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ORIGINAL PAPER

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Polysaccharide-based composite coating formulations for shelf-life extension of fresh banana and mango

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Abstract The effects of four different composite coating formulations based on polysaccharides on maintaining quality and an extended shelf-life of banana and mango at 27±2 °C were investigated and compared with commercial Waxol-coated and uncoated fruits. The formulations consisted of modified starch, cellulose and chitosan, blended with a suitable lipid component and a wetting agent. Quality parameters measured included firmness, total soluble solids and titratable acidity. Physiological parameters measured were CO2 evolved and weight loss due to respiration and transpiration. The polysaccharide-based coatings displayed retarded colour development, lower acidity and greater firmness values compared to Waxol and control. CO₂ evolution and loss in weight were also reduced significantly. The data were also subjected to PCA, to differentiate the characteristics of the five types of films. Chitosan-based coatings were much superior in prolonging the shelf-life and quality of banana and mango.

Keywords Chitosan · Starch · Cellulose · Coating formulations · Shelf-life

Introduction

The world food shortage and continually increasing demand for high quality nutritious foods have paved the way for improved methods of storage for fresh fruits and vegetables. A longer market season would be desirable for both domestic and export trades, so as to make the commodity available to the consumer for a longer period

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of time. The predominant methods used to preserve fresh fruits during handling and subsequent marketing include controlled atmosphere and modified atmosphere packaging (MAP) techniques in conjunction with refrigerated storage. MAP reduces respiration due to the change in the concentration of O_2 and CO_2 in the fruit surroundings, and delays senescence. Though economical, MAP is more difficult to implement because of rather complicated interactions between the product and the packaging material. Also, polymeric films with a wide range of permeability characteristics are rather limited in number. Another limitation involved is environmental concern about using plastic materials.

A recent approach has been the use of a coating that is edible and semipermeable to CO_2 and O_2 . Banks [1] reported that an edible coating mixture composed of sucrose fatty acid ester (SFE) and sodium carboxymethyl cellulose (TAL-Prolong) would produce, after application, a semipermeable modified atmosphere within fresh fruits. SFEs have been tested extensively on banana [2, 3, 4]; changes in the internal concentrations of CO_2 , O_2 and ethylene, and delay of ripening have been noted. Delayed ripening was also reported in pears and apples coated with Nutri-Save (N,O-carboxymethyl chitosan) [5]. EI Ghaouth [6] reported increases in firmness and titratable acidity with reduced CO₂ production in strawberries after treatment with chitosan. Encouraging results were also obtained when Nutri-Save was applied to tomato [7], pepper, squash, cauliflower, sprouts and broccoli [8].

Banaras et al. [9], however, reported that neither Nutri-Save nor Semperfresh (an improved form of TAL-Prolong) was effective in extending the post harvest shelf-life of bell pepper or long green pepper stored at 21 °C. Krishnamurthy et al. [10] reported that a post harvest application of TAL-Prolong delayed ripening in banana; however, the colour and texture were inferior compared to the control. Such contrasting results were attributed to differences in maturity of the fruit and cultivar and permeability of the coatings per se. The objectives of this study were to optimize and evaluate edible coatings based on starch, cellulose and chitosan derivatives in maintaining the quality and extending the shelf-life of freshly harvested banana and mango.

Materials and methods

Fruits and chemicals. Optimally matured banana (Musa robust-ana) and mango (Mangifera indica ev Alphonso) grown on local farms were harvested and washed thoroughly with water. Fruits of uniform size, free of physical damage and fungal infection, were randomly distributed into groups, individual in the case of mango and hands of eight to ten in the case of banana. Each represented one replicate, and for each treatment, six to eight replicates were used. Polysaccharide derivatives such as carboxymethyl cellulose, carboxymethyl starch, hydroxypropyl starch and N,O-carboxymethyl chitosan were prepared as per the reported methods [11, 12, 13, 14, 15]. Chitosan was prepared by heterogeneous alkaline N-deacetylation of chitin [16].

Coating formulations. To prepare 100 ml of coating solution (2–3% total solids for banana and 1.5–2.5% for mango, w/v), 1.0–2.0 g of polysaccharide derivatives were dissolved in double distilled water and blended with glycerol monostearate (0.8%). Tween-80 (0.2 ml) was added to emulsify and improve wettability. The solution was stirred for 30 min and the insolubles were removed by filtration [17, 18]. Coatings based on chitosan and N,O-carboxymethyl chitosan were designated as F1 and F2, whereas S1 and S2 refer to carboxymethyl derivatives of cellulose and starch, and hydroxymethyl starch and hydroxypropyl starch, respectively.

Coating application. The fruits were dipped into the above coating solutions, Waxol (positive control, 6% for banana and 3% for mango) or water, the excess solution was drained and the coated fruits were air-dried. A 1% chitosan solution was applied at the fascicle region of banana to curtail water loss and to prevent fungal growth. After drying, the fruits were stored at 27±2 °C, at a RH of 65% for different lengths of time. At regular intervals the fruits were removed and analysed.

Quality attributes. A sample of four to five fruits in total were randomly removed from each and assessed each week for banana, and on alternate days for mango. The presence of mould was evaluated visually. Cumulative physiological losses in weight (PLW) of the fruits were determined by difference in weight after 24 h. Firmness was measured with Instron Universal Testing Machine (Model 4301, Instron, Conton, USA) and was expressed as kilogram force. Total soluble solids (TSS) content of the fruits was determined by Abbe's refractometer. Acids were titrated to phenolphthalein end point with 0.1 M NaOH and expressed as percentage malic acid. Reducing sugar, after extraction with 80% hot ethanol was determined by the DNS method [19] and expressed as milligrams of reducing sugar as glucose/1 g pulp. Respiration rate was determined on alternate days and expressed as milligrams of CO₂/kilogram/hour [20]. Sensory attributes like colour, texture, flavour and taste were assessed by a panel of six laboratory personnel familiar with banana and mango grades. The fruits were rated on an hedonic scale of 0-7 (7=excellent, 1=poor). Data were evaluated by using ANOVA.

The correlation between sensory and instrumental data was studied by PCA through "Teach/Me data analysis" software [21] and Microsoft Excel. The bubble graphs were obtained using Statistica version 5 (Statsoft, Okla., USA). Two different PCA models were developed separately for instrumental and sensory attributes. An $n \times p$ data matrix of p=6 variables containing pulp to peel ratio, TSS, texture, titratable acidity and reducing sugar for instrumental, and colour and appearance, texture, flavour, taste, and overall quality for sensory attributes; with five different types of films, viz., Waxol, F1, F2, S1, S2 plus control (untreated fruits) as n, was the starting point for further analysis.

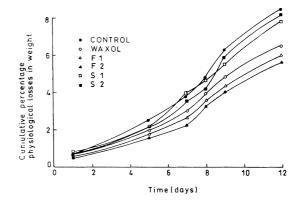


Fig. 1 Cumulative percentage physiological loss in weight (PLW) of banana during storage at 27±2 °C and 65% RH.

General. All the chemicals used were of reagent grade. Tween-80, propylene oxide and cellulose were from Fluka, Switzerland, Waxol is an emulsion formulation prepared in-house for shelf-life extension studies, and glycerol monostearate was from Tate and Lyle, England.

Results and discussion

Analysis of PLW

Polysaccharide coatings alone have no significant influence on the fruit weight loss and other quality attributes (data not shown). Addition of a lipid component such as glycerol monostearate, palmitic acid or SFE significantly enhanced the effectiveness of these coatings, indicating their regulation of the hydrophilic-hydrophobic balance, which would in turn restrict the water loss. Of the series of coating formulations with different levels of percentage of total solids tested, those in the range 2–3% were found to be suitable only for banana. Mangoes treated with the same formulations showed significant reduction in PLW, but during the storage period, off-flavour developed due to anaerobiosis (data not shown) suggesting that these coatings have less air permeability and water vapour transmission rates. Formulations with a lesser total solid levels of 1.5-2.5% however, were effective in reducing the weight loss without adverse effects on the quality of mango. Usually, coatings have an optimum percentage of solid levels, according to the type of fruit or vegetable cultivar [22]. Fig. 1 shows the influence of polysaccharide-based composite coatings on the weight loss of banana stored up to 12 days. Compared to control (uncoated, only water dipped) and Waxol (positive control), F1 and F2 showed PLW of 30-35%, whereas S1 and S2 exhibited least reduction (<5%). Waxol-coated fruits, though, showed less weight loss than S1 and S2, and the rate of weight loss was slightly higher than that of F1 and F2. Fig. 2 shows the weight loss for mango over a period of 7 days. As can be seen, F2 and S1 showed least weight loss compared to Waxol and control. The differences in the ability of these coatings to reduce weight loss could be attributed to differences in the

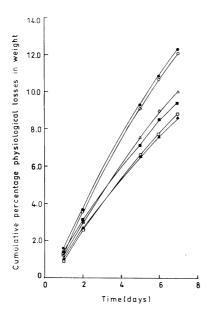


Fig. 2 Cumulative percentage PLW of mango during storage at 27 ± 2 °C and 65% RH

permeability characteristics, which in turn could be due to differences in the chemical nature of polysaccharides and their relative concentrations. Overall, the use of the above coatings improved the sensory characteristics of banana and mango by reducing the water loss and maintaining dark green colour, with glassy, shining and moist-like appearance. When viewed under a light microscope, a uniform coating of the fruit surface without cracks and pinholes was observed.

Effect on quality attributes

The above coatings had beneficial effects on firmness, titratable acidity and reducing sugar content of banana (Table 1). Banana fruits treated with F1, F2 and S2, were firmer, lesser in reducing sugar content and higher in titratable acidity after 12 days than control- and Waxoltreated fruits. They delayed ripening as indicated by changes in TSS, reducing sugar content, texture, and pulp to peel ratio values. The changes in fruit texture

Table 1 Effect of polysaccharide-based composite coating formulations on quality attributes of banana at 27 ± 2 °C, 65% RH, after 15 days of storage. Coatings based on chitosan and *N,O*-carboxy-

during ripening results from both alteration in cell wall structure and the degradation of starch. As the ripening progresses, bound carbohydrate fractions, especially pectic substances and hemicelluloses are rapidly depolymerized by hydrolases [23]. To some extent this trend was reflected in the pulp to peel ratio and firmness values (Table 1). The coated fruits had higher firmness (>150 kg force), lower pulp to peel ratios (<2.0) and acidity values when compared to control. During ripening starch is degraded rapidly by the combined action of amylases, starch phosphorylase and α-1,6-glucosidase and sucrose synthase to sugars such as sucrose, glucose, and fructose along with traces of maltose. In the pulp, sucrose is the predominant sugar at the start of the ripening, and its formation precedes accumulation of glucose and fructose [24, 25]. The reducing sugar content and TSS of polysaccharide-based coated fruits were lower than control, suggesting that the former synthesized reducing sugars at a slower rate than the control (21.5–25.5 mg of glucose/g of pulp as against 32.5 mg of glucose/g of pulp for control). A similar trend was also observed in the case of mango (data not shown). Thus, the results demonstrate that the polysaccharide-based coating formulations slow down the metabolism to give prolonged storage life. Furthermore, these fruits later developed yellow colour, and coatings neither affected appearance nor caused phytotoxicity even after storage of 21 days for banana and 8 days for mango. Etheral treatment after the desired storage period was found to be advantageous for a uniform colour development. However, such an extended storage resulted in control fruits turning black with a collapsed structure due to overripening and fungal infection.

Chitosan-based coatings, in addition to delaying ripening, had the added advantage of an antifungal property. The fascicle region of banana in particular is more susceptible to fungal growth. A number of antifungal agents have been used to control the mould growth. Application of an additional 1% chitosan coating to the fascicle region, however, significantly reduced the incidence of mould growth (results not shown). This was attributed to either fungistatic property of chitosan per se or its ability to induce defence enzymes (i. e. chitinase and β -1,3-glucanase) and phytoalexins in plants or a combination of both.

methyl chitosan were designated as F1 and F2, whereas S1 and S2 refer to carboxymethyl derivatives of cellulose and starch, and hydroxymethyl starch and hydroxypropyl starch, respectively

Coating	Pulp to peel ratio	Total soluble solids °brix	Texture kg force	Titratable acidity % malic acid	Reducing Sugar mg/g of pulp
Control	2.20	25.0	111.8	0.22	32.5
Waxol	1.74	25.0	150.0	0.18	25.5
F1	1.83a	19.2 ^{a,b}	154.6a	$0.12^{a,b}$	$22.0^{a,b}$
F2	1.73a	$10.0^{a,b}$	177.6 ^{a,b}	$0.11^{a,b}$	23.0a
S1	1.64a	$7.5^{a,b}$	168.9a,b	0.15a	21.5 ^{a,b}
S2	$1.46^{a,b}$	4.5 ^{a,b}	157.7a	0.15a	22.0^{a}

^aMean values within the same column are significantly different (P<0.05) between control and experimental

bMean values within the same column are significantly different (P<0.05) between Waxol and experimental

Table 2 Effect of coating formulations on the sensory qualities (scored 1–7) of banana after 21 days of storage at 27±2 °C and 65% RH

^aMean scores within the same column are significantly different (*P*<0.05) between control and experimental

Coating	Colour and appearance	Texture	Flavour	Taste	Overall quality
Control	2	2	3	3	3
Waxol	6	6	6	6	6
F1	6^a	7a	7a	7a	7a
F2	6^a	7a	7a	7a	7a
S1	3a	6a	6 ^a	6a	6^{a}
S2	6 ^a	7a	6 ^a	6a	6a

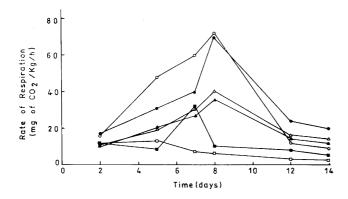


Fig. 3 Rate of respiration of banana during storage at 27 \pm 2 °C and 65% RH

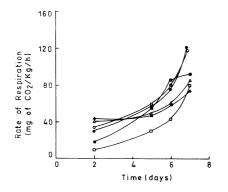


Fig. 4 Rate of respiration of mango during storage at 27 ± 2 °C and 65% RH

Respiration rate

Coating fruits with semipermeable film has generally been shown to retard ripening by modifying the levels of endogenous CO₂, O₂ and ethylene [3, 4]. From the CO₂ production rates (Fig. 3) it is clear that F1- and F2-coated banana showed characteristic climacteric peak on the 8th day, that for S2, the peak was seen on the 7th day and that for S1, however, no such trend was observed. Although, their respiration patterns were generally similar to control, their CO₂ production rates were significantly lower than that of Waxol-treated and control fruit at any stage of storage. In the case of the control, the CO₂ production rate increased rapidly from an initial value of 15 mg CO₂/kg/h to a peak value of 75 mg on day 8, whereas F1, F2 and S2 showed 40, 35 and 32 mg CO₂/kg/h, respectively. A similar trend was observed in

Table 3 Factor loadings for the first three factors in the principal component analysis

Variable	Factor 1	Factor 2	Factor 3
Pulp to peel ratio Total soluble solids Texture Titratable acidity Reducing sugar Colour and appearance Texture Flavour Taste Overall quality	0.4499	-0.3858	-0.5168a
	0.4009	-0.6660 ^a	0.5653
	-0.4707a	-0.2805	0.3493
	0.4256	0.5533	0.5279
	0.4838a	0.1509	-0.1126
	0.4040	0.9053 ^a	0.1310
	0.4547	-0.0740	-0.8875a
	0.4583a	-0.2414	0.2550
	0.4583a	-0.2414	0.2550

^a Indicates the respective principal component factors to which they belong

mango; the control showed a peak value of 120 mg on day 7, whereas F1, F2, S1 and S2 had CO₂ production rates of 83, 77, 80 and 86 mg/kg/h, respectively (Fig. 4). On the other hand, Waxol had no influence on the rate of respiration of either banana or mango. This suggested the polysaccharide-based coatings had the dual effects of showing less air permeability in restriction of CO₂ diffusion and causing beneficial secondary physiological changes during the ripening process. Kader [14] has listed the processes affected by elevated CO₂ in fruits and vegetables. High CO₂ concentrations reduced respiration rates, and prevented or delayed responses to ethylene.

Sensory evaluation

Sensory evaluation of fruits coated with these polysaccharide-based formulations revealed significant differences in colour, texture, flavour and taste compared to Waxol-coated and control fruits (*P*<0.05). F1- and F2treated fruits had maximum freshness, surface colour, texture and taste and were best even after 21 days of storage while those treated with S1 and S2, though comparable in taste and flavour, had inferior colour and texture. Waxol-treated fruits, though ripened to an acceptable quality, were of inferior texture. Uncoated (control) fruits on the other hand, blackened due to overripening and fungal infection (Table 2), and exhibited a very soft, collapsed texture.

The PCA [26, 27] indicated that the three components (see later) met the criteria of their eigen value exceeding 1.0 and the cumulative percentages of total variance of

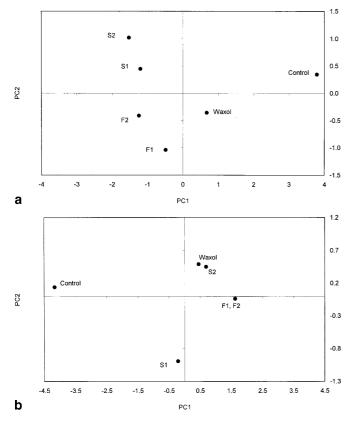


Fig. 5a,b Two dimensional plots, a instrumental and b sensory, of scores on the first two principal components

the three factors were 98.16% and 100% for instrumental and sensory models respectively, where the total variance is the sum of the individual variances of each of the original variables.

The loadings for each component give an indication of the importance of the original value to that component. The loadings are presented in Table 3 for both instrumental and sensory attributes. Examination of this indicates that for instrumental attributes, component 1 (PC1) primarily involved texture and reducing sugar; component 2 (PC2) involved total soluble solids and titratable acidity; and component 3 (PC3) involved pulp to peel ratio. Similarly, for sensory attributes PC1 involved flavour, taste, and overall quality; PC2, colour and appearance; and PC3, texture.

The scores of the first two principal components which explain 92.36% of the total variance of instrumental attributes and 99.16% for sensory attributes are plotted in Fig. 5a,b. For instrumental attributes (Fig. 5a), there is a compact grouping of F and S with reference to PC 1 and PC 2. With reference to PC1, F and S are in the same group whereas Waxol and control are in different groups; with reference to PC2, F1 and F2 are in the same group as Waxol, and S1 and S2 are similar to control. However, the dispersions within the same groups were significant, indicating the difference in the five film types. Similarly for sensory attributes (Fig. 5b), F and S are in different groups. With reference to PC1, S1 is in

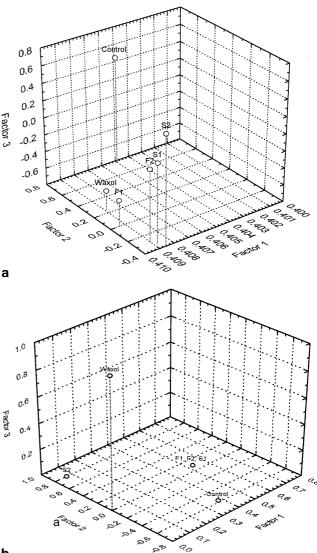


Fig. 6a,b Three dimensional bubble plots, a instrumental and b sensory, of scores on the three principal components

the same group as control, whereas S2, F1 and F2 were in the same group with that of Waxol. With reference to PC2, only S2 matched with both control and Waxol, whereas the difference between F1 and F2 was insignificant. However, with reference to instrumental and sensory attributes both PC1 and PC2 allow differentiation of the two types of films and specifically instrumental analysis allows differentiation within the same group as well.

The above discussion indicates that both instrumental and sensory analyses help in discriminating between the films. Further details are appreciable from three dimensional bubble plots of PCA scores of the first three components, as indicated in Fig. 6a,b for both instrumental and sensory attributes, respectively. Nevertheless, Fig. 6a showed considerable variations due to wide differences in quality attribute values (Table 1), unlike in Fig. 6b, wherein the sensory scoring values had more restricted variations.

In conclusion, starch, cellulose and chitosan-based composite coating were more effective than Waxol in shelf-life extension and maintaining the quality of banana and mango stored at 27±2 °C and 65% RH. Starchand cellulose-based coatings significantly reduced respiration rates; however, their effect on weight loss was insignificant. Chitosan-based coatings, especially F2, on the other hand significantly reduced the weight loss and respiration rate. The coated fruits were fresh, firmer and lower in titratable acidity than the control fruits. Chitosan-based coatings also provided protection against fungal infections. PCA of the data facilitated the differentiation of the five types of films.

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