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A sequential treatment with sodium hypochlorite and a reduced dose of imazalil heated at 50 °C effectively control decay of individually film-wrapped lemons stored at 20 °C

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A B S T R A C T

Storage of individual seal-packaged citrus fruit at room temperature in China is a very common practice that requires a pre-storage treatment with a high concentration of an effective fungicide as imazalil (IMZ) to prevent decay.

In this study, lemons were washed with NaOCl (200 mg L^{-1}) or not, treated with IMZ (50 or 1000 mg L^{-1}) at 20 or 50 °C and individually wrapped with a 16-μm thick extensible polyvinylchloride film (Film A) or two heat shrinkable polyolefinic films, thick 15 (Film B) or 19-μm (Film C).

The sequential treatment with NaOCl and IMZ at 50 mg L^{-1} at 50 °C, was as effective as IMZ at 1000 mg L^{-1} at 20 °C in controlling *Penicillium* decay. Losses for decay in fruit wrapped with the two more permeable films (Film A and Film B) never exceeded 10%, while in those wrapped with Film C (the least permeable) peaked to 41%. All films reduced weight losses, which at the end of storage were 11% in fruit wrapped with Film A and below 4% in those wrapped with the other two films, while were 41% in unwrapped ones. After one week of storage, only 50% of unwrapped fruit were marketable whereas all wrapped fruit were still marketable after 8 weeks. Respiration as well as juice acetaldehyde and ethanol were slightly affected by the two more permeable films, while an abnormal production of CO_2, acetaldehyde and ethanol occurred in those wrapped with film C. Changes in chemical parameters were relevant in fruit sealed with Film C and minor in those with Film A and Film B.

Decay control and quality preservation of lemons stored at room temperature can be achieved for several weeks by a sequential treatment with NaOCl and a heated water emulsion of IMZ at 50 mg L^{-1}, when fruit are wrapped with plastic films highly permeable to gases.

1. Introduction

Lemons are rarely eaten as stand-alone fruit, but are widely used for their juice as well as a natural remedy to prevent and cure a wide range of diseases (Ladaniya, 2008). The flavedo is rich in phenolics (mainly flavonoids) and other nutrients and non-nutrients compounds (vitamins, minerals, dietary fiber, essential oils and carotenoids), with health preventing diseases activity — obesity, diabetes, blood lipid, cardiovascular disorders and certain types of cancer — (González-Molina et al., 2010; Aruoma et al., 2012).

China imports of fresh lemons has markedly increased in the last ten years doubling imports (FAOSTAT http://faostat3.fao.org/browse/T/TP/E). Thus, there is a great interest to increase production and improve storage technologies of lemons. In China most of the product is still handled and sold directly by growers, who normally pick the fruit at the optimal maturity stage and store them in sheds located within the orchards at ambient temperature even for several months, before being sold in local markets. Therefore storage conditions could be un-predictable and far from being optimal and even if fruit in most cases maintain their chemical and eating quality, a high percentage soon becomes

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unmarketable due to transpiration and decay mainly due to *Penicillium digitatum* (Pers.: Fr.) Sacc. and *P. italicum* Wehmer infections.

Storage conditions usually consist of dipping the fruit in imazalil (IMZ) emulsion at concentrations of 1000–2000 mg L^{-1} and closing them individually in plastic bags of different materials. At these concentrations, IMZ residues (whole fruit weight) are within the legal maximum levels (MRLs) in many countries (Ritenour, 2016). Nevertheless, despite the flesh is practically residue-free the use of the peel can be problematic if fungicides exceed the MRLs.

It is therefore important to develop effective postharvest treatments based either on non-fungicidal chemistry or to drastic reduction of IMZ concentrations.

Among the different methods used to decrease infections, immersion of the fruit in hot water for 2–5 min at 45–53 °C could be effective to control various decay-causing pathogens including Penicillium decay (Houck, 1967; Schirra et al., 2011; Palou, 2013). Hot water, at some extent, may indirectly reduce or delay pathogen growth by completely or partially closing the cuticle fractures which represent an important entry avenue for pathogens and removing part of the epiphytic microflora (Ben-Yehoshua and Porat, 2005). It can also increase the host resistance (*i.e.* expression of heat shock proteins, accumulation of inhibitors of pathogens growth) (Usall et al., 2016) or improve the efficacy of biocontrol agents (Zhou et al., 2014). Hot water may also inactivate mycelial growth by killing hyphae on fruit surface but it is not able to damage those inside the tissue and has a limited activity on spores, whose susceptibility depends on their metabolic activity, water content, position on fruit surface and temperature (Ben-Yehoshua and Porat, 2005). Moreover, while germinating spores are easily killed, dry spores are only partially damaged and generally resume their growth in a few days. However, heat treatments do not leave residues and therefore have no protective activity, so new spores carried by contaminated air can infect the fruit causing decay.

IMZ, the most common fungicide in citriculture, is very effective against Penicillium decay (*P.digitatum* and *P. italicum)* showing curative, preventive and antisporulant activity (Erasmus et al., 2015a, 2015b; Kellerman et al., 2016). IMZ curative activity is very high against sensitive strains of both Penicillium species even at very low concentrations, especially when applied as heated emulsion. In addition, it imparts a protective and antisporulant action lasting for the whole storage time, which can be several months, if applied at concentrations of 1000–2000 mg L^{-1} (D'Aquino et al., 2013).

Sodium chloride is worldwide used as a sanitizing and disinfecting agent. It is able to kill spore of pathogens on citrus fruit surface by contact, but it does not act as systemic fungicides and does not have any protective activity (Feliziani et al., 2016).

In wrapped fruit, the film can affect decay positively or negatively. Restricted diffusion of gases could create anaerobic conditions and water condensation, especially if the film is not well fixed to the fruit, favoring pathogens' growth. In addition, anaerobic conditions alter fruit quality. On the other hand, the physical barrier of the film prevents the contact of adjacent decayed fruit avoiding secondary infections.

The objectives of this study were: 1) to evaluate the efficacy of integrated heated treatments with a sanitizing agent as NaOCl and low levels of IMZ, in order to control lemons decay stored at 20 °C and 55–60% RH; and 2) to evaluate the impact of plastic films with different permeability characteristics on decay incidence and fruit quality.

2. Materials and methods

2.1. Fruit

Mature, fully colored yellow lemons cvs Femminello Siracusano and Zagara Bianca were collected from the experimental station of the Institute of Sciences of Food Production (Central-west Sardinia, Italy) and transported to the laboratory within four hours from harvest.

In the laboratory fruit were selected and homogeneous fruit were dipped in a 10^4 conidia mL^{-1} suspension of a *P.digitatum* strain sensitive to IMZ and left to incubate for 20 h in a storage room at 20 °C and 95% RH before treatments, to simulate natural infections occurring during harvest and transportation on fruit surface.

The strain of *P. digitatum* used, a single-spore isolate collected from a decayed clementine from the same experimental station, was cultured on potato dextrose agar (PDA, 39 g L^{-1} in distilled water, autoclaved; Merck and Co., Whitehouse Station, NY) amended with streptomycin sulfate and oxytetracycline hydrochloride (100 + 100 μg m L^{-1}) to prevent bacterial growth.

2.2. Effect of hot water dipping and IMZ heated emulsion on decay incidence

Two thousand 'Femminello Siracusano' lemons were divided into 5 lots of 400 hundred fruit each and each lot was immersed for 3 min into one of the following treatments: a) water at 20 °C (pH 7.8); b) 50 mg L^{-1} IMZ (pH = 7.84) (DECCOZIL 50, 44.66% ai, Decco Italia, Piano Tavola Belpasso, Catania, Italy) at 20 °C; c) water at 50 °C (pH = 7.85); d) 50 mg L^{-1} IMZ at 50 °C (pH = 7.82);e) 1000 mg L^{-1} IMZ at 20 °C (pH = 7.55). Fruit were left to dry at ambient temperature for two hours, than each lot was divided into 4 sub-lots of 100 fruit, of which one was left unwrapped (control), while the other three were wrapped with: 1) Film A, an extensible polyvinylchloride 16-μm-thick (Omny film, Erre-Ci-A, Monza, Italy); 2) Film B, a 15-μm thick shrinkable polyolefinic film (Bolphane BY, Bolloré, Ergué-Gabéric, France); 3) Film C, a 19-μm thick shrinkable polypropylene film (Opti 320, Sealed Air, Foggia, Italy), whose barrier properties are reported in Table 1. Film A was wrapped but not sealed using a wrapping machine for extensible films (Hand Wrapper SW-500E, Lovero, Gyeonggi-do, Korea), while Film B and Film C were sealed and heat-shrank by a wrapping machine for heat-shrinkable films (SE 850, Tecnopack Equipment-Livorno, Italy). Fruit of each combined treatment were then divided into four plastic boxes (25 fruit per box) and stored at 20 °C and 55–60% RH for 8 weeks. Decay incidence was assessed at weekly intervals and expressed as the cumulative percentage of decayed fruit from all replicated boxes.

Table 1
Barrier properties of the used plastic films and method used for determination.

Film	O_2 permeance[a] (μM s^{-1} m^{-2} kPa^{-1})	CO_2 permeance (μM s^{-1} m^{-2} kPa^{-1})	Water Vapor Transmission Rate (μg s^{-1} m^{-2})
A	0.181 (ASTM D-1434)	1.833 (ASTM 1434)	3703 (ASTM E-96)
B	0.051 (ASTM D-1434)	0.199 (Factory method)	324 (ASTM E-96)
C	0.015 (ASTM D-3985)	0.039 (COV-E68)	92.6 (ASTM F 1249-90)

[a] Data provided by the producers were transformed in units required by the SI.