

# Quality of Fruit and Oil of Black-Ripe Olives Is Influenced by Cultivar and Storage Period

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Black-ripe olives (*Olea europaea* cv. Ascolano, Manzanillo, Mission, and Sevillano), intended for oil extraction, were stored at 5 °C for 6–8 weeks to evaluate their postharvest physiology and quality changes. Also, samples of olives were placed at 20 °C for 2 weeks to determine the deterioration rate of four cultivars at ambient temperature. Fruit quality evaluations included color, visual quality, fruit firmness, mass loss, and water and oil content. Decay incidence, physiological disorders, and respiration and ethylene production rates of the olives were also recorded. Olive oil quality was determined by analysis of titratable acidity, peroxide value,  $K_{232}$  and  $K_{270}$  coefficients, and fatty acid composition of the olives. Fruit and oil quality of Ascolano and Manzanillo cultivars deteriorated more rapidly than that of Mission and Sevillano olives. Black-ripe Manzanillo and Ascolano olives could be stored with good air circulation at 5 °C for 2 and 4 weeks, respectively, whereas Mission and Sevillano cultivars could be stored for 6–8 weeks at 5 °C with maintenance of good fruit and oil quality.

**Keywords:** *Olea europaea*; olive; cultivar; fatty acids; olive oil; quality; storage

## INTRODUCTION

Olive processing in important producing countries (such as Spain, Italy, and Greece) is often not well synchronized with crop harvests due to the number and size of the oil extraction facilities (Garcia and Streif, 1991; Gutierrez et al., 1992). Olives are often piled into large heaps and stored at ambient temperatures for up to several weeks prior to processing for oil extraction (Garcia et al., 1996a), and during this period the greatest deterioration takes place (Olias and Garcia, 1997). Pressure within the olive pile during storage can cause fluid secretion from the fruit that can provide an optimum medium for growth of fungi and bacteria (Olias and Garcia, 1997). Under these conditions, anaerobiosis can occur in the inner part of the pile while aerobic losses occur in the outer part (Garcia and Streif, 1991). Furthermore, heat production from respiratory activity may accelerate the deterioration of the fruit (Garcia and Streif, 1991) and eventually cause the breakdown of cell structure (Gutierrez et al., 1992). Oil extracted from these damaged olives can be high in acidity and low in stability (Garcia et al., 1996a) and can develop a high content of volatile acids (acetic or butyric) that causes a characteristic musty smell (Olias and Garcia, 1997). The resultant oil requires refining, resulting in higher costs and loss of market value (Gutierrez et al., 1992).

Among many known cultivars of *Olea europaea*, only five are commercially important in California, namely, Ascolano, Barouni, Manzanillo, Mission, and Sevillano. Of these, Manzanillo is the most popular and widely grown olive cultivar (Luh and Martin, 1996). During

the harvesting season large quantities of olives have to be processed. Currently, table olives (picked mature-green) are stored in brine until they are processed into the California-style black olives (Luh and Ferguson, 1994). The wastewater from brine contains high amounts of salt, making disposal a major problem (Maxie, 1964; Kader et al., 1989). The use of brines or acidulant solutions (Luh and Ferguson, 1994) for the storage of mill olives has also been tried, but they give the oil a peculiar and unpleasant sensory attribute (Olias and Garcia, 1997). Therefore, storage of fresh olives is more desirable and could allow a more orderly flow to the processing plant (Kader et al., 1989). The possibility of extending the length of storage of olives before oil extraction could increase the yield of good quality oil (Petruccioli and Parlanti, 1987).

Storage of green Manzanillo olives at temperatures below 5 °C causes chilling injury (CI), and thus the minimum safe storage temperature is 5 °C (Maxie, 1964; Kader et al., 1990). These authors also reported that the severity of CI depends on time–temperature, cultivar, maturity, and atmospheric composition. Storage of mill olives at 5 °C has been successfully applied by the olive oil industry in Spain (Garcia et al., 1996b). Black-ripe olives are less sensitive to CI than mature-green olives (Agar et al., 1998), similar to avocado fruit (Kosiyachinda and Young, 1976).

Olives in advanced stages of ripening are generally more susceptible to hydrolytic and oxidative deterioration; the main advantage of obtaining oil from ripe olives is the loss of bitterness. However, the loss of polyphenolic compounds that are responsible for bitterness also provides oil stability (Garcia et al., 1996c).

Although there have been some data published on the storage of mature-green [skin green with reddish spots, stage 2, as described by Garcia et al. (1996b)] olives between harvest and processing, there is very little information on the storage of black-ripe [skin black with

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>50% purple flesh, stage 6, as described by Garcia et al. (1996b)] olives. The objective of this study was to evaluate intercultural differences and effects of cold storage at 5 °C on fruit and oil quality of four commercially important black-ripe olive cultivars produced in California.

## MATERIALS AND METHODS

Black-ripe olives (*O. europea* cv. Ascolano, Manzanillo, Mission, and Sevillano) were hand harvested on November 19, 1996, from trees in the same orchard that received the same cultural practices in the University of California Davis experimental orchard. Olives were immediately transported to the Postharvest Laboratory and sorted to obtain fruit of uniform size and color. The olives were randomly divided into 0.5 kg lots that were placed into 2 L glass jars, with each jar considered as one replicate. The jars were ventilated with humidified air in a flow-through system at a flow rate of 500 mL min<sup>-1</sup> at 5 °C. Additional 0.5 kg lots of olives were placed at 20 °C for 2 weeks to determine the deterioration rate of four cultivars at ambient conditions. Three replicates were used for each cultivar-storage duration combination, and all data points represent the means ± SE of the three replicates.

Evaluations in olives were done initially and after 2, 4, 6, and 8 weeks of storage for the quality and ripening characteristics as described below. The percentages of decayed olives with visible mycelial growth and those exhibiting physiological disorders (mainly CI) were determined.

External skin color (opposite sides) was measured on 20 olives for each replicate with a Minolta Chromameter (model CR-300, Minolta, Ramsey, NJ). The visual quality was determined based on the following hedonic scale: 1 = poor, unusable; 3 = fair, limit of usability; 5 = good, limit of marketability; 7 = very good; and 9 = excellent. A weighted average of individual olive quality scores was used to determine the mean visual quality score for each replicate.

The mass of olives in each replicate was recorded initially and after different treatments and storage durations using a balance, and the difference was used to calculate percent mass loss.

Firmness of black-ripe olives was measured by a momentum transfer generator (MTG) (Washington State University, Pullman, WA). The device is a nondestructive impact sensor developed specifically for measuring the firmness of sweet cherries and other soft fruits (Younce and Davis, 1995). Mitcham et al. (1998) reported that the MTG measured firmness of sweet cherries with greater precision than the UC firmness tester incorporated with an Ametek penetrometer (Ametek, Inc., Matfield, PA). Two firmness measurements were collected from opposite sides of each olive of 20 olives per replicate. Olive firmness is reported in MTG units.

Ten olives from each replicate were transferred into a Petri dish and weighed before the dishes were placed in an oven at 105 °C for 48 h until a constant mass was reached. Water content was determined from the difference between fresh and dry mass and expressed as a percent. Oil content was determined in duplicate by extracting the oil from 15 previously dried olives used for determining olive water content. The olives were ground by mortar and pestle, and 10 g of the paste was put into a Soxhlet cartridge. The oil was extracted with 150 mL of hexane at 70 °C for 6 h. The percent oil content was calculated on dry mass basis from collected hexane.

Carbon dioxide (CO<sub>2</sub>) and ethylene (C<sub>2</sub>H<sub>4</sub>) production rates from olive cultivars were monitored in triplicate. An infrared CO<sub>2</sub> analyzer (model PIR-2000R, Horiba Instruments, Irvine, CA) and a gas chromatograph (model 211, Carle Instruments, Anaheim, CA), equipped with an FID detector, were used to measure CO<sub>2</sub> and C<sub>2</sub>H<sub>4</sub>, respectively.

Olive oil extraction was done according to a procedure simulating the commercial process (Garcia and Streif, 1991). About 400 g of black-ripe olives was triturated with a mortar and pestle, after which malaxation of the paste slowly occurred

for 30 min. Malaxation continued for an additional 30 min following addition of 100 mL of water to the paste. Extraction of the oil occurred by compression of the paste in four layers of cheesecloth. This was repeated, following the addition of a further 50–100 mL more water to extract the remaining oil. The filtrate was centrifuged at 3000g<sub>r</sub> to separate the oil from the water. Oil was collected by a Pasteur pipet under light vacuum, filtered through Whatman No. 2 filter paper, filled in dark glass bottles, and flushed with nitrogen to eliminate O<sub>2</sub>. The bottles were stored at -20 °C until analyses were performed.

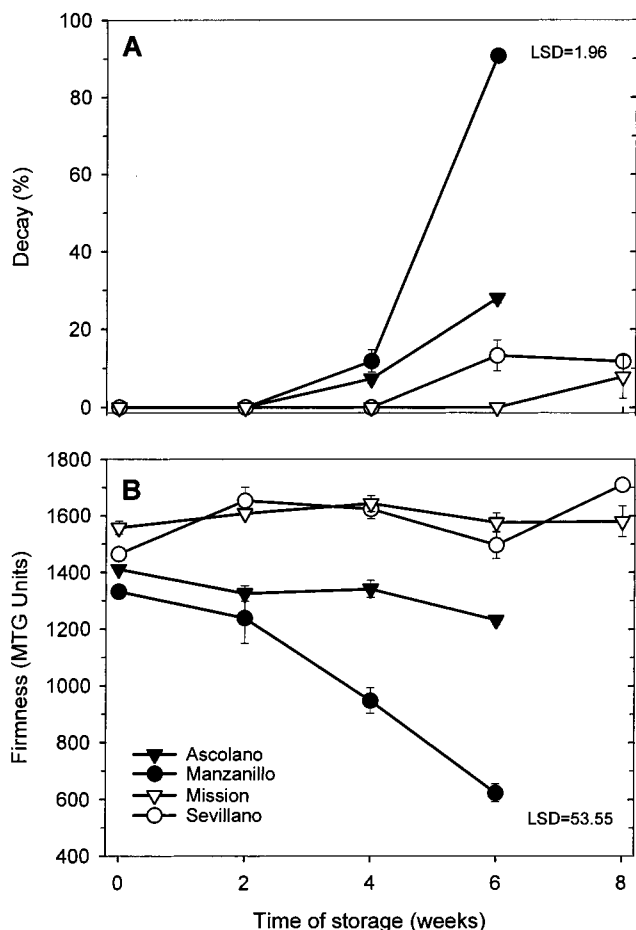
Titrate acidity was determined in triplicate for each cultivar based on the method described by Garcia et al. (1996a). The olive oil sample of 20 g was placed in an Erlenmeyer flask, and 125 mL of a previously neutralized solvent mixture was added. The solvent mixture consisted of equal parts of ethanol and diethyl ether and phenolphthalein as an indicator (1% in ethanol) in the ratio of 2 mL to 125 mL of the solvent mixture. When the sample was completely dissolved, it was titrated with 0.1 N KOH to the first permanent pink color (persisting for at least 10 s) of the same intensity as the neutralized solvent prior to its addition to the sample. The results were expressed as percent of the free oleic acid present in the oil.

Peroxide value was assayed according to the method of Garcia et al. (1996a). A 5 g olive oil sample from each of the three replicates of each treatment was placed in a 250 mL Erlenmeyer flask that had been purged with nitrogen. The sample was shaken and then dissolved in 25 mL of acetic acid/chloroform solution (2:1, v/v), where O<sub>2</sub> was removed by bubbling N<sub>2</sub> through the solution. One milliliter of saturated potassium iodide (KI) solution was added. The mixture was placed in darkness for 5 min and then 75 mL of distilled water was added to stop the reaction. Half a milliliter of freshly prepared starch indicator solution (0.5%) was added to each sample. Finally, the mixture was titrated with 0.01 N sodium thiosulfate until the indicator blue color disappeared. The peroxide value was expressed as milliequivalents of O<sub>2</sub> per kilogram of oil.

The coefficients of specific extinction at 232 and 270 nm were measured according to the following procedure. An oil sample of 250 mg was placed in a 25 mL graduated flask and diluted to 25 mL with cyclohexane (spectrophotometer grade). The sample was then homogenized using a vortex, and the resulting solution was filled into a quartz cuvette. Absorbance at 232 and 270 nm was measured in a spectrophotometer (Shimadzu model 1601) using pure cyclohexane as a blank (Garcia et al., 1996a).

Ten olives from each replicate of each cultivar were randomly selected and analyzed individually for their fatty acid composition. Oil extraction and fatty acid methyl esters (FAMES) were done in one step according to the method of Garces and Mancha (1993). This method allows complete oil extraction and fatty acid transmethylation in the same tube. For oil extraction, 100 mg of flesh was taken from each olive. Samples were boiled at 80 °C for 2 h with a reagent mixture consisting of methanol/heptane/benzene/2,2-dimethoxypropane/H<sub>2</sub>SO<sub>4</sub> (37:36:20:5:2, by volume). Approximately 3 min after the extraction, tubes were placed in a hot water bath and samples shaken vigorously to form one phase. After 2 h, the extraction tubes were cooled to room temperature, at which two phases formed. The upper phase containing the FAMES was transferred to smaller vials, flushed with nitrogen, and capped.

The composition of FAMES was determined by gas chromatography performed on a Hewlett-Packard 5890 GC equipped with an HP 7673 autosampler, a controller, and an FID detector, fitted with a SP-2330 column (30 m, 0.25 mm u.d., 0.2 μm film thickness) (Supelco, Bellefonte, PA). The FAMES were identified on the basis of R<sub>f</sub> of known standards (Sigma). Injector and detector temperatures were 220 and 250 °C, respectively. Oven temperature was held at 190 °C for 7 min, then increased to 210 °C at 10 °C increments, and held for 5



**Figure 1.** Intercultivar differences in the incidence of decay (A) and softening rate (B) of black-ripe olives during storage at 5 °C. Data points represent means of three replicates  $\pm$  SE.

min at the final temperature. Column pressure was maintained at 11 psi.

Analysis of variance (ANOVA) and Duncan's multiple range test with a significance level of  $P < 0.05$  were performed on all of the data using CoStat Statistical Software, ver. 5.01 (CoHort Software, Minneapolis, MN).

## RESULTS AND DISCUSSION

### Decay Incidence and Physiological Disorders.

Cold storage at 5 °C delayed decay incidence on black-ripe olive cultivars (Figure 1A). The four cultivars could be separated into two groups of high and low decay incidence. Manzanillo and Ascolano olives had 12 and 7% decay after 4 weeks, which increased to 90 and 27% after 6 weeks at 5 °C, respectively. Sevillano olives had 13 and 11% decay after 6 and 8 weeks of storage at 5 °C, respectively, whereas Mission olives had only 8% decay after 8 weeks at 5 °C. Similar trends occurred for olives kept at 20 °C (Table 1). Manzanillo and Ascolano olives exhibited 40 and 23% decay, respectively, Sevillano olives had 6% decay, and no decay was observed in Mission olives after 2 weeks of storage at 20 °C.

Garcia et al. (1996a) reported that >80% of mature-green Picual olives decayed after 2 weeks at ambient temperatures, whereas 25% decay was observed after 6 weeks of storage at 5 °C. Similarly, Garcia et al. (1996b) reported that 20% of mature-green Villalonga and Blanqueta olives decayed after 60 days at 5 °C or 20 days at ambient temperature. These results are

comparable to our results with black-ripe Manzanillo and Ascolano olives stored at 20 °C, although much lower decay incidence was observed for black-ripe Sevillano and Mission olives in our study.

Chilling injury (CI), which can be a major cause of deterioration in fresh olives stored before processing, is described as internal browning around the pit or skin at advanced stages (Kader, 1996). In our experiments no visible CI was observed in any of the black-ripe olives of the four cultivars during the entire storage period.

**Color and Visual Quality.** Storage time and cultivar had no significant effect on the color of black-ripe olives (data not shown). Ascolano and Manzanillo olives had sharper declines in visual quality ratings than the other cultivars. Olives were rated between 6 and 7 after 4 weeks, dropping to between 4 and 5 (limit of marketability) after 6 weeks storage. In contrast, Sevillano and Mission olives ranged between 8 and 9 in visual ratings during the entire storage period. Visual quality of Manzanillo and Ascolano olives was rated as 2 and 4, respectively, after 2 weeks at 20 °C, whereas Sevillano and Mission olives remained between 8 and 9.

**Mass Loss and Water and Oil Content.** Mass loss increased with storage time and ranged between 0.6 (Sevillano) and 1.5% (Manzanillo) in black-ripe olives stored at 5 °C (data not shown), and no visible signs of shriveling were observed. Mass loss ranged between 2.3 and 3% in black-ripe olives stored at 20 °C for 2 weeks. Higher mass loss at 20 °C might be partially due to fungal decomposition of olives resulting in the leakage of cell fluids (Castellano et al., 1993), as well as transpiration.

Water content at harvest was 67.9, 60.5, 52.5, and 61.2% in Ascolano, Manzanillo, Mission, and Sevillano olives, respectively. There were no significant differences in water content of black-ripe olives over storage time (data not shown).

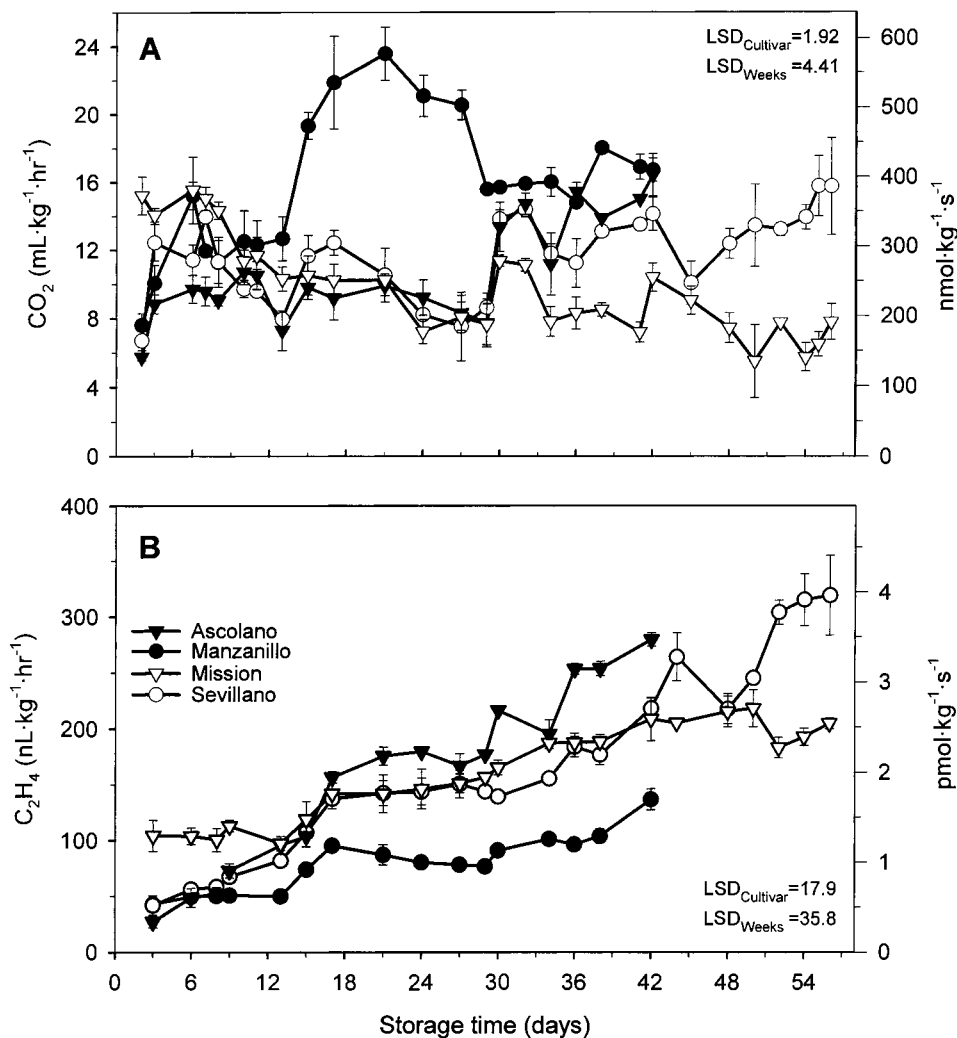
The oil content remained unchanged in the four cultivars during storage at 5 °C. Black-ripe Ascolano and Sevillano olives contained 28.5 and 28.8% oil (on a dry mass basis), respectively, whereas Manzanillo and Mission olives contained 33.3 and 37.7%, respectively. Oil content of black-ripe Manzanillo and Ascolano olives stored at 20 °C for 2 weeks was 29.9 and 26.5%, respectively (Table 1), which was lower than the initial values and those of olives stored at 5 °C, whereas oil content of the other cultivars remained unchanged.

**Fruit Firmness.** Cold storage delayed olive softening (Figure 1B). The firmness of black-ripe Ascolano and Manzanillo olives decreased by 9 and 29% after 4 weeks and by 16 and 53% after 6 weeks of storage, respectively. Mission and Sevillano, the two firmer among the four cultivars, maintained their firmness during the 8 week storage period at 5 °C. Similar results were found at 20 °C, with no loss of firmness in Sevillano and Mission (Table 1), whereas the firmness of Ascolano and Manzanillo decreased by 13 and 44% within the 2 weeks. Garcia et al. (1996a,b) reported that low-temperature storage delays softening caused by endogenous activity of fruit-ripening enzymes and softening caused by exogenous action of pathogens. It is noteworthy that the olive firmness correlated very well with decay incidence, as highest decay was detected in Manzanillo olives, which also softened the most. Sevillano and Mission cultivars, which preserved their initial firmness for as long as 8 weeks, exhibited little decay,

**Table 1. Intercultivar Differences in Fruit and Oil Quality of Black-Ripe Olives Stored at 20 °C for 2 Weeks**

cultivar	storage period (weeks)	decay (%)	firmness (MTG units)	water content (%)	oil content (%)	titratable acidity (% oleic acid)	peroxide value (mequiv of O <sub>2</sub> kg <sup>-1</sup> of oil)	SEC at K <sub>232</sub>	SEC at K <sub>270</sub>
Ascolano	0	0 <sup>a</sup>	1410	67.9	28.5	0.13	3.5	1.13	0.06
	2	23	1221	66.0	26.5	3.20	6.5	1.90	0.17
Manzanillo	0	0	1332	60.5	33.3	0.32	2.6	1.24	0.10
	2	40	742	58.6	29.9	5.70	7.4	1.70	0.21
Mission	0	0	1557	52.5	33.7	0.13	2.6	1.23	0.04
	2	0	1664	52.1	33.8	0.31	5.1	1.86	0.16
Sevillano	0	0	1464	61.2	28.8	0.11	2.6	1.19	0.10
	2	6	1587	61.0	28.9	0.37	5.4	1.43	0.10
LSD <sub>cultivar,0.05</sub> =		2.5	99.4	0.51	1.65	0.75	0.69	0.10	0.020
LSD <sub>time,0.05</sub> =		1.8	70.3	0.27	0.86	0.53	0.49	0.08	0.017

<sup>a</sup> Data points are the means of three replicates.



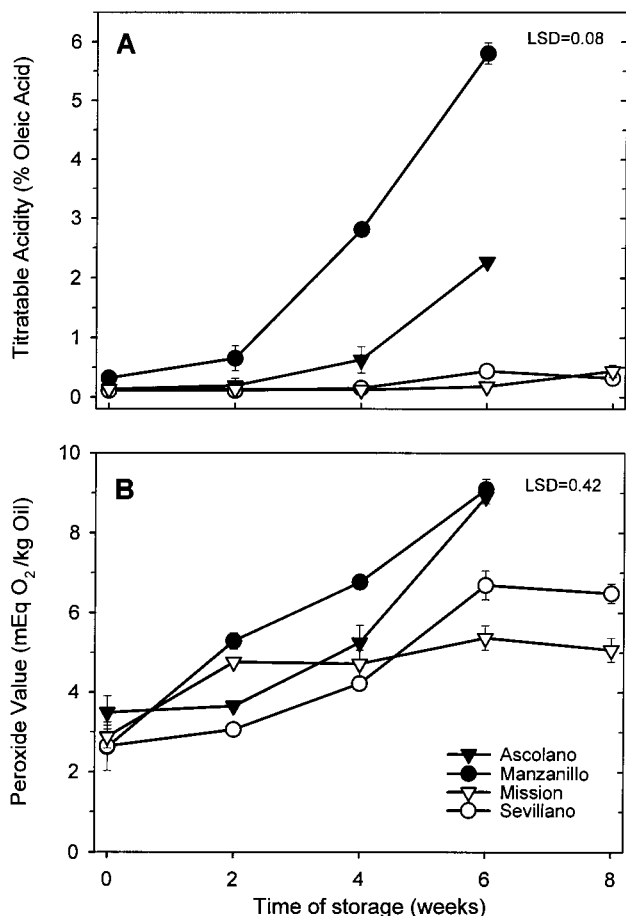
**Figure 2.** Intercultivar differences in respiration (A) and ethylene production (B) rates of black-ripe olives during storage at 5 °C. Data points represent means of three replicates  $\pm$  SE.

whereas Manzanillo had both highest decay incidence and softening rate.

**Respiration Rate and Ethylene Production.** Respiration rate (Figure 2A) fluctuated and ethylene production (Figure 2B) increased with storage time, and both varied among cultivars. Initial respiration rates of Ascolano, Mission, and Sevillano olives ranged between 6 and 16 mL kg<sup>-1</sup> h<sup>-1</sup> for up to 6 weeks of storage at 5 °C. Respiration rates of Mission olives tended to decline over 8 weeks of storage. In contrast, Manzanillo olives exhibited a sharp respiratory increase after 2 weeks to a maximum of 23 mL kg<sup>-1</sup> h<sup>-1</sup>. Manzanillo

olives had a consistently higher respiration rate than the other three cultivars. Ethylene production of black-ripe olives increased after 12 days at 5 °C and continued to rise until the end of the storage period (Figure 2B). Ascolano olives had the highest C<sub>2</sub>H<sub>4</sub> production among the tested cultivars. Although CO<sub>2</sub> production rates of black-ripe Manzanillo olives were highest, C<sub>2</sub>H<sub>4</sub> production was lowest compared to the rest of the cultivars. Mission and Sevillano olives exhibited similar C<sub>2</sub>H<sub>4</sub> production rates for up to 6 weeks at 5 °C.

Initial CO<sub>2</sub> production of black-ripe olives kept at 20 °C ranged between 50 and 80 mL kg<sup>-1</sup> h<sup>-1</sup> and declined



**Figure 3.** Intercultivar differences in acidity (A) and peroxide value (B) of black-ripe olives during storage at 5 °C. Data points represent means of three replicates  $\pm$  SE.

to  $\sim 35$  mL kg<sup>-1</sup> h<sup>-1</sup> after 2 weeks (data not shown). The respiration rate of Manzanillo olives was consistently higher than that of the other three cultivars. Ethylene production rates at 20 °C were also 2–5-fold higher than the initial values of olives stored at 5 °C and declined to between 60 and 140 nL kg<sup>-1</sup> h<sup>-1</sup> levels after 2 weeks (data not shown). The higher respiration rate in black-ripe Manzanillo and Ascolano cultivars may be partially responsible for their faster deterioration. These results on respiration rate, ethylene production, and cultivar variability agree with previous data (Garcia and Streif, 1991; Kader et al., 1990).

**Titratable Acidity.** During the first 4 weeks of storage, acidity values of Ascolano, Mission, and Sevillano olives stored at 5 °C were all below 1%, which is the accepted limit for “extra” quality virgin olive oil (Figure 3A). The acidity of oil obtained from Manzanillo olives reached 2.8% after 4 weeks, which is classified as “semi-fine” virgin olive oil (<3.3% acidity). The acidity continued to increase up to 5.8% after 6 weeks of storage. Oils extracted from Ascolano olives after 6 weeks had 2.7% acidity (semi-fine), whereas acidity levels of oil extracted from Sevillano and Mission olives remained <1% over the 8 week storage period at 5 °C.

The sudden increase of acidity in oils extracted from Manzanillo (5.7%) and Ascolano (3.2%) olives stored at 20 °C correlated well with the development of decay incidence (Table 1). In contrast, Sevillano and Mission olives kept at 20 °C for 2 weeks retained <1% acidity.

Garcia et al. (1996a) reported that mature-green Picual olives stored at 5 °C for 45 days retained

titratable acidity levels <1%, which is similar to our results with Sevillano and Mission olives but contrary to our results with Manzanillo and Ascolano olives kept at 5 °C. It has been well documented that, normally, the titratable acidity of an oil increases with increased storage temperature (Gutierrez et al., 1992; Garcia et al., 1994) and parallels the percentage of decayed olives in the lot from which it was extracted (Garcia et al., 1996b).

**Peroxide Value.** Cold storage at 5 °C significantly delayed the rise in peroxide value of the oils, which is an indication of oxidation (Figure 3B). After 6 weeks of storage, oil of Manzanillo and Ascolano olives had reached  $\sim 9$  mequiv peroxide value, compared to 5 and 6 mequiv for Mission and Sevillano olives. Oil obtained from olives of all cultivars stored at 20 °C for 2 weeks doubled and tripled in peroxide values relative to initial values and ranged from 5.1 to 7.4 mequiv of oxygen/kg (Table 1). Regardless of storage temperature or cultivar, in our study none of the oils analyzed exceeded the maximum peroxide value for extra quality virgin olive oil of 20 mequiv of O<sub>2</sub>/kg of oil. Garcia et al. (1996a,b) also reported that peroxide value of Picual, Blanqueta, and Villalonga olives stored at ambient temperatures and at 8 °C had increased peroxide values relative to those stored at 5 °C.

**Specific Extinction Coefficient at  $K_{232}$  and  $K_{270}$ .** The  $K_{232}$  value is an indication of conjugation of polyunsaturated fatty acids in olives, whereas  $K_{270}$  is an indication of carbonylic compounds (aldehydes and ketones) (Garcia et al., 1996b). UV specific extinction determination permits an approximation of the oxidation process in unsaturated oils (Gutierrez et al., 1992). Extra virgin olive oil has maximum values of 2.40 and 0.20 SEC for  $K_{232}$  and  $K_{270}$ , respectively.

Mission olives showed slower rates of change during storage at 5 °C compared to the other cultivars. Oil from Ascolano olives stored for 6 weeks at 5 °C exceeded extra virgin oil standards. Gutierrez et al. (1992) had observed higher  $K_{232}$  values in mature-green Picual olives stored at ambient temperature than cold-stored olives, whereas Garcia et al. (1996b) found no significant differences in  $K_{232}$  values of the two mature-green cultivars (Blanqueta and Villalonga) stored at 5 and 20 °C for 20 days. In our study, none of the cultivars had oil exceeding the maximum value for  $K_{232}$  after 2 weeks of storage at 20 °C.

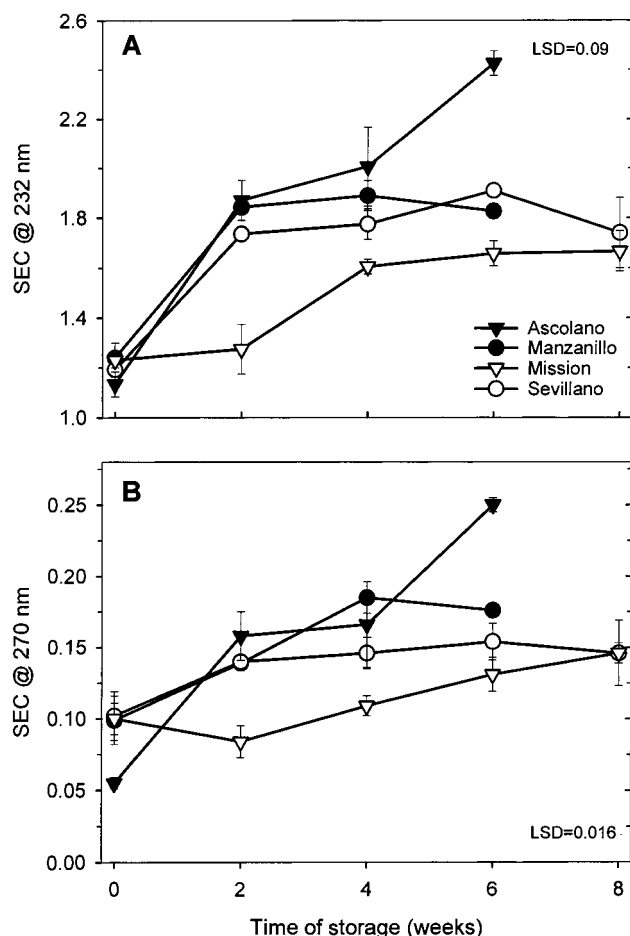
Similar results were observed with  $K_{270}$  values. Values at 20 °C after 2 weeks of storage were generally higher than at 5 °C (Table 1; Figure 4A). Our values for the content of carbonylic compounds measured by  $K_{270}$  value are in accordance with previous reports for mature-green olives (Gutierrez et al., 1992; Garcia et al., 1996a). Garcia et al. (1996b) reported that cold storage had minimal effects on the  $K_{270}$  value of Blanqueta and Villalonga olives.

**Fatty Acid Composition.** Among the eight analyzed fatty acids, only palmitic and oleic acid changed significantly over storage time (Table 2). The fatty acid composition of olives did not change significantly after 2 weeks of storage at 20 °C (data not shown). Palmitic acid is the primary saturated fatty acid in olives (Table 2). Manzanillo (14.3%) and Ascolano (13.9%) olives had a significantly higher content of palmitic acid than Mission (11.2%) olives. The palmitoleic acid content of the cultivars ranged between 0.9 (Manzanillo and Ascolano) and 1.6% (Sevillano and Mission). The stearic

**Table 2. Intercultivar Differences in the Fatty Acid Composition of Black-Ripe Olives Stored at 5 °C**

cultivar	storage period (weeks)	fatty acid composition <sup>a</sup> (%)							
		palmitic 16:0	palmitoleic 16:1	stearic 18:0	oleic 18:1	linoleic 18:2	linolenic 18:3	arachidic 20:0	eicosenoic 20:1
Ascolano	0	15.4	1.5	1.9	60.6	19.4	1.0	0.0	0.2
	2	12.9	1.2	2.2	64.1	18.9	0.3	0.1	0.2
	4	13.9	1.5	2.2	63.6	17.7	0.9	0.0	0.1
	6	13.4	1.0	2.1	66.0	16.1	1.0	0.0	0.2
Manzanillo	0	15.2	1.9	4.1	65.9	10.5	1.3	0.5	0.2
	2	13.7	1.5	3.9	70.1	9.4	0.9	0.3	0.1
	4	14.4	1.6	4.3	68.9	9.2	1.1	0.4	0.1
	6	13.8	1.4	5.3	69.1	9.1	1.0	0.2	0.1
Mission	0	11.1	1.0	2.1	72.8	12.1	0.5	0.1	0.2
	2	11.3	0.9	2.0	72.6	12.5	0.2	0.2	0.2
	4	11.5	0.9	2.2	72.6	12.1	0.4	0.1	0.1
	6	10.7	0.7	2.1	74.7	11.0	0.3	0.1	0.2
Sevillano	0	14.5	1.1	1.9	67.8	13.7	0.7	0.0	0.1
	2	12.2	0.9	2.0	70.5	13.6	0.3	0.2	0.2
	4	12.9	1.1	1.9	69.1	13.9	0.7	0.1	0.2
	6	13.2	0.8	2.0	68.7	14.6	0.5	0.1	0.3
	8	15.5	1.4	1.8	63.8	15.8	1.3	0.0	0.2
LSD <sub>cultivar,0.05</sub> =		0.80	0.16	0.49	2.5	1.37	0.34	0.13	0.07
LSD <sub>time,0.05</sub> =		0.88	0.14	0.50	1.9	1.33	0.21	0.13	0.08

<sup>a</sup> Data points are the means of three replicates.



**Figure 4.** Intercultivar differences in specific extinction coefficient (SEC) at 232 nm (A) and at 270 nm (B) of black-ripe olives during storage at 5 °C. Data points represent means of three replicates  $\pm$  SE.

acid content of Manzanillo olives averaged 4.4% and was 2-fold higher than that of the other three cultivars.

Oleic acid was the main monounsaturated fatty acid found in these black-ripe olive cultivars, and three well-defined groups are distinguished. The average oleic acid

content of Mission olives was 73%, that of Sevillano and Manzanillo olives was 69%, and Ascolano olives contained 63.5% oleic acid after 6 and 8 weeks at 5 °C. A high concentration of oleic acid enhances the stability of the oil, and a diet rich in this fatty acid reduces cholesterol levels.

Polyunsaturated fatty acids, essential for human nutrition, were dominated by linoleic acid in black-ripe olives. The greatest relative difference among the tested cultivars was in linoleic acid content. In the black-ripe Ascolano and Mission olives, the sums of oleic and linoleic acids were nearly identical, but ratios differed, with 63.5% oleic and 18% linoleic in Ascolano and 73 and 11.9%, respectively, for Mission. This inverse relationship between oleic and linoleic acids has also been seen in oilseeds (Garcia et al., 1992). The linolenic acid concentration of the studied cultivars also differed significantly. Mission, Sevillano, Ascolano, and Manzanillo cultivars contained on average 0.35, 0.55, 0.8, and 1.1%, respectively. Traces of arachidic and eicosenoic acid were found in these cultivars.

The composition of fatty acids may vary with cultivar, maturity, and growing location (Luh and Ferguson, 1994). Fatty acid profiles of black-ripe olive cultivars determined in this study are within the required range of extra virgin olive oil, except for linolenic acid. The maximum limit for linolenic acid allowed by the International Olive Oil Council (IOOC) in extra virgin olive oil is 0.9% (Raina, 1995; Spiller, 1996). In our study we obtained slightly higher values for linolenic acid than reported by Luh and Ferguson (1994), especially in Manzanillo and Ascolano cultivars. Because we used chemical extraction for fatty acid analysis in olives, our data correspond to the total lipids of the olive fruit including polar lipids such as glycolipids, which are characterized by high levels of linolenic acid (Mancha, 1974). Virgin olive oil obtained by physical methods might not contain this fatty acid in the quantities we observed.

Manzanillo olives grown in Peru and Picholine Marroca and Picholine Langedoc cultivars from Morocco also have high linolenic acid contents (Ajane et al., 1998). Interestingly, the linolenic acid content of Mis-

sion and Sevillano cultivars increased as the storage time was prolonged; this effect may be due to the senescence of black-ripe olives during long-term storage. Although not a limitation for olives destined for table consumption, the high linolenic acid content must be taken into consideration if olives are destined for oil extraction. All of the research cited in this paper used only olive oil quality criteria such as acidity, peroxide value, and  $K_{232}$  and  $K_{270}$  values for classifying oil because they used cultivars that do not show high contents of linolenic acid. The industry is using acidity as the primary indicator for olive oil quality, probably because it is easy to measure. In California, extra virgin olive oil is currently made from Mission and Manzanillo olives that are blended after pressing to obtain better flavor. Nevertheless, oil of black-ripe olive cultivars with high linolenic acid concentration should be blended with oils from other cultivars to achieve the 0.9% limit set by the IOOC.

In conclusion, decay resistance, fruit and oil quality, and overall storage performance of black-ripe Sevillano and Mission cultivars ventilated with humidified air at both 5 and 20 °C were superior to these characteristics of the other two cultivars. The performance of black-ripe olives stored at 20 °C was remarkable, because all published literature on olive storage reports that decay rates >20% are common for mature-green olive cultivars kept at >20 °C for 10–20 days. This may be due to good air circulation throughout the olives in our study. Black-ripe Manzanillo and Ascolano olives could be stored at 5 °C for up to 4 weeks, whereas Mission and Sevillano cultivars could be stored for 2 weeks at 20 °C or for up to 8 weeks at 5 °C with maintenance of good fruit and oil quality. Effective air flow throughout the olives is needed to prevent respiratory heat accumulation in any area within the stored olives.

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