

„AROMA DALBA” CULTIVAR OF *PIMPINELLA ANISUM* L. DEVELOPED IN MOLDOVA

**GONCEARIUC Maria, Zinaida BALMUS, Ludmila COTELEA,
Svetlana MASCOVTEVA, Violeta BUTNARAS**

Institute of Genetics, Physiology and Plant Protection

Academy of Sciences of Moldova

mgonceariuc@yahoo.com

Abstract. A new cultivar of *Pimpinella anisum* L. named Aroma Dalba has been developed to be proposed for farm cultivation. The variety Aroma Dalba is distinguished by resistance to drought and to diseases. Essential oil separated by hydrodistillation from the seeds, constitutes 7.702% (dry matter). The yield of fruits (seeds) of new variety is 258 kg/ha and the production of essential oil are 20 Kg/ha. The essential oil production Qualitative and quantitative analyses using GC GC-MS attested that the essential oil contains 19 compounds, with an identification rate of 99.85%. There are five major components in the essential oil. The major compound of essential oil is trans-anethole in concentration of 89.0%. Other main compounds include iso-eugenol acetonitrile at a concentration of 4.59%; -himachalene (3.02%); butanoic acid,2-methyl- 4-methoxy-2-(3-methyloxiranyl) phenyl ester (1.13%).

Key words: *Pimpinella anisum*, cultivar, essential oil, trans-anethole, -himachalene., aceto iso-eugenol.

Introduction

Pimpinella anisum L., anise, is an annual aromatic and medicinal species in the family *Apiaceae*. As a farm crop, it existed in the 1st century B.C. on the Crete island and in Egypt [11]. According to other researchers, the species is native to the East, and was acclimatized in the conditions of the mild climate of Europe and Minor Asia, in the north of India, in Japan, and Chile [3]. It grows in the southern Mediterranean region, Western Asia, Middle East, Mexico, Egypt, and Spain [9]. The fruits (seeds) of the species are largely used as a spice and a medication, as well as a drug for preparation of aromatic substances [5, 11]. Anise is mentioned as a remedy by Hippocrates, while Pythagoras appreciated its therapeutic virtues.

Theophrastus, Dioscorides, Columella, and Pliny remarked in their works the qualities of Egyptian and Cretan anise [3]. As a phytotherapeutic species, anise has numerous properties, among them the most important are tonic, antispasmodic, diuretic, appetizing, anthelmintic, emenagogous etc. Anise is known for its antifungal action [7], in addition, it is an excellent galactogogue and a remedy largely utilized in the treatment of various gastrointestinal conditions in children [12]. The species is indicated in asthenia, rheumatism, migraine, nausea, coughs (asthma, convulsive coughs), psychogenic vomit, gastric pains, slow digestion, flatulence, intestinal spasms etc. The essential oil of anise is utilized in the perfumery and cosmetic industry, for production of sanitary and hygienic articles. In the food industry, anise is employed as a flavoring and aromatic agent for products of fish, ice-cream, sweets [9]. Anise fruits are used as an analgesic for migraine, as well as a carminative, stomatal, disinfectant and diuretic in traditional medicine of many countries [2]. In cosmetics, anise is used to produce a better complexion; it is also efficient in teeth bleaching. Anise is mentioned for attenuation or treatment of melancholy, nightmares as well as in the treatment of epilepsy and convulsions [1].

Given the beneficial action of anise fruits and various pharmacological properties, we believe that it would be excellent to cultivate this species in Moldova. Therefore, a new variety of *Pimpinella anisum* L. adapted to the pedoclimatic conditions of this country has been developed and proposed for implementation, cultivation and employment to maintain and strengthen health.

Material and methods

The biological material included various genotypes of *Pimpinella anisum* L. The new variety Aroma Dalba has been developed through individual selections and evaluation of the descendents. The validation of the quantitative trait indices has been performed through trials in comparative competitive crops testing in Comparative Cultures of Contest according to the methods utilized by State Commission for Plant Variety Testing. Integral experiments were set in the last decade of March, 2011, 2012, and 2013 with a seeding rate of 3 kg/ha at a row distance of 70 cm. Fertilization was carried out using Ammophos at a dose of 150 kg/ha applied during plowing. The plot surface in TCCC was 12 sq. m, the number of replicates was four. The essential oil was extracted from mature

seeds ground through hydrodistillation in Ginsberg apparatus. The content of essential oil was recalculated for dry matter and kept in a freezer. The concentrations of the essential oil components were measured using gas-chromatography (GC) and mass-spectrometry (GC-MS). The chromatography conditions were as follows: Gas Chromatograph Agilent Technologies, type 7890, A GC system, MS Agilent Technologies, type 5975 C Mass Selective Detective. The column HP 5MS 30 m x 0.25 mm x 0.25 μ m (5% Phenylmethylsiloxane).

Results and Discussion

The development of a new variety named Aroma Dalba of *Pimpinella anisum* L. was finalized in 2013 (Fig.1). The new variety is distinguished through a pivotant root, erect stem, 58.0 cm tall, that is glabrous, striated with 10.8 ramifications in the upper portion that ends in inflorescences. The leaves are differentiated by the level: two-three lower leaves arranged alternately are unbroken, long-petiolate, ovate, with dentate edges, the upper ones, also low in the number, are sessile, twice-thrice pinnate with linearly lanceolate folioles. The flowers are composed of 15.2 compound umbels, without involucre, with 16 umbels on each flower, the flowers lack calyx and have five white petals, ciliated on edges, on the top with a binding lobe, 1-5 mm long. The fruits, small in size, are diachene, ovoid, with hard to separate halves; they have five ribs, slightly prominent of a greenish color. Thousand seed mass is 1.99 g, 1 gram contains 675 diachenes. The variety is resistant to drought and disease. Aroma Dalba variety was approved in the Republic of Moldova in 2014, the registration number – 2003225 [4]. A Plant Variety Patent Application V20130034 2013.11.13 was filed to the State Agency for Intellectual Property.

The Aroma Dalba variety may be employed in the following directions: leaves may be utilized as a spice, a source of vitamins; fruits may be used as a spice, as well as pharmaceutical products, in the production of essential oil from fruits. The seed production is 258 kg/ha, the content of the essential oil in fruits is 7.702% (dry matter). The production of the essential oil of the Aroma Dalba variety constituted 20.0 kg/ha.

Qualitative and quantitative analyses of the essential oil extracted from anise seeds of the new variety Aroma Dalma through gas-chromatography (GC) coupled with mass spectrometry (GC-MS) have

demonstrated that the essential oil contains 19 components, 99.85% of them being identified (Table 1). There are four major components:

- *trans*-anethole at a concentration of 89.0%;
- aceto *iso*-eugenol at a concentration of 4.59%;
- γ -himachalene (3.02%);
- butanoic acid, 2-methyl-,4-methoxy-2-(3-methyloxiranyl)

phenyl ester (1.13%).

As a major component with the highest concentration, *trans*-anethole has been identified in the anise essential oil in the different European regions, the concentration varying between 76.9% and 93.7%. The highest concentrations of more than 90% have been found in the samples from Greece, Hungary, Scotland, Lithuania, Italy, and Germany (Orav et al., 2008;) [6, 10]. Very high concentrations (92.4%) of *trans*-anethole have been found in anise fruits from Algeria (Saibi et al., 2012). The second major component in the essential oil extracted from the variety Aroma Dalba is *iso*-eugenol at a concentration of 4.59%, while the samples of the essential oil from different European countries contain γ -himachalene at concentrations ranging from 0.4% to 8.2% (Aghili Khorasani, Makhzan al Advieh, 2001; Orav et al., 2008; Samojlik, 2012).

This component in our research has been attested at a concentration of 3.02% (Table 1). As regards the other major components of the oil from Aroma Dalba, we can mention that some of them are similar to those of the Algerian anise oil – *iso*-eugenol and γ -himachalene, though their concentrations are lower than in the variety Aroma Dalba.

The minor components, at a number of 15, are contained at concentrations varying between 0.02% (α -Vatirenene) and 0.32% ((+) β -Bisabolene).

As opposed to the variety Aroma Dalba, the oil samples from different European countries contain more components (Orav et al., 2008).

Thus, a new anise (*Pimpinella anisum* L.) variety has been developed and approved to be proposed for implementation, cultivation, employment. The variety is suitable for both fruit production as a pharmaceutical stuff and processing to obtain essential oil of a good quality.

Table 1

Quantitative and qualitative analyses of the essential oil
from *Pimpinella anisum*, var. Aroma Dalba

Nr. pic	Rt. mostra	Component	Area, %	Nr. pic	Rt. mostra	Component	Area, %
1	11.01	Estragol	0.28	11	18.65	(-)- -Cedrene	0.54
2	12.46	<i>cis</i> -Anetol	0.22	12	18.84	-Himachalene	0.16
3	13.47	<i>trans</i> -Anetol	89.00	13	18.97	(+) -Bisabolene	0.32
4	14.72	-Elemene	0.07	14	19.34	-Cadinene	0.06
5	15,08	<i>trans</i> - - Bergamotene	0.04	15	21.81	-Vatirenene	0.02
6	15,61	Ylangene	0.03	16	22.22	(+) -Longipinene	0.03
7	17.59	<i>cis</i> - - Himachalene	0.20	17	26.35	Aceto iso-eugenol	4.59
8	17.67	-Farnesene	0.03	18	27.44	acid butanoic, 2-methyl, 4-methoxy-2-(3-methyl oxiranyl) phenyl ester	1.13
9	18.3	-himachalene	3.02	19	31.26	Sclareol	0.02
10	18.53	Eugenol methyl ester	0.09	Total compound identified			99.85

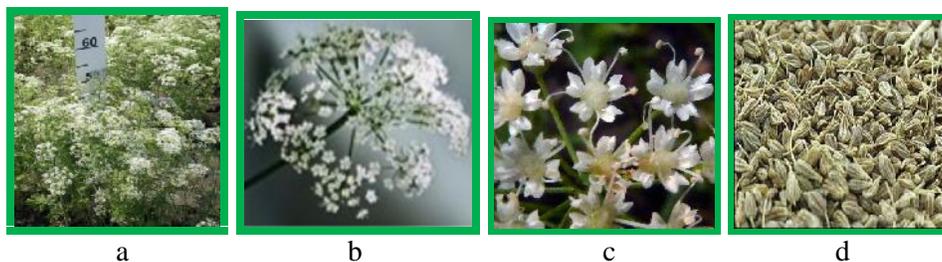


Fig. 1. *Pimpinella anisum* L., var. Aroma Dalba:
a - plants, flowering stage; b-inflorescence; c- flowers, d-fruits.

Conclusion

1. A new variety of anise (*Pimpinella anisum* L.) named Aroma Dalba is proposed for cultivation.
2. The variety Aroma Dalba ensures a production of fruits (seeds) of 258 kg/ha.
3. The content of essential oil in seeds is 7.702% (dry matter).

4. Qualitative and quantitative analyses have established that the essential oil contains 19 components, the rate of identification making 99.85%.
5. The major components of the essential oil extracted from the fruits the variety Aroma Dalba include *trans*-anethole at a concentration of 89.0%, aceto iso-eugenol at a concentration of 4.59%; -himachalene (3.02%), and butanoic acid, 2-methyl-,4-methoxy-2-(3-methyloxiranyl) phenyl ester at a concentration of 1.13%.

References

1. Aghili Khorasani M.H., Makhzan al Advieh (2001). Tehran, Iran: Bavardaran Press. Research institute for Islamic and Complementary Medicine, Iran University of Medical Sciences, p.34-39.
2. Amin G.R. (2005). Popular Medicinal Plants of Iran. Tehran, Iran: Vice-Chancellorship of Research, Tehran University of Medical Science Press, p.78-91.
3. Fischer E. (2000). Anason. Dic ionarul plantelor medicinale. Gema Press, Bucure ti, p.56-59.
4. Machidon M. (2014). Catalogul soiurilor de plante al Republicii Moldova. Edi ie special . Edit. lumina, Chi in u, p.36.
5. Mirheydar H. (2001). Herbal Information: Usage of Plants in Prevention and Treatment of Diseases. Tehran, Iran: Islamic Culture Press Center, p.12-19.
6. Orav A., Raal A., Arak E. (2008). Essential oil composition of *Pimpinella anisum* L. fruits from various European countries. J. Natural Product Research: Formerly Natural Product Letters, Vol. 22, Issue 3, p. 227-232.
7. Özcan MM, Chalchat JC. (2006). Chemical composition and antifungal effect of anise (*Pimpinella anisum* L.) fruit oil at ripening stage. Annals of Microbiology. 56(4):353–358.
8. Saibi S., Belhadj M., Benyoussef EH. (2012). Essential oil composition of *Pimpinella anisum* from Algeria. Analytical Chemistry Letters, Vol.2, Issue 6, p. 401- 404.
9. Salehi Surmaghi MH. (2010). Medicinal Plants and Phytotherapy. Vol. 1. Tehran, Iran: Donyay Taghziah Press, p. 45-48.

10. Samojlik I., Mijatovic V., Petkocic S., Skrbic B., Bozin B. (2012). The influence of essential oil of aniseed (*Pimpinella anisum* L.) on drug effects on the central nervous system. *Fitoterapia*, 83(8), p. 1466-73.
11. Voitkevici S.A. (1999). Anisovoe maslo. Efirnie masla dlea parfiumerii I aromaterapii. *Pishcevaia promishlenosti (rus.)*, v , p.24-27.
12. Zargari A. (1996). *Medicinal Plants. Vol. II*. Tehran, Iran: Tehran University Publications, pp. 592–602.