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EVALUATION OF FATTY ACID PROFILE OF *NIGELLA SATIVA* L. SEEDS

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ABSTRACT

The present investigation was aimed to study the medicinal properties of *Nigella sativa* seeds by determining fatty acid profile. Total fatty acid composition of *Nigella sativa* seeds oil was analysed by using Gas Chromatography. Estimated fatty acid profile showed that *Nigella sativa* seeds oil contained 58.55% of polyunsaturated fatty acids (PUFAs), 24.56 % of monounsaturated fatty acids (MUFAs) while saturated fatty acids (SFAs) only account for 16.73% of total oil contents. The main polyunsaturated fatty acid was Linoleic acid (56.24% of total fatty acid)followed by Oleic acid (23.97%), Palmitic acid (13.10%), Stearic acid (2.80%), Eicosadienoic acid (2.15%), Meristic acid (0.42%), Palmitoleic acid (0.22%), Alpha-Linolenic acid (0.16%) and Arachidic acid (0.16%).*Nigella* seeds are good source of oil and it is rich in polyunsaturated fatty acid which is more healthy.

Keywords: *Nigella sativa,* Fatty acid profile, Polyunsaturated fatty acids, Monounsaturated fatty acids.

Introduction

Nigella sativa L., an annual herb belonging to Ranunculaceae family have been used for thousands of years as a spice and food preservative to a variety of food products as bread, yogurt, pickles, sauces, salads etc. (Hajhashemi *et al.,* 2004). *Nigella sativa* is traditionally used for its galactagogue, appetizer, thermogenic and diuretic effects (Hosseinzadeh *et al.,* 2013). Furthermore, *Nigella sativa* possesses anti-microbial, anti-fungal, anti-oxidative and anti-cancerous properties (Salem and Hossain, 2000).Scientific investigations have depicted that *Nigella* seeds are high in macronutrients as well as micronutrients (Kabir *et al.,* 2019) and are the source of major active compounds like thymoquinone, thymohydroquinone, dithymoquinone, thymol, nigellone, tocopherols, trans-retinol and selenium (Agarwal *et al.,* 2020; Khan *et al.,* 2016). Thymoquinone is the most abundant phytoconstituent, predominantly present in fixed and essential oils of *Nigella*seeds and play a significant role as antioxidants (Varghese and Mehrotra, 2020; Salmani *et al.,* 2014). The oil of *Nigella* seeds has been shown to be effective as functional food and used in treating skin conditions, earaches and chronic colds (Yimer *et al.,* 2019). The present study was conducted to evaluate the fatty acid profile of selected *Nigella sativa* seeds cultivar grown in India namely: Azad Kalaunji-1.

Materials and Methods

Cultivar of *Nigella sativa* seeds namely Azad Kalaunji-1 was procured from C.S. Azad University of Agriculture & Technology, Kanpur, Uttar Pradesh, India. The seeds were cleaned by hand to remove dirt, grit and then packed in air tight plastic containers for further analysis. Oil was extracted in the Soxhlet apparatus by using petroleum etheras the solvent.

Fatty Acid Profile Analysis

Preparation of fatty acid methyl esters (FAMEs)

The methyl esters of fatty acids were prepared according to AOAC method 996.01, Fat (Saturated, Monounsaturated and Polyunsaturated) with some modification (AOAC, 2000a). Oil sample (0.2 g) was dissolved in 3 ml of chloroform and 3 ml of diethyl ether, mixed well and transferred to vial. Placed the vial in water bath at 60 ºC for evaporate. After adding 2 ml of 7% Boron trifluoride and 1 ml of toluene, solution was kept in oven at 100 ºC for 45 min. After cooling to room temperature, 5 ml of HPLC grade water followed by 1 ml of hexane and 1 g of anhydrous sodium sulphate were added. Mixture was allowed to stand until the water and hexane layer becomes separate. The upper layer of methyl esters of fatty acids was transferred to a capped and labelled glass vial for GC analysis.Supelco 37-Component FAME mixture (Supelco, USA) was used as a standard for the identification of fatty acids.

GC-MS Analysis of fatty acid

After methylation, fatty acid analysis was done by Gas Chromatograph/ Flame Ionization detector(GC/FID) (Clarus 500, Perkin Elmer, USA). FAMEs were separated by using a fused-silica capillary SGE column; internal diameter: 0.32mm, length: 30 m, film thickness: 0.25µm (PerkinElmer, USA). Helium was used as carrier gas at flow rate of

1ml/min, and 1 µl of sample was injected. The temperature setting was as follows: 120 ºC for 4 min, then increased to 200 ºC at the rate of 10 ºC/min and later increased up to 250 ºC at the rate of 5 ºC/min for 15 min. The injector and FID detector temperature was set at 220ºC and 280 ºC. The total running time for GC was 80 min. Peaks were identified by comparing the retention times obtained with standard methyl esters and results were expressed as percentage of fatty acid.

Results and Discussion

Fatty acids are the basic components of most naturally occurring lipids in both animals and plants. The diversity of the chain length, degree of unsaturation, geometry and position of double bonds as well as the presence of other groups, render their composition the most definitive characteristics of these lipids (Lima *et al.,* 2002). The determination of fatty acid profile is of considerable importance in lipid analysis.

The fatty acid profile includes saturated and unsaturated fatty acids which were estimated for the *Nigella* seeds and the results are presented in Table 1 and2. Fig. 1 and 2 shows the GC chromatogram of the 37 fatty acid methyl ester standards and GC chromatogram of *Nigella sativa* seeds oil respectively.It contained 58.55% of polyunsaturated fatty acids (PUFAs), 24.56 % of Monounsaturated fatty acids (MUFAs) and16.73% of saturated fatty acids (SFAs) while Total trans-fatty acidsonly account for less than 0.10% of total oil contents. Linoleic, oleic and palmitic acids were found to be rich in higher quantities. The percentage of theses fatty acids were 56.24, 23.97 and 13.10 respectively. According to our findings,*Nigella* oil is rich in linoleic acid categorized as an essential fatty acid. It plays important role in our diet because human body cannot synthesize it. The oil contained 2.80% of stearic acid, 2.15% of eicosadienoic acid, 0.42% of meristic acid, 0.22% of palmitoleic acid, 0.16% of alpha-linolenic acid and 0.16% of arachidic acid. These findings are in good agreement with Ibraheem 2011; Bourgou *et al.,* 2010; Amin *et al.,* 2010. Malhotra *et al.* (2004a) reported *Nigella sativa* was found to contain linoleic acid 44.7-56 %, oleic acid 20.7-24.6 %, linolenic acid 0.6-1.8 %, palmitic acid 12-14.3 %, stearic acid 0.16 %. The source of quantitative and qualitative variability of fatty acids may be due to extraction techniques, genetics (variety grown), seed quality, agricultural conditions. *Nigella sativa* is the richest source of unsaturated fatty acid which is beneficial to human nutrition and health in aspects to behavioural, cognitive functions and inflammatory conditions such as asthama, arthritis and Crohn's disease (Simopoulos, 2002). The higher amount of linoleic acid (essential fatty acid) in the *Nigella sativa* has an enormous potential as a therapeutic constituent in improving of health conditions. *Nigella sativa* oil is considered as one among the newer sources of edible oils, thanks to its vital role in human health and nutrition.

Conclusion

Nigella sativa is an obvious medicinal plant commonly used in the food and pharmaceutical industry as black seed oil. The fatty acid analysis showed that the *Nigella sativa* seeds oil is rich in polyunsaturated fatty acids and monounsaturated fatty acids as well as it contain low levels of saturated fatty acids, signifying this oil as valuable source of nutrition to be used for edible purpose for various health benefits. The health properties of the oil, which were revealed, make interesting avenues of research in future studies.

Figure/Legends

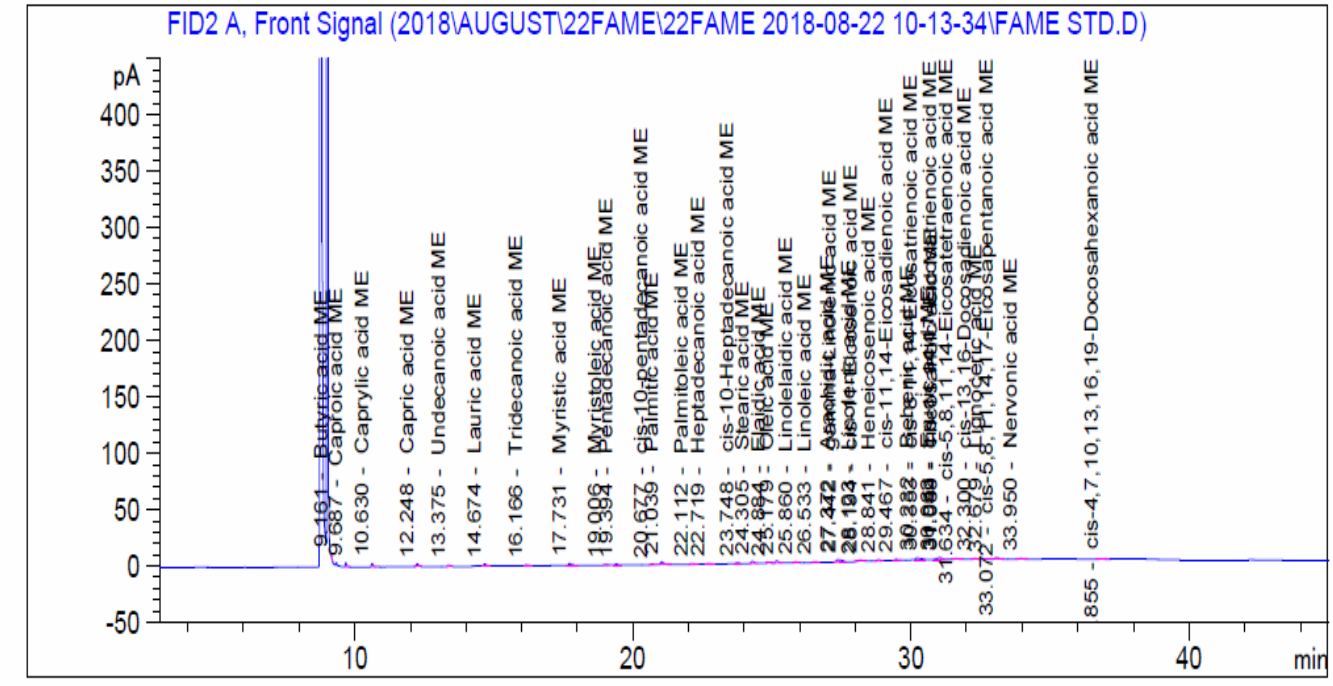

Fig. 1: GC Chromatogram of the 37 Fatty Acid Methyl Ester Standards

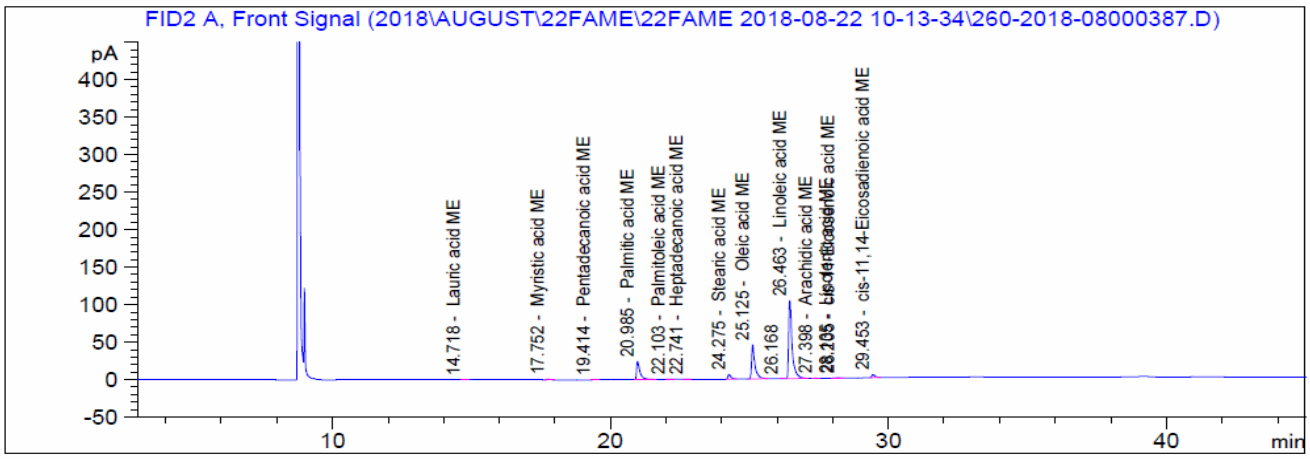

Fig. 2: GC Chromatogram of *Nigella sativa* Seeds Oil

Table 1: Fatty Acid Profile of *Nigella sativa* Seeds

Fatty acid	g/100g
Saturated fatty acids	16.73
Monounsaturated fatty acids	24.56
Polyunsaturated fatty acids	58.55
Total trans-fatty acids	<0.10

Table 2: Fatty Acid Composition of *Nigella sativa* Seeds Oil

Fatty acids	C atoms and double bonds	g/100g
Butyric acid	C_4 : 0	< 0.10
Caproic acid	C_6 : 0	< 0.10
Caprylic acid	C_8 : 0	< 0.10
Capric acid	C_{10} : 0	< 0.10
Undecanoic acid	C_{11} : 0	< 0.10
Lauric acid	C_{12} : 0	0.11
Tridecanoic acid	C_{13} : 0	< 0.10
Myristic acid	C_{14} : 0	0.42
Myristoleic acid	C_{14} : 1	< 0.10
Pentadecanic acid	C_{15} : 0	< 0.10
Pentadecenoic acid + Isomers	C_{15} : 1	< 0.10
Palmitic acid	C_{16} : 0	13.10
Palmitoleic acid	C_{16} : 1	0.22
Margaric acid	C_{17} : 0	< 0.10
Margaroleic acid	C_{17} : 1	< 0.10
Stearic acid	C_{18} : 0	2.80
Oleic acid	C_{18} : 1	23.97
Elaidic acid	C_{18} : 1 n9t	< 0.10
Linoleic acid	C_{18} : 2	56.24
Linolelaidic acid	C_{18} : 2t	< 0.10
Alpha-Linolenic acid	C_{18} : 3 n3	0.16
Gamma- Linolenic acid	C_{18} : 3 n6	< 0.10
Arachidic acid	C_{20} : 0	0.16
Eicosenoic acid	C_{20} : 1	< 0.10
Eicosadienoic acid	C_{20} : 2	2.15
Eicosatrienoic acid	C_{20} : 3	< 0.10
Homo-gamma- Linolenic acid	C_{20} : 3 n6	< 0.10
Aracidonic acid	C_{20} : 4 n6	< 0.10
Eicosapentaenoic acid	C_{20} : 5	< 0.10
Heneicosaenoic acid	C_{21} : 0	< 0.10
Behenic acid	C_{22} : 0	< 0.10
Docosaenoic acid + Isomers	C_{22} : 1	< 0.10
Docosadienoic acid	C_{22} : 2	< 0.10
Docosahexaenoic acid	C_{22} : 6	< 0.10
Tricosanoic acid	C_{23} : 0	< 0.10
Lignoceric acid	C_{24} : 0	< 0.10
Nervonic acid	C_{24} : 1	< 0.10

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Conflict of Interests

The authors declare no conflict of interest regarding the publication of this article.

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