INTEGRATED CONTROL OF *LASIODIPODIA* ROT ON SWEET ORANGE FRUITS USING HOT WATER- FUNGICIDE TREATMENT

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**ABSTRACT**

Integrated control of *Lasiodiplodia* rot on sweet orange fruits using hot – water fungicide treatment was investigated. Sweet orange fruits inoculated with *Lasiodiplodia* sp were drenched separately in hot water mixed with 500 ppm benlate in a water bath at the following hot water treatments: 35, 48, 50, 53 and 55°C for 3 minutes each, 35°C for 20 minutes, 45°C for 15 minutes and 48°C for 10 minutes. There were two sets of control fruits. The first control (control 1) fruits were not drenched, while the second control fruits (control 2) were drenched with unheated water containing 500 ppm benlate. Each treatment was in four replicates. All fruits were placed in sterilized Petri dishes and stored in an atmosphere of 95–100% relative humidity at 28±2°C in sterilized desiccators and assayed daily for disease severity. Results showed that there was significant decay control of fruits drenched in hot water benlate treatment. Mean disease rating for the first seven days of storage were 1.00 ±0.00 for fruits treated at 35°C for 3 and 25 minutes, 45°C for 15minutes, 48°C for 3 and 10minutes each, 50°C for 3minutes and 53°C for 3minutes. Hot water benlated drench treatment at these temperature-time combinations consistently suppressed diseased development by day seven of storage with no sign of decay in treated fruits compared with the control treatments. As storage duration progressed to day 14, only benlated fruits without hot water and fruits drenched at 35°C and 48°C for 3 minutes each had no sign of decay with each treatment having mean disease rating of 1.00. By 21st day of storage, only fruits drenched at 48°C for 3minutes had mean disease rating of 1.00 and showed no sign of infection. This showed that Hot water - benlate drench treatment controlled *Lasiodiplodia* rot on the orange fruits.

**Keywords:** *Citrus sinensis*, *Lasiodiplodia* sp., fungicide, benlate, drench

**INTRODUCTION**

Orange fruits have several beneficial health and nutritive properties (Economos and Clay, 1999). They are rich in Vitamin C and Folic acid, as well as a good source of fibre. They are fat free, sodium and cholesterol free (Economos and Clay, 1999). In addition, they contain vast number of minerals. They may help to reduce the risk of heart diseases and some types of cancer. In short, citrus fruit is enjoyed around the world for its taste, nutritional value and relatively cheap price. However, one of the greatest problems encountered during post harvest storage of sweet oranges is infection by microorganisms.
Researchers have found that control of disease infection in orange fruits could be achieved by post-harvest control practices such as waxing, controlled harvest, controlled atmosphere and good packaging (Kuku, 1985). Babarinsa et al. (2002) worked on the storage of Valencia oranges in brick wall evaporative cooler using the principle of control atmosphere to reduce decay in orange fruits. Pre storage treatment by irradiation has also been reported (Schirra and Maurizio, 1995, Markson et al., 2005). In addition, seal packaging of individual orange fruit using film wrapping before cold storage or prolonged shipment was being practiced (Economos and Clay, 1999).

Although these post-harvest control practices effectively control disease infections in orange fruits, their setbacks cannot be overlooked. For instance, polyethylene film and waxing of fruit with coating if too thick may become so impervious and excessively impede oxygen influx or carbon dioxide efflux thereby unduly interfering with aerobic respiration and induce onset of anaerobic respiration which lead to physiological disorders and a shortened life. More so, synthetic waxes and detergents are not edible and may be hazardous to health if consumed. The other methods are also sometimes injurious to personnel and consumers in addition to high cost and physiological damage to crops. Currently, postharvest diseases are primarily controlled by application of fungicides. Fungicide application is one of the most important steps for successful control of post-harvest decay of Valencia oranges (Grierson and Ben-Yehoshua, 1986). Imazalil and thiabendazole are used in aqueous solution before waxing or in the water emulsion wax to control stem – end rot, green and blue moulds effectively. Also, Smilanick et al. (2003) reported that citrus fruits are generally treated with fungicides to control post-harvest infections. Since postharvest rot is the major factor limiting the extension of storage life of many freshly harvested fruit and vegetables, this investigation was therefore, undertaken to study the integrated control of Lasiodiplodia sp. IMI 503248 which result in substantial post-harvest losses of oranges using hot water – fungicide treatment.

**MATERIALS AND METHODS**

**Isolation of Lasiodiplodia sp. IMI 503248 from Infected Fruit**

The Lasiodiplodia sp. IMI 503248 isolate used was obtained from decayed oranges. The organism was identified based on the structural features as seen on culture plates as well as microscopic characteristics after staining with lactophenol in cotton blue using the method of Markson et al. (2005). This involved cutting out the interface between the healthy and the disease tissue and placing pieces of the affected fruit rind without surface sterilization on plates already containing solidified malt extract agar (MEA). The plates were then benlate incubated at 28±2°C for 10 days, before carrying out morphological and pathological characterizations of the isolate.
Source of Fruits and Preparation
Mature, green apparently healthy orange fruits were harvested from a commercial orchard in Igbatoro, Akure North, Nigeria (7° 51’ 3N; 5° 21’ 46E). Fruits of uniform size and colour were selected. Before treatment, the fruits were washed with fresh water, disinfected superficially for 1 minute in 10% sodium hypochlorite, rinsed with water and allowed to air-dry at room temperature. A ten day old agar slant culture of Lasiodiplodia sp used for inoculum was obtained by inoculating the isolate on already prepared malt extract agar slants. Sterile water was poured into the slant bottle and shaken vigorously to dislodge the spores from the vegetative hyphae. The wash water was collected in a sterilized beaker. One milliliter of the suspension was spread on an area of 1cm² and allowed to dry on a clean microscope slide before counting using the high dry (x40) objective. The spore concentration of the suspension was determined by using the formula of Breed Direct Counting Technique:

\[ \text{Number of spore m}^{-1} = \frac{\text{Average number of spores} \times \text{MCF}}{\text{(about 50 fields)}} \]

\[ \text{MCF} = \frac{\text{Microscope correction factor} = \frac{\text{Area of smear} \times \text{dilution factor}}{\text{Area of Microscope field}}} \]

Area of microscope field = \( \pi r^2 \)

Hot Water Fungicide Treatment
Inoculated fruits were drenched separately in hot water mixed with 500 ppm benlate in a water bath at the following effective and ineffective hot water treatments previously investigated: 35, 48, 50, 53 and 55°C for 3 minutes each, 35°C for 20 minutes, 45°C for 15 minutes and 48°C for 10 minutes. There were two sets of control fruits. The first control (control 1) fruits were not drenched with hot water -benlate treatment, while the second control (benlate only) fruits were drenched with ordinary water containing 500 ppm benlate. Each treatment was in four replicates. All fruits were placed in sterilized Petri dishes and stored in an atmosphere of 95 – 100% relative humidity at 28±2°C in sterilized desiccators and assayed daily for disease severity.

Assessment of Rot Severity
All fruits were placed on sterilized Petri dishes in sterilized desiccators supplying 95% relative humidity at 28 ± 2°C and assessed daily for disease severity using the scale of Arekemase and Oyeyiola (2007) but with slight modification where 1=disease free, 2=slightrots/decay up to 25% of the fruit, 3 = moderate rots /decay up to 50% of the fruit, and 4 = severe rots/fruit completely rotten. Rot / decay was recognized by light brown discolouration on the fruit.
RESULTS
Effect of Hot Water - Fungicide Treatment on Disease Severity of Inoculated Fruits
There was significant decay control of fruits drenched in hot water treatment. Mean disease ratings for the first seven days of storage were 1.00 ±0.00 for fruits treated at 35°C for 3 and 25 minutes, 45°C for 15 minutes, 48°C for 3 and 10 minutes each, 50°C for 3 minutes and 53°C for 3 minutes (Figure 1). Hot water benlate drench treatment at these temperature-time combinations consistently suppressed disease development by day seven of storage with no incidence of decay in treated fruits compared with the control treatments (Figure 1). As storage duration progressed to day 14, only benlate treated fruits without hot water and fruits drenched at 35°C and 48°C for 3 minutes each showed no disease severity with each treatment having mean disease rating of 1.00 (Figure 2). Fruits drenched at 45°C for 15 minutes and 53°C for 3 minutes showed slight disease symptoms as indicated by their mean disease ratings of 1.25 while fruits drenched at 35°C for 20 minutes and 48°C for 10 minutes both had 4.00 as mean disease severity and showed complete decay (Figure 2). By 21st day of storage, only fruits drenched at 48°C for 3 minutes had mean disease rating of 1.00 and showed no sign of infection while control and those drenched at 35°C for 3 and 20 minutes each, 48°C for 10 minutes and benlate treated fruits without hot water, all had mean disease ratings of 4.00 (Figure 3). The fruits were completely decayed.

DISCUSSION
Hot water –benlate drench treatment controlled Lasiodiplodia rot on the orange fruits. However, ordinary benlate drench treatment without hot water performed comparatively better in decay control than the benlate hot water drench treatment.

Most of the hot water drench treatments had shown disease symptoms and complete decay on the treated fruits by 14th day of storage while benlate treated fruits only showed no disease symptoms. This finding with ordinary benlate treatment only disagreed with the earlier view of Schirra and Maurizio (1995) that fungicide was effective in disease and disorders control, its efficacy being greatly enhanced when used in hot solution. However, it is in agreement with the work of (Cohen, 1981) who reported that a fungicide (metalaxyl) used in aqueous preparations alone when applied as a drench effectively controlled fungal decay in citrus fruit than when it was drenched with hot water at 45°C. However, the effectiveness of all the benlate treatments quickly declined, to the extent that by 28th day of storage, all the fruits has shown complete decay. This ineffectiveness of the benlate treatment in extending the shelf life of treated fruits during prolong storage may not be unconnected with the report of Ismail and Zhanj (2004) that the fungicides commonly used in citrus packinghouses such as thiabendazole, imazalil, benlate, fluconale against blue and green moulds have no long-term effect against the rot fungus inside the fruit.
CONCLUSION
Currently, postharvest diseases of perishables are primarily controlled by good sanitation and application of fungicides. It has been reported that fungicides such as thiabendazole, imazalil and guazatine have been used to control post harvest fungal diseases of oranges. Interestingly, this study has equally shown that benlate at 500 ppm drench with hot water is effective for integrated control of Lasiodiploidial rot on orange fruits.

Figure 1: Effects of Hot Benlate Drench Treatment on mature green sweet orange fruits inoculated with *Lasiodiplodia* sp. IMI 503248 after storage at 28±2°C for 7 days. The temperature of treatment in °C and minutes of exposure are given as hyphenated figures. I = Stad. deviation
Figure 2: Effects of Hot Benlate Drench Treatment on mature green sweet orange fruits inoculated with *Lasiodiplodia* sp. IMI 503248 after storage at 28±2°C for 14 days. The temperature of treatment in °C and minutes of exposure are given as hyphenated figures. I = Stad. deviation

Figure 3: Effects of Hot Benlate Drench Treatment on mature green sweet orange fruits inoculated with *Lasiodiplodia* sp. IMI 503248 after storage at 28±2°C for 21 days. The temperature of treatment in °C and minutes of exposure are given as hyphenated figures. I = Stad. Deviation

REFERENCES


