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Oya KÖSEOĞLU¹
Didar SEVİM¹
Mehmet ULAŞ¹
Durmuş ÖZDEMİR²

Determination of Bitterness Index (K_{225}) and Total Fenol Content of Olive Oils Obtained With Different Regions, Varieties and Processing Systems

Farklı Bölge, Çeşit ve Üretim Sistemleri ile Elde Edilen Zeytinyağlarının Acılık İndekslerinin ve Toplam Fenol Değerlerinin Belirlenmesi

¹ Ministry of Food, Agriculture and Livestock Directorship of Olive Research Institute, 35100, İzmir/Turkey
² İzmir Institute of Technology, Faculty of Science Department of Chemistry, 35437, İzmir/Turkey
corresponding author: dcengeler@gmail.com

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Key Words:

K_{225} value, extraction systems, Turkish olive varieties, geographical regions

ABSTRACT

In this work the effect of different growing areas on olive (Ayvalık, Memecik, Gemlik, Beylik, Edincik Su, Girit, Kilis Yağlık, Sarı Ulak, Tavşan Yüreği, Topak Aşı) oil bitterness index (K_{225}) were studied at the South Marmara, South and North Aegean, West and East Mediterranean Regions at two, two and a half (2.5), and three phase extraction system, during 2014/2015 crop season. A total of 41 virgin olive oils samples were collected from these Regions. Total phenol content and bitternes index (K_{225}) were analyzed in the research. A Solid-Phase Extraction procedure were carried out for extraction of the bitter compounds. The results of total phenol content and K_{225} values showed that the Beylik olive oil was determined with the highest total phenol content and bitterness index (K_{225}) with 330.26 mg Caffeic Acid Equivalents (CAE) kg^{-1} oil and 1.21 at 2.5 phase extraction system from Manavgat at the West Mediterranean Region, respectively. After the Beylik variety, the highest total phenol content was determined Ayvalık and Edincik Su olive oil with 291.03 and 270.62 mg CAE kg^{-1} oil, respectively. The Memecik and Ayvalık olive oil bitterness index (K_{225}) was determined 0.86 and 0.85 at two phase extraction system from Muğla and Burhaniye at the South and North Aegean, respectively.

Anahtar Sözcükler:

K_{225} değeri, ekstraksiyon sistemleri, Türk zeytin çeşitleri, coğrafik bölgeler

ÖZET

Bu çalışmada, zeytinyağlarının acılık indeksi (K_{225}) değeri üzerine yetiştirilme bölgelerinin etkisi incelenmiştir. Bu amaçla, 2014/2015 hasat yılında Güney Marmara, Güney ve Kuzey Ege ile Batı ve Doğu Akdeniz Bölgelerinde yetişen zeytinlerden (Ayvalık, Memecik, Gemlik, Beylik, Edincik Su, Girit, Kilis Yağlık, Sarı Ulak, Tavşan Yüreği, Topak Aşı) iki, iki buçuk ve üç fazlı ekstraksiyon sistemi elde edilen, toplam 41 zeytinyağı örneği toplanmıştır. Çalışmada toplam fenol içeriği ve acılık indeksi analizleri yapılmıştır. Acılık bileşenlerinin ekstraksiyonu için katı faz ekstraksiyon sistemi uygulanmıştır. Toplam fenol içeriği ve acılık indeksi (K_{225}) değeri sonuçlarına bakıldığında, Batı Akdeniz Bölgesindeki Manavgat ilçesinden iki buçuk faz ekstraksiyon sistemi ile elde edilen Beylik zeytinyağının 330.26 mg CAE kg^{-1} yağ ile en yüksek toplam fenol ve 1.21 ile en yüksek acılık indeksi değerini gösterdiği tespit edilmiştir. Beylik çeşidinden sonra, en yüksek toplam fenol içeriği 291.03 mg CAE kg^{-1} yağ ile Ayvalık ve 270.62 mg CAE kg^{-1} yağ ile Edincik Su zeytinyağında tespit edilmiştir. İki fazlı ekstraksiyon sistemin ile Güney Ege'de Muğla'dan elde edilen Memecik ve Kuzey Ege'de Burhaniye'den elde edilen Ayvalık zeytinyağlarının acılık indeksleri değerleri sırasıyla 0.86 ve 0.85 olarak tespit edilmiştir.

INTRODUCTION

The olive tree (*Oleo europaea*) is widely cultivated for the production of both olive oil and table olive and they are significantly economic importance for countries. Olive oil which is obtained from the fruit using

only mechanical and physical processes is ready for human consumption and possess unique sensory characteristics and nutritional properties (Inarejos-Garcia et al., 2009). Virgin olive oil is unique among the other vegetable oils because of its high level of

particular phenolic compounds, to which, together with the high content of unsaturated fatty acids, the health benefits of virgin olive oil are attributed (Visioli and Galli, 1998). In olive oils, minor components, especially the phenolic contents are affected by cultivar, climatic and environmental conditions, agronomic practices, maturity index and extraction systems (Sevim et al., 2013; Condelli et al., 2015). Giovacchino et al. (2002) reported that quality parameters of olive oil did not significantly change during the extraction of the fine olives with the two-phase and three-phase centrifugal decanter, while the olive oils obtained with the triple-phase centrifugal decanter had lower total phenol and o-diphenol content than the two-phase centrifugal decanter. The decreases in phenol compounds explained by their water-solubility. Higher water/paste ratios are used in three-phase centrifugation, and therefore larger amounts of phenols are eliminated with water wastes. The three-phase system decanter separates the paste into a relatively dry solid, fruit-water, and oil. Water is added to this system to get it to flow through the decanter (Salvador et al., 2003). For that reason a minimum quantity of water can be added to separate the solid material better and to retain water-soluble phenol compounds as much as possible. Depending on the type of phenols present the intensity of bitterness of olive oils can have high variations (high or low) (Favati et al., 2013). It is generally accepted that the phenolic fraction, secoridoid derivatives such as oleuropein and ligstroside derivatives, of olive oil are mainly responsible for the bitter taste (Morello et al., 2004; Favati et al., 2013). Due to the positive contribution of phenolic compounds to human health, consumers are increasing their consumption of oils with high bitterness attribute (Inarejos-Garcia et al., 2009). As a result, bitterness index (K_{225}) is becoming an important area in olive oil research (Favati et al., 2013). Oil bitterness intensity can be measured with a simple analytical method using spectrophotometric determination at 225 nm (K_{225}) of extraction of bitter compounds.

Out of the many agricultural products of Turkey, olive oil has a remarkable place. In recent years, as a result of confirmation of positive effects of olive and olive oil on human health and nutrition by scientific studies, olive growing has gained a new acceleration in Turkey besides throughout the world. As a result of this trend, new olive orchards have been established. The olive growing regions in Turkey are the South Marmara, South and North Aegean, West and East Mediterranean where Ayvalık, Memecik, Gemlik, Beylik, Edincik Su, Girit, Kilis Yağlık, Sarı Ulak, Tavşan Yüreği, Topak Aşı are the

main cultivars. According to local characteristics the olive harvest is starting in October and continuing until the end of January in Turkey (Kutlu and Şen, 2011).

The aims of this work were to determine the effect of growing regions and processing systems on the total phenol content and bitterness index (K_{225}) of Turkish virgin olive oils. For this purpose, a total of forty-one virgin olive oils samples which extracted with two, two and a half, and three phase system were collected from these Regions, during 2014/2015 crop season. A total phenol content were determined with the Folin-Ciocalteu method and Solid-Phase Extraction (SPE) procedure were carried out for extraction of the bitter compounds (K_{225}).

MATERIAL and METHODS

Materials

Sampling of Extra Virgin Olive Oils

As seen from Table 1 forty one (41) extra virgin olive oils collected from the South Marmara (7), South (8) and North Aegean (15), West (4) and East Mediterranean (7) at 2, 2.5 and 3 phase extraction system, during 2014/2015 crop season. The South Marmara Region contains Gemlik and Edincik Su olive oils, were collected from district of Mudanya, Erdek, Edincik, Osmangazi, İznik. Ayvalık olive oil was collected from the North Aegean Region. This cultivar is grown in Edremit, Ayvalık, Karasınan, Zeytindağ, Küçükkuşu, Ezine, Altınova, Burhaniye and Havran. Memecik olive oil was collected from the South Aegean Region. It is grown at provinces of Aydın, İzmir and Muğla. The East Mediterranean Region contains the Topak Aşı, Sarı Ulak, Ayvalık, Gemlik and Kilis Yağlık olive oils were collected from the city of Tarsus, Adana and Hatay. Beylik, Girit, Gemlik and Tavşan Yüreği olive oils were collected at the locations of Manavgat and Gazipaşa at the West Mediterranean Region.

All oil samples were extracted between October, November and December under industrial conditions in a olive plant in Turkey. Samples were removed from each of three bottles from the same extraction for each samples of olive oils. Each oil samples (contains 500 mL) stored in the dark glass bottles and at +4°C until they were analyzed.

Determination of Total Phenol Content

Analysis of total phenol content of olive oil was determined spectrophotometrically according to the Folin-Ciocalteu method previously described by Gutfinger (1981) and Hrnčirik and Fritsche (2004). 2.5 g of oil sample was dissolved in 5 ml of hexane and after adding 5 ml methanol/water (60:40 v/v) the solution

was shaken for 2 min. The extraction were separated from each other by centrifuging the solution at 3500 rpm for 10 min. 0.2 ml of methanolic phase was put into flask and completed with deionized water to 5 ml, then Folin–Ciocalteu reagent (0.5 ml) was added to the mixture. After 3 min, 1 ml of Na_2CO_3 solution (35%, w/v) was added and diluted to 10 ml with pure water. The solution was incubated for 2 h in a dark place and the absorbance of the solution was read at 725 nm with a spectrophotometer (Shimadzu UV-1700, Japan). The total phenol concentration was calculated from caffeic acid calibration curve. Data was expressed as mg equivalent of caffeic acid per kilogram of oil (mg CAE kg^{-1}).

Determination of Bitterness Index (K_{225})

The compounds responsible for oil bitterness were evaluated spectrophotometrically at 225 nm as absorbance (K_{225} values) with a Shimadzu spectrophotometer UV-1700 PharmaSpec (Japan) according to the Gutierrez et al. (1992). A Solid-Phase Extraction (SPE) procedure were carried out for extraction of the bitter compounds. A sample of 1.0 ± 0.01 g of oil was dissolved in 4.0 mL hexan and passed over a C18 column (Bakerdond spe Columns, J.T. Baker, Phillipsburg, NJ, Holland) previously activated with methanol (6.0 mL) and washed with hexane (6.0 mL). After elution, 10.0 mL hexane was passed to eliminate the fat, and the retained compounds were eluted with methanol/water (1:1) to 25.0 mL in a flask. The absorbance of the extract was measured at 225 nm against methanol/water (1:1) in a 1.0 cm cuvette. Results were expressed as the absorbance of 1.0 g in 100 g (K_{225} values).

Statistical analysis

Data analysis was performed using MINITAB statistical software package version 15. One way analysis of variance (ANOVA) were performed based on the geographical regions of the olive oil samples for both total phenol and bitterness index results. Following ANOVA, Fisher's least significant difference test method (LSD) were used for the pairwise comparison of the five different geographical regions. In addition, simple least squares and multiple regression analysis were also carried out with the same software package where the two and three dimensional scatter plots of bitterness index (K_{225}) were generated with the same software package.

RESULTS and DISCUSSION

Phenolic compounds are the minor compounds in olive oils with high antioxidant activity providing nutritional and sensorial properties. Bitterness is a considered as a positive sensorial attribute of the

virgin olive oils and enhances the whole flavour with related to green olive fruit. Consumers are increasing their consumption of olive oils with high bitterness intensity. As a result of this, bitterness assessment is becoming an important subject in olive oil research (Escuderos et al., 2014).

As can be seen from Table 1, there are five different geographical regions where olive oils were collected which were processed with two and three-phases systems along with two other samples processed with 2.5 phases system coming from East Mediterranean (Manavgat). Beylik olive oil was determined the highest total phenol content ($330.26 \text{ mg CAE kg}^{-1}$ oil) and bitterness index (K_{225}) (1.21) that extracted with 2.5 phase centrifugal system at the West Mediterranean Region. Memecik and Ayvalık olive oils bitterness index (K_{225}) which were extracted with two phase system at South Aegean Region and North Aegean Region, were followed the Beylik olive oil bitter index with 0.86 and 0.85, respectively (Table 1). Total phenol content of Ayvalık and Edincik Su were determined 291.03 and 270.62 mg CAE kg^{-1} oil, respectively. The lowest total phenol content and bitterness index was determined with 12.41 mg CAE kg^{-1} oil and 0.13 on Sarı Ulak and Ayvalık cultivar extracted with three phase system in Tarsus and Edremit at the East Mediterranean and the North Aegean Region, respectively.

According to Aguilera et al. (2005) location do not play an important role on the oil bitterness index (K_{225}) for Frantoio and Lecciono cultivars. A significant effect was reported of olive cultivar and harvest time on the bitterness intensity (spectrophotometric) by some authors (Skevin et al., 2003; Morello et al., 2004; Ilyasoglu et al., 2010; Rotondi et al., 2010; Favati et al., 2013; Condelli et al., 2015). There is no limit set for the bitterness index value at National or International standards. Consumers only refuse or consume the oil products according to their preference. Some consumers prefer to consume olive oils of high bitterness index value, while others prefer to consume olive oils of low bitterness index value. Gutierrez et al. (1992), have reported that K_{225} value ≥ 0.360 correspond to quite bitter olive oils that some consumers are do not choose to consume of these oils. For that all, due to the positive attribution of phenolic compounds to human health, some consumers are increasing their consumption of oils with high bitterness index value (Inarejos-Garcia et al., 2009). It was seen in the study that the K_{225} value of 27 olive oils samples is above 0.360 value. The results of South Aegean Region samples of K_{225} value was a quite higher than this value. The West Mediterranean and the East Mediterranean Regions were followed the South Aegean Region with 0.52 and

0.45, respectively. As reported by authors (Köseoğlu and Unal, 2008; İlyasoğlu et al., 2010, Köseoğlu et al., 2016) different factors, such as olive cultivar, climatic conditions, maturity index, technological processing of oil and harvesting time had a significantly influence on the level of total phenols and also the intensity of

bitterness. The highest total phenol content was determined at the South Marmara Region samples with 200.28 mg CAE kg⁻¹ oil. The South Aegean Region and North Aegean Region olive oil total phenol contents were followed the South Marmara Region with 148.92 and 136.64 mg CAE kg⁻¹ oil (Table1).

Table 1. Total phenol content and bitterness value of olive oil samples according to geographical region and cultivar

Region Name	Subregion Name	Processing Name	Cultivar Name	Total Phenol (mg CAE kg ⁻¹)	Bitterness index (K ₂₂₅)
South Marmara	Mudanya	Three-phases	Gemlik	222.18	0.20
	Erdek		Edincik Su	270.62	0.62
	Edincik		Edincik Su	226.98	0.49
	Erdek		Edincik Su	207.37	0.48
	Orhangazi		Gemlik	94.08	0.23
	İznic		Gemlik	197.36	0.31
	Edincik		Edincik Su	183.35	0.45
Mean				200.28	0.40
North Aegean	Zeytindağ	Two-phases	Ayvalık	126.10	0.29
	Karasinan		Ayvalık	133.71	0.32
	Zeytindağ		Ayvalık	152.52	0.47
	Burhaniye		Ayvalık	291.03	0.85
	Küçükkuyu		Ayvalık	106.08	0.45
	Edremit	Three-phases	Ayvalık	76.86	0.29
	Edremit		Ayvalık	42.83	0.13
	Küçükkuyu		Ayvalık	165.33	0.38
	Ezine		Ayvalık	133.31	0.43
	Altınova		Ayvalık	79.26	0.28
	Havran		Ayvalık	46.84	0.22
	Edremit		Ayvalık	148.12	0.45
	Ezine		Ayvalık	205.76	0.64
	Ayvalık		Ayvalık	184.55	0.49
	Ayvalık		Ayvalık	157.33	0.45
Mean				136.64	0.41
South Aegean	İzmir	Two-phases	Memecik	222.18	0.78
	Aydın		Memecik	139.71	0.74
	Aydın		Memecik	112.89	0.46
	Muğla		Memecik	135.71	0.86
	Aydın	Three-phases	Memecik	155.32	0.76
	Aydın		Memecik	161.73	0.83
	İzmir		Memecik	184.15	0.77
	Aydın		Memecik	79.66	0.48
Mean				148.92	0.71
East Mediterranean	Tarsus	Two-phases	Ayvalık	178.14	0.75
	Tarsus		Gemlik	90.87	0.20
	Adana		Gemlik	134.11	0.52
	Tarsus	Three-phases	Topak Aşı	15.61	0.18
	Tarsus		Sarı Ulak	12.41	0.20
	Hatay		Kilis Yağlık	192.55	0.72
	Hatay		Kilis Yağlık	145.32	0.59
Mean				109.86	0.45
West Mediterranean	Manavgat	2.5-phases	Beylik	330.26	1.21
	Manavgat		Girit	37.63	0.28
	Gazipaşa	Three-phases	Gemlik	49.64	0.30
	Gazipaşa		Tavşan Yüreği	85.67	0.30
Mean				125.80	0.52

Inarejos-Garcia et al. (2009) reported that Cornicabra olive oil K₂₂₅ values was ranged from 0.47 to 0.52 extracted at 28°C, 60 min with two phase extraction

system, Condelli et al. (2015) observed Maiatica, Coratina, O. Vulture, Leccino and O. Bradano olive cultivars K₂₂₅ values 0.12, 0.32, 0.24, 0.21 and 0.22,

respectively. Morello et al. (2004) determined Arbequina variety (harvested first week of November to second week of January) K_{225} values was ranged from 0.15 to 0.37, Favati et al. (2013) defined K_{225} values of Coratine, Ogliarola, Maiatica, Leccino and Blend range from 0.17 to 0.60, 0.15-0.45, 0.06-0.16, 0.07-0.32 and -0.06-0.35, respectively. Aguilera et al. (2005) observed Frantoio and Lecciono cultivars K_{225} values range 0.41-0.37 and 0.38- 0.43, respectively. In this study, K_{225} values of Turkish olive cultivars were ranged from 1.21 to 0.13. Some cultivars of the regions like Edincik Su, Ayvalık, Memecik, Kilis Yağlık and Beylik were determined more higher than Italian and Spanish varieties.

In the research it was determined that the total phenol content in Küçükuyu sample was low but the bitterness index value was high or vice versa. This may be due to that the proportion of the phenolic compounds causing the bitterness value is high or low in the oil samples. Phenolic acids, phenolic alcohols, flavonoids and hydroxy-isochromes are the phenolic compounds that are found in extra virgin olive oil. These compounds are play very important role in bitterness and pungency value of olive oils. Important phenolic compounds in olive oil are hydroxytyrosol, tyrosol and oleuropein. Their concentrations in the olive oil depending on the olive variety, the olive maturity, the time of harvest and the transportation methods and the processing technology (Boskou, 2009).

For a general evaluation of bitterness index (K_{225}) as function of total phenol content of the olive oil samples, it is reasonable to analyze the data with simple least squares (SLS) method. Figure 1 shows the scatter plot of total phenol content versus bitterness index (K_{225}) of 41 olive oil samples.

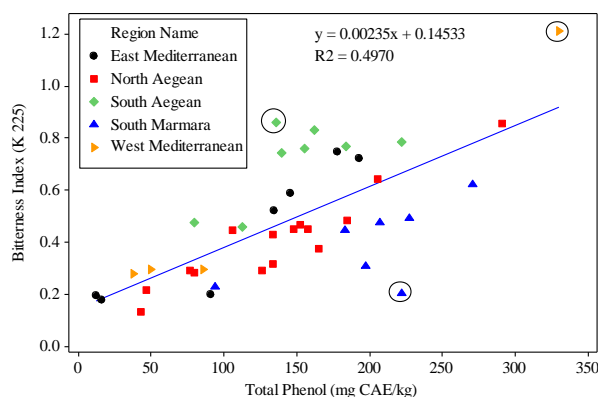


Figure 1. Simple least squares regression plot of total phenol content vs bitterness index (K_{225}) of olive oil samples collected from five different geographical region.

The regression coefficient of the least squares model indicates that there is a correlation between total phenol content and bitterness index as expected

but due to the the differences among the several geographic regions, this correlation gave an R^2 value of 0.497. On the other hand, there are a few samples which were labelled with a circle around them are causing significant deviations from the regression line and this is the another reason for low regression coefficient. It is expected that when these three sample were left out regression model a better regression models could be obtained. Figure 2 shows the scatter plot of this reduced data set when these three samples were left out.

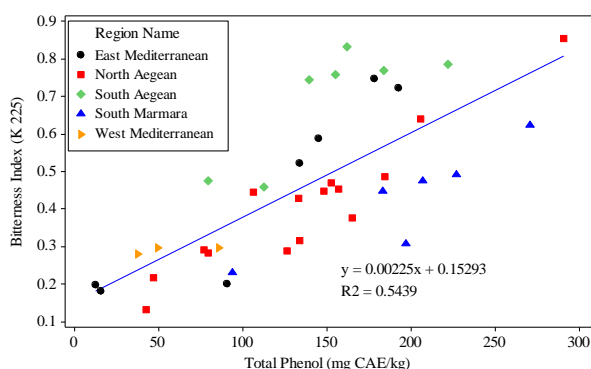


Figure 2. Simple least squares regression plot of total phenol content vs bitterness index (K_{225}) of olive oil samples after removing three outlier samples.

From Figure 2, it is clear that there is a slight increase in R^2 value of regression but this increase is not sufficient to explain total variability in the bitterness index of the olive oil samples as the samples are collected at various geographical regions. In addition, most of the samples were processed with two different methods (two and three phase) and two of them with 2.5 phase system. Besides these differences, there were 10 different cultivar though most of the samples were Ayvalık and Memecik. As a result of these differences, it is expected that a multiple regression model that accounts not only linear effects but also quadratic contributions would generate a better model for bitterness index. Therefore, a second order polynomial model equation (Equation 1) which is composed of geographical region (x_1), processing method (x_2), cultivar (x_3), and total phenol (x_4) was proposed to describe bitterness index (K_{225}) (y). However, there is only one sample with 2.5 phase processing method and therefore no square term were used for x_2 term.

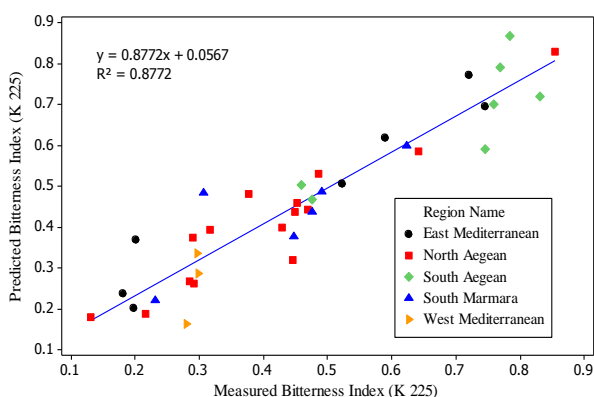
As seen from equation 1, there are 14 coefficients including four linear terms, three square terms and six binary interaction terms along with an intercept term. The statistical significance of the model equation was evaluated by multiple regression with the F-values for analysis of variance (ANOVA) given in Table 2.

$$y = b_0 + b_1x_1 + b_2x_2 + b_3x_3 + b_4x_4 + b_{11}x_1^2 + b_{33}x_3^2 + b_{44}x_4^2 + b_{12}x_1x_2 + b_{13}x_1x_3 + b_{14}x_1x_4 + b_{23}x_2x_3 + b_{24}x_2x_4 + b_{34}x_3x_4 \quad (1)$$

Table 2. Results of ANOVA

Terms	Coefficients	Standard Error of Coefficients	t-values	P-values
Constant	0.626	0.037	16.893	<<0.05
Region	-0.022	0.076	-0.293	0.772
Processing	0.028	0.026	1.075	0.293
Cultivar	0.116	0.079	1.466	0.156
Total Phenol	0.435	0.059	7.307	<<0.05
Region*Region	-0.103	0.083	-1.243	0.226
Cultivar*Cultivar	0.069	0.108	0.637	0.530
Total Phenol*Total Phenol	0.012	0.099	0.124	0.902
Region*Processing	0.045	0.054	0.829	0.415
Region*Cultivar	-0.241	0.127	-1.893	0.070
Region*Total Phenol	0.043	0.130	0.327	0.747
Processing*Cultivar	0.003	0.042	0.076	0.940
Processing*Total Phenol	-0.012	0.066	-0.185	0.855
Cultivar*Total Phenol	0.050	0.098	0.506	0.617

ANOVA evaluations of this model, shown in Table 2, indicates that the most dominating factor of the model is total phenol content as expected from the previous simple regression analysis. On the other hand, none of the other terms of the model equation has P-values lower than suggesting that at 95% confidence level, they are statistically insignificant. Nevertheless, following total phenol content type of cultivar seems to be the second important linear term of the model and it is followed by processing method and geographical region. In terms of nonlinear contribution of the factors, geographical region has the largest absolute coefficient whereas region*cultivar interaction is also quite important at least 90% confidence limit (P-value is less than 0.10). The predicted versus reference bitterness index (K_{225}) values plot is given in Figure 3.

**Figure 3.** Measured vs. predicted bitterness index (K_{225}) by the multiple regression model.

As seen from figure 3, the multiple regression model proposed for the bitterness index produced an R^2 of 0.877 indicating that about 88% of the total variability within the given data were explained with the current model. This is a much better value compared to the simple least squares model generated with just total phenol content given in Figure 2. Response surface plots of the four factor namely geographical region, processing method, cultivar type and total phenol are shown in Figure 4. While two of the four factor being plotted in the three dimensional surface plots, the other two factor were held constant at their middle values.

As seen from Figure 4, there are strong nonlinearities in the top two figures where geographical region and processing type (top left) and geographical region and cultivar (top right) whereas the other figures are mainly dominated by linear effects. In addition, it is seen that total phenol is the main factor which significantly determines the bitterness index (K_{225}) of the olive oil samples.

CONCLUSION

It is generally known that total phenol content was greater in the centrifuge-extracted oil than in the pressure extracted oil. The concentration of total phenol content was slightly lower in the oils extracted using the three-phase decanter than those from the two-phase decanter. This is due to the addition of water to the olive paste processed with the 3-phase centrifugal decanter. In the research it was determined that the total phenol content in Küçükuyu sample was low at two-phase

decanter and high at three-phase decanter. This maybe a minimum quantity of water could be added to the three-phase decanter to separate the solid material better and to retain water-soluble phenol compounds as much as possible.

In this study, Turkish olive oils total phenol content were found to be ranging from 12.41 to 330.26 mg CAE kg^{-1} oil and bitterness index (K_{225} value) ranging from 0.13 to 1.21. The results were demonstrated that total phenol content and bitterness index (K_{225}) of olive oils were affected from cultivars, location and regions. Especially, Beylik, Memecik and Ayvalık olive oils K_{225} values were found be highest that consumers may refuse to consume them in the past. However, in the majority of the reports given in the literature it is stated that bitterness, pungency and astringency are

positive sensorial attribute of the extra virgin olive oils and related to the phenolic compounds in the olive oil. Therefore, due to the positive affect of phenolic compounds to human health, consumers make their preference to oils with more bitterness in recent years. For Beylik cultivar that was located in Manavgat, in which bitterness is so excessive as to cause consumer rejection of the olive oil. Appropriate control of the some factors like maturiy index, technological variables to produce a desirable reduction of intensity of this attribute and hence improve consumer preference. In addition, some Turkish cultivars like Edincik Su, Ayvalık, Memecik, Kilis Yaglık and Beylik were found have higher bitternes values than Italian and Spanish varieties.

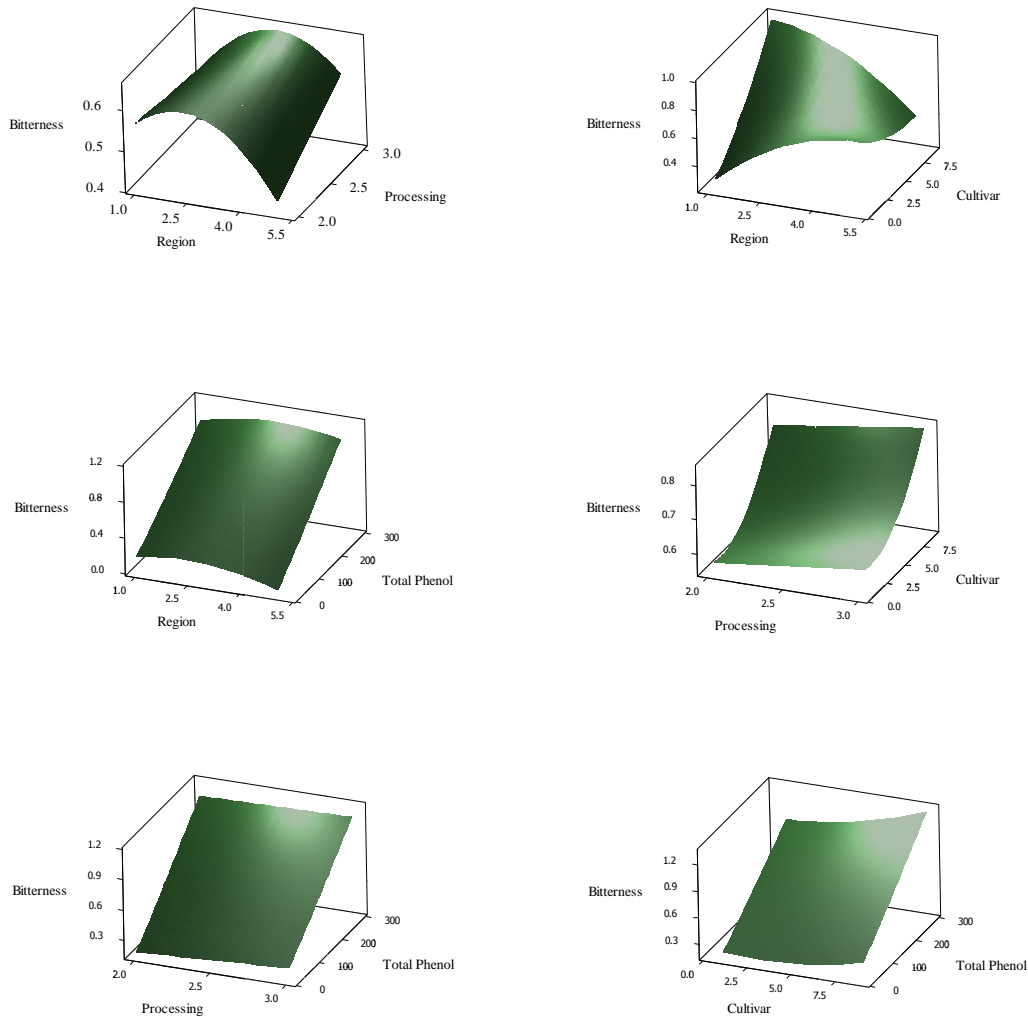


Figure 4. Response surface plots of geographical region, processing type, cultivar and total phenol content as a function of bitterness index (K_{225}).

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