Ripening behaviour and consumer acceptance of 'Conference' pears during shelf life after long term DCA-storage

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Abstract

With the increasing demand for ready to eat fruit, understanding how pear quality evolves during shelf life (SL) is of paramount importance for retailers. Accordingly, the relationships between physicochemical quality parameters, the emission of volatile compounds and consumer satisfaction were investigated in 'Conference' pears from different orchards and stored at 20 ºC following 8 months of cold storage (-0.5 ºC) under dynamic controlled atmosphere (DCA). Our results showed that DCA storage strongly inhibits firmness loss (<5 %) without negatively affecting other quality traits. Upon removal from cold storage and ripening at 20 ºC, 'Conference' pears loss nearly 80 % of its initial firmness in only 5 d. Firmness evolution from harvest to 5 d of SL was successfully fitted with a reverse Gompertz equation ($R^2 > 0.96$). Prolonged DCA storage of Conference did not completely impede ripening as indicated by the reducing trend of I_{AD} and the ethylene postclimacteric behavior of the fruit during SL. In parallel to the decrease of firmness during SL, there was a consistent increase in most ester-type volatiles and especially in hexyl acetate and butyl acetate. Generally, the highest consumer satisfaction after DCA cold storage of 'Conference' pears was reached after 3 d at 20 ºC. In this sense, the most appreciated pears by consumer were those showing high flavour in combination with firmness values in the range of 10-30 N. The Partial Least Square (PLS) model showed that total soluble solids (TSS), the ratio TSS/TTA (total titratable acidity), consumer flavour perception and some particular volatile compounds (i.e. methyl, ethyl and hexyl acetates as well as ethyl trans,cis-2,4-decadienoate) were positively correlated to consumer's overall liking while firmness, TTA and index of absorbance difference (I_{AD}) had a negative correlation and higher prediction capability.

Keywords: esters; overall liking; physicochemical parameter; PCA; PLS; reverse Gompertz, VOC

1 Introduction

‘Conference’ is the most grown pear variety in Europe, representing more than 30 % of the pear yield (Chiriboga et al., 2013) . Pear production in Spain in 2017 was higher than 360 000 t (MAGRAMA, 2018). Pears are climacteric fruit and most European pear varieties require a chilling period after harvest, that may vary from a few days to months depending on the variety, to initiate the autocatalytic ethylene production and thereby ripen (Lindo-García et al., 2019). Under this scenario and to guarantee the supply of pears all year round, long-term cold storage under controlled atmosphere conditions are common practices employed by the pear industry. Storage under dynamic controlled atmosphere is undoubtedly the new storage trend in most pear producing countries (Saquet, 2019). Long term cold storage can reduce pear volatile compounds emission (Zlatić et al., 2016) and has been reported to damage to some extent the aroma of some pear varieties such as “Passe-Crassane” (Rizzolo et al., 1991), “Packham’sTriumph” (Chervin et al., 2000) and “Doyenne du Comice” (Lara et al., 2003).

Pear consumption has been steadily decreasing over the past 5 years (MAGRAMA, 2018). The lack of flavour is among the main reasons for the reported decrease in consumption. Consumers demand a closer relationship between the visual appearance, firmness and organoleptic characteristics (Zerbini, 2002). In this sense, the flavour of pears consist of a complex interaction between taste and odour (Yao et al., 2018) where esters play an important role (Lara et al., 2003; López et al., 2001). In relation to the odour or aroma, methyl and hexyl esters of decadienoate are characteristic compounds of European pears such as Conference (Kahle et al., 2005; Rapparini and Predieri, 2003). In addition, hexanal, 2-methylpropyl acetate, ethyl acetate, hexyl acetate, 3-methylbutyl-2-methyl butanoate, ethyl butanoate, and butanol were also identified as impact volatiles in “Conference” pears (Rizzolo et al., 2005), the concentration of which was largely affected by the fruit maturity at the time of harvest as well as postharvest storage conditions.

Several studies are available describing consumer acceptance of pears just after harvest (Brückner, 2008; Kappel et al., 1995), or how consumer acceptance is affected by long term storage (Hájos, 2012; Moya-León et al., 2006). However, scarce information is found about the temporal variations in the fruit quality and the levels of consumers satisfaction during post-cold storage ripening of pears (Zlatić et al., 2016), a period that under regular shelf-life conditions (20 ºC) may be as short as 5 to 15 d depending on the variety. A better understanding of the post-cold storage ripening of Conference pears may provide crucial information for retailers to schedule the distribution of ready-to-eat fruit in order to deliver it at the time of optimal quality in terms of consumer acceptance. Accordingly, this study aimed at: 1) To assess the evolution of quality attributes such as physicochemical parameters, aroma volatile compounds emission and consumer's overall liking during shelf life at 20 ºC under long term DCA storage, and 2) To find out which of these experimentally measured quality attributes have the greatest influence on consumer's satisfaction.

2 Materials and methods

2.1 Plant material and storage conditions

'Conference' pears (*Pyrus communis* L.) were harvested in august 2018 from five different commercial orchards (L1, L2, L3, L4 and L5) in la Rioja (Spain). Fruit was picked up at optimum commercial maturity according to local growers recommendations which are basically assessed in terms of firmness and sugars content (firmness≈ 55-65 N and total soluble solids>13 %). Thereafter, fruit was transported to IRTA research institute with a refrigerated lorry at 0 ºC and stored at -0.5 ºC for 8 months (34 weeks) under a dynamic controlled atmosphere (DCA) at 90-95 % of relative humidity (RH).The initial set values were 1 kPa O_2 and 0.5 kPa CO_2. An ACR system (Van Amerongen, Netherlands) was used to measure the respiration quotient (RQ) every 4 d. When RQ was higher than 2, the O_2 levels were increased by 0.1 kPa, when the RQ was between

1.5 and 2 the O_2 levels were maintained and when it was lower than 1.5 the O_2 level was lowered 0.1 kPa. After storage, fruit was kept at 20 ºC, 70 % RH, and physicochemical parameters, ethylene production, aroma volatile compounds emission, consumer overall liking and some sensory attributes were determined.

2.2 Physicochemical parameters

Physicochemical parameters (firmness, apparent maturity (I_{AD}), total soluble solids (TSS) and total tritatable acidity (TTA)) were measured each sampling day on 20 fruit from each orchard. Samples were taken upon arrival of fruit to IRTA at harvest, after 8 months of cold storage (0 d) and at 1, 2, 3, 4 and 5 d of shelf life (SL) at 20 ºC. Consumers satisfaction tests were carried out at 1, 3 and 5 d of SL. On these days each fruit was divided in two halves, one was used to measure physicochemical parameters and the other half for the consumer evaluation test.

Firmness was determined on two opposite sides of each fruit after removing the peel, using a hand-held penetrometer (Turoni, Italy) fitted with an 8 mm diameter plunger. The semi-spherical plunger was introduced into each fruit and the maximum force was measured. The apparent maturity of each fruit was measured with a DA-Meter (TR Turoni, Forli, Italy), based on the index of absorbance difference ($I_{AD} = A_{670} – A_{720}$), as described by Turpin et al. (2016).

At each sampling date four juices per orchard were prepared crushing together five halves of fruit. From each obtained juice, total soluble solids (TSS, %) were measured using a digital hand-held refractometer (Atago, Tokyo, Japan), and acidity content (TTA) was measured by titration of 10 ml of juice with 0.1 N sodium hydroxide (NaOH) to pH 8.2 using phenolphthalein. TTA results were expressed as g malic acid L^{-1}.

2.3 Ethylene production

Fruit ethylene production capacity upon removal from cold storage (0 d) was measured daily in 15 flasks (3 flasks per orchard) for 8 d. In each 1.5 L flask, 2 weighted fruit were introduced. Flasks were continuously aerated with humidified air at a constant flow rate of 250 mL min^{-1}, and kept in an acclimatized chamber at 20 °C.

The amount of ethylene produced by the fruit was measured by taking a 1 mL sample of gas from the headspace of each flask and injecting it into a gas chromatograph fitted with a FID detector (Agilent Technologies 6890, Wilmington, Germany) and an alumina column 80/100 (2 m × 3 mm) (Teknokroma, Barcelona, Spain) as described by Giné Bordonaba et al. (2014).

2.4 VOCs analysis

Volatile organic compounds (VOCs) emission was determined at 1, 3 and 5 d during the SL period on fruit from orchards L1, L2 and L3. About 2 kg (6-7 fruit per container) of selected fruit free from defects were introduced in an 8-L Pyrex container. A total of 9 containers (3 per orchard) were kept at 20 ºC up to 5 d. A nitrogen stream (150 mL min^{-1}) was forced for 1 h for the VOC's acquisition. The resulting effluent circulated through an adsorption tube filled with 350 mg Tenax TA (2, 6-dipheyl-1-p-henylene oxide) and Carbograph 1TD (Markes International Ltd., Llantrisant, United Kingdom). Adsorption tubes were kept at 4 ºC until were desorbed (Cano-Salazar et al., 2013).

Volatile compounds desorption was done using an automated UNITY Markes thermal desorption system (Markes International Ltd., Llantrisant, United Kingdom) at 275 ºC for 15 min. Identification and quantification were done with an Agilent 7890B gas chromatograph coupled to a 5977A mass spectrometer (MSD) (Agilent Technologies, Inc., Barcelona, Spain). Volatile compounds separation was performed with a capillary column with cross-linked free fatty acid as the stationary phase (FFAP; 50 m×0.2 mm× 0.33 μm). Helium was used as the carrier gas, at a flow speed of 42 cm s^{-1}, with a split flow of 20 mL min^{-1}. Both the injector and detector were kept at 240 ºC. The analysis was conducted according to the following program: 40 ºC (1 min); 40-115 ºC (2.5 ºC min^{-1}); 115-225 ºC (8 ºC min^{-1}); 225 ºC (10 min). Mass spectra was obtained by electron impact ionization at 70 eV, using the same flow of helium and following the same temperature gradient program as the ones used in the separation. Volatile compounds identification was carried out by comparing the spectrometric data recorded to those from the original NIST HP59943C library mass spectra. Quantification was performed using individual calibration curves, with correlation coefficient higher than 0.95, for each identified compound.

2.5 Sensory analysis

As explained in the physicochemical parameters section, consumer evaluation tests were carried on the remaining halves from fruit used for the physicochemical analysis. Briefly, sensory evaluations were conducted as described by Echeverría et al. (2008). Each half of the fruit was peeled and cut into pieces which were used for the sensory evaluation and evaluated separately by one consumer. All of them were regular consumers of pear. Each plate was therefore presented with five pieces of fruit at one time (one from each orchard). Pieces were identified using three digits and were presented to each consumer in a randomized order. The panel of consumers consisted of 56 experienced volunteers from the staff working at the IRTA research institute. Nearly 80% of the members own more than 15 years of experience in this types of tests. In this sense, this may actually be considered as a semi-trained panel. Consumers (30 % men, 70 % women) were asked to rate the overall liking according to a nine-point hedonic scale (1, dislike extremely; 5, neither like nor dislike; 9, like extremely) (Lopez et al., 2011) and to evaluate firmness and flavour separately through a five-point hedonic scale (1 very low intensity; 2 low; 3 regular; 4 moderate; 5 very high intensity) (Echeverría et al., 2015).

2.6 Statistical and data analysis

Means were compared by analysis of variance (ANOVA), when the analysis was statistically significant, the Tukey's Honestly Significant Difference (HSD) test at $P \leq 0.05$

was performed for separation of means using JMP® 13.1.0 SAS Institute Inc. (SAS, 2013). Correlations between experimental variables were checked using Spearman's rank correlation and, if required, presented as Spearman's correlation coefficient (r) and P value based on a two-tailed test. Unless otherwise stated, significant differences were $P \leq 0.05$.

A Principal Component Analyses (PCA) was conducted in order to establish a preliminary relationship between physicochemical parameters and VOC's. The analyzed data included all measured variables along days of SL (1, 3 and 5) and orchards (L1, L2, L3). A Partial Least Square (PLS) model was used to correlate physicochemical parameters and volatile compounds with sensory evaluation. The physicochemical parameters, volatile compounds, sensorial firmness and flavour were selected as X variables in the PLS model. This model contained consumer's overall liking as response variables (Y). The non-linear iterative partial least squares (NIPALS) algorithm was used for computing the first few factors. KFold validation was used to select the number of factors that minimize the Root Mean PRESS statistic. As a pre-treatment, data were centered and weighed by the inverse of the standard deviation of each variable in order to avoid dependence on measured units. All analyses were carried out with the PLS platform of JMP® 13.1.0 SAS Institute Inc. (SAS, 2013).

The reverse Gompertz function (Eq. 1) was used to fit the evolution of fruit firmness (F, N) as a function of time (t, d),

$$F(t) = a\left(1 - \exp\left(-exp\left(\frac{e \cdot r_m}{a}(\lambda - t)\right)\right)\right), \qquad \text{Eq. 1}$$

where *e* is the base of natural logarithms, λ (d) represents the time at which the maximal firmness decay rate r_m (N d^{-1}) is achieved and parameter *a* (N) refers to the ceiling firmness value of the fruit. The confidence intervals for the estimated parameters were obtained by the Monte-Carlo method as described by Illa et al. (2012).

3 Results and discussion

3.1 Physicochemical parameters evolution during shelf life

According to Kappel et al. (1995) physicochemical parameters of pears such as firmness, I_{AD}, total soluble solids content, total titratable acidity and ethylene are important parameters affecting consumer preferences.

In our study, the initial firmness at harvest was in the range 50-62 N (Fig. 1). Fruit from L3 had the lowest firmness values at harvest, yet after 240+3 d of SL differences only existed between L4 and L5 that showed slightly but significantly higher firmness than the rest of orchards ($p<0.05$). After 8 months of cold storage under DCA only fruit from orchard L1 lost 5 % of the initial firmness while no significant losses were observed in the other orchards. In contrast, Saquet (2018) reported a firmness loss higher than 10 % in Conference pears after only 6 months of storage under different CA conditions (0.5 to 3 kPa O_2 and 0.5 to 6 kPa CO_2). It is therefore likely that our more restrictive storage conditions (DCA with average 0.5 kPa O_2 and 0.5 kPa CO_2) better preserve the firmness of Conference pears. Results reported by Goliáš et al. (2015) on Conference pears stored under regular air for 80 d at 1 ºC and 90 % RH showed a firmness decrease around 50 % during the storage period. Their reported values at 7 d of SL were in line to the ones found in our experiment at 5 d of SL. We found that DCA storage, at the conditions described above, better preserves the fruit firmness of Conference pears during long term storage without negatively affecting other quality attributes or leading to fermentative-related physiological disorders. Indeed, fruit did not shown a significant incidence of internal breakdown disorders (data not shown). Further investigation regarding which are the O_2 threshold levels supported by the fruit under DCA storage is warranted since our storage conditions were far more restrictive than those recommended by Saquet (2019) (2 kPa O_2 and lower than 0.7 kPa CO_2).

The overall evolution of fruit firmness reported herein followed an inverted sigmoidal pattern with a clear inflexion point during the SL (Fig. 1). Predieri and Gatti, (2009) analysed firmness decrease on ‘Abate Fetel’ pears during SL after 13 and 23 weeks storage in regular air at (0-1 ºC) and 95 % RH. They reported that pears stored for 13 weeks also followed an inverted sigmoid curve; however, most curves after 23 weeks did not show the inflexion point. Galvis-Sánchez et al. (2004) reported a similar yet slower firmness loss and no inflexion points during the SL of ‘Rocha’ pears stored during 9 months under different controlled atmospheres (2 and 4 kPa O_2 with 0.5 and 1.5 kPa CO_2) at a temperature between 0-0.5 ºC and RH in the range of 90-95 %. Differences between both studies are likely related not only to cultivar differences but also to the different storage conditions and different data points being measured.

Table 1 shows the best fit parameter values for Eq. 1 and its confidence intervals. All firmness fits had a determination coefficient higher than 0.96. To fit the function, firmness values from harvest, after cold storage and during SL were used. When the fitting was performed without the harvest point, a maximum deviation in the function of 1.3 % at the time t=240+0 d was found in orchard 5. It should be highlighted that in that case the fitted parameter values were not significantly different but the confidence intervals were wider than when including data point from harvest.

The index of absorbance difference in the range of 670-720 nm at the fruit skin (I_{AD}) measures the light absorbance due to chlorophyll. The I_{AD} for Conference pears presented a clear decrease trend during the 8-month cold storage and followed a soft decline throughout the shelf life period (Fig. 2A). Costa et al. (2016) reported that I_{AD} values were a useful tool for assessing postharvest ripening of ‘Abbé Fétel’ pear fruit. Similar observations were made by Saquet (2019) when reviewing the use of this non-destructive parameter as a quality indicator during postharvest storage of pears.

In a study carried out by Jaeger et al. (2003), consumers described the ideal pear as juicy and sweet, the key characteristics of ripeness. Sweetness is mostly related to TSS concentration and the balance between TSS/TTA. In our study, TTA values clearly decreased during the storage period in fruit from all orchards. During the SL period the non-uniform evolution in each orchard followed a global slightly decreasing trend (Fig. 2B). TSS/TTA ratio increased during the DCA cold storage in all orchards, but no clear trend was observed during the SL period (Fig. 2C). The unsteady trend in TSS/TTA ratio during SL has already been reported by Bolte-Lombardiz et al. (2000) in Shinsseiki pears and was attributed to TTA variations.

Ethylene is known to be a major factor regulating fruit ripening, and its sharp increase is considered to control the aroma biosynthesis and other biochemical and physicochemical process (Moya-León et al., 2006; Rapparini and Predieri, 2003). Similar ethylene production rates to those described herein have been previously reported in Spanish Conference pears (Chiriboga et al. 2013b). In our study, fruit had a postclimateric behaviour with the highest ethylene production rate immediately upon removal from cold storage and a decline thereafter, except for the fruit from L3 orchard (Fig. 3). In Conference pears stored under regular air, a typical climacteric behaviour during post-cold storage ripening has been observed up to 90-120 d following cold-storage but not later (Chiriboga et al. 2013b).

3.2 Volatile organic compounds emission

Thirty-four volatile compounds were identified and quantified during the SL period (1, 3 and 5 d) of ‘Conference’ pears previously stored under DCA conditions for 8 months (data not shown). These volatile compounds included 20 esters, 4 alcohols, 1 aldehyde, 4 terpenoids, 2 hydrocarbons and 3 acids. However, only those quantitatively more important and following some remarkable trend over the shelf-life period are shown in table 2.

According to previous works, the aroma of pears is mainly caused by esters (El Hadi et al., 2013; Kahle et al., 2005; Maarse, 1991; Zlatić et al., 2016). The main esters detected in our study (Table 2) were butyl, ethyl and hexyl acetates as straight esters, and methyl and ethyl trans, cis-2,4-decadienoate as branched esters. Similar results were obtained by Rapparini and Predieri (2003) and Kahle et al. (2005), who reported that the methyl esters of decadienoate were the characteristic compounds of european pears. Further, hexyl and butyl acetates were also found important volatile compounds in the pear aroma (Rapparini and Predieri, 2003). High concentrations of these acetates were reported by Saquet (2017) on 'Conference' pear after 2 months of storage plus 7 d of SL. An increase in the ethyl acetate concentration was observed as SL period lengthened (Table 2). Other authors also identified ethyl acetate and hexyl acetate as impact volatiles in 'Conference' pears stored for up to 22 weeks in air and controlled atmosphere (Rizzolo et al., 2005).

In our study, the highest emission rates of ethyl trans, cis-2,4-decadienoate were detected after 5 d of SL in fruit from all the orchards. Similarly, Hendges et al. (2018) observed a high content of this volatile compound in Conference pears treated with 1-MCP after 7 months of storage under normal air and controlled atmosphere plus 7 d at 20 ºC and 60 % RH.

Ethanol was the main alcohol present in the headspace from Conference pears, however, it did not contribute to the fruit odour pattern, owing to its very high odour threshold concentration. Zerbini, et al. (1993) also reported that in 'Conference' pear ethanol was the main alcohol. Ethanol is a marker of fermentative paths if produced in high amounts (Perata and Alpi, 1993). However, the concentrations detected in this work were well below its odour threshold which is 10000 $\mu g\ L^{-1}$ The ethanol emission rates detected in this work during SL period for the three orchards were similar. In our experiments, 3-hydroxydodecanoic acid was also found as a characteristic acid of

'Conference' pears and thereby agree with the results from Heinz and Jennings (1966) on other pear varieties (Barlett).

3.3 The relationship between physicochemical parameters and VOC production

A PCA model was used to obtain a global overview of the relationship between physicochemical parameters and the profile of volatile compounds in a reduced dimension plot. In this data set, 19 variables were used for the PCA: 5 physicochemical parameters (F, I_{AD}, TSS, TTA TSS/TTA ratio), ethylene production and the 13 volatile compounds showed in Table 2. The biplot of the two principal components (PC1 and PC2) captured 71.9% of the total variability (Fig. 4). This biplot showed three groups along the first component, differentiating samples from different SL periods. On the left of the first component are located the samples at 1 d of SL, which were mainly characterized by higher values of firmness, titratable acidity, I_{AD} and ethylene production, meaning that this fruit was less mature. In the middle of the graph are situated the samples at 3 d of SL and on the right the ones at 5 d of SL. These last samples, especially the pears from L1 and L2, were related to high concentrations of some of the most important volatile compounds (hexyl butanoate, ethyl trans, cis-2,4-decadienoate, hexyl and butyl acetate), together with high TTS and TSS/TTA ratio values. The variability among samples increased with time, samples at 1 d of SL were quite homogenous compared with samples at 3 and 5 d of SL. All the volatiles emissions were positively correlated among themselves, while were negatively correlated with firmness. Similar aromatic volatiles were described for Barlett pears by Li (2012). The observed increase of the variability seems to be mainly due to the biosynthesis of some particular volatile compounds, to the erratic pattern changes in the TSS values and thereby also by the TTS/TTA ratio. The volatile compounds emitted by fruit of the L1 orchard, located in the upper part of the two groups (at 3 and 5 d of SL period), showed higher concentrations of butyl butanoate, methyl trans, cis-2,4-decadienoate and butyl acetate, and less TTS and TTS/TTA content. All these compounds possess a strong "pear-like" aroma

(Suwanagul, 1996). The second component discriminated the three different sources. At the top, lied the samples from orchard L1, which were more immature at harvest based on the I_{AD} index. In the middle of the plot, were located fruit from L2. Finally, at the bottom there were samples from L3 orchard. The variability among orchards was lower than among days of SL, since the later cluster was the one that represented most of the variability along the PC1.

3.4 Consumer acceptance

Figure 5A shows that consumer's overall liking depended on the interaction between the sensorial firmness and flavour. In our study, the most pleasing pears, or the ones that obtained higher overall liking scores, were those with a moderate-low sensorial firmness (consumers rated from 1 to 3 in a 5-points hedonic scale) and with a high flavour (consumers rated as 5 in a 5-points hedonic scale). This higher overall liking was obtained between 3 and 5 d of SL (Fig. 5B), regardless of the orchard. Thus, for long term stored 'Conference' pears higher marketability will be reached after being 3 d in retail.

A PLS model is a useful tool to identify which are the indicators that a consumer value more in terms of overall liking (Abdi, 2003). Similar approaches have been done with other fruit including apples (Altisent et al., 2011) and peaches (Cano-Salazar et al., 2013), but to the best of our knowledge this information is lacking for pears. We used a PLS to correlate consumer overall liking (Y variable) with a set of potentially explanatory variables: physicochemical parameters, ethylene production, volatiles organic compounds and sensory attributes (X variables).

Based on PLS method, the X data set was reduced to two principal factors. The first factor explained the 74.58 % of the variation while the second explained the 9.01 %. Thus, the cumulative variation explained by two principal factors was 83.6 % (Fig. 6).

The correlation between measured and predicted overall liking was R^2=0.836, demonstrating the goodness of the model (Fig. 6 insert). This figure showed that consumers preferred fruit at 5 d of SL from orchards L3. Interestingly, those fruit were harvested at I_{AD} values of 1.8 which is the reported optimal harvest values to maximise consumer acceptance in other pear varieties ('Abbé Fétel'; Costa et al. (2016)). The variable importance plot (VIP) (Fig. 7) showed that TSS, TSS/TTA ratio, methyl, ethyl and hexyl acetates, hexyl butanoate, ethyl hexanoate, ethyl trans,cis-2,4-decadienoate and flavour (sensory attribute) were variables positively correlated, with a high weight, to consumers overall liking. In contrast, fruit firmness, TTA and sensorial firmness were negatively correlated to consumer global satisfaction. All of them, were among the most powerful X variables in the determination of the PLS model. All these variables had values above 1 and therefore were the greater contributors that explained the variation (Chong and Jun, 2005).

4 Conclusions

Long term storage of 'Conference' pears at -0.5 ºC under DCA reduces the decay of firmness at levels below 5 % without significantly altering other quality traits yet without completely impeding fruit ripening. Accordingly, upon removal from cold storage and ripening at 20 ºC, eating quality, in terms of flesh firmness, is reached in no longer than 5 d. The massive decrease in the fruit firmness during shelf-life is parallel by a substantial increase in most ester-type volatiles and especially in butyl acetate and ethyl-trans,cis-2,4-decadienoate (5-fold higher at 5 d of SL than at 1 d). The highest consumer appreciation of Conference pears during SL occurred at 3 d of SL when pears had a moderate-low sensorial firmness (equivalent to 25 N of instrumental firmness) and high flavour. The PLS model showed that TSS, TSS/TTA ratio, consumer flavour perception and some particular volatile compounds (i.e. methyl, ethyl and hexyl acetates, ethyl trans,cis-2,4-decadienoate) were positively correlated to consumer's overall liking while firmness, TTA and I_{AD} had a negative correlation yet with higher prediction capability.

Overall, the results from this study may be of paramount importance for retailers aiming to distribute ready-to-eat Conference pears at the time of optimal quality in terms of consumer acceptance.

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References

Abdi, H., 2003. Partial Least Squares (PLS) Regression . Encycl. Soc. Sci. Res. Methods 1–7. doi:http://dx.doi.org/10.4135/9781412950589.n690

Altisent, R., Graell, J., Lara, I., López, L., Echeverría, G., 2011. Comparison of the volatile profile and sensory analysis of “Golden reinders” apples after the application of a cold air period after ultralow oxygen (ULO) storage. J. Agric. Food Chem. 59, 6193–6201. doi:10.1021/jf2005029

Bolte-Lombardiz, S.R, Munt-de-Mores, D., Camelatto D., 2000. Avaliação Do Crescimento E Da Maturação Pós-Colheita De Pêras Da Cultivar Shinsseiki. Pesqui. Agropecuária Bras. 35, 2399–2405.

Brückner, B., 2008. Consumer acceptance of fruit and vegetables: the role of flavour and other quality attributes. Fruit Veg. Flavour 11–17. doi:10.1533/9781845694296.1.11

Cano-Salazar, J., López, M.L., Crisosto, C.H., Echeverría, G., 2013. Volatile compound emissions and sensory attributes of “Big Top” nectarine and “Early Rich” peach fruit in response to a pre-storage treatment before cold storage and subsequent shelf-life. Postharvest Biol. Technol. 76, 152–162. doi:10.1016/j.postharvbio.2012.10.001

Chervin, C., Speirs, J., Loveys, B., Patterson, B.D., 2000. Influence of low oxygen storage on aroma compounds of whole pears and crushed pear flesh. Postharvest Biol. Technol. 19, 279–285. doi:10.1016/S0925-5214(00)00096-X

Chiriboga, M.-A., Saladié, M., Giné Bordonaba, J., Recasens, I., Garcia-Mas, J., Larrigaudière, C., 2013. Effect of cold storage and 1-MCP treatment on ethylene perception, signalling and synthesis: Influence on the development of the evergreen behaviour in ‘Conference’ pears. Postharvest Biol. Technol. 86, 212–220. doi:10.1016/j.postharvbio.2013.07.003

Chong, I.G., Jun, C.H., 2005. Performance of some variable selection methods when multicollinearity is present. Chemom. Intell. Lab. Syst. 78, 103–112. doi:10.1016/j.chemolab.2004.12.011

Costa, G., Vidoni, S., Rocchi, L., 2016. Use of non-destructive devices to support pre- and postharvest fruit management. Acta Hortic. 1119, 329–335. doi:10.17660/ActaHortic.2016.1119.45

Echeverría, G., Cantín, C.M., Ortiz, A., López, M.L., Lara, I., Graell, J., 2015. The impact of maturity, storage temperature and storage duration on sensory quality and consumer satisfaction of ‘Big Top®’ nectarines. Sci. Hortic. (Amsterdam). 190, 179–186. doi:10.1016/J.SCIENTA.2015.04.022

Echeverría, G., Graell, J., Lara, I., López, M.L., 2008. Physicochemical measurements in ‘Mondial Gala®’ apples stored at different atmospheres: Influence on consumer acceptability. Postharvest Biol. Technol. 50, 135–144. doi:10.1016/J.POSTHARVBIO.2008.05.002

El Hadi, M.A.M., Zhang, F.J., Wu, F.F., Zhou, C.H., Tao, J., 2013. Advances in fruit aroma volatile research. Molecules 18, 8200–8229.

doi:10.3390/molecules18078200

Galvis-Sánchez, A.C., Fonseca, S.C., Morais, A.M.M.B., Malcata, F.X., 2004. Sensorial and physicochemical quality responses of pears (cv Rocha) to long-term storage under controlled atmospheres. J. Sci. Food Agric. 84, 1646–1656. doi:10.1002/jsfa.1798

Giné Bordonaba, J., Cantin, C.M., Larrigaudière, C., López, L., López, R., Echeverria, G., 2014. Suitability of nectarine cultivars for minimal processing: The role of genotype, harvest season and maturity at harvest on quality and sensory attributes. Postharvest Biol. Technol. 93. doi:10.1016/j.postharvbio.2014.02.007

Goliáš, J., Kožíšková, J., Létal, J., 2015. Changes in volatiles during cold storage and subsequent shelf-life of “conference” pears treated with 1-MCP. Acta Hortic. 1079, 465–471. doi:10.17660/ActaHortic.2015.1079.61

Hájos, T., 2012. Determination of quality in stored pear fruits by chemical analysis and sensorial judgement. Int. J. Hortic. Sci. 18, 27–31.

Heinz, D.E., Jennings, W.G., 1966. Volatile Components of Bartlett Pear. V. J. Food Sci. 31, 69–80. doi:10.1111/j.1365-2621.1966.tb15417.x

Hendges, M.V., Neuwald, D.A., Steffens, C.A., Vidrih, R., Zlatić, E., do Amarante, C.V.T., 2018. 1-MCP and storage conditions on the ripening and production of aromatic compounds in Conference and Alexander Lucas pears harvested at different maturity stages. Postharvest Biol. Technol. 146, 18–25. doi:10.1016/J.POSTHARVBIO.2018.08.006

Illa, J., Prenafeta-Boldú, F.X., Bonmatí, A., Flotats, X., 2012. Empirical characterisation and mathematical modelling of settlement in composting batch reactors. Bioresour. Technol. 104, 451–458. doi:10.1016/J.BIORTECH.2011.10.031

Jaeger, S.R. , Lund, C.M., Lau, K., Harker, F.R., 2003. In Search of the “Ideal” Pear (pyrus spp.): Results of a Multidisciplinary Exploration. JFS Sens. Nutr. Qual.

Food 68, 1108–1117.

Kahle, K., Preston, C., Richling, E., Heckel, F., Schreier, P., 2005. On-line gas chromatography combustion/pyrolysis isotope ratio mass spectrometry (HRGC-C/P-IRMS) of major volatiles from pear fruit (Pyrus communis) and pear products. Food Chem. 91, 449–455. doi:10.1016/J.FOODCHEM.2004.06.026

Kappel, F., Fisher-Fleming, R., Hogue, E.J., 1995. Ideal pear sensory attributes and fruit characteristics. HortScience 30, 988–993.

Lara, I., Miró, R.M., Fuentes, T., Sayez, G., Graell, J., López, M.L., 2003. Biosynthesis of volatile aroma compounds in pear fruit stored under long-term controlled-atmosphere conditions. Postharvest Biol. Technol. 29, 29–39. doi:10.1016/S0925-5214(02)00230-2

Li, G., 2012. Characterization of aromatic volatile constituents in 11 Asian pear cultivars belonging to different species. African J. Agric. Reseearch 7. doi:10.5897/AJAR12.563

Lindo-García, V., Larrigaudière, C., Echeverría, G., Murayama, H., Soria, Y., Giné-Bordonaba, J., 2019. New insights on the ripening pattern of ‘Blanquilla’ pears: A comparison between on- and off-tree ripened fruit. Postharvest Biol. Technol. 150, 112–121. doi:10.1016/J.POSTHARVBIO.2018.12.013

Lopez, G., Hossein Behboudian, M., Echeverria, G., Girona, J., Marsal, J., 2011. Instrumental and sensory evaluation of fruit quality for “Ryan’s sun” peach grown under deficit irrigation. Horttechnology 21, 712–719.

López, M.L., Miró, R., Graell, J., 2001. Quality and Aroma Production of Doyenne du Comice Pears in Relation to Harvest Date and Storage Atmosphere. Food Sci. Technol. Int. 7, 493–500. doi:10.1106/FLWJ-18CN-8TR9-480G

Maarse, H., 1991. Volatile Compounds in Foods and Beverages. New York: Marcel Dekker.

MAGRAMA, 2018. http://www.mapama.gob.es/es/alimentacion/temas/consumo-y-comercializacion-y-distribucion-alimentaria/panel-de-consumo-alimentario/ultimos-datos/ [WWW Document]. Inf. del Consum. Aliment. en España 2017.

Moya-León, M.A., Vergara, M., Bravo, C., Montes, M.E., Moggia, C., 2006. 1-MCP treatment preserves aroma quality of 'Packham's Triumph' pears during long-term storage. Postharvest Biol. Technol. 42, 185–197. doi:10.1016/J.POSTHARVBIO.2006.06.003

Perata, P., Alpi, A., 1993. Plant responses to anaerobiosis. Plant Sci. 93, 1–17. doi:10.1016/0168-9452(93)90029-Y

Predieri, S., Gatti, E., 2009. Effects of cold storage and shelf-life on sensory quality and consumer acceptance of 'Abate Fetel' pears. Postharvest Biol. Technol. 51, 342–348. doi:10.1016/J.POSTHARVBIO.2008.09.006

Rapparini, F., Predieri, S., 2003. Pear fruit volatiles. In: Janick, J. (Ed.), Horticultural Reviews. John Wiley & Sons, Hoboken, NJ, USA, pp. 237–324.

Rizzolo, A., Cambiaghi, P., Grassi, M., Zerbini, P.E., 2005. Influence of 1-methylcyclopropene and storage atmosphere on changes in volatile compounds and fruit quality of conference pears. J. Agric. Food Chem. 53, 9781–9789. doi:10.1021/jf051339d

Rizzolo, A., Sodi, C., Polesello, A., 1991. Influence of ethylene removal on the volatile development in passa crassana pears stored in a controlled atmosphere. Food Chem. 42, 275–285. doi:10.1016/0308-8146(91)90070-5

Saquet, A.A., 2019. Storage of pears. Sci. Hortic. (Amsterdam). 246, 1009–1016. doi:10.1016/J.SCIENTA.2018.11.091

Saquet, A.A., 2018. Storability of 'Conference' Pear Under Various Controlled Atmospheres. Erwerbs-Obstbau 60, 275–280. doi:10.1007/s10341-018-0369-7

Saquet, A.A., 2017. Aroma volatiles of ' Conference ' pear and their changes during regular air and controlled atmosphere storage 55–66. doi:10.26669/2448-4091121

Suwanagul, A., 1996. Ripening Pear Flavor Volatiles: Identification. Biosynthesis and Sensory Perception. Diss. Oregon State Univ. Corvallis.

Turpin, S.R., Stefanelli, D., Jones, L., Norton, J., Probst, R., Konings, J., Langford, G., 2016. Perfect pears for the next generation of consumers. Acta Hortic. 1120, 507–513. doi:10.17660/ActaHortic.2016.1120.77

Yao, M., Zhou, X., Zhou, Q., Shi, F., Wei, B., Cheng, S., Tan, Z., Ji, S., 2018. Low temperature conditioning alleviates loss of aroma-related esters of 'Nanguo' pears by regulation of ethylene signal transduction. Food Chem. 264, 263–269. doi:10.1016/J.FOODCHEM.2018.05.024

Zerbini, P.E., Balzarotti, R., Rizzolo, A., Spada, G.L., 1993. Effect of picking date on quality and sensory characteristics of pears after storage and ripening. Acta Hortic. 326.

Zerbini, P.E., 2002. The Quality of Pear Fruit. Acta Hortic. 596, 805–810. doi:10.17660/ActaHortic.2002.596.139

Zlatić, E., Zadnik, V., Fellman, J., Demšar, L., Hribar, J., Čejić, Ž., Vidrih, R., 2016. Comparative analysis of aroma compounds in "Bartlett" pear in relation to harvest date, storage conditions, and shelf-life. Postharvest Biol. Technol. 117, 71–80. doi:10.1016/j.postharvbio.2016.02.004

List of tables

Table 1: Estimated parameter values of the reverse Gompertz equation and corresponding confidence intervals (c.i.) at 95 % confidence when fitting firmness evolution of Conference pears during 8 months of cold storage and 5 d of SL at 20ºC as function of time. Coefficient of determination r^2 reflects the goodness of the fits.

Orchard	λ (d)	λ (c.i.)	*a* (N)	*a* (c.i.)	r_m (N d^{-1})	r_m (c.i.)	r^2
L1	2.4	2.1-2.7	59.8	56.1-63.9	15.6	12.9-19.7	0.984
L2	2.3	2.0-2.7	59.6	56.0-64.0	15.4	12.7-19.4	0.984
L3	2.3	2.5-2.7	50.6	48.6-52.7	10.4	9.3-11.7	0.994
L4	2.5	1.9-2.9	62.3	57.0-68.4	12.7	9.9-17.4	0.966
L5	2.6	2.0-3.1	62.3	55.9-67.4	11.7	9.1-15.8	0.966

Table 2: Mean (n=3) values of major VOC's emission rate (µg kg^{-1} h^{-1}) by 'Conference' pears from orchards L1, L2 and L3 at 1, 3 and 5 d of SL. Means within the orchard and days of SL preceded by the same small letters are not significantly different at p≤0.05 (HSD test). No letter indicates the absence of significant differences. (-, values under the detection threshold).

	L1			L2			L3		
	1	3	5	1	3	5	1	3	5
Methyl acetate	-	^{ab}1.228	^{a}1.868	-	^{cd}0.183	^{d}0.107	^{d}0.141	^{bc}0.963	^{ab}1.524
Ethyl Acetate	2.329	3.353	5.508	0.802	1.418	1.775	0.956	3.233	5.527
Butyl acetate	^{bc}2.992	^{ab}11.392	^{a}14.371	^{c}1.578	^{c}2.195	^{bc}3.407	^{bc}3.267	^{abc}7.137	^{abc}9.951
Pentyl acetate	^{b}0.344	-	^{a}1.022	^{b}0.033	^{b}0.084	b0.150	-	^{b}0.097	^{b}0.179
Butyl butanoate	-	0.489	0.136	-	0.056	0.017	-	0.034	-
Ethyl hexanoate	-	0.017	0.023	-	0.002	0.006	-	0.008	0.012
Hexyl acetate	-	^{ab}2.898	^{a}3.412	^{a}b0.312	^{ab}0.569	^{ab}0.641	^{ab}0.307	^{ab}2.698	^{a}3.313
Butyl hexanoate	0.055	-	0.114	-	0.035	0.046	-	0.027	0.077
Hexyl butanoate	-	0.014	0.035	-	0.042	0.017	-	0.037	0.063
Methyl trans,cis-2,4-decadienoate	-	^{a}3.896	^{c}0.859	-	^{c}0.109	^{c}0.465	-	^{bc}0.943	^{ab}2.649
Ethyl trans,cis-2,4-decadienoate	-	^{bc}2.088	^{ab}3.233	-	^{d}0.573	^{d}1.166	^{d}0.267	^{c}3.074	^{a}5.132
Ethanol	3.780	2.402	2.962	1.973	1.384	2.214	1.584	1.548	1.753
3-Hydroxydodecanoic acid	-	^{a}0.345	^{abc}0.178	-	^{bc}0.060	^{bc}0.037	^{ab}0.257	^{abc}0.134	^{bc}0.120

List of figures

Figure 1: Fits of the mean measured fruit firmness at harvest (0), after 8 months of cold storage (240+0) and during SL period (1 to 5 d) as a function of time with the reverse Gompertz equation (lines) of fruit from orchards: L1, L2, L3, L4 and L5. Error bars represent the mean ± standard deviation (n=20).

Figure 2: Postharvest evolution of physicochemical parameters in ‘Conference pears: A) I_{AD} Index; B) titratable acidity, TTA; C) total soluble solids, TSS/TTA ratio at harvest (0), just after 8 months of cold storage (240+0) d and during SL period (1, 2, 3, 4 and 5 d). The vertical bar at the upper right corner represents the significant difference length according to the Tukey HSD test value.

Figure 3: Ethylene production rate evolution during SL at 20 ºC of ‘Conference’ pears from different orchards: L1, L2, L3, L4 and L5. The vertical bar at the upper right corner represents the significant difference length according to the Tukey HSD test value.

Figure 4: Score plot of PC1 and PC2 from a full data PCA model considering instrumental quality and VOC’s (n=19). Data were identified in three different cluster groups: 1 d of SL (red continuous line), 3 d of SL (yellow dashed line) and 5 d of SL (blue dotted line). Data representing three different orchards: L1 (●), L2 (▲) and L3 (■) is contained in the circumference of the correlation circle (black dashed circle).

Figure 5: A) 3D plot of the interaction between sensorial firmness (Y) and flavour (X) through a five-point hedonic scale (1, very low intensity; 5, very high intensity) with consumers overall liking (Z) based on a nine-point hedonic test (1, dislike extremely; 5, neither like nor dislike; 9, like extremely). B) Overall liking during the SL period of ‘Conference’ pears at 240+1 d, 240+3 d, 240+5 d. Error bars represent the standard error. Different letters indicate significant differences ($p < 0.05$) for each day of SL.

Figure 6: Partial Least Squares (PLS) correlation loading plots of the 2 factors. Data was identified in three different groups: 1 d of SL (red continuous line), 3 d of SL (yellow dashed line) and 5 d of SL (blue dotted line), representing fruit from three different orchards: L1 (●), L2 (▲) and L3 (■). The measured vs the predicted overall liking through the model and its correlation coefficient is given in the insert.

Figure 7: Variable importance plot (VIP), the number of VIP>1 (continuous black line) indicates that the indicators are influential in determining the two factors used in the model.

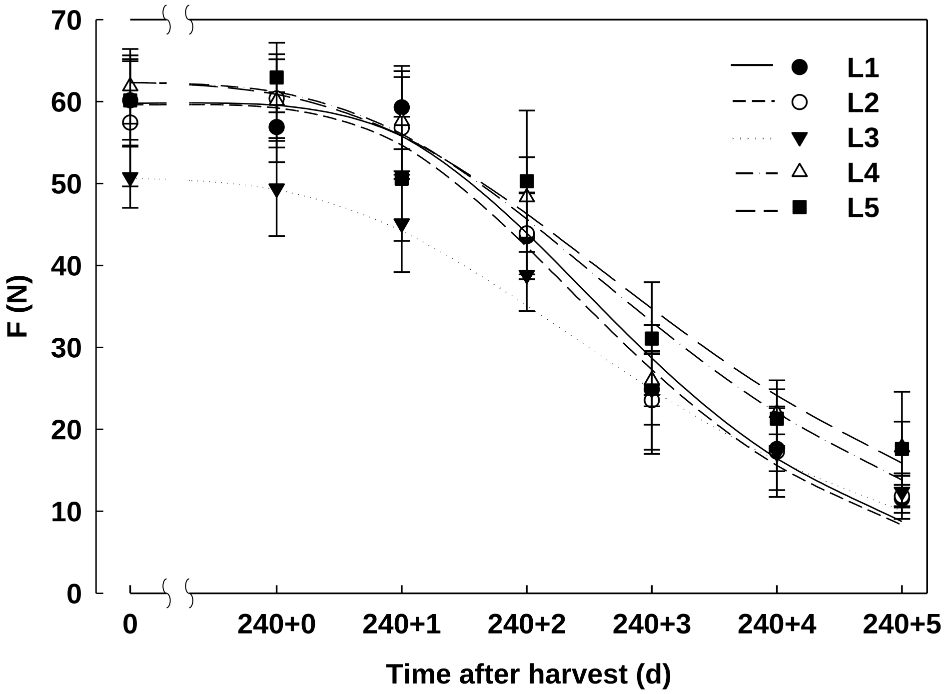
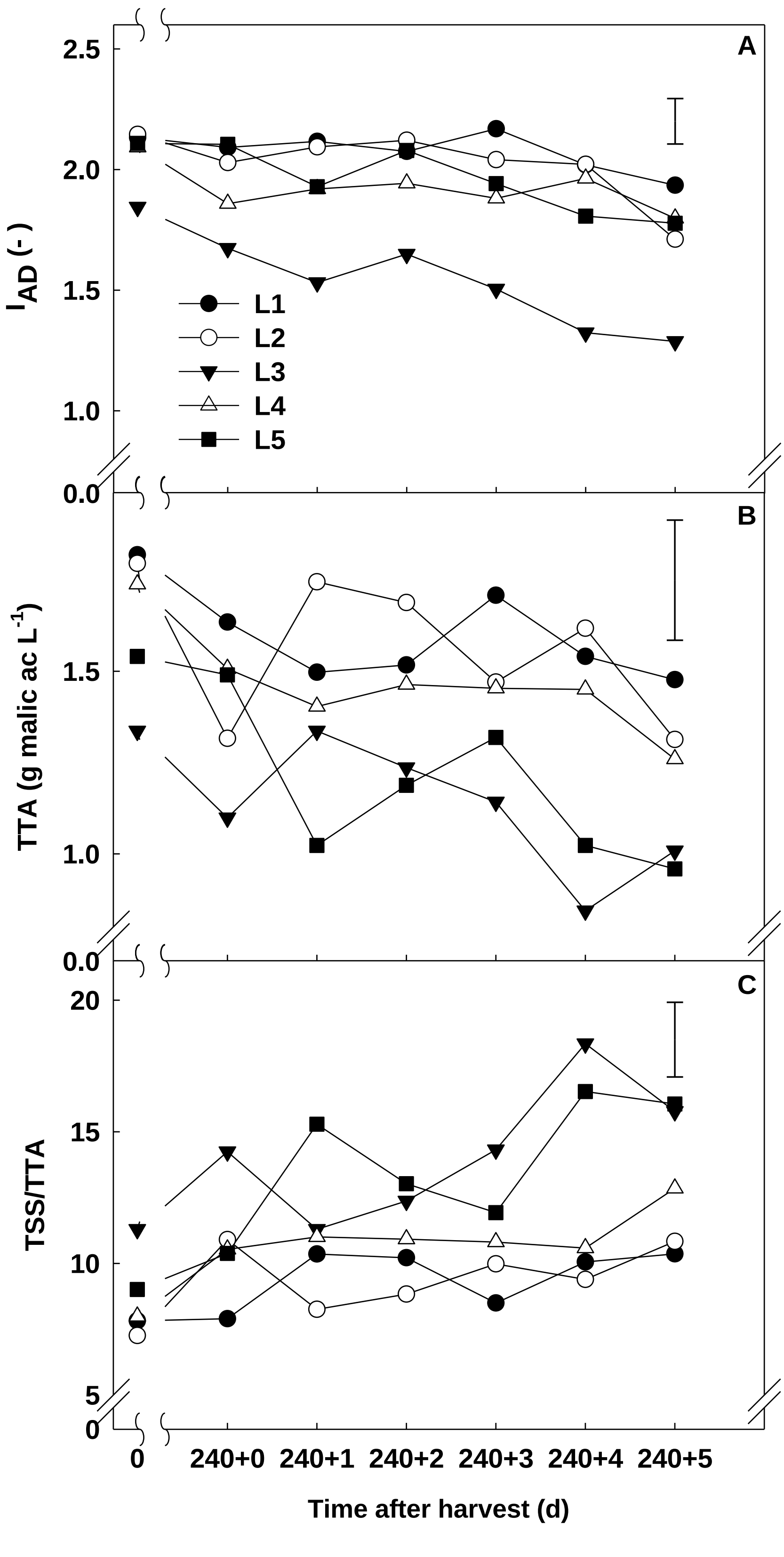

F (N)
70
60
50
40
30
20
10
0
0
240+0
240+1
240+2
240+3
240+4
240+5
Time after harvest (d)
L1
L2
L3
L4
L5

Figure 1



597

598 **Figure 2**

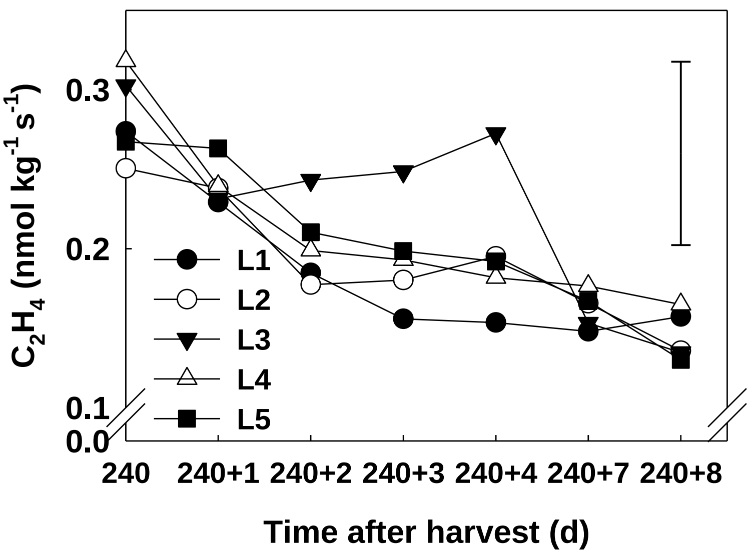

C2H4 (nmol kg-1 s-1)
0.3
0.2
0.1
0.0
L1
L2
L3
L4
L5
240
240+1
240+2
240+3
240+4
240+7
240+8
Time after harvest (d)

Figure 3

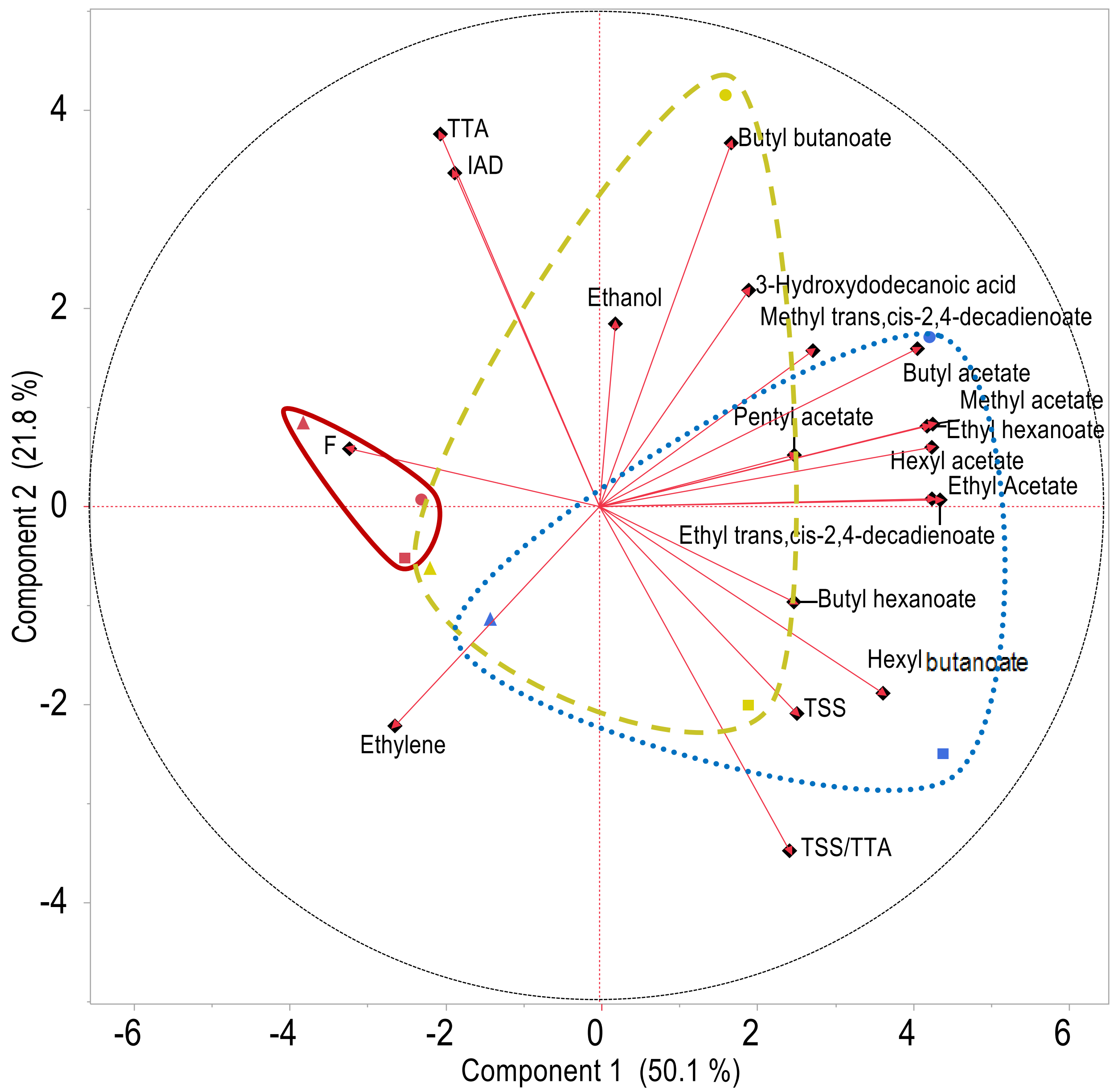

TTA
IAD
Butyl butanoate
Ethanol
3-Hydroxydodecanoic acid
Methyl trans,cis-2,4-decadienoate
Butyl acetate
Methyl acetate
Ethyl hexanoate
Pentyl acetate
Hexyl acetate
Ethyl Acetate
F
Ethyl trans,cis-2,4-decadienoate
Butyl hexanoate
Hexyl butanoate
TSS
Ethylene
TSS/TTA
4
2
0
-2
-4
-6
6
Component 2 (21.8 %)
Component 1 (50.1 %)

Figure 4

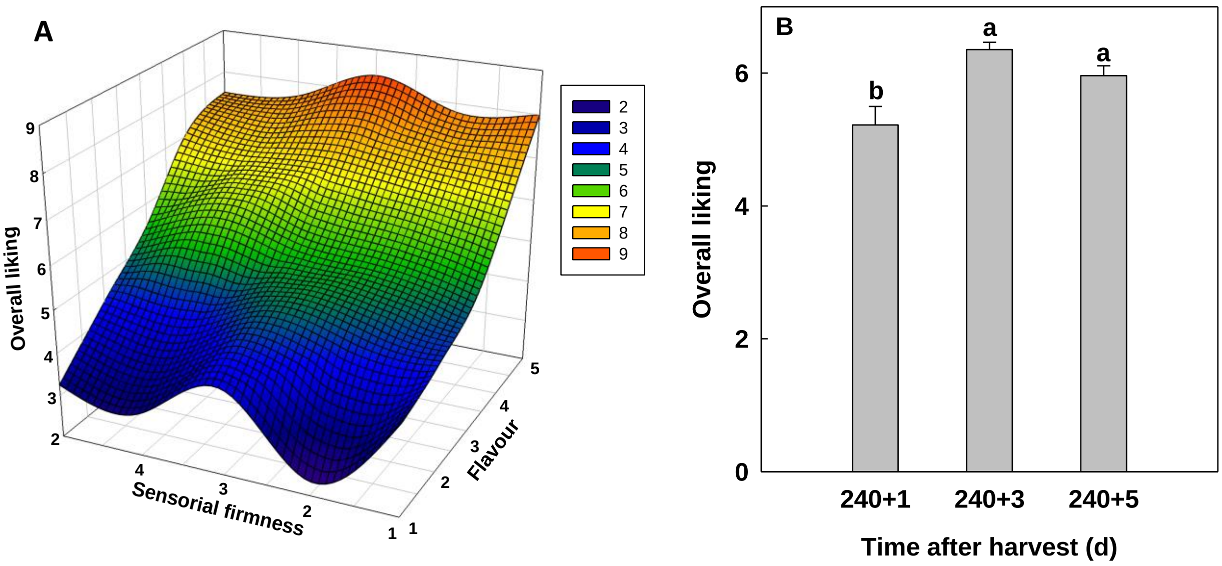
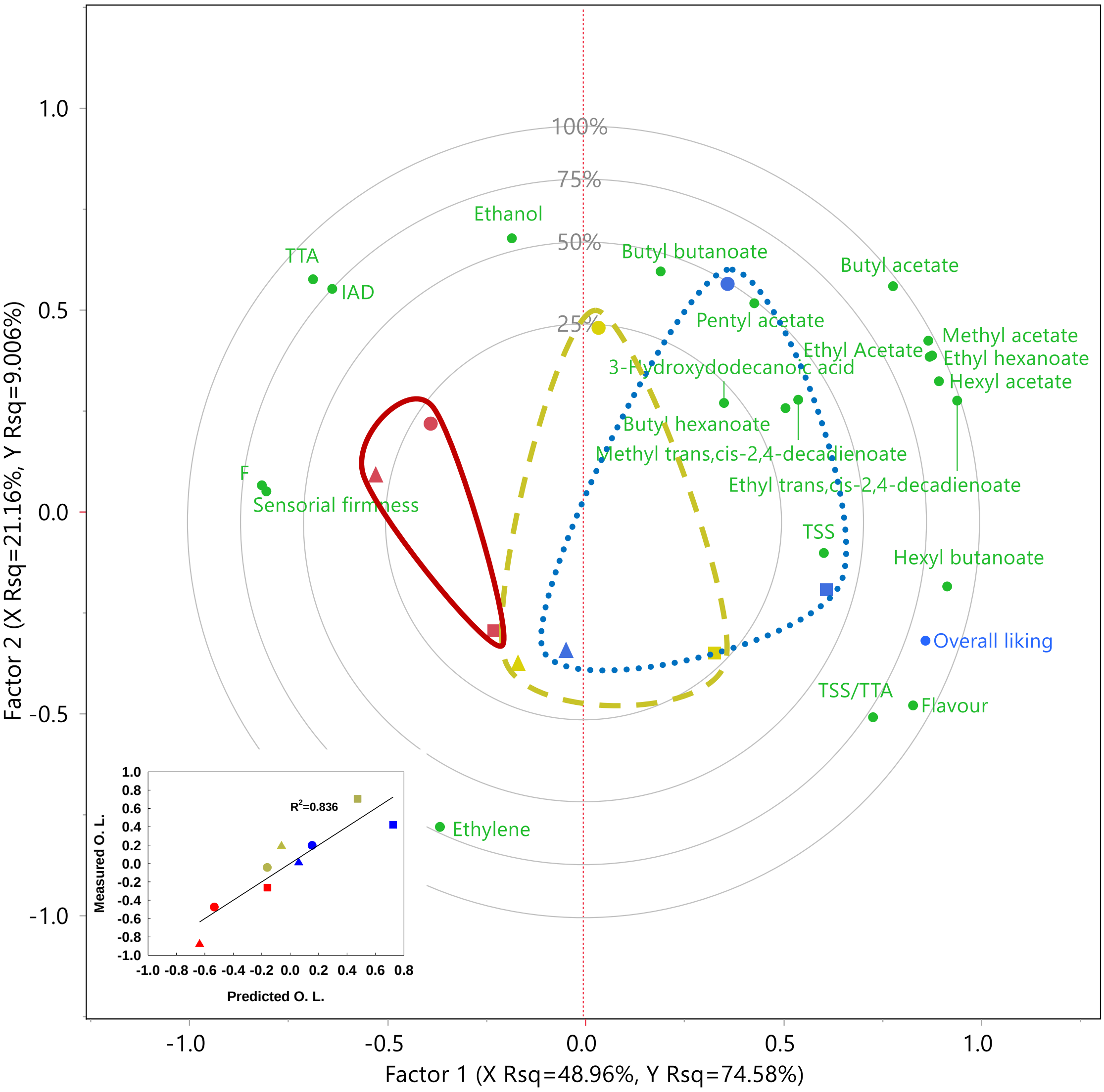

A
Overall liking
Sensorial firmness
Flavour
B
Overall liking
Time after harvest (d)
240+1
240+3
240+5
b
a
a

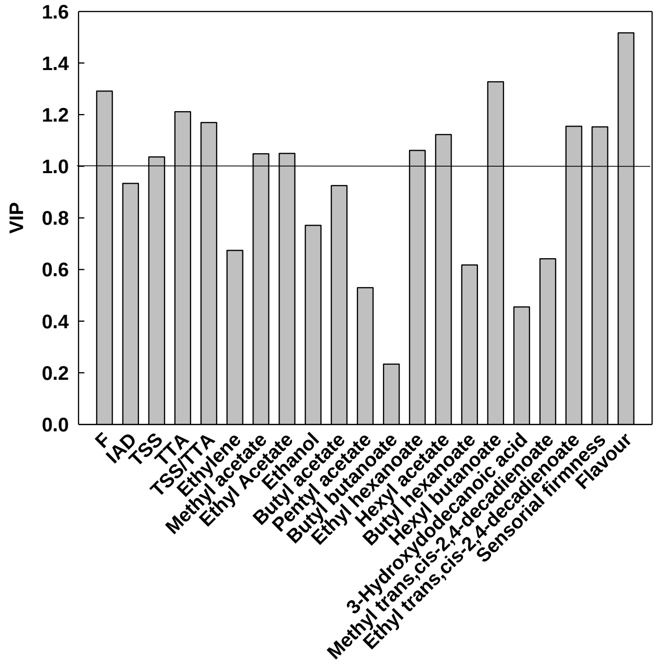
Figure 5



Factor 2 (X Rsq=21.16%, Y Rsq=9.006%)
Factor 1 (X Rsq=48.96%, Y Rsq=74.58%)
1.0
0.5
0.0
-0.5
-1.0
100%
75%
50%
25%
Ethanol
TTA
IAD
Butyl butanoate
Butyl acetate
Pentyl acetate
Methyl acetate
Ethyl Acetate
Ethyl hexanoate
3-Hydroxydodecanoic acid
Hexyl acetate
Butyl hexanoate
Methyl trans,cis-2,4-decadienoate
Ethyl trans,cis-2,4-decadienoate
F
Sensorial firmness
TSS
Hexyl butanoate
Overall liking
TSS/TTA
Flavour
Ethylene
R²=0.836
Measured O. L.
Predicted O. L.
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
-0.6
-0.8
-1.0

Figure 6

664



665

666 **Figure 7**