



An insight into the role of fruit maturity at harvest on superficial scald development in 'Beurré D'Anjou' pear



Gabriela Calvo^a, Ana Paula Candan^a, Marcos Civello^b, Jordi Giné-Bordonaba^c, Christian Larrigaudière^{c,*}

^a INTA Alto Valle, Postharvest, CC 782 (8338), General Roca, Argentina

^b INFIVE (CONICET-UNLP), Diag. 113 y 61 (1900), La Plata, Argentina

^c IRTA Postharvest, PCITAL, Parc de Gardeny, Edifici Fruitcentre, 25003 Lleida, Spain

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ABSTRACT

To assess the influence of the fruit maturity at harvest on superficial scald incidence, 'Beurré D'Anjou' pears were harvested on January 24 (H₁); February 7 (H₂) and February 21 (H₃). Ethylene production, α -farnesene (AF), conjugated trienols (CTols) content and total antioxidant capacity (DPPH) were determined at harvest and during storage and further related to superficial scald (SS) development. Early picked fruit (H₁) had significantly lower scald incidence than H₂ or H₃. The difference in scald sensitivity between harvests was associated to the capacity of the fruit to produce ethylene and to the accumulation pattern of AF and CTols, which in turn was not exclusively ethylene-dependent. Collectively the results presented herein showed that the relationship between scald and maturity in 'Beurré D'Anjou' pears may be opposite to that observed in apples. Changes in the fruit antioxidant capacity during storage rather than the initial antioxidant potential at harvest should be considered to fully understand the biochemical basis of superficial scald in 'Beurré D'Anjou' pears.

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1. Introduction

Superficial scald is one of the main physiological disorders affecting postharvest quality of pears and apples leading to great economic losses worldwide (Wang and Dilley, 1999; Lurie and Watkins, 2012; Giné Bordonaba et al., 2013). This disorder, which manifests as brown or dark patches on the fruit skin, affects the fruit appearance and makes the fruit unsuitable for sale as a fresh commodity (Emongor et al., 1994).

Superficial scald development in apples and pears has been intimately associated with the accumulation of α -farnesene oxidation products within the fruit, the conjugated trienols (CTols) (Huelin and Murray, 1966; Anet, 1972; Whitaker, 2007; Whitaker et al., 2009). In apples, ethylene is known to play a key role in the development of superficial scald since this hormone is involved in the synthesis of α -farnesene by regulating gene expression of hydroxymethylglutaryl-CoA reductase (HMGR) and/or α -farnesene synthase (Ju and Curry, 2000). Accordingly, scald-susceptible pears and apples typically exhibit higher rate of

α -farnesene (AF) than resistant fruit, shortly after they are placed in low temperature storage (Huelin and Coggiola, 1970; Whitaker et al., 1997; Lurie et al., 2005; Gapper et al., 2006; Giné Bordonaba et al., 2013).

Given that superficial scald is considered to be the result of an oxidative process, it is accepted that superficial scald severity is proportional to the degree of oxidation of AF and that the disorder should not appear during storage if the fruit maintains sufficient antioxidants to prevent or limit AF oxidation. Thus, the balance between the content of oxidative species and antioxidants within the fruit skin has an important role in the development and progression of superficial scald in both apples and pears (Ju et al., 1996; Rao et al., 1998; Diamantidis et al., 2002; Zubini et al., 2007; Whitaker et al., 2009; Silva et al., 2010). Low temperature storage weakens the antioxidant mechanisms of plant tissues (Mir et al., 1999) including pears (Chiriboga et al., 2013) hence making the fruit more prone to develop this physiological disorder. Research about the biochemical mechanisms responsible for the development of scald have suggested that apples and pears follow a similar pattern (Rowan et al., 2001; Isidoro and Almeida, 2006; Whitaker, 2007; Whitaker et al., 2009; Zubini et al., 2007). Although such similarities are well documented, there are some differences in ethylene induction and antioxidant content that could result in differences in

* Corresponding author.

E-mail address: christian.larrigaudiere@irta.cat (C. Larrigaudière).

the fruit sensitivity to superficial scald between these species. Most apple varieties produce ethylene after harvest, but many pears, including 'Beurré D'Anjou', generally require exposure to low temperatures to initiate its autocatalytic ethylene production (Abeles et al., 1992). Moreover, it is well accepted that pears typically have a lower content of antioxidants than apples (Leontowicz et al., 2003).

Maturity and harvest date have a significant influence on scald development in apples, with greater susceptibility to this disorder consistently found in earlier harvested fruit (Huelin and Murray, 1966; Anet, 1972; Chen et al., 1985; Wang and Dilley, 1999). This behaviour has been associated both with a lower activity of the antioxidant system in less mature fruit (Barden and Bramlage, 1994a,b) but also to faster kinetics of accumulation of oxidation products (Anet, 1972). In this way, delayed harvest is commonly used as a preventive measure to control superficial scald in highly sensitive apple varieties. However, the relationship between maturity and scald susceptibility has been poorly documented in pears, and seems to be less predictable than in apples (Salunkhe and Desai, 1984; Zoffoli et al., 1998; Whitaker et al., 2009).

Accordingly, the aim of this study was to clarify the existing relationship between fruit maturity at harvest and scald development in 'Beurré D'Anjou' pears. For this purpose, the changes in ethylene production, α -farnesene, conjugated trienols and antioxidant capacity during storage of fruit from different maturities were related to superficial scald incidence.

2. Materials and method

2.1. Plant material and storage conditions

'Beurré D'Anjou' pears (*Pyrus communis* L.) were harvested from a commercial orchard located in Alto Valle (39° 01' S, 67° 44' W), Rio Negro, Argentina from 10 years old trees, planted at 4 × 2 m, on seedling rootstock trained in a modified Trellis system. The experiment was carried out in 2013 with fruit harvested at different stages of maturity: January 24 (H₁); February 7 (H₂) and February 21 (H₃). Fruits were transferred immediately after each harvest to the Postharvest Laboratory of INTA Alto Valle. Fruits of uniform size and free from defects were packed in cardboard boxes with two trays (20 fruit each) covered with non-perforated low density polyethylene liners of 30 μ m and stored in regular air at -0.5°C and 95% RH for 240 days.

2.2. Maturity determinations

Fruit quality parameters at harvest were determined at each date on 5 replicates of 10 fruit each. Flesh firmness (N) was measured using a fruit texture analyzer (Model FTA-GS14, Güss Manufacturing Ltd., Strand, South Africa), with an 8 mm diameter plunger. Two measurements were obtained per fruit on opposite sides, after removal of 2 mm of peel. Soluble solid content (SSC) and titratable acidity (TA) were determined using freshly prepared juice from each individual fruit. SSC (%) were measured using a temperature-compensated digital refractometer (Pal1, Atago, Japan) and TA (g L^{-1}) expressed as malic acid content was measured by titrating 10 mL juice with 0.1 N NaOH to an endpoint of pH 8.2, using a calibrated pH meter. Fruit surface colour was determined with a colorimeter (CR-400, Minolta, Japan) using the CIE L* a* b* colour space coordinates. Hue angle was calculated as $\text{tg}^{-1}(b^*/a^*)$. Starch degradation (%) was determined by comparison to specific tables for this variety (INTA) after inserting a fruit slice of 1–1.5 mm from the equatorial zone in a lugol solution and the percentage of starch degradation was then determined. Fruit weight (g) was determined with an electronic precision balance.

2.3. Ethylene

Ethylene production was measured in 5 replicates of one fruit each from each harvest after 0, 15, 30, 60, 90, 120, 150, 180, 210 and 240 days at -0.5°C and for 30 days shelf life at 20°C . Each fruit was sealed in 1.5 L airtight jar for 30 min. Gas samples of 1 mL were extracted with a syringe from the air headspace. The sample was analyzed with a gas chromatograph (GC14-A, Shimadzu, Japan) equipped with an FID detector, an activated alumina column and injector operating at 240°C , 40°C and 110°C respectively. Helium was used as carrier gas. Ethylene production curves were drawn to determine the "delay" (days) as the time required for the fruit to begin producing values higher than $1 \text{ nL g}^{-1} \text{ h}^{-1}$ and the "climacteric" (days) as the time required to reach maximum production of ethylene.

2.4. Superficial scald

Superficial scald was visually assessed following 120, 180, 210 and 240 days at -0.5°C and 7 days on 5 replicates of 10 fruits. The incidence was expressed as the percentage of fruit affected and the severity was classified according to the following scale: Grade 1 (very slight): less than 25% of the total surface area stained; Grade 2 (mild): more than 25% and less than 50%; Grade 3 (moderate): more than 50% and less than 75%; Grade 4 (severe): over 75%. The superficial scald index (SSI) (Pesis et al., 2009) was calculated:

$$\text{SSI} = \frac{\sum (\text{severity grade}) \times (\text{number of fruit per grade})}{\text{Total number of fruit}}$$

2.5. α -farnesene (AF) and conjugated trienols (CTols)

Determinations of AF and CTols were performed following the method described by Anet (1972), with some modifications, in 5 replicates of 10 fruits each, after 0, 15, 30, 60, 90, 120, 150, 180, 210 and 240 days at -0.5°C . A strip of peel of 2 mm thick was removed along the equatorial zone of the fruit. A 10 mm diameter discs were removed from the peel using a cork borer. Then, five discs excised from each fruit were immersed in 10 mL of HPLC grade hexane for 10 min with constant stirring. Then, 1 mL of this solution was diluted in 4 mL of hexane.

Measurements were performed calibrating first the equipment with HPLC grade hexane. Absorbance at 232 nm (α -farnesene) and 281–290 nm (conjugated trienols) were then recorded using a UV-spectrophotometer (1001 Plus, Milton Roy, USA). Concentrations of α -farnesene and conjugated trienols were calculated using the molar extinction coefficients $E_{232 \text{ nm}} = 27,700$ for α -farnesene and $E_{281-290 \text{ nm}} = 25,000$ for conjugated trienols (Anet, 1972) and expressed as nmol cm^{-2} of fruit.

2.6. Total antioxidant capacity (DPPH)

Total antioxidant capacity was determined using the DPPH test. Measurements were performed on 5 replicates of 10 fruits each, after 0, 15, 30, 60, 90, 120, 150, 180, 210 and 240 days at -0.5°C . The entire peel of the fruit was removed, frozen with liquid nitrogen, lyophilized and ground to a fine powder and then homogenized with 10 mL of 80:20 methanol:water (v/v). Samples were then left for 2 h at room temperature in a constant shaking bath and then centrifuged at 20°C (24,000 g) for 15 min. The supernatant obtained was then filtered and diluted with milli-Q water (1:4; v/v). After diluting the samples, a 20 μ L aliquot of the diluted extract was then mixed and stirred with 980 μ L 1-diphenyl-2-picrylhydrazyl (DPPH; Sigma-Aldrich, Steinheim, Germany) in the dark for 30 min at 4°C .

Table 1Indexes of full maturity and antioxidant capacity (DPPH) of pears 'Beurré D'Anjou' harvested on 24 January (H₁); February 7 (H₂) and February 21 (H₃).

Harvest	Firmness (N)	SSC (%)	AT (g/l)	Colour (Hue)	Starch (%)	Weight (g)	DPPH (%)
H ₁	71.2 a	10.4 b	4.84 a	118.5 a	12.5 b	158.7 b	19.4 b
H ₂	63.2 b	11.0 a	3.44 b	118.2 ab	35.6 a	176.9 ab	18.2 b
H ₃	54.8 c	11.2 a	2.88 c	116.9 b	47.0 a	185.6 a	30.0 a
p-value	<0.0001	0.0003	<0.0001	0.0305	0.0003	0.0146	<0.0001

Table 2Time required to start the ethylene production (delay), to reach the maximum production (days) and value of this maximum (nl g⁻¹ h⁻¹) in fruit stored at 20 °C after different periods of storage at -0.5 °C. Means with the same letter within column are not significantly different, Tukey $\alpha = 0.05$.

Ethylene	Harvest	Storage time at -0.5 °C (days)				
		90	120	180	210	240
Delay (days)	H ₁	nd	28 a	5 a	0	0
	H ₂	15 a	6 b	2 b	0	0
	H ₃	10 a	6 b	2 b	0	0
	p-value	0.1083	0.0018	0.0004	–	–
Climacteric (days)	H ₁	nd	11 a	11 a	9 a	8 ab
	H ₂	23	14 a	9 b	8 a	7 b
	H ₃	17	12 a	9 b	10 a	9 a
	p-value	0.1242	0.0096	<0.0001	0.5364	0.0138
Climacteric (nl g ⁻¹ h ⁻¹)	H ₁	nd	nd	55 a	47 a	43 a
	H ₂	39a	37a	37 a	35 a	43 a
	H ₃	21a	58a	33 a	28 a	51 a
	p-value	0.2773	0.1153	0.0548	0.1023	0.4815

nd: Not detectable within 30 days of shelf life (20 °C).

Initial absorbance (A_i) was measured at 517 nm on the blank and final absorbance (A_f) was measured after the incubation period using a UV-spectrophotometer (1001 Plus, Milton Roy, USA) following calibration with double distilled water. Inhibition (%) was calculated as follow: (A_i) - (A_f)/(A_i) × 100. Under these conditions, an increase in the DPPH oxidation inhibition percentage corresponded to increase in the total antioxidant activity.

2.7. Statistical analysis

Data were subjected to analysis of variance (ANOVA) using INFOSTAT/Professional version 2006p.1 software. Mean comparisons at $p = 0.05$ were performed using Tukey test.

3. Results

3.1. Maturity at harvest

As on-tree maturation occurs the fruit increased in size (26.9 g from H₁ to H₃), and decreased in firmness (16.5 N), acidity (1.96 g L⁻¹) and green colour (hue 1.6). In addition, starch degradation value increased (34.5%) in parallel to an increase in soluble solids content (0.8%). Our data showed that H₁ fruits were significantly less mature than H₃, according to all maturity indexes, while H₂ showed an intermediate maturity being generally closer to H₃ (Table 1).

Antioxidant potential (DPPH) in the peel of the fruit at harvest was significantly lower in fruit from H₁ and H₂ (19.4% and 18.2%, respectively) than in fruits from H₃ (30%, Table 1).

3.2. Ethylene production

Exposure to low temperatures promoted ethylene synthesis depending on the fruit maturity at harvest. For all harvest dates ethylene production was undetectable at harvest and after 15, 30 or even 60 days of storage at low temperatures (data not shown). Fruits from H₁ required 120 days at low temperatures and 28 days at 20 °C to start ethylene production, while in fruits of H₂ and H₃ this induction occurred after 90 days of cold storage plus 15 and 10

Table 3Superficial scald index (SSI) of 'Beurré D'Anjou' pears harvested on 24 January (H₁); February 7 (H₂) and February 21 (H₃) after different periods of storage at -0.5 °C and 7 days at 20 °C.

Harvest	120	180	210	240
H ₁	0.00 c	0.68 b	2.52 a	2.86 b
H ₂	1.08 b	2.84 a	2.94 a	3.60 a
H ₃	2.84 a	2.80 a	2.80 a	3.42 a
p-value	<0.0001	<0.0001	0.2640	0.0039

days at 20 °C, respectively. After 210 days of storage, fruits from all harvests produced ethylene immediately after removing from cold storage.

After 120 and 180 days of storage, the number of days needed to reach the climacteric peak was longer in H₁ than in the later harvests (Table 2). These results are in agreement with the maturity analysis performed at harvest (Table 1) and confirm the differences in the maturity indexes observed between harvest dates. Collectively these results showed that fruits of H₁ were clearly less physiologically mature than fruit from later harvests.

3.3. Superficial scald

No superficial scald was observed in any of the evaluations performed immediately after removing the fruit from cold storage (data not shown). After 120 and 180 days of cold storage plus 7 days at 20 °C, significant differences between harvest dates ($p < 0.0001$) were observed regarding superficial scald incidence. Pears of the later harvests developed more scald than those from the early harvest. After 120 days of cold storage plus 7 days at 20 °C, early harvested fruit (H₁) showed no symptoms of scald, while fruit from H₂ and H₃ harvests exhibited 70 and 98% of incidence respectively, with greater severity in fruit from an advanced maturity (H₃) (Table 3). Similar results were observed after 180 days of storage, where H₁ fruit had a significantly lower percentage of scald (38%) (Fig. 1) and lower severity index than fruit harvested later (Fig. 1 and Table 3).

Accordingly, superficial scald incidence was clearly related to the fruit maturity at harvest and to the fruit ethylene production

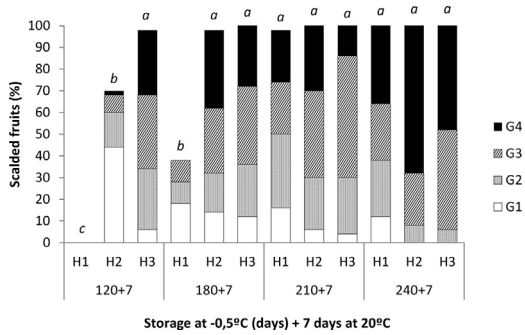


Fig. 1. Incidence (% of affected fruit) and severity (G1, G2, G3, G4) of superficial scald of 'Beurré D'Anjou' pears harvested on 24 January (H₁); February 7 (H₂) and February 21 (H₃) after different periods of storage at -0.5°C and 7 days at 20°C . Means with the same letter are not significantly different Tukey alpha=0.05.

capacity. In this case, the days of delay to start ethylene production and not the days to reach the maximum of ethylene production may be employed as potential indicators of the sensitivity of the fruit to develop superficial scald. After 210 days of storage superficial scald affected more than 90% of the fruits regardless of the harvest date (Fig. 1).

3.4. α -Farnesene (AF) and conjugated trienols (CTols)

AF levels detected at harvest were low, but significant differences between H₁ (1.20 nmol cm^{-2}) and H₃ (4.14 nmol cm^{-2}) were observed. Fruits from H₂ and H₃ began to accumulate AF from day 15 of storage, while fruit of the H₁ initiated AF accumulation only after 30 days of cold storage. Levels of AF for the three harvest dates increased up to 90 days of storage and decreased thereafter, especially when the fruit were picked at H₃ (Fig. 2). Irrespective of the harvest date, very low levels of CTols (less than 1 nmol cm^{-2}) were observed up to 30 days of storage. Fruits from H₁ accumulated significantly lower amounts of CTols from 60 to 120 days of storage if compared to fruit harvested later on H₂ and H₃.

3.5. Fruit antioxidant capacity

At harvest, fruit antioxidant capacity was greater in H₃ fruit than in fruit harvested earlier (Table 1 and Fig. 3). Despite the differences encountered at harvest, the antioxidant capacity of the skin tissue increased, for all harvests, in response to cold storage to reach maximum values after 120 days of cold storage and generally decreased thereafter. The most pronounced decrease in fruit antioxidant capacity and consequently the lowest values towards the end of storage were found in fruit from the latest harvest (H₃).

4. Discussion

4.1. Maturity at harvest as an indicator of scald susceptibility in 'Beurré D'Anjou' pear

In the Alto Valle region, Argentina, 'Beurré D'Anjou' pears are considered mature when the flesh firmness reaches 69 to 73.4 N , soluble solids exceed 10–11%, the malic acid is $3.5\text{--}4\text{ g L}^{-1}$ and starch is degraded by 20–25% (Benítez, 2001). According to these threshold values, fruit from H₁ were slightly immature yet with acceptable firmness values (Table 1) and H₂ fruit were within the range of commercial maturity. Finally, fruit from the later harvest (H₃) were slightly over-mature. It is generally admitted that the interaction between ethylene, AF or CTol content and antioxidant potential depends on the maturity stage and that it affects superficial scald susceptibility. In apples, this interaction explains why early harvested fruits are more susceptible to scald (Chen

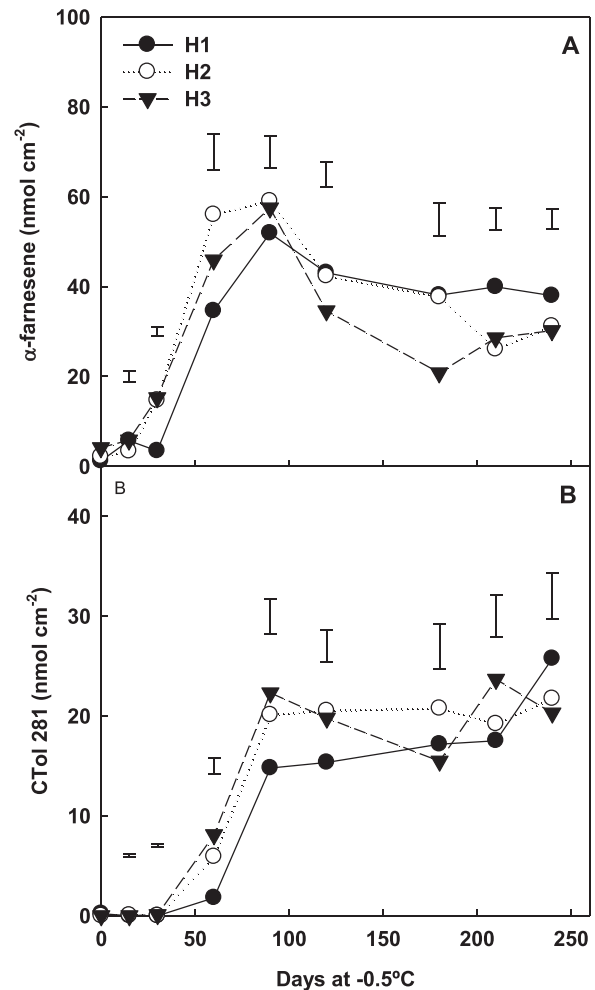


Fig. 2. Content of α -farnesene (A) and conjugated trienols (B) in the skin of 'Beurré D'Anjou' pears harvested on 24 January (H₁); February 7 (H₂) and February 21 (H₃) after different periods of storage at -0.5°C . Values represent the mean of 5 replicates (10 fruit per replicate) and vertical lines indicate the LSD value ($p < 0.05$).

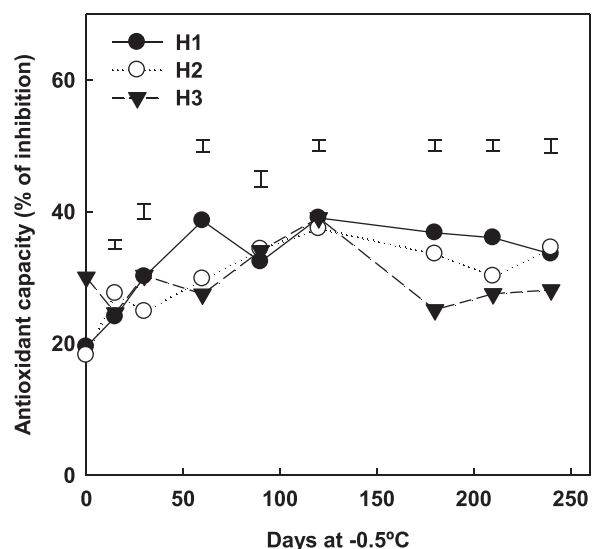


Fig. 3. Changes in the skin antioxidant capacity (% of inhibition) of 'Beurré D'Anjou' pears harvested on 24 January (H₁); February 7 (H₂) and February 21 (H₃) after different periods of storage at -0.5°C . Values represent the mean of 10 replicates and vertical lines indicate the LSD value ($p < 0.05$).

et al., 1985; Ingle and D'Souza, 1989; Wang and Dilley, 1999). In pears, however, these interactions are less evident. While some researchers have observed that pears 'Beurré D'Anjou' (Boonykiat et al., 1987; Zoffoli et al., 1998), 'Packhams Triumph' (Tindale, 1967; Zoffoli et al., 1998) and 'Rocha' (Isidoro and Almeida, 2006) showed a similar behaviour than apples (less mature fruit more prone to develop superficial scald), there are other authors that found opposite results. For instance, Gamrasni et al. (2010) observed that scald incidence was 49.3% and 5% after 6 months of storage in 'Spadona' pears harvested with 48.1 N and 57 N, respectively. Raese and Drake (2000) in 'Beurré D'Anjou' pears and Bower et al. (2003) or Whitaker et al. (2009) in 'Bartlett' pears reported higher scald incidence in fruits harvested later. In summary, the results presented herein confirm that the relationship between scald and maturity in 'Beurré D'Anjou' pears may be opposite to that observed in apples. In this sense, early harvested fruit appeared to be less susceptible to scald than those harvested at an advance maturity stage.

4.2. Relationship between ethylene, α -farnasene, conjugated trienols and superficial scald

The pattern of α -farnasene (AF) accumulation observed in this experiment was similar to that previously described for apples (Huelin and Coggiola, 1968; Anet, 1972; Whitaker et al., 1997; Giné Bordonaba et al., 2013) and pears (Chen et al., 1990; Isidoro and Almeida, 2006; Whitaker et al., 2009) with low initial values at harvest that increased during the first 3 months of cold storage and then decreased due to its *in vivo* oxidation. Conjugated trienols (CTols) have been identified as the predominant *in vivo* oxidation products of AF both in apples and pears (Whitaker, 2007) (Fig. 2). As it occurs in apples, it is assumed that scald incidence in pears is proportional to AF oxidation (Anet, 1972) and that susceptible cultivars are those that exhibit higher levels of CTols (Chen et al., 1990; Whitaker et al., 1997; Gapper et al., 2006; Tsantili et al., 2007). In this work, we observed that CTols, and especially AF, accumulated faster in fruit of advanced maturity (H_2 and H_3), hence explaining, at least in part, the higher incidence and severity of superficial scald of these fruit (Figs. 1 and 2).

Despite decades of research, the way by which AF synthesis is controlled at the molecular level is not fully elucidated. The enzyme AFS1 (alpha farnesene synthase) appears to play a key role in the synthesis of AF and its expression level has been clearly associated to ethylene production in apples (Pechous et al., 2005). In addition, it was demonstrated that not only ethylene production but ethylene perception *per se* are involved in the regulation of AF synthesis and induction of scald in apples (Du and Bramlage, 1994; Watkins et al., 1993; Whitaker et al., 1997; Ju and Curry, 2000). In fact, it is important to notice that, in the present study, the increase in AF content occurs when fruit were not yet capable of producing detectable amounts of ethylene upon removal from cold storage (Table 2). Therefore, it may be that AF synthesis in 'Beurré D'Anjou' does not exclusively depend on ethylene and other factors, such as low temperature storage, may modulate AF synthesis (Table 2 and Fig. 2). Like in other winter pear varieties, cold storage may lead to significant changes at the ethylene perception level by increasing the expression of ethylene receptors such as *PcETR1*, *PcETR5* and *PcERS1* (El-Sharkawy et al., 2003; Chiriboga et al., 2013) which do not directly affect ethylene production (Table 2) but somehow may affect AFS1 expression at the molecular level. Accordingly, using antisense apples (for ACC synthase or ACC oxidase), Pesis et al. (2009) showed that reduced autocatalytic ethylene production was related to lower AF levels and also to less superficial scald. Since ethylene production was not totally suppressed, those authors could not conclude on whether AF synthesis was due to residual ethylene production or to other regulatory elements. Similar conclusions were drawn by Gapper et al. (2006) after detecting certain Pc-

AFS1 transcription levels in 1-MCP treated 'Beurré D'Anjou' pears that showed undetectable ethylene production. It is then clear that further studies are encouraged to assess the AFS activity or expression at the enzymatic or transcript level, and see whether this may explain the differences in scald susceptibility of 'Beurré D'Anjou' pears.

4.3. Critical values of CTols

Another factor to be considered when trying to understand the etiology of superficial scald in pear is associated with the critical values of CTols needed to trigger the disorder. Although these values have never been clearly defined (Gine Bordonaba et al., 2013), there are some studies that quantify this relationship in pears. Chen et al. (1993) observed that in 'Beurré D'Anjou' pears grown in Hood River (USA), scald occurred when CTols content was greater than 2 nmol cm^{-2} . In fruit of the same variety grown in Alto Valle region (Argentina), a threshold value 20 nmol cm^{-2} CTols has also been defined (Calvo and Candan, unpublished data). In this experiment, fruit from H_2 and H_3 showed scald after 120 days of storage when CTols content was 20.8 and 19.9 nmol cm^{-2} , respectively and H_1 scalded after 180 days when CTols levels were $16.8 \text{ nmol cm}^{-2}$ (Fig. 2). The differences observed in the threshold or critical CTol values between studies may be attributed to differences in the methodology used or to regional growing conditions, but it seems clear that in both cases there exists a critical CTols value which if surpassed led to superficial scald.

Although similar CTols levels were found after 120 days of storage between harvests, scald incidence was very different after 180 days of storage (38% incidence in H_1 versus 95–100% in H_2 and H_3 fruit). This result suggests that once the threshold has been reached, scald progresses independently of the levels of CTols and that others compounds such as, for instance, antioxidants may be determinant. Similar results were found by Guerra et al., (2012) in 'Rocha' pear.

4.4. Relationship between fruit antioxidant capacity and superficial scald

The oxidative process associated with scald and triggered by CTols likely involves the activity of several reactive oxygen species (ROS), leading to the unrecoverable disruption of membranes and other cell components (Piretti et al., 1996; Rao et al., 1998; Isidoro and Almeida 2006; Whitaker, 2007; Zubini et al., 2007; Whitaker, 2000; Whitaker et al., 2009). It is generally accepted that late harvested apples are less susceptible to scald and that this difference may be due to higher content of antioxidants (Barden and Bramlage, 1994a,b). In agreement, and despite the known differences in total amount of antioxidants between apples and pears (Campanella et al., 2003; Leontowicz et al., 2003; García-Alonso et al., 2004), late harvested pears (H_3) exhibited higher antioxidant content at harvest but lower antioxidant capacity values towards the end of storage together with greater incidence and severity of scald (Figs. 1 and 3). This result indicates that the initial fruit antioxidant potential was not directly related to the capacity of the fruit to develop scald. It is hence likely, that as specified in apples (Giné-Bordonaba et al., 2013), specific antioxidants rather than the fruit antioxidant potential itself are key elements in determining the fruit susceptibility to superficial scald. For instance, Busatto et al. (2014) recently showed that, in apples, the actual manifestation of superficial scald symptoms resulted from an accumulation of chlorogenic acid and its further oxidation carried out by a polyphenol oxidase (PPO). Whether greater or lower amounts of antioxidants in the fruit are beneficial to prevent the appearance of superficial scald in 'Beurré d'Anjou' pears, needs to be further confirmed. However, these preliminary results confirm that the capacity of the fruit to better maintain its antioxidant potential (as

observed in H₁ fruit) may have a positive influence on the prevention of superficial scald.

5. Conclusion

Collectively the results presented in this work showed that the model generally used for apples to explain the relationship between fruit harvest maturity and superficial scald incidence may not be extrapolated to 'Beurré d'Anjou' pears. Early picked pears were less prone to develop superficial scald than fruit of later harvests. Differences in scald sensitivity between harvests were also associated to the capacity of the fruit to produce ethylene and to the accumulation pattern of AF and CTols. Our results also indicate that AF synthesis in 'Beurré d'Anjou' do not exclusively depends on ethylene. In this sense, other factors, most probably associated with lower storage temperature, account for the accumulation of this sesquiterpene in the skin of the fruit. The involvement of specific antioxidants or the fruit antioxidant capacity remains unclear and should be further investigated yet our results show that the lower susceptibility of early harvested fruit to develop superficial scald was allied to the capacity of the fruit to better maintain its antioxidant potential during storage.

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