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Genotypic differences in sweet cherries are associated with the susceptibility to mechanical damage

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The rheological properties and structure of the flesh tissue at harvest-ripe fruit of the sweet cherry *Prunus avium* L. cultivars Bing, Lapins, Regina, Santina, Sweetheart and Van were measured using a textural analyzer equipment and described by light microscopy, respectively. The rheological measurements inflicted reproducible levels of mechanical damage (pitting). The structural and rheological properties of the mesocarp and epidermis correlate with their susceptibility to mechanical damage. Epidermal cell width and area of external mesocarp cells were negatively associated with susceptibility to mechanical damage while cell number, quantified in 1 mm^2 of external mesocarp, was positively associated. Fruit of Bing and Regina are the least susceptible to mechanical damage among the cultivars under study. The least sensitivity of Regina fruit was associated with its wide epidermal cells and high value of modulus of elasticity, while in Bing with its low number of cells as well as high values of stress and strain at bioyield point. The high susceptibility to mechanical damage of Sweetheart and Lapins fruit was associated with their large number of cells in the external mesocarp tissue and with the lowest values of strain at bioyield point, therefore with the lowest deformation capacity of the tissue.

1. Introduction

The sweet cherry (*Prunus avium L.*) is a highly-valued, highly-seasonal product of cool-temperate climates. It is a premium product that commands high prices at market. The fruit pedicel (stalk) should be fresh and green and the fruit firm, shiny and of brilliant mahogany color. The fruit should be free of visible defects, especially cracking and bruising. All these make the product attractive to the consumer and govern the value. Unfortunately, not only are sweet cherries especially subject to postharvest deterioration due to dehydration and decay, but the fruit skin is very susceptible to surface pitting and this often becomes the critical quality criterion even after quite short periods of storage.

Pitting develops when impact or compression injury occurs; skin depressions overlie necrotic lesions in the fleshy mesocarp (Wade and Bain, 1980), making sweet cherries very susceptible to mechanical bruising. Laboratory and field studies have shown that visible symptoms of surface pitting are not usually evident at harvest but develop during the first 10 days of storage at 0 °C (Porritt et al., 1971). Pitting affects not only the cosmetic aspects of the fruit but it also shortens shelf life and reduces market quality at point of sale. Mitchell et al. (1980) found increasing respiratory activity in proportion to the severity of injury. This also leads to premature decay and softening during storage (Owaga et al., 1972).

Several factors explain the high variability of the tissue to the damages in different years, for different cultivars (Kappel et al., 2006; Toivonen et al., 2004) and even for different trees of the same cultivar (Porritt et al., 1971). In a survey, Facteau and Rowe (1979) found negative correlations between pitting incidence and fruit weight and soluble solids concentration. On the other hand, pitting sensitivity increases as fruit temperature decreases (Crisosto et al., 1993; Facteau, 1982; Facteau and Rowe, 1979; Grant and Thompson, 1997; Lidster and Tung 1980; Porritt et al., 1971; Zoffoli and Rodriguez, 2014).

In general, the texture of a fruit tissue is a function of the mechanical properties of its cell walls, of the cells themselves – cell turgor and cell:cell bonding involving the middle lamella (Harker et al., 1997; Vanstreels et al., 2005). A fruit is composed of different types of cells. These may be loosely or tightly bound, possess thinner or thicker walls, which are more or less elastic, being comprised of cellulose and other polysaccharides (Reeve, 1970). The mechanical properties of a plant material, such as a fruit, are the physical characteristics related to deformation, disintegration and flow of the fruit under a force, and are measured objectively by function of

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mass, time and distance (Blahovec, 1999; Bourne, 1982). According to Abbott (1999) under mechanical loading, fruit and vegetables exhibit viscoelastic behavior, which depends both on the amount of force applied and also on the rate of loading. A fruit can exhibit different mechanical behaviors under uniaxial compression or tensile loading. Force/deformation methods are widely used for objective measurement of the textural properties of solid biomaterials. Recently, biaxial tensile tests of excised fruit skin, simulating the growth stresses and strains occurring in vivo have also been used to characterize the mechanical properties of the sweet cherry cultivars (Brüggenwirth and Knoche, 2016a,b). Mechanical properties of fruit tissue are proved to be influenced by turgor, temperature and ripening stage of the fruit (Brüggenwirth and Knoche, 2016a).

During ripening tissue fracture changes from involving principally failure of the cell walls to failure of cell:cell bonding (Hallett and Harker 1998; Harker et al., 1997). Another important factor having a major influence on tissue strength is the internal pressure of the cells (turgor) (De Belie et al., 2000; Lin and Pitt, 1986). Cell turgor does not affect the tensile strength of soft tissue, as it has been demonstrated in ripe sweet cherry fruit (Knoche et al., 2014) but it has a profound influence on the tensile strength of unripe tissue, as has been demonstrated in pear fruit (De Belie et al., 2000).

A plant tissue's susceptibility to physical damage is an intrinsic characteristic of that tissue. Thus, fruit tissue morphology seems to explain the bruising susceptibility observed in apples. Whose large intercellular airspaces of the outer parenchyma weaken it because of the associated reduction in cell:cell contact area (Alvarez et al., 2000; Mitsuhashi-Gonzalez et al., 2010). This makes apples vulnerable to damage by external forces. Irregularities in tissue structure also weaken the cellular matrix making it more vulnerable to damage. Comparisons between a number of different plant tissues and genotypes (carrot, celery, cucumber and apple) under the same external loads demonstrated that carrot had the toughest tissue. This toughness was attributed to its small parenchyma cells and lack of intercellular spaces (Alvarez et al., 2000). Pierzynowska-Korniak et al. (2002) found that different apple cultivars have different fruit parenchyma cell sizes and shapes, which also contribute to their unique sensory characteristics. The apple is a pome, its parenchyma cells and intercellular spaces are loosely arranged in a net–like pattern which is anisotropic (non-homogeneous in the three orthogonal directions) and this arrangement has been attributed to cultivar sensitivity to mechanical damage (Khan and Vincent, 1990). The mechanical properties of fruit flesh have also been described for other species. Konstankiewicz and Zdunek (2001) reported a relation between the mechanical properties of potato tissue and the intracellular pressure and cellular structure.

In contrast, say, to apples, sweet cherry is a drupe with a hard endocarp (pit), a stomatous exocarp (skin) and an edible mesocarp (flesh) formed from the ovary wall. They are harvested when they are fully ripe and show distinctly different contents of cell wall material associated with firmness texture (Choi et al., 2002; Salato et al., 2013) these may explain the markedly different physical characteristics that result in their very different susceptibilities to surface pitting and other problems such as fruit cracking.

Sweet cherryís hypodermal cells are described as small compared to the mesocarp cells and with thicker walls, mesocarp cells have tangential elongation near the epidermis and radial elongation near the pit (Glenn and Poovaiah, 1989). The epidermis is a single layer of cells covered by a thin cuticle, which is continuous except where interrupted by stomata (Bukovac et al., 1999; Knoche et al., 2000, 2001; Tukey and Young, 1939). The mechanical properties of the skin are influenced by the epidermis and the hypodermis (Brüggenwirth et al., 2014). The strained exocarp holds the mesocarp and endocarp under gentle compression (Grimm et al., 2012). The epidermis and the hypodermis are the structural components of the skin supporting the mechanical properties of the mesocarp as it expands (Brüggenwirth et al., 2014; Grimm et al., 2012).

Table 1
Fruit maturity parameters of sweet cherry cultivars at harvest.

	[1]Soluble solids	[1]Firmness	[1,2]Color	
Cultivar	(%)	(0–100)	Chroma (C)	Hue (°)
Bing	23.9 d	81.5 c	25.7 b	14.3 b
Lapins	18.3 ab	72.9 a	28.3 b	16.9 b
Regina	20.4 c	81.1 c	17.7 a	9.8 a
Santina	16.8 a	75.0 ab	25.4 b	16.5 b
Sweetheart	19.9 bc	77.7 bc	29.5 b	16.7 b
Van	19.9 bc	75.9 ab	25.1 b	16.1 b
P	<0.0001	0.0032	0.0002	<0.0001

[1] Mean of five replicates of 50 fruit per year. Values followed by the same letter were not significantly different according to the Fisherís least significant difference (LSD) test ($P \leq 0.05$).

[2] Average color was described at the lab according to the Commission International de líEclairage (CIE), with a Minolta colorimeter (Minolta, CR-400, Japan).

Phenotypic characterization of sweet cherries involving both microscopy and rheological studies of the genetic resources available are required if breeders are to improve the handling properties of sweet cherries, to increase their resistance to surface pitting.

The objective of this study was to evaluate the available genetic material of sweet cherries with a view to identify if there are tissue structures and/or rheological features that may explain or correlate with susceptibility to mechanical damage. The focus of this study was to characterized cultivars of firm flesh to demonstrate that even in those cultivars, differences in their rheological properties and cell characteristics are influencing its susceptibility or resistance to mechanical damage, so cultivars of extreme soft flesh were avoided.

2. Material and methods

2.1. Fruit material

The study was conducted in sweet cherry (*Prunus avium* L.) fruit from orchards located in central and south-central Chile, over two growing seasons, 2012/2013 and 2013/2014. Fruit of six sweet cherry cultivars, Bing, Lapins, Regina, Santina, Sweetheart and Van were harvested from mature trees of moderate vigor. Two orchards per cultivar were harvested in year-1 and, to increase the variability, three more orchards per cultivar were added in year-2. Orchards with similar agronomic practices, in term of crop load, gibberellic acid application rate were selected. The cultivars were harvested at commercial maturity – optimal for long-term storage (Zoffoli et al., 2006), considering red and dark mahogany colors, resembling color number 4 and 5 according to CTIF color chart (Centre Technique Interprofessionel des Fruit et Legumes, Paris, France) respectively.

For the evaluations, groups of around 100–128 fruit were harvested by hand from the exterior of the canopy, of each cultivar, from each orchard. The selected fruit were free of visible defects and were uniform in diameter (26–28 mm).

Fifty fruit from each orchard, of each cultivar were used to characterize the maturity parameters at harvest (Table 1). Fruit firmness was measured within a range from 0 (soft) to 100 (firm) using a durometer (type A, Durofel, Agro-technologie, Tarascon, France). Soluble solids concentration (%) was measured using a digital thermo-compensated refractometer (PAL-1, Atago Co. Ltd, Tokyo, Japan). Fruit diameter (mm) was measured using digital calipers (model 500-196, Mitutoyo Corp., Kawasaki, Japan). Skin color was measured on each cheek. Average color was described at the lab according to the Commission International de líEclairage (CIE), with a Minolta colorimeter (Minolta, CR-400, Japan) (Table 1). The fruit were harvested at pulp temperature between 20 and 23 °C and transported to the lab in a 5 °C portable cooler. Then, the fruit