

# The effect of thyme essential oil ,calcium chloride and storage time on quantity and quality of sapodilla fruit ( Manilkara zapota L. ) Var. Oval

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**ABSTRACT:** Sapodilla (Manilkara zapota L.) is the tropical fruit from Sapotaceae. This plant is now cultivated in Southern Iran. One of the important problems of sapodilla fruit is short postharvest maintenance period. In this study the effect of calcium chloride and thyme essential oil treatments on fruit quality and quantity characteristics of sapodilla fruit were investigated. The fruits were harvested at physiological maturation stage and treated with :(1) control (no treatment), (2) dipping in 2% (w/v) CaCl<sub>2</sub>,(3) 250 ml/l thyme essential oil,(4) 500 ml/ thyme essential oil, (5) 2% CaCl<sub>2</sub>+250 ml/l thyme essential oil,(6)2% CaCl<sub>2</sub>+500 ml/l thyme essential oil and storage at 13±2°C and 70±5% relative humidity. The analyzed parameters were: Weight loss(%),firmness,total titratable acidity, pH, total soluble solids (TSS),TSS/TA ratio, malondialdehyde and panel test . Also after 30 days fungal contamination of the samples was evaluated. The results showed that the used concentration of thyme essential oil alone or in combination with calcium chloride completely inhibited the growth of fungi on fruit. The use of thyme essential oil at low concentration(250 µl/l) and calcium chloride had a positive effect on some parameter such as fruit firmness, weight loss, malondialdehyde and fruit appearance. Calcium chloride treatments affected flesh color parameters, malondialdehyde and fruit firmness.High concentration of thyme essential oil could change the color and somewhat flesh, taste the fruit and undesirable flavor fruit. Based on the obtained results it can be inferred that thyme essential oil with or without the presence of low concentrations of calcium chloride can replace with chemical fungicides for control of pathogenic fungi on sapodilla fruit and thereby increase its shelf life, but achieve appropriate levels of these essential oils alone and in combination with calcium chloride is needed.

**Keywords:** Calcium Chloride, Manilkara zapota L., Postharvest, Thyme essential oil.

## INTRODUCTION

The Sapodilla (Manilkara zapota ( L.)P.Royen) is a fruit species native the tropical Americas which is said to have originated from southern Mexico. The fruit is sweet with a pleasant aroma and is greatly appreciated by a significantly large group of consumers (Balerdi and shaw,1998, Ma; et al.,2003) . Also fruits in general are important sources of antioxidants, vitamins and minerals, and usually have industrial and medicinal applications. India is the main producer of sapodilla, with a production of 1,346,000 tons (Indian Horticulture Database,2010). Physiologically, Sapodilla fruit exhibit a climacteric behavior in postharvest (Lakshminarayana,1979); ripening occurs rapidly and is characterized by a significant increase in the respiration rate and ethylene production, all of which classify it as a highly perishable fruit with a short shelf life, making its commercialization more difficult. Depending on the variety and the agro-climatic conditions of production, the fruit ripens at 26°C between 8 and 10 days after harvesting (Morais; et al., 2006). Any increase in the shelf-life of this product, therefore, would contribute to an improvement in its commercialization. The fact that the sapodilla is highly perishable due to its climacteric nature, has made it necessary to study technologies of postharvest management that prolong its shelf life and favor its commercialization in more distant markets (Ganjyal; et al., 2003).

Essential oils are the wide range of secondary metabolites. In most cases, they possess antimicrobial, allelopathic, antioxidant and regulatory environment. The essential oils are a complex mixture of a variety of chemicals, including hydrocarbons, alcohols, ketones, aldehydes and other compounds are present in. Fungicidal activity of essential oils has been shown in numerous experiments; however, a few studies on the effect of essential oils on fruit quality were documented in this report. There is clear evidence now due to the higher level of knowledge has increased the demand for healthy and organic food products. The increased interest in such studies as an alternative to synthetic chemicals that are harmful to human health effects have been found to be clear (Asghari marjanlo; et al., 2008).

Alizadeh; et al., 2010, the effect of plant extracts of sage, savory and thyme Shiraz etiologic agents inhibit the growth of fungal mycelium examined and concluded that the extract of thyme and savory resulted in complete inhibition of fungal growth was etiologic agents.

Karimi and Rahemi, 2008, Effect of thyme and clove extracts and fungicide Imazalil on blue mold decay of citrus fruit in cold storage were studied. The results showed that application of thyme and clove extract reduced the percentage of fruit rot. All of which were statistically significant compared to control. The comparative effects of these extracts were not significantly different with fungicide. The use of these extracts also cause ulcers in the control of dental caries; however, severe burns at the site were used.

Hassani; et al., 2009, Effect of essential oils of thyme, basil and rosemary on disease control of gray mold (*Botrytis*) studied in pear. The results showed that the essential oil treatments had no significant effect on pear fruit gray mold disease. The number of infected fruits and severity of disease in the treated fruits decreased. Fungal growth was decreased with increasing concentration of essential oils. The results also showed essential oil of thyme and rosemary were the most and least effective in reducing the incidence and severity of gray mold had infected fruits. *Zataria multiflora* belongs to the Lamiaceae family that geographically grows only in Iran, Pakistan and Afghanistan (Ali; et al., 2000). This plant has the local name of 'Avishan Shirazi' (in Iran) and traditional uses such as antiseptic, anaesthetic and antispasmodic (Hosseinzadeh; et al., 2000). The main constituents of the essential oil of this plant are phenolic compounds such as carvacrol, thymol and eugenol (Shaffiee and Javidnia, 1997).

Calcium has been extensively reviewed as both an essential element and its potential role in maintaining postharvest quality of fruit and vegetable crops (Kirkby and Pilbeam, 1984; Bangarth, 1979). Calcium has an important role in postharvest quality, because of its role in plant metabolism and membrane stability (Kirkby and Pilbeam, 1984) by contributing to the linkages between pectic substances within the cell-wall (Demarty; et al., 1984).

(Sudha; et al., 2007), Effect of calcium chloride treatments on fruit physical characteristics studied. The outcome of this treatment was to hold the highest fruits firmness.

(Mahmoud; et al., 2004), Effect of calcium chloride treatments on fruit ripening mango examined. The outcome of this treatment will delay the ripening fruit.

In this study an attempt is using postharvest technology to increase storage life and maintaining an acceptable quality of *Sapodilla* fruit.

## MATERIAL AND METHODS

*Sapodilla* fruits were harvested in July 2011, from orchard located in the Agricultural Research Station of Minab. Harvested fruits are mature but unripe and were uniform in size and were harvested carefully with minimum mechanical injury. The harvested fruits were washed with water to remove the latex and shade dried until no water was visible on the fruit surface. The samples were transported to the laboratory for analyses.

Treatments included: (1) control (no treatment), (2) dipping in 2% (w/v)  $\text{CaCl}_2$ , (3) 250 ml/l thyme essential oil, (4) 500 ml/l thyme essential oil, (5) 2%  $\text{CaCl}_2$ +250 ml/l thyme essential oil, (6) 2%  $\text{CaCl}_2$ +500 ml/l thyme essential oil and storage at  $13\pm 2^\circ\text{C}$  and  $70\pm 5\%$  relative humidity. The analyzed parameters were: Weight loss(%), firmness, total titratable acidity, pH, total soluble solids (TSS), TSS/TA ratio, malondialdehyde and panel test. Also after 30 days fungal contamination of the samples was evaluated.

Weight loss(%)

the following equation:

$$\text{Weight loss(\%)} = [(A-B)/A] * 100$$

fruit weight at initial period = A

fruit weight at sampling period = B

Firmness

Fruit firmness was determined on whole and unpeeled fruits using a Fruit Pressure Tester (Model FT 327, Made in Italy). The force was determined at two equidistant points on the equatorial region of each fruit. The mean values of the firmness were expressed as Newton (N).

**Soluble solids content (SSC)**

SSC was determined by a digital refractometer and expressed as °Brix. The refractometer was calibrated with distilled water before reading.

**pH**

pH value was determined by pH meter(691, Metrohm, Swiss Made). The pH meter was calibrated using buffer solutions.

**Titrateable acidity (TA)**

TA was determined by titrating 5 mL of juice with 0.1 N NaOH, to pH 8.1 and expressed as grams of malic acid per 100 g fresh pulp.

**Malondialdehyde (MDA) content**

MDA content was measured by the thiobarbituric acid method described by Hodges et al;1999. MDA content was expressed as  $\mu\text{mol}^{-1}$  g fresh weight(fw).

$$\text{MDA} = (\text{OD} / 155 * 4) ( \text{Vtotal} / \text{Fresh Weight} ) ( 1000 / \text{Vsample} )$$

OD= Absorbance at 600 nm-Absorbance at 532 nm

sample weight Fresh Weight =

Vtotal= The total volume of sample

Vsample= The volume of sample

**Panel test**

Panel test were conducted to assess the marketability of fruits. And of characters such as appearance,color,sweetness,astringency,tenderness,firmness and overall acceptability was evaluated. To panel test of 20 people were considered and the fruit were tested by grading method.

**Statistical analysis**

The experiment was a completely randomized design(CRD) with factorial in three replication .The means were compared by was evaluated by Duncan's Multiple Range Test  $P < 0.01$ .

**RESULT AND DISCUSSION**

**Weight loss**

Maximum weight loss occurred in 2% CaCl<sub>2</sub>, while lowest loss (0.2%)was recorded in 2% CaCl<sub>2</sub>+thyme essential oil 250 $\mu\text{l/l}$  and 2% CaCl<sub>2</sub>+thyme essential oil 500 $\mu\text{l/l}$ .Weight loss was highest during the 15th and 20<sup>th</sup> days.Overall highest weight loss occurred in 2% CaCl<sub>2</sub> during the 20 day (Fig.1). The result can be seen weight loss although calcium chloride is used as an ingredient in fruit flesh firmness but simplyit cannot prevented from evapotranspiration. When used from combination Calcium chloride and thyme essential oil decreased weight loss. Probably the interaction between calcium chloride and thyme essential oil decreased cell survival activities such as respiration and thus reduced the percentage of weight loss.

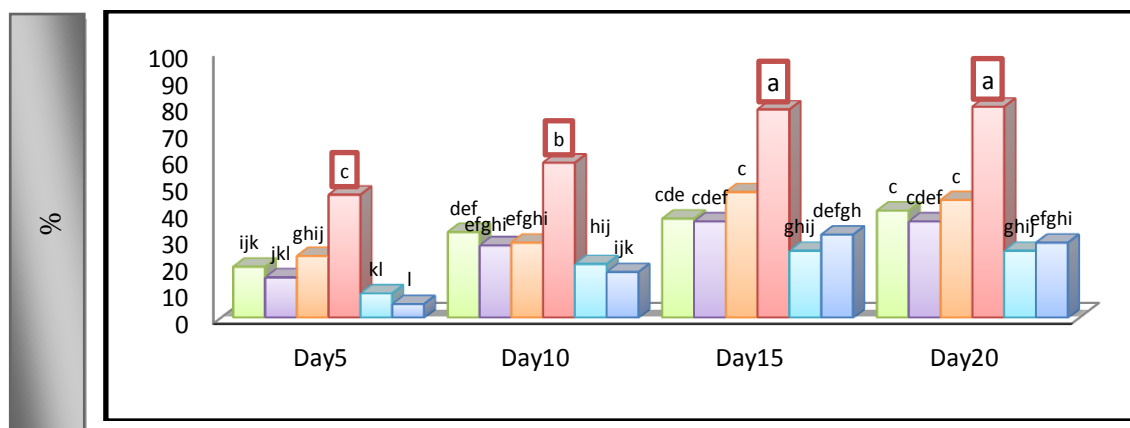


Figure1. Effect of calcium chloride, thyme essential oil and storage time on weight loss of sapodilla fruit.

**Fruit firmness**

Maximum firmness was recorded in combination 2%CaCl<sub>2</sub> and thyme essential oil 500 $\mu\text{l/l}$ . Maximum firmness was recorede in during 5<sup>th</sup>.The lowest firmness was recoreded in control treatment(Fig 2).

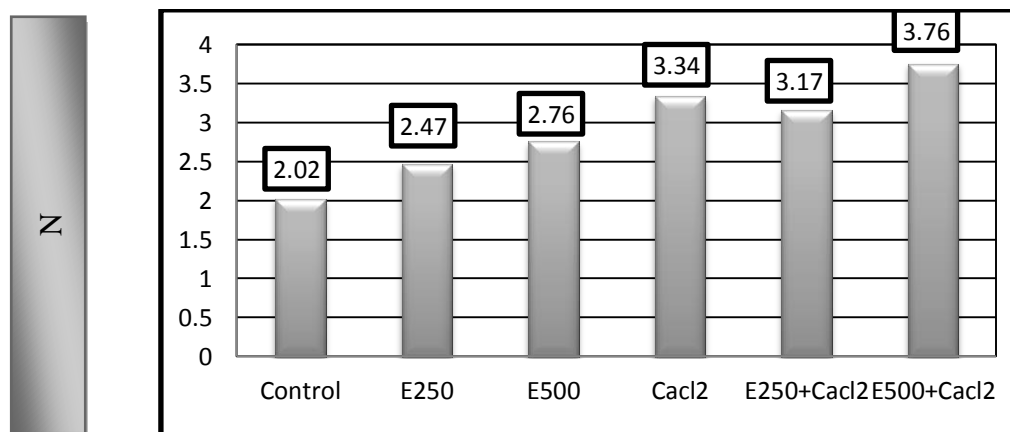


Figure2. Effect of calcium chloride, thyme essential oil and storage time on fruit firmness of Sapodilla fruit.

**Ph, Tss,Ta And Tss/Ta**

The no statistically significant differences were observed in pH,TA,TSS and TSS/TA.

**MDA**

Maximum malodialdehyde(MDA) was recorded in control treatment. The lowest MDA was recorded in 2%Cacl2 and combination 2%Cacl2 and thyme essential oil 250µl/l treatment. MDA was highest during the 15th and 20<sup>th</sup> days.

The cell membrane plays an important role in maintaining cell structure and function of the post-harvest fruits and vegetables. Two key indicators of cell membrane integrity are permeability and malonaldehyde (MDA) content.

During the storage time, much free radical in the cell accumulates and harms to cell membrane owing to the destruction of equilibrium mechanism between production and elimination of the radical.

Lipid peroxidation is an oxidation process of unsaturated fatty acid through free radical action. The reaction produces lipid peroxide that is poisonous to cell. MDA, one of final products in lipid peroxidation reaction, can severely harm cell membrane. MDA content reflects the active status of free radical of post-harvest fruit and vegetable. The more the MDA content is, the higher the level of free radical such as ·OH and O<sub>2</sub><sup>-</sup>.

Malondialdehyde was measured as an indicator to estimate the amount of damage to the cell membrane. The result suggest that using 2%Cacl2 alone or in combination thyme essential oil 250µl/l is to protect cell membrane from postharvest damage.

**Fungal infections**

The results showed essential oil is used in both concentration plays an important role in controlling fungal contamination of fruits. In fact , thirty days of storage was observed in control fruit and fruit treated with 2%Cacl2 had the highest infection rate, but none symptoms of fungal contamination was observed in essential oil treatment and combine essential oil and 2%Cacl2.

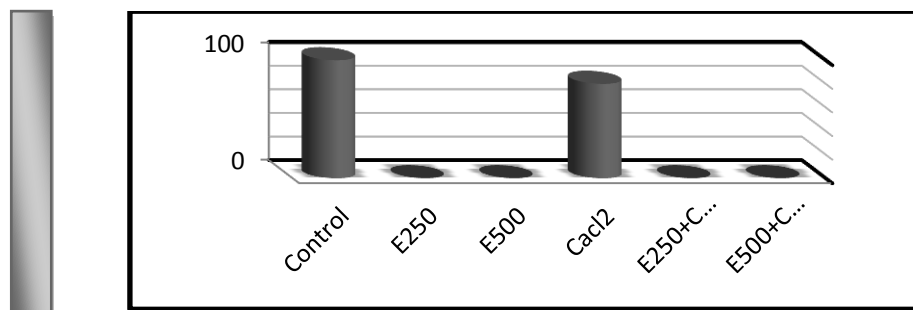


Figure3. Effect of calcium chloride and thyme essential oil on Fungal infections of Sapodilla fruit.

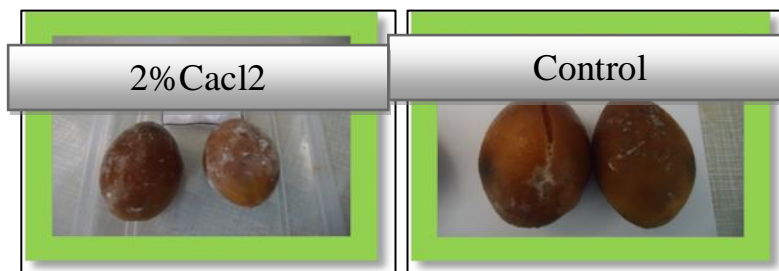


Figure4. Symptoms of fungal contamination of samples in storage for thirty days.

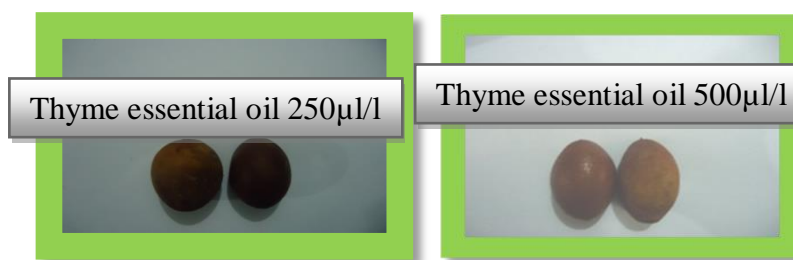


Figure5. Absence fungal contamination of samples in storage for thirty days.

The use essential oil in the control of postharvest diseases of fruits and vegetables has been introduced as a biological in recent years and to be an effective and safe at the same time to have drawn the attention of many researchers. Using essential oils can also decrease waste product health and safety of the fruit. Susceptibility of fungal species is varies, depending on the concentration of essential oils. Differences in antifungal activity of essential oils depends on their composition. The combination may be cause alone or in combination with other antifungal activity of essential oils. (Plotto; et al .,2003 ).

(Moskoki and Mortazavi, 2004), was useful the effect of thyme and Ammi oil for control *Aspergillus parasiticus* on pear and also other research showed that the *Salvia* and *Artemisia* not good efficacy on four pathogenic fungi such as *Botrytis cinerea* on postharvest fruit.

The spray thyme essential oil of the orange contaminated with *penicilium* to controlled and the effect is no different with fungicide TBZ.

#### **Panel test**

The effect of treatments on fruit appearance, color, sweetness, aroma, firmness and finally the acceptance rates of fruit sapodilla is significant at the 1% level.

The best appearance of the fruit on the 2%CaCl<sub>2</sub> and combined 2%CaCl<sub>2</sub> and concentration 250 µl/l essential oil and control. The wrost appearance of the treated fruit by concentration 500µl/l eessential oil.

The best color of the flesh has been treated with calcium chloride, and other treatments were not significantly different from the color of the flesh.

The highest of acceptability rates of fruits on the 2%CaCl<sub>2</sub> and combined 2%CaCl<sub>2</sub> and concentration 250 µl/l essential oil and control and the concentration 250 µl/l essential oil, 500µl/l essential oil and the combination 500µl/l essential oil and 2%CaCl<sub>2</sub> decreased acceptability.

The essential oil treatments can cause adverse effects on fruit appearance which was not accepted by consumers, if using essential oil with low concentration and in combination with calcium chloride appeared better preserved fruit the consumer accepts it.

The essential oil similar any chemical substance when used in high concentrations can cause burns on the surface of the fruit. The results correspond (Asghari Marjanlu; et al., 2008). They reported that when used basil oil at high concentrations (500 and 1000 µl/l) sepals of the burn area of strawberry fruits.

Quality is the most important factor in determining the market value of appearance. As people see the shop have learned from past experience that the quality of visual characteristics associated with some of their favorite. Evaluation of rapid eye of appearances through the experience and criteria of size, shape, condition (such as being fresh) damages done. Since the appearance of the product is the most important index to evaluate product in market, it is better if you use essential oils to be used as the minimum concentration of essential oils no adverse effect on product appearance. Use of essential oil alone can cause adverse effects on

fruit flavor and the sweetness of the fruit overshadowed, especially "The effects when used with a high concentration be higher.

## CONCLUSIONS

The results of this study and related research studies it seems that the use of calcium chloride treatment can have a positive effect on parameters such as fruit firmness and malondialdehyde were reduced, resulting in lower membrane lipid degradation occurs. The color treated fruits maintained. Combination treatment calcium chloride with low concentrations essential oil (250 µl/l) weight loss can reduce and increase fruit firmness. The decreased malondialdehyde and desirable appearance is preserved fruits. Using a combination of calcium chloride and essential oil in high concentration(500µl/l) led to the highest fruit firmness However, the use of essential oil at high concentrations (500 µl/l) alone causes skin discoloration and somewhat flesh, and create an unpleasant odor, taste the fruit, fruit. All concentrations of essential oil alone or in combination with calcium chloride completely inhibited the growth of fungi on fruit. Essential oil can replace chemical fungicides in controlling pathogenic fungi important to the Sapodilla fruit. However, given that the treatments affected fruit flavors are essential oils therefore, the adoption of fruit by consumers is difficult .However, efforts should be made using appropriate methods to be used at a concentration of essential oils has negative effects on fruit quality will be minimized.Based on the obtained results it can be inferred the essential oil of low concentrations with or without the presence of calcium chloride can replace chemical fungicides in the control of pathogenic fungi on fruit Sapodilla and thereby increase its shelf life. However, to achieve the proper concentration of these essential oil further research is needed.

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