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Internal fruit damage in table olive cultivars under superhigh-density hedgerows

María Rocío Jiménez*, Laura Casanova, María Paz Suárez, Pilar Rallo, Ana Morales-Sillero

Departamento de Ciencias Agroforestales, ETSIA, Universidad de Sevilla, Carretera de Utrera, km 1, 41013 Sevilla, Spain

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ABSTRACT

This study describes fruit damage of the Manzanilla de Sevilla and Manzanilla Cacereña cultivars under super high-density conditions at morphological and histological levels. Fruits were harvested both manually and by a grape straddle harvester. Dark spots and tissue ruptures were found throughout the mesocarp of both cultivars, particularly after mechanical harvesting. Quantitative traits previously described by our group were used to evaluate internal damage. 'Manzanilla de Sevilla' always showed the highest total damaged area in the mesocarp, as well as the highest sum of the areas of all of the tissue ruptures, and number of tissue ruptures intersected by the second circumferential arc. Tissue ruptures in the exocarp were also observed in this cultivar as well as a general increase in this type of damage over time (from 2 to 24 h after harvesting). Investigation of the cuticle and epidermal cell dimensions in undamaged fruits shows that 'Manzanilla Cacereña' presents a thicker cuticle and a greater cuticle area per epidermal cell than 'Manzanilla de Sevilla'. The relationship between these results and the different bruising susceptibilities of the two cultivars studied is discussed.

1. Introduction

Mechanical harvesting is necessary in many fruit crops to reduce high production costs. This is particularly true for olive orchards, in which hand harvesting may represent more than 60% of the total cost. Currently, different types of harvesters (trunk shaker, over-row, etc.) are frequently used in olive orchards for oil production (Gil-Ribes et al., 2009; Ferguson et al., 2010; Zion et al., 2011), but not for table olive production. The main disadvantage in this case is the high percentage of fruits damaged by the impact (commonly called bruising) that occurs during harvesting (Castro-García et al., 2015).

The olive fruit is a drupe consisting of a thin external protective layer named the epicarp or exocarp (skin), a fleshy mesocarp (also called the pulp or flesh), and the endocarp (the stone or pit; King, 1938). Bruising damage has been observed in both the exocarp and mesocarp layers. Dark spots appear on the exocarp of the fruit a few minutes after harvest and progress throughout the mesocarp (Castro-García et al., 2010; Jiménez et al., 2016), so that the commercial value of the fruit dramatically decreases (Riquelme et al., 2008) particularly for table olive consumption. The cultivar is one of the most significant determining factors for bruising susceptibility. In this sense, 'Manzanilla de Sevilla,' the leading table olive cultivar grown worldwide (Barranco, 2008), is highly susceptible to bruising damage (Jiménez et al., 2016). For this reason, Rejano et al. (2008) recommended an immediate immersion of fruits in a cold diluted solution of lye (NaOH) after harvest with trunk shakers to stop bruising progress.

Superhigh-density hedgerows (*SHD*) were developed in the 1990s for olive oil production, and they currently occupy over 100,000 ha worldwide, 45,000 ha of which are in Spain (López, 2015). These orchards are characterized by high tree density, usually more than 1500 trees per hectare. One of the main advantages of these type of orchards is the low cost of mechanical harvesting. Straddle harvesters adapted from grape cropping are usually used, and most fruits are removed in no more than two or three hours per hectare. It is assumed that low-vigor and early bearing cultivars with low alternance production are better adapted to superintensive conditions (Connor et al., 2014).

Recently, Morales-Sillero et al. (2014) studied the suitability of 'Manzanilla de Sevilla' and 'Manzanilla Cacereña', a double use cultivar (table olives and olive oil) grown mainly in Spain and Portugal (Barranco et al., 2005), to be harvested by a straddle harvester in an orchard with a density of 1.975 trees ha^{-1}. This work showed that super-high density production is a possible alternative for table olives. Hedgerows of both cultivars were continuous, highly productive (~10.000 kg ha^{-1} and 18.000 kg ha^{-1} for 'Manzanilla de Sevilla' and 'Manzanilla Cacereña' hedgerows, respectively), and efficiently harvested; almost of the fruits were removed (98%) in two hours or less. Olive fruits were harvested at the green maturity stage, and no fruit loosening agent was applied. Concerning fruit damage, both cultivars

* Corresponding author.
E-mail addresses: rjg@us.es (M.R. Jiménez), laucaler@us.es (L. Casanova), maripaz@us.es (M.P. Suárez), prallo@us.es (P. Rallo), amorales@us.es (A. Morales-Sillero).

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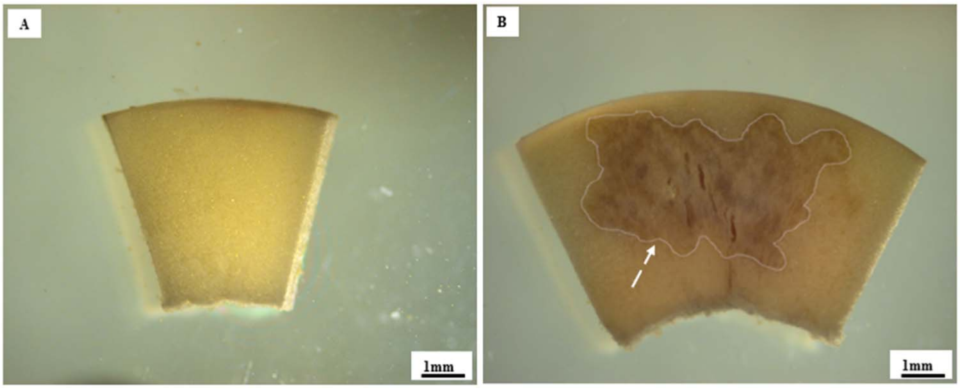

Fig. 1. Fixed mesocarp portions obtained prior to histological procedure. Undamaged tissues (A) were used as control. The total damaged area (*TDA*, mm^2) (— →) was determined in damaged tissues (B) with an image analysis system connected to the binocular microscope.

showed different susceptibility to bruising: 'Manzanilla de Sevilla' fruits showed the highest proportion of damage in both the hand-picked and mechanically harvested fruits. Approximately 89% of fruits of the latter method showed damage over more than 1/4 of the skin after mechanical harvesting in contrast to 'Manzanilla Cacereña' (16% of fruits). However, despite the high percentage of fresh damaged fruits reported, particularly interesting was the low proportion of bruised fruits ($\leq$3%) once the fruits were processed as Spanish-style green olives, regardless of the cultivar.

The damage to fruits harvested by grape straddle machines is probably related to the direct contact with the rods, which radiate from a vertical cylinder, extend into the canopy and rotate to shake shoots and small branches to remove fruits. Connor et al. (2014) suggested that the firmness of fruits and the thickness of skin (exocarp) are related to the level of damage. However, no differences in the firmness of 'Manzanilla de Sevilla' and 'Manzanilla Cacereña' fruits were found before harvesting in the aforementioned study. Hammami and Rapoport (2012) also suggested that the different susceptibility to bruising among olive cultivars might be related to the skin thickness.

The skin of the olive comprises the cuticle, a thin extracellular layer that covers the surface that is composed of an insoluble polymer named cutin and waxes that represent 45–70% of the skin weight (Bianchi, 2003). Under the cuticle there is a layer of cells called the epidermis, and also a subepidermal region, with 1–4 cell layers with similar characteristics as the epidermis (Hammami and Rapoport, 2012). Both the cuticle and epidermis thickness have been related to prevention against biotic and abiotic external factors (Hong et al., 2008; Vichi et al., 2016). Hammami and Rapoport (2012) reported that both tissues are thinner in 'Manzanilla de Sevilla' fruits than in fruits of 'Hojiblanca', a double-aptitude cultivar with low susceptibility to bruising damage (Jiménez et al., 2016). On the other hand, it has been suggested that internal damage, and also the loss of quality in the extracted oils, may be explained by different physiological responses of fruits after harvest by grape straddle machines (Yousfi et al., 2012; Morales-Sillero et al., 2015; Morales-Sillero and García, 2015). Internal damage in fruits could also explain the loss of quality in oils from fruits harvested by trunk-shakers in intensive conditions (Dag et al., 2008). However, to our knowledge no study has described internal damage in fruits harvested by any type of harvester.

A recent methodology developed by our group based on different traits of olive fruit portions and histological sections of the mesocarp allows an exhaustive description and quantification of bruising damage in the fruits (Jiménez et al., 2016). The aim of this work was first to explore, following the aforementioned methodology, the type and severity of the internal damage associated with bruising in 'Manzanilla de Sevilla' and 'Manzanilla Cacereña' fruits harvested both by hand and by a grape straddle harvester in a super-high density orchard. Second, we aim to clarify if the different thickness of the skin explains, at least in part, the different response to damage of each cultivar after mechanical harvesting.

2. Material and methods

2.1. Plant material

Fruits of 'Manzanilla de Sevilla' and 'Manzanilla Cacereña' were harvested in September 2012 in a five year old orchard located near Campo Maior (Portugal; long. 38°56′N; lat. 7°02′W; altitude 201 m). Trees were planted in a standard layout of 3.75 by 1.35 m (1975 trees ha^{-1}), in a north-south orientation under irrigation and non-limiting nutrient conditions. Orchard management is fully described in Morales-Sillero et al. (2014).

2.2. Experiment design and fruit harvest

The trial was designed taking three (replicate) random rows of trees (approximately 90 trees/row) per cultivar ('Manzanilla de Sevilla' and 'Manzanilla Cacereña') and per harvesting treatment (hand and mechanical).

Fruits were harvested at the green ripening stage, when they reached maturity index = 1 (Ferreira, 1979). Samples of 5 kg fruits were taken after hand and mechanical harvesting. Fruits from hand harvesting were manually picked from 12 trees randomly chosen per row. Mechanical harvest was performed by means of a grape straddle harvester (Model VX 7090; CNH Global, Belgium) with a travel speed of 3.5 km h^{-1} and 480 beats per minute.

2.3. Fixation process and internal damage quantification in tissue portions prior to histological procedure

Damaged fruits from each cultivar and replicate were fixed for their preservation at 2 and 24 h after harvest (both, hand and mechanical) in an FAE solution according to Berlyn and Miksche (1976). Two mesocarp portions of each fruit from undamaged (control; Fig. 1A) and damaged areas (Fig. 1B), were cut according to the methodology described by Jiménez et al. (2016). Photographic images of all fixed portions of mesocarp were taken with a Nikon Digital camera (Sight DS Ri 1) connected to a binocular loupe Nikon (Nikon, SMZ 1270) and processed with Nis-Elements AR 3.2 image analysis software (Fig. 1). A total of 480 photographic images were obtained: 10 samples per cultivar (Manzanilla de Sevilla and Manzanilla Cacereña), harvesting treatment (hand and mechanical), time after harvest (2 and 24 h), type of tissue portion (damaged and undamaged) and three replicates.

To quantify the extent of internal damage, the total damaged area (*TDA*, mm^2) in the mesocarp portions was measured according to the methods described in Jiménez et al. (2016) with software (Nikon NIS-ELEMENTS AR 3.2) connected to the binocular microscope. This area was easily distinguished by its darker discoloration in the damaged zone of the mesocarp (Fig. 1B).