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Safe Postharvest Treatments for Controlling *Penicillium* Molds and its Impact Maintaining Navel Orange Fruits Quality

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Abstract: Postharvest diseases caused by *Penicillium digitatum* (green mold) and *Penicillium italicum* (blue mold) are the most important negative factors affecting market local and export of citrus fruits in Egypt. The effectiveness of postharvest treatments of 4% chitosan, 2 % potassium sorbate (PS), or 2% potassium sorbate in wax to control green and blue molds decay and keeping quality were studied on Navel oranges during storage. Intact orange or orange artificially inoculated with *Penicillium digitatum* or *Penicillium italicum*, coated and stored up to 45 days at 5°C and relative humidity (RH) 90-95%. The results indicated that fruit rot and quality characteristics (weight loss and juice percentages, Total soluble solids (TSS), total acidity (TA), firmness, vitamin C content) were affected positively by application of the antifungal coatings. Antifungal coatings significantly reduced incidence and severity of both green (GM) and blue (BM) molds. Coating fruits decreased decay, weight loss and delayed changes loss in the firmness, acidity, ascorbic acid, juice percentage and total soluble solids concentration compared with control fruits. The study suggests that coating might be a promising candidate for controlling decay, maintaining navel orange quality and extending its postharvest marketing life.

Key words: Navel orange · Chitosan · Wax · Potassium sorbate · Quality · Postharvest decay · Green mold · Blue mold

INTRODUCTION

Navel orange (*Citrus sinensis* L. Osbeck) as one of the most important citrus fruits in Egypt, is a subtropical fruit of high commercial value on the international fruit market and is commonly eaten fresh. Under Egyptian conditions, it is a common practice to store mature navel orange fruits until the suitable time for marketing. Injuries sustained to navel orange fruit during harvest allow the entry of wound pathogens, including *Penicillium italicum* causal agents of blue mold and *Penicillium digitatum* causal agents of green mold. These two pathogens are considered to be the most important postharvest diseases and cause serious losses annually [1]. They cause serious problems for the harvested citrus fruits during handling, transportation, exportation and the storage process. Although the use of chemical fungicides gave satisfactory control against mold infection, the fungicide residual can have a harmful effect on human and the environment [2]. Numerous natural safe alternatives have been investigated against different postharvest diseases of citrus fruit such as Chitosan is used as biodegradable film or coating, it has been widely used in the food and cosmetic industries.

Chitosan (poly-β β-(1→4) N-acetyl-d-gluco amine), is a natural antimicrobial compound. On one hand, it can be obtained from crustacean shells (crabs, shrimp and crayfishes) either by chemical or microbiological processes and on the other hand it can be produced by some fungi [3]. The Chitosan coating is safe [4] and shows antifungal activity against several species [5]. Chitosan coating has been used to prolong the shelf life of and control decay in citrus [6, 7], maintained postharvest quality and beneficially influenced firmness, total soluble solid content, titratable acidity, ascorbic acid content and water content of citrus fruit after 56 days of storage at 15°C [8], decreases the respiration rates, inhibits fungal development and delays ripening because

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of a reduction in the evolution of ethylene and carbon dioxide [9]. The use chitosan coating for mandarin was in the range of 1 to 2% [10].

Potassium sorbate (PS) is a wide spectrum antimicrobial food additive with an effect against molds and yeasts, mostly within the pH range of 3-6.5. Bandelin [11], has been tested PS alone or in combination with commercial fungicides against the major postharvest pathogens of citrus fruit and found that PS was not only compatible with these fungicides but also improved their performance against *P. digitatum* green mold. Potassium sorbate solution is applied at room temperature or moderately heated for relatively long immersion times (2-3 min) resulted in relatively effective control of the diseases [12]. Wax (edible coatings) have been used as an effective technology to enhance the overall appearance and quality of the fruit by reducing water loss and preventing shrinkage, reducing internal O_2 levels and increasing internal CO_2 [13]. Coatings could effectively retard the loss of the water, titratable acidity and ascorbic acid of sweet cherries [14], improve firmness, titratable acidity, ascorbic acid and the water content for citrus (Murcott tangor) that stored at 15°C for 56 days [6] and reduce molds caused by *P. digitatum* and *P. italicum*e [15].

The purpose of this research is to investigate the effects of chitosan, wax, potassium sorbate alone or potassium sorbate in wax coating against green and blue rots disease and its role in improvement the quality of navel orange fruits under cold storage condition.

MATERIALS AND METHODS

Plant Material: W ashington navel orange fruit (*Citrus sinensis* L. Osbeek) freshly harvested from a private orchard (Greena) in Wadi Alnatroon, (El-Beheira Governorate). Mature orange fruits undamaged, apparently healthy, similar in shape, weight and color, were harvested at late of December 2010 as well as the second successive season of 2011 at full color stage and the TSS ratio of 8 or higher [16], average weight of 300 gm, total soluble solids content (TSS) (11%), titratable acidity (TA) (0.90%) firmness[13 (lb/in^2)], juice ratio (35 %) and ascorbic acid (V.C.) [48 mg/100 ml juice] were evaluated at harvest time. Fruits transferred to laboratory of Agriculture Development Systems (ADS) project at the Faculty of Agriculture, Cairo University.

Fungi and Culture: One isolate *Penicllium digitatum* (green mold disease incident) and *P. italicum* (blue mold disease incident) obtained from Postharvest Diseases Department, Plant Pathology Research Institute, Agricultural Research Center (ARC), Giza, Egypt were used in present study. All fungi were cultured on Petri-dishes containing sterilized potato dextrose agar medium (PDA) then packed in polypropylene bags and incubated at 25±1°C under 85-90% RH until use.

Preparation of Anti Fungal Fruit Coating Solution: Wax (E-wax) Exported wax which used in citrus packinghouses was obtained from Egyptian Company for Mechanical and Electrical Industries. The composition of the Export wax in the form of water emulsion, posses 22% solids materials was contained (shellac, kalaphonia, polyethylene emulsifier and water). Chitosan was obtained from Sigma Chemicals Company. It was dissolved at 4% (w/v) in 1% HCl by stir-ring overnight at room temperature and the original solution was then diluted to a series of 0.5, 1.0 and 2.0, 4.0% and the pH value of each solution was adjusted to 5.4 with 1 mol/L NaOH. Potassium Sorbate (P.S) 2% (w/v) in deionizer water were used the solution was kept at a natural of 7.8 pH according to previous results [17].

***In vitro* Studies:** Chitosan, Potassium sorbate and wax were evaluated for their capability to suppress fungal growth of *P. digitatum* and *P. italicum* in vitro. Each tested material was amended in PDA medium at concentrations of 0.50, 1.0, 2.0 and 4.0% certain volumes of chitosan, Potassium sorbate and wax taken from stock solution were added individually to different PDA flasks, to achieve the proposed concentration, just before solidifying, then poured into sterilized Petri- dishes. The stock solutions were prepared using sterilized water and tween 80 as emulsifier agent at the rate of 0.05%. Treated media of chitosan and Potassium sorbate were poured into 3 Petri dishes as replicates per each tested concentration. The control plates contained only tween 80 at 0.05%. After medium solidification; Petri dishes were inoculated individually at the centre with fungal inoculums discs (5 mm) of 7 days -old culture of *P. digitatum* and *P. italicum.* All plates were in incubated at 25±1°C until the mycelium of control treatment cover the medium surface in the Petri dish. Growth inhibition was calculated as the percentage of inhibition of the radial growth in relative to the control. After incubation period, the linear growth (mm) of each fungus was recorded and reduction percentages in colonies diameters caused by the tested treatments were calculated using the formula suggested by Fokemma [18] as follows:

$$\text{Reduction percentage} = \frac{(de - di)}{de} \times 100$$

Where:

de = Linear growth in control set.
di = Linear growth in treatment set.

Fruit Inoculation: Seven days -old cultures of *P. digitatum* and *P. italicum* were used to obtain a spore suspension in sterile distilled water containing a drop of a wetting agent (Tween 80) per liter. Spore suspensions were adjusted to 5 x10^6 spores/ml. Fruits were sterilized with 70% ethanol, then artificially infected by wounding the flavedo once on the equator of each fruit with a steel rod (2mm deep by 0.5 mm wide). Fruits were divided to 3 groups prior to treatments, each group containing 450 fruits. The first group as naturally infected fruits, the second inoculated with *P. digitatum* and the third group inoculated with *P. italicum* (Artificial infection). The inoculation was previously prepared carried out by spraying the surface of fruits with previously prepared spore suspensions. Inoculated fruits were kept at room temperature and allowed to air dry for 24 hour before antifungal application treatments.

Antifungal Treatments and Fruits Storage: Orange fruits of three groups, naturally infected fruits, artificially inoculated with each of *P. digitatum* and *P. italicum*, were treated with the following 5 treatments; each treatment consisted of 90 fruits. Treatments:

- Wax
- Chitosan 2% (w/v)
- Potassium sorbate 2 %, 3 min dipping in 2% (w/v) solution (at 21°C, pH of 7.8)
- Potassium sorbate 2 % (w/v).Then or + in wax (3 min dipping in solution at 21°C)

Control Dipping in Distilled Water: Coated fruits were then dried with an electric fan at room temperature (20±2°C) for 30 min. Ninety coated fruits of each treatment were placed into three perforated cartoon boxes each (30×40×20 cm), as box for determining decay, the second for determining weight loss and the third for fruits analysis, each box replicated three times, each of 10 fruits. This experiment was repeated twice (2010 and 2011seasons). Orange fruits were stored at 5°C and relative humidity 90-95% for 45days in the cooled storage room of Agriculture Development Systems (ADS) project, Faculty of Agriculture, Cairo University. The quality measurements were determined at the end of cold storage 45 days for naturally infected fruits, inoculated with *P. digitatum* and inoculated with *P. italicum* (Artificial infection) under different treatments. Disease incidence of the fruit and lesion diameter on the each fruit was recorded. When the visible rot zone out-sides the wounded area on fruit was more than 0.5 mm wide, it was counted as decayed fruit. Disease incidence was measured according to the method of Zeng *et al.* [19]. Severity of infection was estimated as percentage of the external rotten area in proportion to the total area of the fruit [20]. Efficiency percentage measured according to the equation:

$$\%\,\text{Efficiency} = \frac{\text{Control disease severity - Treatment disease severity}}{\text{Control disease severity}} \times 100$$

Disease severity and efficiency% were determined at the end of cold storage 45 days and after 5 days from the end of storage at ambient temperature (20°C and 65% RH) as a marketing life.

The results of the previous experiments were analyzed using the software CoStat version 6.400- CoHort Software. The mean of all treatments were compared by the least significant difference (L.S.D.) at 5% level of probability according to Oehlert [21].

Fruit Quality Assessments

Weight Loss: Fruits were periodically weighed and the loss in mass weight was recorded for each replicate. Data were calculated as percentage.

Fruit Firmness: IT was measured on the two opposite sides of Navel orange fruit samples (6 fruits) by using a hand Magness Taylor pressure tester (lb/in^2).

Total Soluble Solids (TSS): Flesh of individual navel orange fruits were ground in an electric juice extractor for freshly prepared juice. Soluble solids content was measured using a Digital refractometer Instrone (Model PR32 Brix-readings 0-32 ranges Atago Palete ATago. CO. LTD. Japan.

Titratable Acidity (TA): Total acidity (expressed as citric acid %) was determined by titrating 5-ml juice with 0.1N sodium hydroxide using phenolphthalein as indicator [22].

Juice Ratio: Fruit weighed with an electronic balance. Juice was extracted by machine and weight was taken. The juice percentage was expressed as percentage of total weight at the time of measurement. Weight juice/weight fruit x 100 [22].

Vitamin C (Ascorbic Acid): Its content was measured using 2, 5-6 dichlorophenol indophenols' method described by A.O.A.C. [22].

The design for fruit quality experiment was completely randomized factorial design with three replications. Data were analyzed with the Analysis of Variance (ANOVA) procedure of MSTAT-C program. When significant differences were detected, treatment means were compared by LSD range test at the 5% level of probability in the two investigated seasons [23].

RESULTS AND DISCUSSION

Effect of Safe Treatments Chitosan, Potassium Sorbate and Wax on Linear Growth of P. *digitatum* and *P. italicum in vitro*: The inhibitory effect of chitosan, potassium sorbate and wax on the linear growth of tested fungi *P. digitatum* and *P. italicum* were evaluated under *in vitro* conditions. Results expressed as reduction in fungal linear growth are presented in Table 1. Potassium sorbate showed the best effectiveness, where it reduced completely the mycelial growth of *P. digitatum* and *P. italicum* at tested concentrations (0.5, 1, 2 and 4%). Meanwhile, chitosan had less effectiveness, but the inhibition was realed to its concentrations, where it reduced the mycelial growth of *P. digitatum* and *P. italicum* by 100% with 4%concentration.On the other hand, wax did not affect the mycelial growth all the tested concentrations. The obtained results are in accordance with those reported by El-Mougy *et al.* [24]. Therefore sorbate and chitosan may help in control *P. digitatum* and *P. italicum* infection on navel oranges fruits *in vivo*.

Data presented in Table 2 showed that, all treatments significantly suppressed the decay development on naturally infected navel oranges, during cold storage (2010-2011 seasons). It was observed that the efficacy of most treatments was almost constant along the two seasons results, in spite of different infection percentages were obtained. This finding is attributed to the different percentages of infection of the control fruits. Tested treatments proved their higher efficiency to control postharvest fruit rots on navel oranges more than untreated fruits and those treated with wax only. Similar results were revealed by many investigators on different fruits, where potassium sorbate and chitosan were found to be effective against *Penicillium* spp. [6, 24].

Chitosan, potassium sorbate, potassium sorbate in wax and only wax were tested for their curative activity protective effect against citrus fruit molds. Results in Table 3 and 4 indicated that all treated fruits showed complete reduction in molds incidence. It is clear that efficacy of fruit coating with chitosan, potassium sorbate, potassium sorbate in wax were able to highly curative both molds pathogens on navel orange fruits under stress of artificial inoculation for over one month, (the period of experimental test). The present investigation showed that applying chitosan as fruit coating obviously gave complete protection for citrus fruits against mold infection, even under artificial infection with the pathogenic fungi under 45 days storage (Tables 3 and 4). Coating fruits with chitosan decreased postharvest decay caused by fungal infection of citrus [6]. Chitosan, a by- product from the seafood industry, is a safe material as specified by toxicological studies [4]. The mode of action of chitosan was explained by Du, *et al.* [9] since coating of fruit significantly reduced the respiration rate, ethylene production and interval O_2 level of peach, pear and kiwi fruits. The present findings demonstrated that potassium sorbate either alone or in wax as carrier have the potential to be environmentally compatible, nontoxic postharvest fungicides to be used against citrus mold incidence of stored navel orange fruits. Also, Sofos and Busta [25] found that potassium sorbate (PS) is the best characterized of all food antimicrobials as to their spectrum of action. It inhibits certain bacteria and food-related yeasts and mold species. PS was used commercially to retard citrus postharvest decay, but its use did not become popular because its efficacy was sometimes low and it was reported to delay, rather than stop green mold infections in some reports [17].

Fruit Quality Parameters

Weight Loss Percent: Weight loss is mainly associated with respiration and evaporated through the skin. Results presented in Table 5 indicated that with ending of storage period 45 days at 5°C, weight loss of Navel orange was increased. The average weight loss percentage significantly reduced in all coated fruits in both studied seasons. The minimum decrease percent of fruit weight loss was observed in PS + wax followed by PS alone, chitosan then wax treated fruits. The maximum weight loss was found in control treatment. Also, statistical analysis of results indicated that naturally infected oranges significant decrease percent of fruit weight loss compared with inoculated fruits with *P. italicum* and *P. digitatum*. The interaction between treatments and inoculation significantly caused decrease in weight loss in all treated fruits compared with the control specially PS + wax as it much inhibited fruit water decrease from skin surface and more affected on infected fruits with *P. italicum*

Table 1: Effect of safe postharvest treatments on linear growth of *P. digitatum* and *P. italicum in vitro*

		Fungal growth			
Treatments	Concentration	*P. italicum*	% Reduction	*P. digitatum*	% Reduction
Chitosan	0.5	34.17*	62.04	32.33*	64.07
	1	19.67	78.15	18.50	79.44
	2	15.00	83.33	14.17	84.26
	4	0.00	100.00	0.00	100.00
Potassium Sorbate	0.5	5.00	94.44	5.00	94.44
	1	0.00	100.00	0.00	100.00
	2	0.00	100.00	0.00	100.00
	4	0.00	100.00	0.00	100.00
Wax	0.5	90	0.0	90	0.0
	1	90	0.0	90	0.0
	2	90	0.0	90	0.0
	4	90	0.0	90	0.0
Control		90.00	0.00	90.00	0.00
LSD 0.05		4.090	--	1.802	--

*Colony diameter (mm)

Table 2: Effect of safe postharvest treatments on natural infection orange fruit molds at the end of storage and marketing life during 2010 and 2011 seasons

	Natural infection							
	Season 2010				Season2011			
	End storage		Marketing life		End storage		Marketing life	
Treatments	DS*	E**	DS*	E**	DS*	E**	DS*	E**
Chitosan	0.0	100	00	100	0.0	0.0	00	100
Potassium sorbate	0.0	100	00	100	0.0	0.0	00	100
PS+ wax	00	100	00	100	0.0	0.0	00	100
Wax	1.33	79.06	8.33	67.44	0.86	85.50	8.07	67.20
Control	6.35	-	25.58	-	5.93	-	24.60	-
LSD at 0.05	2.657		5.547		1.664		5.794	

*DS = % Disease Severity

**E = % Efficiency

Table 3: Effect of safe postharvest treatments for controlling blue mold on navel orange fruits (2010-2011seasons)

	Penicillium italicum							
	Season 2010				Season 2011			
	End storage		Marketing life		End storage		Marketing life	
Treatments	DS*	E**	DS*	E**	DS*	E**	DS*	E**
Chitosan	0.66	97.18	3.81	95.17	0.28	99.10	3.60	95.53
Potassium sorbate	0.35	98.50	2.48	96.86	0.42	98.65	3.17	96.07
PS+ wax	0.16	99.32	2.36	97.01	0.14	99.55	1.80	97.77
Wax	17.66	47.09	62.77	79.23	12.15	61.00	22.33	72.30
Control	33.38	-	78.96	-	31.15	-	80.62	-
LSD at 0.05	3.138		7.069		7.930		11.471	

*DS = % Disease Severity

**E = % Efficiency

Table 4: Effect of safe postharvest treatments for controlling green mold on navel orange fruits (2010-2011 seasons).

	Penicillium digitatum							
	Season 2010				Season 2011			
	End storage		Marketing life		End storage		Marketing life	
Treatments	DS*	E**	DS*	E**	DS*	E**	DS*	E**
Chitosan	0.35	98.32	4.71	94.83	0.12	99.42	2.75	96.96
Potassium sorbate	0.26	98.75	2.73	97.00	0.07	99.66	1.52	98.32
PS+ wax	0.13	99.38	1.93	97.88	0.07	99.66	1.49	98.35
Wax	14.33	31.30	19.49	78.59	19.33	5.80	28.83	68.16
Control	20.86	-	91.04	-	20.52	-	90.55	-
LSD at 0.05	7.636		11.613		2.23		8.62	

*DS = % Disease Severity

**E = % Efficiency

Table 5: Effect of safe coating on weight loss and firmness of natural infected (N.I) Navel orange fruits, inoculated with *P. italicum* (P.I) and *P. digitaum* (P.D) at the end of cold storage during 2010 and 2011 seasons

	Weight loss %								Firmness (lb/in^2)							
Inoculation	2010				2011				2010				2011			
Treatments	N.I	P.I	P.D	Mean	N.A	P.I	P.D	Mean	N.A	P.I	P.D	Mean	N.A	P.I	P.D	Mean
Chitosan	1.40	2.70	3.10	2.41	1.60	3.0	3.20	2.70	12.0	11.0	10.2	11.0	11.5	10.8	10.1	10.8
Potassium Sorbate	1.30	2.0	2.70	2.0	1.40	2.50	2.90	2.26	12.6	11.3	10.4	11.4	12.3	11.0	10.2	11.1
PS+ wax	0.70	1.40	1.60	1.23	0.85	1.60	1.90	1.45	13.2	11.6	11.0	11.9	13.0	11.4	11.0	11.8
Wax	1.90	3.0	3.30	2.73	2.10	3.10	3.50	2.90	11.7	10.3	10.0	10.6	11.2	10.1	9.8	10.3
Control	5.80	7.80	8.0	7.20	6.0	8.30	8.50	7.60	10.0	9.5	9.0	9.5	9.5	9.0	8.6	9.0
Mean	2.22	3.38	3.74	--	2.53	3.90	4.20	--	11.9	10.7	10.1	--	11.5	10.3	9.9	--
L.S.D at 0.05%																
Treatments (A)	1.405				1.660				1.885				1.009			
Inoculation (B)	0.206				0.230				0.624				0.109			
A*B	0.357				0.398				1.862				0.159			

(blue mold) and *P. digitatum* (green mold). Weight loss of perishables is a serious concern in storage because loss of moisture decreases visual quality. This result confirms the findings of Hagenmaier and Shaw [26] who reported that waxing citrus fruit after harvest in order to enhance their shine and to reduce weight loss and shrinkage. Also, Galed *et al.* [7] established that chitosan on the mandarins and oranges produced excellent results of weight loss percentage and visual appearance. Coating act as barriers, there by restricting water transfer and protecting fruit skin from mechanical injuries, as well as sealing small wounds and thus delaying dehydration [27].

Firmness: At the end of 45 days storage, firmness of Navel orange fruits were decreased. Potassium sorbate in wax treatment maintained highest fruit firmness level followed by potassium sorbate (PS), chitosan and wax each alone in both seasons (Table 5). There was significant difference in fruit firmness among all the treatments compared with control. The highest firmness was achieved by naturally infected fruits compared with inoculated fruits with blue and green molds. As for the interaction, data referred that all treatments significantly increased this parameter specially PS + wax and PS treatments as they were the most effective on inoculated fruits with both *Pencilium* spp. for increasing fruit firmness compared with infected control. Loss of firmness is one of the main factors limiting quality and the postharvest shelf-life of fruits. Present findings were consistent with that of Chien *et al.* [6] who reported that, wax enhances and improve s firmness in citrus (*Murcott tangor*). Low molecular weight chitosan coating beneficially influenced firmness of citrus stored 56 days at 15°C [8]. Coating with potassium sorbate in wax or sorbate alone may protect fruit skin from decay and thus delaying dehydration and improve quality.

Table 6: Effect of safe coating on T.S.S and Juice ratio of natural infected (N.I) Navel orange fruits, inoculated with *P. italicum* (P.I) and *P. digitaum* (P.D) at the end of cold storage during 2010 and 2011 seasons

Inoculation	T.S.S %								Juice ratio %							
	2010				2011				2010				2011			
Treatments	N.I	P.I	P.D	Mean	N.A	P.I	P.D	Mean	N.A	P.I	P.D	Mean	N.A	P.I	P.D	Mean
Chitosan	12.3	13.5	13.7	13.1	12.6	13.6	13.8	13.3	33.6	32.3	32.0	32.6	33.3	31.7	31.5	32.1
Potassium sorbate	11.7	12.5	12.6	12.2	12.0	12.8	13.0	12.6	34.0	32.8	32.5	33.0	33.6	32.4	32.2	32.7
PS + wax	11.4	11.9	12.2	11.8	11.6	12.3	12.6	12.1	34.5	33.4	33.2	33.7	34.0	33.2	33.0	33.4
Wax	12.7	13.4	13.6	13.2	13.0	13.8	14.2	13.6	33.3	31.6	31.3	32.0	33.0	31.2	31.0	31.7
Control	13.0	14.3	14.5	13.9	13.5	14.6	14.9	14.3	32.5	29.6	29.4	30.5	31.2	26.8	26.3	28.1
Mean	12.2	13.1	13.3	--	12.5	13.4	13.7	--	33.5	31.9	31.6	--	33.0	31.1	30.8	--
L.S.D at 0.05%																
Treatments (A)	1.198				1.196				0.673				0.193			
Inoculation (B)	0.219				0.130				0.093				0.168			
A*B	0.380				0.226				1.166				0.291			

Total Soluble Solids (TSS %): It is clear from Table 6 that total soluble solids content of Navel orange fruits were increased at the end of 45 days cold storage in both seasons. However, it was noticed that, the changes in TSS content were more slowly in coating treatment than in control. The PS + wax, PS alone, chitosan and wax treatments significantly maintained levels of the juice TSS% without high changes compared with those of the control as highest significant juice TSS %. Naturally infected fruit attained lower TSS content than inoculated fruits. Interaction results show that, inoculated fruits by blue and green mold were more affected by PS +wax treatment in maintaining TSS% without noticeable changes. TSS percentage is a function of total dissolved solids and moisture content of the fruit, the increase in TSS may be due to faster maturation process and higher water loss of the fruit. It may explain the relatively smaller increase in coating treatments which also showed the least weight loss. The coating treatments had higher effect in maintaining of TSS% without high changes at the end of storage. This was probably due to the permeable semi coating film that may have formed on the surface of the fruits and barrier to moisture loss thus delaying dehydration and improves quality. The results obtained in this study can be explain due to the considerable loss of water experienced by orange fruits especially with the uncoated fruit (control treatment). Our results are in line with those of Barman *et al.* [28] who found that carnauba wax and putrescine of pomegranate had lower SSC compared to untreated fruits. Also, Zhang *et al.* [8] who reported that a slow rise in SSC was recorded in citrus fruit treated with chitosan.

Juice Ratio %: The effects of the postharvest treatments on orange juice content were found to be statistically significant ($p<0.05$). At the end of the 45 days storage period, it was clear that the Juice content of Navel orange fruits were decreased in both seasons (Table 6). However, decline was much higher in control. Meanwhile, coating treatments gave effective results by maintaining higher juice recovery over control. The highest juice content obtained from the PS + wax followed by either PS alone, chitosan or Wax treated fruits compared with control. Also, the maximum averages fruit juice% was achieved by naturally fruit (non-inoculated fruits) with significant differences in decayed orange fruits with both *P. italicum* and *P. digitatum*. Interaction data clarify that, in general all treatments maintained juice ratio in inculcated and non-inoculated fruits. Our results in respect of coated orange fruits, the increase in juice recovery after 45 days of storage might be due to the effect of coating which helped in maintaining higher juice recovery by reducing moisture loss from the fruits. These results are confirmed by Barman *et al.* [28] who reported that carnauba wax + putrescine treated pomegranate fruits had higher juice recovery after 30 days of storage. Waxing could improve the water content for Murcott tangor stored at 15°C for 56 days [6]. Chitosan coatings have been barrier to moisture loss and therefore retarding dehydration of papaya fruits [29]. Coating with potassium sorbate showed complete reduction in mold incidence that may protect fruit skin from decay and thus delaying dehydration and improve quality.

Titratable Acidity %: The quantity of organic acids expressed as citric acid that decreased in orange fruits with increasing fruit maintenance period in cold storage (45 days). The control had a significant decrease in TA % and all treatments inhibited the decline of TA specially sorbate in wax or sorbate alone treatments which significant had higher ability in maintaining low decrease in the TA content of fruits compared with control.

Table 7: Effect of safe coating on acidity and Vitamin C of natural infected (N.I) Navel orange fruits, inoculated with *P. italicum* (P.I) and *P. digitaum* (P.D) at the end of cold storage during 2010 and 2011seasons

	Acidity %								Vitamin C (mg/100ml juice)							
Inoculation	2010				2011				2010				2011			
Treatments	N.I	P.I	N.I	P.I	N.I	P.I	N.I	P.I	N.I	P.I	N.I	P.I	N.I	P.I	N.I	P.I
Chitosan	0.82	0.74	0.72	0.76	0.80	0.71	0.68	0.73	46.3	44.8	44.3	45.00	45.8	44.5	44.0	44.7
Potassium Sorbate	0.85	0.75	0.72	0.77	0.83	0.73	0.70	0.75	46.8	45.3	44.8	45.6	46.0	45.0	44.3	45.1
PS+ wax	0.87	0.79	0.77	0.81	0.85	0.78	0.75	0.79	47.2	46.0	45.4	46.2	46.5	45.5	45.0	45.6
Wax	0.80	0.72	0.68	0.73	0.75	0.70	0.65	0.70	45.6	44.0	43.5	44.3	44.5	42.6	42.0	43.0
Control	0.75	0.66	0.62	0.68	0.72	0.60	0.60	0.63	44.3	42.3	42.0	42.7	43.2	41.4	41.0	41.9
Mean	0.82	0.73	0.70	--	0.79	0.70	0.67	--	45. 9	44.4	44.0		45. 2	43.8	43.2	--
L.S.D at 0.05%																
Treatments (A)		0.030				0.030				0.288				0.146		
Inoculation (B)		0.028				0.028				0.246				0.068		
A*B		0.053				0.053				0.499				0.119		

TA content in natural infected fruit was more than inoculated fruits. Interaction data were significant in both considered seasons (Table 7). Therefore using PS+ wax and PS alone, chitosan and wax treatments inhibited respiration of fruits and subsequent decline production of acids and fruits decay compared to control fruits. The obtained results [30] indicated that wax treatment caused an inhibition in TA changes of Valencia and local oranges that stored at 6+1°C for 3 months. Meanwhile, chitosan treatment had higher ability in maintaining the decrease in the TA content of citrus fruit after 56 days of storage at 15°C [8]. Coating with potassium sorbate in wax or sorbate alone were able to highly protect navel orange fruits against both molds incidence and protected fruit skin from decay, thus delaying ripening and improve quality.

Vitamin C: Results presented in Table 7 indicated that with ending of cold storage period (45 days), Vitamin C content in all coating treatments maintained in stored navel orange fruits higher than in control. The minimum decrease of Vitamin C content was observed in PS + wax treatment. Also, naturally infected fruits (non-inoculated) contained higher Vitamin C content than inoculated fruits with blue and green molds. Interaction data show significant differences between various treatments and inoculated fruits, the highest Vitamin C content was obtained from orange fruits treated with PS + wax. As for inoculated fruits with green mold and blue mold, PS + wax were the best treatment compared with the other coating treatments. This study has demonstrated that the coating treatments, delayed the loss of ascorbic acid at the end of stored orange. This may be due to the effect of coating on retarding of ripening and slowing down the metabolism activity and physiological disorders which causing oxidation of ascorbic acid. These results are in agreement with those obtained by Chen and Paull [31] who reported that tissues entering senescence or ripening have low biosynthesis of ascorbic acid. The decrease in ascorbic acid level was associated with reducing capability of preventing oxidative damage and with the incidence of physiological disorders during storage [32]. Wax significantly ceased an inhibition in ascorbic acid reduction of Valencia oranges [14]. Meanwhile, chitosan coating beneficially influenced ascorbic acid content [8] on citrus (Murcott tangor) fruit [33] on navel orange fruits.

CONCLUSION

Coating citrus fruits with chitosan, wax, potassium sorbate or potassium sorbate + wax hold great promise postharvest treatments for controlling blue and green molds of Navel orange fruits and maintenance their quality. They are easily applied tools that could be suitable for packinghouses, in storage and subsequent marketing without using pesticides and might be a promising candidate for maintaining orange quality and extending its postharvest marketing life.

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