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Full Length Research Paper

The effect of physical treatments on control of *Penicillium digitatum* decay orange cv. Valencia during storage period

Sajad Fatemi* and Hassan Borji

Faculty of Agriculture, Islamic Azad University, Khorasgan (Esfahan) Branch, Isfahan, Iran.

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The aim of this study was to assess the effect of hot water treatments on some of the quality characteristics and control of green mold decay orange cv. Valencia during storage period. The research was conducted in a completely randomized design with 26 treatments and 3 replications at two levels of inoculation and un-inoculation. Treatments included: hot water (45, 50, 55 and 60°C) with the duration (1, 2 and 3 min), control treatment. Fruits were sprayed with suspension solution green mold. 24 to 48 h post-inoculation through immersion in hot water fruits were treated. Following drying, (lapse of 12 h), each treatment was separately packed in plastic bags and along with two control treatments (1- immersion in spore-free distilled water and 2- distilled water containing spore) was transferred to cool storage (temperature 6°C, relative humidity 85 to 90%). Results showed that for all characteristics, un-inoculated treatments had significant difference. Therefore, at harvest and post-harvest, the pathogenic triangle (temperature, environment and the pathogenic agent) were controlled, so that damage due to *Penicillium* infection was minimized.

Key words: Citrus, hot water, fungal decay, storage life, fruit quality.

INTRODUCTION

For growth of micro organisms in fruits and vegetables, the most suitable conditions of the environment include high moisture, temperature and foodstuff (Aidoo, 1991). Green mould and blue mould, which are caused by *Penicillium digitatum* and *Penicillium italicum*. These moulds are seen and observable in every citrus garden and the predominate factors of decaying. The most important damage causing factor in postharvest period is of *Penicillium,* that imposes billions of dollars damage (Pitt, 1981). Citrus fruits, due to their high vitamin C content are very crucial in human nutrition. Post-harvest quality of citrus fruits is influenced by physiologic and pathogenic factors. Post-harvest rots are the main limiting factors in storing horticultural products. Especially, *Penicillium* is one of the major factors limiting storage life of orange. Consumers' knowledge on harm effects of using chemicals to control diseases, pests, and physiologic damages of fruits has augmented.

Therefore, the need for developing effective substances and in the meantime safe for preserving health of horticultural products has made it necessary to apply non-chemical methods to control post-harvest rots. One of the most harmful factors of orange rot in chill-house is usually green mold appearing 5 to 12 weeks post-storage (Cheach and Irvng, 1997). In 1992, for the first time, hot water treatment was employed to control rot on citrus fruits. Pre-storage application of hot water treatment for a short time (about a few minutes), is only effective on pathogenic agents found on external layers of fruit skin (Fallik et al., 1997). The results showed that floating of fruits in hot water 55°C for 2 to 3 min caused the control of green mould and improved the post harvest quality in tangerine (Smilanick et al., 2008). The heat treatments can be an appropriate alternative in stowing Valencia oranges with artificial fungicides (Williams et al., 1999; Nanes et al., 2007). Results showed that hot water treatment (45°C) for 150 s, in addition to controlling rot, to a great level improves the fruit cortex (Larrigaudiere,

*Corresponding author. E-mail: sajad_6689@yahoo.com.

2002). Immersing fruits in not water for 3 min in a temperature of 52°C significantly decreased green mold and fruit loss and also preserved fruit firmness and ascorbic acid content (Alemzadeh and Feridon, 2007).

In the treatment of hot water bruising which is a modern procedure used for post-harvest control of fruit damage, which considerably decreased the green rot resistance in inoculated grape fruits (cv. Star Ruby) (Pavoncello, 2001). Immersion of oranges inoculated with *P. digitatum* in 75°C hot water for 150 s decreased infection by 90% compared with controls (Palou, 2001). Results obtained from studies has shown that immersion of fruits in 55°C hot water for 2 and 3 min and 50°C for 3 min had the best effect in controlling and delaying activity of the tangerine green mold activity (Inkha, 2009). Also, immersing fruits in 60°C hot water for 1 min decreased activity of green mold fungus, clemenules variety compared with controls at 95% (Montesinos-Herrero, 2009). Thermal treatments can be a good substitute for disinfecting Valentia oranges instead of synthetic fungicides (Nanes, 2007). This study was conducted with the purpose of investigating the effect of various thermal levels of water on various factors causing rot in storage and effectiveness mechanism of these treatments on qualitative and quantitative indices of fruit as well as slowing down the trend of green mold development in the chill house.

MATERIALS AND METHODS

Fruits were provided from an orangery in Jahrom-Fars Province. Fruits were divided into two inoculation and un-inoculation categories. Then, through boring a 1 to 2 mm hole in the wound location of joining point of fruit to stalk, the fruits were sprayed with suspension solution containing green mold isolated and purified at a concentration of 1×10^5 at the 24 to 48 h post –inoculation, through immersion in hot water (45, 50, 55, and 60°C) for periods of 1, 2 and 3 min samples were treated. After drying (lapse of 12 h), each of the treatments were packaged separately in plastic bags and along with two control treatments (immersion in spore-free distilled water and immersion in spore containing distilled water) were transferred to the chill-house (with 6°C and 85 to 90% relative humidity). At the end of the third month, appraised parameters including fruit rot percentage, percentage of total soluble solid (TSS); vitamin C percentage and total acidity (TA), pH and decreased weight loss of fruits were evaluated. Fruit rot percentage was determined through separating polluted treatments from healthy fruits and counting them.

To determine fruit weight loss decrease, healthy fruits remained of each replication, were counted and weighed with a scale of 1/100 precision. Then, the secondary weight that remained of healthy fruits, was deducted from their primary weight and obtained figures were added up and averaged. pH was determined with hand-digital pH-meter and to measure percent percentage of total soluble solid (TSS), the hand refractometer was used and results were expressed in Breaks degree. To determine total acidity (TA) according to (A.O.A.C) methods, amount of 10 ml fruit extract was titrated with 0.3 N sodium hydroxide (NaOH) in the presence of Phenolphthalein and expressed as a percent of citric acid according to (A.O.A.C) methods. Vitamin C level was assessed with titration according to (A.O.A.C) methods. All obtained data was analyzed using SPSS software and means were compared using Duncan test at the 5% probability level.

RESULTS AND DISCUSSION

Effects of treatments on fruit rot percentage

Effect of various treatments on fruit rot percentage in both inoculation and un-inoculation cases showed a significant difference at 5% level. In inoculated fruits, the highest fruit rot percent was that of the inoculated control fruits (100%) and the least rot level was relevant to non-inoculated treatments. No doubt hot water treatments in the non-inoculation groups were effective as in fungicide treatments. Also, in inoculation group 3, 60°C hot water treatments performed better compared with other hot water treatments (Table 1).

Effect of treatments on fruit weight loss

Effect of treatments on fruit weight loss % in both inoculation and non-inoculation cases, showed a significant difference at 5% level. The highest weight loss was that of the inoculated 45°C samples treated for 2 min (6.0867%) and the least amount was that of the 45°C sample treated for 3 min (0.17%) in un-inoculated fruits (Table 1).

Effect of treatments on solute percentage

As a whole, no significant difference was seen between inoculated and un-inoculated treatments. The highest level of solutes was that of the 60°C inoculation treatment for 3 min (11.9167%) and the least level was that of the 45°C inoculated sample treated for 2 min (9%) (Table 1).

Effect of treatments on total acid percent

Results showed that the highest total acid content was that of the 60°C sample treated for 1 min (1.806%) in the un-inoculated group and the least acid level was seen in the inoculated group treated in 45°C hot water for 1 min (1.053%) (Tables 1 and 2).

Effect of treatments on vitamin C levels

Effect of various treatments on percentage of fruit weight loss in both inoculation and non-inoculation cases, showed a significant difference at 5% level. The least vitamin C level was seen in the inoculated control treatment and the highest vitamin C level was seen in the non-inoculated samples treated with 50°C hot water for 3

Table 1. Mean comparison levels hot water treatment on characteristics quality and quantity in the fruits inoculation and un-inoculation.

Treatment	ROT (%)	Weight loss(%)	VIT C(%)	TSS(%)	TA (%)	PH
HW 45° 1 min Inoculation	49.99^{cd}	4.81^{efg}	47.29^{bcd}	10.58^{cde}	1.053^{b}	3.39^{bcd}
HW 45°2 min Inoculation	66.66	6.09^{g}	46.74^{bc}	9^{a}	1.20^{bc}	3.37^{bcd}
HW 45°3 min Inoculation	49.99^{d}	4.85^{efg}	47.71 b	10^{c}	1.55^{ef}	3.41^{bcd}
HW 50° 1 min Inoculation	57.22^{cd}	5.111^{fg}	47.50^{bcd}	11.17^{defg}	1.23^{bc}	3.38^{bcd}
HW 50° 2 min Inoculation	44.433^{c}	5.27^{fg}	48.24^{bcde}	10.58^{cde}	1.28^{c}	3.22^{bc}
HW 50° 3 min Inoculation	55.57^{cd}	5.02^{efg}	45.62^{bc}	10.75^{cdef}	1.34^{cd}	3.37^{bcd}
HW 55 c° 1 min Inoculation	38.89^{bc}	5.35^{fg}	48.03^{bcd}	10.67^{cdef}	1.22^{bc}	3.53^{d}
HW 55 c°2 min Inoculation	22.22^{ab}	4.49^{efg}	44.52^{bc}	11.08^{cdefg}	1.25^{c}	3.48^{cd}
HW 55 c° 3 min Inoculation	22.22^{ab}	4.58^{efg}	50.02^{bcdef}	10.33^{cd}	1.41^{cdef}	3.53^{d}
HW 60 c° 1 min Inoculation	22.22^{ab}	3.60^{defg}	45.48^{bc}	10.83^{cdefg}	1.61^{f}	3.96^{e}
HW 60 c° 2 min Inoculation	22.22^{ab}	3.07^{bcdef}	45.34^{bc}	10.83^{cdefg}	1.39^{cd}e	4.19^{ef}
HW 60 c°3 min Inoculation	16.66^{a}	3.27^{cdef}	51.84^{bcdefg}	11.92^{g}	1.31^{cd}	4.21^{ef}
Control Inoculation	100^{a}	100^{h}	0^{a}	0^{a}	0^{a}	0^{a}
HW 45 c° 1 min un inoculation	5.55^{a}	0.76^{abc}	59.06^{efgh}i	11^{cdefg}	1.34^{cd}	3.20^{bc}
HW 45 c°2 min un inoculation	0^{a}	0.23^{a}	58.06^{defghij}	10.67^{cdef}	1.41^{cdef}	3.22^{bc}
HW 45 c° 3 min un inoculation	0^{a}	0.18^{a}	62.50^{ghij}	10.83^{cdefg}	1.33^{cd}	3.27^{bcd}
HW 50 c°1 min un inoculation	0^{a}	0.32^{ab}	67.2467^{ij}	11.58^{efg}	1.51^{def}	3.18^{b}
HW 50 c°2 min un inoculation	0^{a}	0.67^{abc}	67.5067^{ij}	10.58^{cde}	1.38^{cde}	3.27^{bc}
HW 50 c° 3 min un inoculation	0^{a}	0.63^{abc}	69.51 ^{j}	10.67^{cdef}	1.40^{cd}e	3.42^{bcd}
HW 55 c° 1 min un inoculation	0^{a}	0.21^{a}	62.93^{hij}	10.58^{cde}	1.30 ^{c}	3.43^{bcd}
HW 55 c° 2 min un inoculation	0^{a}	0. 80^{abc}	52.42^{cdefg}	10.83^{cdefg}	1.28^{c}	4.30^{f}
HW 55 c°3 min un inoculation	0^{a}	0.77^{abc}	60.2^{ghij}	10.5^{cde}	1.26^{c}	3.47^{bcd}
HW 60 c° 1 min un inoculation	0^{a}	1.04^{abcd}	47.31^{abc}	10.5^{cde}	1.8^{g}	3.47^{bcd}
HW 60 c° 2 min un inoculation	0^{a}	1.39^{abcd}	51.61^{cdefg}	10.5^{cde}	1.42^{cdef}	4.26^{f}
HW 60 c° 3 min un inoculation	0^{a}	1.72^{abcd}	52.1^{defg}	11.5^{efg}	1.28^{c}	4.17^{ef}
Control un inoculation	16.66^{a}	2.31^{abcde}	52.03^{cdefg}	11.75^{fg}	1.27^{c}	3.43^{bcd}

Means with common letters in the same column are not significantly different at $p<0.05$. HW: hot water; Vit C: vitamin c; TSS; total soluble solids; TA: total acidity Rot: rot decay.

Table 2. Analysis of variance of table of treatments on decay control decay and characteristics quality, quantity in the orange "Valencia" .

Source	Mean square						
	df	ROT	VIT C	TA	TSS	PH	W loss
Treatment	25	2212.618**	514.029**	0.276**	14.378**	1.863**	1105.670**
Error	52	133.808	36.673	0.012	0.325	0.023	2.103

**Significant in the level 5%, *no significance.

min (69.51%) (Tables 1 and 2).

Effect of treatments on pH

As a whole, no significant difference was seen between inoculated and un-inoculated treatments. The highest pH was observed for higher temperatures and the lowest level was seen in both inoculated and un-inoculated cases (Tables 1 and 2).

Conclusion

Today, due to environment pollutions and fungicides, dangers on fruits threaten the health of consumers. It is highly recommended that hot water treatments should be replaced with chemical materials. Treatment of floating fruits in hot water through direct inhibition of pathogenic agent and promoting special defensive reactions have been known as methods to control post-harvest diseases. Part of the efficacy of hot water is due to its potential to remove spores from the wounds and also the direct effect of high temperature on the pathogenic agent. No doubt, stimulating resistance in fruit cortex can play a role as well. During the past decades, benefit of most of the chemical treatments due to their efficacy, low cost and easy application has been documented. Recently however, many factors have contributed to decreased dependence on chemical compounds. Generally, fruits which underwent un-inoculation hot water treatments from the aspect of all characteristics were superior to inoculated ones.

The most crucial disease prevalent in orange is green and blue molds (*P. digitatum, P. italicum*), and thermal treatments can be used to control them (Ben-Yehoshua, 1989; Inkha, 2009). In this research project, the immersion in hot water treatment significantly decreased the level of fruit infection by *Penicillium* species, a finding which correspond with those obtained by other researchers like Ben-Yehoshua (1989), and GanjiMoghadam and Rahemi (1995). Thermotherapy treatment had no negative effect on oranges, since it decreases the respiration rate of fruit and also preserves the level of fruit sugar and solutes, a finding that corresponds with those of Shahbake (1991). As a whole, it is probable that the reason for preserving qualitative characteristics of fruits in un-inoculated treatments and partially in inoculated treatments is that plastic bags decrease the rate of water loss and minimized water condensation on fruit surface and also make possible exchange of such gases as oxygen, carbon dioxide and ethylene and respiration activity continues without being affected by used storage temperature, and through decreased water loss, fruit remains more consistent and brighter and due to decreased transpiration, fruit senescence is also delayed and as a result taste, shape an brightness of fruit is maintained for a longer time. Weight loss causes vitamin C loss which might be due to increased oxidation resulting from water decrease. 55 and 60°C hot water treatments were identified as best treatments for preventing fruit infection.

Inhibiting fruit contamination (to pathogens) is considered as a general principle for all control methods either physical, chemical or using essential oils. All factors preparing conditions for growth and development of pathogenic agents should be eradicated. As the general conclusion, use of thermal treatment due to ease of application and being free of risks due to fungicide residues are of special importance and also leave beneficial effects on fruit. Therefore, regarding the effect of hot water treatments in decreasing post harvest losses and to increase internal quality of fruit, application of such treatments can be recommended.

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