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Antioxidant Components of *Fumaria* Species (Papaveraceae)

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***Abstract*—**The genus *Fumaria* L. (Papaveraceae) in Iran comprises 8 species with a vast medicinal use in Asian folk medicine. These herbs are considered to be useful in the treatment of gastrointestinal disease and skin disorders. Antioxidant activities of alkaloids and phenolic extracts of these species had been studied previously. These species are: *F. officinalis*, *F. parviflora, F. asepala*, *F. densiflora*, *F. schleicheri*, *F. vaillantii* and *F. indica*. More than 50 populations of *Fumaria* species were sampled from nature. In this study different fatty acids are extracted. Their picks were recorded by GC technique. This species contain some kind of fatty acids with antioxidant effects. A part of these lipids are phospholipids. As these are unsaturated fatty acids they may have industrial use as natural additive to cosmetics, dermal and oral medicines. The presences of different materials are discussed. Our studies for antioxidant effects of these substances are continued.

Keywords*—Fumaria,* Papaveraceae, fatty acid, antioxidant, Iran

I. Introduction

THE chemical composition of fats and oils which confers on them specific properties has made them suitable for use as foods, fuels and lubricants. They are naturally occurring substances which consist predominantly of esters of fatty acids and glycerol. Their sources include substances of vegetable, animal and marine origin. Vegetable oils in particular are natural products of plant origin consisting of ester mixtures derived from glycerol with chains of fatty acid contain about 14 to 20 carbon atoms with different degrees of unsaturation. Knowledge of their specific characteristics provides insight into their being prone to auto oxidation reactions, thus necessitating the use of antioxidants as one major means of ensuring oxidative stability. Vegetable oils are obtained from oil containing seeds, fruits, or nuts by different pressing methods, solvent extraction or a combination of these. Crude oils obtained are subjected to a number of refining processes, both physical and chemical. Combating the issue of oxidative instability in vegetable oils for industrial use is a continuing research area. In the United States, for instance, three avenues are being pursued. These are:

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- Genetic modification of oils to give higher mono unsaturated compounds
- Chemical modification
- The use of various additives and property enhancers

Genetic modification has been made possible by recent advances in biotechnology. DuPont Technology has developed a soybean seed that presents 83% oleic acid as against having the more unsaturated linolenic acid as the major constituent. This new seed provides oils that show about 30 times the oxidative stability and viscosity stability of the conventional oil. High oleic varieties of rapeseed, canola and sunflower seed oils are increasingly being used as base stocks for lubricant formations [1].

Fatty acid is a carboxylic acid with long aliphatic tail. Fatty acid is divided into saturated and unsaturated acid, depending on the presence of unsaturated double bond in the fatty acid chain [2]. Essential fatty acids are polyunsaturated fatty acid. Linoleic acid (C18:2) and α-linolenic acid (C18:3) are the parent compounds of the omega-6(ω-6) and omega-3(ω-3) fatty acid series, respectively [2]. They are essential in the human diet since they cannot be synthesized by the body. The essential fatty acids are very important to human immune system, to help regulate blood pressure. The ω-3 and ω-6 fatty acid are found in some food; fish, shellfish flaxseed (linseed), soya oil, canola (rapeseed) oil, hemp oil, chia seed, pumpkin seed, sunflower seed, cotton seed oil, leafy vegetables and walnut. The significance of fatty acid analysis has gained much attention because of the nutritional and health implications. The most common procedure for the analysis is the conversion of fatty acid components to methylester in order to improve their volatility [3].

There are many papers focusing the analysis of fatty acid in plant seeds such as flaxseed (*Linum usitatissimum* L.) [4], grape seed oil [5], Thai Durian aril (*Durio zibetbinus* Murr.)[6], and China chestnut (*Sterculia monosperma* vertenat) [7].

Fumarioideae consists of 19 genera according to a recent proposal. The subfamily is variable in seed-coat (hardness, color, and surface) and arils (presence or absence, size, and shape), which have long been sources of taxonomic characters [8].

Fumraia with 7 species in Iran is distributed in different habitats of Iran. These species are morphologically very similar and it could be concluded that these have experienced a recent speciation event. Sometimes *Fumaria* species identification is very difficult or impossible due to a vast modification after specimen collecting. This is true for species like *F. asepala* and *F. in*dica or *F. vaillantii* and *F. parviflora*.

According to the literature, *F. vaillanti* contains alkaloids, vitamins, tanning substances, and lipids. The closely related plant *F. officinalis* L. (drug fumitory) is used in folk medicine.

Mirzabaeva, Umarov and Shukurova [9] works on *F. vaillantii* in S.S.R. This plant is widely distributed in the Namangan, Samarkand and Tashkent. The physicochemical indices of the oil are close to those for the oils of *Glaucium fimbrilligerum* and *Roemeria refracta* from the same family. The compositions of the fatty acids of the oil isolated, included Myristic, Palmitic, Stearic, and Oleic acid. Linoleic acid predominates in the oil of the seeds of *F.vaillantii* . The UV spectrum of the fatty acids of the oil showed the presence of 3.65% of conjugated dienic acids in it.

Gazizov and Glushenkova (1997) [10] founded that the oil from one seed weighed about 2 mg, with a moisture content of 5.6%; the total yield of phospholipids (PhLs) was 0.25% of the air-dry seeds. Six classes of PhLs were detected in the seeds, the main ones being phosphatidylcholines (PhCs) (64.1%) and phosphatidylinositols (Phls) (27.5%). All the classes of PhLs were isolated from the total PhLs by column and thin-layer chromatographies. The compositions of their total fatty acids (FAs) were determined. The FAs in the PLs were represented mainly by palmitic, oleic, and linolenic acids. With respect to the level of unsaturated fatty acids, the highest place was occupied by the PhCs: 66.1%, the second by the phosphatidylethanolamines: 60%, and the third by the Phls: 53.9%; then followed the lyso-PhCs: 46.8%, an unidentified PhL: 41.2% and, finally, phosphatidic acid: 38%.

When evaluated by measuring reducing power, ability to inhibit linoleic acid peroxidation, and 2,2-diphenyl-_-picrylhydrazyl radical scavenging activities, the alkaloid extracts of *Fumaria capreolata* and *Fumaria bastardii* demonstrated strong total antioxidant activity, with effectiveness marginally less than that of the common synthetic antioxidant butylated hydroxyanisole, and better than quercetine and caffeine. These species have wide distribution in the Mediterranean region and have a reputation for effectiveness in treating hepatobiliary disfunction and gastrointestinal disorders via local therapies [1].

Therefore, the objective of this research is determination of Palmitic acid, Linoleic acid, Lignoceric acid, Arachidonic acid, Myristic acid, Lauric acid, Decanoic acid, and oleic acid in *Fumaria*.

II. Material And Methods

A. *Plant Seeds*

Fumaria parviflora were brought from local place then dry, clean and grind.

B.*Instrumentation*

A Shimadzu gas chromatograph was performed with DB-wax fused silica capillary column (30 m x 0.25 mm i.d.., 0.25 mm film thickness). The injector and flame ionization detector were 250 ^{o}C. The column temperature program was started from 150 oC hold for 1 min, then ramp to 200 ^{o}C with the heating rate of 25 oC/min hold for 3 min and final temperature increase to 230 ^{o}C with at a rate of 15 oC/min and hold 5 min. The pressure of nitrogen carrier gas was 100 kPa.

C. *Fatty Acid Extraction and Methylation*

The procedure was base on Blight and Dayer method. A 0.10 g of samples were weighted into screw-tap glass bottles then added 5 ml of toluene and 5 ml of fresh solution of methanolic hydrochloric. The bottles were closed and placed in water bath at 70 ^{o}C for 1 h. Then 5 ml of 6% potassium carbonate solution and 1 ml of toluene were added and thoroughly vortexed for 1 min. The organic phase was separated by using centrifugation at 1100 rpm for 5 min, dried organic phase with sodium sulphate anhydrous and filtered by a millipore 0.45 mm. A 1 ml aliquot was injected into gas chromatograph.

D. *Standard Calibration Curve*

The standard mixture of fatty acids (2, 5, 10, 25 and 50 mg/ml) were prepared by methylation similar to the sample preparation.

III. Results And Discussion

Analysis of fatty acids parameters were perfumed by gas chromatography with DBwax capillary column (30 m x 0.25 mm id.). The calibration ranges were 0-50 mg/ml with the correlation coefficient of 0.9918-0.9993. The detection limits were in the range of 0.08- 0.54 mg/ml.

Figure 1 showed chromatogram of standard fatty acids and

Figure 2 showed chromatogram of fatty acids extracted from *Fumaria parviflora*. Analysis of 0.1 g of plant seeds, the results were found Palmitic acid, Linoleic acid, Lignoceric acid, Arachidonic acid, Myristic acid, Lauric acid, Decanoic acid, and oleic acid in *Fumaria*.

Lignoceric acid, or tetracosanoic acid, is the saturated fatty acid with formula C23H47COOH. It is found in wood tar, various cerebrosides. The fatty acids of peanut oil contain small amounts of lignoceric acid (1.1%–2.2%). This fatty acid is also a byproduct of lignin production [11].

Arachidonic acid (AA, sometimes ARA) is a polyunsaturated omega-6 fatty acid 20:4(ω-6). It is the counterpart to the saturated arachidic acid found in peanut oil, (L. arachis – peanut.) [11].

Linoleic acid (LA) is an unsaturated n-6 fatty acid. It is a colorless liquid at room temperature. In physiological literature, it is called 18:2(n-6). Chemically, linoleic acid is a carboxylic acid with an 18-carbon chain and two cis double bonds; the first double bond is located at the sixth carbon from the methyl end.

Linoleic acid is one of two essential fatty acids that humans and other animals must ingest for good health because the body requires them for various biological processes, but cannot synthesize them from other food components [12]. Linoleic acid is used in making soaps, emulsifiers, and quickdrying oils. Reduction of linoleic acid yields linoleyl alcohol. Linoleic acid has become increasingly popular in the beauty products industry because of its beneficial properties on the skin. Research points to linoleic acid's anti-inflammatory, acne reductive, and moisture retentive properties when applied topically on the skin. Since children with cystic fibrosis suffer from Essential Fatty Acid Deficiency due to mal-absorption, it was hypothesized that high doses of LA might aid in their growth. Dermatitis is one of the first signs of an Essential Fatty Acid deficiency in both humans and animals.

Oleic acid is a mono-unsaturated omega-9 fatty acid in various animals and vegetables. It has the formula $CH_3(CH_2)_7CH=CH(CH_2)_7COOH$ [11]. It is odorless colourless oil. The trans-isomer of oleic acid is called elaidic acid (hence the name elaidinization for a reaction that switches cis-isomers to trans-isomers). The term Oleic means related to, or derived from, oil or olive. Oleic acid may hinder the progression of ALD, or adrenoleukodystrophy, a fatal disease that affects the brain and adrenal glands. Oleic and monounsaturated fatty acid levels in the membranes of red blood cells have been associated with increased risk of breast cancer [13]. Oleic acid may be responsible for the hypotensive (blood pressure reducing) effects of olive oil.

Palmitic acid, or hexadecanoic acid in IUPAC nomenclature, is one of the most common saturated fatty acids found in animals and plants [14]. Its molecular formula is $CH_3(CH_2)_{14}COOH$. As its name indicates, it is a major component of the oil from palm trees (palm oil, palm kernel oil and coconut oil). The World Health Organization claims that dietary intake of palmitic acid increases risk of developing cardiovascular diseases. However, another study showed that palmitic acid has no hypercholesterolaemic effect if intake of linoleic acid is greater than 4.5% of energy. On the other hand, it was shown that, if the diet contains trans fatty acids, the health effects are negative, causing an LDL cholesterol increase and HDL cholesterol decrease [15]. Cetyl palmitate occurs in spermiceti. Retinyl palmitate is an antioxidant and a source of vitamin A added to low fat milk to replace the vitamin content lost through the removal of milk fat. Palmitate is attached to the alcohol form of vitamin A, retinol, to make vitamin A stable in milk. Palmitate esters are also the major component of palm oil. The word palmitate is from the French "palmitique", the pith of the palm tree. Recently, a long-acting antipsychotic medication, paliperidone palmitate (marketed as INVEGA Sustenna), used in the treatment of schizophrenia, has been synthesized using the oily palmitate ester as a long-acting release carrier medium when injected intramuscularly. The underlying method of drug delivery is similar to that used with decanoic acid to deliver long-acting depot medication, in particular, neuroleptics such as haloperidol decanoate.

Myristic acid, also called tetradecanoic acid, is a common saturated fatty acid with the molecular formula $CH_3(CH_2)_{12}COOH$. A myristate is a salt or ester of myristic acid. Myristic acid is also commonly added co-translationally to the penultimate, nitrogen-terminus, glycine in receptor-associated kinases to confer the membrane localisation of the enzyme. The myristic acid has a sufficiently high hydrophobicity to become incorporated into the fatty acyl core of the phospholipid bilayer of the plasma membrane of the eukaryotic cell. In this way, myristic acid acts as a lipid anchor in biomembranes. The ester isopropyl myristate is used in cosmetic and topical medicinal preparations where good absorption through the skin is desired. Reduction of myristic acid yields myristyl aldehyde and myristyl alcohol. *Lauric acid* is the main acid in coconut oil and in palm kernel oil (not to be confused with palm oil), and is believed to have antimicrobial properties.

Decanoic acid, or capric acid, is a saturated fatty acid. Its formula is $CH_3(CH_2)_8COOH$. Salts and esters of decanoic acid are called decanoates. The term capric acid arises from the Latin "capric" which pertains to goats due to their olfactory similarities. Decanoate salts and esters of various drugs are available. Since decanoic acid is a fatty acid, forming a salt or ester with a drug will increase its lipophilicity and its affinity for fatty tissue. Since distribution of a drug from fatty tissue is usually slow, one may develop a long-acting injectable form of a drug (called a Depot injection) by using its decanoate form. Some examples of drugs available as a decanoate ester or salt include nandrolone, fluphenazine, bromperidol, haloperidol and vanoxerine.

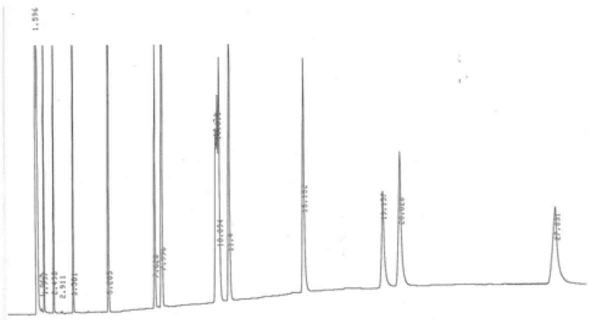

Fig. 1 Chromatogram of standard fatty acid by DB-wax fused silica capillary column (30 m x 0.25 mm i.d., 0.25 μm film thickness)

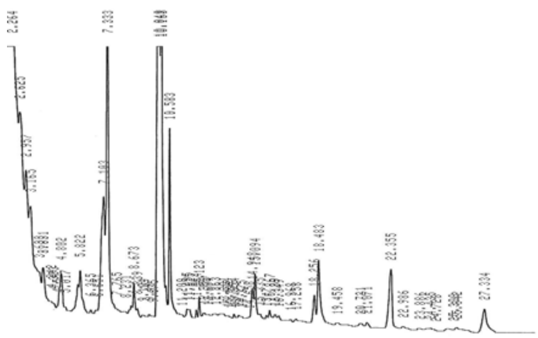

Fig. 2 Chromatogram of fatty acid

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