

The effect of rosemary essential oil on physico-chemical properties of extra-virgin olive oil stored in colourful bottles

F. Al Juhaimi¹, N. Uslu², M.M. Özcan^{2*}, K. Ghafoor¹ and E.E. Babiker¹

¹Department of Food Science & Nutrition, College of Food and Agricultural Sciences, King Saud University, P.O. Box 2460, 11451 Riyadh, Saudi Arabia; ²Department of Food Engineering, Faculty of Agriculture, Selcuk University, 42031 Konya, Turkey; mozcan@selcuk.edu.tr

Received: 10 June 2015 / Accepted: 27 July 2015

© 2015 Wageningen Academic Publishers

RESEARCH ARTICLE

Abstract

The aim of the study was to investigate the effect of both storage material colour (red, green, yellow and transparent glass bottle) and rosemary essential oil on the physico-chemical properties of extra-virgin olive oil. For this reason, free fatty acid, peroxide value, viscosity, colour and fatty acid composition of oils were measured at regular intervals. Free fatty acid and peroxide values of olive oils stored in different coloured bottles increased partly during storage. After 90 days of storage, free fatty acid values of samples changed between 0.78 and 0.89 mg KOH/g oil. By the 90th day of storage peroxide values of samples had changed from 32.75 to 79.46 meq O₂/kg oil, whereas the peroxide value of the control group on the 90th day was 94.55 meq O₂/kg. On the first day (0 day), 'L*', 'a*' and 'b*' values of control groups were determined to be 70.81, -3.69 and 38.26, respectively. During storage, these values partly increased. Linoleic acid (40.95-43.92%), oleic acid (33.04-34.99%) and palmitic acid (12.38-13.58%) were the major fatty acids of olive oils. In view of the analysis, the best results were obtained from oil stored in a green bottle. The addition of rosemary essential oil reduced lipid oxidation and showed an antioxidant effect when compared with the control group.

Keywords: essential oil, glass bottle, oxidation, storage

1. Introduction

Olive oil is one of the few edible oils consumed in the natural state due to its nutritional properties, health value, and pleasant flavour. The high levels of monounsaturated fatty acid, minor nutraceutical components and its sensory properties constitute the nutritional value of olive oil (Angerosa, 2002). Edible oils deteriorate easily because of some chemical reactions caused by the effect of heat, light, enzyme and metal ions (Bandoniene *et al.*, 2002). Lipid oxidation is the most significant reaction that reduces the shelf life of oils (Kristott, 2000). In addition, lipid oxidation causes rancid odours, off-flavours, discolouration and a decrease in nutritional quality (Kathirvel and Rupasinghe, 2011). As a result of the oxidation process, breakdown products such as peroxides, aldehydes and ketons form, making the oil unacceptable for consumption (Özcan and Arslan, 2011). One of the ways of reducing lipid oxidation

is to add antioxidants (Karpinska *et al.*, 2001). Sources of natural antioxidants are spices, herbs, teas, oils, seeds, cereals, fruits and vegetables. The maximum antioxidant capacity is provided by herbs and spices (Inanç and Maskan, 2012). Essential oils are also used as natural antioxidants for slowing down the oxidation process and preserving the polyunsaturated oils (Karpinska *et al.*, 2001). There is a great interest in obtaining and using the natural antioxidants because they are safe, are extracted from natural resources and have functional and sensory properties (Olmedo *et al.*, 2009). The purpose of this study was to investigate the effect of the storage materials (red, green, yellow and colourless glass bottle) and rosemary essential oil on the physico-chemical properties of extra-virgin olive oil stored in several coloured bottles.

2. Materials and methods

Materials

Extra-virgin olive oil was obtained from Mut, district of Mersin in Turkey. The dried leaves of rosemary (*Rosmarinus officinalis* L.) were purchased from a local market in Konya. All reagents and solvents were analytical grade and obtained from Sigma-Aldrich Co. (St. Louis, MO, USA).

Methods

Extraction of essential oil

Dried rosemary leaves (about 300 g) were ground into small pieces using a blender. Ground samples were placed in a flask (2 l) with deionised water (1.5 l). The mixture was hydrodistilled for 4 h in a Clevenger-type apparatus (İldam, Ankara, Turkey). The essential oil was kept in a freezer at -18 °C.

Experimental design

Glass bottles (1 l) in four different colours (red, green, yellow and transparent) were filled with 500 ml extra-virgin olive oil. Rosemary essential oil was added at 0.03% (w/w). A control sample was prepared without added antioxidant under the same conditions in a transparent bottle. The bottled oils were thoroughly mixed and located in a place exposed to sunlight. Samples were stored for 3 months at room temperature and analysed on day 0, 30, 60 and 90 after bottling to determine the physico-chemical properties (colour, viscosity, fatty acid composition, free fatty acid and peroxide values).

Colour analyses

Colour values were measured with a Minolta Chroma meter CR 400 (Konica Minolta, Inc. Osaka, Japan) according to the International Commission on Illumination L*, a*, b* scale (Rizzo *et al.*, 2014).

Viscosity analyses

Viscosity values of samples were measured using a viscometer (SV-10; AND, Tokyo, Japan) and denominated mPa·s (Akbulut *et al.*, 2009).

Analysis of free fatty acid and peroxide value

Free fatty acids and peroxide values were determined using the standard AOCS (2009) method Ca 5a-40.

Fatty acid composition

Olive oil was esterificated according to the ISO-5509 (ISO, 1978) method with some modifications. Fatty acid methyl esters of samples were analysed by gas chromatography (GC-2010; Shimadzu, Tokyo, Japan) equipped with a flame-ionisation detector and capillary column (TR-CN100, 60 m × 0.25 mm, film thickness 0.20 µm; Tecnocroma, Barcelona, Spain). The temperature of the injection block and detector was 260 °C. The mobile phase was nitrogen with 1.51 ml/min flow rate. Total flow rate was 80 ml/min and split rate was also 1/40. Column temperature was programmed at 120 °C for 5 min and increased to 240 °C at a rate of 4 °C/min and held for 25 min at 240 °C. A standard fatty acid methyl ester mixture (Sigma-Aldrich Co.) was used to determine sample peaks. Commercial mixtures of fatty acid methyl esters were used as reference data for the relative retention times (AOAC, 1990).

3. Results and discussion

The free fatty acid values of virgin olive oils (added essential oil and control samples) stored in glass bottles of different colours (red, green, yellow and transparent) for a period of 90 days are given in Table 1. Free fatty acid and peroxide values of olive oils stored in different coloured bottles increased partly during storage. At 90 days of storage, free fatty acid values of samples changed between 0.78 to 0.89 mg KOH/g oil. Free fatty acid values of the control group ranged from 0.39 to 1.22 mg KOH/g oil during storage. Generally, at 90 days of storage of samples, free fatty acid values of samples were found to be lower than the control group. As the time increased from 0 to 90 days, an increase in acidity was determined in all of the storage oils, which was in accordance with the results of Asensio *et al.* (2011) and Rababah *et al.* (2011). As a result of the hydrolysis process, free fatty acids, glycerol, mono- and di-glycerides formed, therefore the deterioration of lipid was observed due to the development of rancidity (Gertz, 1996). During the storage time, the maximum increase was registered in the control sample (1.22 mg KOH/g oil) without rosemary essential oil. It was ascertained that rosemary essential oil (0.3%) showed an antioxidant effect in olive oil. In the experiments reported by Terpinc *et al.* (2009) and Visentin *et al.* (2011), carnosic acid is the most effective antioxidant component in rosemary extracts. From the standpoint of the bottle colour, the minimum increase in the acidity values was found in oil stored in green (0.78 mg KOH/g oil) and yellow (0.79 mg KOH/g oil) bottles. Values of the control sample (added essential oil) and oil stored in red bottle were closest for the last month, while the highest acidity for the first and second month was observed in the red bottle. Rababah *et al.* (2011) studied olive oil stored in a clear glass bottle for two months at 25 °C; the increase in acidity values was 0.04% for the first month and 0.28% for second month. When the glass bottle was compared

with tinplate and plastic bottle, oils in the glass container exhibited the lowest acidity throughout the storage period. According to the study of Chen *et al.* (2014), the inhibitory effect of rosemary essential oil on free fatty acids was higher than butylated hydroxytoluene, but lower than butylated hydroxyanisole and tert-butylhydroquinone.

The peroxide values of virgin olive oils were shown to increase during storage time as illustrated in Table 1. Peroxide values of samples at 90 days of storage had changed from 32.75 to 79.46 meq O₂/kg oil, whereas the peroxide value of the control group at 90 days was 94.55 meq O₂/kg. So, peroxide values of samples were found to be lower than the control group. Also, it can be said that rosemary essential oil affected the stability of olive oils stored in several coloured bottles for 90 days. The presence of oxygen in the headspace of the bottle or dissolution of the oxygen in the oil causes a reaction with unsaturated fatty acids and consequently consists of hydroperoxides and peroxides (Savarese *et al.* 2013). High temperature and light lead to the formation of peroxides (Gharby *et al.*, 2011). Olive oil without added essential oil and stored in a transparent bottle showed the maximum increase in peroxide value. It was indicated that rosemary essential oil had an antioxidant effect on olive oil. When comparing the increase in peroxide values with regard to the colour of bottle, the best results were obtained from olive oil stored in a green bottle. Peroxide value was found according to decreasing orders in oils in transparent (without essential oil) > yellow > transparent (with essential oil) > red > green bottles. Rizzo

et al. (2014) investigated quality changes in extra virgin olive oil packaged in coloured (clear, green, orange, white and blue) polyethylene terephthalate (PET) bottles during storage period. The lowest peroxide values were observed for oils stored in blue, white and green bottles. According to Rababah *et al.* (2011) the best protection was provided with the glass bottle, followed by plastic and tinplate containers. Savarese *et al.* (2013) reported that red bottles did not show sufficient protection against light compared to transparent PET bottles. In addition, Makni *et al.* (2011) stated that the best bottles for packing of oils were opaque glass bottles including antioxidant additives.

Colour values of olive samples stored in several bottles are given in Table 2. On the first day (0 day), L*, a* and b* values of control groups were determined to be 70.81, -3.69 and 38.26, respectively. During storage, these values partly increased. According to the values, green bottles can be recommended for storage. Also, rosemary essential oil protected against colour change during storage. In general, L* values of oils increased, while a* and b* values decreased with the effect of storage time and colour of bottle. Reduction in the pigment content (chlorophyll and carotenoid) caused a change in the colours of oils during the storage time. Rizzo *et al.* (2014) determined that there was not a significant change because of bottle colours and storage conditions.

The fatty acid compositions of virgin olive oils stored in colourful glass bottles are shown in Table 3. According to

Table 1. Free fatty acid and peroxide values of olive oils.

Treatments	Free fatty acid values (mg KOH/g oil)				Peroxide values (meq O ₂ /kg oil)			
	0	30	60	90	0	30	60	90
Control (without essential oil)	0.39	0.77	0.77	1.22	10.59	20.96	48.76	94.55
Control (essential oil added)		0.77	0.75	0.89		19.36	37.16	65.85
Stored in red bottle		0.83	0.85	0.89		19.66	23.19	63.95
Stored in green bottle		0.73	0.79	0.78		15.93	23.76	37.25
Stored in yellow bottle		0.75	0.77	0.79		18.03	30.22	79.46

Table 2. Colour values of olive oils.

Treatments	Day 0			Day 30			Day 60			Day 90		
	L*	a*	b*	L*	a*	b*	L*	a*	b*	L*	a*	b*
Control (without essential oil)	70.81	-3.69	38.26	81.59	-3.39	24.31	83.89	-2.97	19.24	86.16	-2.27	8.70
Control (essential oil added)				80.78	-3.66	23.09	89.75	-1.29	7.55	83.59	-2.79	16.01
Stored in red bottle				78.36	-4.25	25.48	70.36	-3.44	31.73	81.75	-3.27	22.78
Stored in green bottle				78.76	-3.69	7.44	88.09	-2.96	4.51	87.08	-2.22	1.60
Stored in yellow bottle				61.33	-4.63	26.34	84.25	-3.37	13.86	86.06	-2.18	9.28

Table 3. Fatty acid compositions of olive oils stored in different coloured glass bottles.¹

Fatty acids	Fatty acid composition (%)														
	day 0					day 30					day 60				
	C-1	C-2	Red	Green	Yellow	C-1	C-2	Red	Green	Yellow	C-1	C-2	Red	Green	Yellow
Myristic acid	0.27	0.26	0.23	0.27	0.28	0.28	0.27	0.27	0.27	0.26	0.28	0.27	0.28	0.27	0.28
Palmitic acid	13.58	13.18	12.38	12.97	13.49	13.53	13.45	13.47	13.19	13.33	13.51	13.30	13.40	13.35	13.51
Stearic acid	1.98	1.94	1.94	1.91	1.95	1.97	1.95	1.93	1.97	1.97	1.95	1.97	1.99	1.98	1.96
Oleic acid	34.99	34.06	34.60	33.04	33.87	33.71	34.51	34.04	34.38	34.16	34.32	34.36	34.55	34.80	34.61
Linoleic acid	43.92	43.37	40.95	43.23	43.16	43.42	43.74	43.46	43.04	43.22	43.81	43.90	43.80	43.65	43.57
Arachidic acid	0.36	0.23	0.21	0.38	0.36	0.36	0.36	0.36	0.37	0.35	0.34	0.37	0.38	0.36	0.36
γ-Linolenic acid	0.22	0.23	0.20	0.34	0.27	0.22	0.22	0.23	0.22	0.23	0.23	0.23	0.23	0.22	0.23
Linolenic acid	2.46	2.44	2.27	2.50	2.47	2.43	2.41	2.39	2.44	2.44	2.42	2.45	2.47	2.44	2.43
Behenic acid	0.19	0.21	0.20	0.13	0.18	0.18	0.18	0.16	0.18	0.18	0.16	0.17	0.18	0.18	0.19

¹ C-1 = control sample stored in transparent bottle without essential oil; C-2 = control sample stored in transparent bottle with added essential oil.

the results, rosemary essential oil did not effect the fatty acid composition of the samples. Generally, all fatty acid levels in samples were similar. While oleic acid content change between 33.04 (red bottle for 30 days) and 34.99% (for 0 days), linoleic acid contents ranged from 40.95 to 43.80% (control sample stored in a transparent bottle with added essential oil for 30 or 90 days, respectively). Linoleic acid (40.95-43.92%), oleic acid (33.04-34.99%) and palmitic acid (12.38-13.58%) were the major fatty acids of olive oils. Levels of oleic and linoleic acids decreased while a slight increase in palmitic acid content was observed. The reason for this situation was the degradation of double bonds in unsaturated fatty acids because of oxidation. But the levels of phenolic compounds and antioxidants in virgin olive oil provide the high oxidative stability. Therefore, fatty acid compositions of virgin olive oils did not alter dramatically.

Viscosity values of virgin olive oils are given in Table 4. Viscosity values of olive samples showed differences during storage. On the first day (0 day), the viscosity value was 47.67 mPa.s. While viscosity values of olive oils stored in red, green and yellow bottles at 30 days of storage are 42.27, 60.00 and 63.93 mPa.s, these values were determined to be 32.17, 36.43 and 35.63 mPa.s, respectively. These differences are probably due to a reaction between essential oil, measurement temperature and polymerisation. While viscosity values of all samples increased at 60 days of storage, these values partly decreased at 90 days of storage. But essential oil affected the viscosity values of oil samples, and protected the viscosity of oil samples.

Table 4. Viscosity values of olive oils.

Treatments	Viscosity (mPa.s)			
	Time (days)			
	0	30	60	90
Control (without essential oil)	47.67	46.30	56.03	39.80
Control (essential oil added)		48.97	57.90	47.07
Stored in red bottle		42.27	54.83	32.17
Stored in green bottle		60.00	62.07	36.43
Stored in yellow bottle		63.93	57.03	35.63

Acknowledgements

The authors would like to extend their sincere appreciation to the Deanship of Scientific Research at King Saud University for funding this research group (no. RG-1435-049).

References

- Akbulut, M., Özcan, M.M. and Çoklar, H., 2009. Evaluation of antioxidant activity, phenolic, mineral contents and some physico-chemical properties of several pine honeys collected from Western Anatolia. *International Journal of Food Science and Nutrition* 60: 577-589.
- American Oil Chemists' Society (AOCS), 2009. Official methods and recommended practices of the American Oil Chemists' Society (5th Ed.). AOCS, Champaign, IL, USA.
- Angerosa, F., 2002. Influence of volatile compounds on virgin olive oil quality evaluated by analytical approaches and sensory panels. *European Journal of Lipid Science and Technology* 104: 639-660.
- Asensio, C.M., Nepote, V. and Grosso, N.R., 2011. Chemical stability of extra-virgin olive oil added with oregano essential oil. *Journal of Food Science* 76: S445-S450.
- Association of Official Analytical Chemists (AOAC), 1990. Official methods of analysis (15th Ed.). AOAC, Washington, DC, USA.
- Bandoniene, D., Venskutonis, P., Gruzdienė, D. and Murkovic, M., 2002. Antioxidative activity of sage (*Salvia officinalis* L.), savory (*Satureja hortensis* L.) and borage (*Borago officinalis* L.) extracts in rapeseed oil. *European Journal of Lipid Science and Technology* 104: 286-292.
- Chen, W., Zhang, Y., Zu, Y., Yang, L., Lu, Q. and Wang, W., 2014. Antioxidant effects of rosemary extracts on sunflower oil compared with synthetic antioxidants. *International Journal of Food Science and Technology* 49: 385-391.
- Gertz, C., 1996. Chemical changes of oils and fats at elevated temperatures. In: Bell, B.M. (ed.) *Fat in the diet*. Barnes and Assoc. Inc., Bridgwater, UK, pp. 15-21.
- Gharby, S., Harhar, H., Guillaume, D., Haddad, A., Matthaus, B. and Charrouf, Z., 2011. Oxidative stability of edible argan oil: a two-year study. *LWT-Food Science and Technology* 44: 1-8.
- Inanç, T. and Maskan, M., 2012. The potential application of plant essential oils/extracts as natural preservatives in oils during processing: a review. *Journal of Food Science and Engineering* 2: 1-9.
- International Organization for Standardization (ISO), 1978. Animal and vegetable fats and oils preparation of methyl esters of fatty acids. Method ISO 5509. ISO, Genève, Switzerland.
- Karpinska, M., Borowski, J. and Danowska-Oziewicz, M., 2001. The use of natural antioxidants in ready-to-serve food. *Food Chemistry* 72: 5-9.
- Kathirvel, P. and Rupasinghe, H.P.V., 2011. Plant-derived antioxidants as potential omega-3 PUFA stabilizers. In: Dijk, M.V. and Vitek, J. (eds.) *Fish oil: production, consumption and health benefits*. Nova Science Publishers, Inc., Hauppauge, NY, USA, pp. 158-185.
- Kristott, J., 2000. Fats and oils. In: Kilcast, D. and Subramaniam, P. (eds.) *The stability and shelf life of food*. CRC Press, Boca Raton, FL, USA, pp. 279-309.
- Makni, M., Haddar, A., Fraj, A. and Zeghal, N., 2011. Physico-chemical properties, composition, and oxidative stability of olive and soybean oils under different conditions. *International Journal of Food Properties* 18: 194-204.
- Olmedo, R.H., Asensio, C.M., Nepote, V., Mestrallet, M.G. and Grosso, N.R., 2009. Chemical and sensory stability of fried-salted peanuts flavored with oregano essential oil and olive oil. *Journal of the Science and Food Agriculture* 89: 2128-2136.
- Özcan, M.M. and Arslan, D., 2011. Antioxidant effect of essential oils of rosemary, clove and cinnamon on hazelnut and poppy oils. *Food Chemistry* 129: 171-174.
- Rababah, T.M., Feng, H., Yang, W., Eriefej, K. and Al-Omouh, M., 2011. Effects of type of packaging material on physicochemical and sensory properties of olive oil. *International Journal of Agriculture and Biology Engineering* 4: 66-72.
- Rizzo, V., Torri, L., Licciardello, F., Piergiovanni, L. and Muratore, G., 2014. Quality changes of extra virgin olive oil packaged in coloured polyethylene terephthalate bottles stored under different lighting conditions. *Packaging Technology Science* 27: 437-448.
- Savarese, M., De Marco, E., Caporaso, N. and Sacchi, R., 2013. Extra virgin olive oil overall quality assessment during prolonged storage in PET containers. *Global Virtual Conference* 1: 674-679.
- Terpinc, P., Bezjak, M. and Abramovic, H., 2009. A kinetic model for evaluation of the antioxidant activity of several rosemary extracts. *Food Chemistry* 115: 740-744.
- Visentin, A., Cismondi, M. and Maestri, D., 2011. Supercritical CO₂ fractionation of rosemary ethanolic oleoresins as a method to improve carnosic acid recovery. *Innovative Food Science and Emerging Technologies* 12: 142-145.

