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Research Article

Variation of components in laurel (*Laurus nobilis* L.) fixed oil extracted by different methods

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ABSTRACT

In the study, it was aimed to determine the constituents of laurel fixed oil obtained from the different genotypes of laurel (*Laurus nobilis* L.), which is one of the natural plants of the region and which is widely found in the flora of Hatay, by traditional, cold press and soxhlet extraction methods. When the GC/MS analysis results of these obtained oils were examined, the main components of the fixed oils in the traditional method were found as capric acid (2.49%), lauric acid (1.17%), myristic acid (0.16%), palmitic acid (13.69%), stearic acid (2.39%).), oleic acid(55.01%), linoleic acid (10.56%) and linolenic acid(0.11%). In cold press method, fixed oil components was capric acid (0.24%), lauric acid(9.24%), myristic acid(0.98%), palmitic acid (18.41%), stearic acid (2.84%), oleic acid (38.59%), linoleic acid (23.67%) and linolenic acid (2.19%), while it was determined as capric acid (0.46%), lauric acid (11.16%), myristic acid (1.54%), palmitic acid (18.39%), stearic acid (3.58%), oleic acid (36.92%), linoleic acid (23.02%) and linolenic acid (2.54%) in soxhlet extraction method. As a result, while the components of laurel fixed oil did not change according to the fixed oil extraction methods, the amounts of these components changed. Therefore, it was determined that the method of oil extraction in laurel was important.

Keywords: Laurel, *Lauris nobilis* L., GC-MS, Hatay, aromatic.

Farklı yöntemlerle çıkarılan defne (*Laurus nobilis* L.) sabit yağ bileşenlerindeki değişim

ÖZ

Çalışmada Hatay florasında yoğun bir şekilde bulunan ve bölgenin doğal bitkilerinden biri olan defne (*Laurus nobilis* L.) genotiplerinden geleneksel, soğuk sıkım ve soxhlet ekstraksiyonu yöntemleri ile elde edilen defne sabit yağı bilileşenlerinin belirlenmesi amaçlanmıştır. Elde edilen bu yağların GC/MS analiz sonuçları incelendiğinde sabit yağların ana bileşenleri geleneksel yöntemde, kaprik asit (%2.49), laurik asit(%1.17), myristic asit(%0.16), palmitik asit(%13.69), stearik asit(%2.39), oleik asit(%55.01), linoleik asit (%10.56), linolenik asit(%0.11), soğuk preste, kaprik asit (%0.24), laurik asit(%9.24), myristic asit(%0.98), palmitik asit(%18.41), stearik asit(%2.84), oleik asit(%38.59), linoleik asit (%23.67) ve linolenik asit (%2.19) ve soxhlet ekstraksiyonunda kaprik asit (%0.46), laurik asit(%11.16), myristic asit(%1.54), palmitik asit(%18.39), stearik asit(%3.58), oleik asit(%36.92), linoleik asit (%23.02) ve linolenik asit (%2.54) olduğu tespit edilmiştir. Sonuç olarak defne sabit yağının bileşenleri sabit yağ çıkarama yöntemlerine göre değişmezken bu bileşenleri miktarları değişiklik göstermiştir. Bu yüzden defnede yağ çıkarma yönteminin önemli olduğu tespit edilmiştir.

Anahtar Kelimeler: Defne, *Laurus nobilis* L., Hatay, aromatik.

1. INTRODUCTION

From the past to present, people have used plants for therapeutic purposes either by chance or as a result of curiosity, in order to find a cure for diseases. In recent years, medicinal and aromatic plants, which have created awareness all over the world, have become an important focus of attention in Turkey as well. Medicinal and aromatic plants produce active substances that provide drugs to humans to prevent diseases, maintain health, and treat ailments. Essential and fixed oils obtained from leaves and fruits are used in domestic and foreign markets rather than the direct use of bay leaves and fruits, which have an important place in our foreign trade. Laurel and

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its products are used in many fields such as food, medicine, cosmetics and chemistry. In addition, both domestic and foreign trade of laurel products is increasing recently in Turkey.[1]

Bay leaves has compelling features as antioxidant, antiseptic, antibacterial, anti-inflammatory, anticonvulsant, antifungal, analgesic, diaphoretic, anti-migraine, relieving stomach ailments and treating diabetes. In addition, It has been shown in many studies that the laurel plant is also good for diseases such as weakness, indigestion, insomnia, menstrual irregularities and rheumatism.[2-9] The fixed oil extracted from the anthocyanin fruit of the laurel is used in cosmetic medicine and food industries. It is also considered as a natural dyestuff.[10-13] The origin of the laurel plant is the Mediterranean countries according to some references while in some references its orgin is Minor Asia (Anatolia) and Balkans. Our country is very suitable for the cultivation of many cultural plants in terms of climatic conditions, in addition, the rugged topography contributed to the crop aroma, taste, quality and yield characteristics of plants.[14-17] There are 45 genera and 1000 species in the Lauraceae family. There are 2 species belonging to the genus Laurus and they are *L. nobilis* and *L. angustifolia* and *L. angustifolia* has 4 subspecies.[18-21]

Turkey is a country with a significant potential to meet the demand for laurel oil production and its products. Laurel fruit oil is commonly produced in Hatay and exported abroad, either directly as oil or to be used in soap making. 20% of the production of laurel berry oil, which contains some volatile components and is called fixed oil and used in the soap industry.[1,22]

Hafizoglu and Reunanen[23] stated that there are more than 20 fatty acids in laurel fruits and the main components are lauric acid (54.2%), oleic acid (15.1%) and linoleic acid (17.2%). Ayanoglu and co-workers[24] determined that average value of lauric acid was 16.57%, palmitic acid was 18.57%, oleic acid was 38.08%, and linoleic acid was 23.90% in the fruits of 145 female laurel plants collected from different locations in Hatay. Beis and Dunford[25] used supercritical and petroleum ether extracts of laurel fruit oils obtained from Turkey, and the main components were found as lauric acid (43.1-44.8%), oleic acid (37.2 – 37.3%) and linoleic acid (14.7%-13.3%). On the other hand, Yazıcı (2002) determined that components of laurel fixed oil extracted using hexane as a solvent were lauric acid (12.31-14.96%), oleic acid (44.12-45.90%), linoleic acid (21.97-23.05%) and palmitic acid (14.39-14.86%) . The researcher also reported that as the altitude increases in the region, the ratio of saturated fatty acids decreased, while the ratio of unsaturated fatty acids increased. Timur[26] determined that laurel fixed oil components were 8.5-13.0% for lauric acid, 13.1-20.8% for palmitic acid, 36.3-48.3% for oleic acid and 52.8-29.9 for linoleic acid. Castilho and co-workers[27] reported that laurel fruit contains 30% oleic acid, 20% linoleic acid, 18% lauric acid to 22.5% palmitic acid and 84% β-sterol. In a study conducted by Erden [28], laurel fruits were harvested every week from October to December and dried in the sun to extract the oil by soxallet extraction method. Petroleum ether was used as solvent in this study, and the highest oil yield was obtained at the end of December with 25.55% (mass/mass). Nurbaş and Bal[29] investigated the effects of hexane, petroleum ether, carbon sulfide and benzene used as solvents on the efficiency of the soxallet extraction method in oil extraction from laurel fruit. In the trials, used hexane as solvent has the highest efficiency value with 32.12% fixed oil rate. Koçer and Ayanoğlu[9] determined that the main components of fruit and seed fixed oils of laurel found as lauric acid, oleic acid, palmitic acid and linoleic acid. They reported that lauric acid was not found in fruit flesh.

2. MATERIALS AND METHODS

In this study, fruit fixed oil of laurel genotypes was extracted by traditional, cold pressing and soxhlet extraction methods. The traditional method was made according to the boiling method performed by the region villagers. For the cold pressing and extraction method, laurel fruits were firstly dried in an oven at 70 °C. In the study, the extraction method is based on the principle of removing the solvent as a result of the extraction of the sample with solvent (hexane) and weighing the remaining residue (fixed oil). The laurel fruits, which were dried in an oven, were ground by a grinder and 5 g of sample was used in the extraction method. Hexane was used as the solvent in the extraction method. At the end of the extraction, the beaker containing oil was taken and kept in an oven set at 80 ºC for 1 hour.[30,31]

100 µl of oils obtained from laurel fruits were taken, 3 ml of N-Heptane and 400 µl of 2N methanolic KOH solution were added and esterification was applied, and the components of the oils were analyzed by GC/MS. Determination of fixed oil components was performed with Thermo Scientific ISQ Single Quadrupole model gas chromatography device under the following conditions. TR-FAME MS model, 5% Phenyl Polysilphenylene-silohexane, 0.25 mm inner diameter x 60 m length, 0.25 µm film thickness was used. Helium (99.9%) was used as the carrier gas at a flow rate of 1 mL/min. The ionization energy was set to 70 eV, the mass range m/z 1.2-1200 amu. Scan mode was used for data collection. The MS transfer line temperature was 250°C, the MS ionization temperature was 220°C, the injection port temperature was 220°C, the column temperature was initially 120°C and kept there for 1 min. It was increased to 175 °C by increasing 10 °C per minute and kept there for 10 minutes. It was increased to 210 oC by increasing 5 ºC per minute and kept there for 5 minutes. Then, it was increased to 230 ºC by increasing 5 ºC per minute, and the analysis was concluded by

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keeping it here for 6 minutes. The total analysis time is 38.5 minutes. The structure of each compound was defined using the Xcalibur program using mass spectra (Wiley 9).[32]

3. RESULTS AND DISCUSSION

3.1. Traditional Method

When the fatty acid components of the oils extracted by the traditional boiling method from the laurel fruits (seeds) by the villagers living in Hatay were examined, 8 main components were determined (Table 1). The chromatogram of these components is shown in Figure 1. The highest value among these components was found as oleic acid with a rate of 55.01%. Also, other components were determined as palmitic acid (13.69%), linoleic acid (10.56%), capric acid (2.49%), stearic acid (2.39%), lauric acid (1.17%), myristic acid (0.16%) and linolenic acid (0.11%).

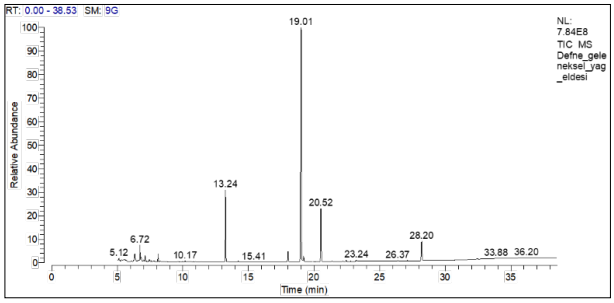

Figure. 1. *Laurus nobilis* L. (Laurel) fixed oil chromatogram obtained by traditional method.

Table 1. *Laurus nobilis* L. (Laurel) fixed oil components and values obtained by traditional method.

RT	Compound Name	CAS	Area (%)
6:72	Kaprik asit, C10	110-42-9	2.49
8:10	Laurik asit, C12	111-82-0	1.17
10:17	Myristic asit, C14	124-10-7	0.16
13:24	Palmitik asit, C16	112-39-0	13.69
18:01	Stearik asit, C18:0	112-61-8	2.39
19:01	Oleik asit, C18:1	112-62-9	55.01
20:52	Linoleik asit C18:2	112-63-0	10.56
22:20	Linolenik asit, C18:3	301-00-8	0.11

As seen in Table 1, Laurel fixed oil components and their quantities were similar to literature reports.[9,23-26] In addition fixed oil component quantities was different from each other. The reason for this may be that lauric acid is found in the fruit seed, as stated in previous studies, and that the fixed oil in the fruit seed cannot be completely removed in the traditional method.

3.2. Cold Press method

When the fatty acid components of the oils extraced from laurel fruits by cold pressing method were examined, 8 main components were determined (Table3). The chromatogram results of these components is shown in Figure 3. The highest value among these components was found as oleic acid with a rate of 38.59% similar to the traditional method. However, oleic acid ration in cold pressing method was lower than traditional method. Other main components in cold pressing method were determined as linoleic acid (23.67%), palmitic acid (18.41%), lauric acid (9.54%), stearic acid (2.84%), linolenic acid (2.19%), myristic acid (0.98%) and capric acid (0.24%). In cold pressing method, some components were lower than traditional method while some components was higher.

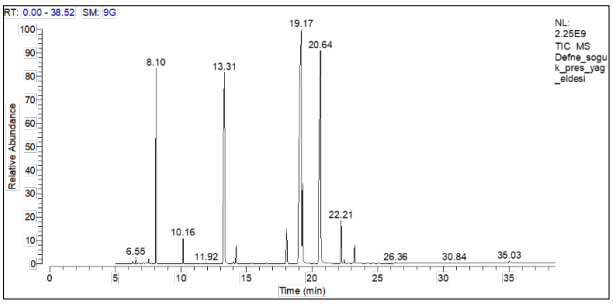

Figure 2. *Laurus nobilis* L. (Laurel) fixed oil chromatogram obtained by cold press method.

Table 2. *Laurus nobilis* L. (Laurel) fixed oil components and values obtained by cold press method.

RT	Compound Name	CAS	Area (%)
6:55	Kaprik asit, C10	110-42-9	0.24
8:10	Laurik asit, C12	111-82-0	9.54
10:16	Myristic asit, C14	124-10-7	0.98
13:31	Palmitik asit, C16	112-39-0	18.41
18:06	Stearik asit, C18:0	112-61-8	2.84
19:16	Oleik asit, C18:1	112-62-9	38.59
20:63	Linoleik asit C18:2	112-63-0	23.67
22:21	Linolenik asit, C18:3	301-00-8	2.19

As seen in Table 2, laurel fixed oil extracted by cold press method was similar to some research findings.[9,23-26] On the other hand, component quatities were different from each other. The difference in the amounts of the components can be attributed to both plant genetic structures and climatic conditions.

3.3. Soxhlet extraction

When the fatty acid components of the oils extracted from the laurel berries were examined, 8 main

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components were found (Table3). The chromatogram results of these components are shown in Figure 3. The highest value among these components was found as oleic acid with a rate of 36.92% similar to the traditional and extraction methods. But, oleic acid value changed according to the methods. Other main components were determined as linoleic acid (23.02%), palmitic acid (18.39%), lauric acid (11.16%), stearic acid (3.58%), linolenic acid (2.54%), myristic acid (1.54%) and capric acid (0.46%).

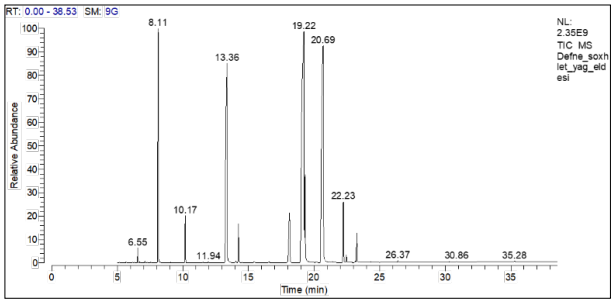

Figure 3. Fixed oil chromatogram of *Laurus nobilis* L. (Laurel) obtained by soxhlet extraction method.

Table 3. *Laurus nobilis* L. (Laurel) fixed oil components and values obtained from soxhlet extraction.

RT	Compound Name	CAS	Area (%)
6:55	Kaprik asit, C10	110-42-9	0.46
8:11	Laurik asit, C12	111-82-0	11.16
10:17	Myristic asit, C14	124-10-7	1.54
13:36	Palmitik asit, C16	112-39-0	18.39
18:12	Stearik asit, C18:0	112-61-8	3.58
19:22	Oleik asit, C18:1	112-62-9	36.92
20:69	Linoleik asit C18:2	112-63-0	23.02
22:23	Linolenik asit, C18:3	301-00-8	2.54

As seen in Table 3, laurel fixed oil components were similar to literature reports[9,23-26] and component quatities were different from each other. The difference in the amounts of the components can be attributed to both plant genetic structures and climatic conditions.

4. CONCLUSION

When the GC/MS analysis results of traditional cold pressing and soxhlet extraction methods are examined, the main components of fixed oils were determined as capric acid (2.49-0.24-0.46%, respectively), lauric acid (1.17-9.54-11.16%, respectively), myristic acid (0.16-0.98-1.54%, respectively), palmitic acid (13.69-18.41-18.39%, respectively), stearic acid (2.39-2.84-3.58%, respectively), oleic acid (55.01-38-59-36.92%, respectively), linoleic acid (10.56-23.67-23.02%, respectively), linolenic acid (0.11-2.19-2.54%, respectively).

When these results were investigated, it was seen that the amount of lauric acid was less and the amount of oleic acid was higher in the traditional method compared to the other methods. The reason for this is thought to be due to the absence of lauric acid in the fruit flesh and for this reason, not all of the fixed oil in the fruit could be extracted with the traditional method. Since the laurel plant is not cultivated, the production of standard raw materials from this plant is restricted. In addition, unconscious harvesting and excessive slaughter also lead to the destruction of genetic resources.

The production of laurel, which is an important export product, should be increased and its planting should be encouraged for a quality production. Especially in the new afforestation areas, the laurel plant should be emphasized in the re-establishment of the degraded and burned forest areas. In the new agricultural facilities to be established for the production of laurel oil and laurel leaves, as well as in the reforestation of forest areas, plantings should be made with selected, high-quality types, and laurel leaf production.

Conflict of interest

The authors declare that there is no conflict of interest.

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