Essential Oil Composition of Syrian Aniseed (*Pimpinella anisum* L.)

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Abstract

Pimpinella anisum (anise), is an aromatic plant which belonging to *Umbelliferae* family, and it is a very popular spice and herb, which is widely applied in the food and pharmaceutical industry. In this study, the hydro distillation essential oil components were determined using GC-MS analysis. The essential oil yield was 2.93% (v/w). Five compounds, comprising more than 99.09% of the composition of oil were identified in the aniseed's essential oils. The main components of the essential oil were Estragole 0.29%, trans-anethole 96.11%, γ-himachalene 1.83%, α-zingiberene 0.53%, α -Cadinol 0.33%.

Keywords: Essential oil, *Pimpinella anisum*, GC-MS, Trans-anethole.

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التركيب الكيميائي لزيت الأساس المستخلص من بذور اليانسون السوري

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الملخص

في هذه الدراسة حددت مكونات الزيت العطري لبذور اليانسون المستخلص بالجرف بالبخار، وحُلل باستخدام GC-MS. تم الحصول على مردود قدره 2.93 % (حجم/ وزن)، شُخصت خمسة مكونات شكلت نسبة 99.09 % من مكونات زيت الأساس لبذور اليانسون. والمكونات الأساسية في هذا الزيت هي: استراجول (0.29 %)، ترانس- أنيثول (96.11 %)، غاما-هيماتشالين (1.83 %)، الفا-زنجيبرين (0.53 %)، الفا-كادينول (0.33 %).

الكلمات المفتاحية: بذور اليانسون، زيت الأساس، GC – MS، ترانس أتينول.

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1. Introduction:

One of the actions to counter the emergence of the drug resistance problem is the development of new antimicrobials. The antimicrobial, antiseptic, and other therapeutic applications of plants are well recognized since the prehistoric times, widely used by all civilizations throughout the millennia [1]. Many aromatic plants and spices are well known for their various beneficial effects on human health. Their use in phytotherapy is mostly related to different activities of their essential oils, such as antimicrobial, spasmolytic, carminative, antiviral, antimutagenic, anticarcinogenic, etc. [2,3].

Essential oils of various plants contain about 20 to 80 components with quiet different concentration from which 2 to 4 components are the main components with higher concentrations than other components of the oil. Generally, these major compounds determine the biological properties of the essential oils [3]. In addition, the formation of essential oil in the plant, and consequently the yield and composition of the produced oil depends on many factors. Genetic differences among plants of the same species that are otherwise indistinguishable (chemotypes) can result in widely different oil types [4].

The *Umbelliferae* (*Apiaceae*) are mostly temperate herbs usually with umbellate inflorescences comprising more than 300 genera and more than 3000 species. The species are commonly further distinguished by the presence of hollow stems and sheathing petioles. The family has a cosmopolitan distribution, but most of its members are confined to northern temperate regions and high altitudes in the tropics [5]. Plants belonging to this family are extensively used for food and medicinal purposes. Some plants of this family such as carrots, parsley, and celery are common vegetable crops, while other members like anise, dill, coriander, fennel, and cumin are famous for their medicinal and aromatic properties 5, 6].

Pimpinella anisum L., a plant belonging to the *Umbelliferae* family, is one of the oldest medicinal plants. It is an annual grassy herb with 30–50 cm high, white flowers, and small green to yellow seeds, which grows in the

Eastern Mediterranean Region, West Asia, the Middle East [7]. *P. anisum* is primarily grown for its fruits (aniseeds) that harvested in August and September. Aniseeds contain 1.5–5% essential oil and used as flavoring, digestive, carminative, and relief of gastrointestinal spasms. Consumption of aniseed in lactating women increases milk and also reliefs their infants from gastrointestinal problems [9]. In the food industry, anise is used as flavoring and aromatic agent for fish products, ice cream, sweets, and gums [7, 9].

The aim of this study was to determine the chemical composition of the essential oil extracted from Syrian seeds of *P. anisum*, by GC-MS analysis.

MATERIALS AND METHODS

Plant Material

The anise seeds (*Pimpinella anisum*) which cultivated in Syria were purchased from the local herbs market in Damascus (albzouryia). The anise seeds identity was confirmed in Department of Botany, Faculty of Science - Damascus University. The dirt was removed with distilled water. Collected seeds were dried in the shade for about 7 days. The dried seeds were ground into fine grinding by a grinder machine.

Essential Oil Extraction

The essential oil was extracted by hydro distillation in a Clevenger-type apparatus, according to the literature [10]. 100 g seeds was subjected to hydro-distillation in a Clevenger-type apparatus for 3 h. in accordance with the standard procedure described in the European Pharmacopoeia. The sample was added to distilled water 1000 ml in a 2 L round – bottomed flask and heated to boiling, after which the essential oil was evaporated together with water vapor, and finally collected in a separating funnel. The upper phase that contained the essential oil was separated from the lower one and the essential oil was dried over anhydrous sodium sulfate and preserved in a sealed sample tube and stored in the fridge at 4^{o}C until analysis.

GC-MS analysis of Essential Oil

Gas chromatography analysis was carried out with an Agilent 6890 N gas chromatograph (GC) equipped with Agilent 5973 mass selective detector (MSD), Agilent Auto sampler 7683 and Agilent HP-5MS capillary

column (30 m, 0.25 i.d., 0.25 µm film thickness) (Agilent Technologies, Santa Clara, CA, USA). The MS detector was operated in electron impact (EI) mode at 70 eV with interface temperature of 280 ^{o}C; the scan range was 50–550 amu. The injection port temperature was set at 250 °C. But GC was performed in split less mode; carrier gas was helium at a constant flow rate of 1ml/min. The column temperature was programmed as follows: an initial temperature of 60 ^{o}C increased to 280 ^{o}C at rate of 3 ^{o}C/min. The injection volume was 1.0 µL.

The analysis of main components of anise essential oil by GC-MS was used for the analysis of other components of anise essential oil, which were present in minor quantity. Electronic integration of the flame ionization detector peak areas was used for determination of quantitative data. MSD Chemstation software (Agilent Technologies) was used for data analysis. Components were identified using their retention times to n ‑alkanes (C8- C24; sigma, Germany) compared to those of Wiley 275 library and those described by Adams (2004) [11].

Results and Discussion

The essential oil yield from sample of anise seeds was 2.93%, and was pale-yellow in color. The analysis of main components of anise seeds oil by GC-MS was used for the analysis of components of anise seeds essential oil, which were present in minor quantity. For identification of these components, the retention time of the desired substance can be placed between the retention times of two adjacent homologous alkanes which were already determined (Table 1 and fig 1). The retention index is a good comparison to identify the samples of unknown substance. The results in Table 1 showed that the oil contained mainly phenylpropanoids and sesquiterpenoid hydrocarbons. Compounds are listed in order of their elution. Five components were identified, accounting for 99.09 %. The major constituents of the essential oil were Estragole 0.29%, trans-anethole 96.11%, γ-himachalene 1.83%, α-zingiberene 0.53%, α-Cadinol 0.33%. It is of interest to note the presence of trans-anethole (96.11%) in very high percentages, which was distinctive *P. anisum*. The essential oil fraction of the *P. anisum* showed differences and similarities from many

anise species growing in the several regions of world, in regards to compounds, in the variety of its components and their relative quantity.

Table 1 Chemical constituents of essential oils from anise seeds.

No. Peak	RI	Compounds	Molecular formula	Area (%)
1	1173	Estragole	$C_{10}H_{12}O$	0.29
2	1296	Trans-anethole	$C_{10}H_{12}O$	96.11
3	1485	γ-himachalene	$C_{15}H_{24}$	1.83
4	1496	α-zingiberene	$C_{15}H_{24}$	0.53
5	1681	α -Cadinol	$C_{15}H_{26}O$	0.33
	Total			99.09

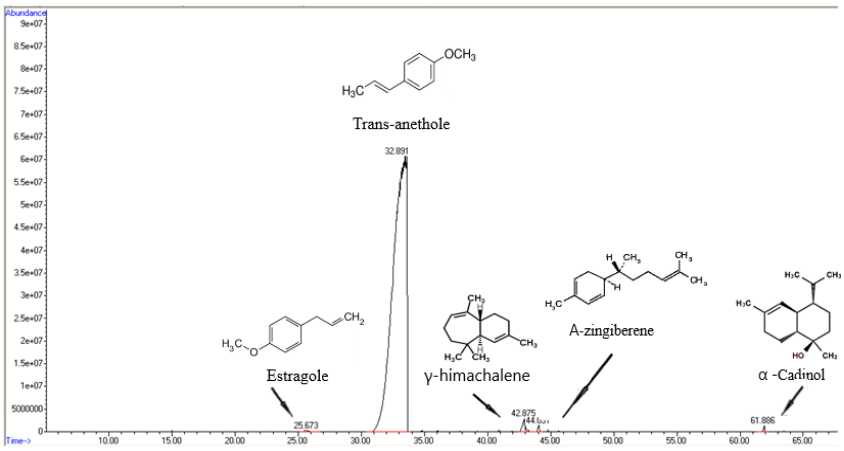

Fig 1. GC/MS hromatogram of anise seeds essential oil.

Aniseed contains 1.5–6.0 mass % of a volatile oil consisting primarily of trans-anethole [12]. The major component of *P. anisetum* essential oil seen in the current study was trans-anethole, which was similar to major component of many regions of the world. In addition, we found that the major compounds of *P. anisetum* essential oil were orderly Estragole,

trans-anethole, γ-himachalene, α-zingiberene, α -Cadinol. The content of trans-anethole is higher than those found in the essential oils of Turkey 95.4%, 89.5% respectively [13,14], Portugal 92.5% [15], Algeria 92.4% [16], and Iran (92.9%) [17]. Other studies have demonstrated the presence of eugenol trans-anethole, methylchavicol, anisaldehyde, estragole, coumarins, scopoletin, umbelliferone, estrols, terpene hydrocarbons, polyenes, and polyacetylenes as the major compounds of the essential oil of anise seed [18].

In another study for determination of the composition of essential oil of *P. anisum* L. fruits obtained from different geographical areas of Europe, in addition to the major components (trans-anethole (76.9–93.7%) and γ-himachalene (0.4–8.2%), some other compounds such as trans-pseudoisoeugenyl 2-methylbutyrate, p-anisaldehyde, and methylchavicol were also identified in essential oil [19].

Results of other studies were completely consistent with the present study, although there were some differences in components and their quantities. This variation can be due to the various factors affecting on essential oils chemical composition such as differences in climate, seasonal, and geographic conditions [20].

Conclusion:

Pimpinella anisum is one of the medicinal plants, which have been used for different purposes in traditional medicine of Syria. So far, different studies were performed on the extracts and essential oil of *P. anisum* to identify the chemical compounds and pharmacological properties of this plant, and various properties such as antimicrobial. In this study, the chemical composition, of aniseed essential oil was analyzed, and the present results indicate that trans-anethole can be identified and determined by GC-MS, Further investigations to determine the other medicinal active compounds of the plant and experimental as well as clinical studies are warranted.

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