

VARIABILITY OF CONTENT AND CHEMICAL COMPOSITION OF THE ESSENTIAL OILS WITH HETEROSIS EFFECT IN HYBRIDS OF *Lavandula angustifolia* MILL.

GONCEARIUC Maria, BUTNARAȘ Violeta, MAȘCOVȚEVA Svetlana, BALMUȘ Zinaida, COTELEA Ludmila, BOTNARENCO Pantelimon

Abstract. We have developed and assessed intraspecific *Lavandula angustifolia* Mill hybrids. The indices of the quantitative characters have been shown to vary between the hybrids and maternal forms. The content of essential oil quantified via hydrodistillation in the Ginsberg apparatus is much higher (3.939 – 5.480%, dry matter) in the hybrids produced than in the maternal forms from which they originated (2.722 – 3.413%). The effect of heterosis for the essential oil content varies considerably in different hybrids derived from different maternal parents and constitutes +37.8 – +110.5%. The quantitative and qualitative analyses of the essential oil assessed with the aid of GC-SM techniques have demonstrated considerable differences of the oil in different hybrids depending on the maternal form. The number of the components identified in the essential oil ranges between 18 and 38. The major components in the essential oil are as follows: linalool (24.15 – 50.84%); linalyl acetate (27.29 – 44.40%); α – terpineol (3.82 – 4.84%); terpinen-4-ol (1.11 – 9.21%); lavandulol acetate (0 – 2.22%). The hybrids with a very high content (4.939-6.164 %, dry matter) of essential oil, different vegetation period from early- to late-ripening and resistant to drought have been selected. Their cloning has resulted in new varieties named Fr.5S8-24, VM-18V and Fr.8-5-15V.

Keywords: *Lavandula angustifolia*, hybrid, heterosis, essential oil, chemical composition.

Rezumat. Variabilitatea conținutului și componenței chimice a uleiului esențial la hibridi cu efect al heterozisului de *Lavandula angustifolia* Mill. Au fost creați și evaluați hibridi intraspecifice de *Lavandula angustifolia* Mill. S-a demonstrat că indicii caracterelor cantitative variază între hibridi și formele materne. Conținutul în ulei esențial, determinat prin hidrodistilare în aparate Ginsberg, este mult mai ridicat (3,939 – 5.480% s.u.) la hibridii creați decât la formele materne de la care au provenit (2.722 – 3.413). Efectul heterosis la conținutul de ulei variază considerabil la diferiți hibridi, derivați de la diferite forme parentale și constituie +37.8 – +110.5%. Analiza cantitativă și calitativă a uleiului esențial, evaluat prin tehnici GC-SM a stabilit diferențe considerabile ale uleiului la diferiți hibridi în funcție de forma maternă. Numărul componenților identificați în uleiului esențial variază de la 18 până la 38. Componenții majori în uleiul esențial sunt următorii: linalool, 24,15 – 50,84%; linalyl acetate, 27,29 – 44,40%; α – terpineol, 3,82 – 4,84%; terpinen-4-ol, 1,11 – 9,21%; acetat de lavandulol, 0 – 2,22%. Au fost selectați hibridi cu conținut foarte ridicat (4.939-6.164 % s.u.) de ulei esențial, cu perioada de vegetație diferită de la timpurie până la tardivă, rezistenți la secetă. Clonarea acestora a rezultat elaborarea soiurilor noi Fr.5S8-24, VM-18V, Fr.8-5-15V,

Cuvinte cheie: *Lavandula angustifolia*, hibrid, heterosis, ulei esențial, componența chimică.

INTRODUCTION

Lavandula angustifolia Mill., lavender is a perennial semi-shrub from the Lamiaceae family, a medicinal, aromatic, as well as melliferous and decorative species of Mediterranean origin. Lavender and the products derived from this species have been used for ages as a therapeutic agent (CAVANAGH & WILKINSON, 2005; GONCEARIUC, 2008) in traditional medication in Asia, Europe, antique Greece and Rome. The utilization of lavender for medicinal, phytotherapeutic and pharmacologic reasons is due to the essential oil and its components separated from inflorescences (GONCEARIUC, 2008). The essential oil is appreciated as a disinfectant, analgesic and scar-healer; it is also known for its choleric, diuretic, carminative and sudorific actions (PĂUN, 1995). Lavender and lavender oil are well-known for tonic, antifebrile and antiseptic properties (STANOJEVIĆ et al., 2011). Like other essential oils, lavender oil is an excellent anti-inflammatory (HANCIANU et al., 2013), antimicrobial (CESUR TURGUT et al., 2017) and antifungal (antimycotic) agent (STANOJEVIĆ et al., 2011; BEHMANESH et al., 2015). The pronounced antiseptic action and beneficial capacity of the essential oil in wound healing have been known since World War I (VOITKEVICI, 1999). Some researchers believe that lavender oil is the strongest antibacterial and the best inhibitor of bacterial growth (CZERWIŃSKA & SZPARAGA, 2015; VARBAN et al., 2017). Studies have demonstrated a sedative action of the essential oil on the central nervous system (LOPEZ, 2017), efficiency in treating depression, stress and anxiety (AKHONDZADEH et al., 2003; KHITAN et al., 2006; SETZER, 2009; KASPER, 2010, 2013; HANCIANU et al., 2013; ANDRYS & KULPA, 2016), including postnatal depression; other disorders of the central nervous system (WHEATLEY, 2005; LOPEZ et al., 2017; RAHMATI et al., 2017) and improvement of spirit and general health condition (CONRAD & ADAMS, 2012; UEHLEKE et al., 2012). Preclinical studies have found anxiolytic effects of lavender essential oil (CHIOCA et al., 2013; RAHMATI et al., 2017).

The employment of lavender to treat neurological disorders such as migraine, stress, anxiety, including depression, have been traditional (AKHONDZADEH et al., 2003; KASPER et al., 2010; KASPER, 2013; ANDRYS & KULPA, 2016; RAHMATI et al., 2017). The findings of a thorough study have proved that lavender oil is a viable option for dealing with such neurological dysfunctions as stroke due to its antioxidant properties (WANG et al., 2012). Both essential oil and inflorescences are used in treating migraines, headaches, heart disorders with a nervous substrate through regulation of anxiety conditions of some internal receptors; in neuro-vegetative dystopias, digestive

disturbances, abdominal bloating, insomnias, kidney diseases, rheumatic diseases, biliary dyskinesia, influenza, cold, coughing, bronchial asthma and stomatitis (BOJOR & ALEXAN, 1994; PĂUN, 1995; GRIGORESCU & SILVA, 1997; STEVENSON & WAITE, 2011). Externally, lavender essential oil is used to treat burns, frost-bite and allergic rash. The researches show that it serves as a natural treatment for the Alzheimer disease, as well. The studies carried out on rats demonstrate that inhalation of lavender essential oil vapours may help prevent stress and improve cognitive disorders (HANCIANU et al., 2013; KASPER, 2013; PAN XU et al., 2017). In aromatherapy and massage, this oil is also frequently used, due to the major benefits, in treating central nervous system disorders as a sedative, antioxylic and modulator of mood condition.

Lavender is a valuable melliferous species since the flower nectar is rich in carbohydrates, while the blossoming period is quite long making 35-40 days. Lavender is also known and employed as a decorative plant (CUCU et al., 1982; GONCEARIUC, 2004, 2008; GONCEARIUC et al., 2011). The excellent aspect of evergreen plants, inflorescences with flowers of different hues of blue, quite long blossoming period, delicate smell of the flowers are exploited with a particular success in southern France and in whole Europe, as well.

The above mentioned influences constantly the studies on the chemical composition, qualities, benefits of the utilization of flowers, essential oil and other derivatives of lavender, as well as the diversity of essential oil employment depending on its qualitative and quantitative composition.

A particular area includes the studies carried out to develop new hybrids and cultivars that are resistant to abiotic factors that would ensure an enhanced production of inflorescences with a higher content of essential oil and a quality that corresponds to the purpose proposed for utilization. Such studies have evidently intensified during the last decade as a consequence of the climatic changes, slow but steady processes of global warming, transformation of some zones into a desert including in south-eastern Europe where farm crops are increasingly impaired by drought and scorching heat while *L. angustifolia*, *Salvia sclarea* and *S. officinalis* cultivars have ensured more enhanced production of high quality essential oil during the dry years than in the years with normal atmospheric precipitations. This work also belongs to this area of research.

MATERIAL AND METHODS

The biologic material is represented by polycross hybrids of the first generation of *Lavandula angustifolia* derived from six maternal forms of different genetic and geographic origin. The evaluation of the biomorphological characters (quantitative) have been accomplished in integral experiments according to applicable requirements. Moldovan cultivars named Moldoveanca 4, Vis Magic 10 and Alba 7 (GONCEARIUC, 2014) served as witnesses. The content of essential oil was quantified in fresh inflorescences at the technical maturation stage through hydrodistillation in Ginsberg apparatus for 45 min. The findings were recalculated for dry matter. The qualitative and quantitative composition of essential oil was determined by GC GC-MS. The analysis equipment included gas-chromatograph Technologies Agilent 7890 equipped with Selective Mass Detector with Quadruple MSD Agilent Technologies 5975C, capillary column (30 m/0.25 mm/0.25 µm) with non-polar stationary phase HP-5ms. The analysis was performed at a temperature of 250 °C; the injector and detector temperature was 280 °C, temperature gradient from T1 = 70 °C (2 min), T2 = 200 °C (5 °C/min), T3 = 300 °C (20 °C/min, 5 min) was used. Mobile phase: Helium 1 ml/min, injected volume was 0.03 ml of essential oil, split rate - 1:100. Identification of chromatographic peaks was performed using the software package AMDIS™, coupled with the NIST database.

RESULTS AND DISCUSSION

The earlier studies have demonstrated that intraspecific hybridization is an efficient technique to develop valuable genotypes due to pronounced variability of the indices of biomorphological character values including those of the content, qualitative and quantitative composition of essential oil. This technique is quite difficult to perform for *L. angustifolia* subspecies *angustifolia*. Lavender flowers are small in size; therefore emasculation works on them are meticulous. Mostly, this operation, as well as further manual pollination damages a floral organ, which leads to very low setting of hybrid seeds. For these reasons, and because of the lack of the staff trained to carry out this kind of activity, it has been decided to develop more hybrids through free pollination, this being a new stage of inducing genetic variability in lavender and in the development of promising genotypes. The difference between the conventional free pollination and the method of producing polycross hybrids we have used consists in the fact that in the hybridization nursery, the parental forms assessed earlier were involved and selected for particular biomorphological characters to be inherited by the hybrids resulted from the hybridizations, including a large number of floral stems per plant, enhanced content of essential oil, enhanced content of linalyl in the oil, resistance to frost, wintering, drought, diseases etc. Ten parental forms selected are of different genetic and geographic origin and are distinguished not only by the high values of the indices of the above biomorphological characters but by the vegetation period, the colour of flower corolla, the colour and density of leaf pubescence etc.

L. angustifolia is an allogamous species, thus, each parental form might have been, in the case of some hybrids, a maternal form and a paternal form for other hybrids. While collecting hybrid seeds from a particular genitor, it was known that it was a maternal form. Hybrids of the first generation (F1) were produced and assessed from six maternal forms in

further studies. Each form was attributed a cipher Fr.1, Fr.5, Fr.8, Cr.13, Cr.26 and VM. The maternal forms Fr.1, Fr.5 and Fr.8 are of French origin. The maternal forms Cr.13 and Cr.26 are generative offsprings of the Ukrainian cultivar Crîmcheanca. The cipher VM identifies the Moldovan cultivar Vis Magic 10 used as a maternal form.

The newly developed and assessed hybrids differ from the maternal forms by vegetation duration (Table 1) and a number of biomorphological characters including essential oil content (Table 1; Fig. 1).

The same late-ripening maternal forms (Fr.1 and Fr.2) have been found to produce hybrids with different vegetation duration, both late- and early ripening ones, while the technical maturity period being average. The difference between the vegetation duration calculated from the beginning of vegetation and up to technical maturity makes from 2 to 19-20 days (Table 1, Fig. 1) (BUTNARAȘ, 2016). Such early-, mid- or late-ripening lavender hybrids propagated vegetative may become the cultivars that would produce a conveyer during harvesting. Each cultivar might be harvested in optimal dates, which allows elimination of raw material and essential oil losses. Simultaneously, the cultivated-processed areas under lavender might be extended without increasing the number of agricultural machinery and the capacity of processing equipment.

Table 1. Essential oil content in the lavender hybrids derived from the maternal forms (♀) Fr.1, Fr.8.

Vegetation period	Hybrid, maternal form	Essential oil content, % (dry matter*)			
		2011	2012, dry year	2013	X
early	Fr.1-3-2V	4,141	5,386	5,290	4,939
	Fr.1-3-23V	5,165	5,613	5,436	5,405
	Fr.8-5-15V	4,214	5,080	4,965	4,753
	Fr.8-5-23V	4,492	5,682	4,682	4,952
middle	Fr.1-3-9V	4,648	5,568	5,425	5,214
	Fr.1-3-20V	3,939	4,403	4,497	4,279
	Fr.8-5-21V	4,158	4,337	5,444	4,646
	Fr.8-5-26V	3,950	4,645	4,560	4,385
	Fr.8-5-40V	4,172	4,765	4,965	4,634
late	Fr.1-3-5V	4,274	5,382	4,324	4,659
	Fr.1-3-13V	4,125	4,250	5,269	4,543
	Fr.8-5-34V	5,096	5,157	5,287	5,181
	Fr.1.♀	3,353	3,379	3,012	3,248
	Fr.8.♀	2,555	3,123	3,833	3,168

Legend: * Essential oil content, recalculated to dry matter.

All the first generation hybrids selected and presented in Table 1, 2 and Figure 1 have recorded a much more enhanced content (3.621– 5.682%, dry matter) of essential oil than the maternal forms Fr.1, Fr.8, Fr.5, Cr.26 and Cr.13 from which they originate.

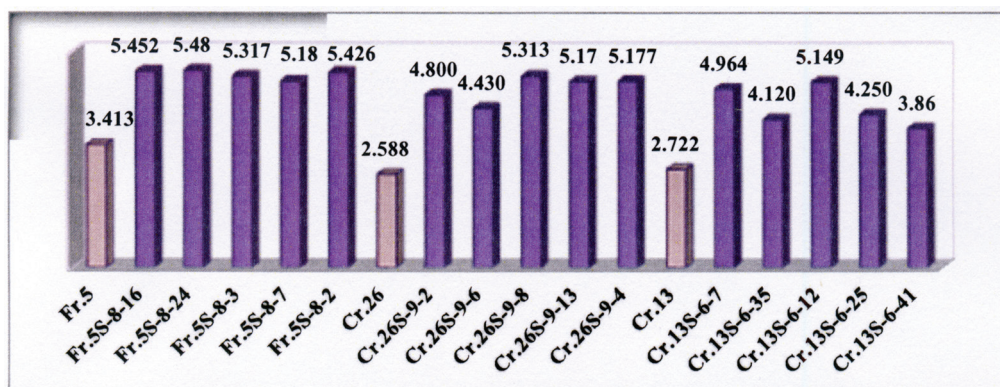


Figure 1. Essential oil content (% dry matter) in *L. angustifolia* F1 hybrids in relation to the *L. angustifolia* maternal forms Fr.5, Cr.26 and Cr.13.

Importantly, the hybrids developed are greatly resistant to drought. Thus, in the dry years, the content of essential oil is higher in all the hybrids than in the previous year (in this case, 2011, Table 1). Acute drought is known to have a negative impact on species and perennial farm crops in both the year of major deficit of atmospheric precipitation and in the subsequent year (years). In the case of the lavender hybrids, we have developed and selected, the consequences of the 2012 year drought were different. In 2013, all the F1 hybrids recorded a higher content of essential oil in comparison with the maternal forms from which they originate (as in the years 2011 and 2012), while five of these hybrids synthesized and accumulated even higher content of essential oil in the year 2013 than in 2011 and 2012.

The similar results (Table 2) were recorded in the years 2015-2017 while testing the promising hybrids Fr.5S8-24, VM-18V and Fr.8-5-15V in comparison with the patented cultivars.

Table 2. Essential oil content in the *Lavandula angustifolia* varieties, comparative competitive crops.

Hybrid F ₁ , variety	Essential oil content, % (dry matter)			
	2015, dry year	2016	2017	2018
Fr.5S8-24	6.164	5.786	5.915	5.955
VM-18V	5.103	4.924	4.829	4.952
Fr.8-5-15V	5.803	4.691	5.454	5.316
Vis Magic 10, witness	4.575	4.597	4.518	4.563
Alba 7, witness	5.762	5.915	5.256	5.644
Moldoveanca 4, witness	5.404	4.318	4.981	4.901

Considerable variability of the F₁ hybrids is attested for the character vegetation period. For example, the hybrids derived from the late-ripening maternal forms Fr.1 and Fr.8 have an average vegetation period ranging between 54.6 days and 70 days (Fig. 2). The high content of essential oil is due to the heterotic effect exhibited in the lavender hybrids from the first generation F₁. The long-term studies have demonstrated that the F₁ hybrids produced from polycross hybridization manifest heterosis in a number of quantitative characters that have a direct impact on the productivity. All the hybrids derived from all the maternal forms exhibit heterosis for essential oil content (Figs. 3 and 4), it being supported by the biomorphological quantitative characters.

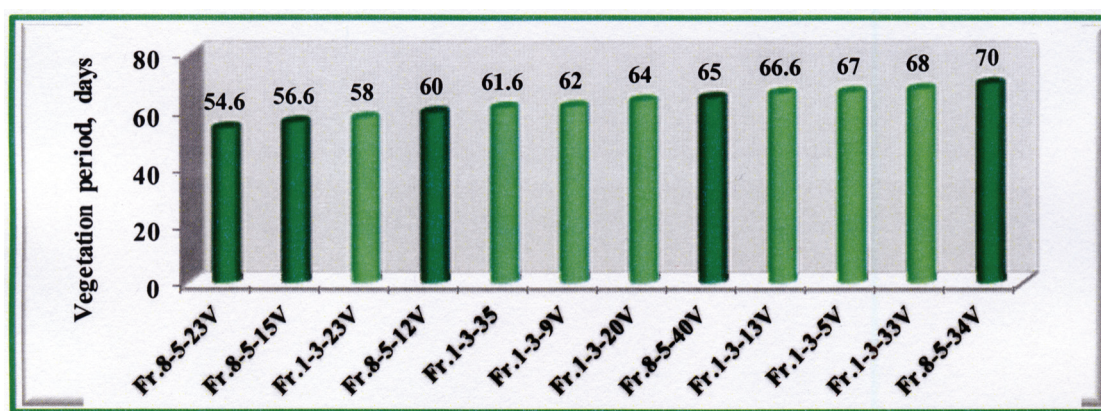
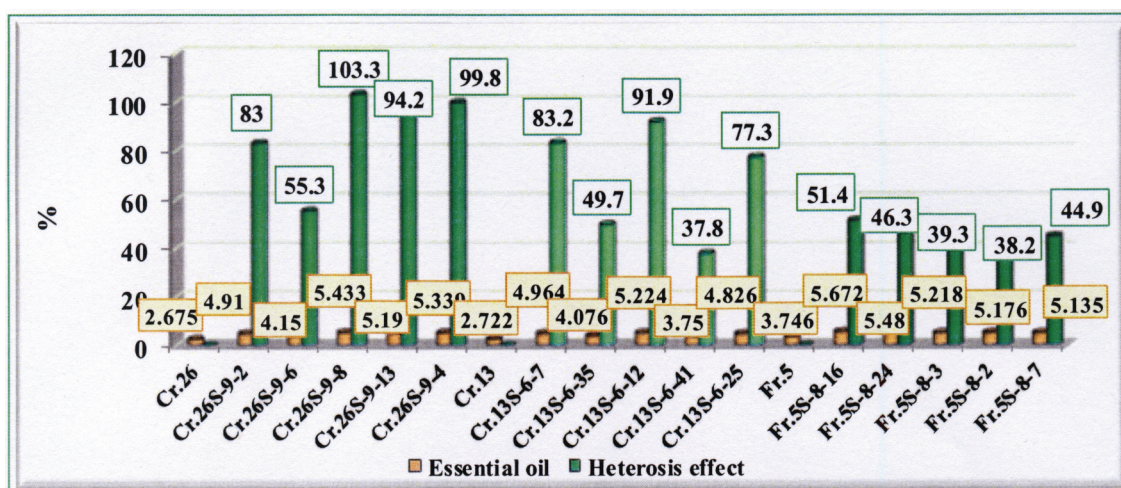


Figure 2. Vegetation period (days) of the hybrids derived from the late-ripening maternal forms Fr.1 and Fr.8.

The amplitude of the heterosis recorded in each first generation hybrid (F₁) for the character essential oil content is influenced by the maternal form. This statement is confirmed by the fact that the hybrids that originate from the maternal form Cr 26 exhibit a heterotic effect for this character within +55.3% and +103.3% (Fig. 3).

Figure 3. Heterosis effect (%) for essential oil content (% dry matter) exhibited by the *L. angustifolia* F₁ hybrids derived from the maternal forms Cr. 26, Cr. 13 and Fr. 5.

In the hybrids derived from the maternal form Cr.13, the heterotic effect for the essential oil content varies between +37.8 and 91.9% depending on the hybrid. The hybrids with a higher heterotic effect for this character belong to the form Fr.5, the heterotic effect varying in different hybrids between +38.2% and +51.4%. These results can be explained by the fact that Fr.5 is a maternal form with the highest content of essential oil (3.746%, dry matter).

Importantly, some hybrids derived from this form, such as Fr.5S-8-16 are distinguished by a very high content of essential oil (5.672%, dry matter) (Fig. 3).

The expression of the heterosis recorded in the hybrids derived from the maternal form Fr.8 is more significant, the variation being less spectacular than in the hybrids originated from the maternal forms Cr.26, Cr.13 and Fr.5. Thus, the heterotic effect attested in the hybrid Fr.8-5-5V is very high making +65.3%, so this value is the lowest vs. other hybrids of this maternal form in which this important index makes +70.1 – +110.5% (Fig. 4).

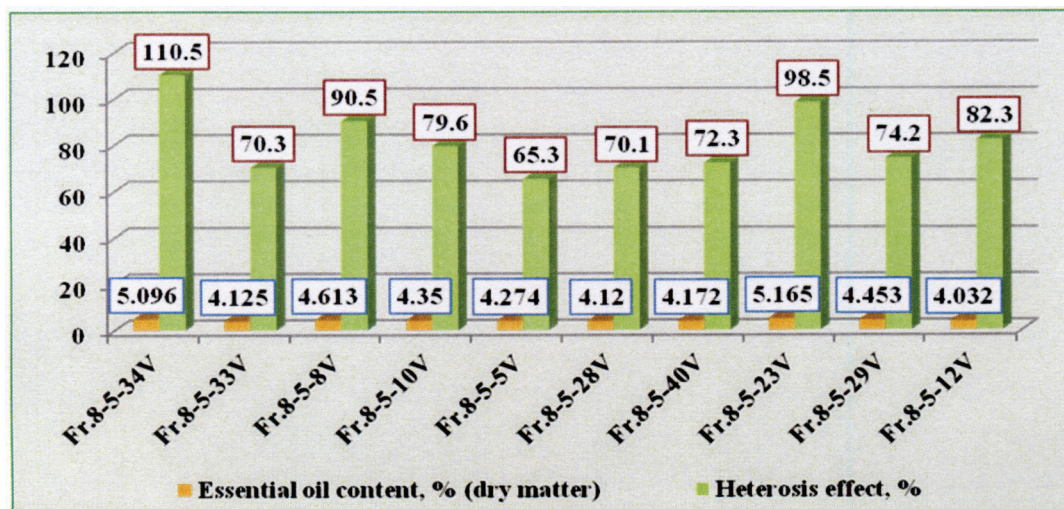


Figure 4. The heterosis effect to the essential oil content by *L. angustifolia* F₁ hybrids originated from the maternal form Fr.8.

We can state that a new generation of hybrids has been developed, some of them recording a high heterotic effect for essential oil content from 37.8% up to 110.5%. The same hybrids exhibit positive heterotic effects for both the character length of floral spike (+13.3 – +23.0%) and the number of verticiles per floral spike (+5.4 – +18.1%), including other biomorphological characters (MASHCOVTZEVA & GONCEARIUC, 2012, 2013; BUTNARAȘ, 2012, 2016; BUTNARAȘ et al., 2013; MAȘCOVȚEVA, 2018).

Taking into consideration the fact that valuable hybrids can be reproduced vegetatively through cloning and may serve a basis for the development of new varieties that inherit all the biomorphological and biochemical characters of the initial hybrid genotypes, this genetic material is invaluable. For example, the hybrids Fr.5S-8-24, Fr.8-5-15V and VM-18V which were further propagated vegetatively and, thus, became cultivars (clone cultivars) are distinguished by accumulation of considerable content of essential oil from 4.691 up to 6.164% (dry matter) (Table 2) including in the dry years.

Evaluation of the content of essential oil in these cultivars (F₁ hybrids) vs. the certified and patented cultivars has confirmed the results obtained earlier (Table 2). (It should be mentioned that our certified and patented cultivars are also F₁ hybrids). The findings confirm the fact that the hybrids and cultivars developed are characterized by a very high content of essential oil with values ranging between 4.691 and 6.164% (dry matter) in both the dry year 2015 and the following years 2016 and 2017.

The qualitative and quantitative analyses of the essential oil separated through hydrodistillation in laboratory conditions performed using gas chromatography coupled with mass-spectrometry (GC-MS) have demonstrated that the number of the components identified in the essential oil varies from 21 up to 38 (Tables 3 and 4). The concentration of each component varies with the hybrid, distinctness being attested between both hybrids and maternal forms from which they have derived and between the maternal forms of the hybrids (GONCEARIUC et al., 2011; BUTNARAȘ et al., 2013; BUTNARAȘ, 2016; MAȘCOVȚEVA, 2018). A pronounced variation of the concentration of some components such as linalyl acetate depending on the year of evaluation has been found. For example, the findings published earlier (BUTNARAȘ et al., 2013) show that the content of linalyl acetate in the essential oil isolated from the F₁ hybrid Fr. 1-3-23V makes 25.44% while the further studies (BUTNARAȘ, 2016) demonstrate that its concentration constitutes 37.27%. The GC-MS analyses have recorded a content of camphor making within 0.25 and 0.75%.

The European Pharmacopoeia, 6th edition envisages the following content of the constituents in the essential oil of *L. angustifolia*: linalool (20.0-45%); linalyl acetate (25.0-46%); α - terpineol below 2.0 %; lavandulol acetate above 0.1 %; camphor below 1.2 % (ROBU, 2012). Thus, the major components in the essential oil isolated from the F₁ hybrids we have developed meet the pharmacological requirements with the exception of the α - terpineol content. This compound has a pronounced smell of lilac. Terpineols are generally intermediate compounds of flavour enhancer synthesis; they are also components of food essences. These compounds are used as solvents and floating agents. Terpineols have antimicrobial properties. Terpineol esters and acetic acid (terpinyl acetate) as a mixture of isomers are used to produce perfume compositions and perfumes.

Summing up the results obtained in assessing the most advanced F₁ hybrids we can conclude that the qualitative and quantitative composition of the major constituents in the essential oil varies within the following limits: linalool: 24,15 – 50,84%; acetat de linalyl acetate: 27,29 – 44,40%; α -terpineol: 3,82 – 4,84%;

Summarizing the results of the evaluation of the most promising F₁ hybrids developed we can conclude that the qualitative and quantitative composition of the major constituents in the essential oil varies within the following range. Terpinen-4-ol at high concentrations (above 2%) is considered to diminish the perfumery value of essential oil, rendering it an herbal flavour and scent. In our view, herbs have a smell of freshness and lavender oil is particularly appreciated due to the freshness and delicacy of the aroma. On the other side, this oil is used not only in perfumery.

Table 3. Variation of the qualitative and quantitative components of the essential oil in the *Lavandula angustifolia* first generation hybrids vs. maternal forms VM.

No.	Components	Aria (%)									
		Fr.8*♀	Fr.8-5-21V	Fr.8-5-34V	Fr.1*♀	Fr.1-3-23V	Fr.1-3-9V	VM*♀	VM-9V	VM-32V	VM-56V
1	o-xylene	0,16	-	0,28	-	0,17	0,13	0,14	-	-	-
2	α -Pinene	-	-	-	-	0,05	-	-	-	0,02	-
3	Camphene	-	-	-	-	-	-	-	0,03	0,04	0,04
4	1-Octen-3-ol	-	-	-	-	0,04	0,51	0,12	-	0,11	0,08
5	α -Phellandrene	-	-	-	-	-	-	0,46	-	0,02	-
6	β -Pinene	-	-	-	-	-	0,09	-	-	-	-
7	3-Octanone	0,31	0,34	0,47	1,11	0,08	-	0,09	-	-	0,02
8	β -Myrcene	0,28	0,21	0,40	-	0,28	0,41	0,19	0,33	0,25	0,34
9	4-Methyl-3-octanol	0,13	0,14	0,11	-	0,26	0,09	0,07	-	-	0,08
10	Hexyl acetate	0,41	0,18	0,36	-	0,36	0,42	0,15	0,14	0,08	0,20
11	ρ -Cymene	-	-	-	-	-	0,11	0,08	-	-	-
12	Limonene	0,16	0,13	0,20	0,92	2,84	0,57	-	0,31	0,65	0,65
13	β -Cadinene	-	-	-	0,51	0,50	-	-	-	-	-
14	1,8-Cineole	0,87	0,34	0,31	-	-	1,56	1,61	0,51	1,23	0,61
15	<i>trans</i> - β -Ocimene	1,16	0,15	4,68	1,83	3,12	2,2	0,66	1,71	0,34	0,63
16	<i>cis</i> - β -Ocimene	0,41	0,21	1,24	1,18	0,52	1,58	0,35	1,17	0,16	0,30
17	γ -Terpinene	-	-	-	-	0,27	0,11	0,10	-	-	-
18	α -terpinen terpinolen	-	-	-	-	0,16	0,10	0,12	-	-	-
19	(+) Linalool oxide	0,15	-	-	-	-	0,30	-	-	-	-
20	Δ^3 -Carene	0,25	0,12	0,15	-	0,11	0,38	0,27	-	0,12	0,23
21	(-)-Linalool	39,95	50,84	36,66	42,96	27,44	28,63	40,98	32,30	38,83	35,83
22	1-Octen-3-yl acetate	0,23	0,72	0,19	1,31	1,24	1,85	-	0,10	0,11	0,11
23	(+) Camphor	0,35	0,25	0,20	-	0,28	0,20	0,54	0,34	0,23	0,29
24	Lavandulol	-	0,95	-	-	0,98	0,37	-	-	0,91	-
25	Borneol	1,16	0,47	0,75	0,84	0,75	1,04	2,22	1,22	1,21	1,58
26	Terpinen-4-ol	0,72	6,08	5,55	6,54	6,97	5,76	6,40	9,21	7,39	9,59
27	Krypton	0,15	0,31	0,13	-	0,09	0,39	0,49	-	-	-
28	α -terpineol	4,71	4,77	3,82	4,74	4,84	4,22	3,45	4,02	2,77	3,96
29	Nerol	0,49	0,27	0,30	0,68	0,38	0,54	0,34	0,27	0,20	0,39
30	Iso-borneol	0,09	-	-	-	-	-	0,14	0,02	0,02	0,02
31	Geraniol	0,09	-	-	-	-	0,22	0,19	0,14	-	-
32	Linalyl acetate	39,98	38,47	37,00	29,28	37,27	40,62	33,92	43,42	41,05	37,40
33	Borneol acetate	0,27	-	0,32	0,05	0,18	-	0,12	0,22	-	-
34	Lavandulyl acetate	0,42	-	1,95	3,25	0,26	1,57	0,77	1,56	0,91	0,90
35	Nerol acetate	0,82	0,30	0,64	0,96	0,48	0,83	0,50	0,62	0,37	0,65
36	Geranyl acetate	1,58	0,64	1,22	1,77	0,88	1,64	0,95	1,22	0,76	1,32
37	β -Caryophyllene	2,00	2,49	1,44	1,61	2,45	1,86	1,66	1,41	1,59	1,59
38	(E)- β -Farnesene	0,60	0,76	0,81	-	-	-	0,88	0,75	0,57	1,77
39	Germacrene D	-	-	0,20	-	0,12	-	0,20	-	0,15	0,27
40	Caryophyllene oxide	0,16	-	-	0,52	0,55	0,14	0,74	-	0,39	0,47
	Nr. components	29	22	26	18	31	31	33	23	28	29
	Identification, %	99,41	99,99	99,96	99,99	99,36	99,66	99,90	99,96	99,70	98,53

Legend: *maternal forms♀: Fr.8, Fr.1, VM.

Table 4. Qualitative and quantitative composition of essential oil in the *L. angustifolia* F₁ hybrids vs. maternal forms.

Compound	Aria (%)						
	Cr.26*♀	Cr.26 S-9-8	Cr.13*♀	Cr.13S-6-12	Fr.5*♀	Fr.5S-8-16	Fr.5S-8-24
o-xylene	0,014	0,018	0,100	0,020	—	0,125	0,120
α -Pinene	—	—	—	—	—	0,050	—
Camphene	0,060	—	—	—	—	—	—
1-Octen-3-ol	—	0,210	—	—	—	0,230	0,050
β -Myrcene	0,265	0,220	0,270	0,120	0,365	0,350	0,360
4-Methyl-3-octanol	—	0,120	—	—	—	0,090	—
Hexyl acetate	0,145	—	—	—	—	—	—
ρ -Cymene	0,090	—	—	—	—	0,090	—

Limonene	0,197	0,080	—	0,100	—	0,290	0,270
β - Phellandrene	0,960	—	—	—	—	—	—
8-Cineole	1,550	0,210	—	—	—	1,900	0,390
<i>trans</i> - β -Ocimene	1,599	1,020	1,630	2,250	1,501	0,310	0,417
α - terpinen	0,128	—	—	—	0,310	0,430	0,170
<i>cis</i> - β -ocimen	0,918	0,610	1,630	0,250	0,535	0,330	0,354
γ - Terpinene	0,128	0,080	0,140	0,140	—	1,370	1,120
δ - Terpinene	0,070	0,080	—	—	—	0,430	—
Linalool oxide	—	0,270	—	—	0,227	0,070	0,090
(-)-Linalool	39,554	42,570	45,500	43,560	34,196	24,150	37,250
1-Octen-3-yl acetate	—	0,210	—	—	—	0,230	0,050
(+) Camphor	0,248	0,210	—	—	0,753	0,390	0,186
Borneol	0,596	0,570	0,420	0,510	2,491	5,040	0,374
Terpinen-4-ol	9,721	4,490	12,580	10,500	8,449	3,830	1,118
α -Terpineol	3,551	3,340	2,720	2,790	4,110	3,740	3,420
Nerol	0,132	0,390	0,300	0,290	0,470	0,620	0,410
<i>iso</i> - borneol	—	—	—	—	—	0,070	—
Linalyl acetate	33,533	38,800	27,290	32,930	31,497	33,730	44,403
Borneol acetate	0,135	—	—	—	0,918	1,960	1,380
Lavandulol	—	0,210	—	—	—	0,330	0,410
Neryl acetate	0,555	—	—	0,350	0,740	0,720	0,599
Geraniol acetate	—	1,100	—	—	—	1,910	1,460
Nerol acetate	—	0,530	0,420	—	—	0,360	0,220
β- Caryophyllene	1,265	1,280	1,060	1,430	2,609	3,720	1,598
Caryophyllene oxid	0,201	0,520	—	—	0,890	0,710	0,455
(E)- β -Farnesene	—	0,610	0,590	0,400	—	0,480	0,290
Germacrene D	0,527	—	0,190	—	0,255	0,640	0,133
3-Octanol	—	—	—	—	0,194	—	—
Δ^3 -Carene	—	0,310	—	0,140	0,350	0,380	0,130
Eucalyptol	0,437	—	—	—	0,229	—	—
Hexyl acetate	0,145	—	—	—	—	—	—
n-Octen-acetate	0,605	—	0,750	0,400	1,230	0,805	0,365
Carvacrol	—	—	—	—	—	—	—
Lavandulol acetate	0,981	—	0,980	1,080	4,511	2,220	0,707
Timol	—	—	—	—	—	—	—
Geranyl acetate	0,976	—	0,830	0,670	1,417	1,310	1,203
p-xylene	—	—	0,100	—	—	0,100	0,100
n-Octen-1-ol	—	—	0,180	0,140	0,186	0,200	0,148
Krypton	—	—	—	—	—	1,970	0,250
Nr. components	29	27	20	21	26	38	34
Identification, %	99,495	97,628	97,900	97,890	98,441	97,050	100,0

Legend: * maternal form Cr.26, Cr.13, Fr.5

Terpinen-4-ol is known to have pronounced antimicrobial (HAMMER et al., 2012) and antifungal (MONDELLO et al. 2006), properties, these qualities being successfully exploited. Importantly, the content of camphor in the essential oil of all the hybrids developed and assessed meet the ISO standards and the requirements of the European Pharmacopoeia varying within the limits of 0-0.753%. Camphor at concentrations of more than 2% is known to certainly diminish the quality and aroma of essential oil.

CONCLUSIONS

F₁ hybrids of *Lavandula angustifolia* were created with high (3.939 - 5.480%) content of essential oil. The indices of the quantitative characters have been shown to vary between the hybrids and maternal forms. The effect of heterosis for the essential oil content varies considerably in different hybrids derived from different maternal parents and constitutes from +37.8 up to +110.5%. The quantitative and qualitative analyses of the essential oil assessed with the aid of GC-SM techniques have demonstrated considerable differences of the oil in different hybrids depending on the maternal form. The number of the components identified in the essential oil ranges between 18 and 38. The major components in the essential oil are as follows: linalool (24.15 – 50.84%); linalyl acetate (27.29 – 44.40%); α - terpineol (3.82 – 4.84%); terpinen-4-ol (1.11 – 9.21%); lavandulol acetate (0 – 2.22%). The hybrids with a very high content (4.939-6.164 %, dry matter) of essential oil, different vegetation period from early- to late-ripening and resistant to drought have been selected. Their cloning has resulted in new varieties named Fr.5S8-24, VM-18V and Fr.8-5-15V2.

REFERENCES

AKHONDZADEH S., KASHANI L., FOTOUHI A., JARVANDI S., MOBASERI M., MOIN M., KHANI M, JAMSHIDI AH., BAGHALIAN K., TAGHIZADEH M. 2003. Comparison of *Lavandula angustifolia* Mill.

- tincture and imipramine in the treatment of mild to moderate depression: a double-blind, randomized trial. *Programe Neuropsychopharmacol Biology Psychiatry*. Elsevier. Paris. **27**(1): 123-127.
- ANDRYS D. & KULPA D. 2016. *Lavandula* spp. essential oils – its use, composition and genetic basis of production. *Folia Pomeranae Universitas Technologiae Stetinensis, Agric., Aliment., Pisc., Zootech.* **328**(39)3: 7-22 (Accessed February 15, 2018).
- BEHMANESH F., PASHA H., SEFIDGAR A., TAGHIZADEH A., MOGHADAMNIA A., ADIB RAD H., SHIRKHANI L. 2015. Antifungal effect of lavender essential oil (*Lavandula angustifolia*) and clotrimazole on *Candida albicans*: An *In Vitro* Study. *Scientifica* (Cairo). Article ID: 261397. Epub. 2015 Oct.13. Vol. 2015, 5 p. doi: 10.1155/2015/261397. (Accessed 10 February, 2018).
- BOJOR O. & ALEXAN M. 1994. *Plante medicinale de la A la Z. Ediția a III-a*. Edit. Ulpia Traiană. București. 253 pp.
- BUTNARAŞ V. 2012. Manifestarea heterozisului la hibridii policross F₁ de *Lavandula angustifolia* Mill. *Buletinul Academiei de Ştiințe a Moldovei. Ştiințele vieții*. Tipografia ASM. Chişinău. **2**(317): 84-90.
- BUTNARAŞ V., GONCEARIUC MARIA, MAŞCOVŢEVA S. 2013. The perspective policross hybrids of *Lavandula angustifolia* Mill. *Oltenia. Studii și comunicări. Ştiințele naturii*. Muzeul Olteniei Craiova: **29**(1): 66-70.
- BUTNARAŞ V. 2016. *Crearea și evaluarea hibridilor policross de Lavandula angustifolia* Mill. Autoreferatul tezei Ph.D. Institutul de Genetică, Fiziologie și Protecție a Plantelor. Edit. Prin-Caro. Chişinău. 26 pp.
- CAVANAGH H. & WILKINSON J. 2005. Lavender essential oil: a review. *Healthcare Infection*. Universitas Press. London. **10**(1): 35-37.
- CESUR TURGUT A., EMEN F. M., SECILMIS CANBAY H., DEMIRDOĞEN R. E., CAM N., KILIC D., YESILKAYNAK T. 2017. Chemical characterization of *Lavandula angustifolia* Mill. as a phyto-cosmetic species and investigation of its antimicrobial effect in cosmetic products. *Journal of Turkish Chemical Society (OTCSA)*. Universitas Press. Ankara. **4**(1): 283-298.
- CHIOCA L., FERRO M., BARETTA I., OLIVEIRA S., SILVA C., FERREIRA J., LOSSO E., ANDREATINI R. 2013. Anxiolytic-like effect of lavender essential oil inhalation in mice: participation of serotonergic but not GABA/benzodiazepine neurotransmission. *Journal Ethnopharmacology*. Elsevier. Paris. **147**: 412-418.
- CONRAD P. & ADAMS C. 2012. The effects of clinical aromatherapy for anxiety and depression in the high risk postpartum woman - a pilot study. *Journal Complementary Therapies in Clinical Practice*. **18**(3): 164-168. doi: 10.1016/j.ctcp.2012.05.002. (Accessed 10 February, 2018).
- CUCU V., BODEA C., CIOACĂ C. 1982. Plante medicinale și aromatice. În: *Tratat de biochimie vegetală*, 1982. Partea a II-a. Edit. Academiei RSR. București. **4**: 219-225.
- CZERWIŃSKA E. & SZPARAGA A. 2015. Antibacterial and Antifungal Activity of Plant Extracts. *Rocznik Ochrona Srodowiska*. Snapshot Press. Kiev. **17**: 209-229.
- GONCEARIUC M. 2004. Genotipuri noi de levănțică (*Lavandula angustifolia* Mill.). *Probleme actuale ale Geneticii, biotehnologiei și ameliorării*. Centrul Editorial UASM. Chişinău: 258-262.
- GONCEARIUC M. 2005. Ameliorarea plantelor Aromatice și Medicinale în Moldova – Realizări și Perspective. *Genetica Ameliorarea Plantelor, Animalelor și Microorganismelor*. Congresul VIII SŞGA din R. Moldova. Tipografia Centrală. Chişinău: 329-334.
- GONCEARIUC M. 2008. *Lavandula angustifolia* Mill. În: Plante medicinale și aromatice cultivate. Centrul Edit. UASM: 99-120.
- GONCEARIUC M. 2014. Medicinal and aromatic plant varieties developed in the Republic of Moldova. *Oltenia. Studii și comunicări. Ştiințele Naturii*. Muzeul Olteniei Craiova. **30**: 29-34.
- GONCEARIUC M., MAŞCOVŢEVA S., BUTNARAŞ V., BALMUSH Z. 2011. The biodiversity of *Lavandula angustifolia* Mill. F₁ hybrids. *Oltenia. Studii și comunicări. Ştiințele naturii*. Muzeul Olteniei Craiova. **27**(1): 7-12.
- GRIGORESCU E. & SILVA F. 1997. De la etnomedicină la fitoterapie. *Tezaurul verde al medicinei*. Edit. Spiru Haret. Iași. **1**. 560 pp.
- HAMMER K., CARSON C., RILE T. 2012. Effects of *Melaleuca alternifolia* (Tea Tree) essential oil and the major monoterpene component terpinen-4-ol on the development of single- and multistep antibiotic resistance and antimicrobial susceptibility. *Antimicrobial Agents Chemotherapy*. Feb. **56** (2): 909-915. doi: 10.1128/AAC.05741-11. (Accessed February 10, 2018).
- HANCIANU M., CIOANCA O., MIHASAN M., HRITCU L. 2013. Neuroprotective effects of inhaled lavender oil on scopolamine-induced dementia via anti-oxidative activities in rats. *Journal Phytomedicine*. Mar. **15**. **20**(5):446-452. doi: 10.1016/j.phymed.2012.12.005. (Accessed February 11, 2018).
- KASPER S. 2013. An orally administered lavender oil preparation (Silexan) for anxiety disorder and related conditions: an evidence based review. *International Journal Psychiatry Clin Pract.* Taylor Francis Online, Nov, 17. Suppl. 1:15-22. *Pub Med*. doi: 10.3109/13651501.2013.813555. (Accessed February 5, 2018).
- KASPER S., GASTPAR M., MÜLLER W., VOLZ H., MÖLLER H., DIENEL A., SCHLÄFKE S. 2010. Silexan, an orally administered *Lavandula* oil preparation, is effective in the treatment of 'subsyndromal' anxiety disorder: a randomized, double-blind, placebo controlled trial. *International Clinical Psychopharmacology*: Sep. **25**(5):227-287. *Pub Med*. doi:10.1097/YIC.0b013e32833b3242. (Accessed February 10, 2018).
- KHITAN J. H., SHAHLAH JIHAD A. AL-SH., RAAD A. KASKOOS, JAVED AHAMAD, MOHAD JAMEEL, SHOWKAT M. 2013. Essential oil composition and antioxidant activity *Lavandula angustifolia* from Iraq.

- International Research Journal Pharmacology*. **4**(4): 117-120. doi: 10.7897/2230-8407.04421 (Accessed February 16, 2018).
- LÓPEZ V., NIELSEN B., SOLAS M., RAMÍREZ M., JÄGER A. 2017. Exploring pharmacological mechanisms of lavender (*Lavandula angustifolia*) essential oil on central nervous system targets. *Frontier in Pharmacology*. Edit. Institute of Genetics and Animal Breeding (PAS). Cracovia. **8**: 1-280.
- MASCOVTEVA S. & GONCEARIUC M. 2012. The expression of heterosis in the perspective F₁ polycross hybrids of the *Lavandula angustifolia* Mill. *Proceedings of 7th CMAPSEEC, Conference on Medicinal and Aromatic Plants of Southeast European Countries*, 27th - 31st May, 2012 Subotica, Serbia. Edit. Institute of Medicinal Plants Research "Dr Josif Pancic" & AMAPSEEC, Belgrade: 374-380.
- MAȘCOVȚEVA S. & GONCEARIUC M. 2013. Research on essential oil content and chemical composition of F₁ polycross hybrids of *Lavandula angustifolia* Mill. *International conference on natural products utilization: from plants to pharmacy shelf, ICNPU*, 3-6 Nov. Bansko, Bulgaria 2013: Book of Abstracts. Edit. The Phitochemical Society of Europe, AMAPSEEC: 46.
- MAȘCOVȚEVA S. 2018. *Influența formei materne în crearea hibridilor performanți de Lavandula angustifolia* Mill. Thesis Ph.D, 150 pp.; Abstract of the thesis Ph.D Institutul de Genetică, Fiziologie și Protecție a Plantelor. Edit. Print-Caro, Chișinău. 28 pp.
- MONDELLO F., DE BERNARDIS F., GIROLAMO A., CASSONE A., SALVATORE G. 2006. In vivo activity of terpinen-4-ol, the main bioactive component of *Melaleuca lternifolia* Cheel (tea tree) oil against azole-susceptible and -resistant human pathogenic *Candida species*. *BMC Infectious Diseases*. Nov 3; **6**:158.
- PAN XU, KEZHU WANG, CONG LU, LIMING DONG, LI GAO, MING YAN, SILAFU AIBAI, YANYAN YANG, XINMIN LIU. 2017. The protective effect of lavender essential oil and its main component linalool against the cognitive deficits induced by D-galactose and aluminum trichloride in mice. *Evidence-Based Complementary and Alternative Medicine*: 7426538. Doi: 10.1155/2017/7426538. (Accessed February 6, 2018).
- PĂUN E. 1995. Lavanda. În: *Sănătatea Carpaților. Farmacia din cămară*. Edit. F & D Stil Commerce. București: 129-133.
- RAHMATI B., KIASALARI Z., ROGHANI M., KHALILI M., FARIBA ANSARI. 2017. Antidepressant and anxiolytic activity of *Lavandula officinalis* aerial parts hydroalcoholic extract in scopolamine-treated rats. *Journal of Pharmaceutical Biology*. Taylor & Francis Press. London. **55**(1): 958-965.
- ROBU S. 2012. *Contribuții la studiul farmacognostic și biologic a unor specii de Lavandula cultivate în România*. Ph. D. Thesis. Universitatea de Medicină și Farmacie T. Popa. Iași. 40 pp.
- SETZER W. N. 2009. Essential oils and anxiolytic aromatherapy. *Conference Nat. Prod. Commun. US Medicine National Institute PUBMED/MEDLINE*. London. **4**(9): 1305-1316.
- STANOJEVIĆ L., STANKOVIĆ M., CAKIĆ M., NIKOLIĆ V., NIKOLIĆ L., ILIĆ D., RADULOVIĆ N. 2011. The effect of hydrodistillation techniques on yield, kinetics, composition and antimicrobial activity of essential oils, from flowers of *Lavandula officinalis* L. Hemijska industrija (Chemical Industry) J. Edit. Association of Chemical Engineers of Serbia. Belgrade: **65**: 455-463.
- STEVENSON A. & WAITE M. 2011. *Concise Oxford Dictionary*. Oxford University Press, Edition 100th Anniversary. 29 pp.
- UEHLEKE B., SCHAPER S., DIENEL A., SCHLAEFKE S., STANGE R. 2012. Phase II trial on the effects of Silexan in patients with neurasthenia, post-traumatic stress disorder or somatization disorder. *Phytomedicine*. Elsevier. Paris. Jun 15: **19**(8-9): 665-671. Doi: 10.1016/j.phymed. (Accessed February 3, 2018).
- VARBAN D., VARBAN R., TOFANĂ M., ROTAR A., SOCACI S., POP C., MOLDOVAN C., GHETE A. 2017. The study of composition and antibacterial activity of the volatile oil of the Sevstopolis lavender variety, cultivated in climatic conditions of Cluj-Napoca. *Hop and Medicinal Plants*. Edit. University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca: **25**: 1-2, 79-86.
- VOITKEVICI S. 1999. Лавандовые масла. *Эфирные масла для парфюмерии и ароматерапии*. (Rus.) (Lavender oils. *Essential oils for perfumery and aromatherapy*). Edit. Food Industry. Moscow: 126-130.
- WANG D., YUAN X., LIU T., LIU L., HU Y., WANG Z., ZHENG Q. 2012. Neuroprotective activity of lavender oil on transient focal cerebral ischemia in mice. *Molecules*. Pub. MDPI online. August, 15: **17**(8): 9803-9817 doi: 10.3390/molecules17089803 (Accessed February 5, 2018).
- WHEATLEY D. 2005. Medicinal plants for insomnia: a review of their pharmacology, efficacy and tolerability. *Journal Psychopharmacology*. Publisher British Association for Psychopharmacology. Oxford, **19**: 414-421. (Accessed February 3, 2018).

Gonceariuc Maria, Butnarus Violeta, Mascalovteva Svetlana, Balmus Zinaida, Cotelea Ludmila, Botnarenco Pantelimon
Institute of Genetics, Physiology and Plant Protection, Chișinău 20, Pădurii St., Chișinău, MD-2002, Republic of Moldova.
E-mail: gonceariuc.maria@gmail.com

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