Effect of chitosan coating on some quality properties of Thomson orange during storage (a case study in Iran)

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Abstract: Most of the world's consumers have used the natural coatings instead of chemical coatings. In this research, some quality properties of Thomson orange were evaluated using Chitosan edible coating (at concentrations of 0.5%, 1% and 2%) and chemical coating of fungicide (Ortho Phenyl Phenol) as compared to uncoated samples during storage for three months. Fruit was maintained in cold storage at 6°C and 85%-90% relative humidity, then pH, total soluble solid (TSS) of orange juice and percentage of fruit weight loss were measured during storage every month. The results showed that weight loss of fruit with 2% chitosan coating was lower than other treatments. Also, this treatment had high resistance against fungal attacks due to high values of pH, so 2% chitosan coating can be able to protect the fruit quality during storage. **Keywords:** chitosan, citrus, fungicide, quality, storage

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1 Introduction

Citrus is a subtropical and tropical fruit. Citrus fruit include oranges, lemons, limes and grapefruit, in addition to tangerines and pomelos. The orange is the fruit of the citrus species. Thomson navel orange is the result of a genetic mutation of Washington navel orange. Also, it is cultivated in many parts of the world, including California, Australia, Spain, Indonesia, Iran and etc. (FAO, 2012). Thomson Navel oranges have an especial importance in comparison with other cultivars of citrus. The orang is one of the most important horticultural crops in Iran. According to FAO State more than 1285000 tons of orang are produced in Iran (FAO, 2012).

Consumers worldwide demand for the natural coating instead of chemical coating for protection of fruit quality (Galed et al., 2004). Many edible coatings are commonly used for fruit and fresh vegetables in recent years (Baldwin, 2007; Arnon et al., 2015). Corn starch coating was used to study the post-harvest quality of Assam lemon fruits by different concentration (1%, 2%, 3%, 4%, 5% and 6%) (Ghosh et al., 2015). An example of natural coatings is chitosan. Chitosan is derived from chitin and is natural biopolymer. Chitin is founded in the shells of crustaceans and for commercial purpose is extracted from shellfish waste (Galed et al., 2004; Tripathi and Dubey, 2004; Muzzarelli and Muzzarelli, 2003). Chitosan is edible and secure that it has numerous applications in agriculture, medicine, food, environment and etc. (Devlieghere et al., 2004). Chitosan film has been used as a coating of crops such as mandarin, strawberries and fresh fruits (Fornes et al., 2005; Ribeiro et al., 2007; Kerch, 2015).

Postharvest losses of fruit was happened between harvest and consumption (Park, 1999). The most harmful citrus during storage pathogen is *Penicillium digitatum* that it produces green mold disease (Kinay et al., 2007; Porat et al., 2000). Green mold is the most important post-harvest disease on citrus (Holmes and Eckert, 1999). Pathogenic fungi attacks citrus fruit and causes decay. Because of the low pH and high moisture content, citrus fruit was unfitted for consumption (Tripathi and Dubey,

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2004).

Chitosan coating is formed a semi- permeable film and provides an alternative to modifying atmosphere storage (Del-Valle et al., 2005; Hagenmaier, 2005). Then transpiration losses and fruit respiration rates were decreased. Therefor the quality of the fruit is effective on long- term storage of fruit was controlled (El Ghaouth et al., 1992; Du et al., 1997). Chitosan has antimicrobial activity (Ait et al., 2004; Plascencia-Jatomea et al., 2003) and the fruit coated by the chitosan restricts fungal growth (No et al., 2007).

Chitosan effectiveness in decay control has the best performance at the highest concentration (Romanazzi, 2010). The chitosan for coating of cucumber and bell pepper fruit with different concentration was used. Research results showed that increasing the concentration of chitosan from 1.0% to 1.5% reduced the respiration rate of fruit and had the more effectiveness in preservation of fruit quality (Chien et al., 2007). Permeability of low molecular weight chitosan (LMWC) is higher than high molecular weight chitosan (HMWC) and expressed that LMWC had the highest bactericidal activity rather than pathogenic bacteria (Chien et al., 2007).

As, little research has been performed on effectiveness of chitosan different concentration on fruit quality properties. Also, no researchers have been effect proposed the of chitosan coating on physiochemical property of Thomson orange during storage, according to our knowledge. Therefore, this study aimed to investigate the effect of different coatings on the physiochemical properties of Thomson orange during storage.

2 Material and methods

Thomson navel orange was hand-harvested from an orchard at Dinek village, Mazandaran, Iran. Samples were chosen randomly from a type of citrus (Thomson Navel) with uniform appearance and without disease.

Chitosan was provided from company of Sigma Aldrich with low molecular weight (43 kDa). The solution of chitosan was prepared in three levels (0.005%, 0.01% and 0.02%) that each level was obtained with adding 0.5, 1.0 and 2.0 g chitosan in 50 mL of glacial

acetic acid, 900 mL distilled water and 1 g tween80 was produced. The composition of 0.5, 1.0 and 2.0 g chitosan are called 0.5C, 1C and 2C coating, respectively.

Before coating, fruit was washed with tap water then were dried in environment temperature and were dipped into the chitosan composition by hand. Coating on fruit surface was dried at environment temperature $(12^{\circ}C)$ along 12 h then fruit was stored at 5°C and 85%-90% relative humidity for 90 days. The evaluations were conducted using three levels of chitosan solution (coated samples) during storages (for 3 months, interval 1 month) in four replications as compared to uncoated samples.

During storage, fruit quality properties were measured. Quality properties included the chemical tests, weight losses percentage and fruit compression test. The measurements were performed in Citrus Research Central of Ramsar, Mazandaran. Chemical tests included pH and total soluble solid (TSS) of fruit juice. Fruit was washed and cut in half then their juices were extracted using a hand juicer. The values of pH for 20 mL of sample juices were measured at laboratory temperature (20°C) using a digital pH meter (Inolab pH 720, German), Also the values of TSS were determined with a digital refract meter at 20°C (Atago - ATC - 20E, Japan).

The numbers of 32 samples were stored in cold storage. Fruit was periodically weighted by digital scale with an accuracy of 0.001 g. Weight loss percentage is calculated according Equation (1):

Percentage of weight loss = $(W_1 - W_2)*100/W_1$ (1) where, W_1 = Fruit weight before test; W_2 = Fruit weight after test.

A full-factorial design was used for experimental analysis. The obtained data were analyzed using SPSS software (Version 17; SPSS Inc., Chicago, IL, USA). Duncan's multiple range tests were used to determine the differences between mean values at a confidence level of 95%. All experiments were performed in three replicates.

3 Results and discussion

Table 1 shows analysis of variance (ANOVA) of some quality properties of orange fruit during storage. According to Table 1, influence of pretreatment and storage time on all of the characteristics were statistically significant (P<0.01).

| Dependent Variable | pH | | Total soluble solid, % | | Weight losses, % | | Moisture content, dry% | |
|-----------------------|----|---------------------|------------------------|---------------------|------------------|---------------------|------------------------|-----------------------|
| Source | df | Mean Square | df | Mean Square | df | Mean Square | df | Mean Square |
| Corrected Model | 19 | 0.112^{**} | 19 | 4.708^{**} | 19 | 70.169** | 19 | 1364.974** |
| Intercept | 1 | 964.406** | 1 | 11925.914** | 1 | 2404.659** | 1 | 5077133.209** |
| Coating | 4 | 0.042^{**} | 4 | 6.095*** | 4 | 10.737** | 4 | 2631.593** |
| Time | 3 | 0.598^{**} | 3 | 18.686** | 3 | 423.484** | 3 | 3799.756** |
| $Coating \times Time$ | 12 | 0.015 ^{ns} | 12 | 0.751 ^{ns} | 12 | 1.651 ^{ns} | 12 | 334.072 ^{ns} |
| Error | 60 | 0.013 | 60 | 0.400 | 60 | 1.109 | 60 | 613.944 |

Table 1 Variance analysis (ANOVA) of treatments on some quality properties of orange fruit

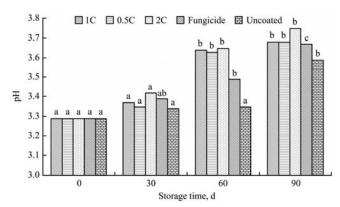
Note: **: Significant (P<0.01), ^{n.s}: Not significant.

As, Table 2 is shown, the pH of orange juice is affected by different coatings and storage times. In the period of 30 days, pH of the sample with fungicide coating had significant difference (P<0.05) with other samples. According to Figure 1, the pH change was statistically determined for each coating. The pH of the sample with 2C coating was higher than the other samples during storage. The pH change in periods 60 and 90 days for all of the samples was significant (p<0.05).

Table 2The pH of orange juice as affected by differentcoatings as compared to uncoated samples during storage time

| Storage time, d | 1C | 0.5C | 2C | Fungicide | Uncoated samples |
|-----------------|-------------------|-------------------|-------------------|--------------------|-------------------|
| 0 | 3.29 ^a | 3.29 ^a | 3.29 ^a | 3.29 ^a | 3.29 ^a |
| 30 | 3.37 ^a | 3.35 ^a | 3.42 ^a | 3.39 ^b | 3.34 ^a |
| 60 | 3.64 ^b | 3.63 ^b | 3.65 ^b | 3.49 ^{ab} | 3.35 ^a |
| 90 | 3.68 ^a | 3.68 ^a | 3.75 ^a | 3.67 ^a | 3.59 ^a |

Note: In each row, values followed by the same letter do not have a significant difference (p < 0.05).



Note: At each coating, the means having the same letter do not have a significant difference at the 5% level.

Figure 1 The pH of orange juice at different coatings and storage times as compared to uncoated samples

Where the composition of 0.5, 1.0 and 2.0 g chitosan are called 0.5C, 1C and 2C coating. The pH change of samples was increased during storage. The samples which were exposure more attack fungal disease in the lowest pH value of juice. Thus, the high pH value of juice increases resistance of fungal attacks and fruit edible quality (Aryan et al., 2008; Martin-Diana et al., 2009; Rahemi, 1998; KhoshTaghaza and Taghinezhad, 2016).

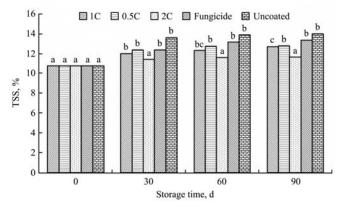
Table 3 shows the mean value of concentrations of total soluble solids (TSS) of juice, for the different pretreatments during storage. The results showed that the values of TSS for samples with 2C and 0.5C coating in periods of 60 days were significant (P<0.05). Also, the sample TSS with 2C and 1C coating in periods of 90 days do not have significant difference.

Figure 3 indicates the TSS value changes for each coating during storage and its changes are indicated using Duncan test with English letters (on each column). According to Figure 3, TSS value changes are not significant for 2C coating compared to other coating. Also, the low and high values of TSS were for samples with 2C coating and uncoated samples, respectively. These findings are in agreement with the results of Ghosh et al. (2015).

 Table 3
 Effect of different coating on TSS of orange juice during storage time

| Storage time, d | 1C | 0.5C | 2C | Fungicide | Uncoated samples |
|-----------------|---------------------|---------------------|--------------------|--------------------|--------------------|
| 0 | 10.80^{a} | 10.80^{a} | 10.80^{a} | 10.80^{a} | 10.80^{a} |
| 30 | 12.03 ^a | 12.38^{a} | 11.45 ^a | 12.40^{a} | 12.40 ^b |
| 60 | 12.33 ^{ab} | 12.75 ^b | 11.67 ^a | 13.20 ^c | 13.20 ^c |
| 90 | 12.70 ^a | 12.80 ^{ab} | 11.65 ^a | 13.38 ^c | 13.38 ^c |

Sugar is formed the highest number of soluble solids in citrus juices. According to Figure 3, amount of sugar increased during storage for all of samples. Increasing the amount of sugar in citrus during storage is due to cell wall hydrolysis with different enzymes (Chundawat et al., 1978; Echeverria et al., 1989; Golshan and Shahbik, 2004; Obenland et al., 2008). It can be concluded due to the low TSS value of samples with 2C coating, water loss was reduced. Therefore, 2C samples with values of TSS 10.80 to 11.65 have more fruit juice as compared to other samples.



Note: At each coating the means having the same letter do not have a significant difference at the 5% probability level.

Figure 2 The TSS of orange juice at different coatings as compared to uncoated samples during storage times

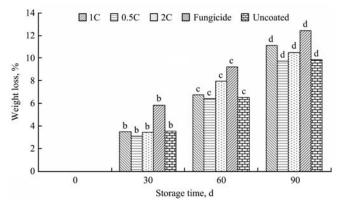
Table 4 indicates effect of different coatings on weight loss percentage of orange fruit during storage. The weight loss values of uncoated sample rather than coated samples in periods of 30, 60 days were significant (P<0.05). At the end of storage time, 2C coating and uncoated samples had the low (9.72) and high (12.43) percent of weight loss, respectively. Figure 3 indicates the effect of coating on the amount of weight loss. The variation trend of fruit weight loss with storage time was significant (p<0.05). These findings are in agreement with the reported observations by Nabifarkhani et al. (2015).

The weight loss of fruit is due to the effect of water loss from the skin or biological changes. Rapid decrease in water content in the fruit, it is a good indicator for fruit senescence. Respiration rate has inversely related with the storage time of fruit. Whatever the fruit respiration rate is lower, the storage time will be higher, but if enough oxygen for aerobic metabolism is not available, it will cause fermentation in the fruit. The weight loss of fruit in different periods of storage has been less than the other samples by using of 2C coating. Therefore, the 2C coating is effective than rest of the coating in preventing the loss of fruit juice.

 Table 4
 Effect of different coatings on the percent of weight loss of orange fruit during storage time

| | | 0 | | 8 8 | |
|-----------------|-------------------|---------------------|-------------------|--------------------|--------------------|
| Storage time, d | 1C | 0.5C | 2C | Fungicide | Uncoated samples |
| 0 | 0.00^{a} | 0.00^{a} | 0.00^{a} | 0.00^{a} | 0.00^{a} |
| 30 | 3.54 ^a | 3.41 ^a | 3.05 ^a | 3.48 ^a | 5.83 ^a |
| 60 | 6.51 ^a | 7.93a ^b | 6.37 ^a | 6.74 ^a | 9.21 ^b |
| 90 | 9.85 ^a | 10.47 ^{ab} | 9.72 ^a | 11.09 ^c | 12.43 ^c |

Note: Values within a row followed by the same letter do not have a significant difference (p < 0.05).



Note: At each coating the means having the same letter do not have a significant difference at the 5% probability level.

Figure 3 The weight loss of orange fruit at different coatings as compared to uncoated samples during storage time

4 Conclusions

In this research, concentrations of chitosan and chemical fungicides are tested on the Thomson orange. The 2C coating had an important effect in maintaining quality and reducing loss of fruit juice during storage due to higher pH value, less concentration of TSS and weight loss percentage than other coatings. Also, it can be replaced as the suitable edible coating instead of fungicides that are commonly used. Therefore, edible 2C coating will be used more than the chemical coating due to high effect in the prevention of fungal disease.

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