İnönü Üniversitesi Sağlık Hizmetleri Meslek Yüksekokulu Dergisi
ISSN: 2147-7892, Cilt 13, Sayı 1 (2025) 9-30
doi: 10.33715/inonusaglik.1544923

Journal of Inonu University Health Services Vocational School
ISSN: 2147-7892, Volume 13, Issue 1 (2025) 9-30
doi: 10.33715/inonusaglik.1544923

Original Article / Araştırma Makalesi

THE EFFECT OF DIFFERENT PROCESSING METHODS ON AMYGDALIN LEVELS AND SOME PHYSICOCHEMICAL PROPERTIES OF APRICOT KERNELS

Acı Kayısı Çekirdeğinin Amigdalin Düzeyine ve Bazı Fiziko Kimyasal Özelliklerine Farklı İşleme Yöntemlerinin Etkisi

Sultan NALÇACI[1] *Mucahit ORUÇ*[2] *Emine ŞAMDANCI*[3]
[1,2]*İnönü University, Institute of Health Sciences, Malatya*
[3]*Gazi University, Faculty of Medicine, Ankara*

Geliş Tarihi / Received: *06.09.2024* ***Kabul Tarihi / Accepted:*** *31.10.2024*

ABSTRACT

In this study, the amygdalin content of the Hacıhaliloğlu, Kabaaşı, and Zerdali varieties was determined using an LS-MS/MS device. Methods to reduce the amygdalin content in raw bitter apricot kernels to a safe level for consumption were identified. To remove bitterness treatments such as alkaline treatment with sodium bicarbonate (4-10% $NaHCO_3$, 7-15 min), ultrasound (30-50 °C, 30-60 min), microwave (240-560 W, 2-5 min) and roasting (120-160 °C, 10-20 min) were performed. After all treatments, protein content, the amygdalin content via HCN, the antioxidant activity, the phenolic compounds and the color values (L, a*, b*, chroma, hue) of the samples were determined. The fat content (%42.99, %44.35, %40.75), protein content (%34.14, %30.36, %29.27) and hydrocyanic acid (HCN) content (mg/kg) of Hacıhaliloğlu, Kabaaşı and Zerdali varieties were 24.56, 30.31 and 1630.66, respectively. Among the methods used to reduce or eliminate the amygdalin content, roasting was found to be the most effective. The amygdalin content of raw Zerdali kernels was 27.64 mg/g, which decreased to 9.77 mg/g after roasting at 160 °C for 10 min. The antioxidant activity was measured at 82.59 mg TE/100 g, but a decrease was observed after treatment with sodium bicarbonate (81.72), ultrasound (57.5), microwave (52.65) and roasting (97.04).

Keywords: Amygdalin, Microwave, Protein, Zerdali.

ÖZ

Bu çalışmada; tatlı çekirdeklere sahip Hacıhaliloğlu ve Kabaaşı çeşitleri ile acı çekirdeklere sahip Zerdali çeşidindeki amigdalin miktarı LS-MS/MS cihazı kullanılarak analiz edildi. Çiğ acı kayısı çekirdeklerindeki amigdalin içeriğini güvenli tüketim seviyelerine düşürmeye yönelik yöntemler belirlendi. Acılığı gidermek için sodyum bikarbonat ile alkali işlem (%4-10 $NaHCO_3$, 7-15 dk), ultrason uygulaması (30-50 °C, 30-60 dk), mikrodalga işlemi (240-560 W, 2-5 dk) ve kavurma işlemi (120-160 °C, 10-20 dk) gibi yöntemler uygulandı. Tüm işlemlerin ardından örneklerin yağ ve protein içeriği, HCN üzerinden amigdalin içeriği, antioksidan aktivite, fenolik bileşikler ve renk değerleri (L, a*, b*, chroma, hue) tespit edildi. Hacıhaliloğlu, Kabaaşı ve Zerdali çeşitlerinin sırası ile yağ içeriği (%42.99, %44.35, %40.75), protein içeriği (%34.14, %30.36, %29.27) ve hidrojen siyanür (HCN) içeriği (mg/kg) 24.56, 30.31, 1630.66 olarak saptandı. Acı kayısı çekirdeklerindeki amigdalin içeriğini azaltmak veya yok etmek için uygulanan uygulamalar arasında en etkili kavurma yönteminin olduğu tespit edildi. Ham Zerdali çekirdeklerinin amigdalin içeriği 27.64 mg/g olarak belirlenmiş ancak 160 °C'de 10 dakika kavurma işlemi sonrasında bu değer 9.77 mg/g'a geriledi. Antioksidan aktivite 82.59 mg TE/100 g olarak ölçülmüş, ancak sodyum bikarbonat (81.72), ultrason (57.5), mikrodalga (52.65) ve kavurma (97.04) işlemleri sonrasında bu değerlerde azalmalar tespit edildi.

Anahtar kelimeler: Amigdalin, Mikrodalga, Protein, Zerdali.

Sultan NALÇACI ✉, sultan.ancin@gmail.com
İnönü University, Institute of Health Sciences, Malatya

Bu makaleye atıf yapmak için (How to cite this article): Nalçacı, S., Oruç, M. ve Şamdancı, E. (2025). The Effect of Different Processing Methods on Amygdalin Levels and Some Physicochemical Properties of Apricot Kernels. *İnönü Üniversitesi Sağlık Hizmetleri Meslek Yüksekokulu Dergisi*, 13(1), 9-30. doi: 10.33715/inonusaglik.1544923

INTRODUCTION

Apricot which is the sweet and juicy fruit of the Rosaceae family (*Prunus armeniaca*) is a small rigid seeded member. Apricot that is consumed both in fresh and dried form, is commonly used worldwide (Uğur & Yaman, 2022). Whereas our country is one of the biggest producer countries in apricot export, Malatya region is the center of apricot and apricot kernel production (Kaya, Okur, Temel, Üstyol & Çaksen, 2012). According to 2022 data from the United Nations Food and Agriculture Organization (FAO), our country leads the way by supplying with 20.8% of the production and 803 thousand tons (FAO, 2022). The use of apricot kernels in the food and health industry increases the importance of that. Sweet kernels are especially used in desserts, yogurts, granolas, and diverse snacks, whereas the use of bitter kernels in health and beauty products is common (Atiş, 2017).

Amygdalin, which is the component that specify the bitterness or sweetness of the kernels, is a natural cyanogenic glycoside (Milazzo, Ernst, Lejeunue & Boehm, 2011). The release of toxic hydrogen cyanide limits the use of apricot kernels in the food industry. In the safety assessment of the *Prunus armeniaca L.* kernel, the plant is on the negative list in Norway, Iceland and Hungary, whereas the positive list in Germany, Croatia, Belgium, Romania, Slovenia (GKGM, 2022). Wild apricot seeds contain 200 mg/100 g of hydrocyanic acid, whereas the seeds of cultivated varieties contain 8.9-11.7 mg/100 g (Alparslan & Hayta, 2008). In terms of acute cyanide poisoning, apricot kernels are the most common source of poisoning specially in young children. (Karsavuran et al., 2014). Hospital in which 13 cases of admission due to poisoning from apricot kernel consumption in terms of, the average age was reported as 5.7 years, were evaluated (Akyildiz, Kurtoğlu, Kondolot & Tunç, 2010).

The European Food Safety Authority (EFSA) has determined a lethal dose of 0.5-3.5 mg per kg of body weight, while setting the acute reference dose (ARfD) at 20 micrograms (µg, $1x10^{-6}$ g) as the safe consumption verge for at a time exposure (EFSA, 2011). In whole apricot kernels, amygdalin and catabolic enzymes are stored in different departments and physical processes like grinding or chewing release hydrocyanic acid. The complete splitting of 1 g of amygdalin reveals 59 mg of HCN (FAO, 2012).

By taking into account the risk due to the amygdalin content of *Prunus armeniaca* L. seeds, it has been recommended that the amount of HCN should not exceed 35 mg/kg in products consisting of whole, ground or crushed, sliced or any extract and extract prepared apricot kernels, on the condition that it is added as positive (P) (GKGM, 2022).

The toxicity risk related to amygdalin, which is found in several plant seeds of the Rosaceae family and some vegetables, has been known for centuries and this risk is eliminated in products prepared using many methods such as peeling, crushing, grinding, soaking, fermentation, and so on. To manage the potential risk of raw apricot kernels, Australia and New Zealand have banned the consumption of raw apricot kernels and all substances derived from raw apricot kernels (ground, milled, crushed, chopped), while ruling out apricots, alcoholic beverages, oil, flavors, juice with seeds, marzipan, cakes, biscuits and confectionery containing raw apricot kernels (FZANS, 2008).

Because of the amount of hydrocyanic acid, the system used by the Ministry to determine the status of exported and returned products was also given warnings. In 2021, apricot kernels comprising a high amount of cyanide (2996 +/-719 mg/kg–ppm) were rejected at the Bulgarian border control under the Rapid Alert System for Food and Feed (RASFF) (GKGM, 2021).

The priority aim of this study is to decrease the level of amygdalin, which is a problem in the food sector, to the appropriate consumption limit and to ensure the use of bitter kernels in the food industry.

MATERIAL AND METHOD

Material

As of materials, the Hacıhaliloğlu and Kabaaşı varieties, which are commonly grown in Malatya and have sweet kernels, as well as the wild apricot variety, which has bitter kernels (Zerdali), have been used. The kernels were obtained from the Apricot Research Institute's garden. The study has been determined 4 trees and 3 replications for each variety. After harvesting the fruit, the kernels have been dried in the sun by breaking. Dried apricot kernels were stored in a cold room at +4 °C for analyses and applications.

Method

It has been aimed to remove and reduce amygdalin with the determined treatments to raw apricot kernels and bitter kernels.

Bitterness removal treatments:

A = Alkaline ($NaHCO_3$) Treatment: Concentration (4% $NaHCO_3$-10% $NaHCO_3$), Duration (7 min- 15 min)

U= Ultrasonic Wave (40kHz) Method: Temperature (30 ºC-50 ºC), Duration (30 min – 60 min)

M= Microwave Method: Power (240 W -560 W), Duration (5 min- 20 min)

K= Roasting Method: Temperature (120 ºC-160 ºC), Duration (10 min- 20 min)

Humidity Ratio

It has been determined by drying the kernel samples in an oven at 105 °C until constant weight (Cemeroğlu, 2010).

Ash Analysis

After recording the tare weight of porcelain crucibles with a constant weight, 5 g of ground kernel samples were placed in them. The samples underwent pre-combustion, followed by combustion at 650 °C in an ash furnace until exact whiteness was obtained, taking at least 12 hours. The crucibles were then transferred to a desiccator, cooled to room temperature, and weighed. The total ash amount (%) was calculated using a formula (Gönül, Altuğ, Boyacıoğlu & Noka, 1988).

Oil Determination

In advance dried in the oven and cooled in the desiccator, a balloon flask was weighed after adding 2 glass beads. Five grams of ground kernel was placed in a cartridge, which was covered with wet cotton. The cartridge is placed in the extractor and solvent (n-hexane) is added to the flasks to make 1.5 flushes. The balloon, cooler and extractor are connected and the heater is turned on. After 8 h of extraction, the remaining solvent is removed. The flask kept in the oven is cooled in the desiccator and weighed and calculated by the formula after constant weighing (Doğan & Başoğlu, 1985).

Protein Determination

According to the Kjheldal method, the protein content in the kernels has been based on the total nitrogen content. The determined %N value is multiplied by a factor of 6.25 to give the percentage of protein (Kacar, 1984).

Amygdalin Amount

The amygdalin content in apricot kernels is quantitatively determined by LC-MS/MS based on the HCN content (Table 1). The rule of the method is that based on the extraction of amygdalin from apricot kernels and products consisting of apricot kernels (whole, ground or crushed, sliced or extracted), filtering and analyzing in LC-MS/MS and reporting with regards to HCN using a conversion factor (Lee, Zhang, Wood, Castillo & Mitchell, 2013).

Table 1. LC-MS/MS Method Parameters

Column	Waters Acquity UPLC BEH C18, 1.7 µm 2.1 x 100 mm
Column temperature	40 °C
Mobile Phase A	0.1% formic acid and 5 mM ammonium formate in water
Mobile Phase B	Acetonitrile
Mobile phase composition	80:20, A:B
Analysis time	4 minutes
Flow rate	0.3 mL/min (isocratic)
Injection volume	5 µL (partial loop with needle overfill)
Source	ESI negative mode
Capillary voltage	3.5 kV
Desolvation temperature	400 °C
Desolvation gas flow rate	800 liters/hour
Detection Mode	MRM

Materials Used

LC-MS/MS device (Shimadzu 8040 LC-MS/MS, Kyoto- Japonya), Acquity UPLC BEH C18, 1.7 µm, 2.1 x 100 mm column, vortex, ultrapure water device, analytical balance, ultrasonic water bath, 2 mL HPLC vials, pipettes (100, 1000, 5000 µL), 50 mL disposable polypropylene test tubes, balloon flask (10 mL), plastic syringe (10 mL), 0.45 µm nylon filter.

Chemicals Used: Amygdalin (Reference Standard Substances, CAS no: 29883-15-6), Water (ultrapure), acetonitrile, methanol (LC-MS purity), acetic acid, ammonium formate, formic acid.

Preparation of Chemicals

1.Mobile phase A (0.1% formic acid and 5 mM water with ammonium formate): 315 mg of ammonium formate was weighed, small amount of water is added and dissolved. Then 1 mL of formic acid is added to the solution and the volume make up to one liter with distilled water. The amounts can be increased or decreased proportionally depending on the volume of mobile phase required.

2. Extraction Solution (70:30 methanol: water + 0.1% acetic acid): To prepare standard solutions and for use in extraction, add 700 mL of methanol and nearly 250 mL of water to a 1000 mL volumetric flask. Add 1 mL of acetic acid and make up the volume with water to the mark.

Preparation of intermediate stock standard solution

The master stock solution is diluted 70:30 with methanol: water + 0.1% acetic acid to prepare an intermediate stock solution with a concentration of 10 mg/L.

Preparation of the calibration curve

The intermediate stock solution with a concentration of 10 mg/L is diluted with extraction solvent (70:30 methanol: water + 0.1% acetic acid) to prepare standards with amygdalin

concentrations of 25, 50, 100, 250, 500, 1000, and 2000 µg/L. The detected amount of amygdalin is reported with regards to (HCN) using a conversion factor. The conversion of amygdalin to HCN is calculated using the formula;

HCN amount=Amygdalin amount x 0.059

Sample Extraction

Apricot kernel sample is homogenized by completely grinding in a blender.1± 0.001 g of the ground sample is weighed into a 50 mL disposable polypropylene tube. 20 mL of extraction solution (70:30 methanol: water + 0.1% acetic acid) is added (20-fold dilution). The tube is placed in an ultrasonic water bath at room temperature for 10 minutes. At the end of the time, it is centrifuged at 5000 xg for 5 min. A few mL of the supernatant solution is passed through a 0.45 µm nylon filter into 2 mL centrifuge tubes. 50 µL of the filtered supernatant solution is taken into a 2 mL vial, 950 µL of the extraction solution (70:30 methanol: water + 0.1% acetic acid) is added and vortexed (20-fold additional dilution) and the vial is analyzed by LC-MS/MS. (400-fold dilution of the sample in the analyzed vial).

Determination of Total Phenolic Content

The extraction of the samples was performed according to the method reported by Zengin et al., (2024). Total phenolic content of the ground kernel samples is determined using the Folin-Ciocalteu (FC) reagent after specific steps, and the absorbance is measured at 765 nm using a spectrophotometer (Shimadzu UV1601). Standard solutions with gallic acid are prepared and the calibration graph is plotted. After attaining the equation, the total amount of phenolic content is calculated and expressed as gallic acid equivalents (mg GAE/100 g dry weight) (Slinkard & Singleton, 1977).

Antioxidant Capacity

In the DPPH method, 100 µL (2.5 mg DPPH and 100 mL methanol) of the ground kernel sample is dissolved in a falcon tube, mixed with DPPH reagent and kept in the dark for 30 min and the absorbance is measured for a wavelength of 517 nm. Calibration graph against standards, equation derivation and calculation are given in mg TE/100 g. (Kumaran & Karunakaran, 2006).

Color Measurement

The color values of the fruits were measured with the CM-700d color gadget (Konica Minolta) according to the L*, a*, b* measurements (CIE) method. In the measurements, L*=0 indicates black, and L*=100 indicates white (bright). The C color value represents the color

intensity of the samples (C=0 indicates a dull color, C=60 indicates a very vivid color). The following formula was used in the calculation. The h° color value represents the hue angle of the samples (0°=red-purple, 90°=yellow, 180°=greenish-blue, 270°=blue)

FTIR Analysis

The ground kernel samples were analyzed with a Perkin Elmer Spectrum One, IR 1.10 version FTIR spectrophotometer with peaks in the range of 400-4000 cm^{-1}.

Scanning Electron Microscope (SEM)

The surface morphologies, pore sizes, and surface distributions of the ground kernels were determined by Scanning Electron Microscope (Leo EVO-40 VPX Carl Zeiss SMT, Cambridge, UK).

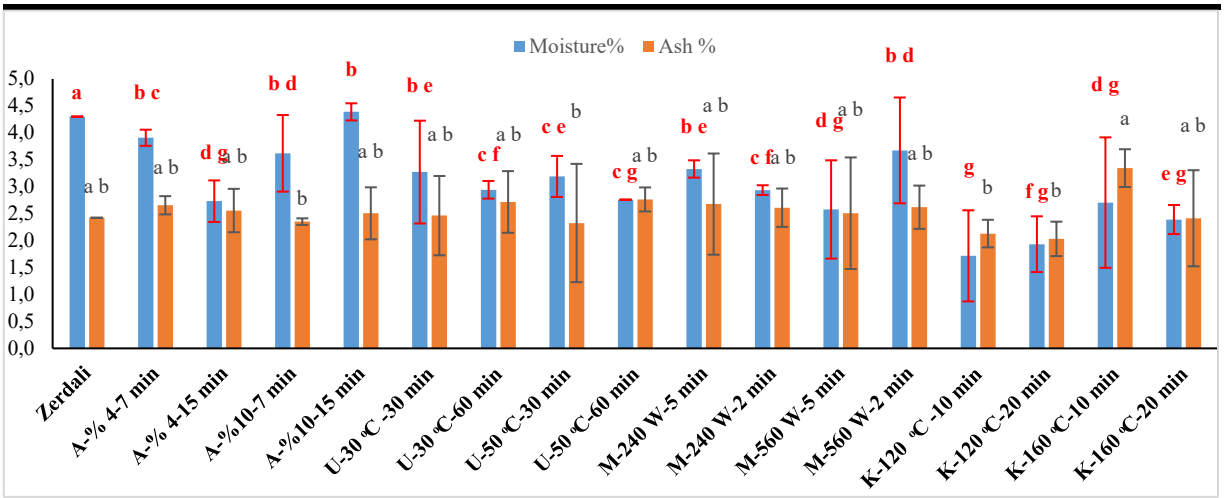
Statistical Analyses

The study data were analyzed as mean ± standard deviation using the JMP 13 package program (SAS Version V.9.4, SAS Institute Cary, N.C.).

RESULTS AND DISCUSSION

The moisture and ash values of the Hacıhaliloğlu variety used in the study were found to be 4.80% and 2.60%, respectively, while they were found to be 4.93% and 2.81% in the Kabaaşı variety.

The moisture and ash values in the treatments aimed at decrasing the amygdalin level of the bitter kernels are shown in Figure 1, and no statistically important difference was found between the treatments. In a study on some apricot varieties grown in Malatya, the moisture amount was found to be 4.86% in Kabaaşı, 4.82% in Hacıhaliloğlu, and 4.88% in wild apricot (Mutlu & Hayaloğlu, 2022).



A=Alkaline, U=Ultrasonic, M=Microwave, K=Roasting
Figure 1. Moisture and Ash Amount of Samples

For reducing the bitterness of raw wild apricot kernels, it was found statistically important ($p < 0.05$) with regards to % oil amount between the treatments (Table 2). The highest amount of oil was observed in the roasting process, with the maximum level seen in the K-160 °C-10 min application.

Table 2. Oil Amount of Samples (%)

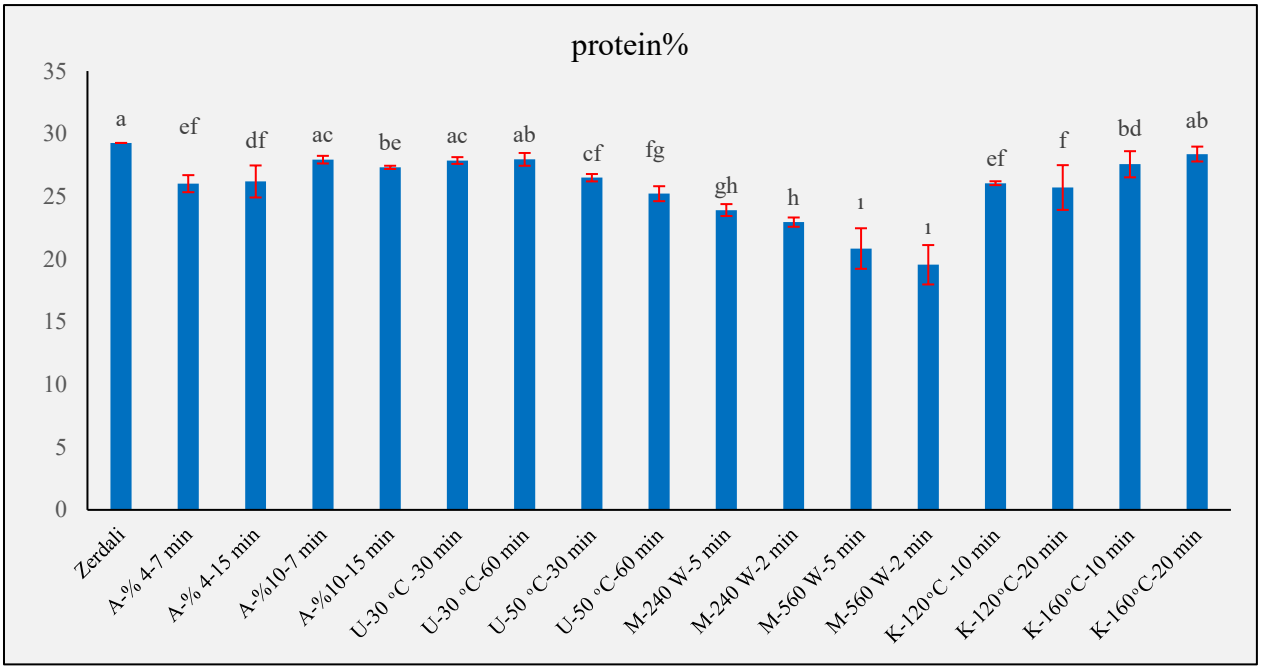
Zerdali	40.75±1.92 **d e**		
A-%4-7 min	29.66±1.35 **h**	**M-240 W-5 min**	41.07±0.44 **d e**
A-%4-15 min	30.11±1.73 **h**	**M-240 W-2 min**	40.88±0.5 **d e**
A-%10-7 min	32.58±0.76 **g**	**M-560 W-5 min**	39.93±0.22 **e**
A-%10-15 min	30.27±3.55 **h**	**M-560 W-2 min**	42.69±1.43 **c d**
U-30 ºC-30 min	37.11±2.49 **f**	**K-120 ºC-10 min**	44.29±0.71 **b c**
U-30 ºC-60 min	30.75±1.09 **g h**	**K-120 ºC-20 min**	45.55±0.17 **a b**
U-50 ºC-30 min	35.8±0.67 **f**	**K-160 ºC-10 min**	46.83±2.88 **a**
U-50 ºC-60 min	36.07±1.44 **f**	**K-160 ºC-20 min**	44.10±0.63 **c b**

A=Alkaline, U=Ultrasonic, M=Microwave, K=Roasting

In a study in which raw apricot kernels were soaked and roasted, the oil amount was found between 17.25-31.05% in soaking and 35.05-38.91% in roasting (Badr, Ramis, Wahdan, Sakr & Elghandour, 2017). In our study, the high oil amount in the roasting process is associated with the literature. Microwave treatment has been reported to change the oil amount and fatty acid composition (Juhaimi, Özcan, Ghafoor & Babiker, 2018), while ultrasonic application has been reported to decrease the oil amount (Zhang, Shi, Yao, Sakr & Zhang, 2020). In our treatments, whereas the oil amount increased slightly with microwave treatment, it decreased somewhat with the loosening and opening of the kernel structure with ultrasonic application.

The % protein amount was found to be statistically important between the bitterness removal processes ($p<0.05$). In the applications, the amount of protein reduces in the microwave process (Figure 2). In a study carried out to remove amygdalin in plum kernel, it was reported

that microwave disrupts the three-dimensional structure of proteins and pave the way the function of proteins to change.



A=Alcaline, U=Ultrasonic, M=Microwave, K=Roasting

Figure 2. Protein Amount of Samples

In our study, to determine the effectiveness of the methods aimed at decreasing the bitterness of wild apricot kernels, amygdalin measurements were carried out using an LC-MS/MS.

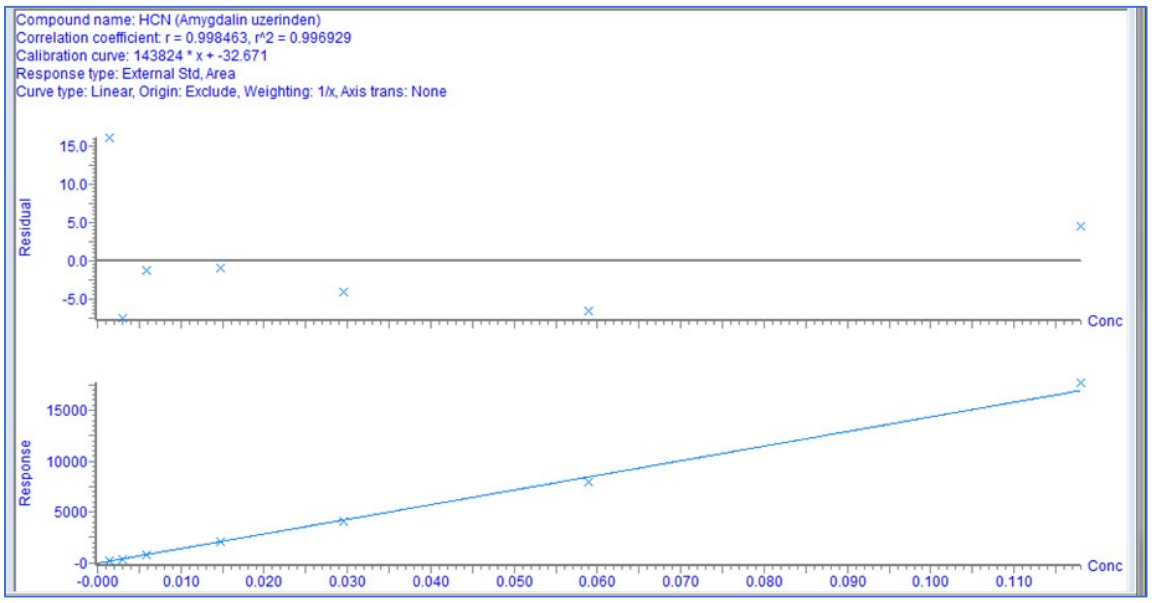


Figure 3. Amygdalin Calibration Graph

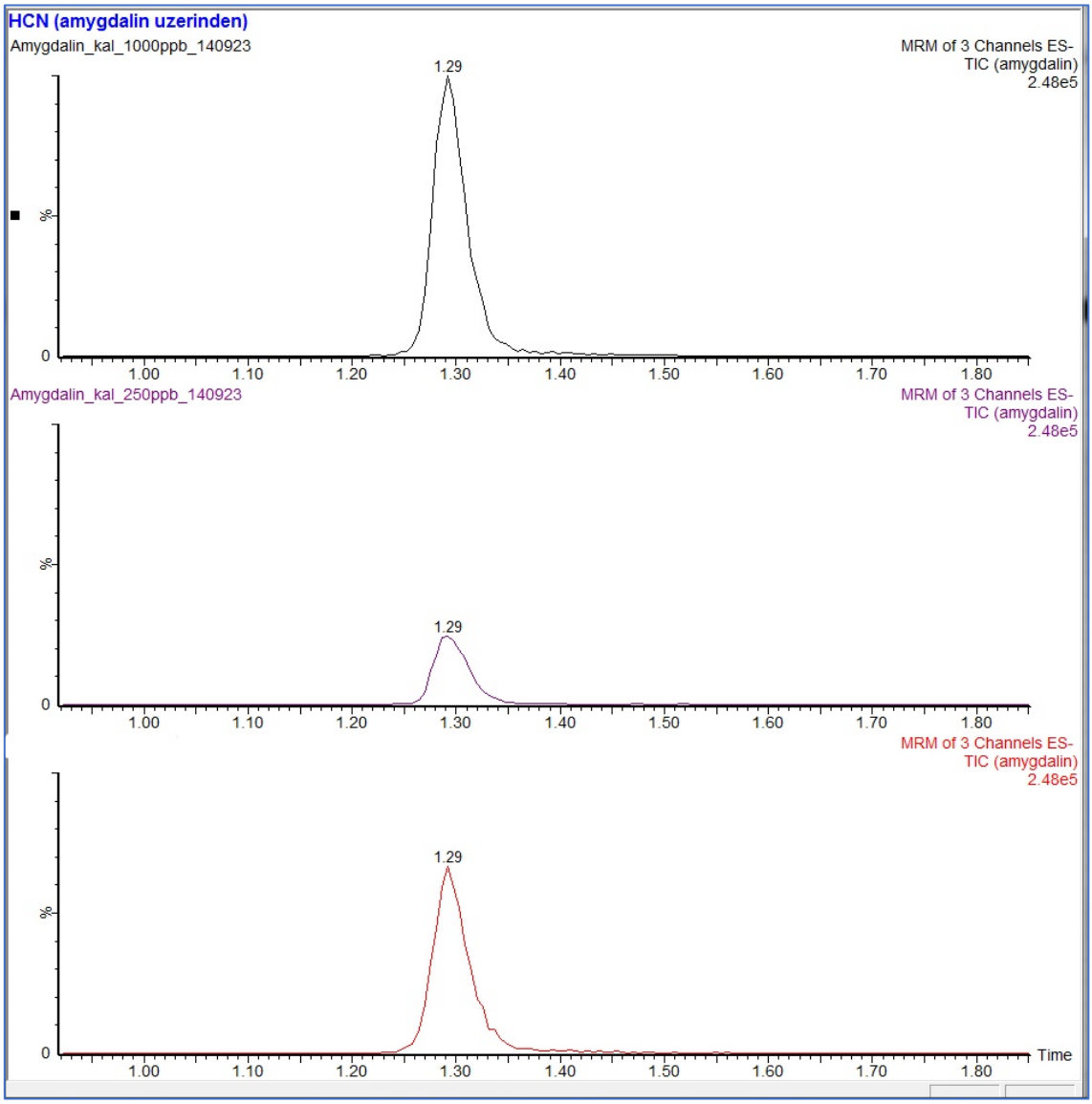


Figure 4. Chromatogram of Amygdalin at Different Concentrations

There was a statistically important difference between the amygdalin values between the treatments ($p<0.05$). As shown in Table 3, the amount of amygdalin was 27.64 in wild apricot, while a small increase was observed in the treatment with alkaline ($NaHCO_3$), which can be explained by the fact that $NaHCO_3$ raises the pH of the medium and enables amygdalin hydrolysis. The amount of amygdalin (mg/g) reduced the most with the K-160 ºC-10 min process 9.77 mg/g.

Table 3. Amygdalin Amount of Samples (mg/g)

Zerdali	27.64±0.49 **a c**		
A-%4-7 min	27.94±0.37 **a**	**M-240 W-5 min**	22.35±0.29 **ı**
A-%4-15 min	27.24±0.33 **c d**	**M-240 W-2 min**	22.70±0.28 **ı**
A-%10-7 min	26.86±0.28 **d e**	**M-560 W-5 min**	25.12±0.25 **g**
A-%10-15 min	27.36±0.38 **b c**	**M-560 W-2 min**	25.83±0.26 **f**
U-30 ºC-30 min	26.48±0.21 **e**	**K-120 ºC-10 min**	24.62±0.29 **h**
U-30 ºC-60 min	26.6±0.11 **h**	**K-120 ºC-20 min**	22.55±0.31 **ı**

U-50 ºC-30 min	27.79±0.33 **a b**	**K-160 ºC-10 min**	9.77±0.26 **l**
U-50 ºC-60 min	20.91±0.29 **j**	**K-160 ºC-20 min**	14.96±0.22 **k**

A=Alkaline, U=Ultrasonic, M=Microwave, K=Roasting

In the literature, there are studies on the determination of amygdalin content of apricot varieties. While the average amount of amygdalin in bitter kernels was found to be 26.27 mg/g, it was found to be 0.16 mg/g in sweet kernels (Poyraz, 2013). The amygdalin amount of the bitter kernels we used in our study was close to these values, while the amount of sweet kernels was found to be 0.42 and 0.51 mg/g.

Plum kernels are the cheapest sources of protein and oil, but cannot be used effectively because of the presence of anti-nutrient factors such as amygdalin (range 0.1-17.5 mg g^{-1}) (García et al., 2014), but the high content of amygdalin makes it a valuable source of medicines (Lee et al., 2013). They performed amygdalin removal in plum kernels using hydrothermal water and microwave method against traditional methods, and described the microwave method as more effective, easier and green detoxification. Both methods did not exactly remove the anti-nutritional compound amygdalin (Bolarinwa, Orfila & Morgan, 2016). In our study, the amount of amygdalin reduced to a certain extent, but it is similar in the sense that it was not exactly removed.

In a study carried out for amygdalin removal in flaxseed, while the initial amount of amygdalin was 377 mg/kg, it was decreased to 63.5 mg/kg by microwave treatment and found more successful than autoclaving, pelletizing and heating processes (Shen, Feng & Fan, 2005). While 83% amygdalin amount was removed with the study, this rate was found to be 65% in our study.

They reported that the rate of amygdalin reduced by 93% when cassava leaves were treated with sodium bicarbonate for 6 hours and that this substance is practical with regards to easy availability at home (Latif & Müller, 2005). However, in our study, it was observed that treatment with sodium bicarbonate had no effect on amygdalin detoxification.

In our study, HCN was found statistically significant between the initial and post-treatment amounts ($p<0.05$). As seen in Table 4, the amount of HCN reduced the most in the kernels roasted at 160 ºC for 10 min compared to the initial amount.

Table 4.HCN Amount of Samples (mg/kg)

Zerdali	1630.66±28.97 **a c**		
A-%4-7 min	1648.16±21.92 **a**	**M-240 W-5 min**	1318.4±16.97 **ı**
A-%4-15 min	1607.33±19.41 **c d**	**M-240 W-2 min**	1339.07±16.47 **ı**
A-%10-7 min	1584.47±16.35 **d e**	**M-560 W-5 min**	1482.07±14.58 **g**
A-%10-15 min	1614.27±22.45 **b c**	**M-560 W-2 min**	1523.8±15.42 **f**
U-30 ºC-30 min	1562.03±12. 63 **e**	**K-120 ºC-10 min**	1452.43±16.97 **h**

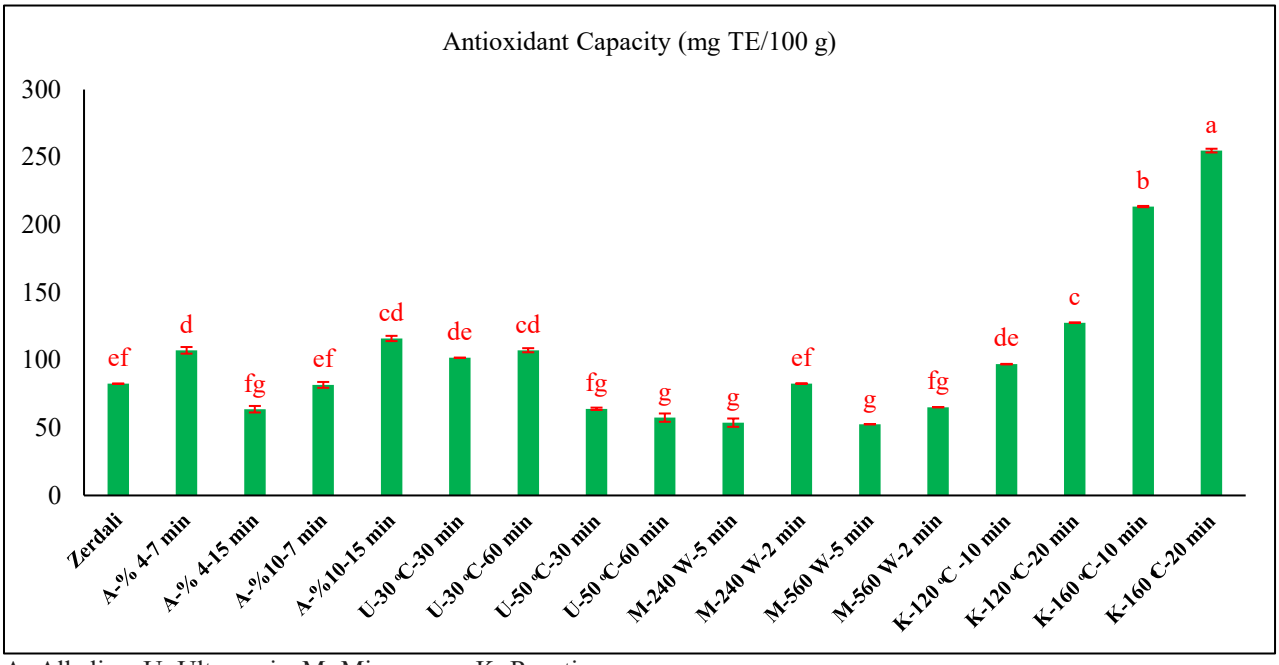
U-30 ºC-60 min	1451.37±6.57 **h**	**K-120 ºC-20 min**	1330.41±18.35 **ı**
U-50 ºC-30 min	1639.89±19.61 **a b**	**K-160 ºC-10 min**	576.2±15.41 **l**
U-50 ºC-60 min	1233.89±17.11 **j**	**K-160 ºC-20 min**	882.35±12.90 **k**

A=Alkaline, U=Ultrasonic, M=Microwave, K=Roasting

In a study where the effects of traditional methods such as grinding, shredding and soaking of hydrogen cyanide, a food contaminant found in flaxseed, cassava, almond and bamboo shoots, were compared with ultrasonic treatment, HCN removal with 4 hours soaking was 61.58%, while 85.43% was determined in a 300 W ultrasound bath (Panghal, Munezzero, Sharma & Chhikara, 2021). In our study, ultrasonic application ensured the highest amygdalin removal rate of 24.50%.

They compared the effectiveness of different thermal treatments (water boiling, microwave, oven heating, autoclaving) on HCN contents in flaxseed (Feng, Shen & Chavez, 2003). While the initial HCN content was 376 mg/kg, they found 22.33, 62.792, 204.33, 300.048 mg/kg, in turn. Water boiling was indicated as the most effective (98% reduction) technique (Noreen, Tufail, Ul Ain & Awuchi, 2023). When we compare the microwave application, they achieved 16% amygdalin removal while in our study it was 20%.

Statistically significant difference was found between the antioxidant activities of the beans after the bitterness removal processes ($p<0.05$). As can be seen in Figure 5, the highest antioxidant capacity was 254.67 in the roasting process, particularly in the 160 ºC 20 min application. While it was 82.59 in raw wild apricot kernels, it varied between 637.34 and 1160.23 in alkaline treatments, between 575.15 and 1073.27 (mg TE/100g) in ultrasonic treatments, and between 526.47 and 826.70 in microwave treatment.



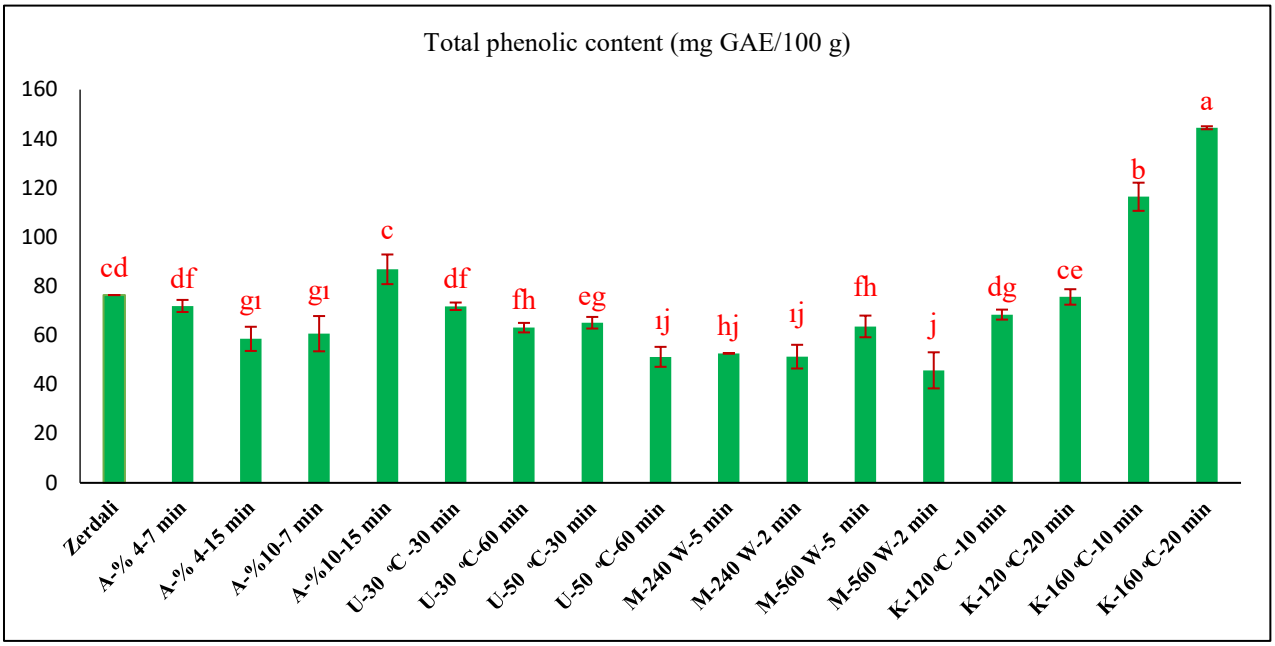
A=Alkaline, U=Ultrasonic, M=Microwave, K=Roasting

Figure 5. Antioxidant Capacity of Treatments (mg TE/100 g)

While the initial amount (mg TE/100 g) in plum kernels was 24.83, it was 36.20 after heat treatment (Sheikh & Saini, 2021). In a study in which the antioxidant capacity of roasted apricot kernels was determined using the Dpph method; it was reported that the scavenging effect of apricot kernel flour extracts on DPPH radical changed significantly with roasting time and even 10 min of roasting was effective in raising the anti-radical power. The increase in antioxidant capacity with roasting was because the formation of maillard reaction products during roasting (Bae & Suh, 2007).

Roasting apricot kernels can raise their antioxidant content because high temperatures can enable the release of certain nutrients. However, the effect of roasting on antioxidant levels is influenced by several factors such as roasting time and temperature. Long-term roasting or roasting at temperatures higher than 200 ºC can decrease the antioxidant content (Thaipong, Boonprakob, Cisneros & Hawkins, 2006).

The total phenolic content was statistically significant among the treatments applied to decrease the amount of amygdalin. Phenolic matter content of raw wild apricot kernels was found to be 76,41 in alkaline treatment (58.5, 86.89), in ultrasonic treatment (51.27, 71.85) and (45.78, 63.65) in microwave treatment (Figure 6). The highest increase in the amount of total phenolic content was 68.46, 144.48 (mg GAE/100 g) in the roasting treatment.



A=Alcaline, U=Ultrasonic, M=Microwave, K=Roasting
Figure 6. Total Phenolic Content of Treatments (mg GAE/100 g)

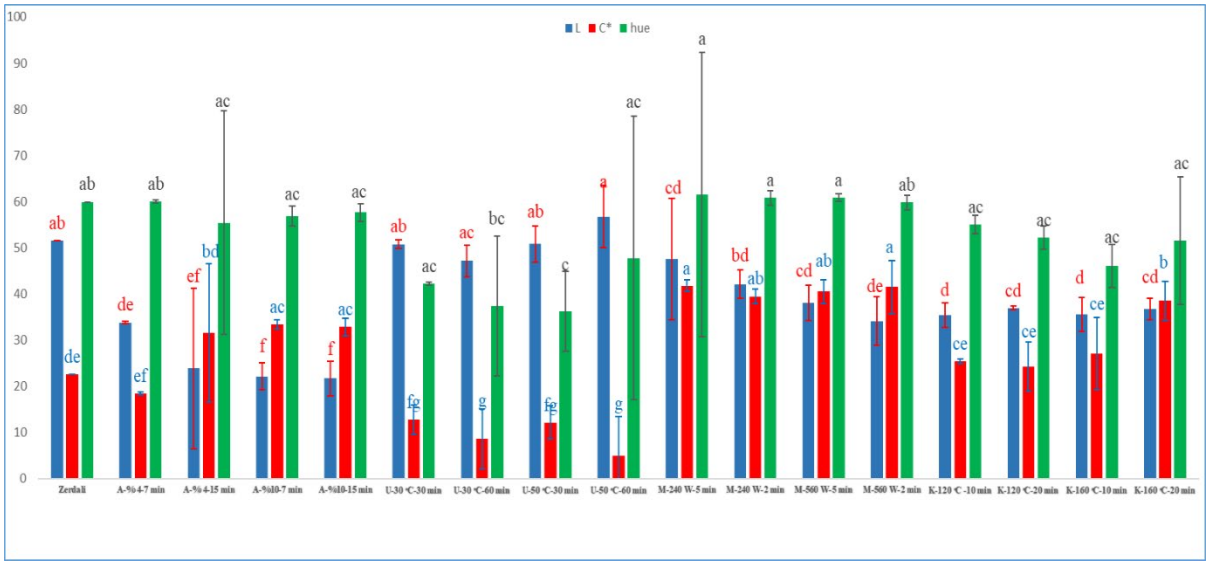
In a study on the effect of soaking and roasting on protein quality of raw apricot kernels, total phenolic matter in raw apricot kernels, roasted kernels, soaked kernels and roasted soaked kernels were 13.68, 26.48, 17.06 and 18.64 mg GAE/100 g, respectively (Badr, Ramis,

Wahdan, Sakr & Elghandour, 2017). In accordance with our study, total phenolic amount increased with roasting.

In a study, hazelnut and peanut samples were roasted at 145 °C for 20 min or 165 °C for 25 min, while black pepper and sesame seeds were roasted at 180 °C for 35 min or 220 °C for 45 min. The results showed that roasting did not significantly ($p > 0.05$) affect the total phenolic content and antioxidant activity of the samples (Turan, Atalay, Solak, Özoğul & Demirtaş, 2021). In our study, roasting significantly increased the amount of both substances.

Different temperature and time (0, 5, 10, 15, 20, 30, 30, 45, 60, 75 and 90 min) combinations are used in roasting (135, 150 and 165 ºC) used in carob powder production. The highest total phenolic content was seen at 165 ºC 75 min. During roasting, important chemical reactions such as sugar caramelization and Maillard reaction occur, which leads significant changes in product quality (Şahin, Topuz, Pischetsrieder & Özdemir, 2009). In our study, the formation of maillard products was decreased by processing in a shorter time at the temperature showing the highest phenolic content.

In a study where date kernels were roasted at 200 ºC for 5, 10 and 15 min and ground and flour was obtained, the phenolic amount of raw date flour was 3616.1 mg GAE/100 g dry matter, while 3237.12, 2080.93 and 1583.66 mg GAE/100 g dry matter were found after roasting processes, while the antioxidant capacity of raw date flour was 10315.06 g TEAC / 100 g dry matter 5343.04, 4902.52, 3858.26 g TEAC/100 g dry matter (Kılıç, 2015). Our study is harmonious with because of the effect of roasting process.



A=Alkaline, U=Ultrasonic, M=Microwave, K=Roasting
Figure 7. Color Value (L*, C*, h º)

The L* value of wild apricot kernels, was 51.63. The main material of the study, while 23.88, 22.16 and 21.74 were found in alkaline treatments, in turn. While the a* value was 11,3 in wild apricot, it reduced the most in ultrasonic treatment 8.52, 6.54, 7.54, 3.20 a* value increased with microwave treatment (19.89, 19.25, 19.63, 20.87). The a* value showed a important increase with 24.12 in K-160 ºC-20 min application. b* value was 19,55 in wild apricot and very low in ultrasonic treatment (9.20, 5.45, 8.93, 3.21). It shows that the color of the kernels is light yellow. It is also thought that these changes can be based upon to the process of transport or removal of phenolic compounds by ultrasonic force from the outer surface of the peeled apricot kernel. b* values of 36,76, 34.48, 35.44, 35.85 in microwave treatments, in turn, showed that the yellowness increased. In alkaline treatments and roasting processes, b* values were found close to the values of wild apricot (Figure 7).

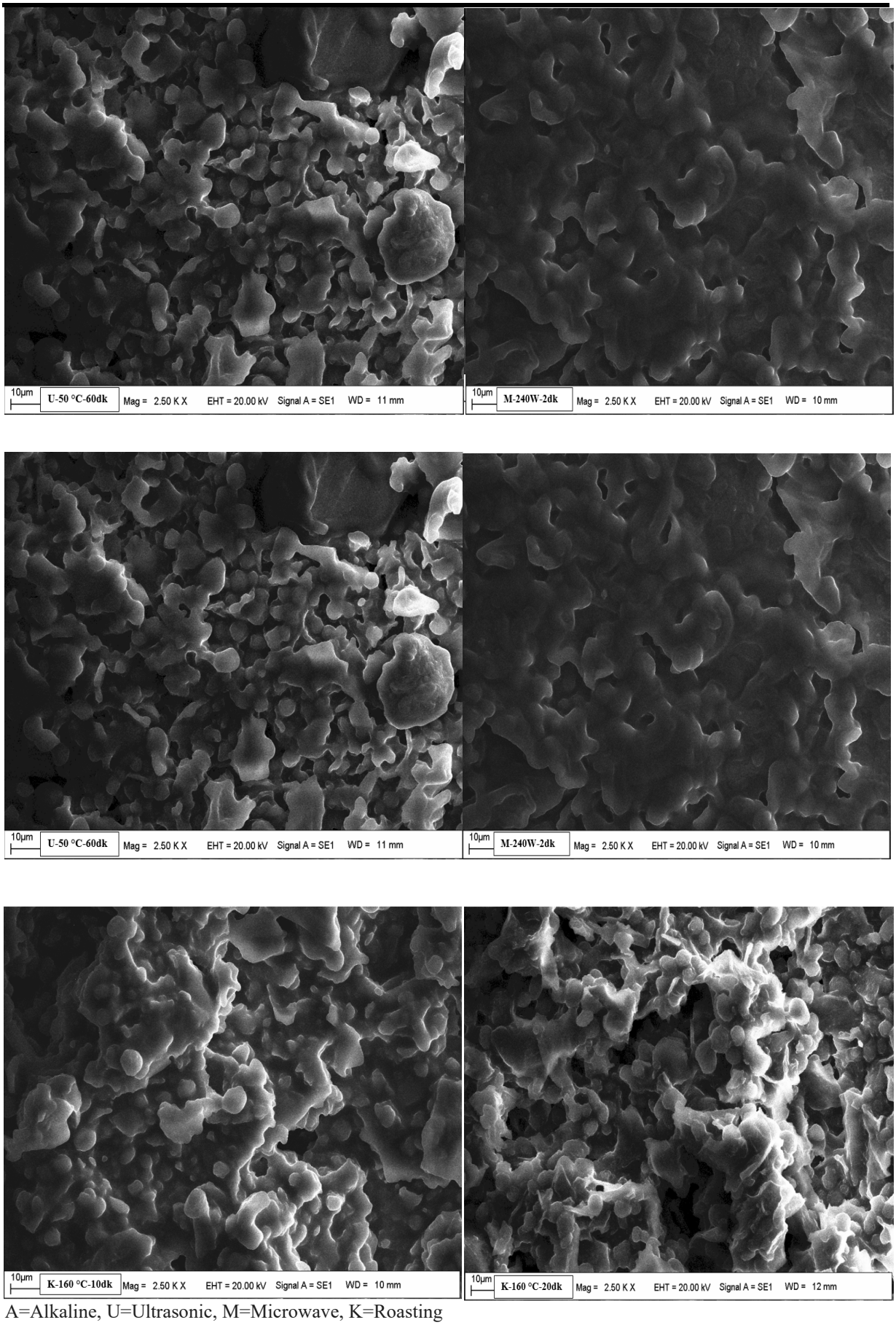
When the C* value of wild apricot was 22.58, it decreased in ultrasonic treatment (12.75, 8.59, 12.16, 5.06), while it decreased with roasting processes (25.41, 24.32, 27.87, 28.52) close to wild apricot and quite increased in microwave treatment (41.81, 39.5, 40.53, 41.50).

While the h° value was 59.97 for wild apricot, it was close to these values in alkaline treatments (60.04, 55.46, 56.7, 57.65) and roasting treatments (55.08, 52.18, 46.08, 51.61). A reduce was observed in ultrasonic treatment (42.23, 37.46, 36.28, 47.79) and an increase in microwave treatment (61.56, 60.82, 60.88, 59.82).

SEM (Scanning Electron Microscope) images of apricot kernels show the surface characteristics of these kernels in detail. SEM ensures high-resolution and three-dimensional imaging, therefore the microscopic structures of the surface, voids, cracks and other details are clearly seen.

SEM analysis of the surface properties of the kernels of the treatments for amygdalin reduction showed in detail the surface structures and voids with high-resolution, three-dimensional imaging. The structures removed from the raw wild apricot kernels by the treatments create voids and cracks.

When SEM images are analyze, it is seen that the fat cells in the structure protect their original form, that is, the processes carried out to reduce the amount of amygdalin are not enough to break down the cell structure. Similarly, in the FTIR spectrum, there is no change in the C=O stretching peaks of fatty acid around 1700 cm^{-1}. When the treated kernels are compared with the SEM of raw wild apricot, it is explicitly seen that there is no change. However, in the surface morphology of the treated ones, there are larger gaps between the units, which is explained by the removal of moisture, amygdalin and small organic compounds that are removed from the structure as a result of the processes (Figure 8).



A=Alkaline, U=Ultrasonic, M=Microwave, K=Roasting

Figure 8.SEM Image of Treatments

According to SEM images, when the amygdalin removal was compared for the difference between roasting processes, the amount of amygdalin reduced more in 160 ºC -10 min application than 160 ºC -20 min application. The large voids in the SEM images in the 160 ºC -20 min application promote that the amount of substance removed is higher than the 10 min application.

As seen in FTIR, in alkaline treatment, 2857-2932 symmetric and asymmetric CH stresses, in turn, 3294 OH stresses belonging to the cellulosic structure, 1744 C=O stresses belonging to the COOH group, and peaks belonging to C-O, C-O-C, M-O-M and M-O stresses at 1051 were drastically unchanged. 2222-2260 cm^{-1} is the C≡N stress belonging to amygdalin and remained the same in all three samples. Thus, no noticeable change was observed in the amygdalin analysis when the peaks of the applications were compared with the raw kernel peaks.

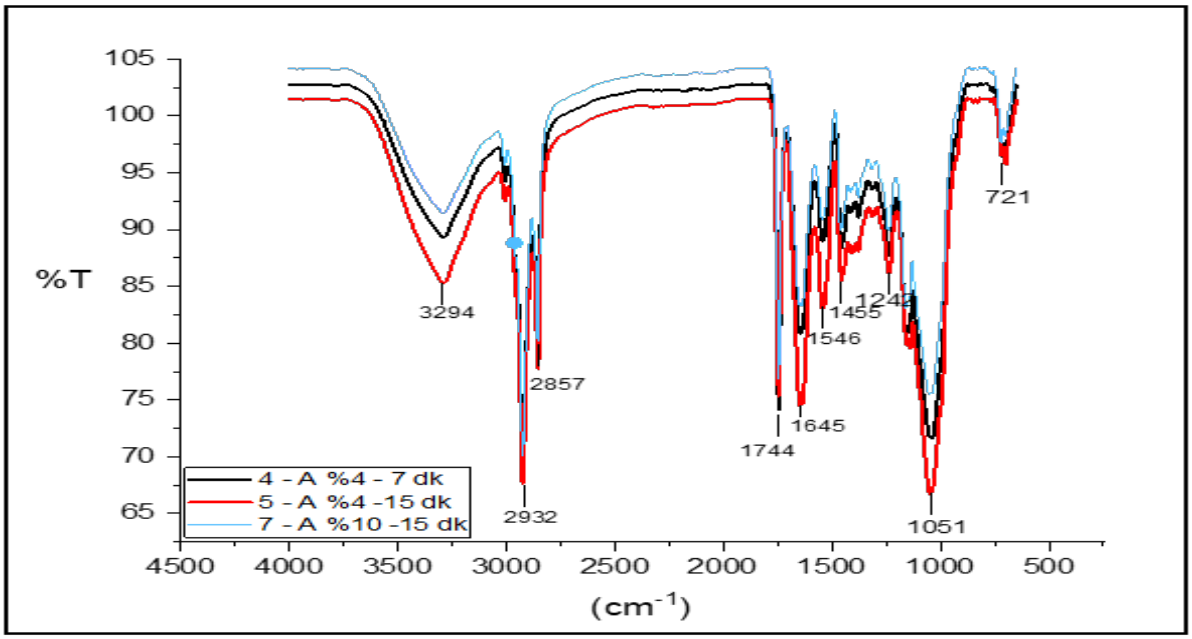


Figure 9. FTIR Results of Alkali Treatments

No significant change in the organic structure of the kernel was observed with ultrasonic treatment. However, a change in amygdalin values in the range of 27.64%- 2.31% was observed depending on temperature and time. The highest change was occured at 50ºC for 60 min. Although the temperature was close to the degradation temperature of amygdalin, the interaction between the liquid medium and the kernel organic structure as a whole remained very limited and a superficial amygdalin removal was observed according to mass balance.

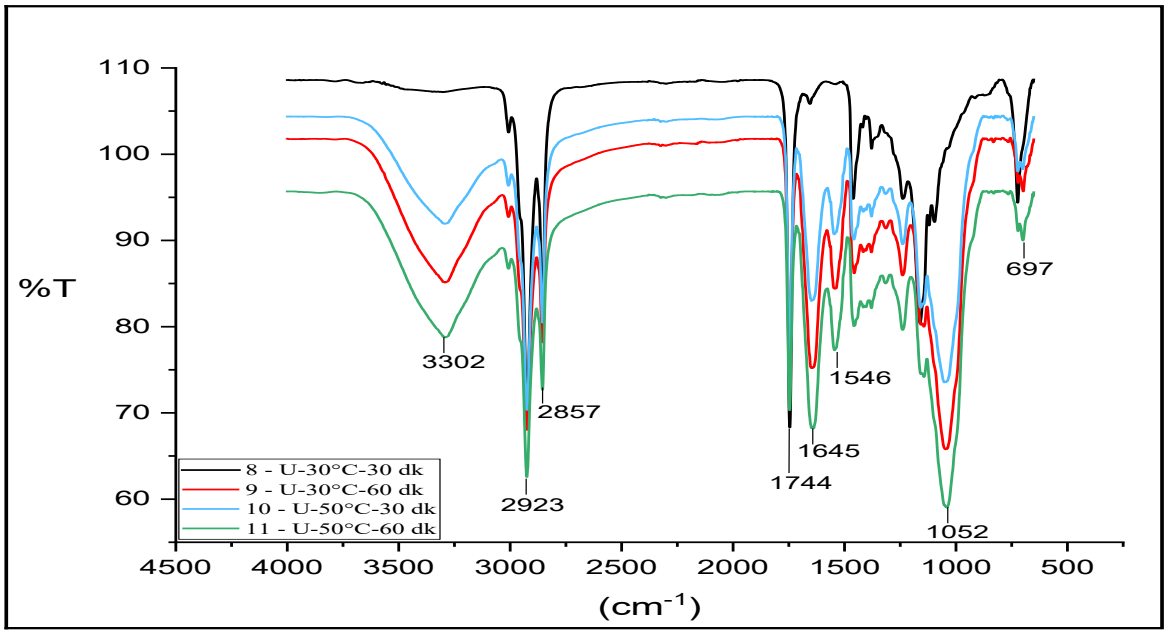


Figure 10. FTIR Results of Ultrasonic Treatments.

The decrease in the amount of amygdalin became more cleared when the amount of energy independent of time increased in microwave treatments. The change in amygdalin was in the range of 1.91%-6.55% by weight. In the FTIR spectrum, there was no significant change in all peaks and very limited change was observed in the amygdalin peak.

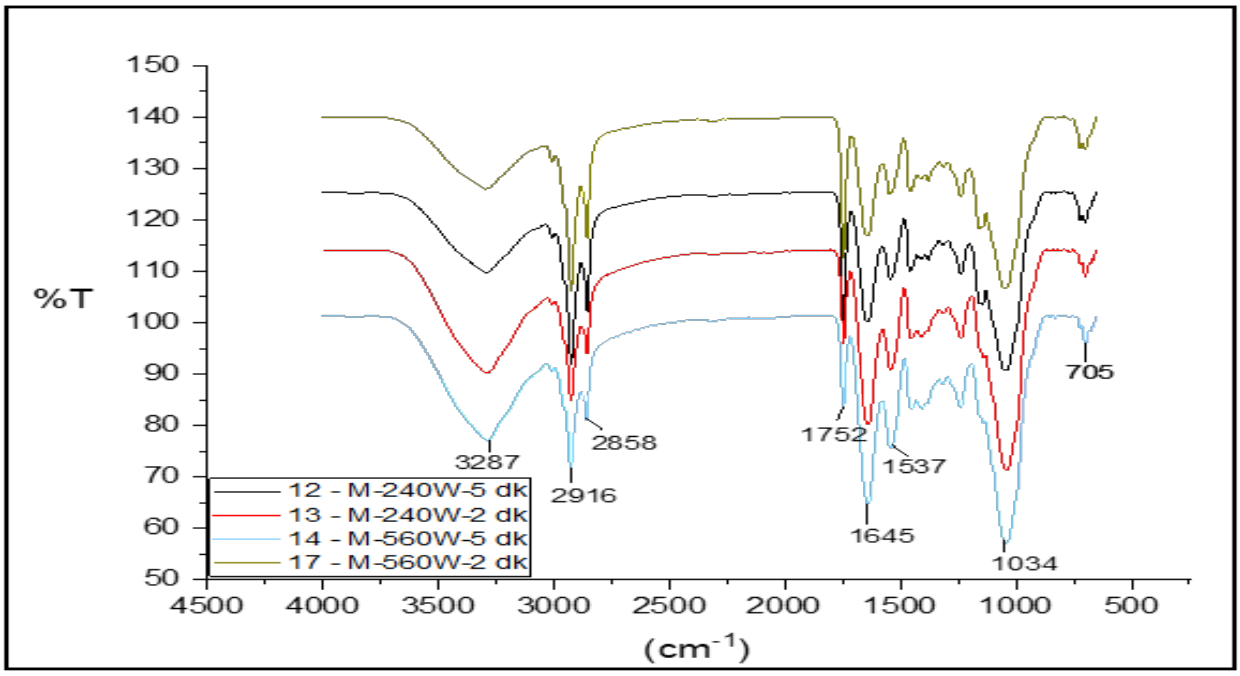


Figure 11. FTIR Results of Microwave Treatments.

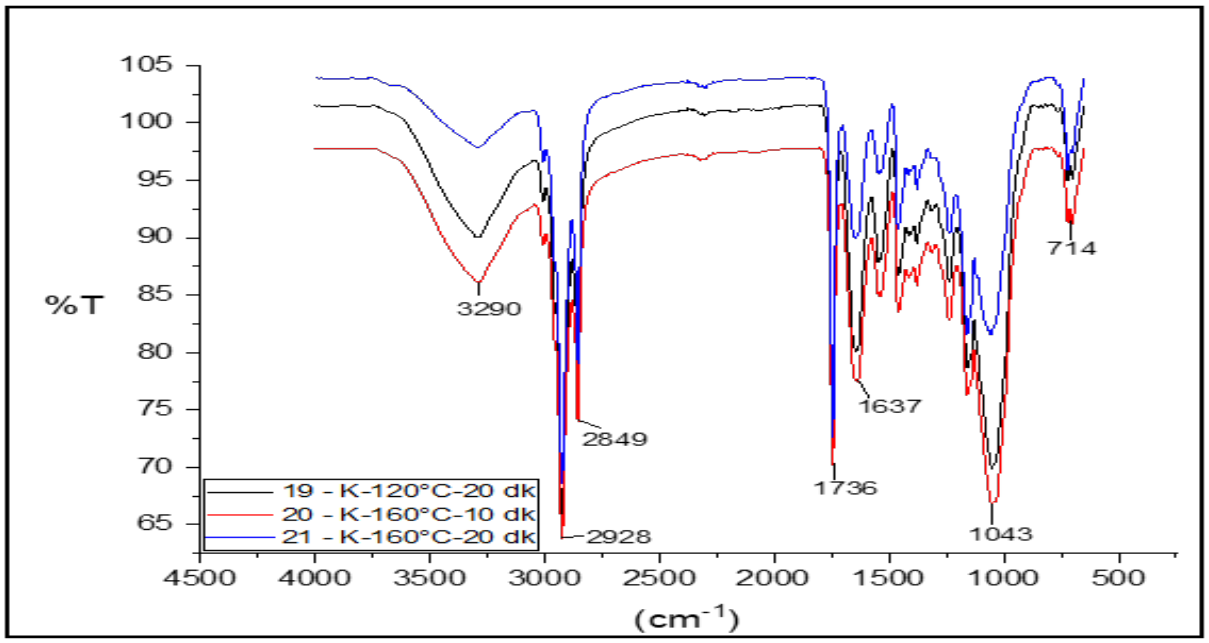


Figure 12. FTIR Results of Roasting Treatments.

Considering the results of the roasting treatment, a significant extent in the organic structure was observed. For 160 ºC-20 min, the OH and water peak at 3290 decreased significantly and the C-O, C-O-C peak at 1043 decreased significantly (Figure 12). This is explained by the change in the organic structure of the kernel near the surface. As can be seen, the amygdalin value decreased by 64.65% in 160ºC-10 min roasting and had a high change. The change in amygdalin peak is harmonious with these results.

FTIR analyses show that the chemical differences between ultrasound-assisted extraction and traditional extraction methods are limited and both methods involve similar functional groups. It is said that all extraction methods have characteristic bands corresponding to certain functional groups (Ali, AL-Hattap & Al-Haydry, 2015). It is harmonious with the functional groups found in our study.

CONCLUSION

In our study, appropriate treatments were selected to reduce the amount of amygdalin in wild apricot kernels, which had limitations in their assessment. Among these treatments, although the amount of amygdalin decreased the most in 160 ºC-10 min application, an exact amygdalin removal could not be succeed. In addition, it could not be brought to the safe consumption limits determined by EFSA. In this field, amygdalin removal should continue to be studied in order to develop new methods in addition to the bitterness removal processes carried out by traditional methods and to ensure their effectiveness.

Conflict of İnterest

The authors declared no conflicts of interest.

Funding

This study was supported by the General Directorate of Agricultural Research and Policies (TAGEM/HSGYAD/A/23/A3/P1/6209).

REFERENCES

Akyildiz, B. N., Kurtoğlu, S., Kondolot, M. & Tunç, A. (2010). Cyanide poisoning caused by ingestion of apricot seeds. *Ann Trop Paediatr.,* 30, 39-43.

Ali, M. A., Al-Hattab, T. A. & Al-Hydary, I. A. (2015). Extraction of date palm seed oil (Phoenix dactylifera) by soxhlet apparatus. *International Journal of Advances in Engineering & Technology*, 8(3), 261.

Alpaslan, M. & Hayta, M., (2006). Apricot kernel: Physical and chemical properties. *Journal Of The American Oil Chemists Society* , 83(5), 469-471.

Atiş, E. (2017). Apricot producing in Kağızman district and its contribution to the economy of territory. *Marmara Geographical Review,* 36, 191-205.

Badr, S. E., Ramis, E. S., Wahdan, O. A., Sakr, D. M. & Elghandour, H. M. (2017). Evaluation of protein quality, phytochemical characterization and the effect of soaking and roasting processes on raw apricot kernels. In The first international conference of Nutrition, Hurghada, *The Egyptian Nutrition Society*, 253-287.

Bae, S. H. & Suh, H. J. (2007). Antioxidant activities of five different mulberry cultivars in Korea. *LWT-Food Sci Technol*, 40(6), 955-62.

Bolarinwa, I. F., Orfila, C. & Morgan, M. R. A. (2014). Amygdalin content of seeds, kernels and food products commercially-available in the UK. *Food chemistry*, 152, 133-139.

Bolarinwa, I. F., Orfila, C. & Morgan, M. R. A. (2015). Determination of amygdalin in apple seeds, fresh apples and processed apple juices. *Food Chemistry* 170:437-442. doi: 10.1016/j.foodchem.2014.08.083.

Cemeroglu, B. (2010). Gıda analizlerinde genel yöntemler, Gıda Analizleri. Ankara: Gıda Teknolojisi Yayınları.

Doğan, A. & Başoğlu, F. (1985). Yemeklik bitkisel yağ kimyası ve teknolojisi uygulama kılavuzu. *A. Ü. Ziraat Fakültesi Yayınları*, 951. 62.

EFSA (European Food Safety Authority). (2011). Use of the EFSA comprehensive European food consumption database in exposure assessment. *EFSA Journal*, 9(3), 2097, 34. doi: 10.2903/j.efsa.2011.2097

EFSA Scientific Committee. (2012). Guidance on selected default values to be used by the EFSA Scientific Committee, Scientific Panels and Units in the absence of actual measured data. *EFSA Journal*,102579, 32 doi: 10.2903/j.efsa.2012.2579

FAO/WHO (Food and Agricultural Organization/World Health Organization). (2012). Safety evaluation of certain food additives and contaminants prepared by the seventy-fourth meeting of the joint FAO/WHO expert committee on food additives. *WHO Food Additives Series*, 65, 1-833.

Feng, D., Shen, Y. & Chavez, E.R. (2003). Effectiveness of different processing methods in reducing hydrogen cyanide content of flaxseed. *Journal of the Science of Food and Agriculture*, 83(8), 836-841. https://doi.org/10.1002/jsfa.1412

FSANZ (Food Standards Australia New Zealand). (2008). Proposal P1002 – Hydrocyanic acid in ready-to-eat cassava chips. Assessment Report. 6 March 2008. FSANZ, Canberra. Available online: http://www.foodstandards. gov.au/code/proposals/Pages/proposalp1002hydrocy3848.aspx.

FSANZ (Food Standards Australia New Zealand). (2014). Survey of cyanogenic glycosides in plant-based foods in Australia and New Zealand, 1-78.

García, M. C., González-García, E., Vásquez-Villanueva, R. & Marina, M. L. (2016). Apricot and other seed stones: Amygdalin content and the potential to obtain antioxidant, angiotensin I converting enzyme inhibitor and hypocholesterolemic peptides. *Food & function*, 7(11), 4693-4701. doi: 10.1039/C6FO01132B

GKGM- Risk Değerlendirme Daire Başkanlığı. (2022).

Gönül, M., Altuğ, T., Boyacıoğlu, D. ve Noka, Ü. (1988). Gıda Analizleri. İzmir: Ege Üniversitesi Mühendislik Fakültesi Çoğaltma Yayın No:64.

Juhaimi, F. A., Özcan, M. M., Ghafoor, K. & Babiker, E. E. (2018). The effect of microwave roasting on bioactive compounds, antioxidant activity and fatty acid composition of apricot kernel and oils. *Food Chemistry*, 243, 414-419. https://doi.org/10.1016/j.foodchem.2017.09.100

Kacar, B. (1984). Bitki Besleme ve Uygulama Kılavuzu. Ankara.

Karsavuran, N., Charehsaz, M., Celik, H., Asma, B. M., Yakıncı, C. & Aydın, A. (2014). Amygdalin in bitter and sweet seeds of apricots. *Toxicological & Environmental Chemistry*, 96(10), 1564-1570. https://doi.org/10.1080/02772248.2015.1030667

Kaya, A., Okur, M., Üstyol, L., Temel, H. & Çaksen, H. (2012). Kayısı çekirdeği yeme sonrası akut siyanür zehirlenme olgusu. *Türk Pediatri Arşivi*, 47(2), 141-142. https://doi.org/10.4274/tpa.2122

Kılıç, H. M. (2015). Endüstriyel bir atık olarak hurma çekirdeği; kavurma prosesinin hurma çekirdeği unu ve hurma çekirdeği kahvesinin antioksidan kapasitesi üzerine etkisi (Master tezi). Fen Bilimleri Enstitüsü.

Kumaran, A. & Karunakaran R.J. (2006). Antioxidant and free radical scavenging activity of an aqueous extract of Coleus aromaticus. *Food Chemistry*, 97(1): 109-114, doi: 10.1016/ j. foodchem.2005.03.032

Latif, S. & Müller, J. (2015). Potential of cassava leaves in human nutrition: A review. *Trends in Food Science & Technology,* 44(2), 147-158. https://doi.org/10.1016/j.tifs.2015.04.006

Lee, J., Zhang, G., Wood, E., Castillo, C. R. & Mitchell, A. E. (2013). Quantification of amygdalin in nonbitter, semibitter, and bitter almonds (Prunus dulcis) by UHPLC-(ESI)QqQ MS/MS. *Journal of Agricultural and Food Chemistry*, 61, 7754-7759.

Mc Guire, R.G. (1992). Reporting of objective color measurements. *Hort Science,* 27, 1254-1255.

Milazzo, S., Ernst, E., Lejeune, S. & Boehm, K. (2006). Laetrile treatment for cancer. *Cochrane Database of Systematic Reviews,* (2). doi: 10.1002/14651858.CD005476.pub3

Mutlu, M. & Hayaloglu, A. A. (2022). Determination of bioactivity of seed protein hydrolysates and amygdalin content for some apricot (*Prunus armeniaca* L.) varieties grown in Malatya, Turkey. *Journal of Raw Materials to Processed Foods*, 3(1), 10-19.

Noreen, S., Tufail, T., Ul Ain, H. B. & Awuchi, C. G. (2023). Pharmacological, nutraceutical, and nutritional properties of flaxseed (*Linum usitatissimum*): An insight into its functionality and disease mitigation. *Food Science & Nutrition*, 11(11), 6820-6829. https://doi.org/10.1002/fsn3.3662

Panghal, A., Munezero, C., Sharma, P. & Chhikara, N. (2021). Cassava toxicity, detoxification and its food applications: a review. *Toxin Reviews,* 1-16. https://doi.org/10.1080/15569543.2018.1560334

Poyraz, N. (2013). Malatya yöresinde yetişen kayısı türlerinin tohumlarında amigdalin miktarının HPLC yöntemiyle belirlenmesi. (Uzmanlık Tezi). İnönü Üniversitesi Tıp Fakültesi, Çocuk Sağlığı ve Hastalıkları AD.

Saini, D., Rawat, N., Negi, T., Barthwal, R. & Sharma, S. K. (2021). Utilization, valorization and functional properties of wild apricot kernels. *Journal of Pharmacognosy and Phytochemistry*, 10(4), 119-126.

Sheikh, M. A. & Saini, C. S. (2022). Combined effect of microwave and hydrothermal treatment on anti-nutritional factors, antioxidant potential and bioactive compounds of plum (*Prunus domestica* L.) kernels. *Food Bioscience,* 46, 101467. http://dx.doi.org/10.1016/j.fbio.2021.101467

Shen, Y., Feng, D., Fan, M. Z. & Chavez, E. R. (2005). Performance, carcass cut-up and fatty acids deposition in broilers fed different levels of pellet-processed flaxseed. *Journal of the Science of Food and Agriculture*, 85(12), 2005-2014. doi:10.1002/jsfa.2155

Slinkard, K. & Singleton, V. L. (1977). Total phenol analyses: Automation and comparison with manual methods. *Am. J. Enol.* Vitic, 28, 49-55.

Şahin, H., Topuz, A., Pischetsrieder, M. ve Özdemir, F. (2009). Kavurma işleminin harnup tozunun fenolik, antioksidan ve esmerleşme özellikleri üzerine etkisi. *Avrupa Gıda Araştırma ve Teknolojisi*, 230, 155-161.

Uğur, Y. & Yaman, R., (2022). Determination of Aflatoxin in Apricot Kernel with UFLC-FD Method and In-Laboratory Method Validation. *Journal of the Institute of Science and Technology*, 12(3), 1734-1742. https://doi.org/10.21597/jist.1086858

Thaipong, K, Boonprakob, U., Crosby, K., Cisneros-Zevallos, L. & Hawkins Byrne, D. (2006). Comparison of ABTS, DPPH, FRAP, and ORAC assays for estimating antioxidant activity from guava fruit extracts. *J Food Compos Anal,* 19(6-7), 669-75. https://doi.org/10.1016/j.jfca.2006.01.003

Turan, S., Atalay, D., Solak, R., Özoğul, M. ve Demirtaş, M. (2021). Ultrasonik destekli ekstraksiyon parametrelerinin kuşburnu (*Rosa Canına* L.) meyvesinin toplam fenolik ve karotenoid miktarları ile antioksidan aktivitesi üzerine etkisi. *Gıda,* 46(3), 726-738.

Zengin, R., Maraş, Z., Uğur, Y., Özhan, O., Karaat, F. E. & Erdoğan, S. (2024). Determination of phytochemical composition in fruits and leaves from different origins: Black Mulberry, Chokeberry and Elderberry genotypes. *Analytical Letters*, 1-23. https://doi.org/10.1080/00032719.2024.2324379

Zhang, Q. A., Shi, F. F., Yao, J. L. & Zhang, N. (2020). Effects of ultrasound irradiation on the properties of apricot kernels during accelerated debitterizing. *RSC advances,* 10(18), 10624-10633. https://doi.org/10.1039%2Fc9ra10965j