Grasas y Aceites
Vol. 46 Fasc. 3 (1995), [redacted] 153-159

INVESTIGACIÓN

Colour changes in different processing conditions of green olives of Chalkidiki variety.

By **G.D. Karaoulanis** and **A. Bamnidou**
University of Thessaloniki, Dept. of Food Science, Box 279, 54006 GREECE

RESUMEN

Cambios de color en diferentes condiciones de tratamiento de aceitunas verdes de la variedad Chalkidiki.

Aceitunas verdes de mesa de la variedad Chalkidiki, sometidas a ennegrecimiento enzimático durante su tratamiento y conservación por métodos comerciales, se recogieron en diferentes cosechas y en dos años distintos. Posteriormente, se trataron con un 1'5%, 1'7%, 2% y 2'3% de NaOH.

Durante la eliminación del amargor, lavado, fermentación y conservación, las aceitunas verdes se guardaron, tanto como fue posible, libre de contacto con el aire. El pH se mantuvo a valores relativamente bajos (~4'0), principalmente durante la conservación de aceitunas verdes. Además, la concentración de salmuera se mantuvo constante durante la etapa de fermentación y conservación (8-10%).

La precaución anteriormente señalada tuvo como resultado la protección del color de la pulpa y de la piel del ennegrecimiento enzimático a un nivel muy satisfactorio.

En cuanto a la textura del fruto, la firmeza se conservó bien.

PALABRAS-CLAVE: Aceituna verde de mesa — Color — Conservación (efecto de) — Chalkidiki (variedad) — Tratamiento habitual (efecto del).

SUMMARY

Colour changes in different processing conditions of green olives of Chalkidiki variety.

Green table olives of Chalkidiki variety, which are subject to enzymatic browning during processing and preservation by the commercial method, were collected in different pickings and in two cultivating years. Then, they were treated with 1.5%, 1.7%, 2% and 2.3% NaOH.

During the removal of bitterness, washing, fermentation and preservation the green olives were kept, as much as possible, away from any contact with air. The pH was kept at relatively low values (~4.0), mainly during the preservation of green olives. Besides, the concentration of brine was kept constant during the stage of fermentation and preservation (8-10%).

The aforementioned precaution resulted in the protection of the colour of the flesh and of the skin from enzymatic browning to a very satisfactory degree.

As for the texture of the fruits, the firmness was well preserved.

KEY-WORDS: Chalkidiki (variety) — Colour — Common processing (effect of) — Green table olive — Preservation (effect of).

1. INTRODUCTION

The cultivation of olive tree in the Mediterranean region dates back to antiquity. Olives are used either for the extraction of olive oil or for consumption as table olives (green or black). The green olive fruits are frequently wounded on the surface or in the flesh during harvest, treatment (processing) or storage. As a result their flesh develops brown spots or brown areas which affect the quality of the fruits and decrease their commercial value (Donaire et al,. 1975).

The brown appearance of the flesh of fruits and vegetables resulting from the mechanical injury during postharvest storage or processing is a wide spread phenomenon (Mathews and Parpia, 1971).

The browning of the flesh is commercially undesirable, because of the unpleasant appearance of the fruits and the concomitant development of off-flavor. The browning potential of various fruits, and some times in different varieties of the same species, has been shown to be directly related to the phenol, the polyphenol-oxidase (PPO) activity or combination of these factors (Sciancalepore, 1985, Sciancalepore and Longone, 1984, Kahn, 1977).

Luh and Phithakpol (1972) suggested for the control of the browning problem in peach canning to wash them thoroughly, cool with water spray and pass through 1% citric or malic acid bath or spray with these acids, so that the pH is always less than 4.

Besides, a combination of high concentration of $C0_2$ and O_2 seems to inhibit significantly the activity polyphenol-oxidase and is the main cause of polyphenol destruction in olives (Maestro, R. et al,. 1993).

In studies of enzymatic browning of fruits, it was noted that many tissues high in carotenoids (i.e. papaya, mango) are characterized by low browning potential relative to tissues low in carotenes (apples, avocado, olives) (Sharon, O. and Kalhn, 1974).

The browning of injured olives could be markedly reduced by dipping the fruits in 0.4% NaOH. Higher NaOH concentrations are also employed in the industrial processing of green olives to remove the bitter glucoside oleuropein. NaOH partially removes the cuticle and apparently inactivates catechol oxidase. Phenolic substrates or their oxidation products may also be leached out (Ben-Shalon et al., 1978).

The pH optimum which have been reported for catechol oxidases are usually around pH 4.5-7 and maximum activity of olive catechol oxidase using 4-methylcatechol as the

substrate was observed at pH 4-5 (Ben-Shalon et al., 1977). Other researchers emphasized the importance of the pH of the system, which may in fact be the only factor determining the resistance of enzymes during the ripening of fruits (Aylward, F. and Haisman, D., 1977).

On the other hand, ripe green olives undergo browning as a result of mechanical injury. Preliminary work showed that blanching the fruit at 90°C for 5 min or the effect of α-irradiation, prevented this phenomenon, an indication that browning is enzymatic (Ben-Shelon, N. et al., 1977, 1978, Karaoulanis, G. unpublished data,1987).

The prevention of the appearence of the brown color in the green table olive fruits of Chalkidiki variety (in Macedonia, Greece) is important and this work aimed at the study of certain parameters during the treatment, which could prevent the appearance of browning without affecting the quality characteristics of the green olives.

For this purpose, at first, we studied the different concentrations of NaOH needed for the removal of bitterness (oleuropein) and we took into consideration the conditions of washing. Besides, during fermentation and preservation of the olives, we made observations of the concentration of brine, pH and acidity of brine, the firmness and the color of the skin and mainly of their flesh.

2. MATERIALS AND METHODS

The olive fruits which were used for the experiments came from an orchard located in the middle of the Chalkidiki peninsula (in Macedonia, Greece) belonging to farmers of the Cooperative Union of Polygiros, Chalkidiki. The olive trees were 25 years old and had received the same cultivating treatments. Pickings took place at the end of October and in the first days of November.

The fruits were of medium size i.e. 120 fruits per Kg and were placed for processing in plastic containers of 3.5 Kg each for the period 1986-87 and of 5 Kg each for the period 1987-88.

Subsequently, the containers were filled with a solution of lye (NaOH) in different concentrations, i.e. 1.5%, 2.0%, 2.3% and 1.7 or 1.8% which is used for processing of green olives in commercial scale for removing the bitter glucoside oleuropein.

For each treatment 3 plastic containers were used. The olives were left in lye until it penetrated almost to the pit. The time required for the lye to penetrate this depth in the flesh ranged from 12 to 14 hours at ambient conditions (20-23°C). As soon as lye reached the pit we started washing the fruits by preventing at the same time their contact with oxygen (air), by means of a net which kept the olives immersed in the liquid (lye or water) for the period of time required for the substitution of lye by water (washing). The duration of the washing was between 27-50 hours.

Immediately after washing and without any contact of the fruits with air, the boxes were filled with brine. The initial concentration of the brine was 10%. During fermentation, the change of the concentration of the brine was corrected and kept at 10%. The brine used for the storage of green olives had also a concentration of 10%. The acidity was also corrected by adding lactic acid, when necessary.

During the fermentation of green olives and also during their storage, the following observations were performed: diameter of the fruits, pH, acidity of the brine, firmness, color of skin and flesh, concentration of brine.

The color of the external surface and mainly of the flesh of the green olives, which is the most important quality characteristic and directly influences consumer preference, was determined by means of a Hunter colorimeter model TCU-COLOR.

The instrument color values were: **L** (darkness-ligthness), **a** (redness,-a greenness) and **b** (+b yellowness, -b blueness). Before each measurement the instrument was standardized with a prototype green plaque, having values: **L**=64.2, **a**=-14.4 and **b**=+5.6.

Titrable acidity of brine was determined with N/10 NaOH in lactic acid. The pH of brine was determined by a table pHmeter.

Firmness, was measured with a pressure tester type Chatillon penetrometer with a modified plunger 2mm in diameter. For this determination 50 fruits were used and measurements were taken on both sides of each fruit in gr/mm².

The experiment was repeated two times in two consecutive growing seasons (2 years). In the second year two experiments, representing two different pickings in 10 day intervals were performed. Every measurement was repeated on three samples and the average value is reported.

The duration of the first experiment (1986-87) was 300 days while the second (1987-88) flasted 250 days and was extended for the study of color and texture for another 250 days.

3. RESULTS AND DISCUSSION

Experiment 1986-1987

1. pH

Fig. 1 shows the values of the pH of the brine in which the fruits of green Chalkidiki olive were kept during the whole experiment.

We observe that the pH values in the first few days of the experiment were quite high in all treatments with lye (NaOH), ranging from 6.3 to 6.0. Following the beginning of the fermentation, the pH became about 5.0. These changes to lower values continued during the fermentation stage and during the conservation of olives for 300 days. At the end of the experiment the pH values were between 3.8 and 4.0.

Among the different treatments there were very small differences in the pH values. The lowest pH value was determined in the treatment with 1.7% of NaOH and the highest in the 2.3% treatment.

In general, the pH values ranged in low levels. This may be one of the reasons for the inhibition of the activity of some enzymes which probably affect the changes of the color of both the skin and flesh of the fruits.

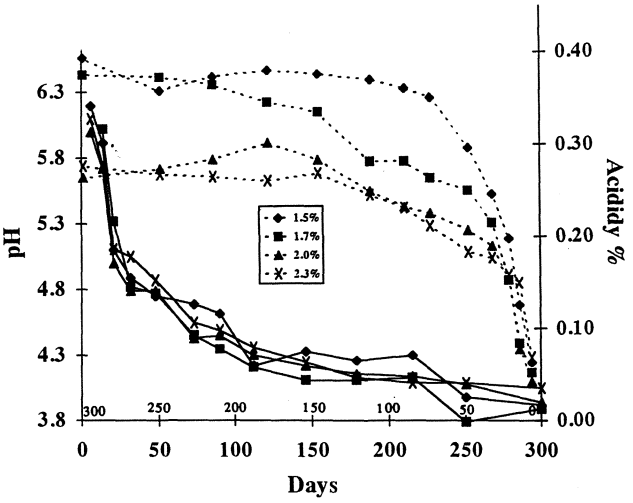

Fig. 1
Changes in the pH and in the acidity of the brine during fermentation and preservation for 300 days of green olives, Chalkidiki variety, after different treatments with lye.

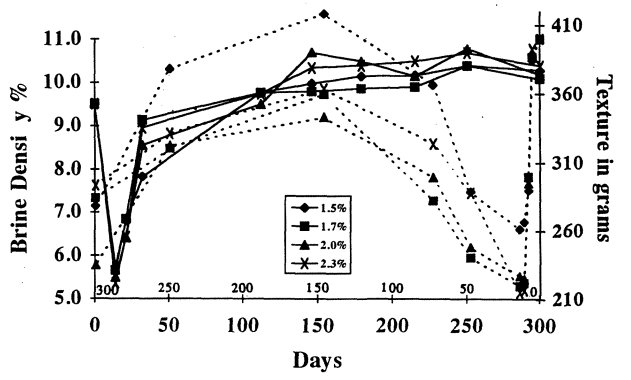

Fig. 2
Changes of the texture of the green olives before and after treatments with lye and changes in the concentration of the brine and of the texture during fermentation and preservation for 300 days.

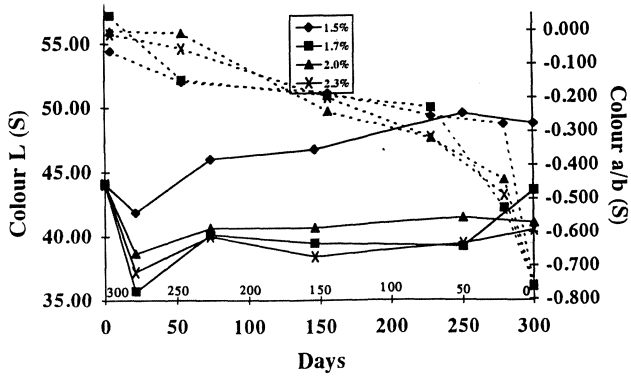

Fig. 3
Changes of the skin color, **L** values and **a/b** ratio values, of the green olives after harvest and treatments with lye and during fermentation and preservation for 300 days.

2. Acidity

Fig. 1 shows the different values of acidity of the brine where the olives were placed after different treatments with lye (NaOH).

The values of the acidity follow a different pattern from those of the pH, i.e. they are very low, 0.04-0.05%, immediately after placing the olives in the brine, increase to 0.20-0.29% during fermentation, continue to increase slightly and finally remain stable. Only in the case of treatment with 2.0% lye we observe a slight decrease of the acidity of the brine in the end of the preservation period.

The highest increase of the acidity was observed in the treatments with 1.5% and 1.7% lye (soda) and the lowest in the treatments with 2.0% and 2.3% lye (NaOH).

In general, the values of the acidity in the different treatments remained low during the whole experiment in accordance to the general behaviour of the pH.

3. Texture

Fig. 2 shows the different values of texture of the olive fruits before treatment, after treatment with lye of different concentrations, during fermentation and during preservation of the olive fruits in brine for 250 or 300 days.

We observe that the values of texture were high before treatment, namely 390 Kgr/mm^2, and decreased considerably after removing the bitterness by washing, reaching the values of 210 gr/mm^2 except for the treatment with 2.3% which had a slightly better value. During fermentation and conservation, the texture is almost restored, the fruits become more hard and the values of texture increase. In the treatment with lye 1.5% the fruits almost regain their initial texture after 150 days from the beginning of the experiment.

In general, in all treatments, the texture has the highest values after 150 days from the beginning of the experiment.

After this period of time the values of the texture tend to decrease for the rest of the experiment.

In addition to equilibrium other factors can also affect texture restoration.

4. Brine

Fig. 2 shows the changes observed in the concentration of the brine during fermentation as well as during preservation of the olive fruits after different treatments with lye.

In all treatments, the concentration of the brine at the beginning of fermentation was about 10%. Shortly after the beginning of fermentation, the concentration decreased quickly to 5.0% and was kept at a constant level of 8-10% until the end of the experiment by correcting interventions, i.e. by addition of salt.

Among the treatments the highest and more stable concentration of the brine (10%) during preservation was in the case of 2.0% lye followed by the treatments 2.3%, 1.5% and 1.7% lye.

The reduction of the concentration of the brine at the beginning of fermentation is due to the need of the fruits to

compensate the internal pressure of the cells lost after the treatments with lye and washing by absorbing salt from the brine.

5. Color

a. Color of the skin

Figure 3 shows how the color of the skin of the olives changed during the experiment (after harvest and during fermentation and preservation).

The color of the skin at the beginning is deep green, the value of **a** is -14.000 and the ratio **a/b**=-0.800, while the value of brightness is **L**=44.80. During fermentation the color changes to light green while the values of the ratio **a/b** is from -0.500 to -0.300 and of the brightness **L** up to 35.00.

Also during preservation there is a graduate discoloration of the skin color which continues until the end of the experiment, when the color becomes yellow-green and sometimes yellow or yellow-pink.

As for as the value of the brightness is concerned the smallest change appears in the fruits of the treatment with 1.5% lye, while the values of **a** and of the ratio **a/b** show about the same change in all treatments.

Finally, we observe that the values of the measurement of the color during the experiment are quite satisfactory in almost all treatments, despite the changes of the skin in this period of time.

b. Flesh

Figure 4 shows the changes in the color of the flesh of olives from harvest to the end of the experiment.

The color of the flesh after harvesting is deep green, the value of the ratio **a/b** is -3.50 and of brightness **L** is 47.00.

In all treatments, the most pronounced change takes place during fermentation.

The changes continue in lower rate during the next stage i.e. during preservation in the treatments with lye 2.3% and 2.0%. In the treatment with 1.7% lye and near the end of the experiment the changes are more pronounced, while in the treatment with 1.5% lye the worsening is quite strong starting from the stage of fermentation to the end of the experiment leading finally to the yellow color of the flesh.

The value of **L** can be considered as stable with small fluctuations in the treatments with 1.5%, 2.0% and 2.3% lye during the fermentation and the preservation while in the case of the treatment with 1.8% lye, the values of **L** showed strong fluctuations and were considerably reduced at the end of the experiment.

Experiment 1987-1988, a and b

Figures 5a,b, 6a,b, 7a,b, and 8a,b show the results of the observations performed in experiments a and b of the period 1987-88. The result of each observartion is the mean value of analyses of three different samples, each coming from a different container.

1. pH

As shown in figure 5a,b the values of the pH of the brine with the green olives were high and between 5.5 and 5.2 in all treatments 1.5% at the beginning.

During fermentation, the values of the pH gradually but steadily become lower. At the stage of preservation, the reduction of the pH continues almost to the end of the experiment reaching a value of 3.9-4.4. This behaviour of the values of the pH probably contributed to the fact that the olives had a good appearance and good color of skin and flesh.

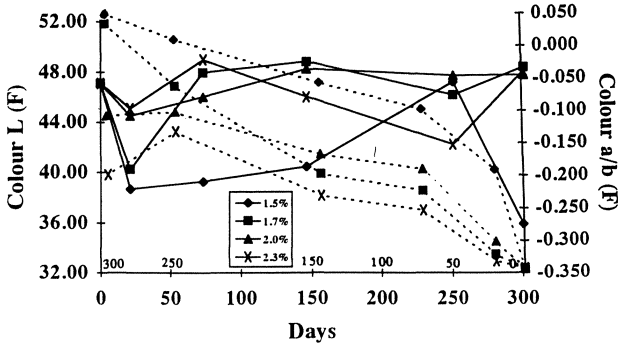

Fig.4.
Changes of the flesh color, **L** values, and **a/b** ratio values of the green olives after harvest and treatmets with lye and during their fermentation and preservation for 300 days.

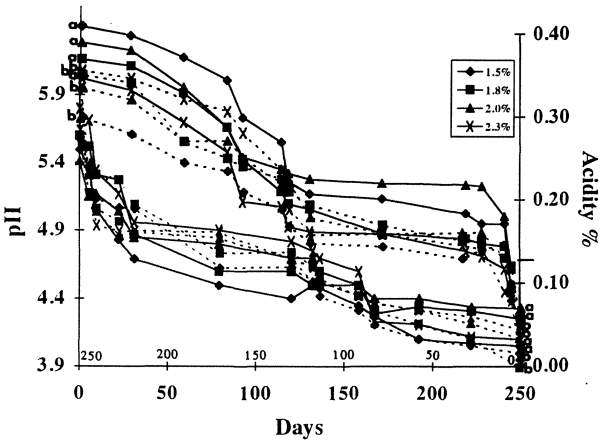

Fig. 5a,b.
Changes in the pH and in the acidity of the brine during fermentation and preservation for 250 days of green olives, Chalkidiki variety, after different treatments with lye.

2. Acidity

The figure 5a,b shows the changes of the acidity of the brine, where the olive fruits were stored, in the different treatments.

At the beginning of the stage of fermentation the values of the acidity are low (0.1%-0.2%) in all cases. Gradually, as the stage of fermentation proceeds they show an arithmetical increase until they reach some maximun values.

Then, during preservation, they remain stable until the end of the experiment.

In general the reduction of the acidity which has high arithmetical values, is actually quite small (0.1%-0.2%) but remains in the levels which inhibit the activation of certain enzymes, which lead to the changes of the green or yellow green color of the flesh of the olive fruits.

3. Texture

Figure 6a,b shows the different values of the texture of olive fruits at the beginning of the experiment and also during its performance.

In all cases the value of the texture is high at the beginning, while when fermentation begins, a sharp reduction is observed due to the treatment for the removal of bitterness and the washing of the fruits (from 450gr/mm^2 to 300gr/mm^2) followed by a relative stabilization due to the absorption of salt by the olive fruits.

In both experiments, better values were observed in the treatment with 1.5% of lye.

In general, the stabilization of the values of the texture to 270-280gr/mm^2 is almost satisfactory.

4. Brine

Figure 6a,b shows the changes of the concentration of the brine in salt from the immersion of the olives in it to the end of the experiment.

During the first days of the immersion of the olives in the brine we observed a reduction of its concentration down to 4-5% in all treatments requiring a daily correction by the addition of salt, until a stable value of 8-10% was established.

The reduction was larger in the cases of 1.8% and 2.0% of lye.

In general, the time for the reduction of the concentration of the brine and the time required for its stabilization were quite similar in all treatments, namely 2-3 days for the reduction and 2-3 for the stabilization.

This phenomenon is considered quite natural after the treatment of the olive fruits with lye for removing the bitterness and after their washing.

5. Color

a. Skin

Figure 7a,b shows the changes of the color of the olive skin from picking to the end of the experiment.

In all cases the color of the fruits starts from a high negative value, which changes sharply during fermentation, and continues to change for quite some time after it.

In the first experiment (a), the cases with 2.3% lye and 1.8% lye show a rather stable change of the color, while the change of the color of the treatment with 1.5% lye is more sharp.

As for the brightness, the values fall steadily until the end of the experiment, except for the case of 2.3% lye.

Morever, in the second experiment (b) the change of the green color is as sharp as for the values of a and b during fermentation and for quite long time after it. Following, the color stays stable until the end of the experiment with the exception of the case with 1.5% lye, where the discoloration is more pronounced.

As for the values of brightness of the skin (Fig. 8a,b) drop after fermentation, that is the color becomes worse

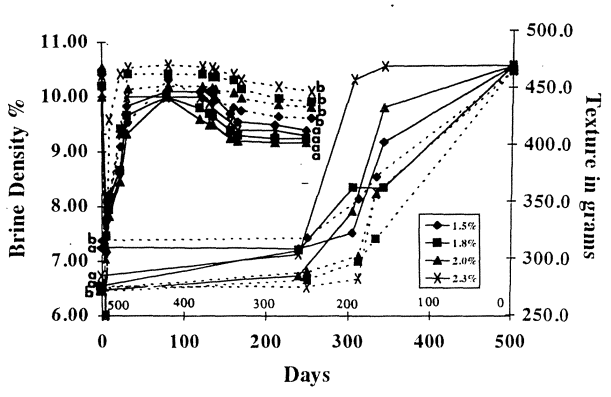

Fig. 6a,b.
Changes of the texture of the green olive fruits before, after treatments with lye and during fermentation and preservation for 500 days. Also changes in the concentration of the brine during fermentation and preservation for 250 days of green olives after different treatments with lye.

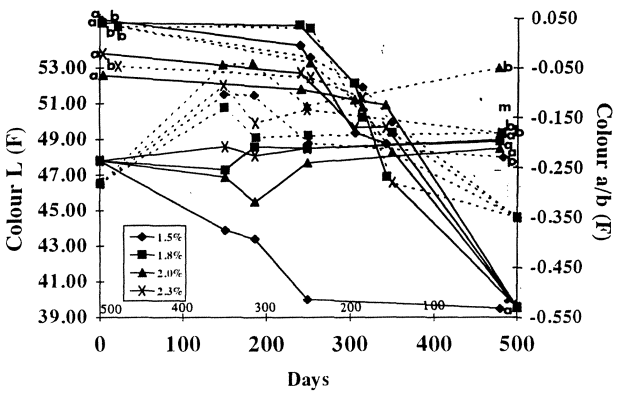

Fig.7a,b.
Changes of the skin and of the flesh color, **L** values and **a/b** ratio values, of the green olives after harvest and treatments with lye and during fermentation and preservation for 500 days.

mainly in the treatments with 2.3% and 1.8% while remain stable in the treatments with 1.5% and 2.0% lye until the 350th day.

After that period of time the brightness for the case with 1.5% is stable until the end of the experiment while it becomes worse in the case of 2.0% lye.

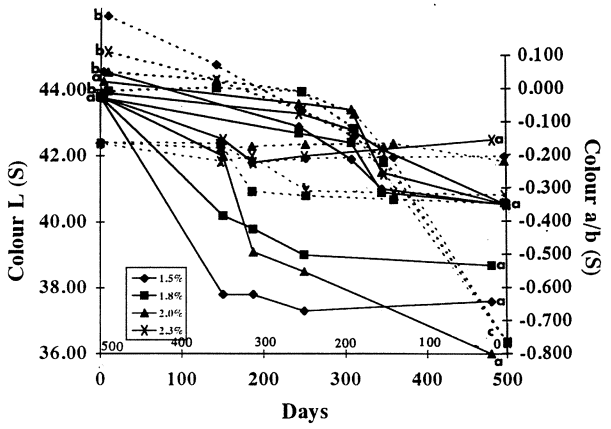

Fig.8a,b
Changes of the skin and of the flesh color, **L** values and **a/b** ratio values, of the green olives after harvest and treatments with lye and during fermentation and preservation for 500 days.

b. Flesh

The color of the flesh in both experiments a and b in all cases changed during fermentation and preservation from dark green (negative high values of **a**) to light green (negative low values of **a**) and sometimes to yellow or light red (positive values of **a**) (Fig. 7a,b).

More precisely, in the experiment a, we observe that in the cases of 1.7% and 2.3% lye, the color of the flesh changes but remains light green or yellow-green (values negative low) while in the cases of 1.5% and 2.0% lye, the changes of the color are more pronounced that is the values are positive. The values of the ratio **a/b** behave in the same way.

This means that the color of the fruit became dark yellow or light red, thus affecting the quality of the green olives and their market value in a negative way.

The values of a (Fig. 8a,b) of the flesh after fermentation show some improvement becoming higher followed by a return to almost the initial values. Thus, the flesh of the olive fruits is, from the point of view of brightness, at satisfactory levels.

4. CONCLUSIONS

The observations on the evolution of the quality characteristics, mainly of the color of the olive flesh, in the experiments of the years 1986-87 and 1987-1988 reported here in lead to the following conclusions.

1. Generally, in all treatments of both experiments the values of the **a** were quite satisfactory, a fact which implies that the color of the flesh was kept good during the whole experiment. Besides, the values of the ratio **a/b** of green olives stayed within satisfactory levels.
2. From all these experiments, one could attribute a basic role for the conservation of the color to the following:

   a. The values of pH during the experiments were kept at quite low levels (Fig. 1, and 5a,b). Thus, the flesh of the green olives were kept in brine with pH=4.0, had very good light green-yellow color, followed by the fruits with pH=4.3 and 4.6. Almost the same behaviour was showed for the color of the skin. Our observation is in agreement with that of Aylward and Haisman (1969) who noted that the pH-optimum of PPO activity varies with the sources of the enzyme and the nature of the substrate in most cases, it is in the pH range 6-7; the enzyme is most inactive below pH 4. Hence, the role of acidulants is to maintain the pH well below that necessary for optimal catalyte activity. Also the Hayakawa et al (1977) observed that in acid foods peroxidase is probably not the enzyme of choise. The effect of low pH of foods in destabilizing peroxidase is clearly within the normal processing range.
   b. The olive fruits during removal of bitterness and washing and also during fermentation and preservation did not come into contact with air because these stages during which this was possible, were performed quickly and carefully.
   c. In the stages of fermentation and preservation the concentration of the brine was approximately 8-10% and the environmental conditions where the fruits were kept were stable.

Thus, under those conditions the activation of two basic enzymes, responsible for the appearance of browning, mainly in the flesh of the fruits but also in their skin, were inhibited.

The same was concluded by the researchers Ben Shalon et al, (1977) who observed that in the industrial processing of green olives the fruits are rinsed in water after soaking in NaOH and are transfered to an acidic solution for fermentation.

Besides, the conservation of a stable level in the concentration of the brine (8-10%) during fermentation and preservation seems to help the preservation of the color and the texture of the fruits.

This can be explained by the fact that the stable concentration of the brine results in a stable internal pressure in the flesh of the olive fruits, thus preventing the interchange of salts or water concentration of the brine. (NaCl 8-10%) retains the internal pressure of the cells of the fruits and thus keeps their texture at good levels. NaCl brine was used to inhibit discoloration of sliced mushrooms in preparation for freeze drying (Farg et al, 1971).

The quality characteristics, of the level of the texture of the green olives and of the color of the flesh and skin are very important for the consumer.

To sum up, the inhibition or prevention from the enzyme action in this particular case (green olives) is achieved by avoiding the exposure to oxygen, combined with low pH values and concentration of brine. The inhibition of enzymatic browning is most frequently used in industry, including presently acids or brine.

ACKNOWLEDGMENTS

We are indebted to the Cooperative Union of Polygyros Chalkidiki, for the provision of raw material and the permission to use their facilities.

BIBLIOGRAPHY

Aylward, F. and Haisnan, D.R. (1969).- "Oxidation systems in fruits and vegetables-their relation to the quality of preserved products".- Adv. Food Res. Vol. 17 p. 1-76.

Ben-Shalon. N, Kahn. V, Harel. E, and Mayer, M.A. (1977).- "Olive Catechol Oxidase-Changes during development-".- J. Sci. Food Agric. **28**, 545-555.

Ben-Shalon. N, Harel. E and Mayer, M.A. (1978).- "Enzymatic Browning in green olives and its Preservation".- J. Sci. Food Agric. **29**, 398-402.

Ben-Shalon. N, Kahn. V, Harel, E and Mayer A.M. (1977).- "Chatechol Oxidase from green olive: Properties and partial purification".- Phytochemistry **16**,1153-1158.

Donaire. J.P, Sánchez. A.J, Lüpez-Goge, J. and Recalde, L. (1975).- "Metabolic changes in fruit and leaf during ripening in the olive".- Phytochemistry, **14**, 1167-1169.

Fang. T.T. and Footrankul, P and Luh, B.S. (1971).- "Effects of blanching chemical treatment and freezing methods on quality of freeze dried mushrooms".- J. Food Sci. **36**, 1044-48.

Hayakawa, K., Timlers, H., Katagiri, M. and Saito, Y. (1956).- "Enzymatic formation of oxolate from oxaloacetate".- J. Am. Chem. Sci. **78**, 5126-5127.

Kahn, V.J. (1977).- "Some biochemical properties of polyphenoloxidase from two varieties differing in their browning rates".- J. Food Sci. **42**, 38-43.

Maestro, R., García, J.M. and Castellano, J.M. (1993).- "Changes in polyphenol content of olives stored in modified atmospheres".- Hort-Science **28**, 749.

Mathews, A.G., Parpia, H.A.B. (1971).- "Food browning as a polyphenol reaction".- Adv. Food Res. Vol. 19 p. 75-132.

Luh, B.S. and Phithakpol, B. (1972).- "Characteristics of polyphenoloxidase related to browning in cling peaches".- J. Food Sci. **37**, 264-268.

Sciancalepore, V. and Longone, V. (1984).- "Polyphenol oxidase activity and browning in green olives".- J. Agr. Food Chem. **32**, 320-321

Sciancalepore, V. (1985).- "Enzymatic browning in five olive varieties".- J. Food Sci. **50**, 1194-1195.

Sharon-Raber, O and Kahn, V. (1983).- "Avocado mesocarp; browning potential, carotenoid content, polyphenol oxidase, catalase and peroxidase activities: Comparison between six avocado cultivars".- J. Food Sci. **48**, 1874-1875.

Recibido: Enero 1994
Aceptado: Mayo 1995