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Original Research Paper

Effect of fungicide and essential oils amended wax coating on quality and shelf life of sweet orange (*Citrus sinensis* Osbeck)

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ABSTRACT

Laboratory research was conducted to study the effect of wax amended coating on the shelf life of *Citrus sinensis* Osbeck during 2017-18 at Rampur, Chitwan. The experiment was conducted in single factor Completely Randomized Design (CRD) with nine treatments and four replications. The treatments consisted of carbendazim and three essential oils viz. lemongrass, mentha and eucalyptus oil at two different concentrations of 0.1% and 0.5%, all of them infused with 10% wax emulsion. The wax treatment devoid of fungicide and essential oils served as control. The application of essential oils with wax improved shelf life and enhanced juice retention, firmness, titratable acidity, vitamin C and disease reduction. But total soluble solid was found higher in fruits treated with wax emulsion only. The highest shelf life and disease control was obtained with wax with 0.5% carbendazim but waxing with 0.5% eucalyptus oil and 0.5% lemongrass oil can be better alternatives considering their superior performance in environmental aspects, consumer preferences and quality parameters like juice retention, firmness, titratable acidity and vitamin C.

Keywords : Carbendazim, Eucalyptus oil, Green mold, Lemongrass oil, Post-harvest

INTRODUCTION

Sweet Orange (*Citrus sinensis* Osbeck) is an economically important citrus fruit of the mid hill region of Nepal. The mid hill region of Nepal (1000 to 1500 masl altitude) has a comparative advantage in the production of sweet orange over traditional crops (rice, wheat, maize etc). Sweet orange is the second most grown citrus crop in Nepal after Mandarin in terms of area and production (MOALD, 2020). The oranges in the Nepalese agricultural market have to compete with products coming from neighboring countries like India and China. The cost of production of sweet orange is higher due to high input costs, the need for hybrid budded and grafted saplings and intensive labor requirements to grow the crop which has forced the grower to think about improving postharvest management practices. The lack of suitable storage and preservation techniques forces the farmers to sell sweet oranges before their horticultural maturity and just after picking. The

unaffordable postharvest preservation has led to a negative effect on the citrus enterprises in Nepal (Kaini, 2013).

Green mold (*Penicillium digitatum* Sacc.) and blue mold (*Penicillium italicum* Wehmer) are the most economically important postharvest pathogens of sweet orange causing significant losses (Abd-El-Khair and Hafez, 2006; El-Otmani *et al.*, 2011; Papoutsis *et al.*, 2019). Currently, the control of green and blue mold is accomplished by pre-and postharvest application of chemical fungicides such as carbendazim, imazalil, thiabendazole, pyrimethanil, fludioxonil, prochloraz and guazatine (Danderson, 1986; Ismail and Zhang, 2004; Smilanick *et al.*, 2006; Smilanick *et al.*, 2008; Berk, 2016; Joshi *et al.*, 2020). Broadly, such fungicides inhibit the ergosterol synthesis, mitochondrial electron transport and synthesis of multi-site enzymes, protein and nucleic acid thereby kill or inhibit fungi or fungal spore



germination (Yang *et al.*, 2011). However, synthetic fungicides are used as the conventional ways of reducing postharvest rots which have many drawbacks including high cost, handling hazards, concern about pesticide residue on fruit and a threat to human health and environment (Tzortzakakis, 2009). Various synthetic fungicides were identified as toxic and carcinogenic by various researchers (Rouabhi, 2010; Singh *et al.*, 2016). Pathogens also developed resistance against extensive use of synthetic fungicides resulting in declining fungicidal efficiency (Fogliata *et al.*, 2000; Hao *et al.*, 2011).

The application of essential oil amended coatings has been developed as a novel and eco-friendly approach to control postharvest microbes, maintain fruit quality and improve shelf life (Alam *et al.*, 2017; Jhalegar *et al.*, 2015). The essential oils do not have only anti-fungal properties, but the secondary metabolites also have antioxidant and bio-regulatory properties (Du Plooy *et al.*, 2009; Jhalegar *et al.*, 2014; Bagamboula *et al.*, 2004; Hendel *et al.*, 2016). The increasing demand for organic fruits encourages replacing synthetic fungicides with safer alternatives. The volatility, ephemeral nature and biodegradability of essential oils make it comparatively advantageous for the treatment of postharvest citrus disease (Ameziane *et al.*, 2007). The synergism between the components in volatiles may be the reason behind the fungitoxic property of essential oils. Therefore, there is a minimal possibility of resistance. The application of essential oils with wax increases its longevity and reduces the amount of essential oils required per fruit. Therefore, the study was made to compare various wax amended treatments, their efficacy and their impact on postharvest parameters.

MATERIALS AND METHODS

Experimental site and fruit material

The present investigation was carried out at Agriculture and Forestry University (AFU), Rampur, Chitwan during the year 2017-2018. The location of the site is 27°40' N and 85°19' E with an elevation of 228 meter above sea level. The experiment was conducted in a cool and humid winter season. The local variety of sweet orange handpicked from the farmers orchard of Sindhuli was transported to Chitwan for the experiment. The fruits were kept in the tagged plastic trays during the storage period at room temperature. The average weight of fruit was 144.56 g. The average seed number was 10. The average juice content of fruit was 84.19 ml.

Experimental design

The experiment was laid out in single factor Completely Randomized Design (CRD) with nine treatments and four replications. There were a total of 36 experimental trays having 12 fruits per tray. The treatments were finalized based on the findings of Tripathi *et al.*, (2004), Jhalegar *et al.*, (2015) and Rokaya *et al.*, (2016).

Treatments details

T1: 10% (w/v) wax emulsion with 0.1% (v/v) Lemongrass (*Cymbopogon flexuosus*) oil

T2: 10% (w/v) wax emulsion with 0.5% (v/v) Lemongrass (*Cymbopogon flexuosus*) oil

T3: 10% (w/v) wax emulsion with 0.1% (v/v) Mentha (*Mentha arvensis*) oil

T4: 10% (w/v) wax emulsion with 0.5% (v/v) Mentha (*Mentha arvensis*) oil

T5: 10% (w/v) wax emulsion with 0.1% (v/v) Eucalyptus (*Eucalyptus sp.*) oil

T6: 10% (w/v) wax emulsion with 0.5% (v/v) Eucalyptus (*Eucalyptus sp.*) oil

T7: 10% (w/v) wax emulsion with 0.1% (v/v) Carbendazim (Bavistin)

T8: 10% (w/v) wax emulsion with 0.5% (v/v) Carbendazim (Bavistin)

T9: Control (dipped in 10% wax emulsion only)

Preparation of 10% wax emulsion

Paraffin wax (58-60°C, Solid LR-Grade) was used for preparing wax emulsion. Five hundred milliliter of water was boiled in a vessel and 50 g of wax was heated in another vessel. Fifteen milliliter of triethanolamine and ten milliliter of oleic acid was added in water as emulsifier and stabilizers. The molten wax was gradually poured into heated water with constant stirring. The stirring was rigorously done until the solution turns milky color. The milky color indicates well prepared emulsion. It was ensured that the heated wax and heated water were at same temperature while mixing. The prepared emulsion was then allowed to cool.

Preparation and application of essential oils and fungicide

The essential oils used in the experiments were prepared at Herbs Production and Processing Corporation Limited (HPPCL), Koteshwor, Kathmandu. The respective herbs collected from the Terai region of Nepal were dried, wilted and steam

distilled to produce oils. One milliliter and five milliliter of essential oils were added in one liter of 10% wax emulsion to prepare 0.1% and 0.5% essential oils with wax emulsion. Similarly, the fungicidal solution of carbendazim was prepared by dissolving 1 g and 5 g of carbendazim (Carbendazim 50% WP) in 1000 ml

of distilled water. One milliliter and five milliliter of fungicidal solution were added in one liter of 10% wax emulsion to prepare 0.1% and 0.5% carbendazim with wax emulsion. The fruits were then dipped in the designated solutions for a few seconds, until a glossy film of wax was formed on the surface of fruits.

Table 1: Chemical constituents of essential oils used in experiment

Essential oils	Chemical constituents
<i>Cymbopogon Flexuosus</i>	Geranial, Neral, Limonene, Caryophyllene, Geranyl acetate, Linalyl acetate, Citral, Isogeranial, <i>p</i> -cymene, Linalool
<i>Mentha arvensis</i>	Menthol, Menthone, Isomenthone, Menthyl acetate, Limonene
<i>Eucalyptus sp.</i>	Eucalyptol, Limonene, Aromadendrene, Phellandrene, Terpinolene, Alpha terpineol

Source: HPPCL website

Data collection and analysis

The data about the Juice content, fruit firmness, Total Soluble Solids (TSS), Titratable Acidity (TA), vitamin C (ascorbic acid) and disease severity scoring was taken at every 5th day interval. The physical (fruit firmness) and chemical (juice recovery percentage, TSS, TA and vitamin C) properties of fruits were measured by destructive sampling technique. The fruit firmness was measured by penetrometer (effigy oil model having 8 mm tip) and TSS was measured by hand refractometer. Acidity and vitamin C was determined as per the procedure outlined by AOAC (2005). The juice recovery percentage and content was calculated by following formulae;

$$\text{Juice recovery (\%)} = \frac{\text{Volume of the juice obtained}}{\text{weight of the fruit}} \times 100$$

The shelf life was evaluated based on the appearance and spoilage of fruits. Fruits were considered to have reached the end of shelf life when fruits showed visible signs of decay irrespective of diameter of symptom (Obagwu and Korsten, 2003).

Disease scoring and identification

Disease scoring was done on 0-5 scale. The assessment was based on the rotted area with respect to total surface area of the sweet orange and expressed in percentage [0 = no infection (fruits are healthy), 1 = infection starts (0-5% rotting), 2 = 6-10% rotting, 3 = 11-15% rotting, 4 = 16-20% rotting, 5 => 20% rotting] (Obagwu and Korsten, 2003;

Abd-El-Khair and Hafez, 2006). The rotted fruits from each replication were removed and counted. Disease severity index of decay fruits by pathogen was calculated by following formulae;

$$\text{Disease severity index (\%)} = \frac{\sum (n \times r_1) + \dots + (n \times r_5)}{MN} \times 100 \quad (\text{Abd-El-Khair and Hafez, 2006})$$

Where, n = number of decayed fruits per category, r_1, r_5 = severity score
M = maximum rating scale number (5), N = total examined fruits

The infected fruits after treatment with fungicide and essential oils were transferred to the pathology lab of AFU for the isolation of fungi. Isolation was carried out on Martin's medium (Bridson, 1995). Small pieces (1-1.2 cm thickness) of rotted fruits were sterilized by dipping into 2% sodium hypochlorite solution for 5 minutes and then washed several times with distilled water and finally dried on sterile filter paper (Abd-El-Khair and El-Mougy, 2003). The fully sterilized pieces were then transferred onto the surface of the medium in sterilized Petri-plates. Inoculated plates were incubated at 25°C for 3-5 days. Hyphal tip technique was followed for purification of the isolated fungi. Barnett and Hunter technique was used to identify fungal cultures (Barnett and Hunter, 1987).

The temperature and relative humidity of the experimental room was recorded daily. The average minimum temperature was recorded 12.88°C while the average maximum temperature was recorded 16.16°C. The average minimum humidity was 86.04% while the average maximum

humidity was 91.73%. The climate was mostly cloudy during the experiment with a few instances of drizzles. The data were entered into Microsoft Excel 2016 and analysis was carried out by using R- Studio version 4.0.2. Both descriptive and inferential analysis was carried out. Interpretations were made based on results, which were assisted by qualitative and quantitative data/information.

RESULTS

Juice recovery percentage

Juice recovery percentage decreased significantly in all treatments with the advancement of storage

time (Table 2). On the day of the experimental setup, the juice recovery percentage was found to be 58.24%. The juice recovery percentage was not significant between treatments for the storage period time of 5 days and 10 days. At 30 days after storage, maximum juice recovery percentage was observed in wax coating with 0.5% lemongrass (42.86%), which was statistically at par with the wax coating with 0.5% eucalyptus (42.81%). The lowest juice recovery percentage was seen in control fruits (33.49%).

Table 2: Effect of postharvest treatments on juice recovery percentage of sweet orange fruits

Treatments	Per cent juice content of fruits on days indicated						
	1	5	10	15	20	25	30
T1	58.24	57.04	50.17	47.04 ^{abc}	45.05 ^c	43.82 ^{bc}	41.45 ^{cd}
T2	58.24	56.95	49.56	47.81 ^a	46.21 ^a	44.10 ^{ab}	42.86 ^a
T3	58.24	56.73	49.60	46.67 ^{bc}	45.30 ^{bc}	43.69 ^c	41.18 ^c
T4	58.24	57.38	51.70	47.92 ^a	45.93 ^{ab}	44.25 ^a	41.63 ^c
T5	58.24	57.10	50.60	47.60 ^{ab}	45.46 ^{abc}	43.64 ^c	42.29 ^b
T6	58.24	57.23	50.82	47.55 ^{ab}	46.16 ^a	44.10 ^{ab}	42.81 ^a
T7	58.24	56.98	49.80	46.42 ^c	43.50 ^d	39.71 ^e	38.50 ^e
T8	58.24	57.20	51.20	46.12 ^c	44.10 ^d	42.02 ^d	41.16 ^d
T9	58.24	56.59	50.63	46.08 ^c	40.67 ^e	36.10 ^f	33.49 ^f
LSD		0.60 ^{ns}	0.85 ^{ns}	1.06 ^{**}	0.70 ^{***}	0.31 ^{***}	0.28 ^{***}
CV		0.73	1.47	1.55	1.08	0.51	0.47
Mean		57.01	50.45	47.02	44.71	42.38	40.59

LSD = Least Significant Difference, CV= Coefficient of Variation, Means within the column followed by same letters do not differ significantly at 5% level of significance by DMRT, Significance codes ***at 0.001, **at 0.01, *at 0.05

Fruit firmness

The fruit firmness decreased with the advancement of the storage period in all treatments (Fig. 1). On the day of the experimental setup, the fruit firmness

was found to be 5.35 kg/cm². On the 30th day after storage, firmness was highest for wax with 0.5% eucalyptus (3.50 kg/cm²) and lowest in control (2.25 kg/cm²) followed by wax with 0.1% carbendazim (2.75 kg/cm²).

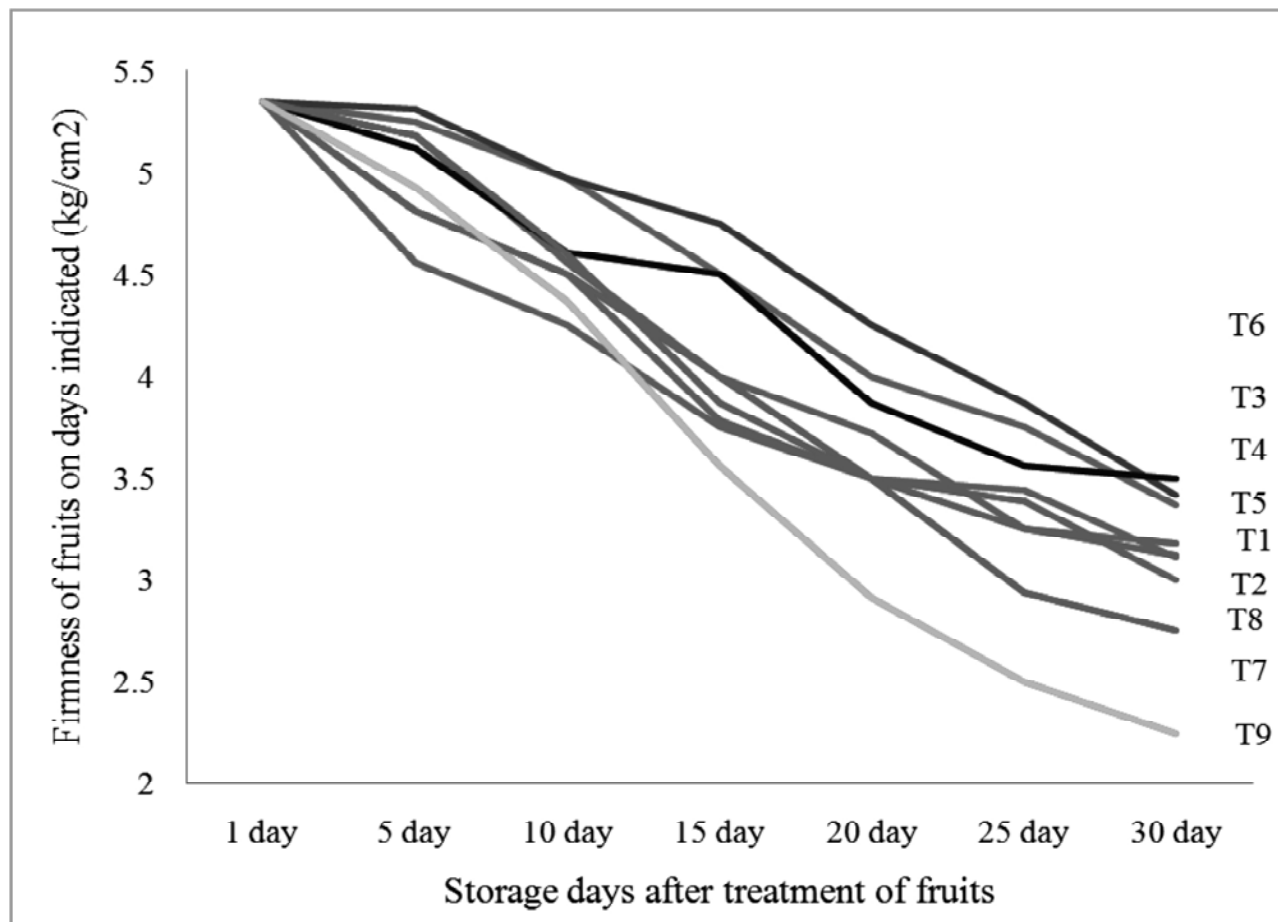


Fig.1 : Effect of postharvest treatments on firmness of sweet orange fruits

Total soluble solids (TSS)

Total soluble solid directly influences the taste of sweet orange. The TSS of fruit on the first day of storage was 11.20 °Brix. TSS increased with increment in the storage period in all treatments from 10 days onwards (Table 3). However, TSS was found to decrease on the 5th day of storage for the treatment wax with mentha. There was no significant difference between treatments on the 5th and 10th day of storage. The highest TSS was observed in control fruits (12.41° Brix) followed by wax with 0.1% carbendazim (12.28° Brix) while the lowest TSS was observed in wax with 0.5% lemongrass (11.97° Brix) at 30th day after storage.

Titrateable Acidity (TA)

The titrateable acidity is an important factor that is directly related to organic acid present in the fruit and also determines the quality of sweet orange. The TA was 1.12 on the first day of the experiment. The effect was significant only after the 10th day of treatment. There was a gradual decrease in TA of sweet orange along with the storage time. At the end of storage life i.e. 30th day, TA was highest for wax with 0.5% lemongrass (0.94%), which was statistically at par to wax with 0.1% lemongrass (0.92%) and 0.5% carbendazim (0.91%). The lowest TA was shown by control (0.72%) at the end of storage life (Table 4).

Table 3: Effect of postharvest treatments on TSS of sweet orange fruits

Treatments	TSS of fruits on days indicated						
	1	5	10	15	20	25	30
T1	11.20	11.25	11.30	11.40 ^{bcd}	11.69 ^b	11.82 ^e	12.21 ^c
T2	11.20	11.25	11.30	11.40 ^{bcd}	11.60 ^c	11.72 ^f	11.97 ^d
T3	11.20	11.00	11.20	11.41 ^{abc}	11.65 ^b	11.97 ^{bc}	12.22 ^{bc}
T4	11.20	11.00	11.30	11.45 ^{ab}	11.78 ^a	11.8 ^e	12.19 ^c
T5	11.20	11.25	11.30	11.37 ^{cd}	11.65 ^{bc}	11.9 ^d	12.20 ^c
T6	11.20	11.25	11.30	11.40 ^{bcd}	11.61 ^c	11.8 ^d	12.10 ^d
T7	11.20	11.20	11.25	11.34 ^d	11.65 ^{bc}	12.02 ^b	12.28 ^b
T8	11.20	11.25	11.30	11.37 ^{cd}	11.61 ^c	11.95 ^c	12.19 ^c
T9	11.20	11.25	11.30	11.47 ^a	11.78 ^a	12.19 ^a	12.41 ^a
LSD		0.20 ^{ns}	0.08 ^{ns}	0.06 ^{**}	0.04 ^{***}	0.04 ^{***}	0.06 ^{***}
CV		1.82	0.53	0.37	0.27	0.23	0.35
Mean		11.18	11.28	11.40	11.66	11.91	12.20

LSD = Least Significant Difference, CV= Coefficient of Variation, Means within the column followed by same letters do not differ significantly at 5% level of significance by DMRT, Significance codes ***at 0.001, **at 0.01, *at 0.05.

Table 4: Effect of postharvest treatments on TA of sweet orange fruits

Treatments	TA on days indicated						
	1	5	10	15	20	25	30
T1	1.12	1.05	1.03 ^{bc}	1.02 ^{ab}	0.95 ^{ab}	0.93 ^{ab}	0.92 ^{ab}
T2	1.12	1.04	1.05 ^b	1.03 ^{ab}	0.99 ^a	0.95 ^a	0.94 ^a
T3	1.12	1.03	0.95 ^{de}	0.92 ^{de}	0.91 ^c	0.84 ^d	0.84 ^d
T4	1.12	1.04	0.99 ^d	0.94 ^d	0.93 ^{bc}	0.88 ^c	0.82 ^d
T5	1.12	1.05	0.95 ^{de}	0.9 ^e	0.86 ^d	0.85 ^d	0.82 ^d
T6	1.12	1.02	1.04 ^{bc}	0.98 ^c	0.95 ^{abc}	0.91 ^b	0.86 ^{cd}
T7	1.12	1.02	1.05 ^{bc}	1.01 ^b	0.96 ^{ab}	0.93 ^{ab}	0.87 ^{bcd}
T8	1.12	1.03	1.08 ^a	1.04 ^a	0.97 ^{ab}	0.93 ^{ab}	0.91 ^{abc}
T9	1.12	1.03	0.89 ^e	0.85 ^f	0.76 ^e	0.74 ^e	0.72 ^e
LSD		0.00 ^{ns}	0.12 ^{**}	0.01 ^{***}	0.03 ^{***}	0.02 ^{***}	0.05 ^{***}
CV		4.86	1.85	1.30	2.75	2.01	4.00
Mean		1.03	1.00	0.96	0.92	0.88	0.85

LSD = Least Significant Difference, CV= Coefficient of Variation, Means within the column followed by same letters don not differ significantly at 5% level of significance by DMRT, Significance codes ***at 0.001, **at 0.01, *at 0.05.

Ascorbic Acid (Vitamin C) content

Vitamin C content is an important nutritive parameter in citrus fruits and it was decreased gradually during the advancement of storage days (Fig. 2). On the first day of storage, the vitamin C content was measured to be 40 mg/100ml of orange

juice. On the 30th day, the highest vitamin C was found in fruits coated with wax and 0.5% eucalyptus (30.31 mg/100ml), followed by wax with 0.5% mentha (29.50 mg/100ml) and 0.1% mentha (29.18mg/100ml), while the lowest vitamin C was observed in control fruits (24.5 mg/100ml).

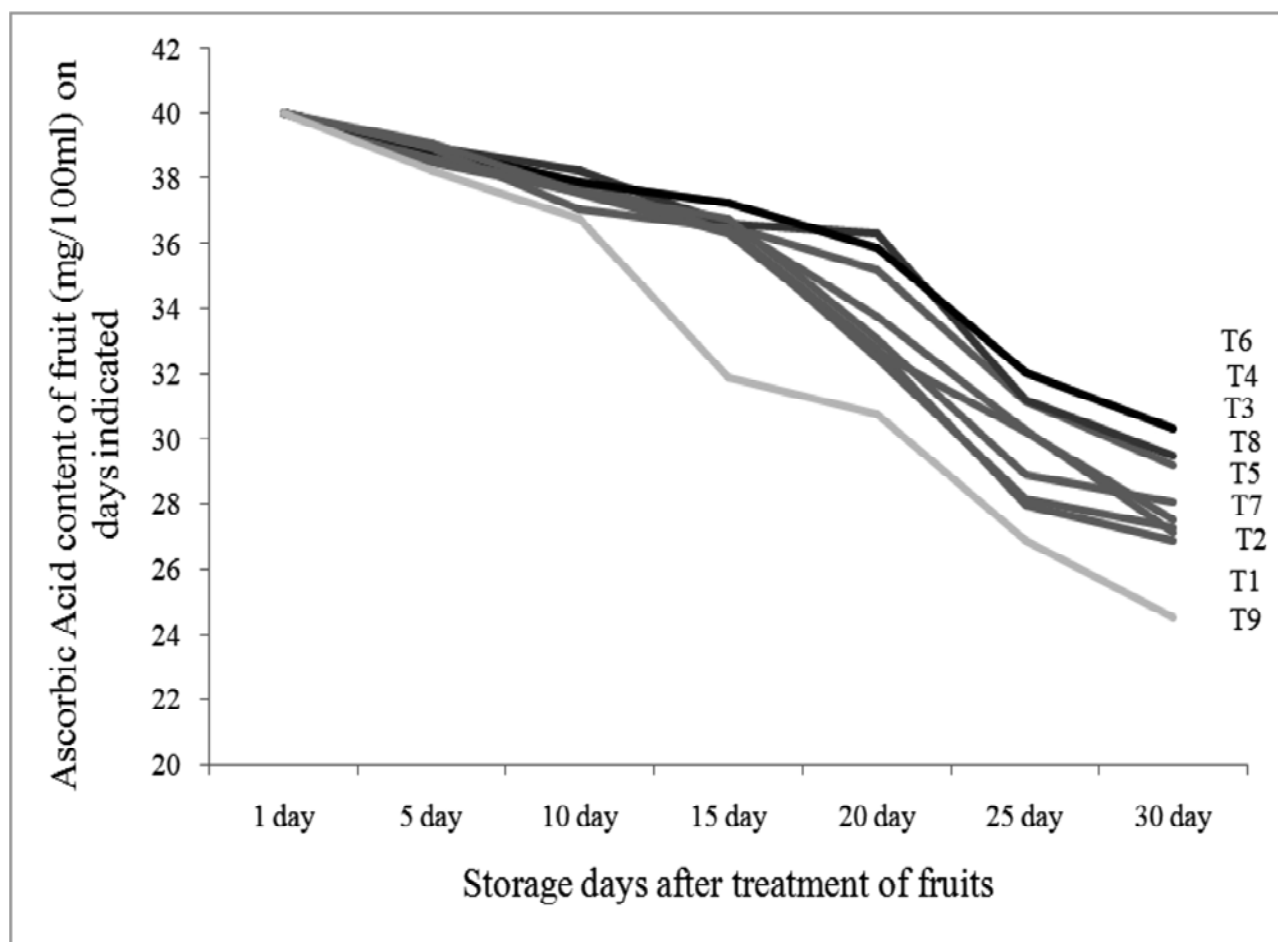


Fig. 2: Effect of postharvest treatments on Ascorbic Acid content of sweet orange fruits

Disease severity index

The disease occurrence in sweet orange was increased with the storage days (Table 5). The green mold (*P. digitatum*) was confirmed through the lab culture of a pathogen. The fruits treated with essential oils and fungicide were found to be more resistant to postharvest fungal diseases. On the 30th day of storage, almost all treatments

exhibited noticeable disease occurrence, control being the highest infected (0.372%) followed by wax with 0.1% mentha (0.180%). The treatment of wax with 0.5% carbendazim (0.004%) was most effective against fungal pathogen and wax with 0.5% eucalyptus oil (0.025%) and 0.5% lemongrass oil (0.025%) being the most effective essential oils.

Table 5: Effect of postharvest treatments on disease severity index in sweet orange fruits

Treatments	Disease severity index of fruits on days indicated						
	1	5	10	15	20	25	30
T1	0.00	0.00	0.00	0.00	0.025 ^{bc}	0.075 ^b	0.123 ^b
T2	0.00	0.00	0.00	0.00	0.004 ^{de}	0.012 ^{cd}	0.025 ^d
T3	0.00	0.00	0.004 ^b	0.012 ^b	0.038 ^b	0.075 ^b	0.180 ^b
T4	0.00	0.00	0.00	0.00	0.017 ^{cd}	0.046 ^{bc}	0.114 ^{bc}
T5	0.00	0.00	0.00	0.012 ^b	0.038 ^b	0.058 ^b	0.114 ^{bc}
T6	0.00	0.00	0.00	0.00	0.008 ^{de}	0.012 ^{cd}	0.025 ^d
T7	0.00	0.00	0.00	0.00	0.00	0.000	0.033 ^{cd}
T8	0.00	0.00	0.00	0.00	0.00	0.000	0.004 ^d
T9	0.00	0.00	0.042 ^a	0.054 ^a	0.096 ^a	0.207 ^a	0.372 ^a
LSD			0.009 ^{***}	0.013 ^{***}	0.013 ^{***}	0.038 ^{***}	0.080 ^{***}
CV			121.76	105.94	35.646	48.471	50.13
Mean			0.005	0.008	0.025	0.054	0.110

LSD = Least Significant Difference, CV= Coefficient of Variation, Means within the column followed by same letters do not differ significantly at 5% level of significance by DMRT, Significance codes ***at 0.001, **at 0.01, *at 0.05.

Shelf life

The wax treatment with carbendazim and essential oils had a significantly better shelf life as compared to the control treatment (Table 6). Wax with 0.5% carbendazim (28.25 days) being the highest and significantly better than other treatments. It was followed by wax with 0.1% carbendazim (25.75 days), wax with 0.5% lemongrass oil (20.00 days) and wax with 0.5% eucalyptus oil (19.75 days). The control fruits (8.25 days) were observed to have the lowest shelf life.

Discussion

A significantly lower juice recovery percentage of control fruits might be due to the fact that the essential oils act as a barrier which checks the loss of moisture from the fruit surface due to the clogging of natural openings (Castillo *et al.*, 2014). Additionally, the lower incidence of disease in essential oils and fungicides treated fruit ensure lower metabolism, which might have contributed to a higher juice recovery percentage. The present finding was supported by (Bisen *et al.*, 2012). The control fruit also had a wax coating and the transpiration process was very slow, so there was an insignificant difference in juice recovery percentage between treatments before 15th day of storage. The moisture

Table 6: Effect of postharvest treatments on shelf life in sweet orange fruits

Treatments	Shelf life
T1	17.25 ^d
T2	20.00 ^e
T3	10.00 ^f
T4	16.75 ^d
T5	14.25 ^e
T6	19.75 ^c
T7	25.75 ^b
T8	28.25 ^a
T9	8.25 ^g
LSD	0.92 ^{***}
CV	3.55
Mean	17.80

LSD = Least Significant Difference, CV= Coefficient of Variation, Means within the column followed by same letters do not differ significantly at 5% level of significance by DMRT, Significance codes ***at 0.001, **at 0.01, *at 0.05.

loss was found significantly lower in fruit treated with essential oil enriched coatings (Du Plooy *et al.*, 2009; D Antunes *et al.*, 2012; Castillo *et al.*, 2014).

In general, coating formulations that minimize weight loss are also better at maintaining firmness, since this attribute is highly influenced by water content. Fruit firmness decreased gradually and significantly along with increasing storage period in all treatments. The decelerated damage may be due to the anti-microbial properties of essential oils. The lowest fruit firmness in control fruit might be due to the rapid degradation of cell walls due to the action of wall-degrading enzymes such as pectinesterase, pectinmethyl-esterase and polygalacturonase which are produced by fungi. Essential oil amended coating maintains cell wall carbohydrate metabolism during storage which is related to decreased susceptibility to infection by fungal pathogen and therefore improves quality. The essential oils together with commercial wax coating maintain the organoleptic integrity along with firmness as mentioned by Jhalegar *et al.* (2014). The essential oils affect the portioning of the lipids of the plasma membrane and changing of its integrity, permeability and inorganic ion equilibrium due to their hydrophobic nature (Lambert *et al.*, 2001) which might be the reasons for greater firmness in the fruits treated essential oils. The present findings were supported by Chafer *et al.* (2012) on the firmness of Navel Powell orange and Castillo *et al.* (2014) on lemon fruits.

The gradual increase of TSS with extending of the storage period might be attributed to concentrated juice content results from dehydration and hydrolysis of polysaccharides. The increased respiration rate due to microbial spoilage, degradation of fruits and increased ethylene production ultimately increased the TSS during ripening and senescence which might be the reason for slightly higher TSS in control fruits as compared to other treatments. The present result was in agreement with the findings of Chafer *et al.* (2012) on Navel Powell orange and Castillo *et al.* (2014) on lemon and Tao *et al.* (2014) on Satusma mandarin, as the essential oils did not show a significant effect on TSS. The present finding was also inconsistent with Asghari *et al.* (2009) who reported insignificant results in TSS while using cumin essential oil on strawberry.

The decrease in titratable acidity with storage is due to the oxidation of organic acids and further

utilization in the metabolic process in the fruits (Hafeez *et al.*, 2012). A gradual declining trend in titratable acidity content of fruit during storage for any treatment was observed by Ansari and Feridoon (2007) and Obenland *et al.* (2008) in citrus. The decreased in titratable acidity of fruits during storage could be due to the consumption of organic acids in the respiration process as stated by Zokaei *et al.* (2006) and Ishaq *et al.* (2009). Similarly, Baiea (2013) on Washington Navel orange detected a decrease in the acidity of fruits during storage. Fruits treated with essential oils showed higher retention of titratable acidity during the storage period which might due to delayed in physiological ageing and alteration in metabolism. The present results are in line with Mahajan *et al.* (2010) suggesting that organic acids were used in the respiratory process. The higher titratable acidity in wax with lemongrass treatment is aligned with the finding of Fatemi *et al.* (2012) who reported that the thymol oil delayed the changes in titratable acidity of Valencia orange. The present finding was also supported by Jhalegar *et al.* (2014) on Kinnow mandarin. Abd El wahab *et al.* (2014) also reported bergamot oil delayed the changes in titratable acidity during cold storage of Crimson seedless grape.

Adisa (1986) stated that vitamin C decreased over time in storage which is similar to the experimental outcome. The decreased in ascorbic acid content of fruits during storage could be due to the conversion of dehydroascorbic to diketogulonic acid by oxidation as reported by Ishaq *et al.* (2009). Under stress, such as a pathogen or chemical exposure, ascorbate oxidase levels were increased, which decreased the level of vitamin C (Loewus and Loewus, 1983; Loewus *et al.*, 1987). The maximum retention of vitamin C was observed with essential oils treatments due to the antioxidant property of essential oils which prevent ascorbic acid from oxidation (Shao *et al.*, 2013). The result was similar to Lin *et al.* (2011) who found that the decrease in vitamin C level was associated with a reduced capacity of preventing oxidative damage which is triggered by the incidence of physiological disorders during storage. The degradation of vitamin C was highest in control fruits which might be due to fruit senescence accompanied by rapid respiration, ethylene production and decay. These results are similar to those reported on the effect of thyme and

clove oil in maintaining ascorbic acid as for orange (Zeng *et al.*, 2012; Baiea and Ei-Badawy, 2013).

The result was similar to the report of Abd-El-Khair and Hafez (2006), as they reported the lemongrass and eucalyptus essential oils significantly reduced the incidence of fungus *P. digitatum* in Washington navel orange during storage. Abdolahi *et al.*, (2010) and Al-Samarrai *et al.*, (2013) found various plant extracts including lemongrass extract could inhibit the mycelial growth of pathogenic fungus *P. digitatum*. The phenolic compounds and their derivatives of essential oils altered the microbial cell permeability by interacting with membrane proteins which would cause deformation in cell structure and function

and permit the loss of macromolecules from their body (Fung *et al.*, 1997; Rattanapitigorn *et al.*, 2006) which might be the reasons of lower microbial growth in the essential oils treated fruits compare to control. Amit and Malik (2010) indicated that the vapour of lemongrass oil damaged the cell membrane mainly due to membrane deformation. However, the variation in the antifungal effect of the essential oils depends on the solubility and capacity to interact with the cytoplasmic membrane (Tripathi and Shukla, 2007). The efficacy of lemongrass was also found superior by Jhalegar *et al.* (2015). Similar results were reported by Du Plooy *et al.* (2009), Fan *et al.* (2014), Jhalegar *et al.* (2014) and Gandarilla-Pacheco *et al.* (2020) in citrus fruits.

Table 7: Summary of studies on the effect of essential oils on major post-harvest pathogens of citrus

Fruit	Target pathogen	Essential oils	References
Orange cv. Tomango	<i>P. digitatum</i>	Mentha oil	Du Plooy <i>et al.</i> (2009)
Orange, Lime	<i>P. italicum</i>	Mentha oil	Tripathi <i>et al.</i> (2004)
Washington Navel Orange	<i>P. digitatum</i>	Lemongrass oil, Eucalyptus oil	Abd-El-Khair and Hafez (2006)
Valencia Orange	<i>G. citri-aurantii</i>	Lemongrass oil	Regnier <i>et al.</i> (2014)
Kinnow Mandarin	<i>P. digitatum, P. italicum</i>	Eucalyptus oil	Jhalegar <i>et al.</i> (2014)
Kinnow Mandarin	<i>P. italicum and P. digitatum</i>	Lemongrass oil, Eucalyptus oil	Jhalegar <i>et al.</i> (2015)

The wax with 0.5% carbendazim with its prominent disease resistance had the longest storability. The superiority of shelf life of 0.5% lemongrass and eucalyptus oils treated fruits might be due to the antifungal properties of essential oils (Tzortzakos and Economakis, 2007; Jhalegar *et al.*, 2014; Jhalegar *et al.*, 2015). In addition to this, postharvest decay is positively correlated with ethylene production and respiration rate which were found to be decreased by the application of essential oils (Jhalegar *et al.*, 2014; Jhalegar *et al.*, 2015). The present result was inconsistent with Tripathi *et al.* (2004) and Tavakoli *et al.* (2019).

The green mold is the major postharvest pathogen in sweet orange. The chemical fungicides have been found effective against such pathogens, but the health hazard of such pesticides is alarmingly high. The use of essential oils as an alternative for chemicals can be an environment friendly technique for prevention of the health hazards. The shelf life of sweet orange can be extended by infusing wax with carbendazim or essential oils. But the superiority of essential oils especially wax with 0.5% eucalyptus oil and 0.5% lemongrass in qualitative parameters as well as in consumer's preferences, organic requirements and environmental aspects make them a better alternative.

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