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Aroma Composition in Relation with Quality of 'Peento' Flat Peach during Fruit Development

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ABSTRACT

'Peento' flat peach (*Prunus persica* L.), increasingly encountered in Tunisian agrosystems, is characterized by tasty and flavoured fruit with a marked and specific aroma that develops when ripe. Volatile compounds of 'Peento' peaches were separated using liquid-liquid extraction with pentane and di-ethyl ether and analysed by gas chromatography/flame ionisation detection (GC/FID). Peach samples produced an average of 3674 µg aroma/g fresh weight (FW) at full ripeness. Lactones was the major class of volatiles described with δ-dodecalactone as the main compound (18.4%) followed by δ-undecalactone (15.3%). Other constituents, including ketones (2), esters (1), alcohols (1), monoterpenes (1) and sesquiterpenes (1), were also identified. 'Elegant Lady' fruits, gathered during the same period, produced 2072 µg aroma/g FW. Methyl isovalerate (13.0%) and δ-undecalactone (7.9%) were the major compounds identified. δ-Dodecalactone was less abundant in the fruit aroma (3.3%) compared to flat peaches. Changes in volatile composition showed a significant increase until maturity. Besides, volatile concentrations decreased. Lactones, the greatest contributors to the peachy note, continued to rise independently of total volatiles produced. The quality of 'Peento' flat peach is influenced by its aroma composition, in particular to the level of lactones in the parenchyma. The date of harvest seemed to be of great importance in preserving fruit quality attributes. Physiological maturity is considered to be the optimal time for harvest. Anticipated as well delayed harvest could result in a dramatic loss of scent and aroma developed by the fruit. The emission of volatile compounds appears to be a useful index for determining the physiological maturity of 'Peento' peaches in an orchard prior to harvest.

Keywords: peach *Prunus persica*, flavour, lactones, δ-dodecalactone, physiological maturity
Abbreviations: ANOVA, analysis of variance; **FW**, fresh weight; **GC/FID**, gas chromatography/flame ionisation detector; **HS/SPME**, head-space/solid phase micro extraction; **IS**, Internal standard; **KI**, Kovàts Index; **SAS**, Statistical analysis system

INTRODUCTION

The ultimate objective of production, handling and distribution of fresh fruits is to satisfy consumers. It is generally agreed that consumer satisfaction is related to product quality (Shewfelt *et al*. 1997). The sensory quality of the peach is linked not only to basic organic components (sugars, organic acids, fibres, micro- and macro-elements) but also to flesh texture and in great part to the volatile compounds, which define the flavour impact (Bononi *et al*. 2012). Peach volatile compound patterns have been studied with different aims, including qualitative and quantitative distribution of volatiles (Engel *et al*. 1988), characterization of peach accessions from different origins (Derail *et al*. 1999; Wang *et al*. 2009), changes in aroma composition during ripening and under different postharvest conditions (Farina *et al*. 2007; Raffo *et al*. 2008) and interaction of volatiles with human receptors (Grosch 2001). In terms of fruit quality at consumption, harvest timing is the determining factor for consumer appreciation (Farina *et al*. 2007) and it is assumed that peach fruit expresses the best aroma when completely matured (Visai and Vanoli 1997). Consequently, the identification and quantification of volatile compounds are often considered only from ripe fruits (Eduardo *et al*. 2010).

Aroma development in the fruit is a dynamic process during which concentrations on volatile compounds change quantitatively and qualitatively with ripening (Bayonove 1973). Agronomic parameters like cultivar, harvest date and environmental conditions clearly influence aroma composition of the fruit (Soing and Vaysse 1999). Volatiles produced by peach have been intensively investigated, and approximately 100 compounds have been identified, including alcohols, aldehydes, esters, lactones and terpenes. Among them, lactones, in particular γ- and δ-decalactone, have been reported to play an important role in the overall peach aroma (Robertson *et al*. 1990; Visai and Vanoli 1997; Aubert *et al*. 2003). However, they act in association with other impact compounds, such as C6 aldehydes, *trans*-2-hexenal, *cis*-3-hexenol, aliphatic alcohols, and terpenes, such as linalool, which are responsible for the spicy, grassy and floral characteristics of peach flavour (Rizzolo *et al*. 1995).

In Tunisia, flat peach cultivars are gaining more and more fields among other common cultivars. The fruit is getting more appreciation and acceptance by a large number of consumers despite its unusual form and its gripping stone. 'Peento' flat peach, economically interesting fruit tree, is famous for its melting and juicy texture, sweet flavour and pronounced aroma. Therefore, the fruit is highly favoured by consumers in all Tunisia.

The present study was undertaken to determine aroma composition and changes during ripening of 'Peento' flat peach compared to other common cultivars growing in semi-arid conditions of Tunisia with the main objective to reveal the origin of peculiar taste and fragrance characterizing the fruit at maturity and to elucidate the optimum physiological stage for fruit quality which could be helpful to decide about optimal date for harvest.

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MATERIALS AND METHODS

Plant material

'Peento' flat peaches were harvested from the down valley of Medjerda in the North-East of Tunisia (governorate of *Mannouba*). The trees, 15 years aged, are drafted on GF677 rootstock and conducted in goblet form under spacing of 5 × 2 m occupying in total 16800 m^2. Healthy and homogenous fruits were selected from all compartment of the tree in the last week of July (full ripeness stage). Cultivar 'Elegant Lady', growing in the same conditions, was selected to compare aroma profile released by the two kinds of fruit. 30 fruits from each cultivar were picked for analysis. 'Peento' peaches are medium-sized, not much coloured, firm to quite firm with a white flesh and gripped stone. 'Elegant Lady' peaches are large-sized fruits with a pronounced red skin, a yellow flesh and a free stone. These two cultivars ripen at the same period (last week of July). Aroma profile of 'Peento' peaches was analysed at three stages of maturity: pre-maturity (commercial stage of maturity) corresponding to 112 days after full bloom; full ripeness (physiological stage of maturity) which corresponds to 120 days after full bloom and over-ripeness (128 days after full bloom). 30 fruits were gathered for each stage and stored until analysis.

Solvents and chemicals

Thirty standard solutions purchased from Fluka and Sigma-Aldrich (purity > 98%) were prepared in di-ethyl ether. A phosphate buffer (K_2HPO_4+KH_2PO_4) was prepared (pH7) for samples dilution during volatiles' extraction. Pentane and di-ethyl ether (purity > 98%) were used as solvents for extraction. Terpinolene was used as internal standard (IS).

Aroma extraction

Fresh peaches from the two cultivars were stone pitted, cut in small pieces and mixed with a blender. The puree obtained was then arranged in hermetical glass jars and stored until analysis. 20 ml of the puree was mixed with 10 ml of a phosphate buffer (K_2HPO_4+KH_2PO_4 – pH 7.0). 10 ml pentane + 10 ml di-ethyl ether were added to the mixture. The overall was 15 min. stirred and samples were frozen until the two phases' separation. The organic phase was recovered and the extract was concentrated (Tonder *et al.* 1998). Concentrations step was performed by introducing the extracts in a vigreux column placed in a 38°C water bath. Concentration was stopped until 1 ml of extract.

Analytical

Analysis of volatile compounds was carried out using a HP (Hewlett-Packard) gas chromatograph 6890 series equipped with flame ionisation detector (GC/FID) and a split-splitless injection mode. The capillary column was HP-Innowax, with a polar stationary phase (polyethylene glycol) and the following dimensions: 30 m × 0.25 mm × 0.25 µm. Temperatures of injection and detection were 250°C and 300°C, respectively. Nitrogen was used as gas carrier at a flow rate of 1.6 ml/min. 0.5 µl of extract was injected. Temperature was scheduled from 35°C (held for 10 min) to 205°C (held for 10 min) at 3°C/min.

Identification of volatile compounds

Co-chromatography. Standard solutions were injected for the identification of the peaks given out after 75 min of analysis. Comparison with retention time of known standard solutions made possible identification of volatile compounds released by the extracts.

Kovàts Index. Unknown peaks were identified by the Kovàts index (KI). This index is specific for each compound and depends only on the type of the column used (polar or apolar column). A series of aliphatic hydrocarbons was injected under the same analytical conditions previously described. Retention time of one compound is set between the two hydrocarbons of reference and KI is calculated as:

$$KI_{A} = 100N + 100\,\frac{T_{R(A)} - T_{R(N)}}{T_{R(N+1)} - T_{R(N)}}$$

A: compound A
N: number of carbon atoms of aliphatic hydrocarbon which come out just before compound A
$T_{R(A)}$: retention time of compound A
$T_{R(N)}$: retention time of hydrocarbon appearing just before compound A
$T_{R(N+1)}$: retention time of hydrocarbon appearing just after compound A.

Statistics
Volatile compounds of peach fruits were checked in triplicate. The results are given as means of data results and expressed as µg/kg FW. Data were subject to one way analysis of variance (ANOVA) to make comparisons between the two cultivars and between the three ripening stages. Statistics were established using SAS (Statistical Analysis System) program version 6.10 ©1991.

RESULTS AND DISCUSSION

Aroma profile of 'Peento' flat peaches

More than 18 compounds were separated and identified in the peach puree samples (**Fig. 1**). 'Peento' peaches produced 3674 µg aroma/g FW. The major compounds identified were δ-dodecalactone (18.4%) and δ-undecalactone (15.3%). β-Ionone (5.0%), α-terpinene (3.9%) and 2-octanol (1.1%) were also identified in the aroma (**Table 1**). Lactones are the most represented compounds with more than 69% of total volatiles produced by the fruit (**Table 1**). 'Elegant Lady' fruits produced 2072 µg/g FW with more than 16 compounds separated from samples. The major compound belongs to the class of lactones with concentration reaching 1122 µg/g FW, corresponding to 54.1% of total volatiles produced by the fruit (**Table 1**). Other major

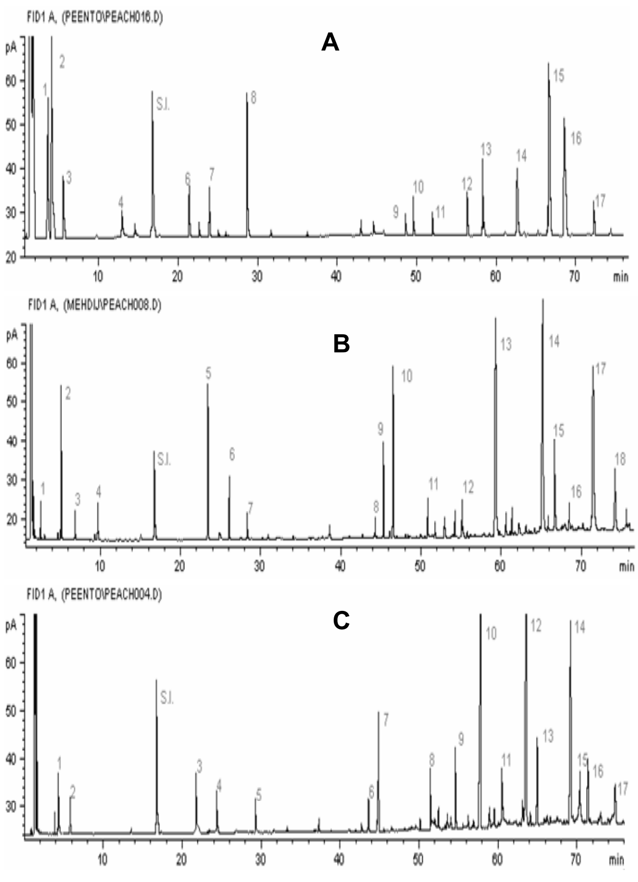

Fig. 1 Chromatograms of peach extracts harvested at three different stages of maturity: (**A**) Prematurity, (**B**) Full ripeness, (**C**) Over-ripeness.

Table 1 Concentrations in volatile compounds identified in the two peach cultivars.

			Peento		Elegant Lady	
Compound	**RT (min)**	**KI**	**Concentration (µg/g FW)**	**Percentage (%)**	**Concentration (µg/g FW)**	**Percentage (%)**
4-methyl-2-pentanone	3.70	-	-	-	83.7	4.0
methyl isovalerate	4.15	-	-	-	269.4	13.0
α-terpinene	4.97	1001	144.3	3.9	-	-
(2)-octanol	26.04	1369	40.4	1.1	-	-
decanal	28.76	1424	-	-	100.5	4.9
geranyl acetate	40.16	1693	-	-	97.0	4.7
ε-caprolactone	45.30	1831	58.8	1.6	-	-
β-ionone	46.48	1864	183.6	5.0	-	-
γ-decalactone	51.71	2015	33.1	0.9	-	-
δ-undecalactone	59.21	>2000	**563.4**	15.3	163.0	7.9
δ-dodecalactone	65.05	>2000	**674.8**	18.4	68.2	3.3
Unknown lactone	66.64	>2000	109.4	3.0	1121.8	54.1
Unknown lactone	67.31	>2000	66.1	1.8	-	-
Unknown lactone	68.20	>2000	235.5	6.4	-	-
Unknown lactone	71.32	>2000	**674.7**	18.4	139.8	6.7
Unknown lactone	74.15	>2000	122.3	3.3	-	-

FW, fresh weight; RT: retention time; KI: Kovàts Index

Table 2 Volatile compounds released by 'Peento' peach in the three ripening stages.

Compound	**RT (min)**	**KI**	**Commercial maturity (µg/g FW)**	**Physiological maturity (µg/g FW)**	**Over ripeness (µg/g FW)**
3-methyl-2-butanone	2.64	-	-	-	50.3
4-methyl-2-pentanone	3.92	-	35.5	-	14.5
methyl isovalerate	4.34	-	**139.3**	-	16.9
α-terpinene	4.97	1001	-	144.3	-
hexanal	5.51	1008	23.0	-	-
1-hexanol	21.45	1283	26.6	-	129.8
(E)-2-hexenol	24.00	1369	24.4	-	-
(2)-octanol	26.04	1369	-	40.4	-
acetic acid	28.73	1423	69.9	-	-
ethyl dodecanoate	43.55	1783	-	-	48.2
ε-caprolactone	44.81	1831	-	58.8	144.3
β-ionone	46.48	1864	-	183.6	-
γ-decalactone	51.71	2015	-	33.1	26.5
δ-undecalactone	59.21	>2000	-	**563.4**	**564.4**
δ-dodecalactone	65.05	>2000	18.3	**674.8**	31.3
Unknown lactone	66.64	>2000	**238.6**	109.4	**534.4**
Unknown lactone	67.31	>2000	36.8	66.1	132.5
Unknown lactone	68.20	>2000	-	235.5	447.8
Unknown lactone	71.32	>2000	-	**674.7**	120.5
Unknown lactone	74.15	>2000	-	122.3	79.5
Total			**613**	**2906**	**2341**

FW, fresh weight; RT: retention time; KI: Kovàts Index

compounds identified were: methyl isovalerate (13.0%) and δ-undecalactone (7.9%).

Compared to 'Elegant Lady', 'Peento' flat peach was richer in volatile compounds (3674 µg/g FW versus 2072 µg/g FW) and differences were significant between the two cultivars (p <0.05). δ-Dodecalactone, the major compound identified in 'Peento' peach, represented only 3.3% of total volatiles produced by 'Elegant Lady' fruit. Lactones, the most contributors to peachy note of the fruit, reached 2544 and 1340 µg/g FW respectively in 'Peento' and 'Elegant Lady' peaches (**Table 1**). Lactones were the major volatile components identified in 'Sudanell' peaches among 35 compounds identified in the fruit (Saura *et al*. 2003). Jia *et al*. (2004), using the Head-Space/SPME technique for extraction, identified 7 main contributing components to the aroma of 'Hakuho' peaches growing in Japan. Volatile concentrates of in-tree ripe peaches (cv. 'Gleason Early Elberta') produced 86 peaks (Do *et al*. 2006) with γ- and δ-lactones, esters, aldehydes, benzyl alcohol and d-limonene being the main volatile components identified. The liquid-liquid extraction used to analyze volatile composition of four cultivars of nectarines revealed aroma patterns composed by 10 lactones, 8 C_6 aldehydes and alcohols, 8 terpenoids, 3 esters, 4 other compounds (Engel *et al*. 1988). γ- and δ-Decalactones were the main constituents. Oblate-peach (cv. 'Xinjiang'), recently subject to aroma analysis, revealed 58 volatiles with lactones and esters being the main characteristic compounds (Cheng *et al*. 2012). In the present study, esters (methyl isovalerate and geranyl acetate) were detected only in 'Elegant Lady' peaches with almost 18% of total volatiles.

Many works on aroma composition showed that γ- , δ-decalactones and other derived lactones were usually considered as the prominent compounds of peach fruit (Riu-Aumatell *et al*. 2004). This was true for flat peaches (cv. 'Peento') grown under semi-arid conditions of Tunisia. 'Peento' fruit is characterized by predominance of lactones, the most specific compounds of the peach aroma. Three among this class represent 52% of total volatiles produced and δ-dodecalactone was the main constituent (18.4%). γ-decalactone, commonly described in peach fruit (Visai and Vanoli 1997; Aubert *et al*. 2003), was little encountered in 'Peento' fresh samples (33.1 µg/g FW). δ-decalactone was not detected in both cultivars using liquid-liquid extraction. However, ε-caprolactone, rarely described lactone, was identified in the fruit samples at small proportions (1.6%) (**Table 1**). 'Elegant Lady' fruit, grown under the same conditions, produced less volatiles while aroma blend is distinguished by abundance of lactones. Other compounds detected belong essentially to norisoprenoids, monoterpenes in 'Peento' peach and esters in 'Elegant Lady' fruit.

Changes in volatile compounds with ripening of the fruit

Chromatographs of 'Peento' peaches obtained according to the harvest date are displayed in **Fig. 1**. More than 17 compounds were separated from samples in the three ripening stages. Total quantity of volatiles produced by 'Peento' peaches at pre-maturity (commercial stage of maturity) was of 613 µg/g FW (**Fig. 1A**). The major compound belongs to the class of lactones and represents 39% of total volatiles (**Table 2**). The other compounds identified were methyl isovalerate and acetic acid with respectively 22.7% and 11.4% of the total. Two compounds from alcohols were also detected (1-hexanol and (*E*)-2-hexenol) and represents together 8% of total volatiles.

Concentrations in volatile compounds increased at full ripeness. 'Peento' peaches produced 3674 µg/g FW at this stage, i.e. six times more than concentrations recorded at commercial maturity. Differences were significant between the two harvest stages ($p < 0.05$). The major compounds identified were: δ-dodecalactone (18.4%), δ-undecalactone (15.3%) and β-ionone (5.0%). Other compounds defined were: α-terpinene (3.9%) and 2-octanol (1.1%) of total aroma. Lactones represent 69.3% of total volatiles produced by the fruit at this development phase.

Besides full maturity, there was a decline in aroma composition of the fruit. At this ripening phase, 'Peento' peaches showed a net decrease in aroma content (2409 µg/g FW) which could affect final quality of the fruit. The major compound described at over-ripeness was δ-undecalactone with concentration exceeding 564 µg/g FW (**Table 2**) followed by two unknown lactones with 22.2% (RT = 66 min) and 18.6% (RT = 68 min) respectively (**Fig. 1C**). These three major compounds represent 64% of total volatiles produced at the over-ripeness. Other compounds belonging to the class of alcohols (1-hexanol) and esters (ethyl dodecanoate) were identified with less proportion (5.4% and 2.0%, respectively). Over-ripeness stage showed an increase in concentrations of lactones. However, the total amount of volatile compounds produced by the fruit decreased. A delay in harvest time could be responsible of a loss in quality of 'Peento' peaches due to the collapse of aroma content.

It was already demonstrated that aroma composition depends on maturity level of the fruit (Bayonove 1973). Volatiles content of peaches increases continuously until ripening. Lim and Romani (1964) comparing volatiles production from peaches picked at three different stages of maturity concluded that non-ripened fruit was the poorest in volatile compounds. Great changes in volatile composition occurred during ripening of 'Glohaven' peaches (Visai *et al*. 1993). Ripe fruit were characterised by high levels of acetoin and (*Z*)-3-hexenol coupled with an increasing amount of lactones. Variation in volatile constituents during maturation of peach (cv. 'Cresthaven' and 'Monroe') showed an increase in concentrations of most compounds with maturity of the fruit (Horvat *et al*. 1990). Linalool, benzaldehyde, γ-decalactone, and δ-decalactone increased significantly during the final stages of maturation in 'Majestic' peaches with γ-decalactone being the principal volatile compound (Chapman *et al*. 1991). It was demonstrated that increased levels of volatiles also closely paralleled to seed and mesocarp growth. Pit lignification was complete about 50 days before the seed and mesocarp fully developed (Chapman *et al*. 1991).

Changes in aroma volatile composition was determined from the immature to mature stage in 'Hujingmilu' peach fruit grown in China (*Zhejiang* Province) (Zhang and Jia 2005). In immature fruits, a C6-aldehydes (*trans*-2-hexenal) and C6-alcohols (*cis*-3-hexenol) were the major components, corresponding to the low ethylene production and high respiration rates. With increasing of fruit maturity, the C6-C12 lactonic compounds, particularly γ- and δ-lactones became the dominant volatile constituents, which increased significantly at the climacteric stage. These facts suggest that ethylene production may be involved in the regulation of lactones production in maturing peach fruit (Zhang and Jia 2005).

Changes in volatile compounds affect both quantitative and qualitative pattern during development of 'Peento' flat peaches. Full ripeness ensures optimum quality of the fruit. The in-tree ripe fruits results on well developed product as volatile compounds emissions are clearly influenced by high temperatures (Crossa-Raynaud 1966) associated with intense sunlight characterizing the summer in all Tunisia. This is undoubtedly enhanced volatile compounds release at maturity. Detection of changes in volatile compounds synthesized by peach may be useful index of maturity reflecting the current physiological stage of fruit development.

CONCLUSIONS

Quality standards of 'Peento' flat peach is closely linked to aroma content of the fruit. 'Peento' peach, when ripe, release an important aroma clearly appreciated when the fruit is still on the tree. Lactones dominate the aromatic profile during the various stages of maturity offering to the fruit its peachy and creamy note. Estimation of aroma potential of peach fruit before harvest is essential to give the right judgment about quality level reached by the fruit. Full ripeness (physiological maturity) remains consequently the optimal stage during which the fruit develops its maximum of flavour. An early harvest is responsible of a dramatic loss in volatiles and thus a presumption of quality deprivation. A delayed harvest has a negative effect on final quality of peach fruit. Indeed, volatile compounds released by flat peach tend to decrease at over-ripeness this implies a transition of the fruit to the senescence. Thus, it remains advisable to make a good decision about the harvest time in the orchard. Volatile compounds released by the fruit on the tree could be a useful index to determine physiological stage of maturity and in fact, to launch the harvest campaign. Better the decision is made in the orchard, better the quality is reached by the fruit when picked and this is certainly advantageous in the post-harvest life.

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