preventing starch degradation (Silva *et al*., 2017). In the present study, chitosan based coating was applied on freshly cut guava fruit samples and various observation were made as an indicator for its freshness during fifteen days storage time.

MATERIALS AND METHODS

Guava fruits of varieties (Allahabadi Surkha) were purchased from local market, Dehradun. The fruits were selected on the basis of their uniformity in size, color and absence of mechanical injury. Tween80, glycerol and chitosan were purchased from Himedia, Mumbai, India. All other chemicals used in the study were of analytical grade. The study was conducted at Department of Food Technology, Uttaranchal University.

Preparation of chitosan nanoemulsion for edible coating

Chitosan (1% w/v) was dissolved in glacial acetic acid (1% v/v) and stirring was done with blender at 800 rpm for 5 min. The pH was kept to 5.2. Glycerol (0.75%) was added as plasticizer followed by addition of few drops of Tween 80 (1 g/100 mL chitosan solution) into the emulsion. Homogenization was done further at 5000 rpm for 10 minutes. The solution was then ultrasonicated at frequency 40 kHz using sonicator (Labman, LMUE-6, India).

Particle size of nanoemulsion

The particle size and zeta-potential value of chitosan emulsions was estimated by using particle size analyzer (Zetasizer Nano-ZS 90, Malvern Instruments, UK). Samples were diluted (1:100) with distilled water to avoid multiple scattering.

Weight loss

Five guava fruits cut samples were used for weight loss (%) determination. Weight loss was calculated by weighing fruits at the 0^{th} day and after fifteen days storage duration. Results were indicated as a percent weight loss.

Titratable acidity and total soluble solids (TSS)

10 g guava cut samples was mixed properly with help of blender with 50 ml distilled water and then after properly filtered. (%) Titratable acidity was determined by titration of obtained filtrate sample with standard solution (0.1 M) of NaOH using phenolphthalein as an indicator. Titratable acidity was expressed as % citric acid.

Total soluble solids expressed as °Brix was assayed using portable refractometer (Erma, India).

Ascorbic acid

Ascorbic acid (mg ascorbic acid per 100 g fruit pulp) content in coated guava samples was estimated by procedure followed by AOAC (1990). Ten gram of guava sample was extracted with 90 mL of 3% metaphosphoric acid. Centrifugation of samples was carried out at 5000×g for 15 min in properly mixed samples. The supernatant (10 mL) was collected for titration against 2, 6 dichlorophenol indophenol dye till pink rose color persists for about 20 s.

Antioxidant activity

Antioxidant activity of guava samples was analyzed by DPPH radical scavenging method (Wani *et al*., 2018). Guava fruit extract (0.1 mL) was added with 3.9 mL DPPH. The reaction mixture containing sample with DPPH was kept for about 30 min. in dark and absorbance was taken at 517 nm using UV-Spectrophotometer (Systronic-AU2701). The results were expressed as (% inhibition) by the equation given below:

$$\%\ \text{DPPH inhibition activity} = \frac{\text{Absorbance of control - Absorbance of sample}}{\text{Absorbance of control}} \times 100$$

RESULTS AND DISCUSSION

Total soluble solids and titratable acidity (%)

TSS and TA (%) of freshly cut samples coated with chitosan samples are shown in Table 1. These parameters are noteworthy for predicting for organoleptic quality of fruit as it helps in determination of soluble sugar and organic acid contents of fruits. The TSS of control fruit samples was observed to increase from 11.06±0.004 °Brix to 13.33±0.004 °Brix during storage (Fig 1). This increase could be featured due to the less respiration rate, deconstructed cell wall and the increase in dry matter due to water loss. Guava categorizes as climacteric fruit and having tendency to show rise in total soluble solids (TSS) content during storage (Singh and Pal, 2008). However, guava samples coated with chitosan were found to observe the gradual decrease in TSS (11.10±0.007 °Brix to 10.45±0.004 °Brix) during 15 day storage kept at 4°C (Fig 1) which was due to slow rate of metabolic activity by presence of functional edible coating (Radi *et al*., 2018).

The TA (%) of the uncoated fruits decreased from 0.92% to 0.70%, while in coated fruits this decrease was comparatively lower up to 0.88% at 15 day of storage. The titratable acidity (TA) was reported to decrease as well by earlier studies over storage since organic acids gets utilized during respiration process (Gol *et al*., 2015). The decrease in acidity was observed in uncoated fruits as the organic acids utilizes in form of substrates for respiratory metabolism as well as energy liberation during storage (Dubey *et al*., 2019). Earlier studies of Lim *et al*. (2011) also reported a decrease in titratable acidity of sweet cherries fruits.

Weight loss

The weight loss during storage in treated samples is presented in Fig 2. In control guava fruit sample the weight loss was found to be 47% after 15 days of storage. However, coated sample had observed less weight loss *i.e.* 8.6% during storage. The less weight loss in chitosan nanoemulsion coated sample was due to barrier properties provided by chitosan network. Wang and Rhim, (2016) observed the same pattern of weight loss in apricots coated with chitosan biopolymer in combination with polyphenolic extract. Chen *et al*., (2016) and Synowiec *et al*., (2014) also demonstrated reduction in weight loss in orange and apple fruit by incorporation of fig fruit and sweet basil seed extract into coating respectively.

Ascorbic acid degradation

Ascorbic acid significantly found in fruits and provides various health activities including immune system, antioxidative characteristics, healthy skins and gums. The decrease in ascorbic acid in guava samples is shown in Table 2. The decrease in ascorbic acid content was found significantly from its original value 300 mg/100g to 100 mg/100 for uncoated samples during 15 day storage. However, the ascorbic acid decreased to 280 mg/100 g from its original value. The decrease in ascorbic acid was mainly due to presence of ascorbic oxidase that causes oxidative deterioration of ascorbic acid. Chitosan had ability to be effective in controlling the losses of ascorbic acid for coating on fruits such as guava (Hong *et al*., 2012) and papaya (Ali *et al*., 2011).

DPPH antioxidant activity

The antioxidant activity decreased to 30% from its original activity of 70% in control sample whereas in coated sample the activity decreased only to 62%. In control sample the phenolic compounds generally degrades due to senescence and high rate of respiration (Ghasemnezhad *et al*., 2010). In our study the antioxidant activity was found more in all

Table 1: Changes in Total soluble solids (TSS) and % Titratable acidity (TA) in guava samples.

Storage	TSS (°Brix)		%TA	
days	Uncoated samples	Coated samples	Uncoated samples	Coated samples
0th	11.06±0.14	11.06±0.4	0.92±0.02	0.92±0.07
7th	12.13±0.23	10.87±0.12	0.84±0.03	0.90±0.03
15th	13.33±0.43	10.45±0.004	0.70±0.12	0.88±0.02

(Mean± standard error, n=3).

Fig 1: Guava samples a): Chitosan coated sample after 15th day b): Control samples after 15th day.

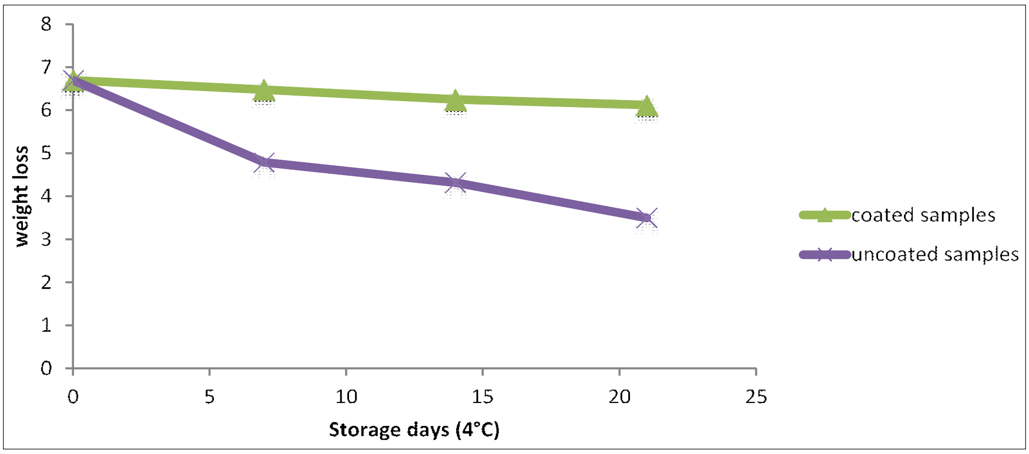

Fig 2: Reduction in weight loss in guava samples during storage at 4°C.

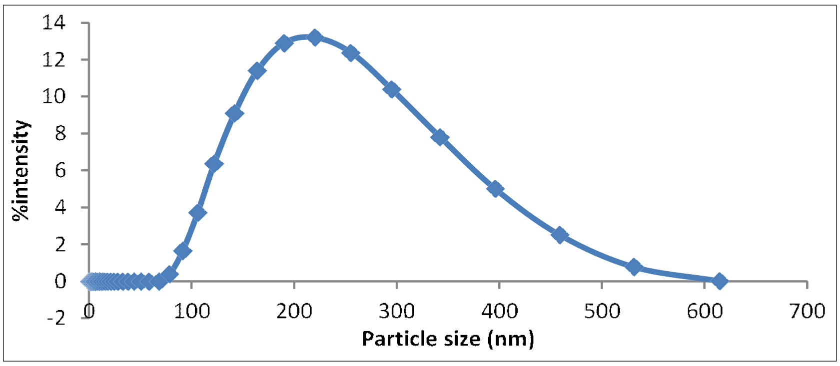

Fig 3: Particle size distribution of chitosan nanoemulsion.

Table 2: Ascorbic acid and DPPH inhibition activity (%) content in guava samples during storage.

Storage	Ascorbic acid (mg/100 g)		DPPH inhibition activity (%)	
days	Uncoated samples	Coated samples	Uncoated samples	Coated samples
0^{th}	300±1.27	300±2.34	70±2.32	70±1.23
7^{th}	220±3.21	292±3.32	53±1.56	67±2.12
15^{th}	100±2.98	280±3.76	30±3.21	62±1.42

(Mean±standard error, n=3).

coated fruits than control Samples. In fact, the coating of fruits modifies the internal atmosphere thereby reducing the risk of oxidative destruction of antioxidant compounds. Wang and Gao (2013) reported higher levels of total phenolic content and antioxidant activity in chitosan coated strawberries during storage. Chiabrando and Giacalone, (2015) observed similar observation in case of Chitosan coated blueberries.

Particle size of chitosan nanoemulsion

The particle size and zeta potential of chitosan nanoemulsion were found to be 202 nm (Fig 3) and -23 mV. The particle size ranges between 78-342 nm. The zeta potential between -30 to +30 mV is considered for stable nanoemulsion (Heurtault, 2003). Zeta potential plays decisive role in providing stability of nanoemulsion. In general, sufficient electrical charge requires to prevent aggregation of droplets by producing the repulsive force between droplet. Ghaderi-Ghahfarokhi *et al*. (2017) obtained the particle size of 235.6 nm produced by chitosan emulsion containing cinnamon essential oil using an ultrasonic water bath for 1 hr.

CONCLUSION

Chitosan based nanoemulsion have small size and large surface thereby having ability to provide antimicrobial and barrier properties against oxygen and moisture thus helps in preventing from deteriorative action in guava fruits. The present study confirmed that the use of chitosan nanoemulsion could augment the quality of the guava fruit. Moreover, degradation of ascorbic acid and antioxidant activity was found to preserve with application of edible coating. Edible coating could be an alternative approach in food industries to reduce post harvest quality losses in fruits.

Conflict of interest: None.

REFERENCES

Ali, A., Muhammad, M.T.M., Sijam, K. and Siddiqui, Y. (2011). Effect of chitosan coatings on the physicochemical characteristics of Eksotika II papaya (*Carica papaya* L.) fruit during cold storage. Food Chemistry. 124(2): 620-626.

AOAC. (1990). Offiial mMethod 967.21. Vitamin C (Ascorbic Acid) in Vitamin Preparations and Juices. (13^{th} ed.). Official Methods of Analysis Washington DC: AOAC International.

Butola, B.S. (2019). Recent advances in chitosan polysaccharide and its derivatives in antimicrobial modification of textile materials. International Journal of Microbiology. 12: 905-912.

Chen, C., Peng, X., Zeng, R., Chen, M., Wan, C. and Chen, J. (2016). Ficus hirta fruits extract incorporated into an alginate-based edible coating for Nanfeng mandarin preservation. Scientia Horticulturae. 202: 41-48.

Candir, E., Ozdemir, A.E. and Aksoy, M.C. (2018). Effects of chitosan coating and modified atmosphere packaging on postharvest quality and bioactive compounds of pomegranate fruit cv. 'Hicaznar'. Scientia Horticulture. 235: 235-243.

Chiabrando, V. and Giacalone, G. (2015). Anthocyanins, phenolics and antioxidant capacity after fresh storage of blueberry treated with edible coatings. International Journal of Food Sciences and Nutrition. 66(3): 248-253.

Chaudhary, S., Kumar, S., Kumar, V., Sharma, R. (2020). Chitosan nanoemulsions as advanced edible coatings for fruits and vegetables: Composition, fabrication and developments in last decade. International Journal of Biological Macromolecules. 152: 154-170.

Chen, C., Peng, X., Zeng, R., Chen, M., Wan, C. and Chen, J. (2016). Ficus hirta fruits extract incorporated into an alginate-based edible coating for Nanfeng mandarin preservation. Scientia Horticulturae. 202: 41-48.

Dipanwita, B. and Kandeepan, G. (2017). Selection of biopolymers to develop a biodegradable and edible film for packaging of luncheon chicken meat slices. Asian Journal of Dairy and Food Research. 36(1): 67-71.

Dubey, P.K., Shukla, R.N., Srivastava, G., Mishra, A.A. and Pandey, A. (2019). Study on quality parameters and storage stability of mango coated with developed nanocomposite edible film. International Journal of Current Microbiology and Applied Sciences. 8(4): 2899-2935.

Gardesh, A.S.K., Badii, F., Hashemi, M., Ardakani, A.Y., Maftoonazad, N. and Gorji, A.M. (2016). Effect of nanochitosan based coating on climacteric behavior and postharvest shelf-life extension of apple cv. Golab Kohanz. LWT-Food Science and Technology. 70: 33-40.

Ghasemnezhad, M., Shiri, M.A. and Sanavi, M. (2010). Effect of chitosan coatings on some quality indices of apricot (*Prunus armeniaca* L.) during cold storage. Caspian Journal of Environmental Sciences. 8: 25-33.

Ghaderi-Ghahfarokhia, M., Barzegara, M., Saharia, M.A., Gavlighia, H.A. and Gardini, F. (2017). Chitosan-cinnamon essential oil nano-formulation: Application as a novel additive for controlled release and shelf life extension of beef patties. International Journal of Biological Macromolecules. 102: 19-28.

Gol, N.B., Chaudhari, M.L. and Rao, T.R. (2015). Effect of edible coatings on quality and shelf life of carambola (*Averrhoa carambola* L.) fruit during storage. Journal of Food Science and Technology. 52(1): 78-91.

Hajji, S., Younes, I., Affes, S., Boufi, S., Nasri, M. (2018). Optimization of the formulation of chitosan edible coatings supplemented with carotenoproteins and their use for extending strawberries postharvest life. Food Hydrocolloid. 83: 375-392.

Heurtault, B. (2003). Physico-chemical stability of colloidal lipid particles. Biomaterials. 24(23): 4283-4300.

Hong, K., Xie, J., Zhang, L., Sun, D. and Gong, D. (2012). Effects of chitosan coating on postharvest life and quality of guava (*Psidium guajava* L.) fruit during cold storage. Scientia Horticulturae. 144: 172-178.

Lim, R., Stathopoulos, C.E. and Golding, J.B. (2011). Effect of edible coatings on some quality characteristics of sweet cherries. International Food Research Journal. 18(4): 1237-1241.

Radi, M., Akhavan Darabi, S., Akhavan, H.R. and Amiri, S. (2018). The use of orange peel essential oil microemulsion and nanoemulsion in pectin based coating to extend the shelf life of fresh cut orange. Journal of Food Processing and Preservation. 42(2): 13441. DOI: 10.1111/jfpp.13441.

Ranganathan, K., Sulaxna, K.C., Gokul, S., Vijayalakshmi, S., Shanmungam, N. (2019). Whey proteins: A potential ingredient for food industry- A review. Asian Journal of Dairy and Food Research. 37(4): 283-290.

Raybaudi-Massilia, R.M., Mosqueda-Melgar, J. and Martín-Belloso, O. (2008). Edible alginate-based coating as carrier of antimicrobials to improve shelf-life and safety of fresh-cut melon. International Journal of Food Microbiology. 121: 313-327.

Reyes, M.U. and Paull, R.E. (1995). Effect of storage temperature and ethylene treatment on guava (*Psidium guajava* L.) fruit ripening. Postharvest Biology and Technology. 6 (3-4): 357-365.

Saini, C.S. and Sharma, H.K. (2016). Effect of pectin coating on colour and quality of dehydrated pineapple during storage. Asian Journal of Dairy and Food Research. 35(2): 120-129.

Silva, G.M.C., Silva, W.B., Medeiros, D.B., Salvador, A.R., Cordeiro, M.H.M., da Silva, N.M., Santana, D.B. and Mizobutsi, G.P (2017). The chitosan affects severely the carbon metabolism in mango [*Mangifera indica* (L.) cv. Palmer] fruit during storage. Food Chemistry. 237: 372-378.

Singh, S.P. and Pal, R.K. (2008). Controlled atmosphere storage of guava (*Psidium guajava* L.) fruit. Postharvest Biology and Technology. 47: 296-306.

Synowiec, A., Gniewosz, M., Kra´sniewska, K., Przybył, J.L., Bączek, K. and Wêglarz, Z. (2014). Antimicrobial and antioxidant properties of pullulan fim containing sweet basil extract and an evaluation of coating effectiveness in the prolongation of the shelf life of apples stored in refrigeration conditions. Innovative Food Science and Emerging Technologies. 23: 171-181.

Thaipong, K., Boonprakob, U., Crosby, K., Cisneros-Zevallos, L. and Byrne, D.H. (2006). Comparison of ABTS, DPPH, FRAP and ORAC assays for estimating antioxidant activity from guava fruit extracts. Journal of Food Composition and Analysis. 19(6): 669-675.

Wang, L.F. and Rhim, J.W. (2016). Grapefruit seed extract incorporated antimicrobial LDPE and PLA fims: Effect of type of polymer matrix. LWT-Food Science and Technology. 74: 338-345.

Wang, S.Y. and Gao. (2013). Effect of chitosan-based edible coating on antioxidants, antioxidant enzyme system and postharvest fruit quality of strawberries (Fragaria × aranassa Duch.). LWT - Food Science and Technology. 52: 71-79.

Wani, S.M., Masoodi, F.A., Ahmad, M. and Mir, S.A. (2018). Processing and storage of apricots: Effect on physicochemical and antioxidant properties. Journal of Food Science and Technology. 55(11): 4505-4514.