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OLIVE OIL VOLATILE COMPOUNDS, FLAVOUR DEVELOPMENT AND QUALITY:

A CRITICAL REVIEW

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ABSTRACT

The unique and delicate flavour of olive oil is attributed to the volatile compounds that develop during and after oil extraction from the olive fruit. The formation of these volatile compounds and the fruit characteristics that affect the formation are examined in this review. The role of extraction time – temperature interactions in volatile development and other factors that impact volatile development such as fruit storage prior to oil extraction are also considered. The volatile compounds that develop during extraction become less dominant during oil storage with the emergence of volatile compounds from chemical oxidation. The presence or absence of particular volatile compounds partly explains quality differences in olive oils.

Keywords: Sensory attributes; odour threshold values; fruit ripeness; cultivar differences; processing conditions.

INTRODUCTION

Olive oil quality depends on market preferences and is based upon consumer perceptions of aroma, taste and colour, which may change over time and with location. Objectionable aroma and taste may lead to product rejection. Hence, the absence, presence and intensity of sensory defects in olive oil determine the quality (Angerosa, 2000). Both positive attributes and sensory defects in olive oil can be associated with volatile compounds.

Volatile compounds in olive oil are mainly produced by oxidation of fatty acids. It is generally agreed that endogenous plant enzymes, through the lipoxygenase pathway, are responsible for the positive aroma perceptions in olive oil whereas chemical oxidation and exogenous enzymes, usually from microbial activity, are associated with sensory defects. Both the processing and storage of the fruit and the oil contribute greatly to the flavour and overall quality of olive oil (Angerosa, 2002; Venkateshwarlu, Let, Meyer & Jacobsen, 2004).

An understanding of the stages at which volatile compounds are formed can be used to control the volatile composition of olive oil, allowing the production and consumption of better quality oils. Selection of premium olive fruit at optimum ripeness and optimum processing conditions are factors that can be used to control the process of volatile compound formation. This review discusses the influence of volatile compounds on olive oil quality and reviews the changes in volatile compound composition of both the fruit and oil that occur during processing and storage.

OLIVE OIL QUALITY

There are several ways of defining quality and perhaps there is no single universal definition that adequately satisfies all situations. In general terms quality is defined as

“The combination of attributes or characteristics of a product that have significance in determining the degree of acceptability of that product by the user” (Gould, 1992).

Olive oil quality may be defined from commercial, nutritional or organoleptic perspectives (Duran, 1990). The nutritional value of olive oil arises from high levels of oleic acid and minor components, such as phenolic compounds, whereas the aroma is strongly influenced by volatile compounds (Kiritsakis, 1998; Angerosa, 2002). Nutritional value and pleasant flavour have contributed to an increase in consumption of olive oil which has fostered cultivation of olives outside the traditional olive oil producing region of the Mediterranean into newer areas where cultivar adaptability, different climatic conditions and different agronomic practices may alter olive quality (Patumi, D'andria, Marsilio, Fontanazza, Morelli & Lanza, 2002).

The International Olive Oil Council (IOOC, 2001) and the EEC (EC, 1991) have defined the quality of olive oil, based on parameters that include free fatty acid (FFA) content, peroxide value (PV), UV specific extinction coefficients (K_{232} and K_{270}) and sensory score. In particular, the quantity of FFA is an important factor for classifying olive oil into commercial grades (Boskou, 1996; Rossell, 1986). The general classification of olive oils into different commercial grades is based on FFA (Table 1) and sensory characteristics (taste and aroma).

The commercial grades separate oil obtained from the olive fruit solely by mechanical or physical means (virgin) from the other oils that contain refined oils.

Providing the olive fruit is sound, at production most olive oil is extra-virgin. When the fruit quality is low, the oil is refined. The classification of olive oil is usually done just after production. However, stability to oxidation is an important requirement excluded in the regulation; such oxidation can lead to a subsequent loss of extra-virgin quality status (Monteleone, Caporale, Carlucci & Pagliarini, 1998). Some parameters that are not included in the IOOC and EC standards (IOOC, 2001; EC, 1991), such as phenolic content, are known to have a significant effect on the stability and sensory characteristics of olive oil. The phenol profile can be followed from the fruit to the oil production and through storage, and may serve as a good indicator of olive oil quality. Indeed, there have been proposals to include phenols in the olive oil standard (Blekas, Psomiadou, Tsimidou & Boskou, 2002; Psomiadou, Konstantinos, Blekas, Tsimidou & Boskou, 2003; Ranalli, Ferrante, De Mattia & Costantini, 1999).

In most cases quality parameters change by the time the oil reaches the consumer (Gutierrez & Fernandez, 2002). Olive oil is susceptible to both hydrolytic and oxidative reactions (Duran, 1990) that can adversely affect oil quality parameters. For instance, an increase in PV, K_{232} and K_{270} values and development or loss of certain volatile compounds is very common between extraction and consumption (Boskou, 1996; Gutierrez & Fernandez, 2002). The presence or absence of particular volatile compounds may also be a good indicator of olive oil quality changes.

VOLATILE COMPOUNDS

Volatile compounds are low molecular weight compounds (less than 300 Da) which vaporise readily at room temperature. Some volatile compounds reach the olfactory epithelium, dissolve into the mucus and may bond with olfactory receptors to give an odour sensation (Angerosa, 2002). The aroma of olive oil is attributed to aldehydes, alcohols, esters, hydrocarbons, ketones, furans and, probably, other as yet unidentified volatile compounds. The major volatile compounds reported in virgin olive oils are the C6 and the C5 volatile compounds. Hexanal, *trans*-2-hexenal, hexan-1-ol and 3-methylbutan-1-ol are found in most virgin olive oils in Europe (Angerosa, 2002; Aparicio, Morales & Alonso, 1997; Kiritsakis, 1998). A study of Italian, Spanish and Moroccan extra-virgin olive oil (Reiners & Grosch, 1998) confirmed the richness of C6 volatile compounds in Italian oils but showed they were poor in fruity esters. The fruity esters, ethyl isobutyrate, ethyl butyrate, ethyl 2-methylbutyrate, ethyl 3-methylbutyrate, and ethyl cyclohexylcarboxylate were rich in Moroccan extra-virgin olive oils (Reiners & Grosch, 1998). It should be noted that the high concentration volatile compounds are not necessarily the major contributors of odour. For instance, Reiners & Grosch (Reiners & Grosch, 1998) reported a concentration of 6770 µg/g for *trans*-2-hexenal with an odour activity value of 16 whereas 1-penten-3-one with a much lower concentration of 26 µg/g had a higher odour activity value of 36.

Volatile compounds, whether major or minor, are crucial to olive oil quality. Volatile compounds that occur in olive oil below their olfactory threshold, and make no direct contribution to the aroma, could be important in understanding the formation and degradation of the volatiles with significant contribution to aroma, and they may provide useful quality markers (Buttery & Takeoka, 2004). This fraction includes C5 carbonyl compounds, pentenols,

hydrocarbons and minor compounds not derived from fatty acid transformations (Angerosa, Camera, D'alessandro & Mellerio, 1998; Buttery & Takeoka, 2004).

Cultivar, geographic region, fruit maturity, processing methods and parameters influence the volatile composition of olive oil. Fruit from different cultivars grown under the same environmental conditions produce oils with different volatile compounds, as does fruit of the same cultivar grown in different geographic regions (Angerosa, Basti & Vito, 1999; Benincasa, De Nino, Lombardo, Perri, Sindona & Tagarelli, 2003; Prenzler, Bedgood, Bishop & Robards, 2002; Ridolfi, Terenziani, Patumi & Fontanazza, 2002; Sacchi, Mannina, Fiordiponti, Barone, Paolillo, Patumi & Segre, 1998). The aroma compounds in an oil are known to increase with the degree of fruit maturity up to a certain point (Aparicio & Morales, 1998; Kiritsakis, 1998; Ranalli, Tombesi, Ferrante & De Mattia, 1998). Apart from the condition of the fruit at harvest, differences in post-harvest handling of the fruit and oil lead to different volatile profiles. Extraction methods and conditions, in particular the malaxation time and temperature, produce olive oils with different flavours (Angerosa, D'alessandro, Basti & Vito, 1998; Di Giovacchino, Sestili & Di Vincenzo, 2002; Ranalli, Pollastri, Contento, Iannucci & Lucera, 2003; Ranalli, Contento, Schiavone & Simone, 2001). Storage of the fruit after harvest and the of oil before reaching the consumer changes the volatile composition of olive oil. Storage of the fruit decreases the aldehyde and ester content that is responsible for the positive aroma. Storage of either the fruit or oil produces volatile compounds that are responsible for off-flavours (Kiritsakis, 1998; Koprivnjak, Procida & Zelinotti, 2000). The absence of the C6 aldehydes, alcohols and esters from the lipoxygenase pathway and the presence of many aldehydes from chemical oxidation, including hexanal from both chemical and enzymatic reactions, characterise the off-flavour

of olive oil. The off-flavour compounds are potentially toxic and have low odour thresholds (Angerosa, 2000; Ha, Nihei & Kubo, 2004).

FORMATION OF VOLATILE COMPOUNDS

Volatile compounds are not produced in significant amounts during fruit growth but arise during the climacteric stage of ripening. During the climacteric period fruits produce ethylene, inducing biochemical, physical and chemical changes and an increase in some proteins and enzyme activities. In olives, the climacteric phase corresponds to a period when oil extracted from drupes gives an elevated oil quality that is rich in aromatic volatile compounds (Ranalli et al., 1998). The majority of these aromatic volatile components are formed through the action of enzymes that are released when the fruit is crushed, and continue to form during malaxation (Olias, Perez, Rios & Sanz, 1993; Tressl & Drawert, 1973).

Volatile production whether during the climacteric phase (or earlier) or during oil processing involves several different pathways (Buttery & Takeoka, 2004) although the (Tressl & Drawert, 1973) volatile compounds in virgin olive oil are mainly formed by chemical and enzymatic oxidation. The volatiles formed from chemical oxidation of the oil are responsible for the off-flavour referred to as oxidative rancidity. In contrast, enzymatic oxidation of olive oils, especially through the lipoxygenase pathway, is considered responsible for the aroma of the oil (Angerosa, 2002; Kiritsakis, 1998). One pathway is the enzymatic splitting of linoleic and linolenic acid into C6 and C9 aldehydes and C9 and C12 oxo acids (Fig 1). Volatile compounds are also formed through fatty acid metabolism producing acids, alcohols, esters and ketones. The lipoxygenase pathway is a biochemical

reaction scheme that accounts for most of the aroma fraction of olive oil containing C6 aldehydes, alcohols and esters (Angerosa, 2002; Kiritsakis, 1998). Although volatile compounds are also formed from amino acids, the action of amino acids in olive oil volatile generation has been given little attention. It is established that valine and leucine are converted to volatile compounds including methyl-branched alkyl and acyl compounds of esters and into methyl-branched alcohols, which have the potential to change the sensory perception (Tressl & Drawert, 1973).

The presence of other minor volatile compounds may provide useful quality markers and lead to an improved understanding of the formation or degradation of the major volatile compounds (Angerosa et al., 1998; Buttery & Takeoka, 2004). Moreover, an understanding of the various pathways can improve the production and storage of premium quality olive oil.

ENZYME ACTION IN VOLATILE COMPOUNDS FORMATION

The lipoxygenase pathway is initiated by the release of enzymes when olive fruit tissues are disrupted. The reaction pathway involves a series of enzymes that oxidise (lipoxygenase) and cleave (hydroperoxide lyase) polyunsaturated fatty acids to yield aldehydes. These are subsequently reduced to alcohols (by alcohol dehydrogenase) and esterified to produce esters (by alcohol acyltransferase). The different stages in the lipoxygenase pathway are detailed in Figure 1.

Acyl hydrolase and lipoxygenase action

In the first step of volatile formation, acyl hydrolase (AH) hydrolyses triglycerides and phospholipids to release free fatty acids. Lipolytic AH is a group of enzymes that include lipases, phospholipases and galactolipases. AH exhibits a narrow range of pH activity in the basic range with optimum activity at pH 8.5 (Table 2).

Hydroperoxides are formed when the fatty acids released by the action of AH are oxidised through the action of lipoxygenase (LOX). LOX shows regiospecificity for the Δ -13 position of both linoleic and linolenic acid, yielding 75-90% of Δ -13 fatty acid hydroperoxides. The enzyme is more active with linolenic acid than linoleic acid by a factor of two (Salas, Williams, Harwood & Sanchez, 1999; Sanchez & Salas, 2000). The higher LOX activity for linolenic acid than linoleic acid supports the biogenesis of more of the six-carbon unsaturated volatile compounds (Fig. 1), which are the major constituents of the virgin olive oil aroma (Salas et al., 1999). Olive fruits show the highest LOX activity 15 weeks after anthesis and the activity decreases during the development and ripening periods (Salas et al., 1999).

The most common LOX activity has been observed in acidic conditions (Table 2) which is similar to the acidic nature of the olive paste (Williams, Salas, Sanchez & Harwood, 2000) encountered during oil extraction. However, the maximum LOX activity has been observed in the alkaline range in olive callus cultures (Williams et al., 2000). The acidic and alkaline pH ranges for LOX activity might suggest the existence of different LOX isoforms (Williams et al., 2000). The observed differences may also be attributed to the different methods used to extract the enzymes as well as the different cultivars tested.

LOX has been shown to be thermally unstable. At 60°C, the LOX activity is reduced to less than 10% within 1 minute (Anthon & Barrett, 2003). The results did not rule out thermal stability of a minor form of the enzyme that is responsible for 13-hydroperoxy lipid formation (Anthon & Barrett, 2003). Different thermal stabilities have been reported for LOX (Table 2) and these might also be attributed to the existence of different isoforms.

Hydroperoxide lyase and *cis-3: trans-2*-enal isomerase action

Hydroperoxide lyase (HPL) catalyses the cleavage of fatty acid hydroperoxides, producing volatile aldehydes and oxoacids (Fig 1). The HPL enzyme can yield C6 aldehydes and C12 ω -oxoacids from the 13-hydroperoxides of linolenic or linoleic acid, or C9 aldehydes and C9 ω -oxoacids from the 9-hydroperoxide derivatives of the same fatty acids, depending on the substrate specificity of the enzyme. The isoform that uses 13-hydroperoxides is the most abundant and widespread HPL enzyme in the plant kingdom (Sanchez & Salas, 2000). The HPL isoform that strictly cleaves 9-hydroperoxides is responsible for the characteristic cucumber-like odour of some fruits and vegetables, whereas the enzyme isoform that uses 13-hydroperoxides produces C6 aldehydes responsible for 'green' aroma (Olias et al., 1993; Salas & Sanchez, 1999; Sanchez & Salas, 2000). The cleavage of 13-hydroperoxides forms C6 aldehydes (Fig 1) that include the saturated aldehyde, hexanal from linoleic acid and the unsaturated aldehyde, *cis-3*-hexenal from linolenic acid. The unsaturated aldehyde, *cis-3*-hexenal is unstable and undergoes rapid isomerisation to a stable compound, *trans-2*-hexenal with the aid of *cis-3: trans-2*-enal isomerase enzyme (Williams et al., 2000). The aldehydes formed through HPL activity and isomerised with the aid of *cis-3: trans-2*-enal isomerase enzyme are further reduced to alcohols.

The highest level of HPL activity is detected in green olive fruits harvested at the initial developmental stages. There is a slight decrease at maturity, but a high activity level is maintained throughout maturation. Consequently, the decrease in the concentration of C6 volatile compounds in the olive oils of mature olive fruits is not attributed to HPL activity (Salas & Sanchez, 1999). This suggests that the limiting factor in volatile aldehyde formation may not be the level of HPL but the availability of the substrate. It has been reported that olive stones contain enzymes other than HPL that metabolise 13-hydroperoxides, resulting in a net decrease of unsaturated C6 aldehydes (Luaces, Perez & Sanz, 2003).

HPL has been shown to be heat labile (Anthon & Barrett, 2003) and displays optimal activity in slightly acidic conditions (Table 2). HPL activity can be rapidly reduced to a few percent of its original activity at elevated temperatures (Anthon & Barrett, 2003). Maximum activity has been observed at 15°C with a clear decline at 35°C (Table 2). Heat treatments between 60 and 68°C have been reported to promote a partial deactivation of the LOX/HPL enzyme system, reducing the synthesis of C6 and C5 compounds (Perez, Luaces, Rios, Garcia & Sanz, 2003).

Alcohol dehydrogenase action

Alcohol dehydrogenase (ADH) catalyses the reversible reduction of aliphatic aldehydes to alcohols (Fig 1). ADH is widespread in the plant kingdom and is responsible for the formation of volatile alcohols that contribute to the aroma of vegetable products (Sanchez & Salas, 2000). There is a decline in ADH activity when the olive fruit colour changes to purple during the ripening process. This supports the analytical observation of a

decrease in the content of C6 alcohols in the aroma of olive oils as the fruit ripeness increases (Salas & Sanchez, 1998). Olive stones seem to be a good source of ADH which is more specific for saturated C6 aldehydes (Luaces et al., 2003). The pH range for ADH activity is between 5.0 and 8.5 with an optimum at 6.8 (Table 2).

Alcohol acetyl transferase action

Alcohols, produced by the action of ADH, can form volatile esters. Alcohol acetyl transferase (AAT) catalyses the formation of acetate esters through acetyl-CoA derivatives. Acetate esters, and esters of alcohols with other fatty acids are important constituents of many fruits. In olive oils, ethyl propionate and hexyl acetate are significant components of the sweet, fruity note (Sanchez & Salas, 2000; Williams, Morales, Aparicio & Harwood, 1998). Olive AAT displays no activity with the short-chain alcohols such as methanol and ethanol and shows low activity towards butanol and 3-methylbutanol (Sanchez & Salas, 2000). Lack of activity towards short chain alcohols explains the scarcity of hexenyl acetate in olive oil although the concentration of the precursors (*cis*-3-hexenol and *trans*-2-hexenol) is dominant among the volatile alcohols (Salas, 2004). This lack of activity also suggests that ethyl acetate, an ester commonly detected in olive oil, may be synthesised through a different pathway (Salas, 2004). Similar observations of lack of AAT activity towards short chain alcohols are made for strawberry AAT where hexanol is the preferred substrate with decreasing esterification rates for butanol, isomyl alcohol, propanol and ethanol (Perez, Sanz & Olias, 1993). Banana AAT is a more selective enzyme forming acetate esters and only very low amounts of propionate and butyrate esters (Perez, Sanz, Olias, Rios & Olias, 1996).

The maximum activity for AAT in olives is found with hexanol and *cis*-3-hexenol; *trans*-2-hexenol is a poorer substrate (Salas, 2004).

The optimum pH range for AAT activity appears in the neutral to basic range (Table 2) with a rapid decrease in the acidic range (Salas, 2004). The AAT activity shows a maximum at 35°C (Table 2), above which there is a marked decrease in activity (Perez et al., 1993). Different temperature ranges for AAT activity were observed when the enzyme activity was monitored through its products, the volatile ester compounds. Heat treatments between 60 and 68°C did not affect AAT activity such that the C6 ester contents in olive oil remained almost constant (Perez et al., 2003). However, there was an accumulation of alcohols when the activity of AAT was lower than the previous enzymes in the pathway. Increasing the AAT activity can enhance the production of volatile esters responsible for the fruity and sweet aroma in olive oil. AAT activity can be enhanced through cultivar selection and by modifying the extraction process such as operating at lower temperatures to prevent enzyme inactivation and promote increased esterification activity (Salas, 2004).

Olive fruit characteristics and volatile formation

Olive fruit maturity and cultivar type influence the formation of volatile compounds in oil and are important factors in determining the olive oil quality. Most of the enzymes involved in the formation of volatile compounds through the lipoxygenase pathway decrease in activity with fruit maturity, for example ADH (Salas & Sanchez, 1998) and LOX (Salas et al., 1999). This supports the observation of a decreased content of C6 volatile compounds, especially those from linolenic acid, in the aroma of olive oils with an increase in the degree of fruit ripeness (Salas & Sanchez, 1998). As noted above, HPL maintains high

activity throughout maturation, ruling out an influence of it on the decrease in C6 volatile compounds in olive oils with fruit maturity (Salas & Sanchez, 1999).

Most of the C6 aldehydes reach a maximum when olive skin pigmentation changes from green to purple (Angerosa & Basti, 2001). At early ripening stages, the amounts of C6 aldehydes are comparable to those of alcohols (Angerosa & Basti, 2001). *Trans*-2-hexenal, the main volatile compound in most European extra-virgin olive oil, decreases with ripeness in most of the cultivars. A decrease in *trans*-2-hexenal with maturity has also been observed in apples (Mattheis, Fellman, Chen & Patterson, 1991). The decrease is observed for most of the aldehydes produced from the lipoxygenase pathway except for *cis*-3-hexenal, which increases with ripeness (Aparicio & Morales, 1998). Preliminary results from our laboratory indicate that the decrease in C6 aldehydes from the lipoxygenase pathway might not be characteristic of all olive cultivars (Kalua, Bedgood, Bishop & Prenzler,). A similar observation (Benincasa et al., 2003) of a cultivar dependence of volatile composition has been made for oil from olive fruits at different stages of maturation. For example, the concentration of hexan-1-ol decreased in cultivar *Nocellara del Belice* but increased in cultivar *Coratina* with ripeness (Benincasa et al., 2003). Regardless of these observations, it has been reported (Aparicio & Morales, 1998) that hexan-1-ol does not contribute to ripeness characterisation. The major reported indicators for ripeness in olive oil are *trans*-3-hexen-1-ol, *cis*-3-hexen-1-ol, *trans*-2-hexen-1-ol, hexanal and hexyl acetate (Aparicio & Morales, 1998).

The differences in C6 and C5 volatile content of oils may also be related to geographic region. For example, the dominance of *trans*-2-hexenal in the volatile profile of European extra-virgin olive oils (Cavalli, Fernandez, Lizzani-Cuvelier & Loiseau, 2004) is

not (Kiritsakis, 1998) always seen in oils from other regions. One study (Reiners & Grosch, 1998) confirmed the richness of C6 aldehydes in Italian oils but found that fruity esters were dominant aroma compounds in Moroccan oils. The variation in levels of C6 aldehydes and alcohols for oil samples from different regions implies that environmental growth conditions may influence the activity of ADH. Differences in the levels of esters in olive oil were not observed between zones in one study, suggesting less dependence of AAT activity on climatic conditions (Vichi, Pizzale, Conte, Buxaderas & Lopez-Tamames, 2003b). It has been suggested that the accumulation of volatile compounds is dominated by the variety with climate and environmental factors probably having an indirect effect by modifying the degree of ripeness (Angerosa et al., 1999). Although there is a link between the variety and the accumulation of volatile compounds, it is not clear if this is the case for all volatile compounds. Vichi *et al.* (Vichi et al., 2003b) observed no difference in the C5 volatile compounds with respect to cultivar, suggesting that the geographic growth area is the main influence on the formation of these compounds.

Fruit storage before oil processing is not encouraged in olive oil production. Good practice in fruit handling recommends that the fruit should be processed as soon as possible after harvest, without storage (Di Giovacchino, 2000). The fruit deteriorates during storage through the action of pathogenic micro-organisms and senescence processes, fermentation and as a result of fruit mechanical damage (Agar, Hess-Pierce, Sourour & Kader, 1998; Garcia, Gutierrez, Castellano, Perdiguero, Morilla & Albi, 1996). The volatile compounds increase due to intensified enzyme activity probably due to a gradual disintegration of the cell structure (Koprivnjak et al., 2000). The oil extracted from degraded fruits usually has high acidity, low stability and a characteristically undesirable odour (Garcia et al., 1996).

Studies in volatile formation during storage have concentrated on the oil with a bias towards the negative aspects of oil quality. Less emphasis has been placed on the effect of fruit handling before oil extraction. Olive fruit storage for 10 days in cool dry air resulted in almost 90% loss of hexanal while *trans*-2-hexenal doubled in concentration and there was a big increase in hexan-1-ol (Koprivnjak et al., 2000). Better sensory properties were observed for air storage of olive fruits for 30 days, with a decrease in 1-penten-3-ol and 2-methyl-1-butanol, compounds that are linked to mouldy and rancid defects (Koprivnjak et al., 2000). It has also been observed that fruit storage in air increases *trans*-2-hexenal and decreases some of the volatile compounds responsible for defects (Koprivnjak et al., 2000); a change that can potentially be exploited to enhance the quality of olive oil.

Processing conditions and volatile formation

It is generally agreed that high quality olive fruit produces a premium olive oil, yet extreme conditions in the processing of olive fruit can affect both the quantity and quality of olive oil, particularly as the production of desirable volatile compounds is dependent on the action of enzymes, which have different optimum temperature for activity (Table 2).

Malaxation temperature and time are the two main parameters that can be controlled during processing to potentially change the sensory properties of the oil. Raising the temperature of the olive paste reduces viscosity, making it easier to separate and obtain high yields.

However, raising the processing temperature reduces the quality of the oil (Amirante, Dugo & Gomez, 2002). A range of views has been published on suitable time-temperature combinations for malaxing olive pastes.

Research has shown that malaxing the olive paste at 30°C achieves both pleasant “green” virgin olive oil and satisfactory oil extraction outputs, but that 35°C introduces numerous defects into the oil without substantially increasing the oil yield (Morales & Aparicio, 1999; Ranalli et al., 2001). Long malaxing times are associated with a significant increase in the total volatile compounds, a decrease in volatile compounds responsible for the pleasant aroma of virgin olive oil and elevated production of 2-methyl butanol and 3-methyl butanol (Angerosa, Mostallino, Basti & Vito, 2001; Ranalli et al., 2003) associated with sensory defects (Table 3). Olive oil extracted from Corregiola grown in Australia showed very little difference between the amounts of volatile compounds produced with malaxation temperatures of 25°C or 35°C, or between malaxation times of 15 and 60 minutes (Tura, Prenzler, Bedgood, Antolovich & Robards, 2004). This is inconsistent with reports from Europe where temperature has been shown to affect the volatile profile (Morales & Aparicio, 1999; Ranalli et al., 2001). Geographic and cultivar influences might explain the high processing temperature tolerance observed (Tura et al., 2004), and different processing parameters may be required in differing geographic growth regions to produce high quality virgin olive oil. It has previously been observed that the optimum processing parameters also vary with cultivar (Servili, Selvaggini, Taticchi, Esposito & Montedoro, 2003).

The pH of olive paste can influence the volatile compound composition of virgin olive oil, but its effect has not been widely explored. Enzymes with an optimum activity in the basic pH range are AH and AAT (Table 2) while those with an optimum in the acidic range are LOX, ADH and HPL (Table 2). Neutralising the acidic olive paste during malaxation has been suggested as a means to enhance AAT activity and promote the production of volatile esters that are responsible for the fruity and sweet aroma in olive oil (Salas, 2004). In addition to its impact on the aroma of virgin olive oil, the alteration of paste pH should take

into account other quality parameters and the minor components in olive oil such as phenolic compounds that are important for both oxidative stability and flavour.

VOLATILE COMPOUNDS AND OLIVE OIL FLAVOUR

The volatile compounds formed during the processing of olive fruit contribute a combined sensation of smell and taste, commonly called flavour. Evaluation of the sensory quality of virgin olive oils involves perception of both favourable and unfavourable sensory attributes, with evaluation of sensory defects being used to classify oils into various grades. The IOOC provides some reference standards to evaluate virgin olive oil sensory quality, however there are some limitations in their range and stability (Angerosa, 2000).

A specific vocabulary has been developed for virgin oil sensory descriptors (IOOC, 1987). The positive attributes of virgin olive oil are explained below.

- (i) **Fruity:** the basic positive attribute of virgin olive oil, characteristic of oil from healthy, fresh fruits, either ripe or unripe. The aroma of the oil from unripe olives is generally characterised by grassy or leafy attributes whereas virgin olive oil from ripe fruits is characterised by aromatic flavours (IOOC, 1987).
- (ii) **Bitter:** the primary taste produced by dilute aqueous solutions of various substances such as quinine, caffeine and many alkaloids. It is the characteristic taste of olive oil from olives that are green or turning colour (IOOC, 1987). Although not contributing to bitter taste, the occurrence of 1-penten-3-one is positively correlated to bitter taste, whereas *cis*-3-hexen-1-ol and hexanal are negatively correlated (Angerosa, 2000).

- (iii) **Pungent:** the biting tactile sensation characteristic of oils produced at the start of the crop year, primarily from olives that are unripe (IOOC, 1987). A volatile compound positively correlated to pungency is 1-penten-3-one whereas *trans*-2-hexenal and hexanal are negatively correlated (Angerosa, 2000).

The sensory quality of the oil is modified due to the presence of defects. The common defects are described using the vocabulary below.

- (i) **Fusty:** a characteristic flavour of oil from olives stored in piles of notable thickness or in jute sacks for long periods before extraction and undergoing an advanced stage of anaerobic fermentation. A common defect, especially with small processing plants that lack sufficient fruit storage space (Angerosa, 2000; IOOC, 1996; Morales, Luna & Aparicio, 2005). The total quantity of volatile compounds is high in fusty oil, with esters and acids contributing significantly to the fusty perception (Morales et al., 2005).
- (ii) **Musty – humid:** a characteristic flavour of oils from fruit infested with large numbers of fungi and yeast as a result of storage at low temperature and high humidity. Fungi have the ability to oxidise free fatty acids to volatile compounds such as 2-heptanone and 2-nonanone. On the other hand, yeasts readily reduce carbonyl compounds (Angerosa, 2000; IOOC, 1996; Morales et al., 2005). Musty-humid oil has a low concentration of *trans*-2-hexenal and contains volatile compounds not present in extra-virgin olive oil such as C8 volatile compounds and short chain fatty acids (Morales et al., 2005)).

- (iii) **Muddy sediment:** a characteristic flavour of oil that has been left in contact with the sediment for a long time (Angerosa, 2000; IOOC, 1996).
- (iv) **Winey – vinegary:** a flavour mainly due to the process of fermentation in the olives, leading to the formation of acetic acid, ethyl acetate and ethanol. It is a flavour reminiscent of wine or vinegar (Angerosa, 2000; Garcia-Gonzalez & Aparicio, 2002; IOOC, 1996; Morales et al., 2005).
- (v) **Metallic:** a flavour, reminiscent of metals that occurs in oil that has been in prolonged contact with metallic surfaces during crushing, mixing, pressing or storage (Angerosa, 2000; IOOC, 1996). 1-penten-3-one has been proposed as a useful marker of metallic off-flavour (Venkateshwarlu et al., 2004).
- (vi) **Rancid:** a flavour of oils that have undergone oxidation. The main contributors are unsaturated aldehydes (Angerosa, 2000; IOOC, 1996; Morales et al., 2005).

Perceived sensory attributes usually arise from the influence and interaction of several volatile compounds, rather than the action of a single compound. Several volatile compounds that contribute to the aroma of virgin olive oil have been identified and quantified (Angerosa, 2002; Venkateshwarlu et al., 2004). A sensory role is reported for *cis*-4-heptenal, which enhances the effect of 1-penten-3-one in developing a metallic defect and also enhances the development of fishy defect with *trans, cis*-2, 6-nonadienal. However, the presence of *cis*-4-heptenal, in the absence of *trans, cis*-2, 6-nonadienal minimises the intensity of fishy odour (Venkateshwarlu et al., 2004).

Flavour perception

The minimum concentration of a compound able to give rise to an olfactory response is the compound's odour threshold value. Odour threshold values for flavour compounds are determined by dissolving the substance in a selected matrix, then identifying the minimum concentration that is reliably detectable to a sensory panel. More than 120 volatile compounds that contribute both positively and negatively to the sensory properties of olive oil have been identified (Aparicio & Luna, 2002). Some of the common volatile compounds with their sensory characterisation and odour threshold are arranged based on their functional groups and carbon number in Table 3.

Comparison of odour thresholds is difficult, since different values may be reported for the same volatile compound (Table 3). Variations in odour threshold value can be attributed to different experimental conditions, particularly variations of the sample matrix. Odour threshold values have been determined in deodorised refined sunflower oil (Aparicio & Morales, 1998; Reiners & Grosch, 1998), refined vegetable oil (Aparicio & Luna, 2002), paraffin oil (Badings, 1970; Meijboom, 1964) and a fully refined deodorised olive oil (Morales et al., 2005). In some cases, the volatile compounds are diluted in water or paraffin oil, depending on solubility, prior to odour threshold determination (Baeten, Hourant, Morales & Aparicio, 1998). A similar matrix to olive oil should be used if odour threshold values are to meaningfully apply to olive oil. The presence of carbohydrates, proteins and other minor components in olive oil can decrease the aroma intensity through sorption, binding and formation of intermolecular complexes (Jung, De Ropp & Ebeler, 2000). Further complicating the comparison of odour thresholds is the difficulty, noted by Angerosa (Angerosa, 2002), in harmonising the sensory definitions across sensory evaluation panels, even where the same vocabulary is used. For instance, the sensory characterisation of

hexanol (Table 3) demonstrates that different (and in some cases contradictory) sensory descriptions exist for the same volatile compound.

Despite the difficulties associated with measurement, several factors can be identified that contribute to the odour threshold value. The odour threshold is dependent on factors that influence the ease of interaction of the molecule with olfactory receptors, and that interaction can be influenced by factors such as chain length and the stereochemistry of the volatile compound, as well as external factors such as matrix effects (Angerosa, 2002; IOOC, 1987; Kiritsakis, 1998). Stereochemical effects are observed with *cis-trans* isomers. The *cis*-isomers of volatile compounds in olive oil display significantly lower odour threshold, for example *cis*- and *trans*-2-nonenal and *trans, cis*-2, 4-decadienal and *trans, trans*-2, 4-decadienal (Table 3).

With olive oil, relationships between aroma and volatile compounds have emphasised the role of C5 to C9 compounds. The most abundant compounds contributing favourably to the aroma of virgin olive oil are the C6 aldehydes and alcohols, which relates to sweetness. C5 aldehydes and alcohols also contribute to the positive attributes of olive oil, providing pungent sensations and correlating with bitterness. Small amounts of C5 ketones, pentene dimers or monoterpenes affect the aroma (Cavalli et al., 2004). Most of the smaller ketones, with five to seven carbon atoms, are linked to positive sensory characteristics. The esters are predominantly linked to the positive fruity aroma of olive oil, except for 2-methylpropyl butanoate, which is associated with the unpleasant winey and fusty odour (Table 3). The ester, 2-methylpropyl butanoate, may result from esterification of butanoic acid, from fermentation of olive fruit, by methyl-branched alcohols from leucine and valine. Carboxylic acids are linked to sour and pungent sensations synonymous with sensory

defects in olive oil. Carboxylic acids with two or three carbon atoms are associated with microbial fermentation and other fruit handling defects, whereas the higher carboxylic acids are linked to oxidative rancidity (Table 3).

Chain length also influences flavour perception, and volatile compounds with 7-12 carbon atoms are important contributors to aroma as the oil ages (Angerosa, 2002). Long chain aldehydes and alcohols characterise the sensory defects associated with oxidation during oil storage. Sensory defects arising from fruit storage are associated with the occurrence of long chain ketones with at least eight carbon atoms (Table 3). Compounds with less than four carbon atoms have not been extensively reported in the literature on olive oil aroma. Low carbon number alcohols and aldehydes are associated with fermented and malty fruit, respectively, which might result from improper fruit handling. Improper fruit handling may also be the source of sensory defects arising from the occurrence of methyl-branched alcohols, formed through conversion of leucine and valine (Tressl & Drawert, 1973)).

Odour thresholds are affected by the degree of unsaturation and the number of atoms other than carbon in the volatile compound. Comparison of mono-unsaturated and saturated volatile compounds has identified an increased odour threshold for the saturated C7, C8 and C10 aldehydes and C8 ketones (Table 3). This corresponds with observations made for C6 aldehydes in oil/water systems (Haahr, Bredie, Stahnke, Jensen & Refsgaard, 2000) in which the release of volatile compounds has also been shown to depend on chain length and degree of unsaturation as well as the matrix. The release of C6 aldehydes decreases with the number of double bonds whereas unsaturation of C9 aldehydes hardly affects the flavour release (Haahr et al., 2000). Occurrence of conjugated double bonds in

the molecular structure greatly increases the odour threshold for some compounds in olive oil, as observed between 2,4-heptadienal and heptanal but this is not true for comparison of decadienal and decanal, where *trans, trans*-2, 4-decadienal and *trans, cis*-2, 4-decadienal have odour thresholds of 180 µg/kg and 10 µg/kg, respectively, while decanal is 650 µg/kg (Table 3).

Oxidation and volatile compounds

Oxidation of the triglycerides and their derivatives in virgin olive oil causes changes in the chemical, sensory and nutritional properties of the oil that affect the quality of the oil (Rahmani & Csallany, 1998; Velasco & Dobarganes, 2002). According to EEC and IOOC regulations (EC, 1991; IOOC, 2001), peroxide values, K_{232} , K_{270} and sensory evaluation assess the oxidative deterioration of olive oil. Sensory evaluation detects oxidative deterioration before changes are observed in these other parameters, and this emphasises the importance of volatile compounds in detecting early stages of olive oil deterioration (Vichi, Pizzale, Conte, Buxaderas & Lopez-Tamames, 2003a).

Olive oil oxidation is influenced by external factors, such as storage conditions, and by oil composition (Gutierrez & Fernandez, 2002; Velasco & Dobarganes, 2002). Some minor components in olive oil have a greater contribution towards the oxidative stability than the major components, the triglycerides (Aparicio, Roda, Albi & Gutierrez, 1999; Salvador, Aranda & Fregapane, 1999; Velasco & Dobarganes, 2002; Rahmani & Csallany, 1998). Minor components in virgin olive oil may act as either anti-oxidants or pro-oxidants, and processing and storage of the oil influence the composition of these minor constituents and hence the oil's stability. This is why virgin olive oils, with identical fatty acid

compositions, can show differences in stability. To control oxidation in olive oils, an understanding of the effects of external factors on oxidation is paramount. The complexity of the situation is indicated by carotenoid pigments; they are antioxidants that strongly inhibit virgin olive oil photo-oxidation but have a pro-oxidant effect in the absence of tocopherols and at elevated temperatures (Psomiadou & Tsimidou, 2002; Velasco & Dobarganes, 2002).

The most important external factors influencing olive oil oxidation are temperature, light and oxygen concentration. At high temperatures, there is an increase in the rate of oxidation but a reduction in the solubility of oxygen. The concentration of alkoxy radicals increases, relative to the initially formed peroxy radicals, and polymeric compounds are formed from alkoxy and alkyl radicals. At low or moderate temperatures, the rate of oxidation is slow. Hydroperoxides are the major compounds formed, and their concentration increases until the advanced stages of oxidation when they decompose into minor volatile compounds, in particular carbonyl compounds that may modify olive oil aroma (Velasco & Dobarganes, 2002).

The moderating effect of light is exerted via minor components in olive oil, such as pigments, which can be electronically excited, through absorption of light, and subsequently transfer their excess energy to the oxygen molecule creating the singlet state favourable for addition to fatty acids (Hamilton, Kalu, Prisk, Padley & Pierce, 1997; Velasco & Dobarganes, 2002). A high oxygen concentration from storage of olive oil in contact with air or frequent opening of oil containers leads to a rate of formation of hydroperoxides that is higher than their decomposition rate. This leads to the production of carboxylic acids (Velasco & Dobarganes, 2002).

From such considerations, it can be deduced that ketones and aldehydes dominate the volatile compounds in oils stored at low temperatures, whereas carboxylic acids dominate the volatile compounds in oils stored in oxygen rich environments, and polymeric volatile compounds are produced at elevated temperatures. These volatile compounds from oxidation modify the sensory quality of olive oils (Vichi et al., 2003a). It has been reported (Angerosa, 2002) that the main factors that characterise off-flavours are the low abundance of the C6 aldehydes, C6 alcohols and esters from the lipoxygenase pathway and the presence of many C7 to C12 aldehydes and other volatile compounds with low odour thresholds. The focus in determining the extent of oxidation has been on volatile compounds that are formed and not necessarily the compounds that are lost as the oil ages. A reduction in *trans*-2-hexenal and an increase in C6 alcohols and C5 ketones have been observed in olive oil stored in the dark, and these compounds were proposed as markers of virgin olive oil quality freshness (Cavalli et al., 2004).

There are volatile compounds that are formed in oxidised olive oil regardless of the external conditions. Pentanal, hexanal, octanal and nonanal are the major compounds (Kiritsakis, 1998; Morales, Rios & Aparicio, 1997) and carboxylic acids such as hexanoic and propanoic acid have also been detected during the oxidation (Gutierrez, Villafranca & Castellano, 2002; Vichi et al., 2003a). Other volatile compounds detected in the late oxidation stages are 2-pentylfuran and 2-ethylfuran, which might be useful in distinguishing oxidation at late stages (Vichi et al., 2003a).

Proposed markers of oxidation include nonanal (Vichi et al., 2003a) and the ratio of hexanal/nonanal (Kiritsakis, 1998; Morales et al., 1997). While most studies have used nonanal as a primary indicator of rancidity, Solinas et al. (Solinas, Marsilio & Angerosa, 1987) observed that 2-pentenal and 2-heptenal were the main rancidity indicators. The

choice of the main indicators for oxidative rancidity has focussed on the presence of the volatile compound that causes the rancid off-flavour and not necessarily on the odour activity. It should be noted that some oxidation markers, such as nonanal and 2,4-heptadienal, have high odour threshold values (Table 3). Volatile compounds with a high odour threshold value have a less significant impact on flavour (Vichi et al., 2003a). The rate of volatile compound formation during oxidation has also been considered in choosing oxidation markers. High rates of volatile compound formation during oxidation are observed for nonanal, hexanal and octane followed by 2-pentylfuran, *trans*-2-propenal and 2,4-heptadienal isomers. All these compounds can be considered as markers of oxidation except for hexanal (Vichi et al., 2003a). The amount of hexanal does not distinguish oxidised oils from virgin oils as this compound originates from both the enzymatic and chemical oxidation pathways (Vichi et al., 2003a; Morales et al., 1997).

CONCLUSION

The challenge of producing a high quality olive oil is defeated if the oil deteriorates in production or storage. Oil quality may be defined in a number of ways, but the consumers' sensory perception of flavour is the ultimate determinant. Largely the range of volatile compounds present in the oil determines aroma. Hence, it is critical to understand the formation of these volatile compounds and promote certain favourable flavour attributes in olive oil.

Producing olive fruit with superior properties and ensuring that the positive attributes are transferred to the oil are essential to ensure a consistently high quality olive oil. Processing parameters can be altered to optimise oil production for a particular fruit.

The changes in processing parameters should take into account differences in cultivars, maturity, agronomic practices, geographic regions and the impact on the overall quality of olive oil.

In addition to improving the volatile compound composition of the fruit, control of sensory defects in olive oil is achieved through good management practices, including post-harvest fruit handling procedures that control exogenous enzyme activity (Monteleone et al., 1998). An understanding of the pathways that produce the volatile compounds is important in enhancing the quality of olive oil. Promotion of certain stages of the lipoxygenase pathway can be used to enhance some desired volatile compounds. For instance, conditions that promote HPL and inhibit ADH and AAT activity can be applied to elevate the 'green' aroma. Similarly, the conditions that promote AAT activity can be applied to enhance the fruity aroma (Salas, 2004).

Currently, most efforts have focussed on understanding the differences in oil quality from olive fruits of different qualities and in the reduction of quality deterioration once the oil is produced. Post-harvest storage of olives has shown to increase the concentration of *trans*-2-hexenal (Koprivnjak et al., 2000). Further investigation should be made in post-harvest fruit handling technologies that enhance the generation of positive volatile compounds in addition to easing pressure on processing plants.

Table 1: General classification of olive oils based on FFA.

Olive oil classification	FFA Limit (as oleic acid) %
Extra-virgin olive oil	0.8 (max)
Virgin olive oil	2.0 (max)
Ordinary virgin olive oil	3.3 (max)
Lampante virgin olive oil	3.3 (min)
Refined olive oil	0.3 (max)
Olive oil	1.0 (max)
Refined olive pomace oil	0.3 (max)
Olive pomace oil	1.0 (max)

Table 2: Temperature and pH ranges for enzyme activities.

Enzyme	pH	Reference	Temperature	Reference
Acyl hydrolase (AH)	8 – 9	(Olias et al., 1993)		
	8.5 (optimum)	(Olias et al., 1993)		
Lipoxygenase (LOX)	5.2 - 6.6	(Olias et al., 1993)	30°C (max)	(Ridolfi et al., 2002)
	5.5 - 6.0	(Georgalaki, Bachmann, Sotiroudis, Xenakis, Porzel & Feussner, 1998)	55°C (max)	(Georgalaki et al., 1998)
	5.0 - 5.5	(Salas et al., 1999)		
	5.0 - 7.0	(Ridolfi et al., 2002)		
	6.1 (optimum)	(Olias et al., 1993)		
	6.0 (optimum)	(Ridolfi et al., 2002)		
	7.5 (max)	(Salas et al., 1999)		
Hydroperoxide lyase (HPL)	5.0 – 7.0	(Olias et al., 1993))	15°C (optimum)	(Anthon & Barrett, 2003)
	6.0 (optimum)	(Salas & Sanchez, 1999)	35°C (max)	(Salas & Sanchez, 1999)

Enzyme	pH	Reference	Temperature	Reference
	5.7 (optimum)	(Olias et al., 1993)		
Alcohol dehydrogenase (ADH)	5.0 – 8.5	(Olias et al., 1993)		
	6.8 (optimum)	(Olias et al., 1993)		
Alcohol acetyl transferase (AAT)	8.0 (optimum)	(Perez et al., 1993)	35°C	(Perez et al., 1993)
	7.5 (optimum)	(Salas, 2004)	(optimum)	
	6.8 (optimum)	(Olias et al., 1993)		

Table 3. Odour thresholds and sensory descriptors of volatile compounds in olive oil.

Volatile compounds	Odour threshold ($\mu\text{g}/\text{kg}$ oil)	Sensory descriptor (aroma)	Reference
Aldehydes			
Acetaldehyde	0.22	Pungent, sweet	(Reiners & Grosch, 1998)
3-methylbutanal	5.4	malty	(Reiners & Grosch, 1998)
2-methylbutanal	5.2	malty	(Reiners & Grosch, 1998)
pentanal	240	woody, bitter, oily	(Morales et al., 2005)
<i>trans</i> -2-pentenal	300	green, apple	(Morales et al., 2005)
	300	green, bitter almond	(Aparicio & Luna, 2002)
hexanal	75	green-sweet	(Aparicio & Luna, 2002)
	80	green apple, grassy	(Morales et al., 2005)
	300	green	(Reiners & Grosch, 1998)
<i>cis</i> -3-hexenal	3	green	(Aparicio & Luna, 2002)

Volatile compounds	Odour threshold (µg/kg oil)	Sensory descriptor (aroma)	Reference
	1.7	leaflike	(Reiners & Grosch, 1998)
<i>trans</i> -2-hexenal	424	green, apple-like	(Reiners & Grosch, 1998)
	420	bitter almonds, green	(Morales et al., 2005)
	1125	green, astringent	(Aparicio & Luna, 2002)
heptanal	500	oily, fatty, woody	(Morales et al., 2005)
<i>trans</i> -2-heptenal	5	oxidised, tallowy, pungent	(Morales et al., 2005)
2,4-heptadienal	3620	fatty, rancid	(Morales et al., 2005)
octanal	320	fatty, sharp	(Morales et al., 2005)
	56	citrus-like	(Reiners & Grosch, 1998)
<i>trans</i> -2-octenal	4	herbaceous, spicy	(Morales et al., 2005)
nonanal	150	fatty, waxy, pungent	(Morales et al., 2005)
<i>trans,trans</i> -2,4- nonadienal	2500	soapy, penetrating;	(Morales et al., 2005)
	2500	deep-fried	(Reiners & Grosch,

Volatile compounds	Odour threshold (µg/kg oil)	Sensory descriptor (aroma)	Reference
			1998)
<i>cis</i> -2-nonenal	4.5	green, fatty	(Reiners & Grosch, 1998)
<i>trans</i> -2-nonenal	900	paperlike, fatty	(Reiners & Grosch, 1998)
decanal	650	penetrating, sweet, waxy	(Morales et al., 2005)
<i>trans</i> -2-decenal	10	painty, fishy, fatty	(Morales et al., 2005)
2,4-decadienal	2150	strong, fatty	(Morales et al., 2005)
<i>trans, trans</i> -2,4-decadienal	180	deep-fried	(Reiners & Grosch, 1998)
<i>trans, cis</i> -2,4-decadienal	10	deep-fried	(Reiners & Grosch, 1998)
<i>trans</i> -4,5-epoxy- <i>trans</i> -2-decanal	1.3	metallic	(Reiners & Grosch, 1998)
Alcohols			
ethanol	30000	alcohol	(Morales et al., 2005)
butan-2-ol	150	winey	(Morales et al., 2005)

Volatile compounds	Odour threshold ($\mu\text{g}/\text{kg}$ oil)	Sensory descriptor (aroma)	Reference
2-methyl butan-1-ol	480	winey, spicy	(Morales et al., 2005)
3-methyl butan-1-ol	100	woody, whiskey, sweet	(Morales et al., 2005)
pentanol	470	fruity	(Aparicio & Luna, 2002)
	3000	strong, sticky, balsamic	(Morales et al., 2005)
3-penten-2-ol	400	perfumery, woody	(Morales et al., 2005)
hexanol	400	fruit, banana, soft	(Aparicio & Morales, 1998)
	400	undesirable	(Aparicio & Luna, 2002)
<i>trans</i> -2-hexen-1-ol	5000	green grass, leaves	(Morales et al., 2005)
	8000	green, grassy, sweet	(Aparicio & Morales, 1998)
<i>trans</i> -3-hexen-1-ol	1500	green	(Aparicio & Morales, 1998)
<i>cis</i> -3-hexenol	6000	green	(Aparicio & Luna, 2002)
	1100	leaf-like	(Reiners & Grosch, 1998)
heptan-2-ol	10	earthy, sweety	(Morales et al., 2005)

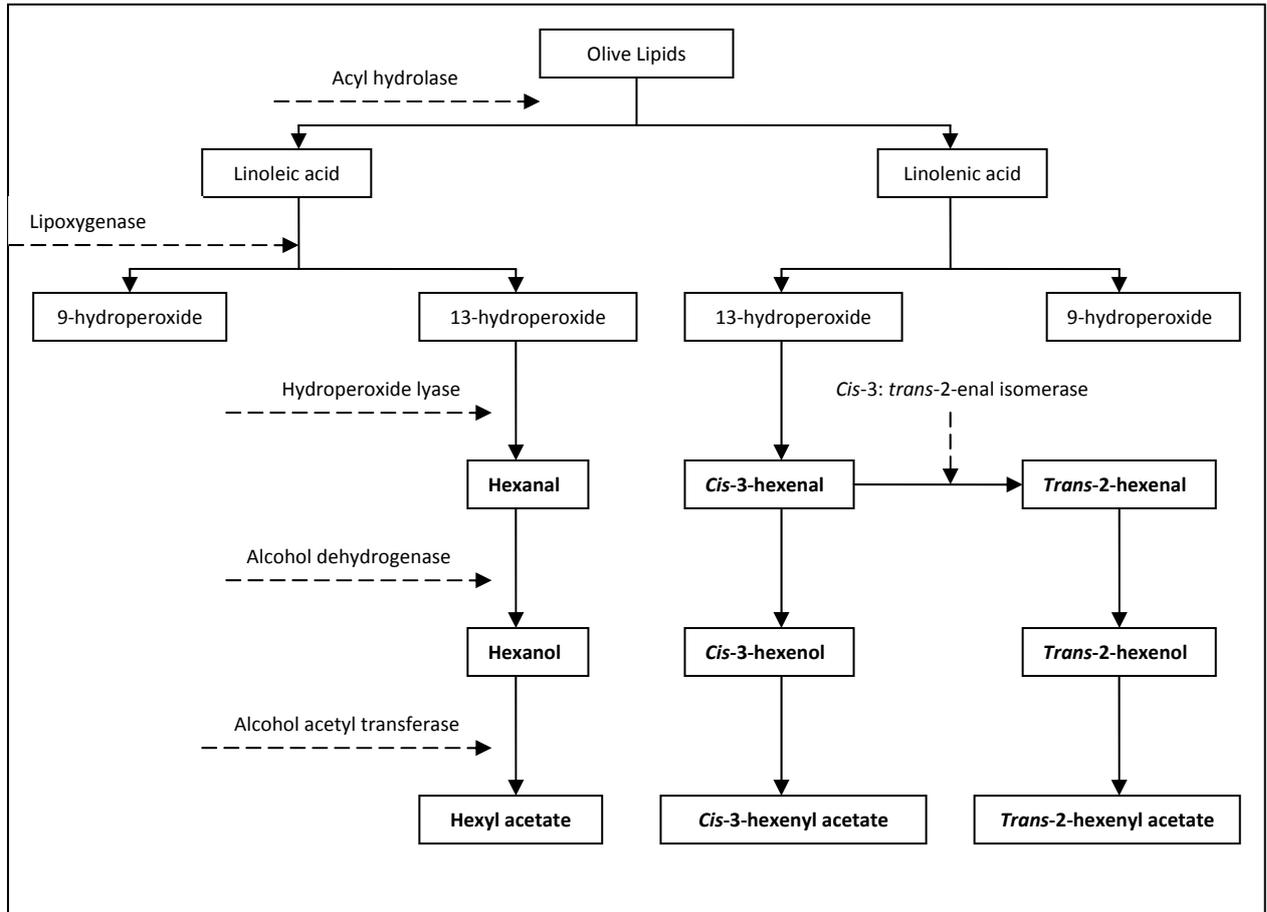
Volatile compounds	Odour threshold ($\mu\text{g}/\text{kg}$ oil)	Sensory descriptor (aroma)	Reference
6-methyl-5-hepten-3-ol	2000	perfumey, nutty	(Morales et al., 2005)
octan-2-ol	100	earthy, fatty	(Morales et al., 2005)
octen-3-ol	1	mouldy, earthy	(Morales et al., 2005)
nonanol	280	fatty	(Morales et al., 2005)
	13500	rancid	(Aparicio & Luna, 2002)
Esters			
ethyl acetate	940	sticky, sweet	(Morales et al., 2005)
butyl acetate	300	green, fruity, pungent	(Morales et al., 2005)
hexyl acetate	1040	green; fruity, sweet	(Aparicio & Luna, 2002)
			(Baeten et al., 1998)
<i>cis</i> -3-hexenyl acetate	750	green	(Aparicio & Luna, 2002)
	200	banana-like	(Reiners & Grosch, 1998)
ethyl propanoate	100	fruit, strong	(Morales et al., 2005)
ethyl butanoate	30	sweet, fruity	(Morales et al., 2005)
	28	cheesy, fruity	(Reiners & Grosch,

Volatile compounds	Odour threshold (µg/kg oil)	Sensory descriptor (aroma)	Reference
			1998)
ethyl isobutyrate	1.2	fruity	(Reiners & Grosch, 1998)
propyl butanoate	150	pineapple, sharp	(Morales et al., 2005)
2-methylpropyl butanoate	100	unpleasant, winey, fusty	(Morales et al., 2005)
ethyl 2-methylbutyrate	0.72	fruity	(Reiners & Grosch, 1998)
ethyl 3-methylbutyrate	0.62	fruity	(Reiners & Grosch, 1998)
ethyl cyclohexylcarboxylate	0.16	aromatic, fruity	(Reiners & Grosch, 1998)
Ketones			
butan-2-one	40000	ethereal, fruity	(Morales et al., 2005)
1-penten-3-one	50	green	(Aparicio & Luna, 2002)
	0.73	green, pungent	(Reiners & Grosch, 1998)

Volatile compounds	Odour threshold ($\mu\text{g}/\text{kg}$ oil)	Sensory descriptor (aroma)	Reference
heptan-2-one	300	sweet, fruity	(Morales et al., 2005)
6-methyl-5-hepten-2-one	1000	pungent, green	(Morales et al., 2005)
octan-2-one	510	mould, green	(Morales et al., 2005)
1-octen-3-one	10	mushroom, mould,	(Morales et al., 2005)
	10	pungent; mushroom-like	(Reiners & Grosch, 1998)
<i>cis</i> -1,5-octadien-3-one	0.45	geranium-like	(Reiners & Grosch, 1998)
<i>trans</i> - β -damascenone	11	boiled apple-like	(Reiners & Grosch, 1998)
Carboxylic acids			
acetic acid	500	sour, vinegary	(Morales et al., 2005)
	124	vinegar-like	(Reiners & Grosch, 1998)
propanoic acid	720	pungent, sour	(Morales et al., 2005)
butanoic acid	650	rancid, cheese	(Morales et al., 2005)
3-methylbutyric acid	22	sweaty	(Reiners & Grosch,

Volatile compounds	Odour threshold (µg/kg oil)	Sensory descriptor (aroma)	Reference
			1998)
pentanoic acid	600	unpleasant, pungent	(Morales et al., 2005)
hexanoic acid	700	pungent, rancid	(Morales et al., 2005)
heptanoic acid	100	rancid, fatty	(Morales et al., 2005)
octanoic acid	3000	oily, fatty	(Morales et al., 2005)
Other compounds			
octane	940	sweety, alcane	(Morales et al., 2005)
4-methoxy-2-methyl-2- butanethiol	0.017	black currant-like, catty	(Reiners & Grosch, 1998)
guaiacol	16	phenolic, burnt	(Reiners & Grosch, 1998)

Figure 1. Pathway for the formation of major volatile compounds in virgin olive oils. Volatiles are shown in bold; enzymes are shown by dashed arrows. (Adapted from (Olias et al., 1993); (Ridolfi et al., 2002)).



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