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Príncipe de Vergara, 154.
28002 Madrid, Spain.
Tel.: 34-915 903 638
Fax: 34-915 631 263
E-mail: iooc@internationaloliveoil.org

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Performance of the Arbequina, Haouzia and Menara olive varieties in rainfed conditions in the Meknès region of Morocco

A. Mahhou[1], Y. Nabil[2], A. Hadiddou[3], A. Oukabli[3] and A. Mamouni[3]

[1] IAV Hassan II, Rabat, a.mahhou@iav.ac.ma & a.mahhou@gmail.com
[2] Office Régional de Mise en Valeur Agricole du Haouz, Morocco.
[3] Centre Régional de la Recherche Agronomique de Meknès, Morocco.

ABSTRACT

The objective of this study was to evaluate the performance of the Haouzia, Menara and Arbequina varieties in rainfed conditions in the Meknès region. The carpometric characteristics of the olives were determined and parameters likely to help olive growers to decide harvest timing were evaluated. For this purpose, the maturity index was calculated and the oil content and polyphenol content were determined during ripening. Oleic and linoleic acid content were likewise determined. Flowering and fruit set were monitored to assess the effectiveness of pollination and fertilisation, which were measured on the basis of recorded yields.

Oil, polyphenol and oleic acid content differed by variety. The Arbequina variety recorded a maximum polyphenol content of 1833 ppm. This was lower than the maximum polyphenol content of Menara and Haouzia, which recorded respective values of 2134 ppm and 2127 ppm. The maximum oil content on a fresh weight basis was 23.6% for Menara, followed by 23.3% for Haouzia and 17.56% for Arbequina. When taken together, these parameters enabled determination of the optimum harvesting period, which is when oil and polyphenol content are at the best levels. For the crop year in question, the optimum harvest period extended from 03 to 19 December for Arbequina and from 03 December to 02 January for Menara and Haouzia. Flowering lasted from 8 to 29 April in the case of the Arbequina variety, and started on 15 April and ended towards the end of the first week of May in the case of Haouzia and Menara. Fruit set was satisfactory, recording rates of 15.36% for Arbequina, 11% for Haouzia and 12% for Menara. Mean yields over a four-year period were 44 kg/tree for Menara, 37.25 kg/tree for Arbequina and 35.5 kg/tree for Haouzia.

Key words: variety; olive oil; olive; carpometric characteristics; optimum harvest date; maturity index; oil content; polyphenol content.

INTRODUCTION

Olive growing is of great socio-economic importance to Morocco where it accounts for 5% of agricultural GDP. Olive orchards occupy 784 000 hectares of land and produce approximately 1,500,000 t of raw olives. Morocco produces 160,000 t of olive oil and 90,000 t of table olives. It exports 17,000 t of olive oil and 64,000 t of table olives to foreign markets and the sector generates more than 15 million work days (MAPM, 2012).

The demand for olive oil and processed olives on international and home markets has seen remarkable growth in recent decades, driven by research into the health-promoting value of these products and the important part they play in the prevention of certain diseases. The Moroccan State

has thus put in place a strategy for olive orchard area to reach 1,220,000 ha by the year 2020 through the implementation of what are known as the Pillar I and Pillar II projects under Morocco's Green Plan.

However, there are numerous, heavy constraints in the way of the development of the olive industry in Morocco. These are largely linked to the shortage of rainfall, the cultural techniques applied, which are often traditional and sketchy, and the processing infrastructure which has not been sufficiently modernised.

In 2009 Morocco adopted a strategy under its Green Plan in order to diminish the national shortfall and consolidate Morocco's position on the international market for olive oil and table olives. This strategy envisages the implementation of 510 integrated projects by 2020, aimed at improving productivity and quality, achieving strong, lasting value creation, establishing two olive research hubs in Marrakech and Meknès and promoting and diversifying exports through stronger extension and applied research programmes.

The domestic olive sector continues to be dominated by the Picholine marocaine, a population variety adapted to Morocco's soil conditions. It is a dual-purpose variety used for making green and black table olives as well as oil. It gives average to good yields if producing conditions are met but has quite a pronounced on/off bearing pattern, particularly in rainfed conditions. For this reason, the *Institut National de la Recherche Agronomique* (INRA) selected two clones of the Picholine marocaine called Haouzia and Menara. The fruit of the dual-purpose Haouzia clone is oval in shape and larger than that of the Picholine marocaine (3.3–5 g). It has an average oil content of 20–24% and gives good quality oil (stable, with a high content of oleic acid and polyphenols).

The moisture and oil content of olives vary according to cultivar and growing conditions. In Morocco, for instance, Arbequina has an oil content of 37% of dry weight and Picholine marocaine has a 30% content (Boulouha, 2006). The oil content of the same variety when grown under irrigation in California ranges between 22 and 27% of fresh weight (Vossen, 2005). Haouzia has an oil content of 20–24% according to the varietal fact card published by INRA (Boulouha *et al*. 2006a) and of 23.2% as reported by Hadiddou *et al*. (2006) as well as in the World Catalogue of Olive Varieties (International Olive Council, 2000). However, lower oil contents have been reported by El Ajel, (2006) and Rafik, (2008) who respectively cite values of 20.3% and 21.8% of fresh weight. Menara has a mean fresh weight oil content of 24% as reported by Hadiddou *et al*. (2006). The same content is cited in the World Catalogue of Olive Varieties (International Olive Council, 2000) and concurs with the value of 23.2% found by Rafik (2008).

Polyphenol content differs according to variety (Cimato *et al*., 1996; Pannelli *et al*., 2001; Sweeney, 2005). Vossen (2005) reports that the polyphenol content of olive varieties ranges from the very high levels found in Koroneiki and Coratina to the very low levels found in varieties such as Picual. It is particularly worthwhile to monitor the changes in the concentration of phenolic compounds because these substances affect the organoleptic characteristics and oxidative stability of olive oil (Chimi, 1987, Chimi *et al*., 1991). Fantozzi and Montedero (1978) reported that the phenolic content of the fruit flesh changes ac-

cording to the degree of pigmentation, ranging from 2065 (mg gallic acid/100 g dry olive paste) at the green stage to 2285 at the semi-black stage and 1997 at the black stage. These authors concluded that polyphenol content is at its optimal level when the olives are at the semi-black stage and that it is associated with better quality oils.

The same tendency in polyphenol content has been observed by Atouati (1991) who documented a rise in total phenolic compound content between the green and semi-black stages, followed by a decrease at the black stage. Conversely, the reverse occurred for oil content, which was at its highest during the black stage. When studying three varieties in irrigated olive orchards in the Settat region Mahhou *et al*. (2011) reported an increase in polyphenol content to a maximum of 1823 ppm in Arbequina, 2192 ppm in Koroneiki and 2113 ppm in Picholine marocaine, followed by a decrease from early December onwards.

In rainfed conditions the Haouzia clone is also productive, giving an average yield of 25–80 kg/tree. The Menara variety is characterised by yields of more than 60 kg/tree (over 11 crop years), an oil content in excess of 20% and good resistance to *Pseudomonas savastanoi* (olive knot). Besides these clones, other foreign varieties have been introduced, particularly Spanish varieties, on account of their early bearing and adaptation to high densities (Arbequina) (Boulouha B. *et al*, 2006).

The objective of this research was to determine the characteristics of olives and olive oils at different harvest times in order to identify the best period to pick three varieties – Arbequina, Menara and Haouzia – grown in rainfed conditions in the Meknès region.

2. MATERIALS AND METHODS

2.1. Site characteristics

This study was conducted on the INRA experimental farm at Ain Taoujdate, situated in the Sais plain 30 km from Meknès in the province of El Hajeb (altitude: 550 m; latitude: 33° ; longitude: 5°) during the 2008/09 crop year.

The soils on the farm are alluvial: deep, reddish brown, non-calcareous, sandy clay.

The Ain Taoujdate region has an average annual rainfall of around 500 mm. Precipitation is spread over September to April, although largely concentrated in the months of November and December. The dry season lasts from June to September. Annual rainfall during the crop year was 700 mm. Mean minimal temperatures ranged from 4º in the coldest month (December) to 18 ºC in the hottest month (July) while the mean maximum temperatures for the same months varied from 15 to 38 ºC.

The study was conducted on two Picholine marocaine clones – Haouzia et Menara – planted in 1989, and the Spanish Arbequina variety planted in 1988 on a spacing of 7x7 m, i.e. at a density of 204 trees/ha.

The **Haouzia** variety is a selection of the Picholine marocaine. It is hardy and has a high rooting capacity (65%). The tree is of medium vigour and has a spreading growth habit. The olives are dual-purpose and have an oil content of 23%. It is partially self-fertile and starts bearing as of the third year. Yields average 60 kg/tree in irrigated conditions and 25–80 kg/tree in rainfed conditions (Central North). It is tolerant of peacock spot, olive knot and drought.

The **Menara** variety is also a selection of the Picholine marocaine. The tree displays medium vigour when grown in rainfed conditions and high vigour when irrigated. It has an erect growth habit and a very high rooting capacity (70%). It displays 30% less alternate bearing than the Picholine marocaine. It starts bearing as of the third year and when mature gives average yields of more than 60 kg/tree under irrigation. In rainfed conditions it yields 35–80 kg/tree. It is resistant to olive knot.

2.2. Sampling

Four trees of each genotype were chosen at random according to a completely random design and marked with paint. A composite sample (1 kg) of olives of each genotype was collected on the dates listed in Table 1; the samples were picked randomly at shoulder height from different shoots.

The samples were placed in plastic sachets and sent to the laboratory on the same day for characterisation. The rest of the sample was stored in the freezer at -20°C until it underwent physico–chemical analysis.

TABLE 1.
Sampling dates

Sample number	Sampling date
1	22/10/2008
2	05/11/2008
3	19/11/2008
4	03/12/2008
5	12/12/2008
6	19/12/2008
7	02/01/2009
8	09/01/2009
9	16/01/2009

2.3. Olive fruit analyses

Determination of the maturity index (MI) of the olives

This determination is based on the colour assessment of 100 olives picked at random from a 1 kg sample. The fruit is arranged according to eight colour stages ranging from a deep green skin colour to a black skin colour and dark flesh colour all the way to the stone.

The maturity index is calculated as follows:

$$\text{Maturity index} = \frac{[(0 \times n_0) + (1 \times n_1) + (2 \times n_2) + \ldots + (7 \times n_7)}{100}$$

Where n_0, n_1, …, n = the number of fruit in each of the following numerical classes:

0: deep green skin colour
1: yellow or yellow-green skin colour
2: yellow-green skin colour with reddish spots
3: red to purple skin colour
4: black skin colour and white-green flesh colour
5: black skin colour and violet flesh colour halfway to the stone
6: black skin colour and violet flesh colour almost to the stone
7: black skin colour and dark flesh colour all the way to the stone

Determination of the carpometric characteristics of the olive fruits

Fruit, stone and flesh weight were determined in a sample of 20 olives. Fruit shape was determined by measuring the length and width of the 20 fruits with callipers. The ratio between the two measurements indicates the shape of the fruit according to the criteria laid down by the International Olive Council (IOC, 2000).

Determination of olive moisture content

Two 40-g samples of olives were collected, weighed (fresh weight) and dried to constant weight in an oven at 75 °C for 48 hours. The samples were reweighed after being removed from the oven (dry weight). The difference between the two weights is the weight of the moisture, the content of which is calculated relative to 100 g of olives.

$$\text{Moisture } (\ ) = \frac{\text{Fresh weight} - \text{Dry weight}}{\text{Fresh weight}} \times 100$$

Determination of fruit oil content

Soxhlet method

Seventy grams of olives were crushed in a mortar and dried to constant weight in an oven at 105 °C (~ 42 h). The oil recovered was weighed (M) and the percentage oil content (fresh and dry) was calculated from the following formulas:

$$\text{OCF } (\ ) = \frac{M \times 100}{M_0}$$

$$\text{OCD } (\ ) = \frac{M \times 100}{M_1}$$

Where:

- **OCF** = oil content on a fresh matter basis
- **OCD** = oil content on a dry matter basis
- **M** = weight of the extracted oil
- $\mathbf{M_0}$ = fresh weight of the sample
- $\mathbf{M_1}$ = dried weight of the sample

Each sample was tested in triplicate to determine the mean oil content of each variety.

2.4. Oil testing methods

Total polyphenols

The method proposed by Vázquez Roncero (1975) was used to determine the total polyphenols. Ten grams of oil were weighed, diluted with 50 ml of hexane and placed in a separating funnel. The polyphenols were extracted three times in a 20-ml methanol: water solution (60%:40%) and were shaken each time for 2 min 30 s. After each extraction, the bottom layers were separated directly into a 100-ml flask and made up with distilled water. This constituted the polyphenol solution. In a 50-ml volumetric flask, 35 ml of distilled water were placed together with 15 ml of the polyphenol solution and 2.5 ml of Folin–Denis reagent. The mixture was homogenised by shaking and left to rest for 3 min. Five ml of the 6% NaOH solution were added, made up to volume with distilled water and mixed well. The control was performed in the same conditions as the oil sample. After being left to rest for one hour (45 min minimum), the absorbance reading was taken with a spectrometer at 725 nm.

Fatty acids

Approximately 0.3 g of oil were weighed into a 50-

ml retort to which 2.5 ml methanol sodium were added to make the solution deep pink in colour. The retort was then placed below a condenser and heated under reflux for 10 min; 2.5 ml of sulphur methanol were added until the pink colouring disappeared and the flask was then heated for a further 10 min. After being cooled with the aid of a funnel, the mixture was transferred to a test tube and the retort was rinsed with 6 ml heptane (2 ml x three times). The mixture in the tube was made up to volume with NaCl, which helps the esters to float. The methyl esters floating on the surface were recovered with the aid of a micro–syringe. The fatty acid composition was determined by gas chromatography.

2.5. Monitoring of flowering and evaluation of fertility

Flowering period

Visual inspections were performed to determine the flowering period of the varieties. These were conducted once a week, from March throughout flowering. The following stages were assessed:

- Start of flowering, 10% open flowers
- Full bloom, 90% open flowers
- End of flowering, start of petal drop

Evaluation of fertility

On 29 April 2009 four shoots (one each facing South, North, East and West) were marked on each of the four trees of each genotype. The initial number of flowers on each shoot was counted on the same day. After physiological drop in June, the number of set flowers or small fruits was counted. The fruit set rate was calculated according to the following equation:

$$\text{Fruit set rate ()} = \frac{\text{Number of set flowers}}{\text{Total number of flowers}} \times 100$$

2.6. Number of determinations and statistical analysis

All the laboratory analyses were performed in triplicate. The results given in the interpretation are the mean of three determinations. The results were analysed using Minitab for analysis of the variance and the calculation of the descriptive statistics: mean, standard deviations, etc.

3. RESULTS AND DISCUSSION

3.1. Maturity index

The maturity index of the olives from the Moroccan Haouzia and Menara clones and the Arbequina variety was monitored between 22 October 2008 and 16 January 2009 (Table 2). The index ranged from 1.4 to 4.9 for the Picholine marocaine clones and from 2.3 to 4.7 for the Arbequina.

TABLE 2.
Changes in the maturity index of Haouzia. Menara and Arbequina olives grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year

Sampling date	Arbequina	Haouzia	Menara
22/10/2008	2.3	1.4	1.4
05/11/2008	2.5	2.7	1.9
19/11/2008	2.5	3.5	3.2
03/12/2008	3.1	3.9	3.9
12/12/2008	3.4	4.3	4.1
19/12/2008	3.5	4.6	4.4
02/01/2009	4.1	4.6	4.6
09/01/2009	4.4	4.8	4.6
16/01/2009	4.7	4.9	4.8

Haouzia and Menara ripened earlier than Arbequina. The maturity index curves of the clones evolved at a similar rate although Haouzia was slightly ahead.

3.2. Carpometric characteristics of the olives

Mean fruit weight (Table 3 and Figure 1) developed at a similar rate for the three genotypes, but differed between the two Haouzia and Menara clones and Arbequina. Analysis of variance revealed an effect of variety on fruit weight. Application of Tukey's method to separate the mean weights for each sample collection date distinguished two homogeneous groups, the two Picholine marocaine clones on the one hand and Arbequina on the other, except on 05/11/2008, 09/01/2009 and 16/01/2009 when differences were detected among the three varieties, thus giving three homogeneous groups. The mean fruit weight of Arbequina, which is an oil variety, was lower than that of the two, dual-purpose clones. Between the start and end of the sample collections, mean fruit weight increased from 0.86 g to 1.61 g, from 2.13 to 2.45 g and from 1.92 to 2.55 g, respectively for Arbequina, Haouzia and Menara, thus recording respective percentage increases of 87, 15 and 33%. This recorded trend in fruit weight concurs with the results reported by Atouati (1991), Idrissi (1994), Lachir and Sidi Baba (1994), El Cadi and El Jamaï (1998), and Faqih and Hmama (1999). Rafik (2008), who conducted research in the same plot the preceding crop year (2006/07), reported similar results, namely a mean fruit weight of 1.7 g for Arbequina, 2.4 g for Haouzia and 2.3 g for Menara. The mean fruit weight of the Arbequina variety is slightly lower than that found by Mahhou *et al*. (2011) in irrigated conditions in the Settat region.

TABLE 3.
Changes in the mean fruit, flesh and stone weight and flesh-to-stone ratio of the Arbequina, Haouzia and Menara varieties grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year

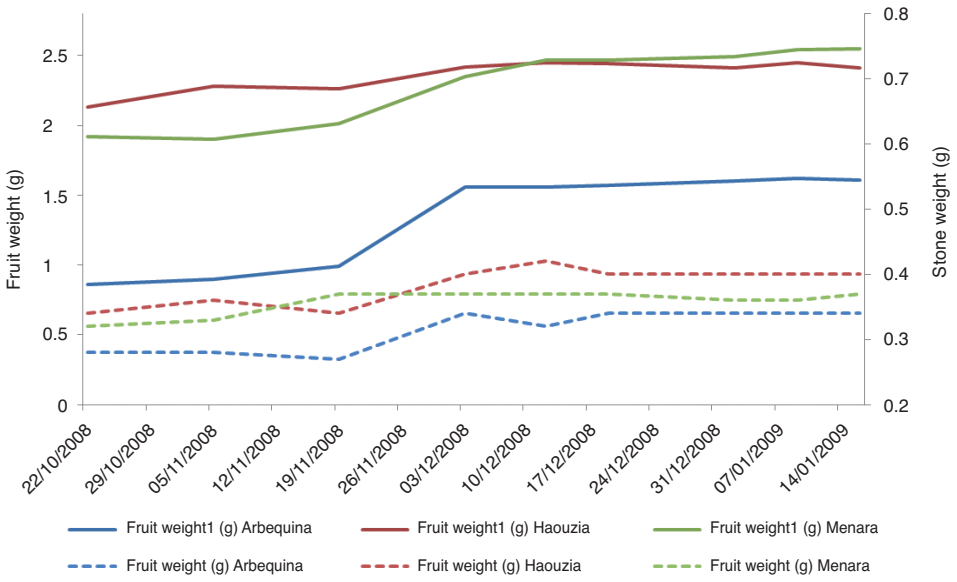
Date	Fruit weight[1] (g)			Stone weight (g)		
	Arbequina	Haouzia	Menara	Arbequina	Haouzia	Menara
22/10/2008	0.86 a	2.13 b	1.92 b	0.28 a	0.34 b	0.32 b
05/11/2008	0.90 a	2.28 c	1.90 b	0.28 a	0.36 c	0.33 b
19/11/2008	0.99 a	2.26 b	2.01 b	0.27 a	0.34 b	0.37 b
03/12/2008	1.56 a	2.42 b	2.35 b	0.34 a	0.40 b	0.37 ab
12/12/2008	1.56 a	2.45 b	2.47 b	0.32 a	0.42 c	0.37 b
19/12/2008	1.57 a	2.44 b	2.47 b	0.34 a	0.40 c	0.37 b
02/01/2009	1.60 a	2.41 b	2.49 b	0.34 a	0.40 b	0.36 a
09/01/2009	1.62 a	2.45 b	2.54 c	0.34 a	0.40 b	0.36 a
16/01/2009	1.61 a	2.41 b	2.55 c	0.34 a	0.40 c	0.37 b

[1] For each date and parameter, means with the same letter are not significantly different (Tukey, 5%).

TABLE 3. (contd)

Date	Flesh weight (g)			Flesh-to-stone ratio		
	Arbequina	Haouzia	Menara	Arbequina	Haouzia	Menara
22/10/2008	0.58 a	1.79 b	1.60 b	2.07 a	5.26 b	5.00 b
05/11/2008	0.62 a	1.91 c	1.57 b	2.22 a	5.30 c	4.76 b
19/11/2008	0.73 a	1.91 c	1.63 b	2.71 a	5.62 c	4.41 b
03/12/2008	1.22 a	2.02 b	1.98 b	3.58 a	5.05 b	5.35 b
12/12/2008	1.24 a	2.03 b	2.09 c	3.87 a	4.83 b	5.65 c
19/12/2008	1.23 a	2.04 b	2.10 c	3.62 a	5.10 b	5.66 b
02/01/2009	1.26 a	2.02 b	2.13 c	3.71 a	5.05 b	5.92 b
09/01/2009	1.28 a	2.05 b	2.18 c	3.77 a	5.13 b	6.05 b
16/01/2009	1.27 a	2.01 b	2.18 c	3.74 a	5.02 b	5.89 b

Figure 1. Changes in fruit and stone growth in the Arbequina, Haouzia and Menara varieties grown in rainfed conditions in the Meknès region during the 2008/09 crop year



Hence, mean fruit weight, which is a trait linked to variety, continues to be under the influence of the annual environmental and tree care conditions which can give rise to large variations.

The mean weight of the Haouzia stones at the black stage was 0.4 g. This falls within the range of 0.3 g – 0.45 g reported in the World Catalogue of Olive Varieties (International Olive Council, 2000) but is 0.75 g lower than the value reported by INRA in irrigated olive orchards at Menara in Marrakech. A mean fruit stone of 0.37 g was found for the Menara clone, which lies inside the range of 0.3 – 0.45 g reported for this clone in the World Catalogue of Olive Varieties (International Olive Council, 2000) while lying 0.5 g below the weight reported by INRA Marrakech in irrigated collections at Menara. The final mean stone weight of the Arbequina variety was 0.34 g. This is greater than the values reported by Romero and Diaz (2005) and by Sweeney (2005), who documented a weight of 0.27 g. Taiebi (2008) recorded a stone weight of 0.26 g in irrigated Arbequina olives. Rafik (2008), however, working in the same rainfed conditions as in the research by the authors of this paper, reported a stone weight of 0.4 g for Arbequina and of 0.5 g for Menara and Haouzia, which is above the weights recorded this year for the two clones. Stone weight thus depends on yearly growing and climatic conditions. Therefore, while being a trait linked to variety, mean stone weight can vary according to the environmental and crop management conditions.

Flesh-to-stone ratios of 3.87, 5.62 and 6.05 were recorded respectively for Arbequina, Haouzia and Menara (Table 3). The ratio of the Haouzia clone agrees with the data contained in the varietal fact card (Boulouha *et al*., 2006a) which cites a ratio of 4 to 6. However, in the case of the Menara clone, the ratio reported in this research is higher than that cited in the fact card (3–5). Barranco *et al*. (1999) state that table olive varieties should have a ratio of at least 5. Hence, under the test conditions, the Moroccan clones recorded a suitable fruit weight and flesh-to-stone ratio for processing as table olives. Tous *et al*. (1998) and Rallo *et al*. (2005) reported a ratio of 3.9 for the Arbequina variety, which is close to the value found in this research for this year (3.87).

Fruit shape is determined by the length/width ratio (L/W), which hardly changed during the fruit development of any of the varieties. The mean values of this ratio were:

- 1.17 for Arbequina, which corresponds to a spherical shape
- 1.26 for Haouzia and 1.31 for Menara, which is an ovoid shape and concurs with the description of the International Olive Council (2000).

3.3. Fruit composition

3.3.1. Changes in moisture

Fruit moisture content decreased in the three varieties through ripening, dropping from 61% to 56% in Arbequina, from 65 to 56% in Menara and from 65 to 58% in Haouzia. Faqih and Hmama (1999) report a slight decrease in moisture at an advanced stage of ripening due to fruit transpiration. Similar observations have been made by other autors (Atouati, 1991; Idrissi, 1994; Lachir and Sidi Baba, 1994; Lamrini, 1995; Rahmani *et al*., 1997; El Cadi and Jamaï, 1998).

According to the classification proposed by Del Río and Caballero (1994) for olive moisture content (very low:< 40; low: 40-50 ; medium: 50-60; high: 60-70; very high:>70), the three varieties come under the medium category.

3.3.2. Changes in oil content

The changes in fruit oil content (as a percentage of fresh matter) in the three varieties are given in Table 5 and Figure 2. Oil content is a varietal characteristic and specific to each variety. Walali *et al*. (1984) reported a difference in oil content among the clones of the Picholine marocaine. Boulouha (2006 b) confirmed a differing dry-matter oil content among the Haouzia (48%), Menara (56%) and Arbequina (37%) varieties. During this crop year and in the test conditions reported in this paper, the oil content of the Haouzia and Menara clones as a percentage of the fresh matter (23%) was higher than that of Arbequina (17.6%). The latter value lies below the contents reported by several authors, namely 22.6% (El Ajal, 2006), and 23% (Ouazzani *et al*., 2002) in irrigated olive orchards in the Meknès region, and 25.7% (Taiebi, 2008) in irrigated orchards in the Settat region. The fresh weight oil content of Arbequina olives grown under irrigation in California ranges from 22 to 27% (Vossen, 2005). The oil content of the Haouzia clone (23.3%) lies within the band of 20–24% published in the IRTA varietal fact card (Boulouha *et al*. 2006a) and concurs with the value of 23.2% reported by Hadiddou *et al*. (2006) and of 23% stated in the World Catalogue of Olive (International Olive Council, 2000). However, El Ajel, (2006) and Rafik (2008) found lower fresh weight contents of 20.3% and 21.8% respectively. An oil content of 23.6% was observed for the Menara clone, which tallies with the mean

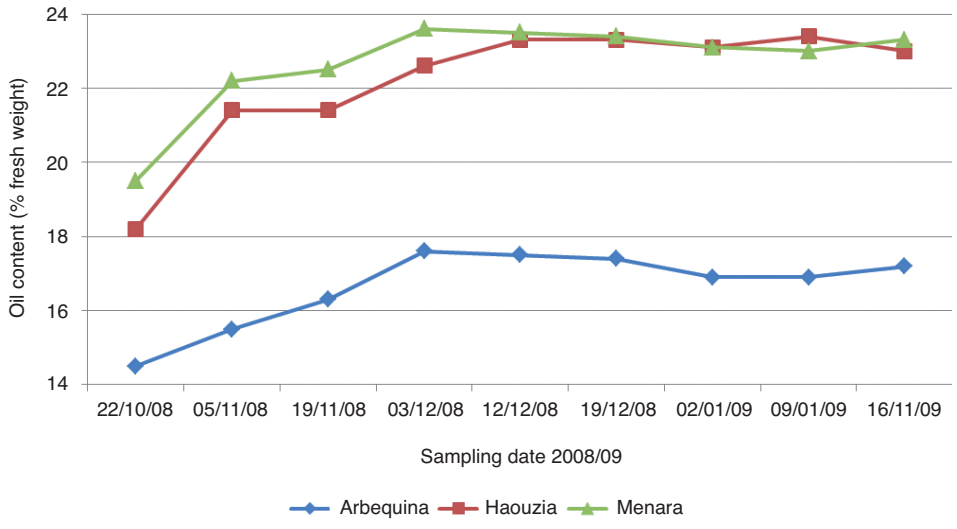
value of 24% reported by Hadiddou *et al*. (2006). The same content is recorded in the World Catalogue of Olive Varieties (International Olive Council, 2000) and concurs with the value of 23.2% reported by Rafik (2008).

TABLE 4.
Changes in the polyphenol content (ppm) of olives belonging to the Arbequina. Haouzia and Menara varieties grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year

Sampling date	Arbequina	Haouzia	Menara
22/10/2008	1731 a[1]	1833 b	1854 c
05/11/2008	1735 a	1933 b	1946 c
19/11/2008	1803 a	2054 b	2123 c
03/12/2008	1833 a	2124 b	2133 c
12/12/2008	1828 a	2127 b	2134 c
19/12/2008	1812 a	2103 b	2125 c
02/01/2009	1755 a	2067 b	2112 c
09/01/2009	1724 a	2013 b	2066 c
16/01/2009	1722 a	1987 b	2032 c
Mean	1771.4	2026.8	2058.3

[1] For each date, means with the same letter are not significantly different (Tukey, 5%).

Figure 2. Changes in the oil content (% fresh weight) of the Arbequina, Haouzia and Menara varieties grown in rainfed conditions in the Meknès region during the 2008/09 crop year



Fruit oil content increased in step with ripening, rising from 14.5% to 17.6% for Arbequina, from 18.1% to 23.3% for Haouzia and from 19.5% to 23.6% for Menara. These maximums were reached in early December, after which oil content levelled off. Walali *et al*. (1984) reported the differences in oil content in five Picholine marocaine clones on different sample collection dates. El Antari (2006) reported this difference between the Menara and Haouzia clones on different sampling dates between October and November. Oil biosynthesis proceeds very rapidly between the time the olives are at the green stage until they turn completely black, after which oil content stabilises (Uceda and Frías, 1975; Suarez, 1984; Civantos, 1999) and even records a small decrease at advanced stages of maturity (Lachir and Sidi Baba, 1994; El Cadi and Jamaï, 1998; Hmama and Faqih, 1999). This decline in oil content can be attributed not only to the accumulation of dry matter in olives at an advanced stage of ripening but also to endogenous lipases (active at the black stage) which hydrolyse the triglycerides and fatty acids (Harrar, 2007). The intensity of oil formation is a genetic trait, but also depends on soil and climatic conditions and crop management (Civantos, 1999).

A strong correlation was observed between the maturity index and oil content of the three varieties. This ratio is best described by the following equations:

- Arbequina: $y = 1.297x^3 - 14.72x^2 + 54.55x - 48.78$ with $R^2=0.953$ (Figure 3)
- Haouzia: $y = -0.480x^4 + 6.358x^3 - 30.24x^2 + 61.83x - 24.72$ with $R^2=0.987$ (Figure 4)
- Menara: $y = 0.517x^5 - 8.465x^4 + 53.40x^3 - 161.8x^2 + 235.3x - 109.5$ with $R^2=0.997$ (Figure 5)

Mahhou *et al*. (2011) also reported the existence of a strong correlation between the maturity index and oil content of Arbequina, Koroneiki and Picholine marocaine olives grown under irrigation in the Settat region.

3.3.3. Changes in polyphenols

Monitoring the changes in the concentration of phenolic compounds is particularly relevant because these substances affect the organoleptic characteristics and oxidative stability of olive oil (Chimi, 1987; Chimi *et al*., 1991). Fantozzi and Montedero (1978) reported that the phenolics content of the fruit flesh varies according to the extent of pigmentation,

Figure 3. Ratio between the maturity index and oil content (% fresh weight) of the Arbequina variety grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year

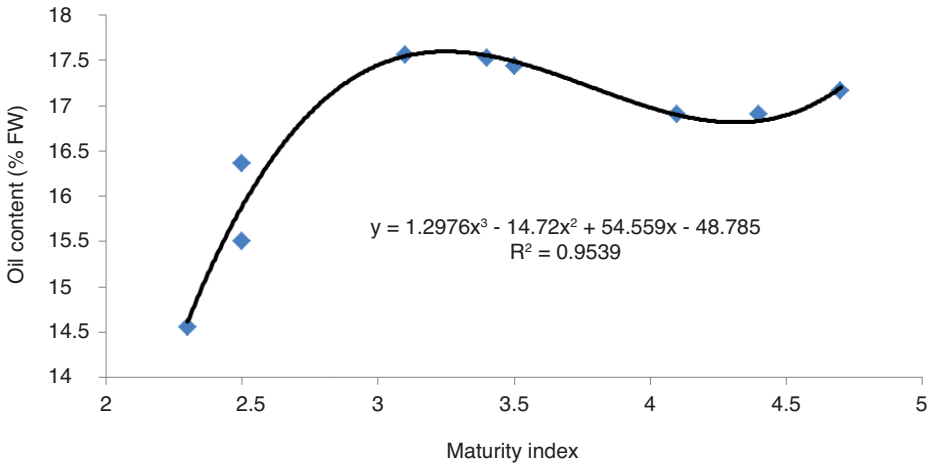


Figure 4. Ratio between the maturity index and oil content (% fresh weight) of the Haouzia clone grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year

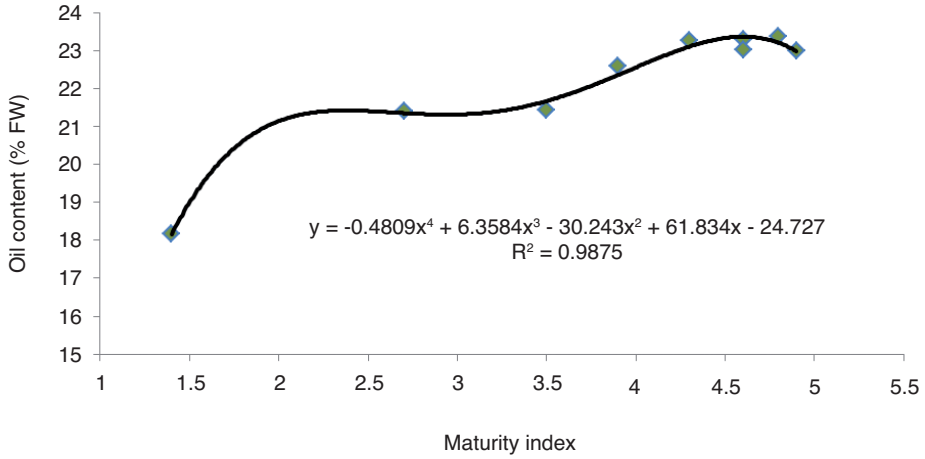
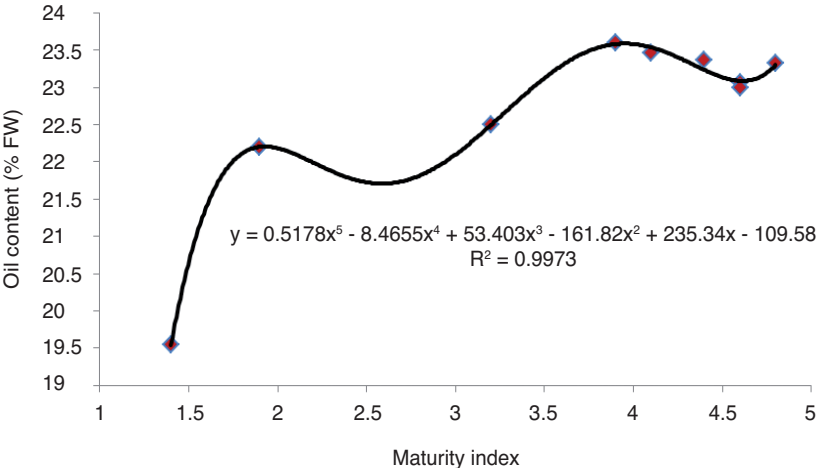


Figure 5. Ratio between the maturity index and oil content (% fresh weight) of the Menara clone grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year



rising from 2065 (mg gallic acid/100 g dried olive paste) at the green stage, to 2285 at the semi-black stage and then dropping to 1997 at the black stage. These authors concluded that the polyphenol content of olives is optimal at the semi-black stage and that it is associated with better quality oils. The polyphenols content of the Haouzia and Menara clones was higher than that of the Arbequina variety (Table 5 and Figure 6). During ripening, the polyphenols accumulated in the olive to reach maximum values of 1833 ppm for Arbequina, 2127 for Haouzia and 2134 for Menara; these values levelled off for some time before starting to fall if the olives were not harvested. The same trend in polyphenol content has been documented by Atouati (1991) who reported that phenolics content increases between the green and semi-black stages, and then drops during the black stage. In contrast, this trend was reversed in oil content, which was at its highest at the black stage. Mahhou *et al*. (2011) reported an upward trend in polyphenols content, which reached a maximum of 1823 ppm for Arbequina, 2192 ppm for Koroneiki and 2113

TABLE 5.
Maturity index, oil content (%) and polyphenol content (ppm) of the Arbequina, Haouzia and Menara varieties grown in rainfed conditions in the Meknès region of Morocco during the 2008/09 crop year

Variety	Sampling date	Maturity index	Oil content (% FW)	Polyphenols
Arbequina	22-10-2008	2.3	14.56	1731
	05-11-2008	2.5	15.50	1735
	19-11-2008	2.5	16.36	1803
	03-12-2008	3.1	**17.56**	**1833**
	12-12-2008	3.4	17.53	1828
	19-12-2008	3.5	17.44	1812
	02-01-2009	4.1	16.90	1755
	09-01-2009	4.4	16.90	1724
	16-01-2009	4.7	17.16	1722
Haouzia	22-10-2008	1.4	18.16	1833
	05-11-2008	2.7	21.40	1933
	19-11-2008	3.5	21.43	2054
	03-12-2008	3.9	22.60	2124
	12-12-2008	4.3	23.26	**2127**
	19-12-2008	4.6	**23.30**	2103
	02-01-2009	4.6	23.03	2067
	09-01-2009	4.8	23.36	2013
	16-01-2009	4.9	23.00	1987
Menara	22-10-2008	1.4	19.54	1854
	05-11-2008	1.9	22.20	1946
	19-11-2008	3.2	22.50	2123
	03-12-2008	3.9	**23.60**	2133
	12-12-2008	4.1	23.46	**2134**
	19-12-2008	4.4	23.37	2125
	02-01-2009	4.6	23.03	2112
	09-01-2009	4.6	23.00	2066
	16-01-2009	4.8	23.33	2032

ppm for Picholine marocaine, then turning downwards in early December in the three varieties studied under irrigation in the Settat region.

The ratios between the maturity index and polyphenol content of the olives were determined. The correlation between these two parameters was found to be very strong for the three varieties and is described by the following equations:

- Arbequina: $y = 53.04x^3 - 621.5x^2 + 2322x - 976.8$ avec $R^2=0.855$ (Figure 7)
- Haouzia: $y = -49.70x^3 + 436.8x^2 - 1075x + 2620$ avec $R^2=0.98$ (Figure 8)
- Menara: $y = -13.05x^3 + 65.34x^2 + 64.77x + 1673$ avec $R^2=0.98$ (Figure 9)

Mahhou *et al.* (2011) also reported the existence of a strong correlation between the maturity index and oil content of Arbequina, Koroneiki and Picholine marocaine olives grown under irrigation in the Settat region.

3.3.4. Fatty acids

The oleic acid content of the Haouzia and Menara olives (76.4% and 76.8%) was higher than that of the

Figure 6. Changes in the polyphenol content (ppm) of olives belonging to the Arbequina, Haouzia and Menara varieties grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year

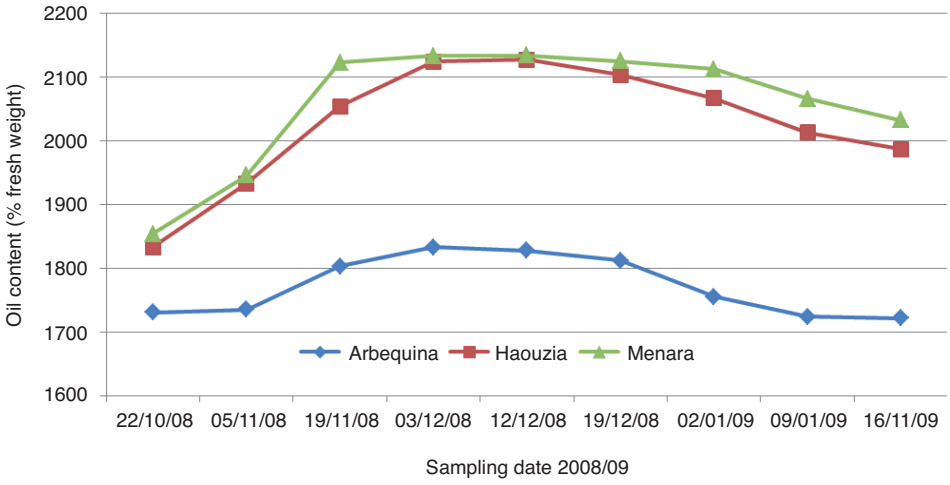


Figure 7. Ratio between the maturity index and polyphenol content (ppm) of the Arbequina variety grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year

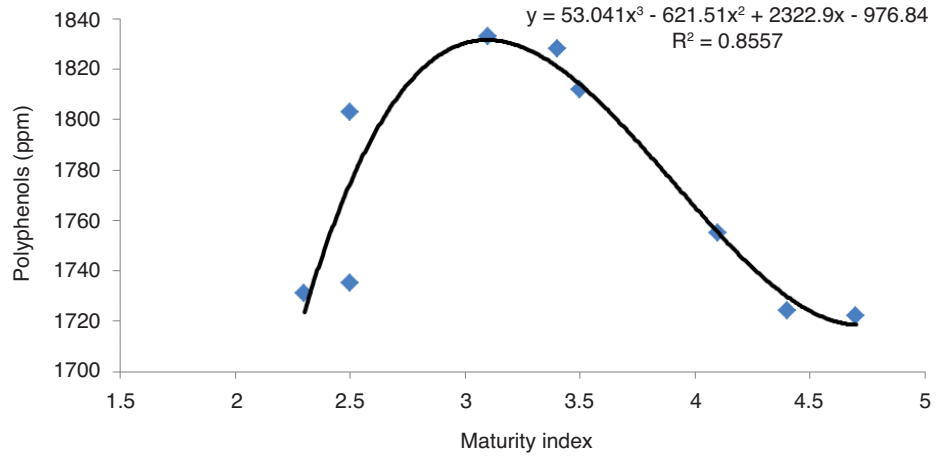
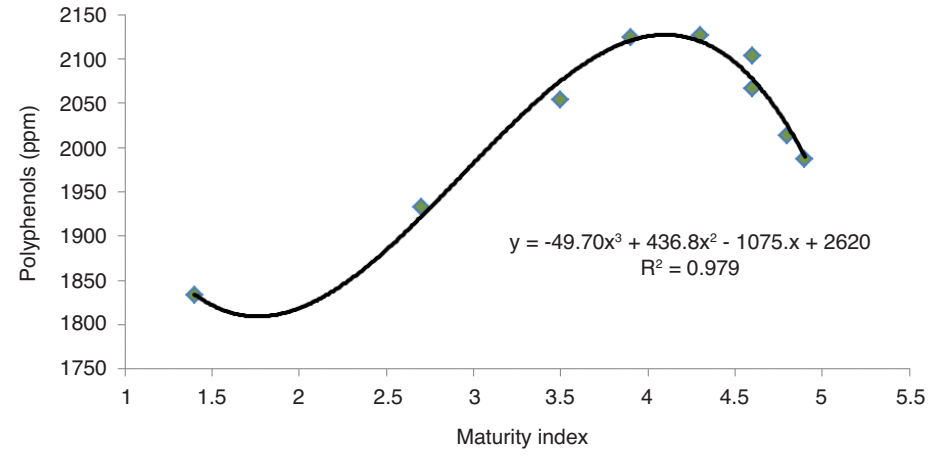


Figure 8. Ratio between the maturity index and polyphenol content (ppm) of the Haouzia variety grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year

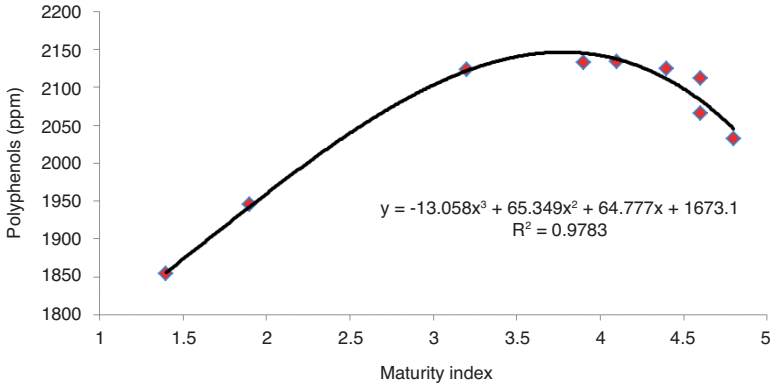


Arbequina olives (66.6%). These values concur with those reported for Haouzia and Menara (76.3% and 76.6%) by El Ajal, (2006). El Alami (2003) observed values of 74.62% for Haouzia and 68.58% for Menara. The oleic acid content of the Arbequina olives found in this research was 66.6%, which is higher than the values of 62.3% (El Antari, 2006) and 60.4% (El Antari, 2003) reported by other authors. However, other authors have reported higher values than the ones reported in this research: 68.2% (Romero and Diaz, 2005); 70.8% (Ouazzani, 2005) and 74.6% (El Ajal, 2006). Each variety of olive has its own distinctive oleic acid content. Romero and Diaz (2005) found differences in this parameter in seven varieties in which it ranged from 61.23% for Blanquetta to 78.28% for Picual. Ouazzani (2005) similarly observed this intervarietal difference ranging from 80.24% for Picual to 70.85% for Arbequina.

The linoleic acid content of the Arbequina variety (13.7%) was higher than that of Haouzia and Menara (around 10%). These values concur with the results found by other authors, namely 9.41% for Haouzia (El Ajal, 2006) and 10.9% for Menara (Ouazzani, 2005). However, El Alamy (2003) has reported values of 11.69% for Haouzia and 15.12% for Menara. In the case of the Arbequina variety, the linoleic acid content of 13.7% found in this paper concurs with the value of 13.98% observed by El Antari (2006) but is much higher than the value of 8.08% reported by El Ajal (2006).

This variation in the fatty acid contents reported by different authors can perhaps be explained by climatic and producing conditions. The oleic and linoleic acid contents of the three varieties rose until the last ten days of November, from which point they levelled off.

Figure 9. Ratio between the maturity index and polyphenol content (ppm) of the Menara variety grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year



3.4. Determination of optimum harvest period

The determination of the optimum harvest period for oil-olives is meant to identify the stage of ripening where the olives have a high oil content (quantity) and a satisfactory level of polyphenols.

Hence, this period has to be determined on the basis of oil and polyphenol content. Table 5 summarises the results for the criteria which enable the determination of the optimum harvest period.

Figures 10, 11 and 12 show the intersection period of the maximum values for oil and polyphenol content relative to the maturity index. This is the best period for harvesting the olives. These figures help to pinpoint the optimal period for harvesting the three varieties by relating the maturity index to the oil and polyphenol content of the olives (Table 6).

TABLE 6.
Optimum harvest periods for Haouzia, Menara and Arbequina on the basis of the maturity index, oil content (%FW) and polyphenol content (ppm) in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year

Variety	Maturity index	Polyphenols (ppm)	Oil content (%FW)	Optimum harvest period
Arbequina	3.1 – 3.5	1812 – 1833	17.44 – 17.56	From 03 to 19 December
Haouzia	3.9 – 4.6	2067 – 2127	22.6 – 23.3	From 03 December to 02 January
Menara	3.9 – 4.6	2112 – 2134	23.03 – 23.6	From 03 December to 02 January

Figure 10. Changes in oil content (% FW) and polyphenol content (ppm) relative to maturity index in the Arbequina variety grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year

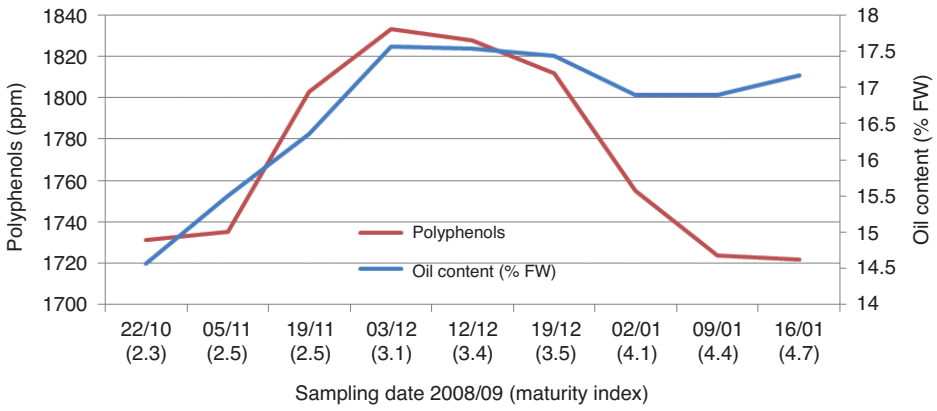
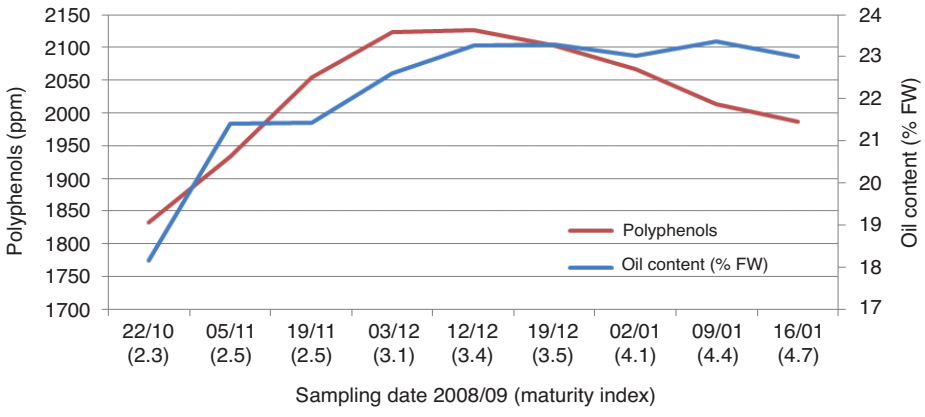


Figure 11. Changes in oil content (% FW) and polyphenol content (ppm) relative to maturity index in the Haouzia variety grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year



3.5. Yields

The crop yields of the three varieties over four crop years are reported in Table 7 and Figure 13.

In the trial conditions, Arbequina was the highest yielding variety in the 2008/09 crop year, giving a yield of 68 kg/tree, followed by Menara with 63 kg/tree and Haouzia with 48 kg/tree. The yields of the Arbequina and Menara varieties were considerably higher than the levels recorded the previous season, showing respective increases of 106% and 50% while Menara recorded a rise of 6%. Taking the means for the four crop years concerned, Menara is the top yielder with 44 kg/tree, followed by Arbequina (37.25 kg/tree) and Haouzia (35.5 kg/tree).

Figure 12. Changes in oil content (% FW) and polyphenol content (ppm) relative to maturity index in the Menara variety grown in rainfed conditions in the Meknès region of Morocco in the 2008/09 crop year

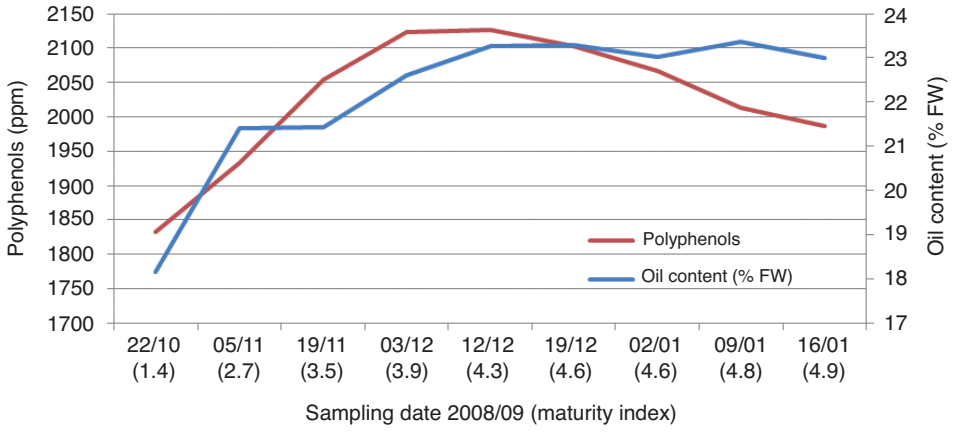


Figure 13. Yield (kg/tree) of the Arbequina, Menara and Haouzia varieties grown in rainfed conditions in the Meknès region of Morocco over four crop years

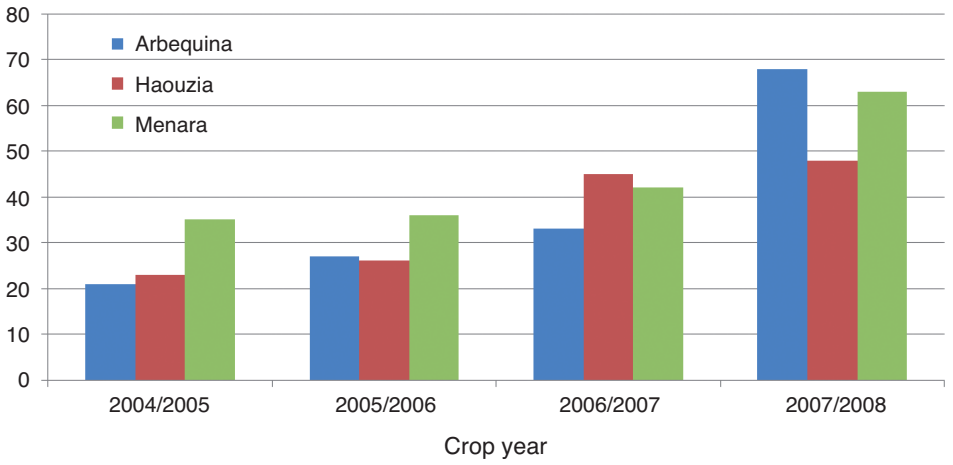


TABLE 7.
Yield (kg(tree) of the Arbequina, Haouzia and Menara varieties grown in rainfed conditions in the Meknès region

Variety	2005/2006	2006/2007	2007/2008	2008/2009	Mean
Arbequina	21	27	33	68	**37.25**
Haouzia	23	26	45	48	**35.5**
Menara	35	36	42	63	**44**

CONCLUSION

This research assessed the performance of two Picholine marocaine clones, Haouzia and Menara, and the Spanish Arbequina variety in rainfed conditions in the region of Meknès during the 2008/09 crop year. Average rainfall is 500 mm in the area; however, 700 mm were recorded for this crop year. Between early October and mid-January the maturity index increased from 2.3 to 4.7 for Arbequina, from 1.4 to 4.8 for Haouzia and from 1.4 to 4.9 for Menara. The moisture content of the olives belonging to the three genotypes tended to diminish as ripening progressed. However, the heavy rainfall recorded in the crop year led to slight increases in the moisture content of the olives at the end of the cycle. The Menara variety had a higher moisture content than the Menara and Haouzia varieties. Maximum fresh weight oil content was reached by Menara in early December when it recorded 23.6%, which is similar to the value of 23.2% found by Rafik (2008). Next highest was Haouzia, which reached a content of 23.3% in mid-December, thus lying above the value of 21.8% recorded by Rafik (2008). The maximum oil content of the Arbequina variety (17.56%) was recorded in early December and was much lower than the value of 25.7% observed by

Taeibi (2008) in irrigated orchards in the Settat region but higher than the value (16.6%) reported by Rafik (2008) in the same orchard as the one studied in this paper. Arbequina recorded a maximum polyphenol content of 1833 ppm, which is lower than the levels recorded for Menara (2134 ppm) and Haouzia (2127 ppm). In the case of the two fatty acids reviewed – oleic acid and linoleic acid – the two clones had higher levels of oleic acid, recording a content of 76.5% for Haouzia and 76.8% for Menara, compared with 66.7% for Arbequina. However, the opposite was observed for linoleic acid, with Arbequina recording a content of 13.7% compared with around 10% for Haouzia and Menara.

The optimum harvest period in rainfed conditions in the region of Meknès was determined on the basis of the oil and polyphenol content. For the crop year concerned, it was:

- From 3 to 19 December for Arbequina, with a maturity index between 3.1 and 3.5.
- From 3 December to 2 January for Menara and Haouzia, with a maturity index between 3.9 and 4.6.

The yields recorded in the 2008/09 crop year were higher than the crops borne in the same orchard in the previous three seasons, lying at 68 kg/tree, 48 kg/tree and 63 kg/tree for Arbequina, Haouzia and Menara, respectively.

The flowering period of the Arbequina variety extended from 8 until 29 April. In the case of the Menara and Haouzia clones, it began on 15 April and lasted until 5 and 8 May, respectively. The overlapping of the flowering periods of the Menara and Haouzia varieties, which are partially inter-compatible, should improve the fruit set rate of these two genotypes. Fruit set rates of 15% (Arbequina), 12% (Menara) and 11% (Haouzia) were recorded.

The findings of this research supplement those reported by Rafik (2008). They provide confirmation that the three genotypes are adapted to rainfed conditions in the region of Meknès; however, the Haouzia and Menara show some degree of superiority owing to their larger sized fruit and content and quality of superior oil. ■

REFERENCES

Atouati, B.Y., 1991. Évolution des caractéristiques carpométriques et de la fraction phénolique totale avec le stade de maturité des olives, variété Picholine Marocaine. Mémoire de 3ème cycle .Option : I.A.A. I.A.V. Hassan II, Rabat.

Boulouha B., Oukabli A., Hadiddou A., Sikaoui E., Ouguas Y., Mamouni A., 2006a. Fiche technique olivier. Edt INRA, Rabat.

Boulouha, B. 2006. Forum Oléa. Marrakech, 25 May.

Chimi H. 1987. Dosage des composés phénoliques de l'huile d'olive vierge et comparaison avec leurs pouvoirs antioxydants respectifs, Mémoire de 3ème cycle Agronomie Option IAA, IAV Hassan II, Rabat, 1987.

Civantos L, 1999. Obtención del aceite de oliva virgen. 2ª Edición. Editorial Agrícola Española, S.A. Artes Gráficas COIMOFF, SA., Madrid, Spain.

International Olive Council, 2000 (Castañeda C., Barranco Navero D., Cimato A., Fiorino P., Rallo Romero L., Touzani A., Serafini F., Trujillo Navas I): World Catalogue of Olive Varieties. Published International Olive Council, 360p.

Del Río, C., J. M. Caballero, M. Dolores García-Fernández. 2005. Chapter 2: « Variedades de olivo en España » (Libro II: Variabilidad y selección).

El Ajal A. 2006. Évaluation des performances agronomiques des variétés étrangères et locales d'olivier cultivées dans les conditions pédoclimatiques de la région de Meknès du Maroc. Mémoire de 3ème cycle, option Sciences et techniques de la production fruitières. ENA Meknès.

Elantari A., El Moudni A., Ajana H., Cert A., 2003 « Étude de la composition lipidique de deux compartiments du fruit d'olive (pulpe et amande) de six variétés d'oliviers cultivées au Maroc », Olivae n°98 pages 20-28.

EL CADI, H., JAMAÏ, M. A., 1998, « Contribution à l'étude de l'amélioration de la qualité de la production oléicole dans la zone d'action de l'ORMVA du Tadla », Mémoire de 3ème cycle Agronomie Option IAA, IAV Hassan II, Rabat.

El Antari, 2006 Étude de la qualité de l'huile des variétés d'olivier sélectionnées par L'INRA-Maroc » Edition Journée Olivier la recherche agronomique et la profession, ensemble pour le dévloppement durable de l'oléiculture nationale.

FAQIH, H., HMAMA, M., 1999, « Contribution à l'amélioration de la qualité de production oléicole dans la zone d'action de l'ORMVA du Tadla », Mémoire de 3ème cycle Agronomie Option IAA, IAV Hassan II, Rabat.

FONTOZZI, P., MONTEDERO, G. 1978 « Dosage des composés phénoliques dans les drupes des olives récoltées à différents stades de maturation ». IAA, 1335-1339.

Hadiddou A., K. Bencheqroun O., Mamouni A., Oukabli A., Mekaoui A., 2006 « Performances des obtentions INRA d'olivier et des variétés étrangères sous les conditions pluviales » Edition Journée Olivier la recherche agronomique et la profession, ensemble pour le développement durable de l'oléiculture nationale.

Harrar, 2007. Contribution à l'amélioration de la qualité de la production oléicole dans la zone d'action de la DPA de Khénifra. Mémoire de troisième cycle pour l'obtention du diplôme d'Ingénieur d'État en Industries Agricoles et Alimentaires, IAV Hassan II, Rabat.

Idrissi, My A., 1994. Diagnostic du Secteur Oléicole au Maroc. Cas du Haouz. Mémoire de 3ème cycle. Département de Chimie-Biochimie alimentaire I.A.V. Hassan II, Rabat.

LACHIR, B., SIDI BABA, A. 1994 « Diagnostic du secteur oléicole au Maroc, cas de la région de Fès », Mémoire de 3ème cycle Agronomie Option IAA, IAV Hassan II, Rabat.

LAMRINI M., 1995 «Détermination de la date optimale de maturité des olives à huile (var, Picholine marocaine)», Mémoire de 3ème cycle Agronomie Option IAA, IAV Hassan II, Rabat.

MAPM, 2012 : Bilan de la campagne oléicole, DPV, Ministère de l'Agriculture et de la Pêche Maritime, Rabat Maroc.

Mahhou & al. (2011) OLIVAE n° 116, 2011, Performance et qualité de production des variétés d'olivier Arbequina, Koroneiki et Picholine marocaine conduites en irrigué dans la région de Settat (Maroc).

Ouazzani N., Idrissi A., El Ghazi N., Lumaret R., 2002. Varietal structure of Moroccan olive germplasm: evidence from genetic markers and morphological characteristics. Acta Hort. 586, 30 October, Valenzano, Italy.

Rafik, 2008, Évaluation du comportement de 11 variétés d'olivier (Carolea, Leccino, Arbequina, Manzanilla, Agvalik, M26, K26, S19), retenues par l'INRA, en zone pluviale (Ain Taoujdate) » Mémoire de troisième cycle

pour l'obtention du Diplôme d'Ingénieur d'Etat en Agronomie option Horticulture. IAV Hassan II, Rabat.

Rallo Romero L., Barranco D., Caballero J. M., Del Rio C. Martin A., Tous J., Trujillo I., 2005. Variedades de olivo en Espana. Eds. Junta de Andalucia, MAPA y Ediciones Mundi-Prensa, Madrid.

Romero Agustí, Joan Tous, Juan F. Hermoso, Joan Plana, 2005 Dept. d'Arboricultura Mediterránia, Centre "Mas Bové", IRTA, Reus-Constantí (Tarragona); Estación Experimental del Ebro, IRTA, Amposta (Tarragona), Espagne.

Rahmani, M. ; Lamrini, M., Csallany, S., 1997, « Mise au point d'une méthode simple pour déterminer l'époque de récolte des olives », Olivae n° 69, pp 48-51.

Sweeney, S. 2005. National olive variety assessment – NOVA- Stage 2. Rural Industries Research and Development Corporation Publication No. 05/155, Project No. SAR-47A.

Suarez M.P., Fernández-Escobar R., Rallo L., 1984. Competition among fruit in olive. II. Influence of inflorescence or fruit thinning and cross-pollination on fruit set components and crop efficiency.Acta Horticulturae, 149:131-143.

Taiebi, 2008 « Production et qualité d'huile de trois variétés d'olivier : Arbequina, Koroneiki et Picholine marocaine dans la région de Settat », Mémoire de troisième cycle pour l'obtention du Diplôme d'Ingénieur d'État en Agronomie option Horticulture. IAV Hassan II, Rabat.

Tous J., Romero A., Plana J., 2005. Vigor – Chapter 2, *in* Variedades de olivo en España (Libro II: Variabilidad y selección).

Uceda M., Frías L., 1975. Época de recolección, evolución del contenido graso del fruto y de la composición y calidad del aceite. II seminario oleícola internacional. Córdoba.

Vázquez Roncero A., Janer Del Valle C., Janer Del Valle M.L, 1973,« Détermination de la teneur en polyphénols totaux dans l'huile d'olive ». Grasas y Aceites, Vol. 24, N° 6, pp 350-357.

Vossen, P. 2005. Producing Olive Oil. In Olive Production Manual, 2nd edition. University of California Ag and Natural Resources publication, 157–173.

Walali L., Chmitah M., Loussert R., Mahhou, A., Boulouha B., 1984, « Caractères morphologiques et physiologiques de clones d'olivier, Picholine maroaine ». Olivae n°3, pp 26-31.

Design and management of olive hedgerow orchards: effect on oil productivity and quality

M. Gómez del Campo[1], E. Trentacoste[1,2] and D. J. Connor[3]

[1] Dpto. Producción Vegetal: Fitotecnia, Universidad Politécnica de Madrid.
[2] Estación Experimental Agropecuaria Junín (INTA), Mendoza, Argentina.
[3] School of Land and Environment, The University of Melbourne, Victoria, Australia.

ABSTRACT

Olive hedgerow orchards designed for harvest with modified grape harvesters are a viable alternative to orchards designed for harvesting with trunk shakers. The main advantages are high yields during early years, low harvesting cost, minor workforce and fast harvesting at optimum times, but these features are only possible with optimal structures, maintained over time. The interception of solar radiation is the decisive factor for olive oil production; so, optimal hedgerow structure has to maximise canopy illumination. Because olive hedgerow orchards are a recent innovation, the response of oil productivity and quality to alternative structures in various locations is unknown. Simulations of oil productivity and quality for different designs will be useful for the sector to establish optimum structures for individual conditions. This paper describes recent progress and remaining challenges for both farmers and researchers.

Key words: super-intensive olive orchard, harvesting mechanisation, hedgerow structure, row width, orientation.

OLIVE HEDGEROWS

Before going into hedgerow characteristics it is important to describe some specific features of the olive canopy. Olive leaves are small and evergreen and have a high specific weight while olive fruits are small and distributed over the canopy, chiefly in well lit areas. Both are positioned on supple shoots. This set of vegetative and fruit-bearing organs can be trained to several shapes, the most common of which are the vase and hedgerow (Figure 1). The hedgerow is a system where the canopy foliage is distributed continuously along the orchard row and has two vertical or slightly angled walls. To achieve a continuous mass of foliage it is necessary to shorten the spacing between the trees along the row. This cultivation system not only increases planting density but also modifies the geometrical arrangement of the canopy, thus affecting the microclimate of the leaves, fruit and soil and modifying intercepted radiation, temperature, wind and moisture. This causes major changes in olive response because the physiology of trees which stand separately differs from that of trees trained to a hedgerow.

Hedgerows differ greatly in size, depending on growing conditions and management (Figure 2), and can be planted at different densities. In commercial olive orchards tree height can range from 2.5 to 5 m and canopy width from 1 to 4 m.

HEDGEROWS DESIGNED FOR GRAPE PICKER HARVESTING

High harvesting costs have been the determinant of hedgerow size. For many crops, olive included, the possibilities offered by continuous harvesting machinery have significantly increased competitiveness. Although Bravigrieri proposed this orchard management system to achieve high yields in Italy as far back as 1961, it was ruled out because tree development

Fig. 1. Different olive growing systems: traditional, low-density orchard trained to a vase shape (*left*); intensive orchard with vase shaped trees planted at twice the density of the traditional orchard (*centre*), resulting in higher external surface area (ESA) and higher canopy volume; super-intensive hedgerow orchard designed for harvesting with a grape picker (*right*) where, although the canopy is half the volume, the ESA is almost double.

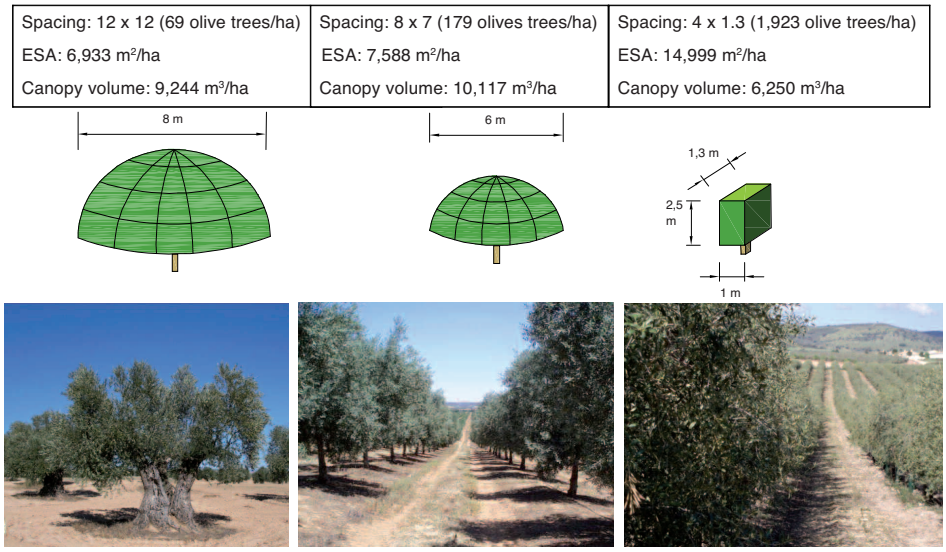
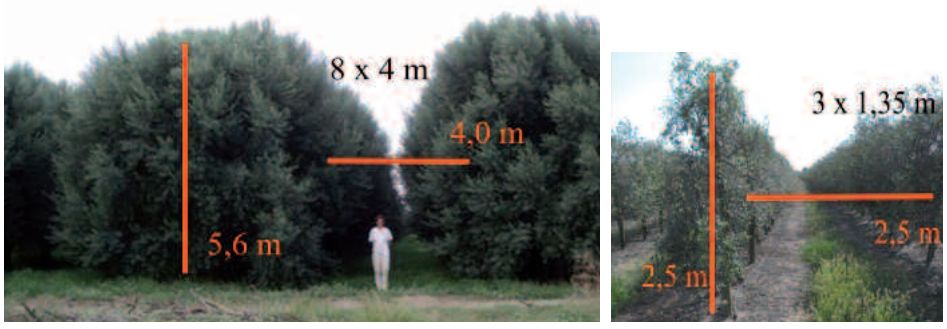


Fig. 2. Different geometrical arrangements of cv. 'Arbequina' hedgerows: tall hedgerows planted on an 8x4 m spacing (313 olive trees/ha) in Argentina and Australia, initially trained to a vase shape, which eventually formed a continuous canopy wall due to high vigour (*left*); smaller hedgerows achieved in olive orchards planted at high densities (1,975 trees/ha) in low-vigour conditions and trained to a central leader (*right*). The horizontal line is the distance between adjacent row walls and the vertical line is the height.



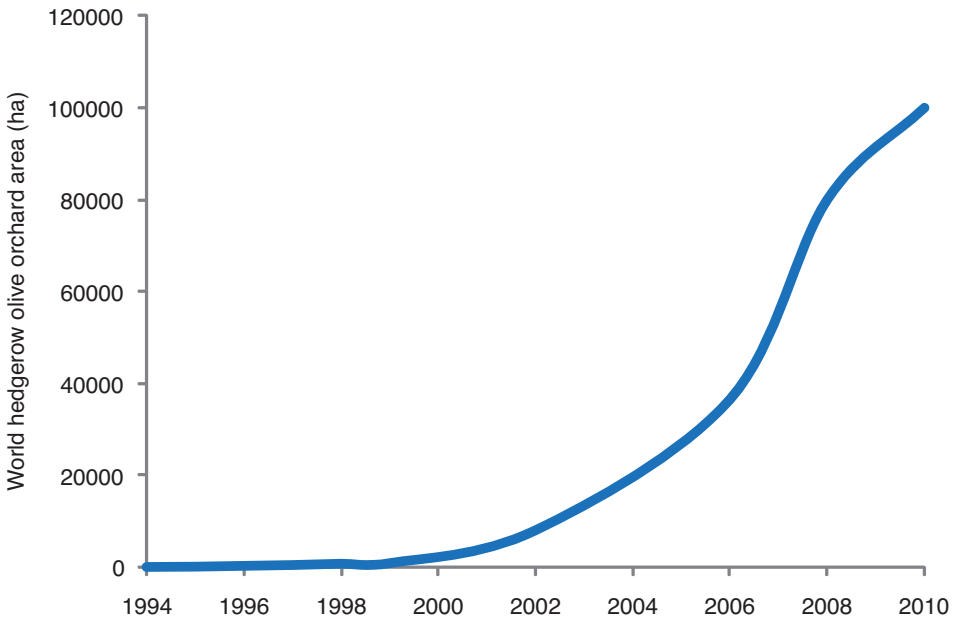
Catamarca, Argentina **Tarragona, Spain**

was excessive and no harvesting solution could be found (Morettini, 1972). Subsequently, in the late 1990s, the olive-growing industry in Spain started to plant hedgerow olive orchards for harvesting with grape pickers (Figure 3). These pickers had been developed 30 years earlier in the United States to harvest grapes and, with some minor modifications, can be used to harvest olives in the early years of orchard establishment. Larger olive harvesting prototypes later made their appearance, paving the way for larger volume hedgerows. Since then, the world area planted with hedgerow olive orchards designed for grape pickers has expanded without interruption (Figure 4). Planting density varies. In irrigated orchards it ranges from 1,250 to 1,975 trees/ha, practically ten times the density of traditional olive orchards, hence the name of super-intensive olive orchards. Currently, in water-short environments rows are spaced up to 5 and 6 m apart with a distance of 1.5–2.0 m between the trees. As a result, planting densities decrease to 833 and 1,333 trees/ha.

Hedgerow olive orchards are a viable alternative to intensive vase shaped orchards (200–350 trees/ha) designed for harvesting with trunk shakers. Hedgerow olive cultivation owes its competitiveness to the higher, early yields obtained, the low harvesting costs and the smaller amount of labour required. Additionally, top quality oil is obtained under this planting system because the olives can be harvested at any time and are picked solely from the tree. At the same time, the high speed of harvesting reduces the time it takes to get the olives from field to mill. The design and management of hedgerow olive orchards nevertheless raises question marks and challenges for farmers and researchers, some of which will now be discussed.

Fig. 3. Hedgerow olive orchards designed for harvesting with modified grape pickers first made their appearance in Spain in the 1990s. The industry managed to achieve a type of orchard design and management suited to harvesting with this continuous-operating machinery, which had undergone innovations and improvements for over 30 years. The orchard has to be managed in such a way as to allow the machinery to work swiftly while causing the least possible damage to the tree. Current grape picker models are capable of harvesting hedgerows up to 3.30 m in height and 1 m in width.

Fig. 4. Trend of world area under hedgerow olive orchards designed for harvesting with grape pickers.



RADIATION AND OLIVE YIELD

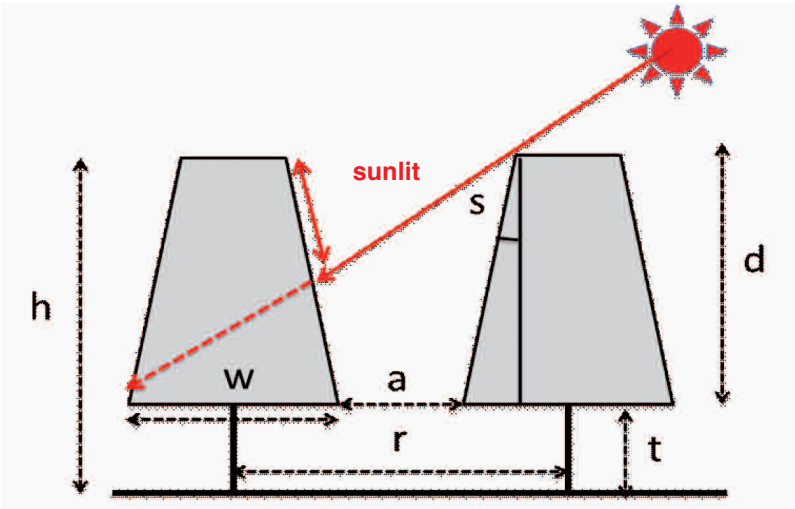
Olive yield depends on radiation interception and distribution inside the hedgerow; however, little research has been performed on olive response to light because radiation is not a limiting factor in traditional orchards. Productivity decreases in hedgerow olive orchards where a proper structure is not maintained over time (Pastor *et al*., 2007). In the first experiments with closely planted hedgerows it was observed that as the hedgerows grew in height the fruit crop gradually moved upwards, leaving the lower parts of the canopy bare of olives. Consequently, hedgerow design and management is a decisive factor in ensuring lasting optimal characteristics and planting system profitability.

In 2006 the Universidad Politécnica de Madrid, in partnership with The University of Melbourne (Australia), undertook preliminary research on hedgerow olive cultivation with the aim of equipping the industry with tools for the design and management of optimally sized hedgerows. The first step was to design a tool permitting rapid assessment of the radiation intercepted by hedgerows of differing characteristics. Connor (2006) developed a theoretical model to predict the radiation intercepted by the different layers of a solid hedgerow (non-porous wall) on the basis of row characteristics (height, spacing and orientation), site latitude and day of the year. The latter two parameters determine solar elevation and solar azimuth (Figure 5). This model is based on the models developed by Cain (1972) and

Jackson and Palmer (1972; 1980). On observing that many hedgerows adapted to grape pickers are narrow and porous, Connor *et al*. (2009) incorporated porosity into the model. As the sun rises and solar azimuth changes, the row walls are illuminated from the highest parts downwards. The sunlight length of row wall is decisive in row illumination and photosynthesis response. The model also allows for diffuse radiation which, though small (around 10%) compared with horizontally incident radiation, is important on overcast days (Connor *et al*., 2009). This model was validated on the basis of daily measurements of incident radiation on rows oriented N-S and E-W. The first model estimates the profiles of incident radiation on the external face of the row, then calculating intercepted radiation (incident radiation less the radiation passing through the row) from the row porosity estimates obtained from photographs taken against a red or white background (Figure 6).

The next step in our research was to determine the response of oil production and oil components at different levels of radiation in order to determine the radiation thresholds for the different physiological processes involved. Earlier research had already reported the radiation sensitivity of floral initiation, vegetative growth, fruit set, fruit size and oil content (Ortega-Nieto, 1945; Tombesi and Standardi, 1977; Tombesi and Cartechini, 1986; Acebedo *et al*., 2000). A linear increase was observed in dry fruit weight and oil yield as the radiation intercepted by the rows increased in October, when oil synthesis is at its height (Figure 7). Yield largely depends, however, on the number of olive fruits, which depends in turn on radiation but varies in response according to orchard and year. Analysis of data for 11 olive orchards gave a value of R^2=0.30 for the relationship between fruit density and radiation; however, when individual ratios were established, R^2 increased to 0.70 (Connor *et al*. 2012).

Fig. 5. Cross-section perpendicular to the hedgerow. Hedgerows are geometrically defined in terms of depth (*d*), slope (*s*) to vertical (*s* = 0 for rectangular canopies) and width at the base (*w*). Row height (*h*) is *d*+*t* where *t* is height above ground level maintained free of canopy. Individual hedgerows are separated in planting lines by distance (*r*), giving a free alley width (*a* = *r* – *w*). Together with row porosity and orientation, these parameters enable hedgerow characterisation.



RADIATION AND OIL QUALITY

Fruit position in the hedgerow likewise determines oil quality (Figure 8). The few data available have to be considered preliminary. Gómez-del-Campo and García (2012) have reported that olives positioned in the upper parts of the canopy give more stable oils due to their higher content of polyphenols. On the other hand, olives located on the lower parts of the row give higher-oleic oils.

OPTIMAL DESIGN AND UTILITY OF YIELD AND QUALITY SIMULATIONS

Orchard design and management should enable the

Fig. 6. Effect of alley width (1, 2 and 3 m) at 35 ºN on the radiation intercepted by a rectangular canopy, 2-m deep and 1-m wide, oriented N-S and E-W. The length of the sunlit row depends on the distance between the row walls. Yearly changes differ in hedgerows of differing geometrical characteristics. Rows oriented N-S receive the same radiation on both sides while side E receives sun in the morning and side W in the afternoon.

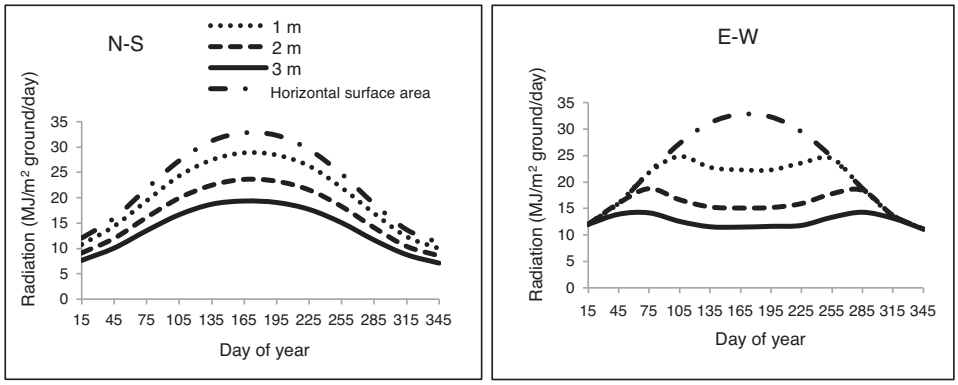


Fig. 7. Relationship between yield components and daily incident radiation in October in various hedgerow orchards, cv. 'Arbequina', orented N-S (Connor *et al.,* 2012). Blank circles represent the upper levels of the row receiving daily radiation in excess of 6 MJ m^{-2}. In the areas receiving more than 6 MJ $m^{-2}d^{-1}$, fruit density (*left*) is horizontal =1000 fruits m^{-2}, below which density decreases linearly. Dry weight (*centre*) and fruit oil content (% dry matter) (*right*) increase linearly with incident radiation.

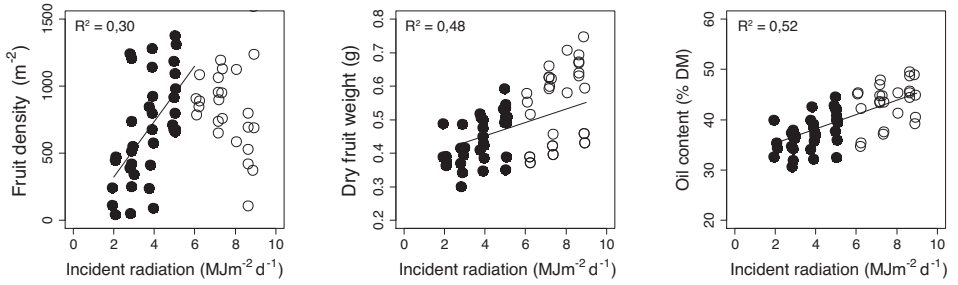
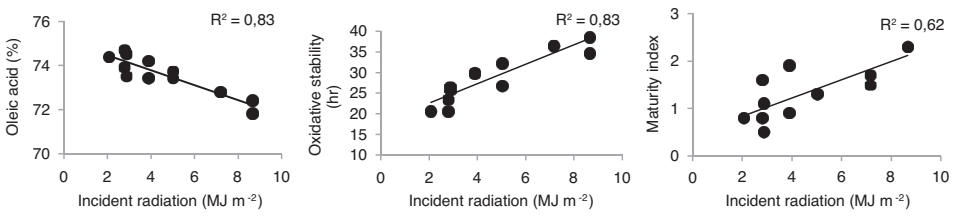


Fig. 8. Relationship between chemical oil characteristics and fruit maturity index and daily incident radiation in October in nine olive orchards, cv. 'Arbequina' (Gómez-del-Campo and García, 2012). Oleic acid concentration (*left*) decreases as incident radiation increases whereas oxidative stability (*centre*) and maturity index (*right*) increase as incident radiation increases



hedgerow to maintain optimal geometrical characteristics to achieve maximum returns. So far, our research has focused on rows oriented N-S, which are therefore illuminated symmetrically on both faces through the day. Maximum yields are obtained when the entire row wall receives radiation above a threshold value and the whole row structure is solid, without any spaces between the bottom or upper parts of the trees, thus achieving maximum photosynthetic area per row length. The radiation model makes it possible to calculate the radiation intercepted by the different layers as well as by the row as a whole (Figure 5). By applying the yield (Figure 7) and quality models (Figure 8) to radiation interception it is possible to simulate yield (Figure 9) and oil quality (Figure 10) in rows of differing geometrical characteristics (Connor and Gómez-del-Campo, 2013).

It is interesting to give a brief description of the information generated by these simulations. In rows oriented N-S, row spacing is optimal when free alley width (row spacing – row width) equals row height (Figure 9). Narrower hedgerows give higher yields because shorter alley widths lead to higher row lengths per hectare. Row illumination can be increased

by sloping the walls at an angle in rhomboidal shapes, chiefly in wider rows. Rhomboidal shapes respond to changes in illumination by achieving higher yields, partly because of the narrower alley width. On simulating yields at latitudes between 30 º and 40 º it was observed that yield decreased with latitude but without change to optimal row spacing.

The few data currently available on the effect of radiation on oil quality are solely for cv. 'Arbequina' (Gómez-del-Campo and García, 2012). The oil produced from this variety is highly rated for its organoleptic characteristics; however, it is sensitive to oxidation and has a low content of oleic acid. Consequently, oxidative stability and oleic acid content are the two quality parameters that have been simulated (Figure 10). Oleic acid content decreases as row spacing increases, but does so to a lesser extent in narrow, shaded rows than in wide rows. Stability and maturity index increase as row spacing increases and the increases recorded are higher in narrow, short rows than in tall, wide rows.

Fig. 9. Simulated oil yield for rectangular rows oriented N-S at 35 ºN of differing depth (2, 3 and 4 m), width (1 m -*left* and 3 m - *right*) and row spacing (2 to 8 m) (Connor and Gómez-del-Campo, 2013). 1-m wide rows (*left*) are designed for harvesting with grape pickers whereas 3-m wide rows have to be harvested with bulkier machinery. All the rows achieve maximum yield when row spacing is equal to their depth. Maximum yield is achieved in the tallest rows. Narrower rows achieve higher yields.

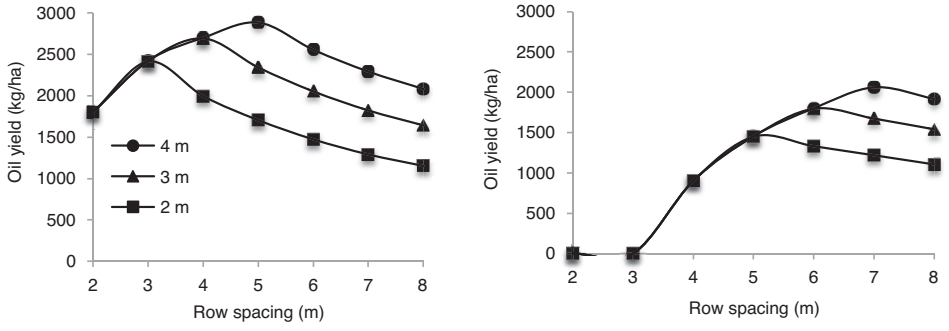
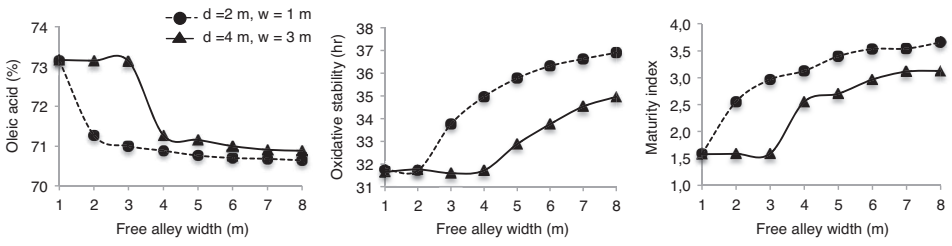


Fig. 10. Simulated effect of free alley width (1 to 8 m) on oil quality and maturity index of rectangular rows oriented N-S at 35 ºN of two depths (d= 2 and 4 m) and widths (w= 1 and 3 m) (Connor and Gómez-del-Campo, 2013).



According to the simulations reported, no single solution exists and maximum yield can be obtained in rows of differing characteristics. However, the optimal design is the one which makes it possible to maximise yield while ensuring easy, cost-effective management. It has to be borne in mind that the highest yielding row will not always be the most profitable. Easy, cost-effective management basically equates with two goals: facilitating the use of cheap machinery for cultural practices (harvesting, pruning, crop health treatments) and achieving a hedgerow that allows air to circulate, so keeping it healthy. It has been observed that the greater the row spacing, the higher and wider the rows need to be to intercept maximum radiation; the taller and wider the row, the more expensive the harvesting machinery and the more difficult it is to prune the trees. This would appear to suggest that low, narrow rows are more profitable.

HEDGEROW ORIENTATION: A QUESTION YET TO BE RESOLVED

Most hedgerows are oriented N-S; however, there are situations (site geometry, slope of the land) where this is not possible and where it

might even be advantageous to modify row illumination by using other orientations. This raises the question of the impact of row orientation on yield and oil quality.

Row orientation will largely determine the amount and distribution of radiation and its effect on the physiological processes in olive. Figure 6 plots the changes in the radiation intercepted by rows oriented N-S and E-W. N-S orientation exposes the crop to high levels of summer radiation whereas rows oriented E-W receive more radiation at the beginning of spring and in the autumn, coinciding with the period of oil synthesis and accumulation in olive.

The greatest differences generated by row orientation occur in the distribution of radiation on both faces of the row. When oriented N-S, both sides receive similar radiation throughout the day, side E is illuminated during the first half of the day and side W after midday. When the row is oriented E-W, the S side (in the northern hemisphere) is exposed to solar radiation for most of the day whereas the N side remains shaded, except for short periods in the morning and afternoon in the summer. Consequently, the N side is dependent on diffuse sky radiation (which has less energy), the radiation reflected by the adjacent row and the radiation transmitted from the sunlit side. These relationships are altered by the presence of pores or foliage-free spaces in the row wall, which allow the solar beams to penetrate through to the other side of the row. Row porosity has a greater effect in E-W orientations and allows the periods of greater radiation interception (winter-spring and autumn) to coincide with the periods when the solar beams are at higher angles, so increasing the radiation transmitted from side S to side N. In rows oriented N-S, the radiation which penetrates through side E and reaches side W in the morning is offset by the radiation in the reverse direction in the afternoon.

Little information is available about the effect of row orientation in olive. In other fruit crops (apple, pear and vine), a 15–25% increase in yield has been reported in N-S orientations compared with E-W (Khemira *et al*., 1993), but this higher N-S productivity cannot be generalised because it is dependent on the phenological cycle of the crop, structural row characteristics and site latitude. The aim is to make row illumination coincide with the critical stages that determine yield and quality.

Row arrangement alters other factors directly related to solar radiation such as temperature. Many biological processes involved in fruit growth, development and quality are dependent on fruit temperature. The difference between fruit and environmental temperature will increase in conditions of greater incident radiation and slower wind speed. The differences in radiation and temperature generated by row orientation can affect the chemical composition of the oils. In recent research conducted in hedgerow olive orchards, cv. 'Arbequina' oriented N-S and E-W, Gómez-del-Campo and García (2012) reported higher oleic acid content and lower palmitic and linoleic acid content in oil extracted from fruit growing on the E side (N-S oriented row) and N side (E-W row) than on the opposite side. In addition, irrespective of fruit position on the row, the oil obtained from fruits on N-S rows had a higher content of phenolic compounds (i.e. antioxidants) than those oriented E-W. To achieve top oil quality, it is essential to determine the optimal harvest time and to ensure that the stage of fruit ripening is uniform. Row orientation alters fruit maturity index in that fruit located in rows oriented E-W ripen earlier than those positioned N-S. This is associated with greater incident radiation in the autumn on the S side of rows oriented E-W. It should be borne in mind, however, that greater differences are

generated in the radiation received on the S and N sides of low-porosity rows oriented E-W; as a result, the stage of ripening is more heterogeneous.

Choosing the right row orientation is more important in radiation-limited environments (narrow hedgerows, high cloud cover). The research cited only investigated N-S and E-W orientations, but clearly there are intermediate orientations which should be studied to ascertain their impact not only on yield and quality, but also on water demand, low-temperature damage and weed management. Data from established trials (Figure 11) will help to clarify these issues.

Fig. 11. Experimental plot planted in 2008 at Puebla de Montalbán (Toledo), cv. 'Arbequina', at four orientations (N-S, E-W, and the intermediate orientations NE-SW and NW-SE), with the support of several companies. The University of Córdoba and the firm Todolivo (Córdoba) subsequently set up similar trials

CHALLENGES

There are some limitations on planting olive hedgerow orchards for harvesting with grape pickers: initial high investment, moderate site slope and need for sufficient water (rainfall or irrigation water). Another question that has to be addressed is the small range of varieties suited to this system. The characteristics sought in a variety intended for hedgerow cultivation are: quick start of bearing; regular, large, high quality yields; and low vigour. Few varieties fulfil these requirements. Currently, super-intensive olive orchards are being planted with 'Arbequina', and to a lesser extent 'Arbosana' and 'Koroneiki'. Planting large areas with a single variety raises problems in terms of harvest and crushing control. Moreover, this system is not feasible in small orchards unless nearby orchards use the same machinery for harvesting.

At present, the two major challenges are to determine the optimal structure and to maintain that structure. To determine the optimal hedgerow design it is necessary to determine the radiation thresholds for oil production. Radiation helps to explain some yield parameters (fruit size and oil content) but radiation levels are not the sole determinant of the number of olives, which is influenced by other potential factors such as temperature. In addition, it is neces-

sary to ascertain the radiation response of yield and oil quality in different varieties because the data available so far are primarily for cv. 'Arbequina' hedgerow olive orchards. Few data are available on rows oriented E-W, and those that are available show that the response to radiation, and probably to temperature, differs from that of N-S rows.

For row structure to be maintained it is important to design the orchard properly in the light of the size of the harvest machinery available as well as of the environmental, soil and crop conditions determining olive growth potential at the site. In regions where the crop shows low vigour, it will be possible to use low, narrow rows suited to smaller machinery whereas in regions conducive to high plant vigour, optimal row size will be bigger and management should aim at illumination of the whole canopy. Vegetative growth can be controlled through proper irrigation and fertilisation management and, in the last instance, through pruning. The results of ongoing research by several teams on deficit irrigation in hedgerow olive orchards will supply information for the application of this strategy and the reduction of vigour.

The biggest challenge, however, faces table olive orchards. The industry has initiated some experimentation on hedgerow table olive orchards. The challenges involved are greater than for oil-olives because besides achieving high yields, the olives have to be the right size and to reach the processing facility intact.

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REFERENCES

Acebedo M.M., Cañete M.L., Cuevas J. 2000. Processes affecting fruit distribution and its quality in the canopy of olive trees. Adv. Hort. Sci. 14: 169-175.

Cain J.C. 1972. Hedgerow orchard design for most efficient interception of solar radiation. Effects of tree size, shape, spacing, and row direction. Search Agric. 2:1-14.

Connor D.J. 2006. Towards optimal designs for hedgerow olive orchards. Aust. J. Agric. Res. 57:1067-1072.

Connor D.J., Centeno A., Gómez-del-Campo M. 2009. Yield determination in olive hedgerow orchards. II. Analysis of radiation and fruiting profiles. Crop Pasture Sci. 60:443-452.

Connor D.J., Gómez-del-Campo M., Comas J. 2012. Yield characteristics of N–S oriented olive hedgerow orchards, cv. Arbequina. Sci. Hort. 133: 31–36.

Connor D.J., Gómez-del-Campo M. 2013. Simulation of oil productivity and quality of N-S oriented olive hedgerow orchards in response to structure and interception of radiation. Sci. Hort. 150:92-99.

Gómez-del-Campo M., García J.M. 2012. Canopy fruit location can affect olive oil quality in 'Arbequina' hedgerow orchards. J. Amer. Oil Chemists 89:123-133.

Jackson J.E., Palmer J.W. 1972. Interception of light by model hedgerow orchards in relation to latitude, time of year and hedgerow configuration and orientation. J. Applied Ecol. 9:341-357.

Jackson J.E., Palmer J.W. 1980. A computer model study of light interception by orchards in relation to mechanized harvesting and management. Sci. Hort. 13:1-7.

Khemira H., Lombard P.B., Sugar D., Azarenko A.N. 1993. Hedgerow orientation affects canopy exposure, flowering, and fruiting of Anjou pear trees. HortScience 28: 984-987.

Morettini A. 1972. Olivicoltura. Ramo Editoriale Degli Agricoltori. Roma. Italia.

Ortega-Nieto J.M. 1945. Poda del Olivo, con aplicaciones especiales a la zona de Ubeda y 'El Condado' (Jaén). Editora El Olivo, S.S.L. Jaén. España.

Pastor M., García-Villa M., Soriano M.A., Vega V., Fereres E. 2007. Productivity of olive orchards in response to tree density. J. Hort. Sci. Biotechnology 82: 555-562.

Tombesi A., Standardi A. 1979. Influenza della defogliazione, della eliminazione dei frutti e della decorticazione anulare sulla differenziazione delle gemme a fiore nell'olivo. Annali della Facoltà di Agraria di Perugia 33: 407-421.

Tombesi A., Cartechini A. 1986. L'effetto dell' ombreggiamento della chioma sulla differenziazione delle gemme a fiore dell' olivo. Riv. Ortoflorofrutticoltura Italiana 70: 277-285.

Antioxidant potential of olive pruning wood extracts, cv. Arbequina, from Catamarca, Argentina

C. Salim Rosales, P. Gómez, E. Gómez, N. Rodríguez and M. Bravo

Facultad de Ciencias Agrarias, Universidad Nacional de Catamarca. analiticafcaunca@gmail.com

ABSTRACT

The olive growing area of Argentina amounts to 105,000 ha, most of which is farmed under the intensive production system where pruning is a customary management practice. Little is known, however, about the potential of olive pruning wood as a source of natural antioxidants. This paper appraises the antioxidant properties of extracts of olive wood, cv. Arbequina, from the Valle Central area of Catamarca (Argentina), given that these are the variety and province with the largest olive crop area in the country. The extracts were prepared with 50% aqueous ethanol and ethyl acetate and assayed for their polyphenol content, DPPH radical scavenging activity and antioxidant activity by adding them to virgin olive oil subjected to oxidation at 80 ºC for five days. The progress of oxidation in the treated and control oils was monitored by determining total polyphenols, peroxide value, ultraviolet absorbance (K_{232} and K_{270}), free acidity and acid profile. The results show that aqueous ethanol has a greater extraction capacity than ethyl acetate, thus highlighting the polarity of the chief active ingredients extracted. Furthermore, the highest extract concentrations of those assayed (100, 300 and 600 ppm) displayed a DPPH inhibition rate (IR) of more than 90%, 15 minutes after mixing the extract with the free radical. When applied to virgin olive oil at ambient temperature, the extracts had a slight protective effect but did not demonstrate antioxidant activity when subjected to forced oxidation. It is recommended adding the extracts to other systems permitting better interface distribution of the supplemental phenols, so stimulating antioxidant protection mechanisms. In view of the growing interest in the use of natural antioxidants, these extracts, or their derivatives, could become a valid marketable alternative for the olive growing industry.

Key words: *Olea europaea* L., wood, extracts, antioxidants, total polyphenols, DPPH.

INTRODUCTION

At present, some 105,000 ha of olives are grown in Argentina, the top producer and exporter of olive oil and table olives in the Americas (Federación Olivícola Argentina, 2011). The chief producing provinces are, by order of crop area, Catamarca, La Rioja, Mendoza, San Juan, Córdoba and Buenos Aires (Federación Olivícola Argentina, 2011; Peter Searles *et al*., 2011). Olive growing is one of the main economic activities in the arid valleys of the Cuyo and North-west regions of Argentina.

Crop acreage in Catamarca expanded significantly from the 1990s, spurred by tax benefit policies (Act 22.021) and favourable conditions on foreign markets. As a result, numerous companies set up intensive, high-technology olive plantations in the province. Nowadays, 24,500 ha are under olive trees in Catamarca (Searles, *et al*., 2011), distributed among the regions of Valle Central (Capayán and Valle Viejo), Bolsón de Pipanaco (Pomán) and the highlands

(Tinogasta). Approximately 80% of the varieties grown are for oil production, notably Arbequina, Frantoio, Barnea and Coratina. The remaining 20% is made up of dual-purpose varieties such as Manzanilla (Matías *et al*., 2012). The large percentage of crop area planted with Arbequina makes this variety preponderant in the province and positions Catamarca as the top producing hub of Arbequina olive oil outside Catalonia (Andrada *et al*., 2008).

In Catamarca, 90% of olive oil output goes to foreign markets while 10% is for the domestic market. In the case of table olives, 70% of the volume processed by the industry is exported to foreign markets, leaving 30% for the home market where it goes to supermarkets, wholesalers and retailers (Cáceres *et al*., 2009). The United States and Brazil (Cáceres *et al*., 2009) are the chief buyers of Argentine olive oil. This order is reversed for table olives, with Brazil in first position and the United States in second (Cáceres *et al*., 2009). However, the present delicate situation of the European economy is affecting the profitability of Argentina's olive sector in that, in a bid to ease the crisis, the traditional olive growing countries are competing with Argentina to win over the markets where it currently sells its products (Pallares, 2012). This makes it necessary to find new market niches, not just for olive oil but also for the by-products of olive production. One innovative option among the feasible alternatives might be to recover the antioxidants contained in olive industry waste (Gómez *et al*., 2008), particularly in olive pruning residue, a biomass which is not currently tapped and which looks set to increase over time because pruning is a customary and necessary cultural practice in olive orchards, especially when managed intensively (Iñiguez Monterde *et al*., 1999).

An antioxidant is defined as any substance which retards or prevents the oxidation of an oxidizable substrate (organic or inorganic molecules) when present at low concentrations with respect to the concentrations of the substrate (Venereo Gutiérrez, 2002). Polyphenolic compounds (PCs) are commonly acknowledged to be multifunctional antioxidants. They are a complex class of secondary metabolites biosynthesised throughout the plant kingdom (Wood *et al*., 2001) which act as phytoalexins, protecting the plants from environment or pest-induced stress (Pelayo Villarejo, 2006; Rugna *et al*., 2007). Each stress situation gives rise to a different metabolic behaviour which affects the production and variability of the metabolites (Harbone, 1994). Hence, individuals of the same species growing in different environments undergo variations in the synthesis of their phenolic compounds (Matsuki, 1996). The extent of polyphenol hydroxylation and the position of the hydroxyl groups in the molecule is one of the most important determinants of polyphenol antioxidant activity (Oliveras López, 2005) in that the polyphenols with an orthodihydroxyphenolic structure exhibit greater antioxidant activity (De la Torre Carbot, 2007).

Olive, like other plants, produces more polyphenolic compounds in response to environmental factors (Halls, 2003). In the past, the phenolic compounds in extracts of different parts of the olive plant (leaf, fruit, flower, bark) were used in popular medicine. Nowadays, the high antioxidant activity of olive leaf infusions is documented in scientific literature (Romani, 1999). This characteristic has led to the marketing of olive leaf extracts owing to their spectrum of uses in plant therapy and cosmetics as well as in the pharmacological and food industries.

More is being learned by the day about the advantages of using natural antioxidants for health and industrial purposes and the disadvantages of synthetic antioxidants because of their volatility and potential carcinogenicity (Venereo Gutiérrez, 2002; Dwyer, 1996), thus making it necessary to explore new natural sources of antioxidant-activity metabolites. Olive pruning wood could be a valid option in this respect; however, little literature currently exists on the phenolic composition of the wood of this species. There are only a few papers on the lignans and glucosides isolated from the bark of different species of the *Olea* genus (Chiba, 1979; Tsukamoto, 1985), the determination of chlorogenic acid by thin-layer chromatography (Ozkaya, 1999) and the volatile fraction of wood and the potential of olive pruning wood as a new source of natural antioxidants (Altarejos, 1997; Pérez-Bonilla, 2003).

Local studies conducted by our research team have highlighted the presence of antioxidant polyphenols in wet pomace from the two-phase extraction of olive oil from Arbequina and Coratina olives (Gómez *et al*., 2007) as well as in the leaves of Arbequina (Gómez *et al*., 2008) and Coratina olives (Reales *et al*., 2010) cultivated in the Valle Central area of Catamarca. The purpose of this paper was to research more deeply into the potential of olive pruning wood as a natural source of antioxidants in order to glean fundamental information for initiating possible applications of a fully renewable but as yet underrated by-product of the olive industry.

OBJECTIVES

The objective was to assess the potential of pruning wood from cv. Arbequina olives cultivated in the Valle Central area of Catamarca as a source of natural antioxidants by determining the total polyphenol content, orthodiphenol content, radical scavenging activity and antioxidant activity of extracts of this plant residue in solvents of differing polarity.

METHODOLOGY

Sampling

Olive pruning residue (*Olea europaea* L.), cv. Arbequina, was used for the research. It was obtained from Agrofresco S.A., a company located in Las Esquinas, in the district of Valle Viejo, Catamarca, Argentina. Samples were taken in August 2007 by simple random sampling. The pruning residue of 20 trees chosen at random was collected. The olive branches were taken to the Chemistry Laboratory at the Faculty of Agricultural Science, National Catamarca University (UNCa), where they were cleaned and dried in an oven at 40 ºC for 48 hours, any leaves were removed and the wood was crushed. The samples were stored in clearly identified containers.

Preparation of extracts; polyphenols, orthodiphenols and total solids content

Extracts of olive wood were prepared in quintuplate in a 50% aqueous solution of ethanol (WE) and ethyl acetate (WAc) at a ratio of 1:10 p/v and were macerated at ambient temperature for 24 hours in beakers covered with cling film and tinfoil. The extracts were then vacuum filtered and 50 ml of fresh solvent were added to the remaining residue. The process was repeated, estimating 48 hours' maceration. The extracts obtained from the first and second maceration were combined and the final volume of each extract was divided into two 50-ml aliquots measured in a volumetric flask. One of the aliquots was used to

quantify the total polyphenols (TPPs) and orthodiphenols (ODPs) while the other was used to determine the quantity of total solids (TSs). The samples were stored at 5–10 ºC in amber coloured containers to protect them from the light. The procedure was repeated to obtain the ethyl acetate extracts.

Polyphenol content was evaluated according to the Folin-Ciocalteu method by absorption spectrophotometry at λ = 725 nm and calculated according to equation (1). In addition, orthodiphenol content was measured at λ = 370 nm with 5% sodium molybdate in a 50% aqueous solution of ethanol. Content was calculated according to equation (2). Caffeic acid was used as the standard in both determinations.

$$TPP\ (ppm) = R \times DV \times TV/(AV \times SM) \quad (1)$$

$$ODP\ (ppm) = R \times DV \times TV/(AV \times SM) \quad (2)$$

where R: curve reading, in ppm; DV: dilution volume; TV: total extract volume; AV: aliquot volume; SM: sample mass.

Total solids content was determined by evaporation to dryness at 105 ºC and subsequent weighing to constant weight.

Descriptive data analysis was performed for each determination on the basis of the median values. Non-parametric analysis of variance was used for inferential statistical analysis, calculating the significant differences of the mean ranges according to the Kruskal Wallis test and considering values of $p<0.05$ to be significant. InfoStat version 1.1.2002 was the statistics software used.

Determination of 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging capacity

Extract solutions at concentrations of 100, 300 and 600 ppm were prepared by diluting the WE and WAc extracts.

Using a stock DPPH solution, a 7.4 x 10^{-5} M solution (Pérez Bonilla *et al*., 2003) was prepared in analytical grade methanol. The capacity of the different extract solutions to scavenge the free radical 2,2-diphenyl-1-picrylhydrazyl (DPPH) was evaluated in quintuplate (Brand Williams *et al*., 1995; Gadow *et al*., 1997) using a stoquiometric extract:DPPH ratio of 1.6:1 (Pérez Bonilla *et al*., 2003). The kinetic reaction of the mixture was monitored for 30 minutes by taking spectrophotometric readings at 515 nm and the inhibition rates were calculated according to equation (3):

$$IR = [A_{t= o\ min} - A_{t = 15\ min})/ A_{t= 0\ min}] \times 100. \quad (3)$$

where $A_{t = 0\ min}$: initial DPPH absorbance; $A_{t = 15\ min}$: absorbance 15 minutes after making the extract:DPPH mixture.

Descriptive analysis was performed using the values of the IR medians. Kruskal-Wallis non-parametric analysis of variance was applied for statistical processing and values of $p<0.05$ were considered significant.

Addition of extracts to a lipid substrate

1250 ml of virgin olive oil (S), which had been previously characterised, were placed in three 2000-ml Erlenmeyer flasks. The WE and WAc extracts were then added separately, agitating continuously and vigorously each time, to a 120 ppm concentration of total phenols. The mixtures of control oil plus olive wood ethanol extract (S-WE) and control oil plus olive wood ethyl acetate extract (S-Mac) were left to rest for 48 hours at ambient temperature and protected from light.

Each system (treatment) was divided into five 250-

ml aliquots and transferred to brown cap-less flasks, which had been previously marked. The controls were divided in the same way.

Lipid oxidation was performed for five days in an oven at 80 ºC and monitored by determination of the following: total polyphenols at λ = 725 nm (Folin-Ciocalteu method), peroxide value (PV) (ISO 3960-COI/T.15-IUPAC 2501), ultraviolet absorbance at K_{232} and K_{270} (COI/T.20/Doc. No 19), free acidity (COI/T.15) and acid profile (COI/T.20/Doc. No 24). The determinations were performed before heating the samples and 1 and 5 days after the start of the trial, except for the acid profile which was only determined when the extracts were added to the oil. The experiment was performed in quintuplicate for subsequent statistical processing purposes. Kruskal-Wallis non-parametric analysis of variance was applied, considering values <0.05 to be significant.

RESULTS AND DISCUSSION

Polyphenols, orthodiphenols and total solids content

As can be seen from Graph 1, the total polyphenols (TPP), orthodiphenols (ODP) and total solids (TS) content of the olive wood extracts varied according to the extraction solvents used in the assays. The highest TPP, ODP and TS rates were recorded for the extracts in 50% aqueous ethanol, which is indicative of the polarity of the extracted phenols (Table 1).

When ethyl acetate was used as solvent, the TPP extraction rate was 71.51% lower than with the 50% water:methanol solution. The values recorded were 5395.13 mg and 18938.85 mg phenols/kg wood, respectively (Graph 1 and Table 1). Additionally, the differences in the median TPP content of the extracts treated with 50% ethanol and ethyl acetate were statistically significant (p<0.0079) (Table 1).

The values recorded for the ODPs behaved similarly (Graph 1). The aqueous ethanol extracted 52.64% more ODPs than did the ethyl acetate. Here too, statistically significant differences

Graph 1. Medians of TPP and ODP yields of WE and WAc extracts

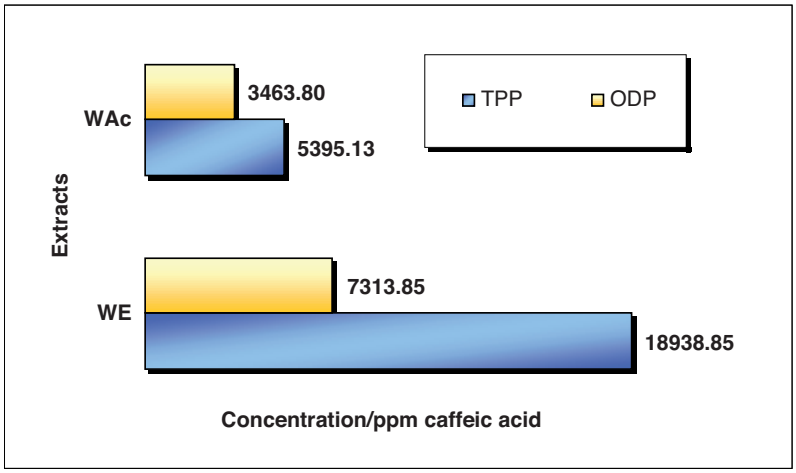


TABLE 1.
Medians of TPP, ODP and TS yields in WE and WAc extracts

Parameters	Medians*		Extraction differences
	WE	WAc	
TPP	18938.85 **A**	5395.13 **B**	71.51
ODP	7313.85 **A**	3463.80 **B**	52.64
TS	17.54 **A**	3.00 **B**	82.9

* Different letters indicate significant differences at 5% (calculated according to the Kruskal-Wallis one-way analysis of variance by ranks)

(p<0.0079) (Table 1) were noted between the medians of the extracts. However, a larger proportion of ODPs was extracted from the wood with ethyl acetate than with aqueous ethanol. The values recorded for TPPs and ODPs show that only 38.62% of the TPPs quantified in the ethanol extract were ODPs whereas this percentage rose to 64.20% (Graphs 2 and 3) in the case of the ethyl acetate extract. This is interesting, bearing in mind that the degree of hydroxylation and the position of the oxyhydryl groups in the molecule are important factors in PC antioxidant activity (Oliveras López, 2005; De la Torre Carbot, 2007).

It was likewise observed that the TS extraction rate with ethyl acetate was 82.90% lower than with 50% aqueous ethanol. Statistical analysis revealed statistical differences (p<0.0079) in TS extraction according to the extraction solvent employed (Table 1 and Graph 4).

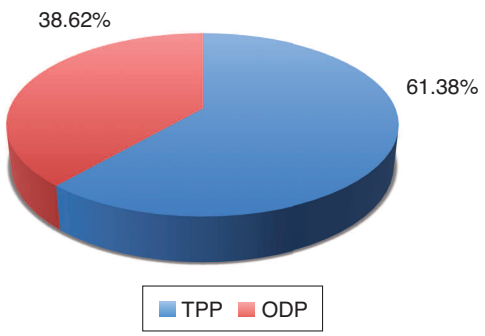
In terms of their chemical composition, the components that can be extracted from pruning wood are largely hydrosoluble phenolics. This was demonstrated by the fact that the 50% ethanol:water mixture achieved more efficient extraction rates and recorded significantly higher TS, TPP and ODP median values. This finding concurs with other research (Pérez Bonilla *et al*., 2003). It should be stressed, however, that ethyl acetate extracted the largest proportion of ODPs from the total extracted phenols.

Importantly, the fact that the aqueous ethanol mixture has the greater TPP, ODP and TS extraction capacity is a plus for the potential applications of the extracts as food preservatives or in the pharmacological and cosmetics industry because ethanol is innocuous while ethyl acetate is toxic.

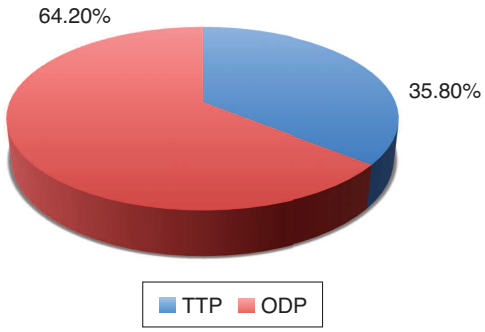
Free radical scavenging activity

Of the six extracts of cv. Arbequina olive wood, the most active ones were WAc 600 ppm and WE 600 ppm, in that order. This coincides with research reporting the excellent free radical scavenging activity of olive wood extracts in the same solvents (Pérez Bonilla *et al*., 2003). Except for treatment WAc 100 ppm, all the wood extracts recorded an IR of more than 50% fifteen minutes after mixing the extract with the DPPH (Table 2); the same value was recorded for butyl-hydroxy-toluene (BHT) at 500 ppm (Rosales Castro M. and Gonzáles Laredo R., 2003).

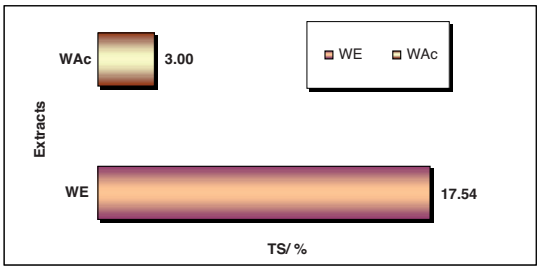
Graph 2. Proportion of ODP in WE extract



Graph 3. Proportion of ODP in WAc extract



Graph 4. Medians of TS yields of WE and WAc olive extracts



At 600 ppm, the ethyl acetate extract displayed the greatest free radical scavenging activity whereas at 300 and 100 ppm the ethanol extracts did so (Table 2).

In addition, as can be seen from Graph 5, the performance of the ethanol-

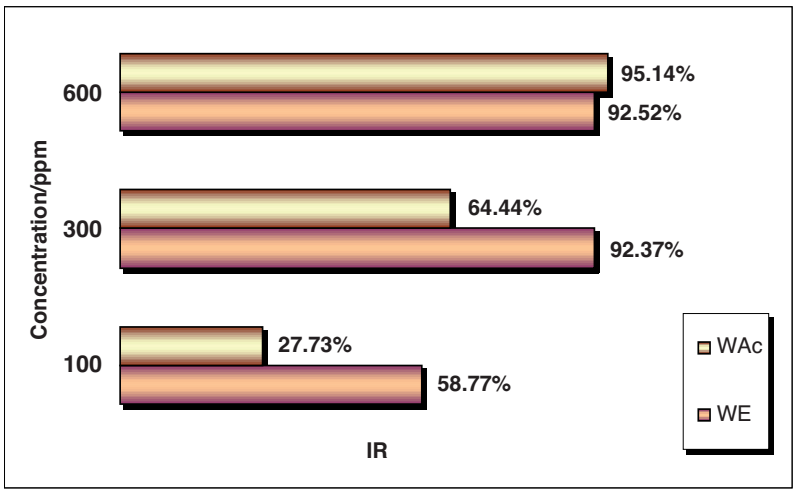
water and ethyl acetate extracts differed to a smaller extent at 600 ppm than at the rest of the concentrations assayed. The activity of the WAc extract was observed to be 2.75% greater than that of the WE extract at 600 ppm. Conversely, at 300 ppm, the free radical scavenging activity of the WE extract was 30.23% higher than that of the WAc. On lowering the concentration of both wood extracts, their inhibitory power decreased sharply; at 100 ppm, WE scavenging power was 52.82% greater than that of the WAc extract (Graph 5).

Graph 6 points to a direct relationship between the concentration of the ethyl acetate extracts and DPPH scavenging activity. Thus, the greatest FR scavenging activity (95.14%) was recorded for the extract at a concentration of 600 ppm. At 300 and 100 ppm, the extracts exhibited a lower antioxidant activity (64.44% and 27.73%) while the IR of the ethanol solutions at 600 and 300 ppm was almost masked by the large quantity of antioxidant PCs in the two extracts. In other words, in the case of this solvent, extract concentration did not have a very pronounced effect on free radical scavenging activity. Only the WE 100 ppm treatment displayed significantly lower activity versus the higher concentrations.

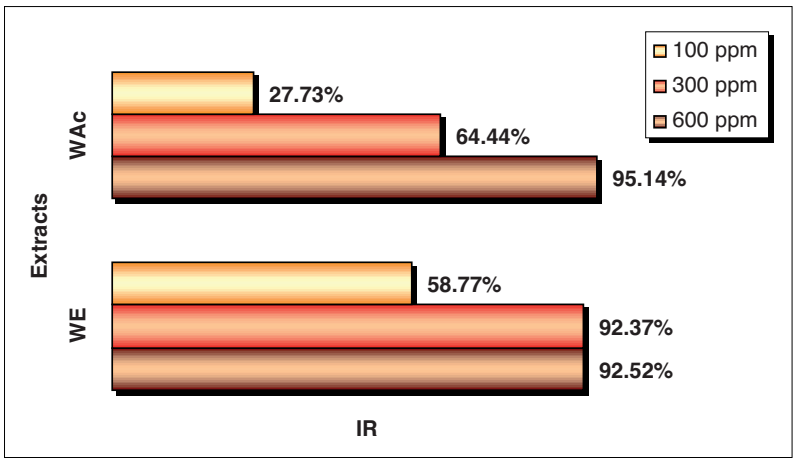
TABLE 2.
IR medians of WE and WAc extracts at 100, 300 and 600 ppm (Extract: DPPH = 1.6:1; 15 min)

Extracts	Extract concentrations	TPP	ODP	% ODP/TPP	IR
WE	600 ppm	11,363.31	4,388.31	38.62	92.52
	300 ppm	5,681.65	2,194.16		92.37
	100 ppm	1,893.88	731.39		58.77
WAc	600 ppm	3,237.08	2,078.28	64.20	95.14
	300 ppm	1,618.54	1,039.14		64.44
	100 ppm	539.51	346.38		27.73

Graph 5. Antioxidant activity of olive wood extracts expressed in terms of IR medians at 100, 300 and 600 ppm (extract:DPPH = 1.6:1; 15 min)



Graph 6. Antioxidant activity of olive wood extracts according to the extraction solvent used, expressed in terms of IR medians at 100, 300 and 600 ppm (extract:DPPH = 1.6:1; 15 min)



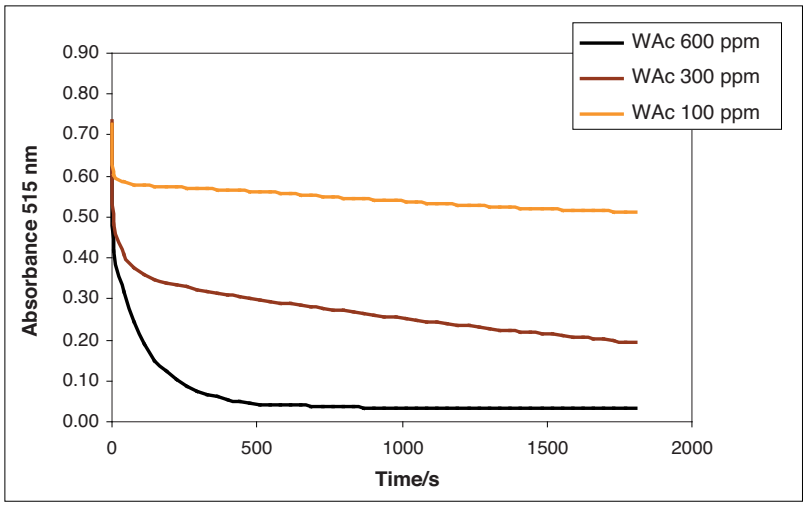
The WAc extract at 600 ppm containing a higher proportion of orthodiphenols recorded the highest IR of the six extracts analysed; however, on performing the Kruskal Wallis assay no significant differences were noted at 5% in relation to the WE treatments at 600 ppm and 300 ppm which had IRs in excess of 90% (Table 3).

TABLE 3.
IR medians of WE extracts at 300 and 600 ppm and of WAc extract at 600 ppm

Extracts	IR medians*
WAc 600 ppm	95.14 **A**
WE 300 ppm	92.37 **A**
WE 600 ppm	92.52 **A**
* Different levels indicate significant differences at 5%.	

A direct relationship was observed between extract concentration and IR in the ethyl acetate extracts, which displayed differing reaction kinetics to the DPPH free radical (Graph 7). At 600 ppm, the absorbance curve of the extract-DPPH mixture decreased to a minimum of 0.032, after which reaction equilibrium started to be established and eventually stabilised at an absorbance of 0.033 with an IR of 95.42%; this activity remained almost constant until the end of the 30-minute reading. At 300 ppm, the absorbance values continued to drop until the end of the 30-minute reading, i.e. WAc at 300 ppm continued to exhibit antioxidant activity without reaching equilibrium. In the case of WAc 100 ppm, the stoquiometric extract-DPPH ratio assayed was observed to be unsuitable for free radical inhibition purposes. The final absorbance readings for this mixture revealed that the curve was reaching equilibrium at scavenging rates of 31.73% and 31.75%. Conversely, the ethanol extracts assayed proved to be extremely active although a pronounced difference was noted in the anti-free radical activity of the most diluted solution of the extract (Graph 8). In the case of treatment WE 600 ppm, the reaction quickly reached equilibrium at an absorbance of 0.055 and an IR of 95.52%; these values remained constant from 1373 seconds until the end of the 30-minute reading. WE 300 ppm also exhibited excellent free radical scavenging activity although the reaction was not as quick as in the case of WE 600 ppm, stabilising at an absorbance of 0.057 and an IR of 92.37%. Lastly, WE 100 ppm was observed to react more slowly than the more concentrated extracts. By 30 minutes, the curve had not stabilised, equilibrium had not been reached and the absorbance continued to de-

Graph 7. Median absorbances, according to time, of the extract:DPPH mixtures (1.6:1) of the ethyl acetate olive wood extracts at 100, 300 and 600 ppm



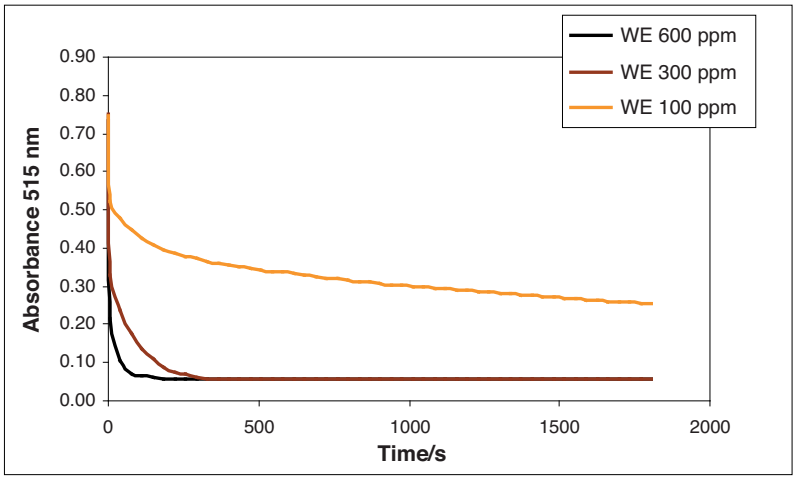
crease; more reaction time was therefore neeeded.

Earlier literature has documented that the bioactivity reported in plant extracts is due not only to mechanisms exerted by their phenolic compounds (flavonoids, tannins, quinones) but also to the synergic effect of any secondary metabolites they contain (alkaloids, terpenes), which are also acknowledged to exert this activity (Murillo *et al*., 2007).

Analysis of the free radical inhibitory power and reaction kinetics of olive wood extracts in 50% ethanol:water and ethyl acetate confirmed that solvent polarity plays an important part in DPPH-scavenging activity. This performance is linked to the nature of the compounds extracted, which could be explained by isolating and identifying the pure compounds present in the extracts and investigating their antioxidant capacity.

This paper shows that, irrespective of the solvent used, high TPP olive wood extracts are excellent free radical scavengers and could be considered a potential source of antioxidants. Nevertheless, all these assumptions need to be confirmed in deeper qualitative and kinetic studies and to be amplified, in particular, by applying such extracts to real biological systems in order to evaluate the effective free radical scavenger activity.

Graph 8. Median absorbances, according to time, of the extract:DPPH mixtures (1.6:1) of the 50% ethanol olive wood extracts at 100, 300 and 600 ppm



Antioxidant activity

Of the 120 ppm of TPPs added, the ethanol treatment only incorporated 1.52% and the ethyl acetate treatment 4.68%. The increase in phenols was lower in treatment S-WE than in S-WAc, which points to a better distribution of the phenolic structures of the WAc extract on the lipid substrate, possibly because the compounds in the ethyl acetate extracts are less polar and therefore more soluble in the oil. One day after being subjected to high temperatures, both S (control virgin olive oil) and the oils containing the extracts recorded an initial decrease in TPPs (Table 4 and Graph 9). On day 0, the phenol increase in the treated oils was not significant vis-à-vis the control phenols. Similarly, significant differences were not recorded in the TPP concentrations of S-WE, S-WAc or S one and five days after beginning the assay (Table 6).

With regard to acid composition, treatments S-WE and S-WAc did not show any changes in acid profile compared with the test oil. Put differently, the addition of the phenolic extracts did not affect the fatty acid composition of the lipid substrate, which retained its properties in this respect (Table 5 and Graph 10).

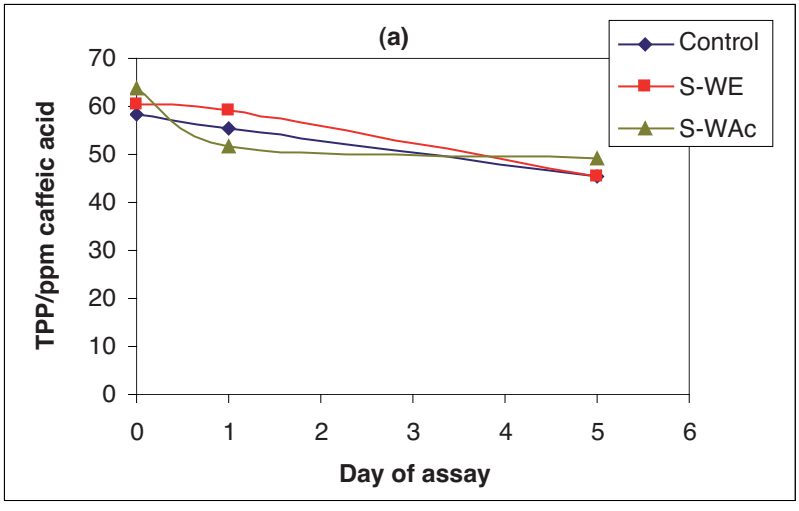
The accelerated consumption of the polyphenols present in the treated oils leads to the assumption that the addition of the extracts helped to protect the oil fat-

TABLE 4.
Medians and percentage changes of the control (S) and treatments (S-WE and S-WAc)

Parameters	Day of assay	S	% change S	S-WE	% change S-WE	S-WAc	% change W-WA
TPP	0	58.17		60.35		63.79	
	1	55.40	*-4.75*	59.12	*-2.03*	51.68	*-18.98*
	5	45.62	*-21.57*	45.39	*-24.79*	49.11	*-23.01*
IR	0	14.47		12.49		12.00	
	1	27.92	*92.95*	27.45	*119.78*	26.94	*124.50*
	5	62.35	*330.89*	73.46	*488.15*	71.92	*499.33*
K_{232}	0	2.38		2.41		2.42	
	1	3.73	*56.72*	3.80	*57.68*	3.77	*55.79*
	5	6.33	*165.97*	8.64	*258.51*	7.88	*225.62*
K_{270}	0	0.23		0.24		0.24	
	1	0.23	*0.00*	0.22	*-8.33*	0.27	*12.50*
	5	0.39	*69.57*	0.53	*130.43*	0.52	*116.67*
% FOA	0	1.07		1.04		1.04	
	1	1.13	*5.61*	1.13	*8.65*	1.10	*5.77*
	5	1.18	*10.28*	1.18	*13.46*	1.18	*13.46*

ty acids from oxidation; however, it was observed that the wood extracts added to the oil only exhibited oxidative protection on day 0, when a significant reduction in IR values was recorded in the treatments compared with S ($p<0.00449$) (Table 6). Before being subjected to the effects of temperature, the treatments lowered IR by 14.19% in S-WE and by 16.27% in S-WAc (Table 4 and Graph 11). On day 1 of the assay, the IR of the treatments remained below that of the control, although not significantly so (Table 6). On day 5, the IR values of the treatments were significantly higher ($p<0.0255$) than S levels whereas significant differences were not observed between the values for this parameter between S-WE and S-WAc (Table 6). This shows that the quantity of antioxidants incorporated via the extracts was not able to diminish the harsh oxidation triggered by the presence of free radicals, the formation of which was catalysed by the high assay temperature. In fact, in conditions of forced oxidation, the extracts even acted like pro-oxidants, raising the concentration of oxidation indicators.

Graph 9. Changes in TPPs in the control oil and treatments S-WE and S-WAc



Between days 0 and 1, the K_{232} absorbance curves of the treated and untreated oils are virtually superimposed in Graph 12. While the protective effect of the extracts was not observed – although this was recorded via the IRs for the same days – the differences between the K_{232} values of the S, S-WE and S-WAc oils were not significant (Graph 13). In the case of K_{270}, the absorbance values are close to each other (Graph 13) and statistical analysis revealed no significant differences between the values recorded for that same day. This shows that supplementation

TABLE 5.
Acid profile of oil (S) and treatments (S-WE and S-WAc)

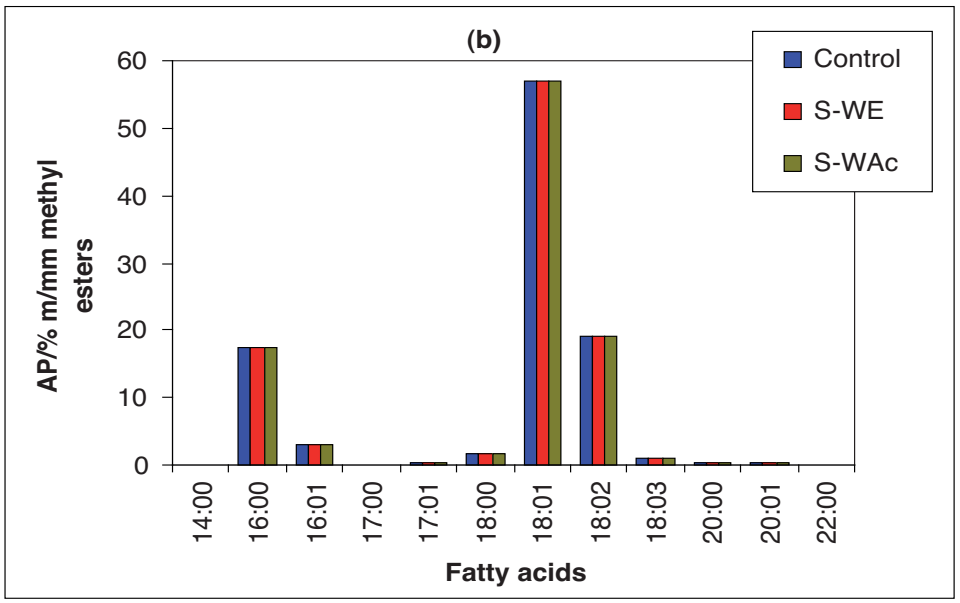
	S	S-WE		S-WAc		
Acids	Mean	SD	Mean	SD	Mean	SD
14:0	0.03	0.00	0.03	0.00	0.03	0.00
16:0	17.33	0.01	17.34	0.02	17.32	0.00
16:1	3.10	0.00	3.12	0.01	3.10	0.00
17:0	0.09	0.00	0.09	0.00	0.09	0.00
17:1	0.21	0.00	0.21	0.01	0.20	0.01
18:0	1.53	0.01	1.54	0.02	1.52	0.00
18:1	57.12	0.00	57.12	0.02	57.10	0.01
18:2	19.07	0.01	19.05	0.01	19.07	0.01
18:3	0.94	0.00	0.94	0.01	0.94	0.00
20:0	0.40	0.00	0.40	0.00	0.40	0.00
20:1	0.29	0.01	0.28	0.00	0.28	0.00
22:0	0.02	0.00	0.02	0.00	0.02	0.00

TABLE 6.
Median values of TTP, IR, K_{232}, K_{270} and % FOA of S, S-WE and S-WAc

Day of assay	Treatments	Medians *				
		TPP	IR	K_{232}	K_{270}	% FOA
0	*S*	58.17 **A**	14.47 **B**	2.38 **A**	0.23 **A**	1.07 **B**
	S-WE	60.35 **A**	12.49 **A**	2.41 **A**	0.24 **A**	1.04 **AB**
	S-WAc	63.79 **A**	12.00 **A**	2.42 **A**	0.24 **A**	1.04 **A**
1	*S*	55.40 **A**	27.92 **A**	3.73 **A**	0.23 **AB**	1.13 **A**
	S-WE	59.12 **A**	27.45 **A**	3.80 **A**	0.22 **A**	1.13 **A**
	S-WAc	51.68 **A**	26.94 **A**	3.77 **A**	0.27 **B**	1.10 **A**
5	*S*	45.62 **A**	62.35 **A**	6.33 **A**	0.39 **A**	1.18 **A**
	S-WE	45.39 **A**	73.46 **B**	8.64 **B**	0.53 **B**	1.18 **A**
	S-WAc	49.11 **A**	71.92 **B**	7.88 **AB**	0.52 **AB**	1.18 **A**

* Different letters within the same parameter and day indicate significant differences at 5%.

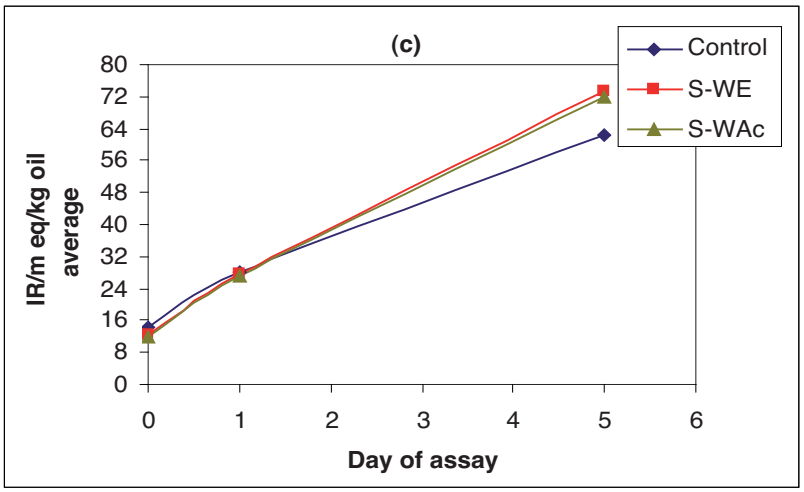
Graph 10. Acid profile of the control oil and treatments S-WE and S-WAc



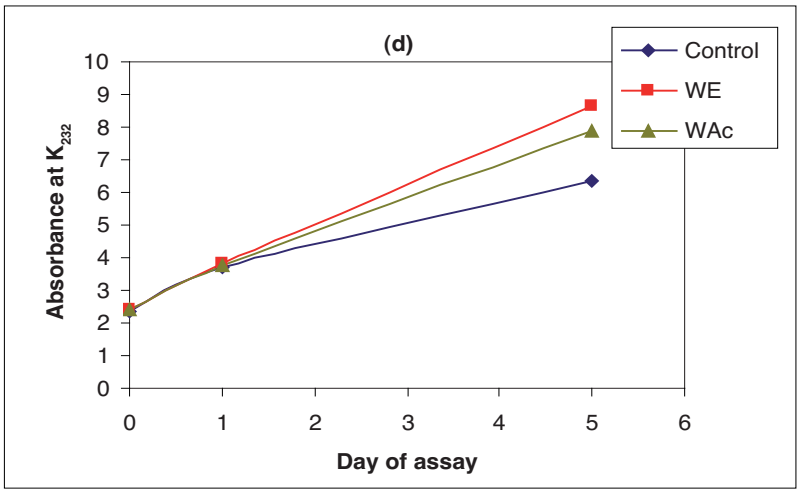
of the substrate with the extracts did not add primary or secondary oxidation products. After heating for 1 day, the K_{270} value of the control oil did not differ significantly from that of the treatments. After 5 days of the trial, the K_{270} value of S was significantly lower ($p<0.0435$) than that of S-WE (Table 6) and the treatments did not provide protection from primary or secondary oxidation processes. In all probability, the smaller increase in K_{232} and K_{270} in S compared with the values for the treatments is due to the fact that the addition of the extracts incorporated compounds which, when subjected to the high temperature conditions of the test, gave rise to primary and secondary oxidation products which increased the pertinent indices.

Statistically speaking, the percentage of free oleic acid (FOA) in S-WE did not differ significantly from that of S-WAc or of the control oil, whereas the percentage of free oleic acid in S-WAc was significantly lower ($p<0.0291$) than that of S (Table 6). From day 1 until the end of the assay, the treatments evolved similarly to S and there were no significant differences (Tables 4 and 6 and Graph 14).

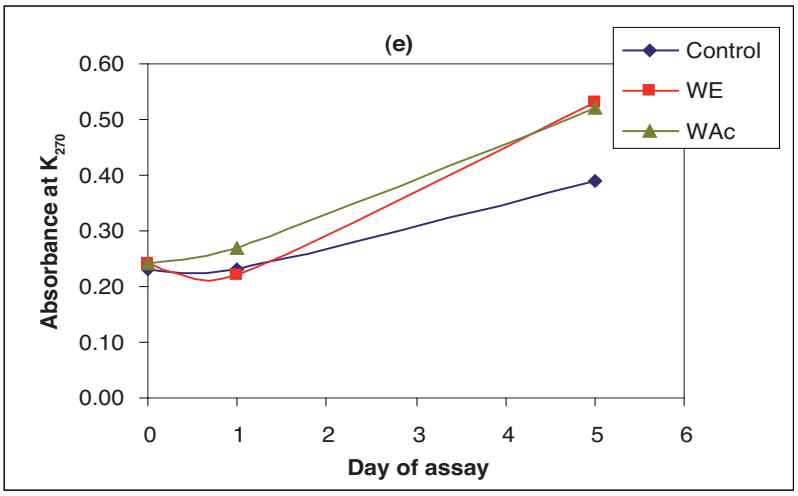
Graph 11. Changes in the IR of the control oil and treatments S-WE and S-WAc



Graph 12. Changes in K_{232} of the control oil and treatments S-WE and S-WAc



Graph 13. Changes in K_{270} of the control oil and treatments S-WE and S-WAc



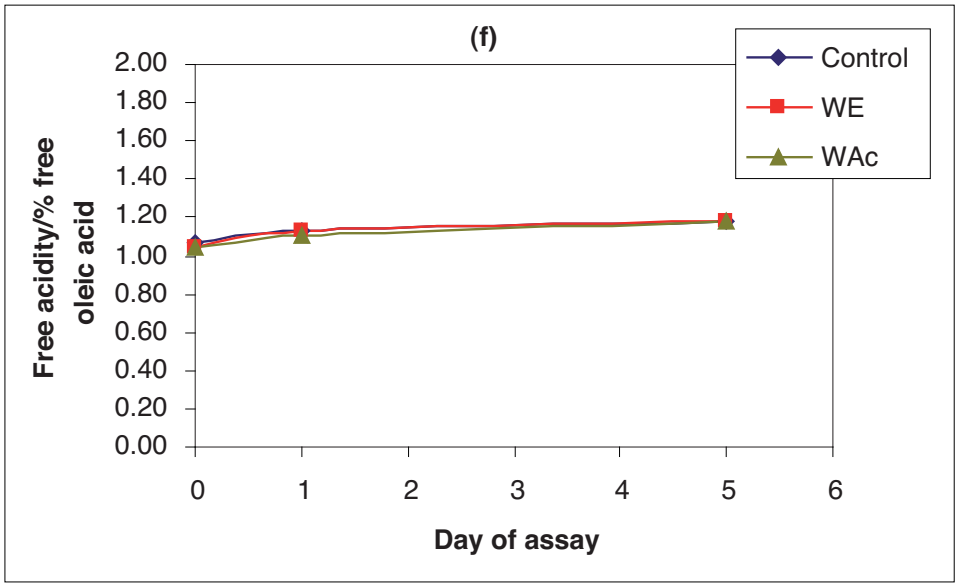
CONCLUSIONS

Olive wood extracts in ethanol-water and ethyl acetate have a high DPPH-scavenging capacity. When added to virgin olive oil at ambient temperature, they exert a mild protective effect; however, they have no antioxidant activity when the lipid substrate is subjected to forced, extreme oxidation and even display a pro-oxidant behaviour. When added, they do not alter the percentage of free oleic acid or the fatty acid composition of the oil used as a substrate.

It can therefore be concluded that ethanol and ethyl acetate extracts of cv. Arbequina olive pruning wood from the Valle Central area of Catamarca in Argentina are a very valuable raw material for the extraction of natural polyphenols. They are at an advantage compared with alternative extraction sources because of the high polyphenol yields and important antioxidant properties.

The results obtained in this paper give an insight into the possibility of considering olive pruning wood from the Valle Central area of Catamarca as a by-product of the olive industry and a raw material for the isolation of phenolic extracts and pure antioxidant compounds instead of as a waste prod-

Graph 14. Changes in % FOA of the control oil and treatments S-WE and S-WAc



uct. Such a strategy would help not only to prevent environmental problems but also to generate returns from a product which is completely wasted at present, thus resulting in sustainable agricultural practices and the possibility of generating competitive industries, with all the ensuing social benefits. ■

REFERENCES

Altarejos J., Nogueras M., Sánchez A. (1997). VIII Simposium Científico-Técnico de Expoliva, Jaén.

Andrada C., Luna M., Gómez P. (2008). "La reconversión productiva de la olivicultura en Catamarca". La Alimentación Latinoamericana. Nº 276. Pp. 54-60.

Brand-Williams W., Cuvelier M., Berset C. (1995). "Use of a free radical method to evaluate antioxidant activity". Lebensmittel-Wissenschaft und-Technologie /Food Science and Technology. Vol. 28. Pp 25-30.

Cáceres R., Novello R., Robert M. (2009). "Estudios Socioeconómicos de los Sistemas Agroalimentarios y Agroindustriales". S ISSN 1852-4605. Nº 2. Pp 11-41.

De la Torre Carbot K. Efecto del consumo del aceite de oliva sobre la composición de las lipoproteínas de baja densidad en individuos de diferentes países europeos. Tesis de Doctorado. Universidad de Barcelona. 21-40, 2007.

Dwyer J. (1996). "Is there a need to change the American Diet?: Dietary phytochemicals in cancer prevention and treatment". Adv. Experimen. Med. Biol. N° 401. Pp. 192-193.

Gómez E., Salim Rosales C., Gómez P., Bravo M. (2008). "Métodos de extracción de polifenoles en hojas y residuos leñosos de Olivos". IV Congreso Iberoamericano de Ambiente y Calidad de Vida. Ed. Universitaria de resúmenes- Línea científica. Catamarca. Argentina. Pp. 296.

Gómez P., Dalla Lasta M., Porcú E., Bravo M., Nieto S., Kaen R. (2007) "Evaluación de la actividad antioxidante de extracto fenólico de alperujo". Aceites y Grasas N° 67. Tomo XVII. Vol.2. Buenos Aires. Argentina. Pp. 336-341.

Gómez P., Dalla Lasta M., Bravo M., Rosales Salim C., Gómez E., Castillo D. (2008) "Efecto de extractos de alperujo de Arbequina y Coratina sobre la estabilidad de aceite de oliva virgen". IV Congreso Iberoamericano de Ambiente y Calidad de Vida. Ed. Universitaria de resúmenes- Línea científica. Catamarca. Argentina. Pp. 171.

Halls S. (2003). "In vinus vitali? Compounds activate life-extending genes". Science. Nº 301. Pp.165.

Harbone, J. (1994). "Introduction to Ecological Biochemistry". 4ta. Edición. Academic Press Ed., London. Pp. 384.

Iñiguez Monterde, A; Sánchez Riquelme, L; Sierra Carrascosa, M. (1999). "Poda e Injertos del Olivo". Cuadernos de tecnología agraria. Serie agricultura. Nº 2. Pp. 5-6.

Matías, C.; Cáceres, R. (2012). "Olivo: el cultivo con mayor crecimiento". INTA Informa. Año XI N° 123. Marzo 2012. Disponible en:
http://intainforma.inta.gov.ar/wp-content/uploads/2012/02/Inta_Informa_123.pdf

Matsuki M. (1996). "Aust" J. Bot. Nº 44. Pp. 613-634.

Mosquera O., Niño J., Correa Y., Buitrago Bejarano D. (2005). "Estandarización del método de captura de radicales libres para la evaluación de la actividad antioxidante de extractos vegetales". Scientia et Técnica. Año XI. Nº 27. Pp. 231-234.

Murillo E., Lombo O., Tique M., Méndez J. (2007). "Potencial Antioxidante de Bauhinia Kalbreyeri Harms (FABACEAE)". Información Tecnológica Nº 6. Vol 18. Pp 65-74.

Oliveras López J. (2005). "Calidad del aceite de oliva virgen extra. Antioxidantes y función biológica". Tesis de Doctorado. Universidad de Granada. 25-56.

Ozkaya M., Celik M. (1999). Acta Hortic. Nº 474. Pp. 477.

Pallares J. H. (2012). "La crisis golpea al sector olivícola". LA NACION. 2012 - lanacion.com. Disponible en:
http://www.lanacion.com.ar/1442206-la-crisis-golpea-al-sector-olivicola.

Pelayo Villarejo E. (2006). "Vigilancia Tecnológica e Inteligencia Económica para la industria de zumos. Aplicación práctica para el caso de zumos funcionales y nutracéuticos obtenidos de olivo, romero y alcachofa". Trabajo de doctorado. Pp 12-13 y 51-53.

Pérez-Bonilla M., S. Salido, P. Linares, T. van Beek, J. Altarejos, M. Nogueras, A. Sánchez. La madera de olivo como nueva fuente de antioxidantes naturales. Foro de la tecnología oleícola y la calidad. Código TEC-26. 2003. Disponible en:
<http://www.expoliva.com/expoliva2003/simposium/com.asp?anio=2003&codigo=TEC>.

Reales N., Gómez P., Salim Rosales C., Bravo M., Gómez E. "Actividad antioxidante de extractos de hoja de olivo, cultivar Coratina, del Valle Central de Catamarca". Revista del CIZAS. Vol 11. N°1. Año 2010. Pp. 27-39.

Romani A., Mulinacci N., Pinelli P., Cimato A. (1999). "Polyphenolic Content in Five Tuscany Cultivars of Olea Europaea L". J Agric. Food Chem. Vol. 47. Pp. 964-967.

Rosales Castro M. y Gonzáles Laredo R. (2003). "Comparación del contenido de compuestos fenólicos en la corteza de ocho especies de pino". Maderas y Bosques. Nº 2. Vol. 9. Pp. 41-49.

Rugna A., Riccio R., Gurni A. y Wagner M. (2007). "Efectos de la radiación solar sobre la producción de polifenoles en ejemplares femeninos de Smilax Campestris Griseb. –Smilacaceae-." Comunicaciones breves. Lat. Am. J. Pharm. Nº 3. Vol. 26. Pp. 420-423.

Searles P., Aguero Alcarás M., Rousseaux M. (2011). "El consumo del agua por el cultivo de olivo (*Olea europaea* L.) en el noroeste de Argentina: una comparación con la Cuenca Mediterránea". Ecología Austral Nº 21. Pp15-28.

Tsukamoto H., Hisada S., Nishibe S. (1985). Shoyakugaku Zasshi. Nº 39. Pp. 90.

Venereo Gutiérrez J. (2002). "Daño Oxidativo, Radicales Libres Y Antioxidantes". Rev. Cuban Medic. Militar. N° 2. Vol. 31. Pp. 126-133.

Venereo Gutiérrez J. (2002). "Daño oxidativo, radicales libres y antioxidantes." Rev. Cubana Med. Milit. 2002; 31(2). Pp. 126-133.

von Gadow A., Joubert E., Hansmann C. (1997). "Comparison of the antioxidant activity of aspalathin with that of other plant phenols of rooibos tea (*Aspalathus linearis*), α-tocopherol, BHT and BHA". J. Agric. Food Chem. Nº 45. Vol. 3. Pp. 632-638.

Waterman P y S. Mole. Methods in ecology. (1994). "Analysis of phenolic plant metabolites". Blackwell Scientific publications. Pp. 237.

Wood J., Senthilmohan S. y Peskin A. (2001). "Antioxidant activity of procyanidin-containing plant extracts at different pHs". Food Chemistry Nº 77. Pp. 155-161.

Ryan D., Robards K., Prenzler P., Antolovich M. (1999). "Applications of mass spectrometry to plant phenols. Trac-Trends in Analytical Chemistry". Cap.18. Pp. 362-372.

Influence of technological factors on virgin olive oil*

M. Servili

Dipartimento di Scienze Economico-Estimative e degli Alimenti, Facoltà di Agraria, Università di Perugia, Via San Costanzo, 06121 Perugia (Italy). servimau@unipg.it

According to European legislation (EC 61/2011) and the International Olive Council (IOC, 2010), the marketable quality of virgin olive oil (VOO) is determined by parameters such as free acidity and oxidative status (peroxide value, K_{232}, K_{270} and ΔK), which identify product deterioration, while other analytical markers such as waxes, sterols, aliphatic and triterpenic alcohols, trans-isomers of fatty acids, fatty acid and triacylglycerol composition and stigmastadienes are taken into account to prevent oil adulteration and fraud.

Sensory analysis was added to the analytical methods to control the occurrence of off-flavours not permitted in extra virgin olive oil (EVOO) under EU regulations. Off-flavours had in fact been well defined by the International Olive Council (IOC, 1987), which also standardized the procedure for their determination on the basis of values assigned through sensory analysis (EU Regulation 1989/03).

The marketable parameters do not take into account the analytical markers that certify the health-related and sensory properties of EVOO even though these represent an important fraction of the exclusive composition of EVOO that makes it so unique compared with all the other common vegetable oils consumed in the world. Examples of such markers are natural antioxidant compounds, monounsaturated oleic acid and squalene. Moreover, these markers are not declared on current EVOO labelling; consumers are not therefore informed about the health properties of the product, which are mainly attributable to its high content of monounsaturated oleic acid, squalene and natural antioxidants as well as of phenolic compounds, tocopherols and carotenoids (López-Miranda *et al*., 2010; Bach-Faig *et al*., 2011; Cicerale *et al*., 2011). The sensory properties (mainly aroma) of EVOO are the result of a complex mixture of volatile compounds, C_5 and C_6 saturated and unsaturated aldehydes, alcohols and esters which are responsible for typical flavours such as 'cut grass', 'hay' and ''floral'', as well as of hydrophilic phenols which cause bitter and pungent notes (Angerosa *et al*., 2004; Servili *et al*., 2004; 2009a). Furthermore, these compounds show great antioxidant activity and play an important role in the prevention and/or reduction of chronic degenerative events based on inflammatory processes as well as of chronic–degenerative diseases such as cardiovascular–cerebral diseases (EFSA, NDA, 2011) and cancer (Servili *et al*., 2009b; Obied *et al*., 2012).

The nutritional importance of EVOO has always been linked to its high content of monounsaturated fatty acids (MUFAs), particularly oleic acid. In the last decade, however, significant variability has been noted in the oleic acid content of EVOO, traditionally fixed within a range of 55%–83% of total fatty acid content. This strong variability is closely related to the expansion of olive cultivation to several new growing areas where the EVOOs produced have a low oleic acid content below 50%, Clearly, this has

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an impact on the health and nutritional properties of EVOO (Terés *et al*., 2008; López-Huertas, 2010). The same remarks apply to the tocopherols and hydrophilic phenols in EVOO (Servili, 2012a).

EVOO polyphenols constitute a group of secondary plant metabolites not often found in other oils and fats. This is the most important class of phenols and includes phenolic alcohols and acids, flavonoids, lignans and secoiridoids (Servili *et al.,* 2004; Obied *et al*., 2008), which are found exclusively in plants belonging to the *Oleaceae* family (of which the olive is the only edible fruit) and are the most important fraction from the biological point of view. In particular, the main secoiridoids are the dialdehydic form of decarboxymethyl elenolic acid linked to 3,4-DHPEA or p-HPEA (3,4-DHPEA-EDA or p-HPEA-EDA), the 3,4-DHPEA-EA isomer of oleuropein aglycone and the ligstroside aglycone (p-HPEA-EA) (De Marco *et al*., 2007; Obied *et al*., 2007; 2008; Servili *et al*., 1999; 2004; 2009b).

Several agronomic factors such as cultivar, ripening stage, geographical and genetic origin of the olives, olive tree irrigation and technological factors such as oil extraction conditions during crushing, malaxation and EVOO separation (Angerosa *et al*., 2004; Servili *et al*., 2004; 2009a; Inglese *et al*., 2011) have a strong influence on the qualitative and quantitative composition of the volatile and phenolic fractions. In fact, during crushing, some endogenous enzymes (polyphenoloxidase (PPO), peroxidase (POD) and lipoxygenase (LOX)), are distributed in different forms in the constituent parts of the drupe; POD in particular is largely contained in the kernel (Servili *et al*., 1999), while PPO is mainly found in the mesocarp. These endogenous enzymes play an important role in determining the amount of phenolic and volatile compounds in EVOO.

Endogenous PPO, POD and LOX activity is regulated by malaxation conditions (temperature and oxygen concentration), which also heavily affect the end concentration of hydrophilic phenols and volatile compounds in EVOO. The phenol concentration of the olive paste and oil is decreased by PPO and POD activity, which catalyses phenol oxidation, while the C_5 and C_6 saturated and unsaturated aldehydes, alcohols and esters correlated with the 'green' sensory notes in EVOO are produced by LOX through a cascade pathway (Angerosa *et al*., 2004; Servili *et al.,* 2007a).

The phenols and volatile compounds in EVOO are heavily affected by the crushing system. According to Servili *et al*., (1999), the phenols are concentrated mostly in the pulp and are only found in small amounts in the olive stone and seed. The hydrophilic phenol content of EVOO can be increased by using a type of hammer that has a differentiated effect on the component parts of the drupes, for instance a blade crusher, toothed crusher, pre-crusher or stone crusher, which decreases the release of POD in the paste, so attenuating seed tissue degradation (Servili *et al.,* 2007a). This is further confirmed by the fact that the phenol concentration of EVOO is higher when the stones have been removed from the olives prior to mechanical extraction of EVOO (Angerosa *et al*., 1999; Lavelli and Bondesan, 2005; Mulinacci *et al*., 2005; Amirante *et al*., 2006; Servili *et al*., 2007a). The crushing system also has a strong impact on EVOO volatile compound concentration. For instance, the use of a hammer crusher, which roughly grinds the pulp tissue, leads to an increase in olive paste temperature and a concomitant decrease in HPL activity (Servili *et al*., 2002; Angerosa *et al*., 2004).

Some authors have reported on the relationships between malaxation condi-

tions (time, temperature and low oxygen concentration in the malaxator head-space) and EVOO volatile and phenolic concentration, which is determined by monitoring the amount of endogenous oxidoreductases, such as PPO, POD and LOX. During malaxation, the decrease in O_2 values (observed in covered malaxators) inhibits PPO and POD activity and raises the concentration of hydrophilic phenols in the olive paste and in the resultant EVOO (Servili *et al*., 2008a; 2008b; Taticchi *et al*., 2013). Furthermore, the natural production of CO_2 caused by olive cell metabolism during malaxation reduces oxidative activity in the paste during this phase (Parenti *et al*., 2006 a; 2006b; Servili *et al*., 2008a). The influence of malaxation temperature on phenolic concentration has recently been studied (Boselli *et al*., 2009; Gómez-Rico *et al*., 2009). When oxygen concentration is low in malaxated pastes, phenol oxidative degradation due to PPO and POD activity is inhibited while the phenolic solubility of EVOO is enhanced by a temperature increase (Taticchi *et al*., 2013). These results show that temperatures above 30 °C partially inactivate PPO. On the other hand, these temperature values could increase the activity of depolymerizing enzymes which promote the release of hydrophilic phenols in the oil and vegetation water by hydrolyzing the olive cell wall (Vierhuis *et al*., 2001; Servili *et al*., 2008a; 2008b). Moreover, it has been observed that the enzymes involved in the LOX pathway are active during malaxation. For this reason, cultivars and malaxation temperature affect the volatile profile and, therefore, the sensory characteristics of the resulting EVOOs (Angerosa *et al*., 2004; Servili *et al*., 2009a). In fact, during malaxation, temperatures above 35 °C decrease the quantity of volatile compounds in EVOOs. In particular, the concentration of aldehydes seems to be affected by the processing temperature: the smallest amounts are observed at 35 °C while the highest concentration occurs at 25 °C. Esters also behave in the same way as the aldehydes whereas alcohol concentration increases with malaxation temperature. Thus, malaxation temperature has to be fixed at approximately 25 °C. However, several studies conducted some years ago on different cultivars revealed that the decrease in aroma production by the LOX pathway (due to high temperatures) is cultivar-dependent. This aspect opens up a new line of research to optimise malaxation operating conditions to allow for cultivar-mediated variability. Preliminary studies have been performed on some Italian cultivars to define the best malaxation conditions in terms of temperature and O_2 concentration. They reveal that the best working temperatures are in the range of 20–33 °C while the oxygen concentration should range between 50 and 30 KPa (Servili *et al*., 2012a).

The amount of phenolic fractions in EVOO is also influenced by the extraction system used, for instance pressing or centrifugation. In traditional centrifugation systems, a large amount of water was added to reduce the viscosity of the paste and to improve oil yield. However, this also lowered the phenolic concentration of EVOO and altered its sensory and nutritional characteristics. In the last twenty years, this extraction system has been adapted to reduce the amount of water added during oil extraction. Centrifuges can be classified in three groups on the basis of this aspect: (a) traditional three-phase centrifuges characterized by the addition of 0.5–1 m^3 of water per ton; (b) new three-phase centrifuges featuring the addition of 0.2–1 m^3 of water per ton at the most; (c) two-phase centrifuges that can operate without the addition of water and which do not produce vegetation water as a by-product of the oil extraction process. The new centrifugation systems produce oils with a higher content of phenolic compounds

than do traditional systems because they reduce hydrophilic phenol loss in the vegetation water. Consequently, temperature monitoring during malaxation and the reduction of the amount of water added before centrifugation are critical points in oil extraction technology which impact strongly on EVOO quality.

TECHNOLOGICAL STRATEGIES FOR THE REUSE OF VIRGIN OLIVE OIL BY-PRODUCTS

In the last two decades the new approach to EVOO extraction is now moving towards the reuse of by-products such as olive pomace and olive vegetation water (OVW), both considered in the past to be waste whose disposal entailed additional cost. This new approach should also be oriented at enhancing by-products in order to improve process profitability. In particular, the innovative reuse of EVOO by-products is important because of the hydrophilic phenols they contain, the amount of which is greatly affected by the agronomic and technological conditions of EVOO production. In point of fact, after crushing and malaxation only a small proportion of the phenols is released in the EVOO – between 1% and 3% of the total phenolic concentration of the olives – while a larger amount is found in the pomace and OVW (Servili *et al*., 1999; 2004; 2007a; 2007b; 2011a). In Italy, three-phase centrifugation is the most common extraction system. It requires dilution of the malaxated paste with water and produces 50–90 L of OVW/100 kg of olive paste and 50–60 kg of olive pomace/100 kg of olive paste. At present, the two-phase system is largely used in Spain and is characterised by a sharp reduction in water consumption during the extraction process: it produces 70 kg of olive pomace/100 kg of olive paste.

The residual oil contained in the pomace is recovered by extraction with organic solvents. New opportunities for pomace reuse include compost production, as fuel for producing thermal energy from a renewable source and as an additive in animal feed (Pauselli *et al*., 2007; Servili *et al*., 2007a).

In the case of OVW, the large amounts of bioactive phenols contained in this by-product can be recovered. OVW is made up of an emulsion of water, oil, mucilage and pectins and contains 3–16% organic substances, 1–8% sugars, 1.2–2.4% nitrogen compounds and 0.34–1.13% phenolic compounds (Naionakis and Halvadakis, 2004). Secoiridoids such as 3,4-DHPEA-EDA and verbascoside are the most abundant phenolic compounds in OVW (Servili *et al*., 2004). The pollution potential of OVW is strictly dependent on its polyphenol content. It is expressed as biochemical oxygen demand (BOD_5) and ranges from 35 to 110 g/L, while the chemical oxygen demand (COD) ranges from 40 to 196 g/L (Niaonakis and Halvadakis, 2004). The recovery of large amounts of OVW phenols is therefore an innovative process for the reuse of a product whose disposal represents a cost to olive oil plants (Roig *et al*., 2006). Various approaches have already been implemented (Turano *et al*., 2002; Kujawski *et al*., 2004; Roig *et al*., 2006; Agalias *et al*., 2007; Paraskeva *et al*., 2007; Russo, 2007; Khoufi *et al*., 2008; Gortzi *et al*., 2008), although there are constraints on their application on a plant scale because of the complexity of OVW pre-treatment and the high costs of treatment and plant installation. A membrane filtration system has recently been applied on an industrial scale to obtain a crude phenolic concentrate (CPC) from OVW after pre-treatment with a depolymerising enzymatic pool (Servili *et al*., 2011a). On applying this process it is possible to obtain an OVW volume ranging from 75 to 80% and a large reduction in the original OVW pollution load

(more than 95%). In particular, the polyphenol concentration of the resultant CPC is four times higher than that of the initial OVW. The polyphenols present in the largest quantities are 3,4-DHPEA-EDA and verbascoside, although the content of the former is heavily affected by prolonged OVW storage due to OVW hydrolysis (Servili *et al*., 2011a). ■

REFERENCES

Agalias A., Magiatis P., Skaltsounis A., Mikros E., Tsarbopoulos A., Gikas E.,. et al., 2007. A new process for the management of olive oil mill waste water and recovery of natural antioxidants. *Journal of Agricultural and Food Chemistry*, 55, p. 2671–2676.

Amirante P., Clodoveo M., Dugo L., Leone G., Tamborrino A., 2006. Advance technology in virgin olive oil production from traditional and de-stoned pastes: Influence of the introduction of a heat exchanger on oil quality. *Food Chemistry*, 98, p. 797-805.

Angerosa F., Servili M., Selvaggini R., Taticchi A., Esposto S., & Montedoro G F., 2004. Volatile compounds in virgin olive oil: Occurrence and their relationship with the quality. *Journal of Chromatography A*, 1054, p. 17–31.

Angerosa F., Basti C., Vito R., Lanza B. 1999. Effect of fruit stone removal on the production of virgin olive oil volatile compounds. *Food Chemistry*, 67, p. 295-299.

Bach-Faig A., Berry EM., Lairon D., Reguant J., Trichopoulou A., Dernini S., … & Serra-Majem L., 2011. Mediterranean diet pyramid today. Science and cultural updates. *Public Health Nutrition*. 14, p. 2274-2284.

Boselli E., Di Lecce G., Strabbioli R., Pieralisi G. & Frega N.G., 2009. Are virgin olive oils obtained below 27 °C better than those produced at higher temperatures? LWT - *Food Science and Technology*, 42, 3, p. 748-757.

Cicerale S., Lucas LJ, and Keast RSJ., 2011. Antimicrobial, antioxidant and anti-inflammatory phenolic activities in extra virgin olive oil. *Current opinion in biotechnology*, 23, (2), p. 129-135.

De Marco E., Savarese M., Paduano A., Sacchi, R., 2007. Characterization and fractionation of phenolic compounds extracted from olive oil mill waste waters. *Food Chemistry*, 104, p. 858-867.

E.U., 2003 November 6, Regulation 1989/03 amending Regulation (EEC) No 2568/91 on the characteristics of olive oil and olive-pomace oil and on the relevant methods of analysis. Official Journal of the European Communities L. 295/57 13/11/2003.

E.U., 2011. Regulation No. 61/2011 of 24 January 2011 amending Regulation (EEC) No. 2568/91 on the characteristics of olive oil and olive-residue oil and on the relevant methods of analysis. Official Journal of the European Union, L23, 1–14.

EFSA Panel on Dietetic Products, Nutrition and Allergies (NDA). (2011). Scientific Opinion on the substantiation of health claims related to polyphenols in olive and protection of LDL particles from oxidative damage (ID 1333, 1638, 1639, 1696, 2865) pursuant to Article 13(1) of Regulation (EC) No 1924/2006. EFSA Journal. 9, 4, 2033, p. 1-25.

Gómez-Rico A., Inarejos-García A. M., Salvador D.M., & Fregapane G., 2009. Effect of malaxation conditions on phenol and volatile profiles in olive paste and the corresponding virgin olive oils (Olea europaea L. Cv. Cornicabra). Journal of Agricultural and Food Chemistry, 57(9), p. 3587–3595.

Gortzi O., Lalas S., Chatzilazarou A., Katsoyannos E., Papaconstandinou S., Dour-

toglou E., 2008. Recovery of natural antioxidants from olive mill wastewater using Genapol-X080. *Journal of the American Oil Chemists' Society*, 85, p. 133–140.

Inglese P., Famiani F., Galvano F., Servili M., Esposto S., Urbani S., 2011. Factors affecting extra-virgin olive oil composition. *In Horticultural Reviews*, Jules Janik Ed., John Wiley & Sons Pubs., 38, p. 83-148.

International Olive Council, IOC;1987. Sensory analysis of olive oil – Method – Organoleptic assessment of virgin olive oil. COI/T.20/Doc. No. 3. Madrid, June 18th.

International Olive Council, IOC, 2010. Determination of the content of waxes, fatty acid methyl esters and fatty acid ethyl esters by capillary gas chromatography. COI/T. 20/NC No 28/Rev. 1, 1–17.

Khoufi S., Aloui F., Sayadi S., 2008. Extraction of antioxidants from olive mill wastewater and electro-coagulation of exhausted fraction to reduce its toxicity on anaerobic digestion. *Journal of Hazardous Materials*, 151, p. 531–539.

Kujawski W., Warszawski A., Ratajczak W., Porębski T., Capała W., Ostrowska I., 2004. Removal of phenol from wastewater by different separation techniques. *Desalination*, 163, p. 287–296.

Lopez-Huertas E., 2010. Health effects of oleic acid and long chain omega-3 fatty acids (EPA and DHA) enriched milks. A review of intervention studies. *Pharmacological Research*, 61, p. 200–207.

López-Miranda J., Pérez-Jiménez F., Ros E., et al., 2010. Olive oil and health: Summary of the II international conference on olive oil and health consensus report, Jaén and Córdoba (Spain) 2008. In Nutrition, *Metabolism & Cardiovascular Diseases*, 20, p. 284-294.

Lavelli V., and Bondesan L., 2005. Secoiridoids, tocopherols, and antioxidant activity of monovarietal extra virgin olive oils extracted from destoned fruits. *J. Agric. Food Chem.*, 53, p. 1102-1107.

Mulinacci N., Giaccherini C., Innocenti M., Romani A., Vincieri F.F., Marotta F., Mattei A., 2005. Analysis of extra virgin olive oils from stoned olives. *J. Sci. Food Agric.* 85,p. 662- 670.

Niaounakis M., Halvadakis C.P., 2004. Olive-mill waste management. Literature review and patent survey. Atene: Typothito-George Dardanos.

Obied H.K., Bedgood Jr. D.R., Prenzler P.D., Robards K., 2007. Bioscreening of Australian olive mill waste extracts: Biophenol content, antioxidant, antimicrobial and molluscicidal activities. *Food and Chemical Toxicology*, 45, p. 1238–1248.

Obied H.K., Prenzler P.D., Omar S. H., Ismael R., Servili M., Esposto S., Taticchi A., Selvaggini R., and Urbani S. 2012. Pharmacology of Olive Biophenols., p. 195-223. *In Advances in molecular toxicology* Vol. 6. Eds. James C. Fishbein and Jacqueline M. Heilman.

Obied H.K., Prenzler P.D., Robards K., 2008. Potent antioxidant biophenols from olive mill waste. *Food Chemistry*, 111, p. 171–178.

Paraskeva C.A., Papadakis V.G., Kanellopoulou D.G., Koutsoukos P.G., Angelopoulos K.C., 2007. Membrane filtration of olive mill wastewater and exploitation of its fractions. *Water Environment Research*, 79, p. 421–429.

Parenti A., Spugnoli P., Masella P. and Calamai L., 2006a. Carbon dioxide emission from olive oil pastes during the transformation process: technological spin offs. *Eur. Food Res. Technol.*, 222, p.521-526.

Parenti A., Spugnoli P., Masella P., Calamai L., and Pantani,O.L., 2006b. Improving olive oil quality us-

ing CO_2 evolved from olive pastes during processing. *Eur. J. Lipid Sci. Technol.*, 108, p. 904-912.

Pauselli M., Servili M., Esposto S., Gervasi G., Mourvaki E., Taticchi A., Urbani S., Selvaggini R., Concezzi L., and Montedoro GF., 2007. Effect of destoned olive cake as animal feed on ewe milk quality. Proc. of the International Conference New technologies for the treatment and valorization of agro by-products ISRIM, Terni-Italy 3-5 October.

Roig A., Cayuela M.L., Sanchez-Monedero M.A., 2006. An overview on olive mill wastes and their valorization methods. *Waste Management*, 26, p. 960–969.

Russo C., 2007. A new process for the selective fractionation and total recovery of polyphenols, water and organic substances from vegetation waters (VW). *Journal of Membrane Science*, 288, p. 239–246.

Servili M., Baldioli M., Selvaggini R., Macchioni A. and Montedoro GF. 1999 Phenolic compounds of olive fruit: One- and Two-Dimensional Nuclear Magnetic Resonance characterization of nüzhenide and its distribution in the constitutive parts of fruit. *Journal of Agricultural and Food Chemistry*, 47, p.12-18.

Servili M., 2012. Olive oil processing technologies and investments. Proc. of the International Seminar on Present and future of Mediterranean olive sector. Mediterranean Agronomic Institute of Zaragoza, Spain, 26-28 November 2012.

Servili M., Esposto S., Fabiani R., Urbani S., Taticchi A., Mariucci F., Selvaggini R., Montedoro GF., 2009b. Phenolic compounds in olive oil: antioxidant, health and sensory activities according to their chemical structure. *Inflammopharmacology*, 17, p. 76-84.

Servili M., Esposto S., Taticchi A., Urbani S. Di Maio I., Sordini B., Selvaggini R., Montedoro GF., Angerosa F., 2009a. Volatile compounds of virgin olive oil: their importance in the sensory quality. *In Advances in Olive Resources*, Berti L. and Maury J. Eds., p. 45-77.

Servili M., Taticchi A., Esposto S., Urbani S., Selvaggini R. and Montedoro GF., 2007a. Effect of olive stoning on the volatile and phenolic composition of virgin olive oil. *Journal of Agricultural and Food Chemistry*, 55, p. 7028-7035.

Servili M., Selvaggini R., Baldioli M., Begliomini L., Taticchi A., Esposto S., Montedoro G.F., 2002. Effect of the time of exposure to the air contact of olive pastes during the mechanical extraction process on the volatile and phenolic compounds of virgin olive oil. Innovation in virgin olive oil processing. *La Rivista Italiana delle Sostanze Grasse*, vol. LXXIX, 439–441.

Servili M., Pauselli M., Esposto S., Taticchi A., Urbani S., Selvaggini R., Montedoro GF., and Concezzi L., 2007b. New approach to the use of stoned olive pomaces for animal feeding. Proc. of the International Conference New technologies for the treatment and valorization of agro by-products. ISRIM, Terni, Italy 3-5 October.

Servili M., Selvaggini R., Esposto S., Taticchi A., Montedoro GF., Morozzi G., 2004. Health and sensory properties of virgin olive oil hydrophilic phenols: agronomic and technological aspects of production that affect their occurrence in the oil. *Journal Chromatography A*, 1054, p.113-127.

Servili M., Taticchi A., Esposto S., Urbani S., Selvaggini R. and Montedoro G.F., 2008b. Innovations in the extraction technology for the improvement of the virgin olive oil quality. Proc. 6th Euro Fed Lipid Congress., Athens, Greece 7-10 September. p. 138.

Servili M., Taticchi A., Esposto S., Urbani S., Sel-

vaggini R., & Montedoro GF., 2008a. Influence of the decrease in oxygen during malaxation of olive paste on the composition of volatiles and phenolic compounds in virgin olive oil. *Journal of Agricultural and Food Chemistry*, 56 (21), p. 10048–10055.

Servili M., Taticchi A., Esposto S., Urbani S., Veneziani G., Fabiani R., Morozzi G. and Montedoro, GF., 2007c. Recovery and chemical characterization of bioactive phenols from virgin olive oil vegetation waters. Proc. of the International Conference New technologies for the treatment and valorization of agro by-products, ISRIM, Terni, Italy 3-5 October.

Servili M., Esposto S., Veneziani G., Urbani S., Taticchi A., Di Maio I., Selvaggini R., Sordini B. and Montedoro GF. 2011a. Improvement of bioactive phenol content in virgin olive oil with an olive-vegetation-water concentrate produced by membrane treatment. *Food Chemistry*, 124, p. 1308–1315.

Taticchi A., Esposto S., Veneziani G., Urbani S., Selvaggini R., Servili M., 2013. The influence of the malaxation temperature on the activity of polyphenoloxidase and peroxidase and on the phenolic composition of virgin olive oil. *Food Chemistry*, 136, p. 975–983.

Terés S., Barceló -Coblijn G., Benet M., Álvarez R., Bressani R., Halver J. E. and Escriba P. V., 2008. Oleic acid content is responsible for the reduction in blood pressure induced by olive oil. *PNAS*, 105 (37), p. 13811–13816.

Turano E., Curcio S., De Paola M., Calabrò V., Iorio G., 2002 .An integrated centrifugation–ultrafiltration system in the treatment of olive mill wastewater. *Journal of Membrane Science*, 209, p. 519–531.

Vierhuis E., Servili M., Baldioli M., Schols H.A., Voragen A.G.J., Montedoro G.F., 2001. Effect of enzyme treatment during mechanical extraction of olive oil on phenolic compounds and polysaccharides. *J. Agr. Food Chem*. 49:1218-1223.

Olive production systems*

R. Fernández-Escobar[1], R. de la Rosa[2], L. León[2], J. A. Gómez[3], L. Testi[3], F. Orgaz[3], J. A. Gil-Ribes[4], E. Quesada-Moraga[5], A. Trapero[1] and M. Msallem[6]

[1] Departamento de Agronomía, Universidad de Córdoba. Campus de Rabanales, edificio C4, 14071, Córdoba, Spain. [2] IFAPA Centro Alameda del Obispo. Avda. Menéndez Pidal s/n, 14004, Córdoba, Spain. [3] Instituto de Agricultura Sostenible, CSIC. Apartado 4084, 14080, Córdoba, Spain. [4] Departamento de Ingeniería Rural, Universidad de Córdoba. Campus de Rabanales, 14071, Córdoba. Spain. [5] Departamento de Ciencias y Recursos Agrícolas, Universidad de Córdoba. Campus de Rabanales, edificio C4, 14071, Córdoba, Spain. [6] Institut de l'Olivier, Tunis, Tunisia.

ABSTRACT

The olive tree is one of the most important oil-producing crops in the Mediterranean region. The fact that olives have been present for centuries in most of the producing countries can be misleading as regards the sustainability, hardiness, longevity and adaptability of olive cultivation. At present, olive trees are planted in every region of the globe located between 30 and 45° latitude in both hemispheres. Olive growing is a complex agrosystem with differing production systems and technical packages of cultivation and genetic resources. Review of how the olive growing sector has evolved reveals that olive culture expanded slowly until the 1950s, after which planting systems switched from traditional to intensive. Since the 1990s, the tendency has been to convert traditional orchards to more intensive ones. Intensive and high density orchards are proliferating in the new growing areas and new producing countries with the objective of shortening the non-bearing period and reducing costs. These changes in growing systems have been accompanied in many cases by the irrational, unsustainable exploitation of natural resources and the introduction of new varieties. This report is a review of the different olive growing systems and the move from traditional to more intensive production systems.

Keywords. Genetic resources – Intensive orchards – Sustainable olive production – Mechanization – Pest and disease control.

INTRODUCTION

Olive orchards can be found under very different conditions, from desert to more humid climates, and occupy an area of about 10 million ha worldwide. In the last few years olive production has increased as a result of the development of modern orchards, the intensification of traditional orchards and the expansion of olive growing into new producing areas. Given the longevity of olive trees, the majority of the producing countries present a mosaic of different types of olive plantations.

Traditionally, the olive has been grown in extensive, dry farming conditions in orchards characterized by densities of up to 150 trees/ha and little mechanization. The profitability of these types of orchards is normally low and they are located in the oldest olive growing areas using local cultivars. A significant proportion is situated in steep, marginal areas. Most of the world's olive orchards are currently cultivated in this system.

From the 1970s, the development of irrigation, management and harvesting techniques brought changes in the new olive production sys-

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tems. This led to the intensification of olive growing with increases in tree density up to 450 trees/ha and final spacing determined by water availability, edaphoclimatic conditions, harvest system and cultivar. There is a strong tendency for harvest mechanization in these intensive orchards. Trees are trained to a single trunk with the canopy starting at around 1m from the ground. Continuous pruning from planting until the formation of the 5-6 year-old-tree is considered a key factor in obtaining a properly shaped canopy. The canopy volume per tree tends to be lower in new plantations whereas the density is higher.

The process of intensification continued in the early 1990s with the advent of high-density, hedgerow orchards, which evolved as a system capable of reducing the amount of labour needed for harvesting. Initially based on densities of around 2,000 trees/ha and spacing of around 3.75 x 1.35 m, the trees are always grown under drip irrigation in this system. Plants are trained to a monoconical shape as soon as they are planted. Straddle machines developed for vineyards have been adapted for use for olive harvesting. The main advantages of this system are the small labour requirements for harvesting and the early start of commercial production, which begins from three years after planting. However, the large amount of investment required for this planting system has led to a reduction in tree density to around 1,200 trees/ha (4 x 2 m spacing). Another drawback is that the excessive vigour of the few cultivars currently used in this system makes long-term productive hedgerows unpredictable (De la Rosa *et al*., 2007). Mechanical pruning has been proposed as a solution to reduce labour in both intensive and hedgerow systems and to keep canopy volume within suitable thresholds. One strategy to control vigour is the development of new low-vigour cultivars specifically designed for this growing system or the use of dwarfing rootstocks. However, the vigour of a cultivar is largely influenced by environment, so specific trials need to be conducted to test the suitability of a given cultivar in a specific environment. This is especially important in those areas outside the Mediterranean Basin where climatic conditions could dramatically affect vigour.

Table 1 summarizes the main characteristics of the growing systems. Although the systems listed are the most popular ones up to now, new systems are emerging as a result of new harvesting solutions. This is the case of orchards with densities of around 500–700 trees/ha designed for continuous mechanical harvesting in an over-the-row configuration (Ravetti and Robb, 2010). More cultivars could be adapted to this system, which lies halfway between

TABLE 1.
Characteristics of olive growing systems

Growing system	Area (%)	Production (%)	Density (trees/ha)	Productivity (kg/ha)
Marginal	20	15	< 80	≤ 1000
Traditional	50	45	80-150	1500-3000
Intensive	29	40	200-450	5000-7000 *(irrigated 8000-12000)*
High density	1		1500-2500	8000-12000

intensive and hedgerow orchards.

In many olive growing countries, the tendency is to convert some traditional orchards to more intensive plantations. Such attempts should be well planned and should take into account all the edaphoclimatic, economic and social aspects before being carried through.

GENETIC RESOURCES

The first olive growers selected individuals with the best characters in wild olive forests, i.e. those with the biggest fruit size, largest proportion of flesh and highest oil content. This process probably occurred simultaneously in different places across the Mediterranean region and gave rise to numerous local cultivars whose dissemination largely remained restricted to their area of origin. For many years, traditional olive orchards were planted with these local cultivars. However, in recent decades, olive growing techniques have evolved considerably, leading to new orchards designed to give higher yields and to facilitate mechanical harvesting.

Changes in growing systems have been accompanied in many cases by the replacement of traditional cultivars by varieties hitherto unknown in these areas. Frequently, this has not been preceded by experimentation to confirm the suitability of the varieties to the new areas, even though several studies show that the agronomic and quality characters of an olive cultivar can change depending on the area of cultivation. The lack of previous experimentation has led in some cases to the commercial failure of new plantings.

High density hedgerow plantings are a good example of the significant changes seen in olive orchards in recent years. However, there are no specific low-vigour cultivars or dwarfing rootstocks adapted to this system. Due to the lack of specific cultivars for this system, early-bearing cultivars such as 'Arbequina', 'Arbosana' or 'Koroneiki' have mainly been used (De la Rosa *et al*., 2007), although they cannot really be considered low-vigour cultivars. This could be a problem, particularly in very favourable growing conditions, and has stimulated the development of breeding programmes to obtain new cultivars to widen the range of available cultivars adapted to modern olive growing systems.

In the last few years, breeding attempts have been initiated in several countries although only a few have managed to complete the breeding process. Several new cultivars have been released recently as a consequence of breeding programmes. Among them, 'Barnea' (Lavee *et al*., 1986), 'Fs17' (Fontanazza *et al*., 1998) and 'Chiquitita'/'Sikitita' (Rallo *et al*., 2008) have been marketed with relative success both in their countries of origin and abroad.

Recently developed genomic tools could help to improve several aspects of olive growing. The most practical use of genomics nowadays is for the authentication of nursery plants. As planting a new olive orchard is very costly, and mistakes in the choice of cultivar used only become evident after 3–4 years of field growth, cultivar authentication is highly advisable, especially when foreign or little known cultivars are used. Additionally, there are molecular and serological tests to check whether nursery plants carry pathogenic fungi, bacteria or viruses. However, there has not been a great demand

for certified plants in most of the olive growing countries. Molecular markers and studies of expressed genes are also being used to discover the genetic basis of the most important olive agronomic traits such as oil content, oil quality and resistance to biotic and abiotic stress. In the future, these studies could help significantly to speed up breeding programmes by greatly facilitating the selection process.

SUSTAINABLE MANAGEMENT OF OLIVE ORCHARDS

Soil management and soil degradation

Soil degradation is one of the major threats to the sustainability of olive cultivation. Losses of top soil in areas characterized by shallow soils lead to damage such as the reduction of soil water storage capacity, which is critical to the survival and productivity of rainfed olives. The decrease of water quality in water courses due to excess sediment and agrochemicals has been noted as a major environmental problem in some olive growing areas.

Soil management in olive has been oriented for centuries at ensuring the productivity and survival of the plantation under conditions of limited rainfall by a combination of low plant density, limitation of tree canopy size through pruning and elimination of adventitious vegetation to limit competition for soil water. Traditional extensive cultivation resulted in a system where the soil was covered by vegetation during part of the year. This system was liable to moderate soil losses, especially if applied in a mosaic type of landscape where the orchards were surrounded by areas of natural vegetation or retention structures. Traditional systems based on integrated use of the orchard with low density intercropping with a field crop or grazing seemed to be in a similar situation. However, erosion rates increased and became unsustainable when olive cultivation was pursued systematically in sloping areas and in landscapes covered solely with olives.

The possibility of all-year-round tillage offered by mechanization combined with the availability of herbicides enabled complete elimination of adventitious vegetation. Measurements on a small catchment scale also indicate high losses affecting the quality of surface waters. The extrapolation from regional analysis to areas with similar conditions and types of soil management indicates that this is a widespread problem around the Mediterranean Basin where olive cultivation is still largely concentrated (Gomez *et al*., 2008).

In recent decades the major drive to control erosion in olive orchards has been to develop and expand the use of ground cover in orchard lanes to prevent erosion and improve soil properties. This cover is controlled by mowing or the application of herbicides in late winter or spring to prevent competition for soil water with the olives. However, in commercial farms the results achieved are still disuniform. Considerable uncertainty remains about the impact of the competition from the cover crop, especially in years of limited rainfall, in orchards planted at different densities or under changing climate scenarios characterized by higher temperatures and scarcer precipitation (Gucci and Caruso, 2011). The expansion of olive irrigation is a relevant change that will affect the use of cover crops in olive growing areas. This provides a margin of safety to implement cover crop soil management without significantly limit-

ing olive yield but does not seem to have been fully explored. The beneficial impact of applying pruning residue mulch has also been demonstrated. This has a similar effect to cover crops, but without the risk of water competition, although the amount of pruning residues needed to prevent erosion is not always available in less productive orchards.

The simplified landscape resulting from the extension and intensification of olive orchards has also led to severe gully erosion. This problem has serious consequences for the sustainability of olive cultivation. The techniques based on increasing ground cover are of little effect in preventing gully erosion because they are much more effective in lowering soil losses than reducing runoff (Gómez *et al*., 2011).

Irrigation and sustainability

Irrigation is a relatively recent practice in olive growing. Although some kind of irrigation has been traditionally applied to olives in very arid areas as an auxiliary practice, the bulk of farm water supply was usually assigned to other less drought-tolerant crops. The appearance of drip irrigation in the 1960s paved the way for easy irrigation of orchards located on sloping terrain. New plantations are mostly irrigated because tree yield responds strongly to even relatively small amounts of irrigation water. An evapotranspiration-yield function indicates an increase of 5 kg/ha of olive oil for each mm of water evapotranspirated during the season (Moriana *et al*., 2003). The relationship is not linear and means that the marginal productivity of water, i.e., the increment in yield for each incremental unit of water used by the crop, is higher under low water use and lower when approaching the full water requirement of the tree. Olive is usually grown in arid and semiarid environments where the irrigation water supplied to the farmers is the main limiting factor on crop production; under these circumstances, farmers naturally tend to apply less water than what is needed for full production and to distribute it over the maximum area while trying to avoid water stress when the crop is more sensitive (e.g. flowering or oil development). This strategy is called "regulated deficit irrigation" or RDI. Excessive water demand is a major risk associated with olive irrigation in that olive growing areas are mainly located in zones already at risk of desertification. The water requirements of olive growing areas must therefore be determined as precisely as possible. Shifting from rainfed to irrigated olive groves entails intensification of the cropping system. Maximum returns can only be obtained from water investment by routing the water to the more productive environments capable of supporting high-density, intensive and productive olive farming.

As many olive orchards are deficit irrigated and typically lie in semi-arid zones, soil salinization is often an environmental risk. The use of saline water in olive irrigation is not necessarily detrimental to the environment, but soil changes must be closely monitored for sustainability purposes. Salinity effects on yield depend on the concentration but even though tolerance is a cultivar-dependent characteristic, most cultivars grown under semiarid conditions may develop well with no significant reduction of yield with an ECe in a range between 3 and 6 dS/m. Olive trees are less sensitive to leaf Cl^- than Na^+, and Ca^{2+} plays an important role in Na^+ exclusion and retention mecha-

nisms. Melgar *et al*. (2009) suggest that highly saline irrigation water can be used for a long time without affecting olive growth and yield through proper management entailing the supply of Ca^{2+} to the irrigation water to prevent Na^{+} toxicity, the application of drip irrigation until winter rest and the use of a tolerant cultivar.

Fertilization, crop quality and environment

Fertilization is a common practice in olive growing because it aims to satisfy the nutritional requirements of the trees when the nutrients required for their growth are not provided in sufficient amounts by the soil. The annual fertilization programme may vary among orchards and among years within an orchard. However, a survey conducted in the Mediterranean region (Fernández-Escobar, 2008) revealed that in 77% of cases the fertilization programme was repeated every year and generally involved applying several mineral elements, even when in most cases the nutritional status of the orchard was unknown. This approach tends to apply more mineral elements than necessary and, at the same time, may cause mineral deficiencies if a specific element is not applied in sufficient amounts. The excessive application of unnecessary fertilizers increases growing costs, contributes unnecessarily to soil and water pollution and may have a negative effect on the tree and crop quality.

Predicting the amount of fertilizers required annually to support optimum productivity is not simple. From a rational point of view, a nutrient must be supplied only when there is proof that it is needed. For this purpose, leaf-nutrient analysis gives an indication of tree nutritional status and is an important tool for determining fertilization requirements (Fernández-Escobar, 2007).

Perennial plants like the olive have nutrient storage organs to help them easily reuse nutrients. This is why their nutrient needs are lower than those of annual plants. *Potassium* deficiency is the major nutritional disorder in rainfed olives because the low soil moisture limits the spread of the potassium ion through the soil solution and prevents its absorption by the roots. It is worse when yields are high because it is the element removed in largest amounts by the crop, around 4.5 g K/kg olives. In rainfed olive orchards, between two and four leaf applications of 1%–2% K have given satisfactory results, although it is usually necessary to repeat the applications in following seasons until K reaches an adequate level in the leaves. In calcareous soils, iron deficiency may occur in addition to potassium deficiency. Trees suffering from iron deficiency, known as *iron chlorosis*, display a characteristic series of symptoms such as yellow leaves, small shoot growth and lower yield. Iron chlorosis is difficult and costly to correct. The best solution for new orchards is to choose a variety that tolerates this anomaly. In established orchards the remedy is to apply iron chelates to the soil or to inject iron solutions into the tree trunk. *Calcium* deficiencies are to be expected in acidic soils. In these situations it is necessary to apply a limestone amendment. The amount required depends on the soil texture and pH. Finally, *nitrogen* is the mineral element required in the largest amounts by plants and consequently, it is commonly used in the fertilization programmes of horticultural crops. However, long-term studies have demonstrated that annual applications of nitrogen fertilizers are not necessary to maintain high productivity and growth. On the contrary, this practice has been reported to have nega-

tive effects on the tree, on crop quality and on the environment (Fernández-Escobar 2011). These studies recommend that the best strategy to optimize nitrogen fertilization in olive orchards, as well as fertilization with other nutrients, is to apply them solely when the previous season's leaf analysis indicates that leaf nitrogen concentrations have dropped below the deficiency threshold.

OLIVE PEST AND DISEASE MANAGEMENT

The olive is a woody crop with a complex agroecosystem in which there is a good balance of many organisms at different trophic levels. Some of these organisms are phytophagous or pathogens of the olive tree, others are entomophagous predators and parasitoids, i.e. antagonists of the pathogens, and some are even species that seek shelter. The phytophagous or pathogenic organisms that feed and/or develop on olive can largely determine whether olive cultivation is economically feasible in certain situations.

Most of the thousands of publications on olive insects in the Mediterranean region concern fewer than a dozen species, which are major pests. These include key pests such as the olive fly (*Bactrocera oleae*), olive moth (*Prays oleae*) and black scale (*Saissetia oleae*) and some secondary, although occasionally key pests such as oleander scale (*Aspidiotus nerii*), the two olive scolytids *Hylesinus oleiperda* and *Phloeotribus scarabaeoides,* and the olive pyralid moth (*Euzophera pinguis*).

There are over 100 olive pathogens, although only a few cause serious economic losses to olive groves. One important group comprises fungal leaf and fruit diseases, mainly scab or peacock spot caused by *Fusicladium oleagineum*, anthracnose due to *Colletotrichum* spp. and cercosporiose due to *Pseudocercospora cladosporioides*. These three diseases, which cause heavy olive defoliation and debilitation and reduce plant productivity and oil quality, are the reason for regular fungicide treatments in olive groves. Another important disease is verticillium wilt caused by the vascular fungus *Verticillium dahliae*. This disease was unknown 30 years ago, but is currently considered the most serious disease and the main challenge to olive growing in some Mediterranean areas. Other diseases that have a moderate impact on Mediterranean olive groves are tuberculosis or olive knot, which is caused by the bacterium *Pseudomonas savastanoi* pv. *savastanoi* and is associated with wounds on leaves and branches, and a root and crown rot caused by several species of the oomycete genus *Phytophthora*, especially prevalent in waterlogged soils (Trapero and Blanco, 2010). These olive pests and diseases clearly restrict olive oil production because they lower yields and raise total production costs. It is estimated that approximately 30% of the olives produced are lost to pests and diseases at an annual control cost exceeding 200 million euro.

Biodiversity tends to be high in traditionally managed olive plantations because their structural diversity provides a variety of habitats. The older trees support a wide diversity and high density of insects and microorganisms which, together with the tree's fruit, provide an abundant supply of food. The low level of pesticide use allows a rich flora and insect fauna to flourish, which in turn is a valuable food source for a variety of bird species. Conversely, the intensive application of production-raising techniques has a very detri-

mental effect on ground flora, microorganisms and insect populations and considerably reduces the diversity and total number of flora and fauna.

When planting olives, it is essential to use **certified material**, particularly to avoid later problems associated with scales, mealy bugs and other biting sucking insects. Certified material is also crucial as regards plant health, particularly so in the case of pathogens that cause systemic infections (*V. dahliae*, viruses and phytoplasmas) and of those that remain associated to the plant material and cannot be easily detected, such as the epiphytic stage of *P. savastanoi*, latent infections of *F. oleagineum* and infections caused by fungi or nematodes on roots. It is not rare for symptoms of some of these diseases to appear months or years after planting the trees.

Fertilization is notorious for its impact on insect incidence. Excessive application of nitrogen fertilization results in the emergence of many new shoots, which helps a high percentage of neonate nymphs of *S. oleae* to find suitable settlement sites. Nonetheless, balanced mineral nutrition improves not only the nutritional status of the olive trees but also their defence mechanisms, thus helping them to avoid those herbivores that develop easily on debilitated trees such as *E. pinguis* or *H. oleiperda*. Likewise, excess nitrogen and deficient potassium are acknowledged to heighten olive susceptibility to fungal foliar pathogens (mainly peacock spot) and verticillium wilt.

Irrigation can influence both the vegetative status of the olive and the soil microclimate by promoting the development of mites, scales and olive fly in the first instance and by heightening the incidence of *O. cribricollis* and white grubs in the second. Irrigation also increases the activity of root pathogens (*V. dahliae*, *Phytophthora* spp., etc) and irrigation water can contribute to pathogen dispersal. Both possibilities have been confirmed for verticillium wilt, which is therefore especially severe in irrigated olive groves.

Soil management systems have been found to influence not only phytophagous populations and soil-borne pathogens but also predators, parasitoids and antagonists. Little information is available, although some cruciferous cover crops have been reported to reduce soil-borne inoculum of *V. dahlia*. Cover crops have also been reported to increase leaf infection by *F. oleagineum* due to higher humidity in the lower parts of the tree canopy. Overall, conventional olive crop management has a negative impact on the abundance of canopy spiders and to a lesser extent on their diversity. Cover crops conversely promote spider populations, although this effect is greater in natural covers than in planted ones. In general, tillage may help to eliminate different stages of soil-dwelling pests and to reduce pathogen inocula that survive on fallen leaves, but it also destroys the nests of natural enemies, thus limiting their beneficial action, and it promotes inoculum dispersal of some soil-borne pathogens such as *V. dahliae*.

Pruning has a great impact on pest and disease incidence and control. It affects phytophagous insects and aerial pathogens by modifying the microclimate of the tree canopy and by reducing the inoculum after removing the affected parts of the tree. Improving tree aeration through pruning lowers the incidence of insects such as *S. oleae*, *P. oleae, L. ulmi* and of the aerial pathogens *F. oleagineum*, *Colletotrichum*, *P. cladosporioides* and *P. savastanoi*. On the other hand, severe pruning can cause intense growth of tender shoots and eventually pro-

mote olive scale activity. The avoidance of pruning wounds will decrease the incidence of *E. pinguis* and wood decay fungi. Pruning remains must be removed and destroyed prior to bark beetle emergence. The olive bark beetles *P. scarabaeoides* and *Hylesinus* spp and the bark mosquito *R. oleisuga* can be controlled by using baited pruning wood, which must be destroyed or treated with insecticides prior to adult emergence. In addition, leaving water sprouts on the tree may protect olive shoots from attack by *O. cribicollis* since it prefers the former.

Harvesting method and **time** may also influence the activity of insects and pathogens. Wounds caused by harvesting with poles promote *R. oleisuga, E. pinguis* and *P. savastanoi* activity. Early harvesting is recommended to reduce olive fly activity and fruit rot caused by *Colletotrichum* spp. and other fruit rot, so indirectly raising the quality of the resultant olive oil.

Planting density can have a great impact on pest and disease development, especially in dense plantations where there are shady areas between the trees, which therefore increase leaf wetness duration and infections by aerial pathogens (Trapero, 2007). Furthermore, in the current scenario of olive growing, the question arises of whether pests and diseases considered to be secondary in conventional plantations might not become a problem in new high density plantations. In most cases this kind of plantation has high, irrigation-related soil humidity that might create a favourable environment for pests and pathogens. Likewise, olive tree susceptibility to the olive fly *B. oleae* is higher under irrigated conditions than under rainfed conditions (Santiago-Álvarez *et al*., 2010). In addition, there are several reports on the need for measures to control new pests (i.e. *Margaronia unionalis*) and diseases (i.e. alternaria fruit rot) in high density olive plantations (León *et al*., 2007). Olive knot is becoming a key disease in the hedgerow systems where control measures need to be intensified due to increased wounds caused by harvesting and pruning.

Current olive **pest and disease management strategies** are still based on the use of chemical pesticides. However, increasing public sensitivity towards environmental pollution and problems derived from the side effects of these products have provided the impetus for the development of alternative, benign pesticides and the development of the concept of Integrated Pest and Disease Management (IPDM or IPM). IPM strategy is based on ecological principles and encourages biological control through natural enemies such as predators, parasites, insect pathogens and non-pathogenic antagonistic or competitive microorganisms. It also involves cultural control strategies to minimize pest and disease entry and spread, the use of tolerant plant species, and the judicious use of chemical pesticides.

Bioinsecticides are considered the most viable alternative for olive pest control. Nonetheless, while viruses, bacteria, and protozoa have to be ingested with food, entomopathogenic fungi enter via the exoskeleton, a mode of action by contact which makes them an attractive alternative to chemicals. Entomopathogenic fungi play a dual role as bioinsecticides because they may also be used as an unexplored source of new insecticide molecules of natural origin. Research has revealed the high occurrence of the mitosporic ascomycetes *Beauveria bassiana* and *Metarhizium anisopliae* not only in the soil of olive crops but also in olive and olive weed phylloplane. Additionally, *B.*

bassiana has been found to be a natural biocontrol agent of the olive moth *Prays oleae* and the olive pyralid moth, *Euzophera pinguis* (Quesada-Moraga and Santiago-Álvarez, 2008). Copper products are being widely used for the control of olive diseases, thus making it necessary to find alternatives to reduce the use of copper in olive groves. Currently, natural products, organic amendments and antagonistic microorganisms are being studied for the control of various diseases, but to date there are very few commercial applications of these products. Consequently, the biological control of olive diseases is a current challenge for modern olive growing.

MECHANICAL HARVESTING OF OLIVE ORCHARDS

Harvesting can account for up to 40% of crop costs. The type of harvesting system used is determined by the type of olive orchards (Table 2).

TABLE 2.
Costs of the olive oil harvesting systems used in the different type of olive orchards

Type of olive orchard	Yield (kg/ha)	Harvesting system	Harvesting cost (€/kg fruit)
Traditional orchards that cannot be mechanized	1,500-3,000	Branch shaker and manual harvesting with rods	0.15-0.25
Traditional orchards adapted to mechanization	4,000-6,000	Tractor-mounted trunk shaker	0.14-0.19
Intensive olive orchards	5,000-10,000	Self propelled trunk shaker	0.09-0.12
High-density olive orchards	8,000-10,000	Straddle harvester	0.04-0.06

Current advances in olive harvesting systems have not focused on traditional olive orchards. This type of orchard, designed for manual harvesting, poses several problems due to the steep slopes where mechanization is not feasible. The harvesting systems used in this kind of plantation include manual aids such as branch shakers and combs.

Vibrating systems are the most common harvesting methods used in olive oil orchards (Fig. 1). The current trend is to use trunk shakers to detach the olive fruits onto canvas or nets placed under the trees. This is the most versatile system because it can be used in both traditional and intensive olive orchards. However, harvesting efficiency is poor in traditional groves because of the large trunk diameter and number of trunks per tree, and the fact that the tree structure is adapted to manual harvesting which strongly influences the transmission of vibration.

Recently, an integral mechanical harvesting system is being developed for traditional olive orchards using canopy shakers (Fig. 2). The removal efficiency exceeds 80%.

One of the most important lines of research in olive harvesting is to maximize the percentage of fruit removed. In many instances harvesting efficiency is dictated by the operational parameters of the machines used and tree suitability for mechanical harvesting (Gil-Ribes *et al*., 2010).

Integral mechanical harvesting includes three meth-

Fig. 1. Tractor-mounted trunk shaker

ods: trunk shakers with inverted umbrella (wrap-around shakers), side-by-side trunk shakers and canopy contact shakers. The inverted umbrella trunk shaker is the most commonly used. The trees must be adapted and have an upright trunk more than 1 m long to facilitate clamping and the use of the catch frame. Side-by-side trunk shakers are another alternative based on vibration and fruit interception (Fig. 3). This machine comprises two separate catch frames which move in parallel to both sides of the row of trees and a trunk shaker mounted on one of the frames.

Olive straddle harvesters are self-propelled with hydrostatic transmission and

Fig. 2. Canopy shaker and catch frame adapted for traditional olive orchards

their structure covers the external surface of the trees (Fig. 4). Fruit is removed by a number of beating heads formed by curved rods, radially arranged on one or several axes, which impart a reciprocating shaking to the canopy at a low frequency and high amplitude. The olives are intercepted in the bottom of the tunnel where the fruit is driven by a circulating conveyor towards containers or continuously discharged on to trailers. During harvesting forward movement, a deformable mechanism allows folding and sealing with the tree trunk, thus avoiding fruit drop and loss.

The main advantage of the straddle harvester is that it permits continuous operation at a speed of 0.4–3 km/h, achieving harvest efficiencies ranging from 90 to 95% of the fruit crop. The tree canopy needs to be small – not more than 2.0–3.5 m high and 0.80 to 1.20 m wide – for these machines to be able to move the rods over the tree canopies. This is one of the major problems. Giant harvesters such as those used in intensive olive orchards have not been successful in Europe. However, they are commonly used and widespread in the new modern olive groves located in Aus-

tralia and Argentina. The results are promising, but due to its size and cost, this machinery can only be used on large, flat orchards where there is little rain during the harvesting period.

Table olive crop places specific limitations on mechanical harvesting due to damage to the tree bark, because of the early timing of harvest, and damage to the fruit (bruising). The best commercial trunk shakers achieve fruit removal rates of between 70 and 75% in the case of table olives. If the machine is adapted to the tree and vice versa, it is possible to achieve 85% harvesting efficiency. ■

Fig. 3. Inverted umbrella trunk shaker (*left*) and side-by-side trunk shaker (*right*) in intensive olive orchard

Fig. 4. Straddle harvester for intensive (*left*) and high-density hedgerow (*right*) olive orchards

REFERENCES

De la Rosa R., León L., Guerrero N., Rallo L. and Barranco D., 2007. Preliminary results of an olive cultivar trial at high density. In: Aust. J. Agr. Res., 58, p. 392-395.

Fernández-Escobar R. 2007. Fertilization In: *Production techniques in olive growing*. Madrid, Spain: International Olive Council, p. 145-168.

Fernández-Escobar R., 2008. Olive fertilization practices in the Mediterranean region. In: *Olivae,* 109, p. 13-22.

Fernández-Escobar R., 2011. Use and abuse of nitrogen in olive fertilization. In: *Acta Horticulturae,* 888, p. 249-258.

Fontanazza G., Bartolozzi F. and Vergari G., 1998. Fs-17. In: *Riv. Fruticoltura,* 5, p. 61.

Gil Ribes J.A., López Giménez F.J., Blanco Roldán G.L. and Castro García S., 2010. Mecanización. In: *El Cultivo del Olivo*. Madrid, Spain: Mundi-Prensa-Junta de Andalucía, p. 434-506.

Gómez J.A., Giráldez J.V. and Vanwalleghen T., 2008. Comments on "Is soil erosion in olive groves as bad as often claimed?"by L. Fleskens and L. Stroosnijder. In: *Geoderma*, 147, p. 93 – 95.

Gómez, J.A., Llewellyn, C., Basch, G, Sutton, P.B., Dyson, J.S., Jones, C.A. 2011. The effects of cover crops and conventional tillage on soil and runoff loss in vineyards and olive groves in several Mediterranean countries. Soil Use and Management 27: 502 - 514.

Gucci R. and Caruso G., 2011. Environmental stresses and sustainable olive

growing. In: *Acta Horticulturae,* 924, p. 19 – 30.

Lavee S., Haskal A. and Wodner M., 1986. 'Barnea' a new olive cultivar from first breeding generation. In: *Olea,* 17, p. 95-99.

León L., De la Rosa R., Rallo L., Guerrero N. and Barranco D., 2007. Influence of spacing on the initial productivity of hedgerow 'Arbequina' olive orchards. In: *Spanish Journal of Agricultural Research,* 5, p. 554-558.

Melgar J.C., Mohamed Y., Serrano N., García-Galavís P.A., Navarro C., Parra M.A., Benlloch M. and Fernández-Escobar R., 2009. Long term responses of olive trees to salinity. In: *Agricultural Water Management*, 96, p. 1105-1113.

Moriana A., Orgaz F., Pastor M. and Fereres E., 2003. Yield responses of a mature olive orchard to water deficits. In: *Journal of the American Society for Horticultural Science,* 128, p. 425 – 431.

Quesada-Moraga E. and Santiago-Álvarez C., 2008. Hongos Entomopatógenos. In: *Control biológico de plagas*. Navarra, Spain: Phytoma Publicaciones de la Universidad Pública de Navarra, p. 98-120.

Rallo L., Barranco D., De la Rosa R. and León L., 2008. 'Chiquitita' olive. In: *HortScience,* 43, p. 529-531.

Ravetti L. and Robb S., 2010. Continuous mechanical olive harvesting in modern Australian growing systems. In: *Adv. Hort. Sci.,* 24, p. 71-77.

Santiago-Álvarez C., Del Rio C., Casado G., Campos C. and Quesada-Moraga E., 2010. Variation of susceptibility to olive fly *Bactrocera oleae* (Gmelin) attack in ten olive Spanish commercial oil cultivars under dry and irrigated conditions. In: *IOBC/wprs Bull* , 59, p. 104.

Trapero A. and Blanco-López M.A., 2010. Diseases. In: *Olive growing*. Junta de Andalucía/Mundi-Prensa/RIRDC /AOA, Australia. p. 521-578.

Trapero A., 2007. Densidad de plantación y enfermedades del olivar. In: *Mercacei*, 51, p. 210-213.


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