

CHEMICAL COMPOSITION AND ANTIMICROBIAL ACTIVITY OF ESSENTIAL OIL FROM SEEDS OF *Anethum graveolens* GROWING IN UZBEKISTAN

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We have previously studied the chemical composition of essential oil (EO) from seeds of *Anethum graveolens* from Xinjiang Autonomous District in the PRC [1]. The component composition of essential oils is known to depend on the habitat. It seemed interesting to compare the composition and biological activity of EO from dill seeds growing in China and Uzbekistan. We used GC—MS to establish the structures of the isolated compounds.

EO from seeds of *A. graveolens* (2007 harvest) that were collected in Tashkent Oblast was isolated by steam distillation in 4.2% yield. The chemical composition of the EO was studied using a Perkin—Elmer Turbo GC—MS. The component content of the oil was calculated using areas of GC peaks of total ion current without correlation coefficients. EO components were identified by comparing retention times and mass spectra of the component obtained in mass scanning mode and by using mass-spectral library data for standard oil components and pure compounds. A total of 22 chemical compounds was identified in EO of *A. graveolens* seeds growing in Uzbekistan. Table 1 lists the chemical composition of the EO.

The principal EO components from dill seed growing in Uzbekistan were carvone (73.61%), limonene (14.69), *cis*-dihydrocarvone (5.87), diplaniol (1-allyl-2,5-dimethoxy-3,4-methylenedioxybenzene) (2.16), and 1,2-diethoxyethane (1.43%), which together made up 99.2% of the total EO component composition. The principal components of EO from dill seed growing in China were *n*-pentacosane (27.96%), dioctylester of 1,2-phenyldicarboxylic acid (25.10), octacosane (13.81), tricosane (9.14), and *n*-nonacosane (6.85%) [1]. A comparison of our data with that obtained earlier indicated that both the qualitative and quantitative composition of the principal EO components of *A. graveolens* growing in different geographic zones differed considerably. The high content in the studied EO of carvone, which is widely used as a growth inhibitor of bacteria [2-4] and certain fungi [5] and as a repellent [6] is noteworthy. Both *S*-(+)-carvone and *R*-(-)-carvone are used in the food industry to produce flavors [4] and in agriculture. For example, *S*-(+)-carvone is used in the Netherlands to prevent premature sprouting of potato tubers and tulip bulbs during storage [7, 8]. Carvone is an available and inexpensive reagent for organic synthesis in both enantiomeric forms. This makes it attractive for asymmetric synthesis of natural compounds [9].

Antimicrobial activity of EO fractions toward *Candida albican* and *Staphylococcus aureus* was estimated using the Barry method to determine the minimal inhibiting concentration (MIC) [10]. Growth of microorganisms decreased markedly upon addition of EO to nutrient medium. The experimental results are given below:

Microorganism	Addition	MIC, mg/mL
<i>Staphylococcus aureus</i>	1:512	0.273
<i>Candida albican</i>	1:51200	0.00273

Complicated mixtures of monoterpenes and sesquiterpenes from *A. graveolens* EO possessed pronounced antimicrobial and fungicidal activities and were a strong barrier against penetration of bacterial and fungal infection in plant seeds during their storage and sprouting. Furthermore, a comparison of the results and the literature showed that EO from dill seeds cultivated in Uzbekistan consisted mainly of carvone, limonene, *cis*-dihydrocarvone, diplaniol, and 1,2-diethoxyethane as the principal components whereas the principal components of EO from seeds of European and Indian dill were limonene and carvone [4, 11].

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TABLE 1. Composition of Essential Oil from *Anethum graveolens* Seeds

Component	Retention time, min	Content, %	Component	Retention time, min	Content, %
1,2-Diethoxyethane	6.807	1.43	<i>cis</i> -Dihydrocarvone	50.12	5.87
Ethylacetate	8.84	0.06	α -Humulene	56.17	0.02
α -Phellandrene	18.66	0.03	D-Carvone	57.57	73.61
β -Myrcene	18.91	0.07	<i>cis</i> -Dihydrocarveol	58.89	0.15
Limonene	21.42	14.69	<i>m</i> -Carveol	62.01	0.04
Sabinene	21.68	0.02	Carvone oxide	62.58	0.02
2,6-Dimethyl-1,3,5,7-octatetraene	21.96	0.07	3,5-Dimethylcyclohexen-1-one	63.11	0.03
1-Methyl-3-(1-methylethyl)benzene	25.33	0.03	<i>cis</i> -Carvone	63.77	0.04
2-Methyl-2-propenylbenzene	36.72	0.02	Farnesene	72.82	0.01
Limonene oxide	37.40	0.02	Myristicin	85.75	0.04
Dihydrocarvone	48.72	1.44	Diplaniol	91.82	2.16

Plant Material and Isolation Method. EO was isolated from dill seeds (100 g) by grinding in a coffee grinder and extracting using steam distillation for 4 h. EO was extracted from the aqueous emulsion using diethylether and dried over anhydrous Na₂SO₄ overnight. Ether was removed by evaporation on a water bath to afford EO (4.2% yield).

Analysis of EO Components by GC—MS. The chemical composition of EO was studied using GC—MS on a Perkin—Elmer Turbo Mass Aid System XL with a quadrupole mass-selective detector. We used a quartz capillary column with copolymer 5%-phenylmethylsilicone (PE-5MS, 30 m × 0.25 mm) and a stationary phase (25 μm thick). The He carrier gas flow rate was 35 cm/s. The column was heated according to a temperature program starting at 75°C for 2 min, heating to 100°C at 2°C/min, to 160°C at 4°C/min, to 220°C at 2°C/min, and holding at that temperature for 2 min. The duration of the final isothermal mode was 20 min at 230°C. Sample volume was 0.2 μL. Vaporizer temperature was 180°C; detector, 220°C; ionization potential, 70 eV, mass scan range, 30-550 *m/z*.

Antimicrobial and fungicidal activity of dill-seed EO was investigated by the Barry method [10].

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