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Determination of Physicochemical Properties of *Nigella sativa* Seed Oil from Balıkesir Region, Turkey

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Abstract

Black cumin seed (*Nigella sativa*) and its oil has been used as a dietary supplement and a preferred traditional remedy in Africa, Asia and Middle East for centuries and is now consumed by millions of people in America and Europe. Scientists are advising consumption of black cumin seed because of the vitamins, effective compounds and the essential fatty acids within its content which have been found in researches. Cold pressed black cumin seed (*Nigella sativa*) oil is used in the European Union and other developed countries as natural supplements and/or phytotherapeutic agents to support the immune system, treat asthma, allergic rinnitis, diabetes etc. In this study the aim is to identify in a scientific way an acceptable alternative source for the *Nigella sativa* seeds which are used in foods, natural supplements and therapeutical applications. With this purpose in the study oil was extracted from Nigella sativa seeds by cold press method without applying heat or chemicals. Physicochemical properties of *Nigella sativa* seed oil from Balıkesir Area, Turkey (peroxide value, free fatty acids, fatty acid composition, refractive index, iodine value, soap value, saponification value, unsaponifiable matters) have been determined and followed at 25 degrees Celsius % 60 RH for 8 months. As a result it is found that *Nigella sativa* oil‘s physicochemical properties from the Balıkesir area in Turkey are consistent with the general physicochemical properties of *Nigella sativa* seed oil which is used for the food industry and supplements.

Keywords: *Nigella sativa*, Black cumin seed oil, cold press

1. Introduction

Nigella sativa, a plant originally native to Southern Europe, North Africa, and Southwest Asia, is today cultivated in many countries in the world, including the Middle Eastern Mediterranean region, Southern Europe, India, Pakistan, Syria, Turkey, and Saudi Arabia(Watson,2015, Ahmad, 2013, El-Tahir and Bakeet, 2006). The seeds are the most important plant part, and have been used since time immemorial in various traditional systems of medicine, such as Unani-Tibb, Ayurveda, and Siddha, to treat various ailments (Ahmad, 2013). The seeds and oil have a long history of folklore usage, being widely used as an antihypertensive; as a liver tonic, diuretic, digestive, antidiarrheal, appetite stimulant, and antibacterial; for skin disorders; and as an analgesic (Ahmad, 2013). Scientific studies carried out in the recent past have validated the ethnomedicinal uses, and reports indicate it to possess antidiabetic, anticancer, immunomodulatory, analgesic, antimicrobial, anti-inflammatory, spasmolytic, bronchodilator, hepatoprotective, renal-protective, gastroprotective, antioxidant, and anticancer properties (Khan,2011). *Nigella sativa* (black cummin, schwarz kümmel) belongs to Ranunculaceae familia and the plant is indigenous to Mediterranean areas, through it is grown in other parts of the world as well (Ramadan, 2007; Cemek et Al., 2008).

2. Material Preparation

2.1 Black Cumin (Nigella sativa) Seed

Black cumin seed was obtained from different domestic sources and evaluated. *Nigella sativa* samples from the

Balıkesir area of Turkey were used for the study considering the herbarium studies results and effectivenes of cold press extraction method. Nigella sativa seeds had been supplied from the manufacturer free of dust and impurities.

2.2. Cold Press Extraction Process

The cold press extraction method is used for the extraction of oil from *Nigela sativa* seeds. The temperature was kept below 40 degrees Celsius and no chemical and heating process was used for the process to protect physicochemical properties of oil. The cold press oil machine head diameter has been chosen at 8 mm and the operation speed was set at 17 rpm. Oil has stored in stainless steal drums for one day to settled out sediments. The next day the oil was purified by using 1 µ pores filtration paper. At the last step the *Nigella sativa* oil was filled to 200 mL amber glass bottles and kept at 25 degrees Celsius % 60 RH for 8 months. The analysis are conducted on a monthly basis.

2.3. Analysis

2.3.1. Physicochemical Analysis

Nigella sativa oil has been analysed for the free fatty acid (% Oleic acid) (Ph. Eur. 8.0/2.5.1), Refractive Index (Ph. Eur. 8.0/2.2.6) , Peroxide Value (Ph. Eur. 8.0/2.5.5), Unsaponification Matter (Ph. Eur. 8.0/2.5.7), Saponification Value (Ph. Eur. 8.0/2.5.6), Iodine Value (Ph. Eur. 8.0/2.5.4), Relative Density (Ph. Eur. 8.0/2.2.5) .

Table 1. Physicochemical Properties of *Nigella sativa* Oil

	Determined Values by Months								Valuee in the Literature
ANALYSIS	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	
FFA (%)(Oleic Acid)	2.86	2.86	2.82	2.85	2.84	2.95	3.05	3.1	7.49 – 22.7
Peroxide (meqO_2/kg)	25.9	25.7	25.9	26.5	26.8	29	31	32	4.35 – 35.32
Refractive index 20ºC	1.4737	1.4737	1.4737	1.4737	1.4737	1.4737	1.4737	1.4737	1.4732 - 1.4738
Refractive index 40ºC	1.4664	1.4664	1.4664	1.4664	1.4664	1.4664	1.4664	1.4664	1.45 – 1.48
Soap (ppm)	0	0	0	0	0	0	0	0	0
Iodine value	122	122	122	122	122	122	122	122	98 – 122
Saponification Value (mg KOH/g)	194	194	195	194	194	194	194	194	190 – 226
Unsaponifiable Matter (g/kg)	6.39	6.23	7.01	7.12	7.05	7.1	7.09	7.07	0.6 – 1.4

2.3.2. Fatty acid composition

Fatty acid composition is determined by an in-house method. A 10 mg oil sample was weighed into the flask and vortexed by adding 2 mL potassium hydroxide. After centrifugation the sample was injected to a GC-FID system. Agilent 6890 GC and SUPELCO SP 2560 column has been used for the fatty acid composition analysis. Identification and quantification of FAME was accomplished by comparing the retention times of peaks with those of pure standards FAME-MIX 37 and analyzed under the same conditions.

The column temperature was programmed from 140 to 240 0C at 4 ^{0}C/min with 5 min holding time. Then the temperature was raised to 240 ^{0}C at 8 ^{0}C/min with a final holding time of 20 min. 1 µm of the sample was injected. Flow rate was 1.1 mL/min and hellium is used as an carrier gas . Choromatogram time was 50 min. The results were expressed as a percentage of the individual fatty acids in the lipid fraction.

Table 2. Fatty Acid Composition of Nigella sativa seed oil

	Determined Values by Months								Valued in the Literature
Fatty Acid Composition	1	2	3	4	5	6	7	8	
Miristic Acid (C 14:0)	0.15	0.14	0.14	0.14	0.14	0.15	0.15	0.15	0.13 – 0.16
Palmitic Acid (C 16:0)	11.94	11.98	11.88	11.64	11.96	11.95	11.93	11.96	12.90 – 13.25
Palmitoleic Acid C 16:1)	0.19	0.19	0.19	0.19	0.2	0.18	0.18	0.18	0 - 0.60
Margaric Acid (C 17:0)	0.061	0.065	0.067	0.062	0.065	0.064	0.064	0.065	0.06
Heptadesenoic Acid (C 17:1)	0.054	0.053	0.059	0.056	0.05	0.05	0.05	0.05	0.03 – 3.29
Stearic Acid (C 18:0)	3.34	3.34	3.34	3.35	3.35	3.32	3.32	3.32	2.56 – 2.80
Oleic Acid (C 18:1)	24.64	24.57	24.59	24.49	24.41	24.34	24.36	24.3	22.63 – 24.51
Linoleic Acid (C 18:2)	56.17	56.19	56.21	55.57	56.37	56.32	56.21	56.33	58.90 – 61.20
Linolenik Acid (C 18:3)	0.25	0.24	0.26	0.23	0.26	0.25	0.23	0.22	0.21 – 0.28
Arashidic Acid (C 20:0)	0.2	0.22	0.21	0.22	0.22	0.24	0.22	0.26	0.13 - 0.15
Eicosenoic Acid (C 20:1)	0.31	0.3	0.31	0.31	0.31	0.25	0.36	0.34	0.27 – 0.35
Eicosadienoi c Acid (C 20:2)	2.55	2.58	2.59	2.58	2.57	2.57	2.57	2.58	1.86 – 9.40
Behenic Acid (C 22:0)	0.039	0.031	0.038	0.036	0.032	0.02	0.02	0.020	0.50 - 1.30
Docosenoic Acid (C 22:1)	0.047	0.036	0.032	0.033	0.048	0.039	0.042	0.044	0.30 – 1.10

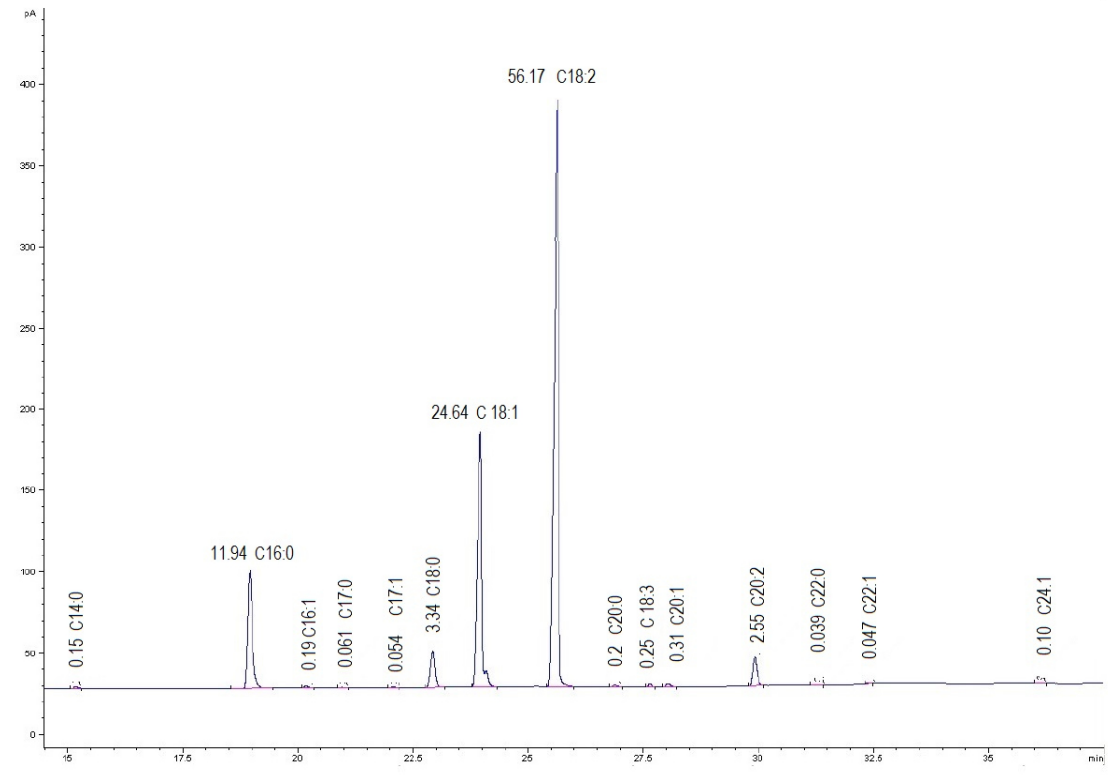

Figure 1. Nigella sativa seed oil chromatogram

3. Conclusion

The data of Table 1 show the physicochemical properties of *Nigella sativa* seed oils and the values in the literature. FFA and Peroxide value are used as the most important indicator for seed oil quality. FFA value is found lower than the literature values reported by Kiralan et al. (2014), Cheikh-Rouhou et al. (2007), Al-Saleh et al.(2006). As the low FFA indicates that the stability of the oil (Khoddami et al., 2011), the oil in the study can be accepted to be more stabile than the other Nigella oils with in the literature.

The peroxide value is found within the limits but higher than some other Nigella oils in the literature. The reason of this stuation can be cultivation differences and also process and transportation conditions.

The refractive index for 20 degrees C- 40 degrees C is found within the range of other studies. The Iodine value is found within the highest level indicating the low level of saturated fatty acids in the composition of oil. Saponification value is found similar with the Nigella seed oil from Egypt.

The unsaponifiable matter value is found higher than the other Nigella seeds oil. Differences in cultivation zones, climate, maturity levels and storage conditions can be effective for the results.

Fatty acid composition (Table 2) is found that similar with the literature values and the most abundant fatty acids found in the oil were the linoleic acid (C 18:2) 56.17% has the highest rate than oleic acid (C 18:1) 24.64% and palmitic acid (C 16:0) 11,94% respectively. After these acids stearic acid (C 18:0) 3.34%, eicosadienoic acid (C 20:2) 2.55%, eicosenoic acid (C 20:1) 0.31%, linolenic acid (C 18:3) 0.25%, arashidic acid (C 20:0) 0.20%, palmitoleic acid (C 16:1) 0.19%, miristic acid (C14:0) 0.15%, margaric acid (C 17:0) 0.061%, heptadesonoic acid (C 17:1) 0.054%, dokosenoic acid (C 22:1) 0.047%, behenic acid (C 22:0) 0.039%.

The results are found similar with the *Nigella sativa* seed oil from Egypt, Iran etc. that is given in the literature, but the reason of differences in the general rate is can be different climate conditions and geographical land zones.

As a conclusion *Nigella sativa* seed oil's physicochemical properties from Balıkesir Region /Turkey is found to be similar to the other sought-after varieties of *Nigella sativa* seed oil which is cultivated in Egypt, Iran etc. . Because of the similarity of the physicochemical characteristics of *Nigella sativa* seeds from Balıkesir Area it can be used as a suitable alternative raw material for both the food and supplement industry. Planning for further studies includes research about the different alternative areas where the *Nigella sativa* plant is grown with similar physicochemical properties. Also planned are more detailed studies of the same area of the Nigella sativa seeds' microbiological content for both seed and oil, mineral and vitamin characteristics and essential oil percentegas.

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