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RESEARCH ARTICLE

Edible skin coating material containing neither ammonia nor morpholine

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ABSTRACT

Ammonia and morpholine have been widely used in fruit coatings as a base to ionize fatty acids, all over the world. However, both have certain limitations now. Morpholine is banned in most countries due to its carcinogenic effects and ammonia based waxes are difficult to make owing to its high volatility. It also causes environmental hazard/irritation for the workers of the waxing factory. So an edible skin coating material without ammonia and morpholine was made in present research. The edible skin coating material was made from ethanol based microemulsion of castor oil, shellac, gum rosin and gum acacia, by atmospheric method. A new laboratory method was developed in the course to prepare an environment friendly edible skin coating material. The newly developed edible skin coating material was applied to mandarins where it successfully reduced the rate of weight loss, decrease in firmness and vitamin C, with better sensory qualities than the uncoated ones. It was also environment friendly for workers of waxing factory than the ammonia based fruit coating. The edible skin coating material was also successfully tested on cucumber and bell-pepper. It could be concluded from the results that good edible skin coating material can be made without ammonia or morpholine.

Keywords: Ammonia, edible skin coating material, ethanol, fruit coating, mandarins

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INTRODUCTION

The edible skin coating material needed to be developed as a result of increasing demand of high quality; low cost nutritious food by consumers in combination with the ecological need to minimize disposable packaging waste (Del-Valle et al., 2005). Composite (microemulsion or nanoemulsion) coatings are promising in improving moisture and oxygen barrier properties of hydrophilic coating materials and improving coating adhesion, durability, and delivery of active compounds. Research on identifying the most compatible material combinations and stability of the emulsion system needs to be continuously investigated for meeting the specific needs of coating fresh fruits and vegetables (Lin and Zhao, 2007). Emulsified composite films of polysaccharides and lipids improve moisture barrier properties and can be manufactured in a single step in contrast to a layer-by-layer process for multilayer films (Hambleton et al., 2009).

Morpholine and ammonia are widely used in fresh fruit coatings. But both have certain drawbacks. Fresh fruits processed in the European Union (EU), cannot be coated with morpholine-containing fruit coatings as morpholine is not approved for use as a food glazing agent for fresh fruits in the EU under European Union food additives legislation (EC Regulation 1333/2008), although such coatings are permitted for use in the United States, Canada and Australia (Anonymous, 2010). Also morpholine has limited approval, because like other amines, it can react to form carcinogens (Hagenmaier, 2004).

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Ammonia on the other hand, evaporates very rapidly (boiling point = -37 °C), especially at high temperatures (about 95°C). Even at ambient temperatures, ammonia evaporates from coating formulations fast enough to make it necessary to keep them tightly closed. Ammonia vapors are unpleasant, toxic and, in addition, can cause false alarms in packinghouses that use human detection of its odor as a warning that the ammonia based refrigeration system is leaking (Hagenmaier, 2004). At present, the area of edible films is not considered a priority by industry due to overall public perception and hesitation about adding more chemicals to fresh produce (Farber et al., 2003). In the past few years, research efforts have focused on the design of new eco-friendly coatings based on biodegradable polymers, which not only reduce the requirements of packaging but also lead to the conversion of by-products of the food industry into value added film-forming components (Vargas et al., 2008). Studies on edible coatings have to focus on aspects such as consumer acceptability and final cost of the coated product (Olivas et al., 2009). The development of new edible coatings with improved functionality and performance for fresh fruits is one of the challenges of the post harvest industry.

The purpose of present paper is to make an SCM which could suit to both fruits and vegetables besides being edible, natural, and workers friendly and simple to manufacture at the same time without the use of advanced and complex technology.

MATERIALS AND METHODS

Selection of ingredients for edible skin coating material (ESCM)

Firstly, different potential natural ingredients (polysaccharides, proteins, lipids and/or resins) for the formulation of ESCM were identified on the basis of easy and abundant availability. Then sorting was done for the development of a new ESCM based on their physico-chemical properties. Proteins were left out in this process because these have properties similar to those of polysaccharides (Krochta, 2002) but are costlier and less abundantly available than polysaccharides. Before finalizing the ingredients for the preparation of ESCM, some ingredients were tested individually for their coating effect to reduce the amount of trial emulsions. In this context, different selected edible oils (castor oil, linseed oil, canola oil, black seed oil, ajwain oil), different gums of plant origin (gum acacia, gum karaya, gum pine, gum tragacanth and guar gum), starch, resins of plant (rosin) and animal origin (shellac) were tested mainly for weight loss and gloss (data not shown). As the properties of emulsions are greatly affected by the type and concentration of the emulsifier, so different emulsifiers were tested (data not shown) in this context and a non-ionic emulsifier from the group of polysorbates was finalized because of its increased stability, formulating flexibility, wider compatibility and non-reactance with active ingredients of the ESCM. The regulatory status of different ingredients of the finalized formulation is shown in table 1.

Preparation of ESCM

The ESCM was prepared by simple atmospheric method in the following way: Sodium hydroxide and ethanol were dissolved in distilled water. Gum acacia and/or starch, rosin and/or shellac were dissolved by mild heating (40°C) in this water one by one and it was named solution A. Non-ionic emulsifier was dissolved in some water and added to castor oil and this was named solution B. Both solution A and B were slowly and continuously mixed while constant stirring was provided throughout to make a homogeneous microemulsion of ESCM. As the ESCM was clear and not milky so there was no need of homogenization or other high energy turbulence after final preparation and it was considered that the ESCM being prepared is fairly a microemulsion with the globule size less than 0.5µm.

Table 1 Regulatory status and function of different ingredients of the finalized formulation

Name of Ingredient	Regulatory Status	
	FDA[a]	EU[b]
Castor oil	21CFR 172.876	E1503
Rosin	21CFR 172.210	E915[c]
Shellac	21CFR 175.300	E904
Gum acacia	21CFR 172.780	E414
Sodium Hydroxide	21CFR 184.1763	E524
Polysorbate 20	21CFR 178.3400	E432
Ethanol	21CFR 184.1293	E1510

[a]Food and Drug Administration; [b]Fed. of European Food Additives and Food Enzymes Industries; [c]South African National Halal Authority

Fruit treatment

Mandarins (*Citrus reticulata* cv. Blanco) were grown in the orchards of Orange Research Institute, Sargodha (Punjab, Pakistan). Cucumber (*Cucumis sativus L.*) and bell pepper *(Capsicum annuum L.*) were grown in the agricultural farms of Haji Sons Farms, Chiniot (Punjab, Pakistan). Washing of citrus fruits was done firstly with simple water and then with the fungicidal solution of thiabendazole (Textar® 60-T by Tecnidex, Valencia, Spain) @2000ppm in washing tank. Subsequent drying was carried out in hot air tunnel at 50°C for 1.50 minutes. For comparison, waterwax Fomesa (Fruitech, s.l., Valencia, Spain) containing 20% total solids (10% oxidized polyethylene, 8% wood rosin and 2% ammonium hydroxide), was chosen because it is well-known market brand and is oxidized polyethylene based which is a synthetic compound. Waxes were applied separately to fruits by a combination of spraying and brushing method. The rate of coating was maintained @1ml/Kg of fruit because this amount of coating was officially recommended by the manufacturer (Fruitech, s. l., Valencia, Spain) and was typical of the amount of coating widely practiced by the citrus industry of Pakistan. The fruits were weighed before and after the coating to monitor the amount applied which was about 0.20g for mandarin. This amounts of coatings resulted in about 0.15mg/cm^2 of dry coating, calculated from wet weight and known solids content (10%). Additional fungicide imazalil (DECCOZIL®50 by Decco Italia, Italy) each @1L/200 L wax were added to Fomesa while no additional fungicide was added to ESCM at the time of application. After waxing, fruits were dried in a hot air tunnel at 55°C for 1.75 minutes. The packed fruits were pre-cooled to internal temperature of 5°C by the blast air in a reefer container and then transported by the same to cold chambers of Post Harvest Research Centre (PHRC), Faisalabad and stored at 5±2°C with 85-90% relative humidity (RH).

Cucumber and bell peppers were washed in our laboratory with 200ppm solution of sodium hypochlorite and dried before waxing. ESCM was applied to each piece of vegetable by spraying method using pressure hand sprayer and all-around coating was ensured by hand (using latex gloves). Coated cucumbers and bell peppers were dried in a hot air oven at 50°C for 02 minutes. The amount of coating applied was about 0.12g for cucumber and 0.18g for bell peppers. Cucumbers and bell peppers were stored at 10±2^{0}C with 90-95% relative humidity for 14 and 35 days respectively. The quality of ESCM was evaluated on the basis of physical characteristics and performance.

Physical characteristics

Physical characteristics and stability were evaluated by creaming and coalescence, turbidity, viscosity, specific gravity and amount of coating lost by flaking. The physical stability of the prepared emulsion was determined by heat-cool cycling between refrigerator temperature and 45°C with storage at each temperature of 24 hrs. Creaming (a reversible separation of the emulsion into dilute and concentrated region) or coalescence phenomenon (an irreversible destruction of emulsion) of the emulsions was observed after each cycle had finished (Viyoch J. et. al., 2003) and separation/destruction was recorded from 1-5 scale (1: Nill, 2: < 5%, 3: 5-15%, 4: < 15-25% and 5: >25%). Turbidity was measured (n=2) with a turbidity meter (2100N Laboratory Turbidimeter, HACH, US). Viscosity was determined by a viscotester (Rion Viscotester, VT-04, Extech International Corp., Boston, USA) with a no. 1 rotor operating at 63.8 rpm. Amount of coating lost by flaking was measured (n = 6) by bumping and rubbing together a pair of fruit from the same treatment, wiping the fruit with a cloth and measuring the nearest 0.1 mg the weight uptake. The reported means are based on 2 measurements each on 3 different pairs of fruits (Hagenmaier, 2004).

Performance

Performance was evaluated by comparing the results of uncoated samples, ESCM coated samples and synthetic wax coated fruits. The parameters compared included physiological weight loss, firmness, vitamin C contents, gloss, sensory analysis (color, flavor, taste, and overall acceptability) and worker friendly aspects (smell and irritation).

Individual packs of mandarins were weighed by a digital weighing balance (Sartorius GM 1501, Precision Weighing Balances, Bradford, MA, US) at the beginning of the study and thereafter weekly until the end of the storage period. The result was expressed as percentage of weight loss relative to the initial value (taken as 100%).

$$\text{Weight Loss (\%)} = \frac{\text{Initial Weight} - \text{Recorded Weight}}{\text{Initial Weight}} \times 100$$

The firmness of fruit/vegetable was determined by using a penetrometer (model 53205, TR di Turoni, Forli, Italy) and results recorded as KgF. The vitamin C contents were determined according to the method as described by AOAC (2000). Gloss was determined (n=50) with gloss meter (BYK Gardner Micro-Tri-Gloss, Silver Spring, MD). The reported means are based on 10 measurements on each of 5 different fruits. Sensory evaluation was conducted by a panel using 9-point hedonic scale (Lee et al, 2003). The worker friendly aspects of both waxes were recorded on a nine-point hedonic scale (Lee et al, 2003). Four personals (foreman, sorter, operator and packer), who were closer to the grading and packing operation area were selected from 4 different factories and the reported values are means of all four factories. The data was analyzed using Statistix 8.1.

RESULTS AND DISCUSSION

Physical Characteristics

Good fruit coatings should necessarily be prepared as microemulsion, so that when the water evaporates the emulsion will have a smooth surface. This implies that the coating appears transparent to translucent (with globule size

<0.02µm diameter) and not milky white (Prince, 1977). Many formulations, among all the formulations prepared, were transparent/clear (all data not shown). Physical Characteristics of some of the good formulations are shown in table 2. Only the best formulation (E) was chosen on the basis of physical characteristics for comparative study, although other good formulations were also tested individually on mandarins alone (data not shown).

Table 2. Physical characteristics of some good formulations (mean values where applicable, p< 0.05, LSD)

Characteristic	Formula					
	A	B	C	D	E	F
Visual appearance	creaming*	creaming*	clear	clear	clear	clear
Creaming	3 [c]	3 [c]	2[b]	2[b]	1[a]	2[b]
Coalescence	3 [c]	3 [c]	2[b]	2[b]	1 [a]	2 [b]
Turbidity (NTU)	434[d]	402[d]	305[c]	275[c]	118[a]	201[b]
Viscosity (cp)	350[d]	325 [d]	225[c]	213[c]	147[a]	195[b]
Specific gravity g/cc	1.005 [c]	1.003[c]	0.999[b]	0.993[b]	0.986[a]	0.989[a]

A= starch, shellac, rosin, castor oil (8%) B= gum acacia, starch, rosin, castor oil (5%)
C= gum acacia, rosin, shellac, castor oil (5%) D= gum acacia, shellac, rosin, castor oil (4%)
E= gum acacia, shellac, rosin, castor oil (2%) F= gum acacia, shellac, rosin, castor oil (3%)
*Separated by gravity into two layers, 90% clear and 10% cream, turbidity is of clear phase only

As all the formulations were made by simple atmospheric method so it was possible to try and rule out many different formulations in a relatively shorter time because the atmospheric method is relatively simple and fast (Hagenmaier, 2004) and it became even easier because neither of the formulations contained ammonia that could pose difficulty in preparation by its vaporization. All of the emulsions were made with the ingredients acceptable by the Code of Federal Regulations and/or EU for use in food and/or fruit coating. Those emulsions which included starch among other ingredients have higher degree of creaming, coalescence and turbidity while the emulsions that did not included starch among their ingredients had lower values for these characteristics (Table 2). This may be due to the poor solubility of starch in alcohol (Anonymous, 2013) as alcohol was mixed in water at the start of manufacturing process. These results showed that starch is not as suitable component for a good ethanol based composite coating as the gum acacia is. Gum acacia was also the least viscous and the most soluble of the hydrocolloids and it produced stable emulsion with most oils. Similarly viscosity and specific gravity were also recorded higher for the emulsions containing starch while lower for the emulsions without starch (Table 2). Higher viscosity and specific gravity are not the desired features for a fruit coating that is meant to be used by spraying method. Also the higher the viscosity and specific gravity, the lower is the coverage area of the fruit coating. The amount of flaked coating for both the coatings increased with the passage of storage time (Fig. 1) but it was lower for the ESCM though it was non-significant.

It was previously found that with minimal plasticizer it may be possible to find a suitable compromise between optimum appearance and shine on one hand, and optimum flavor on the other hand (Hagenmaier, 2004). But the results of present study suggest that it is possible to formulate good fruit coatings without the addition of any particular plasticizer.

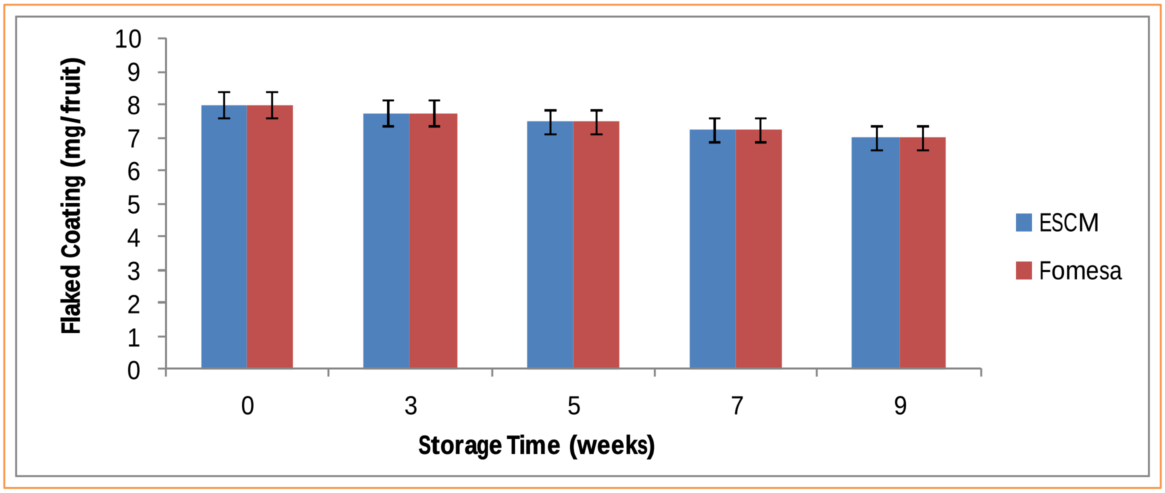

Figure 1. Flaking of coatings on Blanco mandarins, weight of removed coatings

Performance

The control of weight loss is important in that most fresh produce is sold by weight. A coating that will reduce the amount of weight loss will also reduce the amount of overfill required (Khout et al., 2007). The newly developed ESCM successfully reduced the weight loss percentage not only in mandarins where it was at par with the synthetic wax (Fig. 2) but also in cucumber (Fig. 3) and bell peppers (Fig. 4) where it was significantly different from the uncoated samples.

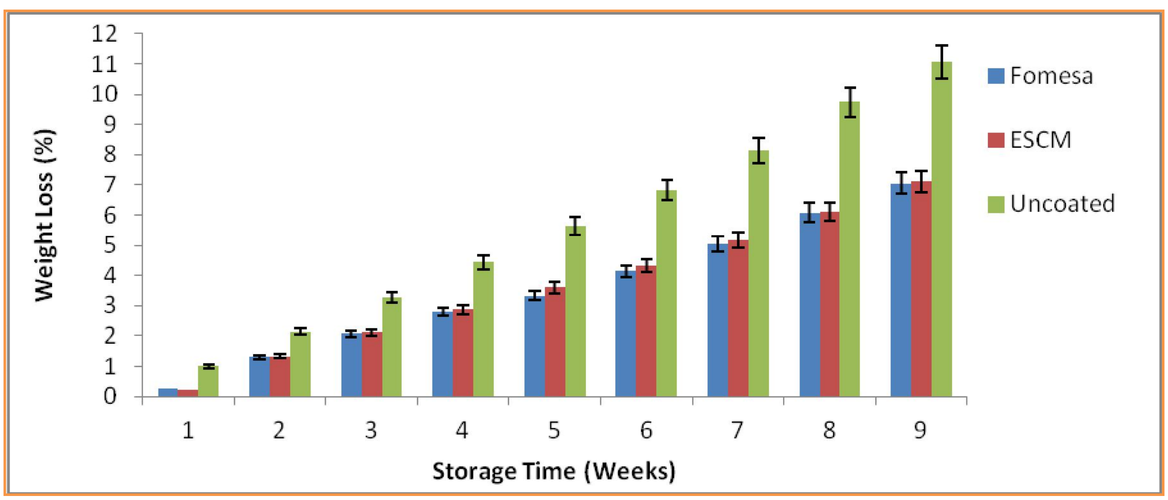

Figure 2. Effect of different treatments on weight loss of mandarins ($p < 0.05$) stored at 5±2°C, 85-90% relative humidity

Coatings enhance the reduction of moisture loss by enhancing the natural barrier, if already present, or replacing it in cases where washing and handling has partially removed or altered it. And also the hydrophobic components such as oils and resins (similar to cutin) emphasize the reduction of moisture loss in coated samples (Baldwin, 1994). Present findings are supported by the earlier results of Arif et al., (2013), and Zulfiqar et al., (2013) who found significant reduction in weight loss of grapefruits and cucumbers coated with an oil based edible skin coating material as compared to the uncoated ones.

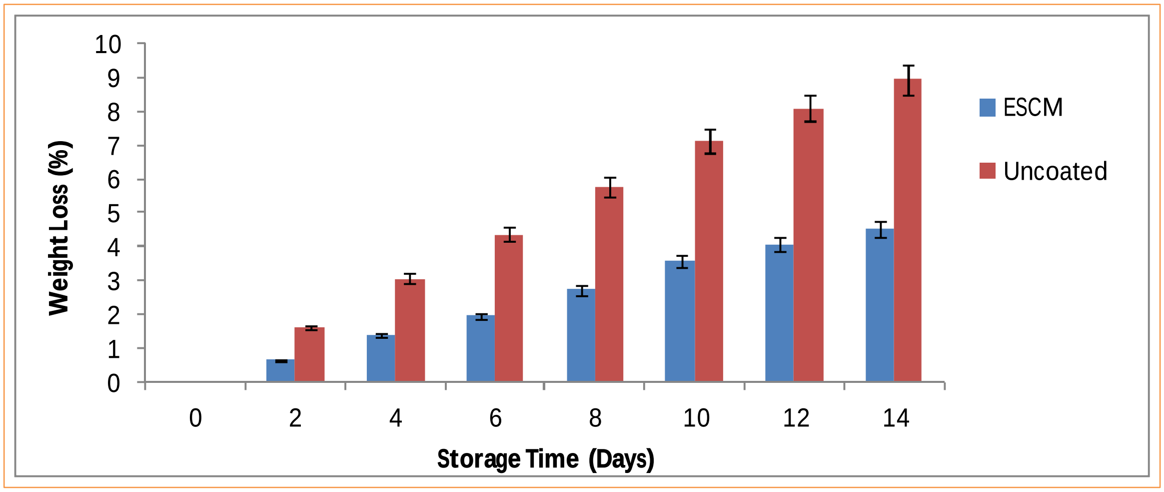

Figure 3. Effect of non ionic-anionic ESCM on the weight loss of cucumbers stored at low temperature

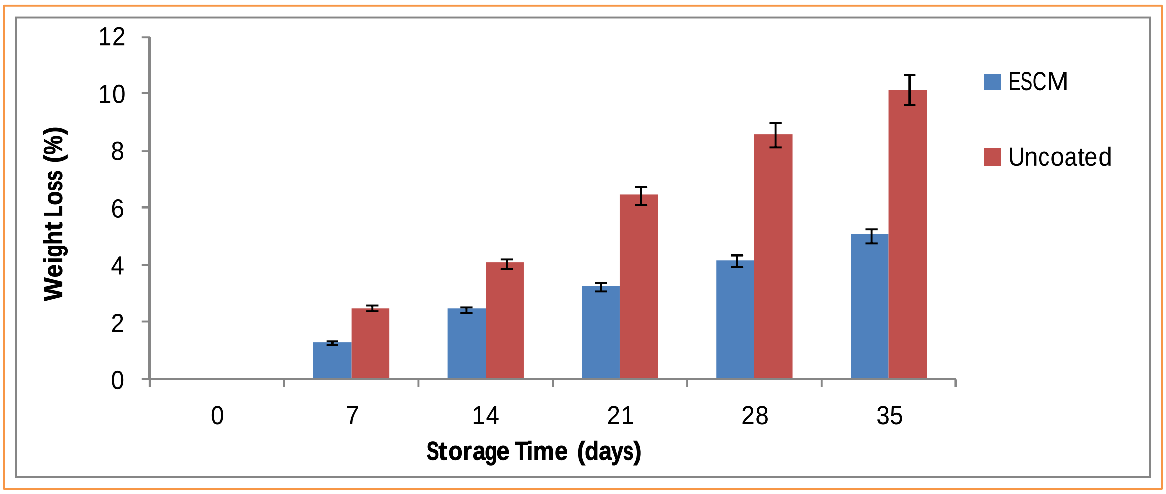

Figure 4. Effect of non ionic-anionic ESCM coating on weight loss of bell peppers ($p< 0.05$) stored at 10±2°C, 85-90% relative humidity

Firmness of fruits and vegetables is important not only for their shipping but also for their eating and cooking quality. Firmness of mandarins coated by ESCM was at par with that of synthetic (Fomesa) coated mandarins while it was significantly different from that of uncoated ones (Figure 5). Firmness is thought to be affected by two factors; it can be a function of cell wall turgor which is directly affected by moisture loss through the skin (Olivas and Cánovas 2009), and/or the loss of firmness is a biochemical process that involves the hydrolysis of pectin and starch by enzymes e.g. wall hydrolases (Yaman & Bayoindirli, 2002). Coatings act as a semi permeable barrier to moisture loss, thereby restricting water transfer (Ribeiro et al., 2007), which helped in maintaining the turgor pressure of the cells and in turn the firmness.

While the coating also resulted in the reduction of O_2 level and increase in the CO_2 level (the modified atmosphere) by acting as a semi-permeable membrane (Baldwin, 1994) that resulted in the maintenance of firmness during storage. Similar results were obtained by Arif et al. (2013) who found that coating of grapefruits with an oil based edible skin coating material maintained their firmness. Vitamin C contents of the mandarin for all the treatments decreased as a function of storage (Figure

6) but this decrease was statistically non-significant between the coated mandarins while significant when compared to the uncoated mandarins. Vitamin C is readily oxidized by oxygen when exposed to it (Hussain et al., 2006).

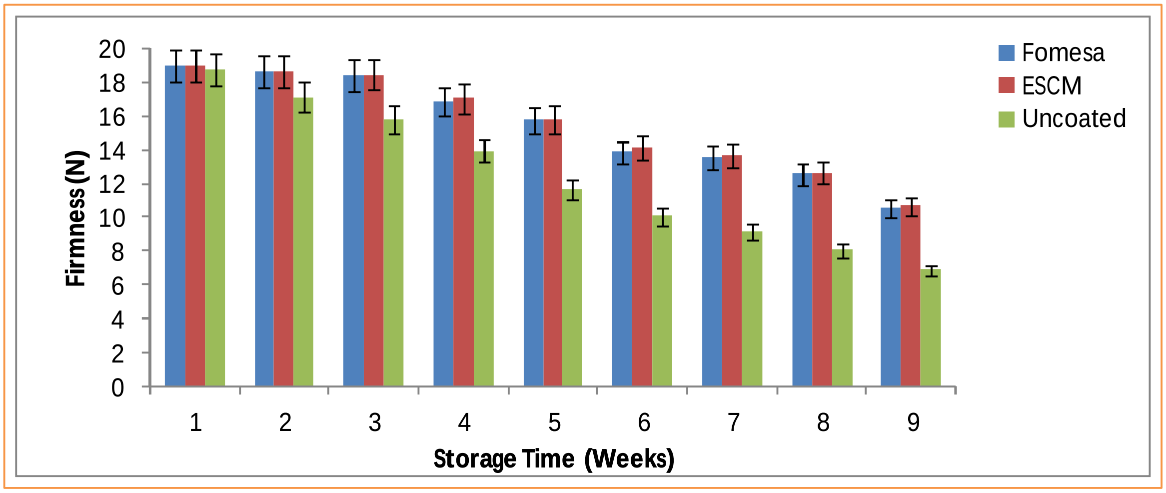

Figure 5. Effect of different treatments on firmness of mandarins ($p< 0.05$) stored at 5±2^{0}C, 85-90% RH

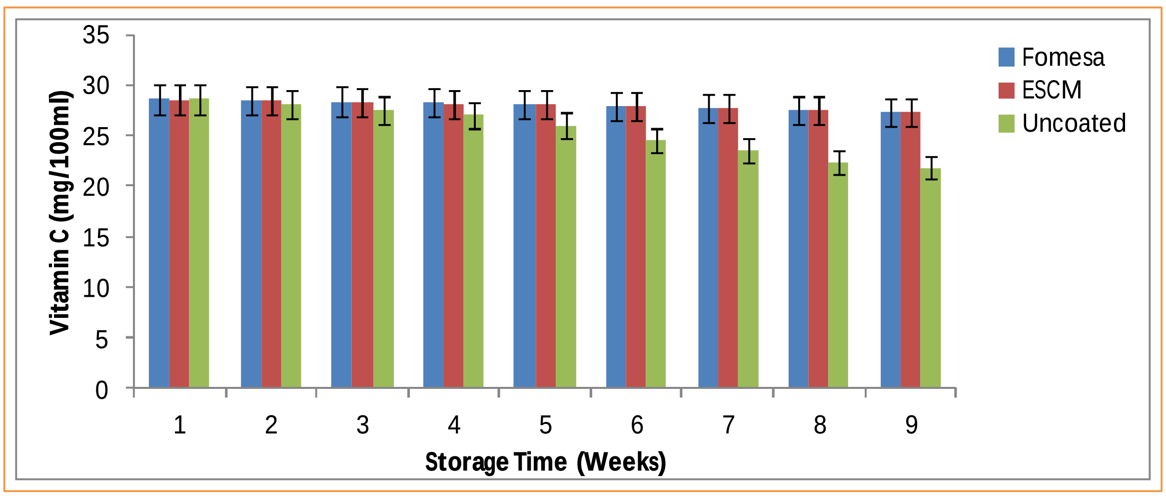

Figure 6. Effect of different treatments on vitamin C contents of mandarins stored at 5±2°C, 85-90% relative humidity for 9 weeks

Creation of modified atmosphere by the coatings limits the exchange of gases that reduces the amount of oxygen reaching to the interior of fruit thus preventing the oxidation of vitamin C (Baldwin et al., 1995). Both of the coatings decreased the oxidation of vitamin C alike. These results are at par with the previous results of studies which found that vitamin C contents of Murcott fruits decreased during storage at low temperature (Yu et al., 2011) and that the vitamin C contents were found higher for coated mandarins than the uncoated ones during storage at low temperature (Mahajan et al., 2005).

The coatings are applied to fruits and vegetables to enhance their outlook and appearance, and in that way improve consumer preference for coated products over the uncoated ones. The gloss imparted by both of the coatings was good but it decreased with the passage of storage time (Figure 7). By comparison, the mean gloss of Fomesa and ESCM were 7.95 and 8 respectively at the start of the study, not significantly different from each other. While the mean gloss of Fomesa and ESCM was 7 for both at the end of storage period, indicating that the newly developed ESCM from natural ingredients is at par with the noticeable market brand in terms of gloss imparted to fruit and that it can also fairly maintain the gloss over prolonged storage period.

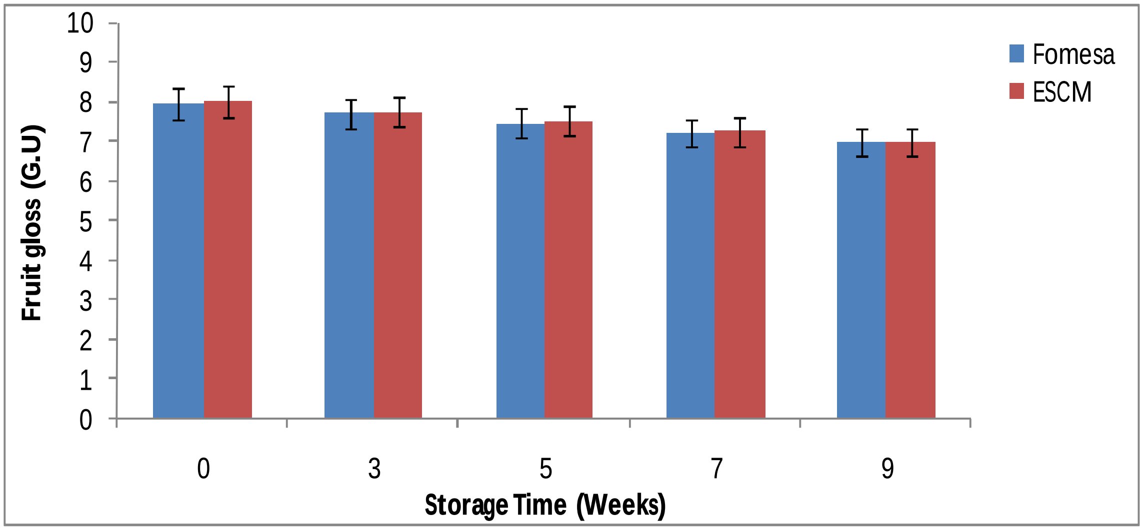

Figure 7. Effect of different coatings on gloss of mandarins stored at 5±2^{0}C, 85-90% RH for 9 weeks

Bajwa and Anjum (2007) have previously reported that polyethylene based wax gives more shine than shellac based wax, but the results of present study suggest that it is possible to get the same shine as of polyethylene based synthetic wax from a coating prepared from natural materials only. Perhaps the hydrophobic components (oils, resins) of the fruit coatings did not allow the water to be accumulated on the surface as opposed to hydrophilic components (polysaccharides, proteins), thus maintaining the gloss over prolonged storage period. Both ammonia and ethanol, the most volatile components of Fomesa and ESCM respectively, readily vaporize at the temperatures which are maintained during different processing operations in grading & packing factories, so that the coatings get readily dried. On vaporization, the volatiles can produce an unfriendly environment for the workers of the factory. The Hedonic scale ratings for the worker friendly aspects for both of the fruit coatings clearly revealed that the newly developed ESCM is readily liked by the workers of all factories as opposed to the synthetic wax (Fomesa), which is not liked by the workers owing to the pungent smell of ammonia, an integral part of Fomesa (Table 3).

Table 3. Mean values for the worker friendly aspects of Fomesa and ESCM by the personals of four factories

Worker	Parameter			
	Smell		Irritation	
	Fomesa	ESCM	Fomesa	ESCM
Foreman	4^c	8^e	4^c	8^e
Sorter	2^a	7^d	2^a	7^d
Operator	2^a	7^d	2^a	7^d
Packer	3^b	8^e	3^b	8^e

The Hedonic scale ratings of ESCM were significantly different from those of Fomesa, statistically. One important function of the coatings is to improve and maintain the sensory qualities of the fruit being coated. Similarly, a decreasing pattern was observed for all the sensory qualities for all the treatments but the non-ionic anionic ESCM fairly maintained the sensory qualities of the mandarins, which were significantly superior to the uncoated mandarins while these were at par with the Fomesa coated mandarins (Table 4).

Table 4. Scores for the effect of different treatments on the sensory qualities of mandarins stored at 5±2°C, 85-90% relative humidity for 9 weeks

Attribute	Treatment	Storage Time (days)									
		0	7	14	21	28	35	42	49	56	63
Visual Appearance	Uncoated	8.00^{a}	8.00^{a}	8.00^{a}	8.00^{a}	8.00^{a}	7.75^{b}	7.75^{b}	7.50^{c}	7.25^{d}	7.25^{d}
	ESCM	8.25^{a}	8.25^{a}	8.25^{a}	8.25^{a}	8.25^{a}	8.15^{a}	8.15^{a}	8.05^{a}	7.95^{b}	7.95^{b}
	Fomesa	8.25^{a}	8.25^{a}	8.25^{a}	8.25^{a}	8.25^{a}	8.15^{a}	8.10^{a}	8.10^{a}	8.00^{a}	7.95^{b}
Color	Uncoated	8.00^{a}	8.00^{a}	8.00^{a}	8.00^{a}	8.00^{a}	7.90^{b}	7.85^{b}	7.70^{c}	7.70^{c}	7.70^{c}
	ESCM	8.10^{a}	8.10^{a}	8.00^{a}	8.00^{a}	8.00^{a}	7.90^{b}	7.85^{b}	7.75^{b}	7.75^{b}	7.75^{b}
	Fomesa	8.10^{a}	8.10^{a}	8.00^{a}	8.00^{a}	8.00^{a}	7.90^{b}	7.85^{b}	7.80^{c}	7.75^{c}	7.75^{c}
Flavor	Uncoated	8.00^{a}	8.00^{a}	8.00^{a}	8.00^{a}	7.90^{b}	7.85^{b}	7.75^{c}	7.70^{d}	7.70^{d}	7.65^{d}
	ESCM	8.00^{a}	8.00^{a}	8.00^{a}	8.00^{a}	7.90^{b}	7.85^{b}	7.75^{c}	7.70^{d}	7.70^{d}	7.65^{d}
	Fomesa	8.00^{a}	8.00^{a}	8.00^{a}	8.00^{a}	7.90^{b}	7.85^{b}	7.80^{c}	7.75^{c}	7.70^{d}	7.65^{d}
Overall Acceptability	Uncoated	8.00^{a}	8.10^{a}	8.10^{a}	8.10^{a}	8.00^{b}	7.95^{b}	7.90^{c}	7.80^{d}	7.80^{d}	7.80^{d}
	ESCM	8.10^{a}	8.10^{a}	8.10^{a}	8.10^{a}	8.00^{b}	7.95^{b}	7.90^{c}	7.80^{d}	7.80^{d}	7.80^{d}
	Fomesa	8.10^{a}	8.10^{a}	8.10^{a}	8.10^{a}	8.00^{b}	7.95^{b}	7.90^{c}	7.90^{c}	7.80^{d}	7.80^{d}

(Values sharing the same letter in columns of specific parameter are not significantly different)

Respiration patterns of different mandarins can affect indirectly the sensory attributes like color and flavor as higher rates of weight loss under the ambient conditions decreased the color scores (Hagenmaier, 2002). Both of the coatings decreased the rate of weight loss in the current study thus minimizing the negative changes on the sensory qualities. Contreras-Oliva et al. (2011) also found similar results, which say that sensory qualities were not affected by the edible coating of 'Valencia' oranges.

CONCLUSION

The edible skin coating material (ESCM) prepared from natural ingredients was equally effective in fairly maintaining the postharvest quality of fruits (mandarins) as well as vegetables (bell-pepper, cucumber). It can safely be a good alternative to the synthetic wax. The ethanol based ESCM was more liked as compared to the ammonia based synthetic wax by the workers of the citrus factories regarding worker friendly aspects. The results of present study also suggested that ammonia or morpholine are not necessary to formulate good fruit coating.

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