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ISOLATION OF GENOMIC DNA FROM THE LEAVES OF THE DISTANT HYBRIDS OF VINE (*VITIS VINIFERA L. X MUSCADINIA ROTUNDIFOLIA* MICHX.)

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Abstract: For the isolation of the genomic deoxyribonucleic acid (DNA) from vine leaves of distant hybrids (*Vitis vinifera* L. x *Muscadinia rotundifolia* Michx.), the DNA isolation protocol based on CTAB method was used. The grouping of distant hybrids in the obtained dendrogram shows that at the DNA level there are some differences between them, differences sometimes unnoticeable at the level of ampelographic characterization. As a result, in the characterization of varieties and hybrids of vines, the ampelographic analysis needs to be supplemented by an analysis at the molecular level, based on DNA amplification techniques. As a result of distant hybrids grouping based on the size of alleles, it was found that there are two distinct main groups denoted by A and B, each having secondary branches. The hybrid F4 BC3 DRX-M4-541 1 is closely akin to the variety Chasellas dóre. Also, the two samples of *Vitis sylvestris* L. have been found to be genetically different, being placed in different subgroups. The hybrid F4 BC3 DRX-M4-536 is genetically close to the male specimen of *Vitis sylvestris*. The distant hybrid DRX-M4-660, which proved to have larger differences at the molecular level, isn't grouped in a cluster with any other hybrid.

Key words: DNA, distant hybrids, leaves, primers, alleles.

IZOLAREA ADN-ULUI GENOMIC DIN FRUNZELE HIBRIZILOR DISTANŢI DE VIŢĂ DE VIE (*VITIS VINIFERA* L. X *MUSCADINIA ROTUNDIFOLIA* MICHX.)

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Rezumat: Pentru izolarea acidului dezoxiribonucleic (ADN) genomic din frunzele de viţă de vie provenite de la hibrizi distanţi (*Vitis vinifera* L. x *Muscadinia rotundifolia* Michx.) a fost utilizat protocolul

de izolare ADN, bazat pe metoda CTAB. Modul de grupare al hibrizilor distanţi în dendrograma generată arată că la nivel de ADN există unele diferenţe între aceştia, diferenţe uneori nesesizabile la nivel de caracterizare ampelografică. Ca urmare, în caracterizarea soiurilor şi a hibrizilor de viţă de vie se impune ca analiza ampelografică să fie completată de cea la nivel molecular, prin tehnici bazate pe amplificarea ADN. În rezultatul grupării hibrizilor distanţi pe baza mărimii alelelor s-a constatat că s-au constituit două grupe principale distincte notate cu A şi B, fiecare dintre acestea având ramificaţii secundare. Hibridul F4 BC3 DRX-M4-541 este foarte înrudit cu soiul Chasellas dóre. De asemenea, cele două probe provenite de la *Vitis sylvestris* L. s-au dovedit ca fiind diferite din punct de vedere genetic, fiind plasate în subgrupuri distincte. Hibridul F4 BC3 DRX-M4-536 este apropiat genetic de Vitis sylvestris exemplarul masculin. Hibridul distant DRX-M4-660 care s-a dovedit a avea diferenţe mai mari la nivel molecular, nefiind grupat în clouster-uri cu nici un alt hibrid.

Cuvinte cheie: ADN, hibrizi distanţi, frunze, primeri, alele.

Introduction

SSR genetic fingerprinting technique can be used successfully in the determination of phylogeny relationships in the biological material analyzed. The representation of the number and size of alleles using the barcode technique gives a clear view of the molecular similarities and differences that occur between hybrids and reference varieties analyzed.

Materials and Methods

Study Material

The distant hybrids of vine (*Vitis vinifera L. x Muscadinia rotundifolia Michx.*): BC1 - DRX-55 (prob. 1); BC4 - DRX-M4-536 (prob. 2), DRX-M4-578 (prob. 3), DRX-M4-545 (prob. 4), DRX-M4-604 (prob. 5), DRX-M4-508 (prob. 6), DRX-M4-660 (prob. 7); BC2 – DRX-M3-3-1 (prob. 8); - *Vitis sylvestris Gmel.* (♀) (prob. 9); - *Vitis sylvestris Gmel.* (♂) (prob. 10); BC3 - DRX-M4-580 (prob. 11), DRX-M4-541 (prob. 12), DRX-M4-507 (prob. 13), DRX-M4-537 (prob. 14) served as study material.

Genomic DNA Extraction

For the isolation of genomic deoxyribonucleic acid (DNA) from vine leaves of distant hybrids (*Vitis vinifera* L. x *Muscadinia rotundifolia* Michx.), the specimens of *Vitis sylvestris L.* and the two international varieties taken as reference, it was used the DNA isolation protocol, based on CTAB method (the protocol of Lodhi et al., 1997, modified by Rodica Pop et al., 2003). The quantification of the quality and quantity of deoxyribonucleic acid (DNA) was performed using Nanodrop ND-1000 Spectrophotometer (Thermo Scientific).

Each sample has been subjected to three readings using Nanodrop with the aim of obtaining an average value used for the dilutions required for PCR amplification. It was used a concentration of DNA of 20 ng/μL.

PCR amplification

PCR amplification was performed in thermocycler type Palm Cycler (Corbett Research) under the conditions of touch down. The primers used were VVS2, MD5, MD7, MD27, ZAG 62 and ZAG 79, synthesized by the company IDT (USA). The selection of primers was done taking into consideration the recommendations of the gene bank European Vitis Database.

The characteristics of the used primers are shown in Table 2.

Table 2.

Characteristics of the used primers

No. crt.	*Name of the primer*	*Nucleotide sequence*	*Tm (melting temperature)*	*Type of fluorochrome for marking*
1	vvs2 forward	5'-CAGCCCGTAAATGTATCCATC-3'	53.3	5' Well Red D2
2	vvs2 reverse	5'-AAATTCAAAATTCTAATTCAACTGG-3'	48.9	-
3	MD5 forward	5'-CTAGAGCTACGCCAATCCA-3'	53.9	5' Well Red D3
4	MD 5 reverse	5'- TATACCAAAAATCATATTCCTAAA-3'	45.9	-
5	MD7 forward	5'-AGAGTTGCGGAGAACAGGAT-3'	56	5' Well Red D4
6	MD 7 reverse	5'-CGAACCTTCACACGCTTGAT-3'	55.6	
7	MD27 forward	5'- CCCCAAGGCTCTGAAAACAAT-3'	55.8	5' Well Red D4
8	MD 27 reverse	5'-ACGGGTATAGAGCAAACGGTGT-3'	58.3	-
9	ZAG 62 forward	5'- ACGGTGTGCCTCTCATTGTCATTGAC-3'	64.7	5' Well Red D4
10	ZAG 62 reverse	5'- CCATGTCTCTCCTCAGTTCTCAGT-3'	57.7	-
11	ZAG 79 forward	5'- AGATTGTGGAGGAGGGAACAAACCG-3'	60.8	5' Well Red D2
12	ZAG 79 reverse	5'- TGCCCATTTTCAAACTCCCTTCC-3'	58.0	-

Improving the amplification protocol consisted in using Touchdown PCR amplification so that the truthfulness of the final results was consistent with the specialized literature. It is worth mentioning that after the optimization of all amplification protocols, all the used primers generated amplification products, which were studied with the help of the genetic analyzer CEQ 8800™ capillary DNA analysis system (Beckman Coulter, Fullerton, CA, USA) in the next stage of experimentation, in order to determine the number of alleles and their size.

In order to identify the optimum temperature of attaching primers, there was performed a heat shock that exceeded by about five degrees Celsius the melting temperature of the forward primer, then the temperature gradually decreased with about one degree Celsius at each amplification cycle until it was reached the temperature at which primers attachment could be more specific.

The optimization of the amplification protocol is important because it helps to avoid obtaining non-specific amplification products. It was also found that the attachment optimum temperature depends on the melting temperature of the most unstable primer, from thermal point of view, of the primer pair.

In Table 3 there are presented the PCR amplification programs which were optimized and used in order to study the migration of the reaction products in the genetic analyzer.

Table 3

Amplification protocol of vine samples analyzed with the primers vvs2, MD5, MD7, MD27, ZAG 62, ZAG 79

No.	Name of the primer	PCR condition	The composition and the volume (µL) of the PCR reaction mixture	DNA quantity used / sample(µL)
1	vvs2	1. 95 ^{0}C - 0:30 min (1 cycle of amplification) 2. 95 ^{0}C – 0:30 min (1 cycle of amplification) 57 ^{0}C→51 ^{0}C – 1:00 min (by one cycle of amplification touchdown) 72 ^{0}C – 1:00 min 3. 95 ^{0}C – 0:30 min 50 ^{0}C – 1:00 min 72 ^{0}C – 1:00 min (25 cycles of amplification) 4. 72 ^{0}C – 5 min 4 ^{0}C – 99 min	$H_2$0- 4 $MgCl_2$- 1.2 dNTP mix – 0.6 Buffer – 4 Primer R – 1 Primer F – 1 Taq Pol. 0.2	3

2	MD5	1. 95 ^{0}C – 0:30 min (1 cycle of amplification) 2. 95 ^{0}C – 0:30 min (1 cycle of amplification) 58 ^{0}C→56 ^{0}C – 1:00 min (by one cycle of amplification touchdown) 72 ^{0}C – 1:00 min 3. 95 ^{0}C – 0:30 min 52 ^{0}C – 1:00 min 72 ^{0}C – 1:00 min (25 cycles of amplification) 4. 72 ^{0}C – 5 min 4 ^{0}C – 99 min	H_20- 4 $MgCl_2$- 1.2 dNTP mix – 0.6 Buffer – 4 Primer R – 1 Primer F – 1 Taq Pol. 0.2	3
3	MD7	1. 95 ^{0}C – 0:30 min (1 cycle of amplification) 2. 95 ^{0}C – 0:30 min (1 cycle of amplification) 60 ^{0}C→56 ^{0}C – 1:00 min (by one cycle of amplification touchdown) 3. 95 ^{0}C – 0:30 min 55 ^{0}C – 1:00 min 72 ^{0}C – 1:00 min (25 cycles of amplification) 4. 72 ^{0}C – 5 min 4 ^{0}C – 99 min	H_20- 4 $MgCl_2$- 1.2 dNTP mix – 0.6 Buffer – 4 Primer R – 1 Primer F – 1 Taq Pol. 0.2	3

4	MD27	1. 95 ^{0}C – 0:30 min (1 cycle of amplification) 2. 95 ^{0}C – 0:30 min (1 cycle of amplification) 59 ^{0}C→56 ^{0}C – 1:00 min (by one cycle of amplification touchdown) 72 ^{0}C – 1:00 min 3. 95 ^{0}C – 0:30 min 55 ^{0}C – 1:00 min 72 ^{0}C – 1:00 min (25 cycles of amplification) 4. 72 ^{0}C – 5 min 4 ^{0}C – 99 min	H_20- 4 $MgCl_2$- 1.2 dNTP mix – 0.6 Buffer – 4 Primer R – 1 Primer F – 1 Taq Pol. 0.2	3
5	ZAG 62	1. 95 ^{0}C – 0:30 min (1 cycle of amplification) 2. 95 ^{0}C – 0:30 min (1 cycle of amplification) 65, 64, 63, 60, 57, 55, 53^{0}C – 1 :00 min (by one cycle of amplification touchdown) 72 ^{0}C – 1:00 min 3. 95 ^{0}C – 0:30 min 55 ^{0}C – 1:00 min 72 ^{0}C – 1:00 min (25 cycles of amplification) 4. 72 ^{0}C – 5 min 4 ^{0}C – 99 min	H_20- 4 $MgCl_2$- 1.2 dNTP mix – 0.6 Buffer – 4 Primer R – 1 Primer F – 1 Taq Pol. 0.2	3

6	ZAG 79	1. 95 ^{0}C – 0 :30 min (1 cycle of amplification) 2. 95 ^{0}C – 0:30 min (1 cycle of amplification) 62 ^{0}C→56 ^{0}C – 1:00 min (by one cycle of amplification touchdown) 72 ^{0}C – 1:00 min 3. 95 ^{0}C – 0:30 min 55 ^{0}C – 1:00 min 72 ^{0}C – 1:00 min (25 cycles of amplification) 4. 72 ^{0}C – 5 min 4 ^{0}C – 99 min	$H_2$0- 4 $MgCl_2$- 1.2 dNTP mix – 0.6 Buffer – 4 Primer R – 1 Primer F – 1 Taq Pol. 0.2	3

Verification of PCR amplification products

PCR amplification products obtained after using the 6 SSR primers mentioned above were verified by migration in agarose gel 1.4% (1.4 g agarose LE Analytical Grade, Promega in 100 ml solution TAE). In Figure 1 there are shown the PCR amplification products obtained with primers pair MD5 and migrated in agarose gel and the ladder of 100 bp used.

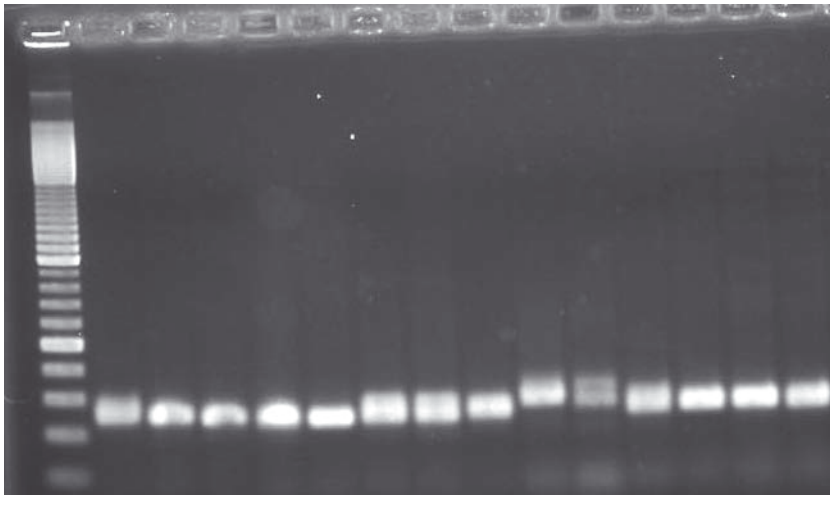


Fig. 1. The image of agarose gel with the PCR products resulting from amplification with the pair of primers MD5 and visualized with the help of the picture-taking system UPV. L-ladder Mass Ruler 100 bp (Promega)- molecular weight marker. 1-14 - the samples migrated according to the numbering from Table 1.

Samples migration in the genetic analyzer

Optimal dilutions of PCR products were obtained by probing and we found that satisfactory results concerning the migration conditions were recorded at the following dilutions:

- PCR products amplified with the primer ss2 were diluted at a ratio of 1:5 and then a volume of 1μL was used for migration;
- PCR products amplified with the primer md5 were diluted at a ratio of 1:20 and then a volume of 1μL was used for migration;
- PCR products amplified with the primer md7 were diluted at a ratio of 1:40 and then a volume of 1μL was used for migration;
- PCR products amplified with the primer md27 were diluted at a ratio of 1:40 and then a volume of 1μL was used for migration;
- PCR products amplified with the primer ZAG 62 were diluted at a ratio of 1:40 and then a volume of 1μL was used for migration;
- PCR products amplified with the primer ZAG 79 were diluted at a ratio of 1:5 and then a volume of 1μL was used for migration;

The PCR products obtained with the help of the six primers were migrated in the genetic analyzer Ceq TM 8800 (Beckman Coulter), using a volume of 0,25 μL standard 400 bp from Beckman Coulter and solution for migration -38,3 μL SLS (sample loading solution).

In order to analyze more accurately the results, there were used for comparison two international vine varieties, Sauvignon Blanc and Chasellas Dóre, whose size and number of alleles are given in the literature.

Methods of statistical interpretation of the results

The determination of the number and size of alleles at the analyzed varieties was performed automatically with the help of the software used for data interpretation included in the genetic analyzer CEQ 8800 ™ from Beckman Coulter Company. The dendrogram on the way of grouping of hybrids was done with the help of the programs PAST and FIG. TREE using the EUCLIDEAN method.

Results and Discussions

Results on DNA isolation

The amount of DNA (ng/μL) and its purity (expressed through the values of the ratio 260/280) obtained from the analyzed vines samples are shown in the images from below:

Sample ID	User ID	Date	Time	ng/ul	A260	A280	260/280	260/230	Constant	Cursor Pos.	Cursor abs.	340 raw
v1.2	Default	3/4/2013	1:26 PM	520.64	10.413	5.780	1.80	1.93	50.00	230	5.395	0.824
v1.2	Default	3/4/2013	1:26 PM	518.40	10.368	5.773	1.80	1.91	50.00	230	5.422	0.842
v1.2	Default	3/4/2013	1:26 PM	534.74	10.695	5.981	1.79	1.88	50.00	230	5.700	0.906
v2.1	Default	3/4/2013	1:28 PM	624.02	12.480	6.063	2.06	1.95	50.00	230	6.388	0.474
v2.1	Default	3/4/2013	1:28 PM	615.58	12.312	5.920	2.08	1.95	50.00	230	6.325	0.539
v2.1	Default	3/4/2013	1:28 PM	609.27	12.185	5.871	2.08	1.93	50.00	230	6.309	0.637
v3.1	Default	3/4/2013	1:29 PM	2325.09	46.502	22.112	2.10	2.08	50.00	230	22.318	0.882
v3.1	Default	3/4/2013	1:29 PM	2282.99	45.660	21.767	2.10	2.07	50.00	230	22.010	1.038
v3.1	Default	3/4/2013	1:29 PM	2241.85	44.837	21.268	2.11	2.07	50.00	230	21.649	0.932
v4.1	Default	3/4/2013	1:30 PM	2261.95	45.239	21.194	2.13	2.16	50.00	230	20.975	0.784
v4.1	Default	3/4/2013	1:30 PM	2240.66	44.813	21.002	2.13	2.15	50.00	230	20.808	0.881
v4.1	Default	3/4/2013	1:31 PM	2956.71	59.134	26.177	2.10	2.12	50.00	230	27.834	3.209
v5.1	Default	3/4/2013	1:31 PM	2142.19	42.844	20.260	2.11	2.06	50.00	230	20.802	1.020
v5.1	Default	3/4/2013	1:32 PM	2082.33	41.647	19.688	2.12	2.05	50.00	230	20.315	1.061
v5.1	Default	3/4/2013	1:32 PM	2769.71	55.394	26.483	2.09	2.04	50.00	230	27.206	1.489
v6.1	Default	3/4/2013	1:34 PM	1918.78	38.376	18.366	2.09	2.05	50.00	230	18.749	0.504
v6.1	Default	3/4/2013	1:34 PM	1892.10	37.842	18.118	2.09	2.04	50.00	230	18.513	0.534
v6.1	Default	3/4/2013	1:34 PM	1868.42	37.368	17.870	2.09	2.04	50.00	230	18.315	0.613
v7.2	Default	3/4/2013	1:35 PM	2030.90	40.618	19.420	2.09	2.01	50.00	230	20.181	0.837
v7.2	Default	3/4/2013	1:35 PM	2013.89	40.278	19.261	2.09	2.01	50.00	230	20.054	0.865
v7.2	Default	3/4/2013	1:36 PM	2179.73	43.595	20.923	2.08	2.01	50.00	230	21.673	0.967
v8.1	Default	3/4/2013	1:36 PM	1555.29	31.106	15.152	2.05	2.00	50.00	230	15.559	0.885
v8.1	Default	3/4/2013	1:37 PM	1541.12	30.822	15.003	2.05	1.99	50.00	230	15.505	0.980
v8.1	Default	3/4/2013	1:37 PM	1577.18	31.544	15.353	2.05	1.99	50.00	230	15.813	1.040
v9.1	Default	3/4/2013	1:38 PM	4532.00	90.640	46.849	1.93	1.96	50.00	230	46.170	1.143

Fig. 2. Centralizing table generated by Nanodrop on the results of DNA quantification at the analyzed vine hybrids (9 samples)

Sample ID	User ID	Date	Time	ng/ul	A260	A280	260/280	260/230	Constant	Cursor Pos.	Cursor abs.	340 raw
v6.1	Default	3/4/2013	1:34 PM	1868.42	37.368	17.870	2.09	2.04	50.00	230	18.315	0.613
v7.2	Default	3/4/2013	1:35 PM	2030.90	40.618	19.420	2.09	2.01	50.00	230	20.181	0.837
v7.2	Default	3/4/2013	1:35 PM	2013.89	40.278	19.261	2.09	2.01	50.00	230	20.054	0.865
v7.2	Default	3/4/2013	1:36 PM	2179.73	43.595	20.923	2.08	2.01	50.00	230	21.673	0.967
v8.1	Default	3/4/2013	1:36 PM	1555.29	31.106	15.152	2.05	2.00	50.00	230	15.559	0.885
v8.1	Default	3/4/2013	1:37 PM	1541.12	30.822	15.003	2.05	1.99	50.00	230	15.505	0.980
v8.1	Default	3/4/2013	1:37 PM	1577.18	31.544	15.353	2.05	1.99	50.00	230	15.813	1.040
v9.1	Default	3/4/2013	1:38 PM	4532.00	90.640	46.849	1.93	1.96	50.00	230	46.170	1.143
v9.1	Default	3/4/2013	1:38 PM	4481.90	89.638	46.307	1.94	1.96	50.00	230	45.819	1.276
v9.1	Default	3/4/2013	1:39 PM	4477.20	89.544	46.064	1.94	1.97	50.00	230	45.491	1.316
v10.2	Default	3/4/2013	1:39 PM	4659.17	93.184	49.161	1.90	1.95	50.00	230	47.892	1.574
v10.2	Default	3/4/2013	1:40 PM	4686.44	93.729	49.168	1.91	1.95	50.00	230	48.011	1.670
v10.2	Default	3/4/2013	1:40 PM	4657.66	93.153	49.039	1.90	1.95	50.00	230	47.846	1.626
v11.2	Default	3/4/2013	1:41 PM	1486.59	29.732	14.317	2.08	1.98	50.00	230	15.012	0.697
v11.2	Default	3/4/2013	1:41 PM	1477.64	29.553	14.260	2.07	1.97	50.00	230	14.964	0.741
v11.2	Default	3/4/2013	1:41 PM	1467.66	29.353	14.187	2.07	1.97	50.00	230	14.874	0.711
v12.1	Default	3/4/2013	1:42 PM	2589.12	51.782	24.903	2.08	2.09	50.00	230	24.749	0.662
v12.1	Default	3/4/2013	1:42 PM	2827.31	56.546	27.354	2.07	2.09	50.00	230	27.096	0.818
v12.1	Default	3/4/2013	1:43 PM	3139.08	62.782	30.655	2.05	2.07	50.00	230	30.266	0.843
v13.1	Default	3/4/2013	1:43 PM	3724.60	74.492	36.437	2.04	2.06	50.00	230	36.145	0.715
v13.1	Default	3/4/2013	1:44 PM	3705.13	74.103	36.205	2.05	2.06	50.00	230	36.001	0.733
v13.1	Default	3/4/2013	1:44 PM	3688.85	73.777	36.051	2.05	2.06	50.00	230	35.895	0.724
v14.1	Default	3/4/2013	1:45 PM	3604.78	72.096	34.926	2.06	2.08	50.00	230	34.658	0.725
v14.1	Default	3/4/2013	1:45 PM	3626.94	72.539	35.253	2.06	2.08	50.00	230	34.875	0.725
v14.1	Default	3/4/2013	1:45 PM	3611.00	72.220	35.037	2.06	2.08	50.00	230	34.754	0.772

Fig. 3. Centralizing table generated by Nanodrop on the results of DNA quantification at the analyzed vine hybrids (9 samples)

After quantification of the samples, DNA dilutions were made so that all the samples used for migration to have a concentration of 20 ng/µL. In Table 4 there are shown the average values of the samples of DNA, and the values of the dilution factor and the volumes of DNA stock and those of sterile double-distilled water used for samples dilution.

Table 4

Summarizing table on stock samples of DNA dilutions in order to achieve PCR amplification

Proba	cantitate ng/µL	Puritate 260/280	Suma	Media	Fdilutie	DNA	Apa
1	520,64	1,8	1573,78	524,59	26,23	3,8	96,2
	518,4						
	534,74						
2	624,02	2,08	1848,87	616,29	30,81	3,2	96,8
	615,58						
	609,27						
3	2325,09	2,1	6849,93	2283,31	114,17	0,9	99,1
	2282,99						
	2241,85						
4	2261,95	2,13	7459,32	2486,44	124,32	0,8	99,2
	2240,66						
	2956,71						
5	2142,19	2,1	6994,23	2331,41	116,57	0,9	99,1
	2082,33						
	2769,71						
6	1918,78	2,09	5679,3	1893,10	94,66	1,1	98,9
	1892,1						
	1868,42						
7	2030,9	2,09	6224,52	2074,84	103,74	1,0	99,0
	2013,89						
	2179,73						
8	1555,29	1,98	4673,59	1557,86	77,89	1,3	98,7
	1541,12						
	1577,18						
9	4532	1,94	13491,1	4497,03	224,85	0,4	99,6
	4481,9						
	4477,2						
10	4659,17	1,9	14003,27	4667,76	233,39	0,4	99,6
	4686,44						
	4657,66						
11	1486,59	2,07	4431,89	1477,30	73,86	1,4	98,6
	1477,64						
	1467,66						
12	2589,12	2,07	8555,51	2851,84	142,59	0,7	99,3
	2827,31						
	3139,08						
13	3724,6	2,05	11118,58	3706,19	185,31	0,5	99,5
	3705,13						
	3688,85						
14	3604,78	2,08	10842,72	3614,24	180,71	0,6	99,4
	3626,94						
	3611						

Genetic fingerprint of the material analyzed using SSR (Simple Sequence Repeats) technique

Experimental data on the identification of the number and size of alleles

The results obtained concerning the number and size of alleles and are shown in Table 5:

Table 5

The analyzed number and size of the obtained alleles of the local and newly created varieties (the red colour indicates the international varieties used as reference in this study)

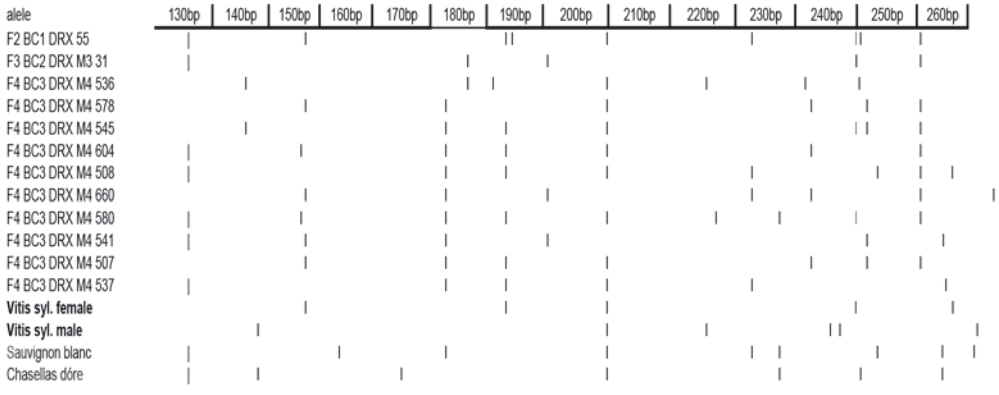
Denumirea probei	ss2		md5		md27		md7		zag 62		zag 79	
	129-155 bp		226-246 bp		173-194 bp		233-263 bp		185-203 bp		236-260 bp	
F2 BC1 DRX 55	137	149	233	239	184	190	244	244	188	204	251	261
F3 BC2 DRX M3 31	137	137	239	239	184	184	244	260	186	194	255	261
F4 BC3 DRX M4 536	139	139	239	239	190	190	226	244	186	204	241	261
F4 BC3 DRX M4 578	149	149	239	239	180	190	252	252	194	204	261	261
F4 BC3 DRX M4 545	139	139	239	239	180	190	244	252	188	204	255	261
F4 BC3 DRX M4 604	137	153	229	239	180	190	240	248	188	204	255	261
F4 BC3 DRX M4 508	137	137	233	233	180	190	248	248	188	204	261	261
F4 BC3 DRX M4 660	139	149	233	239	180	180	268	268	188	194	261	261
F4 BC3 DRX M4 580	137	153	227	237	180	190	244	244	192	204	255	261
F4 BC3 DRX M4 541	137	149	239	239	180	190	244	252	194	204	247	255
F4 BC3 DRX M4 507	149	149	239	265	180	190	252	252	188	194	261	261
F4 BC3 DRX M4 537	137	137	233	263	180	190	250	250	188	204	255	261
Vitis sylvestris female	139	149	233	233	190	206	240	240	190	204	255	255
Vitis sylvestris male	147	147	233	239	196	196	226	260	198	204	255	255
Sauvignon blanc	137	155	233	237	180	190	240	248	194	204	245	247
Chasellas dóre	137	147	229	239	176	190	240	258	194	204	251	257

The representation number and size of alleles of the varieties analyzed by DNA barcode

Data grouping was done using the program "Excel" (Table 6), establishing the time of identification of alleles' size so that it could include all the values obtained after the migration of the samples analyzed in the genetic analyzer Beckman Coulter Ceq 8800 TM.

Table 6

The representation of the number and size of alleles of the distant hybrids analyzed by DNA barcode



The migration of PCR products was performed in the Genetic Analyzer Beckman Coulter Ceq 8800 TM in order to identify the number and size of alleles of vine varieties using the SSR technique. In the figures 4 and 5 are shown some migrated samples so that the heterozygous (at the same locus, allele, Figure 4.) or homozygous state (Fig. 5, sample 2 - F3 BC2 - DRX-M3-3-1) may be highlighted.

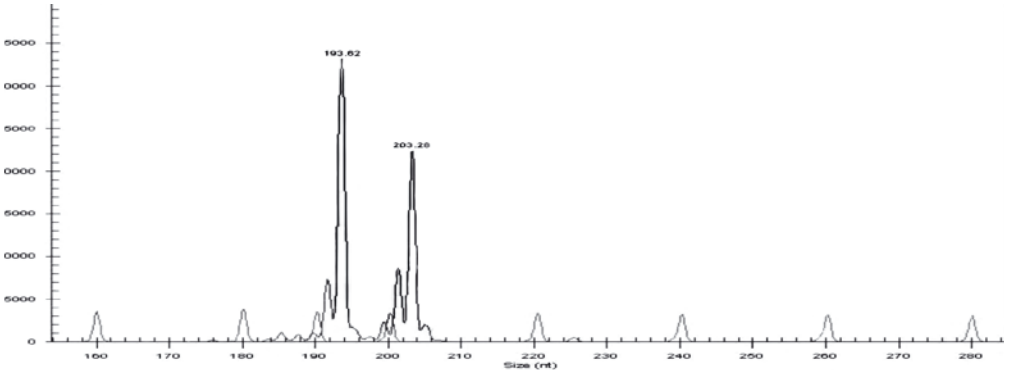


Fig. 4. Migration in the Genetic Analyzer Beckman Coulter Ceq 8800 TM in order to identify the number and size of alleles of vine varieties using the technique SSR with the primer ZAG 62.

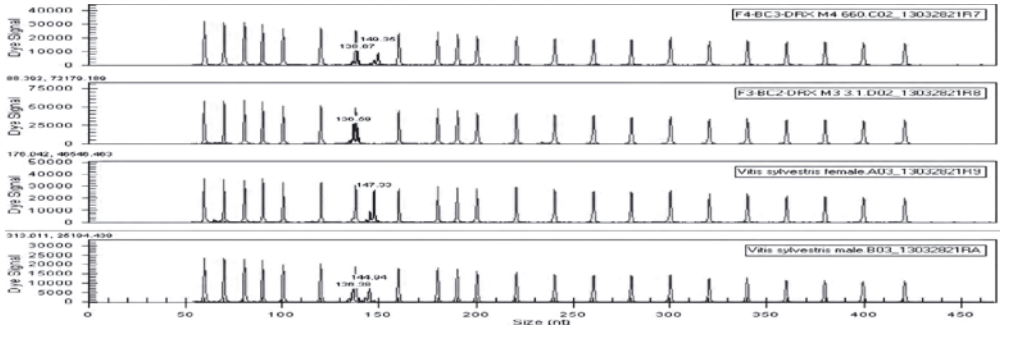


Fig. 5. Migration in the Genetic Analyzer Beckman Coulter Ceq 8800 TM in order to identify the number and size of alleles of vine varieties using the technique SSR with the primer VVS2.

The results on the way of grouping of hybrids and on the varieties used as reference

Grouping distant hybrids based on the size of the alleles identified using SSR technique was performed in order to determine their type of genetic similarity / difference (Fig. 6).

Thus, it can be seen that there were formed two different main groups denoted by A and B, each of them having some secondary ramifications. It is worth mentioning the fact that the hybrid F4 BC3 DRX-M4-541 is very akin to the variety *Chasellas dóre* and it is possible that the latter may have contributed to the formation of the hybrid mentioned above.

Also, the two samples of *Vitis sylvestris* were found to be genetically different, being placed in separate subgroups. The hybrid F4 BC3 DRX-M4-536 is genetically close to the male specimen of *Vitis sylvestris*, and it may have contributed to the formation of the hybrid.

Among the hybrids F4 BC3, DRX-M4-660 stands out, because it has proved to have more differences at the molecular level, being unable to be grouped in a cluster with any other hybrid.

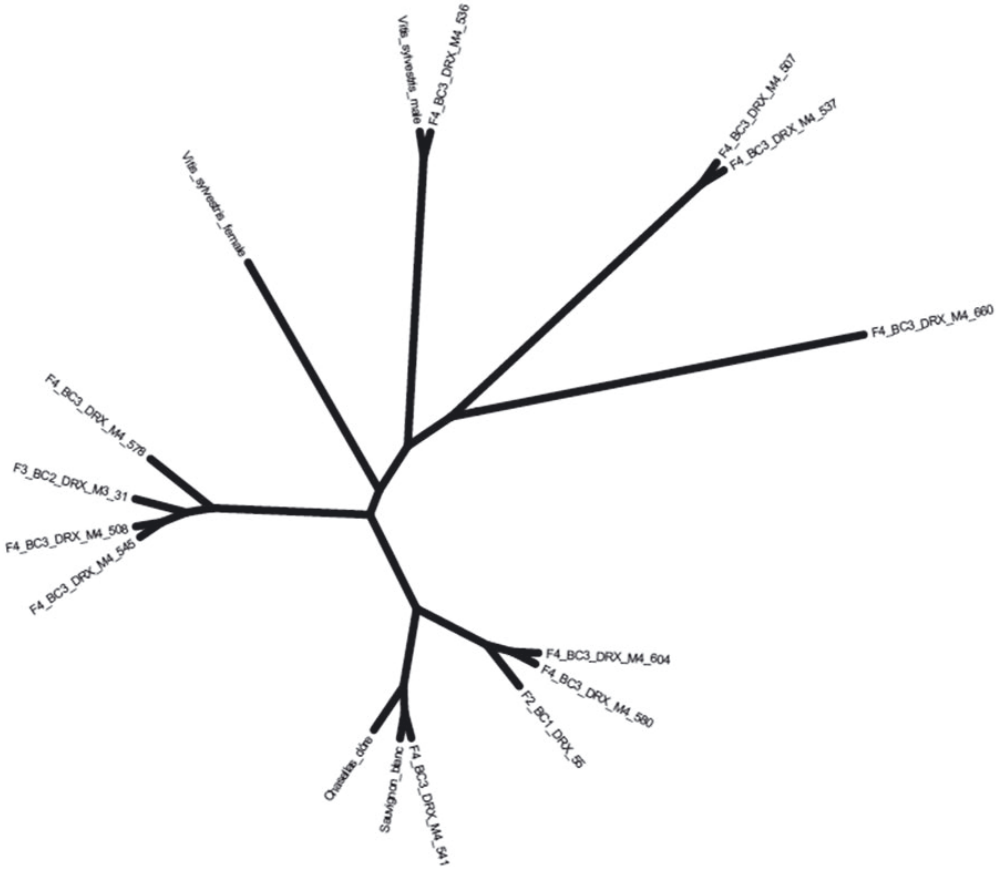


Fig. 6. The dendrogram, drawn up according to the Euclidean method, on the backcross hybrids and the reference varieties analyzed.

Conclusions

1. SSR genetic fingerprinting technique can be used successfully in the determination of phylogeny relationships in the biological material analyzed.

2. The representation of the number and size of alleles using the barcode technique gives a clear view of the molecular similarities and differences that occur between the hybrids and the reference varieties analyzed.

3. The grouping of hybrids in the generated dendrogram shows that there are some differences between them at DNA level, differences sometimes unnoticeable at the level of ampelographic characterization. As a result, the characterization of varieties and hybrids of vine requires the ampelographic analysis to be completed by an analysis at the molecular level, based on DNA amplification techniques.

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II. CONSERVATION OF BIOLOGICAL DIVERSITY

CZU: 502.72(478)+581.5:502.75(478)

THE ARGUMENTATION OF THE EXPEDIENCY OF UNDERTAKING ENVIRONMENTAL RECONSTRUCTION WORKS IN THE "CODRII" NATURAL RESERVE

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Abstract. The article summarizes an analysis on arboreta of the "Codrii" natural reserve performed on the basis of arrangements from 1997 and 2010 of the field data obtained by the itineraries, in order to bring in correspondence the arboreta structure and composition with the site type. The arboreta from 741 subplots framed in 63 plots that constitute the forestry fund of the natural reserve were analyzed. The arboreta have been divided into the categories: natural-fundamental (157 subplots), degraded (9 subplots), partial derived (172 subplots), total derived (180 subplots) and artificial with indigenous species in the area (176 subplots), with indigenous species outside the area (47 subplots). It has been concluded that the environmental reconstruction works are required to be performed over an area of 2236.2 ha in improper arboreta of the senile age (80-100 years). On the basis of the environmental restoration works, the proportion of the main species will increase up to 5% at beech, 15% at pedunculate oak, 45% at sessile oak and the productivity will increase up to 300-350 m^3/ha.

Key words: environmental reconstruction, arboreta, natural reserve.

ARGUMENTAREA OPORTUNITĂȚII EFECTUĂRII LUCRĂRILOR DE RECONSTRUCȚIE ECOLOGICĂ ÎN REZERVAȚIA NATURALĂ „CODRII"

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Rezumat. Articolul rezumă o analiză a arboreturilor din rezervaţia naturală "Codrii" în baza amenajamentelor din 1997 şi 2010 a datelor din teren obţinute pe itinerar, în vederea corespunderii structurii şi compoziţiei arboreturilor tipului de staţiune. Au fost analizate arboreturile din 741 subparcele, încadrate în 63 parcele care constituie fondul forestier al rezervaţiei. Arboreturile au fost repartizate pe categorii:

naturale fundamentale (157 subparcele), degradate (9 subparcele), parțial derivate (172 subparcele), total derivate (180 subparcele) și artificiale cu specii indigene în areal (176 subparcele), cu specii indigene în afara arealului (47 subparcele). S-a tras concluzia că lucrările de reconstrucție ecologică sunt necesare de efectuat pe o suprafață de 2236,2 ha în arboreturile necorespunzătoare de vârstă senilă (80-100 ani). În baza lucrărilor de reconstrucție ecologică se va mări proporția speciilor principale până la 5% la fag, 15% la stejar pedunculat și 45% la gorun, iar productivitatea va spori până la 300-350 m^3/ha.

Cuvinte-cheie: reconstrucție ecologică, arboreturi, rezervație naturală.

"Codrii" natural reserve, the first reserve from the Republic of Moldova, was founded in 1971 by the Council of Ministers of the MSSR from September 27, No. 310, based on the forests received from the Forest District Lozova (the southeastern part thereof) from Strășeni Forestry. The reserve has an area of 2740.0 ha and includes a strictly protected zone of 732.0 ha. "Codrii" Natural Reserve is located in the central region, the western part of the Central Moldavian Plateau, which is a core of forests. The reserve is divided into two parts by the national road Chisinau-Leuşeni. This is where the peaks which separate the three basins: North – Bâc river basin, Southwest - Cogâlnic river basin and Southwest - Botna river basin, begin.

In 1975, by the Decision of the Council of Ministers of the MSSR from January 8, No. 5, the reserve was established covering an area of 5009.0 ha, including the northwest region of Lozova Forest District. Over the years there were made changes by expanding the total area; at the moment "Codrii" Natural Reserve area is of 5170.7 ha, 5040.7 ha of which are covered with forest. Until the establishment of the reserve, the forests from the area have been managed differently, from one stage to another, especially, by performing silvicultural works, especially on the exploitation of forest resources, without paying attention to the regeneration from seeds and proper management of arboreta. Injudicious management has helped to reduce the share of the basic species in the composition of arboreta (evergreen oak, pedunculate oak, beech), weakening the ecological balance because of the repeated regeneration of trees by shoots.

In the first half of the twentieth century the arboreta were managed by coppicing method, using the regeneration of trees from shoots. This way of management, focused only on obtaining wood, led inevitably to the substitution of oaks with mixed and secondary species like ash, linden, hornbeam, and in sunny places - with cherry, maple and even locust tree. The repeated sanitation works in some areas with increased intensity without effective measures of ecological restoration also led to the worsening of the ecological balance and reduction of consistency. Cuttings practiced during the twentieth century had an anti-ecological character. Harvesting of all the arboretum at once caused damage to the environment and, therefore, led to the inadequate regeneration of the forest by the loss of its specific environment of "forest".

The arboreta reproduced from seeds constitute 13% of the area occupied by forests, 5% - from tree planting and the remaining 82% - trees reproduced from shoots. The failure to perform the tending and management of the arboreta, until 1995, led to

overwhelming the basic species and reducing their proportion in the composition of the arboreta. The young arboreta occupy 4% of the area covered by forests, and the old ones - 66%. This is because of the lack of ecological reconstruction works over a long period of time.

According to the "Law on Fund of Natural Areas Protected by State" (Government Decision No. 1538-XIII from February 25, 1998), the arboreta can be controlled through management measures such as tending and regulation of arboreta. Regeneration works (for the fundamental arboreta) and ecological reconstruction works (for inappropriate arboreta) are allowed with the purpose of regenerating the fundamental ecosystem. These management measures can be taken only with the approval of the competent fora in this field.

Within the reserve there are several types of forest sites in the phytoclimatic floor "Hilly, discontinuous, with oak and beech forests of lower limit" from the forest zone. Knowing the characteristics of each site and their importance, the following types of sites have been identified: *6155* –hilly oak woods with sessile oaks, mixed woods with sessile oaks on plateaus, sunny and partly sunny slopes with gray, gray brown + / - brown, low alluvial, middle edaphic soils (Bm): 303,1 ha (6%); *6156* – hilly oak woods with sessile oaks, mixed woods with sessile oaks on plateaus, sunny and partly sunny slopes with gray, gray brown, large edaphic soils (Bs): 2884,3 ha (57%); *6157* - hilly oak woods with sessile oaks, mixed woods with sessile oaks on plateaus, partly shady slopes with gray, gray brown, large edaphic soils (Bs): 1280,6 ha (26%); *6253* hilly oak woods with beech of lower limit, mixed woods with beech, on shady slopes with gray, gray brown, typical brown, low alluvial large edaphic soils - (Bs): 266,8 ha (5%); *6264* – hilly oak woods with pedunculate oak, meadow, hill (valley) poplar woods, on grey soils +/- gleyed, large edaphic soils (Bs): 119,4 ha (2%); *6271* hilly oak woods with pedunculate oak, mixed woods with hornbeam, on valley and on the lower third of slopes with grey, grey brown, large edaphic soils – (Bs): 186,5 ha (4%).

From the above, it appears that the most sites are of superior quality - 94% (4737.6 ha), the rest 6% are of middle quality. The largest area is (83%) occupied by the sites with sessile oak woods and mixed woods with sessile oaks. They are found throughout the whole area of the reserve. The sites with beech, at the lower limit, represent 5% of the forests. The beech, in these sites, is found in a mixture of 10% (in most areas up to 100%) on very small areas. The pedunculate oak woods and mixed woods with sessile oaks are present on about 6% of the land covered with vegetation, on valleys and on the lower third of slopes. The sites with middle quality can be found on small plateaus and on sunny upper slopes of watersheds (6%), being represented, in general, by mixed woods with sessile oaks. The identified forest types and productivity of the arboreta which fit in these types can be seen in the table 1.

Table 1

The types of forest from the "Codrii" natural reserve

No. crt.	Code		Name of the natural fundamental type of forest	Area		Average productivity of arboreta			Waste ground
	Type of site	Type of forest		ha	%	high	middle	low	
1	2	3	4	5	6	7	8	9	10
1.	6155	5323	Mixed woods with sessile oak, Pm	163,9	3	-	157,7	6,2	-
		5326	Hilly mixed woods with sessile oak, Pm	90,4	2	1,4	86,0	3,0	-
		5513	Mixed woods with pedunculate oak and sessile oak, Pm (hornbeam, cherry tree)	48,8	1	-	29,1	19,7	-
Total, type of site				**303,1**	**6**	**1,4**	**272,8**	**28,9**	**-**
2.	6156	5321	Mixed woods with sessile oak, Ps	530,7	10	223,7	273,1	33,3	0,6
		5322	Hilly mixed woods with sessile oak, Ps	1757,5	35	1129,7	600,0	27,8	-
		5512	Mixed woods with pedunculate oak and sessile oak, Ps	596,1	12	357,8	221,6	16,7	-
Total, type of site				**2884,3**	**57**	**1711,2**	**1094,7**	**77,8**	**0,6**

3.	6157	5322	Hilly mixed woods with sessile oak, Ps	1002,6	20	231,2	722,8	48,6	-
		5512	Mixed woods with pedunculate oak and sessile oak, Ps	278,0	6	80,5	190,2	7,3	-
Total, type of site				**1280,6**	**26**	**311,7**	**913,0**	**55,9**	**-**
4.	6253	4311	Beech and hornbeam woods, Ps	17,5	-	15,6	1,9	-	-
		5211	sessile oak and beech woods, Ps	89,3	2	6,8	71,8	10,7	-
		5312	Hilly mixed woods with sessile oak and beech, Ps	160,0	3	100,3	55,0	4,7	-
Total, type of site				**266,8**	**5**	**122,7**	**128,7**	**15,4**	**-**
5.	6264	6121	pedunculate oak woods of meadow from hilly region, Ps	119,4	2	71,7	34,9	12,8	-
Total, type of site				**119,4**	**2**	**71,7**	**34,9**	**12,8**	**-**
6.	6271	6211	Hilly pedunculate oak woods and mixed woods, Ps	131,0	3	58,3	36,4	36,3	-
		6212	Hilly mixed woods with pedunculate oak, Ps	55,5	1	52,2	0,5	2,8	-
Total, type of site				**186,5**	**4**	**110,5**	**36,9**	**39,1**	**-**
Total, reserve				**5040,7**	**100**	**2329,2**	**2481,0**	**229,9**	**0,6**
%				100	-	46	49	5	-

The hilly mixed woods with sessile oak - 57%, followed by the mixed woods with sessile oak - 13% and the mixed woods with pedunculate oak and sessile oak - 18% have the largest share on the reserve territory.

Characteristics of the Arboreta

The arboreta of the "Codrii" natural reserve is characterized by the following features:

- The productivity of 41% of arboreta is high, of 36% - middle and of 23% - low. The majority of the arboreta - 82% - originate from shoots, only 13% - from seeds and the remaining 5% - from planting;
- According to the type of the mixture, 5% of the arboreta of the Reserve are pure or nearly pure, the rest are mixed: on 14% of the surface, the species form a mixture of 50-80%, over 35% - form a mixture of 30-50% and on the 46% of the surface, the species form a mixture of below 30%.

Regarding the horizontal structure of the arboreta, it has been found that, in proportion to the total area covered by forests, the oak woods constitute 32%. The *sessile oak* (24%) is found in a mixture with other species in 9.6% of the area, the rest – in pure or nearly pure arboreta (4%). It originates from shoots (78%). The average density of the arboreta is of 0.79 and the average age is of 94 years, the grade of production is 2.2 with an average volume of 321 m^3/ha and a yearly average growth of 3.8 m^3/ha. The *pedunculate oak* is found in a proportion of 8%. 49% of the pedunculate oaks originate from shoots, 32% - from planting and 19% - from seeds. On 77% of the area, it grows in a mixture with other species; the rest is pure or nearly pure. Its volume is of 298 m^3 per hectare, with an average yearly growth of 3.7 m^3/ha. At an average age of 89 years, the arboreta have an average density of 0.76 and a grade of production of 2.6. At the eastern boundary of the area within the Reserve, the beech grows in proportion of 1%. It grows, generally, in a mixture with other species. It originates from seeds (93%) and shoots (7%). With an average yearly growth of 4.6 m^3/ha, it achieves an average volume of 364 m^3 per hectare at an average age of 109 years. The volume per hectare, the current growth and the productivity achieved by the sessile oak, the pedunculate oak and the beech confirms that, taking into consideration the conditions and the sites from the Reserve, these species are the most suitable to be promoted both: on the areas where they are now and in all the derived sites where they have been replaced by other, less valuable, species. The species with the largest share of the auxiliary species from the Reserve is the *ash tree* (25%). It originates from shoots (93%) and plantations (4%). The *linden*, a species which is usually found in mixtures, is found in a proportion of 13%. It mainly originates from shoots - 97%. It achieves an average volume of 288 m^3 per hectare at the average age of 86 years, with an average grade of production of 2.9. Its average density is of 0.78. The average yearly growth is of 5.2 m^3/ha.

Another species found in mixtures is the *hornbeam* (21%). At an average age of 76 years, it has an average volume of 168 m^3 per hectare; the average yearly growth is of 4.0 m^3/ha. Analyzing the vertical structure of the arboreta, it has been found that the structure of the most of them is relatively uneven aged (54%), uneven aged (11%), even aged (14%) and relatively even aged (21%).

From the data on the structure of the forestry fund protection, there were observed the following: - an improper composition with regard to the quality provided by the site - the presence of lower productivity arboreta; - unbalanced distribution on age criteria.

The regeneration of the main species in the most arboreta from shoots or suckers (49% - pedunculate oak, 78% - sessile oak) helps to maintain the same combination of biotypes, therefore, the realization of natural succession of species is inhibited [6]. In this situation we can mention not only the lower productivity of the arboreta, but first of all, the possibility and value of the functions exercised by the forest and their stability under the conditions of the climate changes [4, 8].

This requires a long process of conversion of arboreta concerning the regeneration from seeds through appropriate environmental reconstructions that restore the valuable forests appropriate to the sites.

Resulting from the general state of the forestry fund of the reserve, it has been decided to carry out an analysis of the arboreta in order to determine the appropriate structure of the arboreta for the type of the site. This is necessary to be performed in order to determine the volume of ecological reconstruction for years and to highlight the main species [3, 5]. There have been used the material of the Management plan of the "Codrii" natural reserve from the years 1985, 1997, 2010, as well as the observations and determinations on the itinerary [1, 2, 9]. The arboreta of 741 subplots framed in 63 plots, constituting the forestry fund of the reserve, have been analyzed. All forest types identified within the reserve are represented by natural-fundamental arboreta as well as by degraded, derived and artificial arboreta. It has been established that only on 25% of the area covered with forests there are natural-fundamental arboreta appropriate from the point of view of the composition and structure, about 2% of which are underproductive [7].

Several types of arboreta have been highlighted: *natural*: - fundamental - 1187.7 ha, 157 subplots; - degraded - 79.3 ha, 9 subplots; - partial derived - 1275.8 ha, 172 subplots; - total derived - 2114.1 ha, 180 subplots and *artificial* - indigenous species in the area - 339.9 ha, 176 subplots; - indigenous species outside the area - 43.9 ha, 47 subplots.

This unsatisfactory situation of the arboreta from "Codrii" natural reserve denotes the wrong direction of the forest development in the past and the high volume of ecological reconstruction which needs to be done on the background of the climatic

changes. In determining the actual volume of ecological reconstruction at the first stage, the older arboreta - 90-100 years – have been taken into account.

For each subplot, where the ecological restoration is planned, the main species and the most effective method of ecological restoration have been established.

Conclusions

1. The reconstruction of inappropriate arboreta, at the first stage, needs to be performed on an area of 2236.2 ha (degraded - 79.3 ha, total derived - 1279.7 ha, partially derived - 652.3 ha, artificial with local species - 16 1 ha, affected by drying - 208.8 ha).

2. The materials obtained allow the ecological restoration of inappropriate arboreta to be planned in time and space.

3. The proportion of the main species will increase as a result of the ecological reconstruction: at the sessile oak - from 24% to 45%, at the pedunculate oak - from 8% to 15% and the beech - from 1% to 5%, and the productivity of the arboreta will increase from 268 m^3/ha up to 300-350 m^3/ha. At the same time, the percentage of the relatively uneven aged and uneven aged arboreta will increase and will balance the arboreta by age groups, which will allow a more effective performance of the multiple functions.

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RARE PLANT SPECIES OF SEMI-DESERT STEPPES IN SOUTHERN MOLDOVA

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Abstract. The article contains the list of 37 rare species of the semi-desert steppes from the Republic of Moldova arranged according to the categories of rarity: IUCN (1994) – 35 species, Law on fund of natural areas protected by state (1998) – 20 species and Red Book of the Republic of Moldova (ed. II, 2002) – 9 species.

Keywords: rare species, categories of rarity, semi-desert steppes.

SPECIILE DE PLANTE RARE DIN STEPELE SUBDEŞERTICE DIN SUDUL MOLDOVEI

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Rezumat. Articolul prezintă lista celor 37 specii rare din stepele semidesertice din Republica Moldova după categoriile de raritate: UICN,1994 – 35 specii, Legea privind fondul ariilor naturale protejate de stat 1998 – 20 specii şi Cartea Roşie a Republicii Moldova ediţia a II-a 2002. – 9 specii.

Cuvinte-cheie: specii rare, categoriile de raritate, stepe semidesertice.

Introduction

Areas with semi-desert steppe vegetation have been preserved in the southern Moldova. They were formed on the south-west facing slopes, of the valleys of the rivers: Prut, Salcia and Ialpug (Postolache, 1994, 1995; Titica 2011). The floristic composition and the structure of the plant communities of the semi-desert steppe are distinguished from the rest of the steppe vegetation. It has been established that the composition of semi-desert steppe flora includes 279 species of vascular plants, 37 of which are rare species. According to the international classification (IUCN, 1994),

Law on fund of natural areas protected by state from 1998 and the Red Book of the Republic of Moldova the second edition in 2002, these plant species have been confirmed and included in the respective lists.

Materials and Methods

The investigation of the semi-desert steppe flora was conducted during the years 2007-2013, by the itinerary method. The surfaces with semi-desert steppe vegetation were investigated during the growing season. Over 400 exsiccatae have been collected. For the most important plant species and plant communities have been established sectors and determined geographical coordinates with the help of GPS-Garmin 300. The studied rare plant species have been assigned to the respective category of rarity according to the IUCN Red List of Threatened Species (1994), the Law on fund of natural areas protected by state (1998) and the Red Book of Moldova (the second edition, 2002).

Results and Discussions

In the semi-desert steppes of southern Moldova have been identified 37 species of vascular plants with different degrees of rarity, making up 13.26% of the total number of plant species. In accordance with the Law on fund of natural areas protected by state (1998) and the IUCN Red List of Threatened Species (1994), they have been grouped into the following categories of rarity: **critically endangered** (CR) - 4 species, **endangered** (EN) - 6 species; **vulnerable** (VU) - 5 species, **rare** (R) - 15 species, **indeterminate** (I) - 2 species, **near threatened** (Nt) - 3 species.

According to the Law on fund of natural areas protected by state (1998), the rare plant species have been assigned to the following categories: category II - 11 species, III - 1 species, IV - 7 species; VII - 1 species; VIII - 2 species. The present plants in the Red Book of the Republic of Moldova, the second edition from 2002, make up only 9 species.

Table 1

List of the rare plant species from the semi-desert steppes

The name of the species in Latin	The name of the species in Romanian	Locality	The category according to IUCN	The category of rarity*)	Rare plants included in the Red Book of the Republic of Moldova, ed. II, 2002
Bellevalia sarmatica (Georgi) Woronov	Belevalie sarmaţiană	Ciumai, Văleni, Giurgiuleşti, Slobozia Mare	CR	II	+

Delphinium fissum Waldst et Kit.	Nemțișori fisurați	Giurgiuleşti, Slobozia Mare (Săvulescu Tr. & Rayss Tcharna 1924-1934)	CR	II	+
Gymnospermium odessanum (DC) Takht.	Leontică-de-Odesa,	Giurgiuleşti,Văleni, Slobozia Mare	CR	II	+
Gypsophila glomerata Pallas ex Adams	Gipsoriță proliferă	Ciumai, Câşliţa Prut, Văleni, Slobozia Mare	CR	IV	+
Amygdalus nana L.	Migdal pitic	Vadul lui Isac, Ciumai, Giurgiuleşti,Văleni, Slobozia Mare	EN	II	-
Asparagus tenuifolius Lam.	Umbra iepurului	Văleni, Slobozia Mare	EN	II	-
Colchium triphyllum G. Kuntze	Brănduşă trifilă	Câşliţa Prut, Giurgiuleşti (Gheideman, 1982), Giurgiuleşti (Proiectul Phare Ro 2004)	EN	II	+
Helichrysum arenarium (L.) Moench.	Siminoc arenicol	Ciumai, Câşliţa Prut, Giurgiuleşti	EN	II	-
Ornithogalum amphibolum Zahar.	Celnuşă dubioasă	Câşliţa Prut, Slobozia Mare, Giurgiuleşti, Etulia (Vitko, 1976); Ciumai (Постолаке, Истрати 1991)	EN	VII	+
Sternbergia colchiciflora Waldst. et Kit.	Ghiocel-de-toamnă	Ciumai (Постолаке Истрати1991); Văleni (Gheideman & Nikolaeva 1973), Ciumai, Văleni	EN	II	-
Astragalus dasyanthus Pallas.	Zăvăcustă	Colibaşi, Ciumai, Câşliţa Prut	VU	IV	+
Adonis vernalis L.	Ruşcuţă vernală	Ciumai	VU	III	-

Asparagus officinalis L.	Sparanghel medicinal	Ciumai, Giurgiuleşti,Văleni, Slobozia Mare.	VU	II	-
****Convolvulus lineatus*** L.	Volbură liniată	Câşliţa Prut (Proiectul Phare Ro 2004), Câşliţa Prut;	VU	II	+
Ephedra distachya L.	Cârcel	Câşliţa Prut,Văleni, Slobozia Mare, Giurgiuleşti	VU	II-III	+
Achillea coarctata Poiret.	Alunele comprimate	Câşliţa Prut, Slobozia Mare, Giurgiuleşti	R	IV	-
Adonis wolgensis Stev.	Ruşcuţă volgeană	Ciumai,Giurgiuleşti,Văleni, Slobozia Mare	R	IV	-
Astragalus corniculatus Bieb.	Cosaşi corniculat	Ciumai, Giurgiuleşti,Văleni, Slobozia Mare	R	-	-
Astragalus ponticus Pall.	Cosaşi pontic	Ciumai, Giurgiuleşti,Văleni, Slobozia Mare	R	-	-
Carex supina Wahlenb.	Rogoz culcat	Ciumai, Giurgiuleşti,Văleni, Slobozia Mare	R	-	-
Centaurea trinervia Steph.	Albăstriţă trinervă	Ciumai, Giurgiuleşti,Văleni, Slobozia Mare	R	-	-
Gagea taurica Stev.	Scânteiuţă-de-Krimeia	Ciumai, Giurgiuleşti,Văleni, Slobozia Mare	R	-	-
Galium volhynicum Pobed.	Drăgaică volâniană	Ciumai, Giurgiuleşti,Văleni, Slobozia Mare	R	-	-
Haplophyllum suaveolens (DC.) G. Don. fil.	-	Ciumai, Brânza, Giurgiuleşti	R	-	-
Onobrychis gracilis Bess	Sparcetă gracilă	Ciumai, Câşliţa Prut	R	-	-
****Ornithogalum boucheanum*** (Kunth) Aschers.	Luşcă	Câşliţa Prut (Vitko 1976), Giurgiuleşti (Gheideman 1982) ; Ciumai (Постолаке, Истрати 1991)	R	VIII	-

Ornithogalum kochii Pari.	Celnuşă koch	Ciumai, Giurgiuleşti,Văleni, Slobozia Mare	R	-	-
Scorzonera mollis Bieb.	Lăptiucă moale	Ciumai, Giurgiuleşti,Văleni, Slobozia Mare	R	IV	-
Tamarix ramosissima Ledeb.	Tamarişcă rămuroasă	Ciumai, Brânza, Văleni, Câşliţa Prut,Giurgiuleşti	R	-	-
Valerianella coronata (L.) D.C.	Fetică coronată	Giurgiuleşti,Văleni, Slobozia Mare	R	-	-
Diplotaxis tenuifolia (L.) DC.	Puturoasă	Vadul lui Isac, Slobozia Mare	I	-	-
Asperula tenella Heuffel	Lipitoare delicată	Slobozia Mare, Brânza, Giurgiuleşti	I	-	-
Astragalus varius S. G.Gmel.	Cosaş variat	Ciumai, Giurgiuleşti,Văleni, Slobozia Mare	Nt	-	-
Dianthus leptopetalus Wild	Garofiţă leptopetală	Ciumai, Giurgiuleşti	Nt	-	-
Hyacinthella leucophaea (C. Koch) Schur.	Zambilă albicioasă	Câşliţa Prut, Giurgiuleşti,Văleni, Slobozia Mare	Nt	VIII	-
Goniolimon besserianum (Schult.) Kusn.	Apărătoare beser	Ciumai, Câşliţa Prut, Slobozia Mare	-	IV	-
Minuartia glomerata (Bieb.) Degen	Mierluţă glomerată	Giurgiuleşti,Văleni, Slobozia Mare	-	IV	-

Symbol from the table: + the species is present; - the species is missing; ***** the species is indicated on the basis of the researches conducted by other authors

Rare plant species

Photo 1. *Goniolimon besserianum*

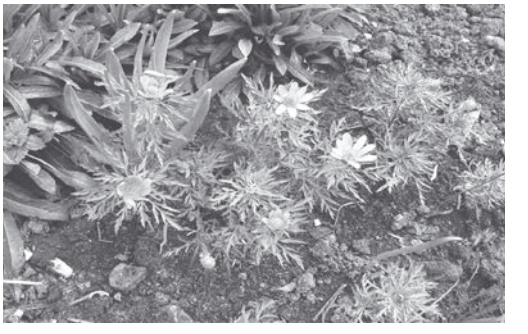

Photo 2. *Adonis wolgensis*

Photo 3. *Bellevalia sarmatica*

Photo 4. *Amygdalus nana*

Conclusions

1. 37 rare plant species have been identified in the semi-desert steppe vegetation of the southern Moldova.

2. The number of rare plant species is different:

a) According to the IUCN Red List of Threatened Species (1994) - 35 species of plants;

b) According to the Law on fund of natural areas protected by state (1998) – 20 species.

c) According to the Red Book of the Republic of Moldova, the second edition from 2002, - 9 species.

3. The plant species *Bellevalia sarmatica, Gymnospermium odessanum, Gypsophila glomerata* and *Ephedra distachya* are very rare in the flora of the Republic of Moldova.

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QUALITATIVE AND QUANTITATIVE ASPECTS OF THE PERIPHYTON ALGAE COMMUNITIES FROM THE LOWER REACH OF THE RIVER ICHEL

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Abstract: The article presents the results of floristic investigations on algal flora in the lower reaches of the river Ichel. 232 species and varieties of algae belonging to 6 phyla were found. These species belong to 91 genera, 47 families, 20 orders and 12 classes. The algal flora from the river forms different biocenosis that change during the year, forming on the surface of the submerged objects a biomass, which weighs up to 1-2 kg/m^2. Species of algae resistant to high concentrations of organic substances dissolved in water develop intensely in the river.

Keywords: periphyton, phytoplankton, algal communities, biomass, biodiversity, eutrophications.

ASPECTE CALITATIVE ŞI CANTITATIVE ALE COMUNITĂŢILOR DE ALGE PERIFITONICE DIN CURSUL INFERIOR AL RÂULUI ICHEL

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Rezumat. În articol sunt prezentate rezultatele cercetărilor floristice asupra algoflorei cursului inferior al râului Ichel. Au fost evidenţiate 232 de specii şi varietăţi de alge ce aparţin la 6 filumuri. Aceste specii aparţin la 91 de genuri, 47 de familii, 20 de ordine şi 12 clase. Algoflora râului formează diverse cenoze, care pe parcursul anului se schimbă evident, formând pe suprafaţa diferitor obiecte submerse o biomasă de până la 1-2 kg/m^2. În râu se dezvoltă intens specii de alge rezistente la concentraţii sporite de substanţe organice dizolvate în apă.

Cuvinte-cheie: perifiton, fitoplancton, comunitate algală, biomasă, biodiversitate, eutrofizare.

Introduction

The small rivers draw the attention of scientists around the world because they have become the main receivers of various wastewaters. The pollution induces considerable changes both in the hydrochemical composition of rivers, and in the structure and composition of algal communities [1].

Algae are a specific group of organisms which plays an important role in the functioning of any biocenosis. They are the basis of food chains participating in the primary production of organic substances and in the circuit of elements in nature, being involved in the processes of self-regulation, self-cleaning and self-pollution of water. The results of investigations in this field have a primary importance in highlighting new species, especially, the invasive ones, which can grow abundantly under intense water pollution with organic substances with high content of nitrogen and phosphorus contained in exaggerated concentrations in the wastewaters and sewage which flows into rivers [5].

Materials and Research Methods

Investigations on algal flora of the river Ikel were carried out during 2011-2013. The samples were collected seasonally at the following research stations: Ciorescu, Făureşti, Goian, Hruşova. A total of 60 algae samples were processed. Collecting and processing of algae samples was performed according to the unified methods of collecting and processing of field and experimental hydrobiological samples [3, 6, 14]. A part of the collected material was brought to the laboratory and analyzed recently (fresh) with the help of the microscope **Nikon YS 100**, another part was fixed in 4% formol solution. The determinants in force were used for species identification [3, 7, 8, 9, 10, 11, 12, 13, 15, 16].

The periphyton number was estimated by counting the algal cells with the help of the hemocytometer "Goreaev". The biomass of the species of filamentous - macrophytes algae is determined by direct weighing in wet state, and the biomass of microscopic algae – by the method of calculating the average cell volume of each species. The cell volume was determined by comparing them with certain geometric figures. Sometimes, we used the volumes already calculated [18]. The biomass was calculated for each species separately and then it was summed up.

Results and Discussions

As a result of the investigations on communities of periphyton algae on some higher plants (*Carex riparia* Curt., *Sparganium erectum* L., *Potamogeton pectinatus* L. etc.), wood and submerged rocks were identified 232 species and varieties of algae that belong to 6 phyla (*Cyanophyta* – 47, *Bacillariophyta* – 97, *Xanthophyta* – 2, *Dinophyta* – 5, *Chlorophyta* – 58 şi *Euglenophyta* – 23) (tab. 1).

Table 1

Taxonomic structure of the algal flora in the lower reaches of the river Ikel

Phylum	Number					
	Class	Order	Family	Genus	Species and varieties	%
Cyanophyta	2	4	13	20	47	20,2
Bacillariophyta	2	3	8	26	97	41,8
Xanthophyta	1	1	2	2	2	0,9
Dinophyta	1	2	2	3	5	2,2
Chlorophyta	5	9	19	33	58	25,0
Euglenophyta	1	1	3	7	23	9,9
Total	12	20	47	91	232	100

The periphyton algal flora of the river Ikel grows abundantly on various types of substrates, both living and inanimate, forming a biomass of 10 g/m^2 up to 1-2 kg/m^2, with the predominance of chlorophyta, cyanophyta, diatoms and euglenophyta. Algae development achieves its maximum intensity in autumn and is reduced during the cold season, when the water temperature is below 10^{o}C.

Thus, in the winter samples collected from the wood and submerged rocks, it was established an intensive development of bacillariophyta with a number of 70-80 species and varieties, the number of cells of which in 2011 amounted to about 51600 mln/m^2 and in 2012-55450 mln/m^2 with a biomass of 46.8 g/m^2 and 57.3 g/m^2, respectively (Table 2). The species: *Rhoicosphenia curvata* (Kutz.) Grun., *Cocconeis pediculus* Ehr., *Diatoma vulgare* Bory., *Melosira varians* Ag., *Cyclotela meneghiniana* Kutz., *Synedra ulna* (Nitzsch) Ehr., *Navicula rhynchocephala* Kutz., *N. viridula* Kutz., *N. cryptocephala* Kutz., *N. confervacea* Kutz., *Cymbella tumida* (Breb.) V. H., *Gomphonema olivaceum* (Lyngb.) Kutz., *G. augur* Ehr., *G. parvulum* Kutz., *G. constrictum* Ehr. *var. capitatum* (Ehr.) Cl., *Epithemia zebra* (Ehr.) Kutz., *Nitzschia dissipata* (Kutz.) Grun., *N. kuetzingiana* Hilse., *N. paleacea* Grun., *N. amphibia* Grun., *N. apiculata* (Greg.) Grun., etc. were frequent in periphyton.

In the winter samples were also highlighted some cyanophyta: *Merismopedia tenuissima* Lemm., *Oscillatoria lacustris* (Kleb.) Geitl., *O. chalybea* (Mert.) Gom., *O. terebriformis* (Ag.) Elenk., *Phormidium foveolarum* (Mont.) Gom. etc., *Lyngbya kuetzingii* (Kutz.) Schmidle, which formed a biomass up to 1.3 g/m^2.

In spring, with increasing temperatures, in the periphyton from the submerged stones and wooden objects, intensifies the development of cyanophyta from the class *Hormogoniophyceae* and especially the chlorophyta from the class *Chlorococcophyceae* and *Ulotricophyceae*: *Carteria multifilis* (Fres.) Dill, *Micractinium quadrisetum* G. S. Smith., *Pediastrum boryanum* (Turp.) Menegh., *Monoraphidium irregulare* (G. M. Smith) Kom.-Legn., *Didymocystis planctonica* Korch., *Chlorella vulgaris* Beier., *Scenedesmus quadricauda* (Turp.) Breb., *S. acutus* Meyen, *S. acuminatus* (Lagerh.)

Chod., *Stigeoclonium tenue* (Ag.) Kutz., *Oedogonium sp., Coleochaete soluta* Pringsh., etc., producing a biomass of 20-50 g/m^2. During this period, the number of diatoms decreased and constituted in 2012 (from March to May) 46300 mln/m^2 - 14350 mln/m^2 with a biomass of 42.5 g/m^2 and 16.0 g/m^2, respectively. This fact was conditioned by the increase of water temperature and the intense development of cyanophyta [17].

Table 2

Qualitative and quantitative dynamics of the periphyton algal flora on rocks and submerged wood

Data	Number of species and varieties of algae					(mln. cells/m^2) g/m^2			
	Cyanophyta	***Bacillariophyta***	***Chlorophyta***	**Other phyla**	**Total**	***Cyanophyta***	***Bacillariophyta***	***Chlorophyta***	**Total**
24.02.11	6	79	2	2	89	-	51600 46,8	-	51600 46,8
27.04.11	12	62	32	5	111	25000 1,2	7000 6,4	6550 20,5	38550 28,1
21.06.11	32	49	47	12	140	78300 5,3	8400 12,1	1200 153,9	87900 171,3
15.10.11	36	57	39	10	142	20300 2,1	27100 38,5	1800 17,3	49200 57,9
02.02.12	8	94	2	4	108	1435 1,3	55450 57,3	-	56885 58,6
19.03.12	11	71	6	6	94	1130 1,8	46300 42,5	1200 7,1	48630 51,4
01.05.12	14	66	32	10	122	87900 25,3	14350 16,0	7390 35,1	109640 76,4
24.07.12	40	51	54	25	170	41500 16,2	9460 11,3	14290 203,0	65250 230,5
19.10.12	43	64	46	21	174	31300 17,7	22950 28,4	7580 51,9	61830 98,0
Total	47	97	58	30	232				

From other phyla of algae, some euglenophyta were frequent in spring: *Euglena viridis* Ehr., *E. polymorpha* Dang., *E. oxyuris* Schmardo., *Phacus caudatus* Hübner. and others, which were met through the filaments of blue algae species of the genera *Oscillatoria* and *Phormidium*.

In summer, on the stems and submerged leaves of aquatic higher plants, clorophyta predominates in the complex: clorophyta – diatoms – cyanophyta or clorophyta – cyanophyta – diatoms with an average number of cells of 56700 mln/m^2 in 2011 and 5192 mln/m^2 in 2012 and with a biomass of 20.1 g/m^2 and 72.1 g/m^2, respectively (Table 3). On stones and submerged wooden objects, the number of cells in summer constituted 87900 mln/m^2 and 65250 mln/m^2, with a biomass of 171.3 g/m^2 and 230.5 g/m^2, respectively. Sometimes, the periphyton algae formed a biomass up to 1-2 kg/m^2. The filamentous green algae: *Cladophora glomerata* (L.) Kutz., *Rhizoclonium hieroglyphicum* (Ag.) Kutz., *Mougeotia sp., Oedogonium sp., Stigeoclonium tenue* were predominant as well as the cyanophyta: *Phormidium foveolarum*, *Anabaena variabilis* Kutz., *A. flos-aquae* (Lyngb.) Breb., *Oscillatoria terebriformis*, *O. chalybea*, *O. tenuis* Ag., *O. geminata* (Menegh.) Gom., *Gomphosphaeria lacustris* Chod., *Coelosphaerium dubium* Grun., *Plectonema terebrans* Born. et Flah., *Calothrix brevissima* G. S. West., etc. Numerous microscopic algae develop among them; most of them occur as sessile forms [4]. Thus, on the epiphytic algae of the first order inhabit epiphytic species of the second order, the most common are: *Coleochaete scutata* Breb., *Oedogonium sp., Uronema confervicolum* Lagerh., *Characium acuminatum* A. Br., *Pseudocharacium acuminatum* Korsch. from clorophyta, *Phormidium foveolarum*, *Lyngbya kuetzingii*, *Plectonema terebrans* from cyanophyta, *Achnanthes hungarica* Grun., *Amphora ovalis* Kutz., *Cocconeis pediculus*, *C. placentula* Ehr., *Cymbella cistula* (Hemp.) Grun., *Gomphonema acuminatum* Ehr., *G. constrictum var. capitatum*, *G. olivaceum*, *Nitzschia dissipata*, *Rhoicosphaenia curvata*, *Synedra ulna* from diatoms, etc.

The diatoms, as in spring, were less numerous on the inanimate substrates; their number of cells was of 8400 mln/m^2 in 2011, with a biomass of 12.1 g/m^2, and in 2012 - 9460 mln/m^2 with a biomass of 11.3 g/m^2. *Navicula cryptocephala*, *N. cryptocephala* Kutz. *var. intermedia* Grun., *N. rhynchocephala*, *Achnanthes hungarica*, *Anomoeoneis sphaerophora* (Kutz.) Pfitz., *Cymbella prostrata* (Berkeley) Cl., *C. turgida* (Greg.) Cl., *Gomphonema olivaceum*, *G. augur*, *Nitzschia amphibia*, *N. paleacea*, *Surirell ovata* Kutz. *var. crumena* (Breb.) V. N and others were the most frequent in periphyton.

In autumn, in the periphyton of the river Ikel, the intense development of cyanophyta was observed (species of the genera *Microcystis, Anabaena, Oscillatoria, Phormidium,* etc.). In this period, among the filaments of cyanophyta *Oscillatoria chalybea* and *O. Terebriformis*, some euglenophyta, such as: *Trachelomonas verruco-*

sa Stokes, *T. dubia* Swir., *Strombomonas fluviatilis* (Lemm.) Defl., *Euglena viridis*, *E. gracilis* Klebs., *E. polymorpha*, *E. oxyuris, Euglenopsis vorax* Klebs., *Phacus caudatus* etc., were developing abundantly and on the filamentous green algae – *Colacium vesiculosum* Ehr.

Table 3

Qualitative and quantitative dynamics of periphyton algal flora on aquatic higher plants
(*Carex riparia* Curt., *Sparganium erectum* L., *Potamogeton pectinatus* L.)

Data	Number of species and varieties of algae					(mln.cells/m²) g/m²			
	Cyanophyta	*Bacillariophyta*	*Chlorophyta*	Other phyla	Total	*Cyanophyta*	*Bacillariophyta*	*Chlorophyta*	Total
27.04.11	10	59	33	5	107	21000 0,4	11600 12,8	8790 17,2	41390 30,4
21.06.11	27	48	40	11	126	43600 1,3	10700 8,6	2400 10,2	56700 20,1
15.10.11	35	50	38	12	135	31400 6,6	13800 13,7	1100 7,4	46300 27,7
01.05.12	13	60	32	8	113	18600 2,4	17200 18,3	8200 22,5	44000 43,2
24.07.12	41	47	49	20	157	36300 18,5	7700 9,0	7920 44,6	51920 72,1
19.10.12	40	61	43	18	162	31500 16,4	18400 19,1	6900 28,3	56800 63,8
Total	42	95	57	24	218				

In this period, on the wooden objects and submerged rocks, the number of cells constituted 49200 mln/m² in 2011 and 61830 mln/m² in 2012, with a biomass of 57.9 g/m² and 98.0 g/m², respectively.

Cyanophyta and euglenophyta (and, sometimes, small amounts of diatoms and chlorophyta) formed, on the surface of the mud from the water, large (120-200 cm², and in some places with a slow flow - 1 to 3 m²), mucilaginous colonies of 0.2 - 0.8 cm thick and blue-green colour. Fragments of such colonies, removed by the water currents from the surface of the mud, were met among some thickets of aquatic higher plants.

From the phylum Dinophyta, 5 species were highlighted in periphyton: *Gymnodinium fuscum* (Ehr.) Stein., *Glenodinium quadridens* (Stein.) Schiller., *G. pulvisculus* Stein., *G. berolinense* (Lemm.) Lind. şi *Peridinium cinctum* (O. F. M.) Ehr, and from the phylum *Xanthophyta* – only two species: *Tribonema affine* (Kutz.) G. S. West and *Centritractus belenophorus* (W. Sch.) Lemm. These species were found in periphyton in summer, with the abundance indices: "rare" and "very rare", so, they played an insignificant role in determining the quantitative parameters of the algal flora of the river Ikel.

Conclusions

The periphyton algal flora of the river Ikel grows abundantly on various types of substrates, forming a biomass of 10 g/m^2 up to 1-2 kg/m^2. The chlorophyta, cyanophyta, diatoms and euglenophyta are predominant in the periphyton. In the river, resistant algae to high concentrations of organic substances dissolved in water develop intensively. The most widespread of them are the species of the genera: *Microcystis, Aphanizomenon, Anabaena, Oscillatoria, Cladophora, Rhizoclonium, Mougeotia, Spirogyra, Diatoma, Melosira, Navicula, Nitzschia, Euglena* etc.

So, in the river Ikel 232 species and varieties of algae belonging to 6 phyla have been highlighted (*Cyanophyta* – 47, *Bacillariophyta* – 97, *Xanthophyta* – 2, *Dinophyta* – 5, *Chlorophyta* – 58 şi *Euglenophyta* – 23).

As a result of eutrophication, in the river Ikel, euglenophyta and cyanophyta (the species of the genera *Phormidium, Anabaena, Oscillatoria, Euglena* etc.) develop intensively and cause the phenomenon called "algal bloom".

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III. INTRODUCTION OF PLANTS AND SUSTAINABLE USE OF PLANT RESOURCES

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BIOLOGICAL PECULIARITIES AND UTILIZATION POSSIBILITIES OF THE CULTIVAR "SOLAR" OF *HELIANTHUS TUBEROSUS*

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Abstract. We studied the biological peculiarities, productivity, nutritional value of the harvested fresh mass and the calorific value of the solid biofuel of the species of Jerusalem artichoke (*Helianthus tuberosus* L.). It was established that the autochthonous cultivar "Solar" at the stage of flower buttons formation achieves a productivity of 14.4 kg / m^2 of fresh mass or 3.7 kg / m^2 of dry matter, it has a moderate content of 9.32% raw protein, balanced with essential amino acids and an increased content of 58.71% nitrogen free extractive substances necessary for animal nutrition. The dry stems can be used to produce solid biofuel with a calorific value of 18.7 MJ / kg.

Key words: Jerusalem artichoke, cultivar "Solar",biological peculiarities, productivity, nutritional value, calorific value.

PARTICULARITĂȚILE BIOLOGICE ȘI POSIBILITĂȚILE DE VALORIFICARE A SOIULUI "SOLAR" *HELIANTHUS TUBEROSUS*

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Rezumat: S-au studiat particularitățile biologice, productivitatea, valoarea nutritvă a masei proaspete recoltate, precum și capacitatea calorică a biocombustibilului solid la specia de nap porcesc (*Helianthus tuberosus* L.). S-a stabilit că soiul autohton "Solar", în faza de formare a butonilor florali atinge productivitatea de 14,4 kg/ m^2 masă proaspătă sau 3,7 kg/ m^2 substanță absolut uscată, conținut moderat de 9,32% proteină brută, echilibrată cu acizi aminici esențiali și conținut majorat de 58,71 % substanţe extractive dezazotate necesare pentru nutriția animalelor. Tulpinele uscate pot fi valorificate la producerea biocombustibilului solid, capacitatea calorică 18,7 MJ/kg.

Cuvinte cheie: nap porcesc, soiul "Solar", particularități biologice, productivitate, valoare nutritvă, capacitatea calorică.

Introduction

Traditionally, for the Republic of Moldova, the agriculture continues to be the basic sector of the national economy with a share of 15.1% of the GDP, 60% of the exports and 40% of the economically active population of the country. One of the major problems of the development of agriculture is the revitalization of the animal husbandry sector, the increase of the crop production, which can provide a guaranteed flow of balanced fodder both quantitatively and qualitatively throughout the year, according to the physiological necessities of animals. In the context of the current climate changes involving stronger droughts and, in future, the aridization of the area, a phenomenon which is already becoming more noticeable in our area, moving towards a sustainable and ecological agriculture requires to increase the diversity of agricultural ecosystems by introducing and expanding of some plant species which are tolerant to adverse environmental factors (temperature and water stress) and to biotic factors, which can value the existing pedoclimatic conditions, increase the productivity and stability of agricultural production with a relatively low anthropogenic input and to ensure the improvement of the quality of life in rural areas.

One of these species is the Jerusalem artichoke, also called topinambour, *Helianthus tuberosus* L., from the family *Asteraceae*, native to North America. Europeans for the first time got acquainted with the Jerusalem artichoke in 1586, during the colonization of the region Virginia in North America. In the early seventeenth century, it is brought to France from where spreads throughout the Europe as an ornamental, food and medicinal plant. In the eighteenth century, it begins to be used in Transylvania, Moldova and Wallachia extending to the East until the Tsarist Empire where was known as "the radish of Wallachia". Today the Jerusalem artichoke is spread on all the continents and occupies an area of over 2.5 million ha. It was established that a hectare of Jerusalem artichoke can absorb from the air and use about 6 tonnes of carbon dioxide, while a hectare of forest – only 3-4 tonnes, which makes possible to provide 30 people with oxygen, has a coefficient of active photosynthetic energy utilization of over 3.5%, surpassing the maize 3 times. It makes possible to achieve the productive potential of 300 tons of fresh mass and about 230 tons of tubers [5, 10, 16, 22]. In the second half of the twentieth century, in our country, some researches on this species were initiated [14,18] and it started to be cultivated industrially, mainly for animal feed, but with the destruction of livestock farms, this plant species lost its popularity, currently being cultivated only by amateurs on private plots. In the Botanical Garden (Institute) of the ASM over the years, it has been accumulated a collection of over 60 taxa of Jerusalem artichoke with different vegetation period, plant habitus, shape and colour of tubers, which serves as material for improving, being selected the most promising forms in order to create new varieties [8].

Materials and methods

The Jerusalem artichoke plants, the autochthonous cultivar "Solar", served as object of study. The experiments were conducted on the experimental field of the Botanical Garden (Institute) of the ASM in 2009-2012. The experiences started in early spring when the soil had reached the physical maturity. The planting was performed according to the scheme 70 cm x 35cm, with medium tubers of approximately 40-50 g at a depth of 7-10 cm. Scientific studies on the growth, development and productivity of the plants were carried out according to the methodical indications [13,15], nutritional value [12,17] and calorific value [2].

Helianthus tuberosus L

Results and discussions

The "Solar" cultivar of Jerusalem artichoke, *Helianthus tuberosus* L., is created in the Botanical Garden (Institute) of the ASM by clonal selection from large populations mainly intended for animal feeding. We found out that in the period of 13-18 days after planting, the buds from the tubers developed and plantlets appeared on the soil surface. The root system development began during the same period. In the first 20 to 30 days after the appearance of plantlets, the development of the aerial part is slow; in this period, a rosette of 4-5 leaves is formed and the development of the stem begins. During this period, the root system grows more intensely and the fibrous roots can extend up to 30 cm. After 40-45 days from the beginning of the vegetation, the growth and development of the aerial part accelerates forming an

erect stem, green with shades of anthocyanin, covered with a bluish-gray waxy film, 300-450 cm tall, 2-5 cm thick, rough, porous, branched in the upper part, with 50-70 leaves. The leaves are dark green, in the lower part of the stem they are opposite and on the upper part - alternate, there are petioles with ovate leaf lamina, middle sized with a sharply toothed edge. The inflorescence is a solitary flowerhead situated on the top of the branches, with a diameter of 4-6 cm at flowering. The period of flowering starts in August. The involucral bracts are imbricate, ovate-lanceolate, sharp, rough and porous on the edges. The flowers are of two types; those at the edge of the disc are lingulate sterile with yellow petals of 3-4 cm long. The disk flowers are tubular, hermaphrodite, consisting of a yellowish white calyx and a corolla composed of ray florets with 5 yellow little teeth. The androecium consists of five stamens with united anthers and the gynoecium has an interior, unilocular ovary, and style finished with a bifid stigma. The fruit is an achene of 5-6 mm long and 1.8-2.1 mm wide, of light gray colour. The weight of 1000 seeds is 6.4 g.

On the underground part of the stem, in late May, the stolons begin to form; the length of the stolons of the cultivar "Solar" is of 10-23 cm and by thickening of their terminal part in July first tubers develop, and the period of formation and growth of tubers lasts untill the end of September. The tubers store the reserve substances and form buds for the next year. The settlement of the tubers in the soil is dispersed. The medium sized tubers weigh about 43-65 grams, they have an oval-oblong shape and the peel is thin and of a cherry color with a high content of anthocyanins and the core is white. On the tuber surface are observed the nodes of the stolon in the form of rings, on each ring there are two opposite buds. The number of rings is from 5 to 9.

In the next years, the Jerusalem artichoke plants begin their vegetation in spring when the soil temperature is about 8-10 ° C. According to the results of the scientific research conducted in 2012 it was established that, at the end of June, it developed 34-36 shoots / m^2, the plants reached 200 cm high, having developed 37-40 leaves, the fresh mass of a shoot was about 305.6 grams and 69.2 grams of dry matter, the leaf content constituted about 45%, the potential production of natural forage was of 10.7 kg / m^2. During the next decade, an accelerated pace of growth and development was observed, so the plants reached a height of 258 cm, they had formed about 50 leaves, a shoot contained about 425.4 grams of fresh mass or about 103.0 grams of dry matter and 14, 9 kg / m^2 of natural forage or 3.6 kg / m^2 of dry matter. During this period, it was found a decrease in the content of leaves of the harvested mass up to 38.2% due to the fact that 11-13 leaves from the base of the stem had dried, at the same time, the dry matter content of natural feed increased from 22.4% to 24.8%. at the stage of flower buttons formation, at the end of July, the plants grew over 290 cm tall, having developed over 60 leaves, 15 - 18 of which were already dry, a fact which influenced the reduction of leaves content in the harvested mass up to 36.1% and the fresh mass of a shoot was of only 410.7 grams, at the same time, the increase of the dry matter content

was insignificant reaching 104.9 grams / shoot of dry matter, the plant productivity was of - 14.4 kg / m^2 of fresh mass, or 3,7 kg / m^2 of dry matter. In the next period, until the end of the vegetation, it was observed a decrease in the harvested fresh mass and content of leaves, the accumulation of fresh mass being insignificant.

It is known that the animal body, in order to maintain the vital functions and give different production, needs permanently exogenous nutrients received from fodder, which, after the process of digestion and assimilation, are used by the body to ensure the following functions: plastic, energetic and biocatalytic. Based on the foregoing, it was determined the dry matter content, the chemical composition, the content of macro and micronutrients, amino acids, nutritional value and metabolizable energy of the fodder from the fresh mass of the Jerusalem artichoke harvested during the formation of flower buttons. The fresh mass of the harvested Jerusalem artichoke, the cultivar "Solar" during this period contains 25.5% of dry matter. Their chemical composition is as follows: 9.32% of raw protein, 1.93% of raw fat, 21.29% of raw cellulose, 8.75% of minerals, 58.71% of nitrogen free extractive substances.

Proteins are very important nutrients, since they are the only source of assimilable nitrogen by the body and which is determined by the content of certain amino acids that ensure biological value of the fodder. It was established that the biochemical composition of proteins of Jerusalem artichoke, the "Solar" cultivar, indicates a superior biological value, containing all the essential amino acids (mg/100 mg dry matter): 0.144 leucine, 0.082 isoleucine, 0.174 phenylalanine, 0.099 lysine, 0.016 methionine, 0.089 threonine, 0.107 valine and tryptophan (was detected but, because of technical conditions, its quantity wasn't determined) and other non-essential amino acids such as: 0.170 asparagine, 0.089 serine, 0.238 glutamine, 0.215 proline, 0.103 glycine, 0.103 alanine, 0.106 tyrosine, 0.091 histidine, 0,079 arginine.

The presence of minerals in animal nutrition is indispensable for their growth and health because they are essential components of all tissues and organs which maintain the osmotic pressure at a constant level, participate in the regulation of acid-base balance, activates a series of enzymes, moderate the neuromuscular activities, and prevent the emergence and development of diseases of animals. It was established that the content of microelements in the dry matter of Jerusalem artichoke is as follows: 16.31 g / kg potassium, 12.65 g / kg calcium, 4.10 g / kg magnesium, 2.53 g / kg phosphorus. The content of some microelements was also determined; it constitutes of 196.84 mg / kg sodium, 2.54 mg / kg copper, 19.68 mg / kg zinc, 59.9 mg / kg manganese, 155.8 mg / kg iron, 28, 4 mg / kg strontium.

In the specialized literature, it is mentioned that the chemical composition of Jerusalem artichoke changes depending on the harvest time of fresh mass, variety, soil and pedoclimatic conditions [1,4,5,6,7,]. Thus, according to the scientific results obtained by the scientists from the Ukrainian Fodder Institute [21] the determination of the nutritive value of Jerusalem artichoke green mass should be guided by the average

chemical composition (M ± m,%): 35.2 ± 4.1 % dry matter, 27.44 ± 0.45% organic matter, 5, 68 ± 0.39% raw protein, 1.36 ± 0.06% raw fat, 25.12 ± 2.45% raw cellulose, 59.58 ± 2.06% nitrogen free extractive substances, about the same data were obtained by the scientists from the National Botanical Gardens from Kiev [19]. In the agro-climatic conditions of North Ossetia Alania, Russian Federation, the "Interest" variety of Jerusalem artichoke contains 26.43% dry matter represented by: 16.59% raw protein, 5.25% raw fat, 28.44% raw cellulose, 13.54 % minerals (including 1.61% calcium and 0.38% phosphorus) and 35.68% nitrogen free extractive substances [11,20].

It is well known that the Jerusalem artichoke tubers are used in the food industry, animal feeding, and in the production of medicines and the naturally dried stems are used as fuel [3,5,9,16,22]. The conducted research has shown that after the first frosts in autumn the dehydration of the leaves and stems accelerates, so that in early December the stems are leafless and their humidity is below 25%. With the establishment of the negative temperatures, the humidity of the stems is less than 15% by the end of the year and they can be collected and processed into briquettes. It was determined that the bulk density of the crumbled stems of Jerusalem artichoke cultivar "Solar" is of 288 kg / m^3 and the density of the solid biofuel reaches 720 kg / m^3. The calorific value of the briquettes is of 18.7 MJ / kg of dry matter.

Conclusions

The local cultivar of Jerusalem artichoke – "Solar", at the stage of flower buttons formation, achieves a productivity of 14.4 kg / m^2 fresh mass or 3.7 kg / m^2 dry matter, having a moderate content of 9.32% raw protein balanced with essential amino acids and an increased content of 58.71% nitrogen free extractive substances necessary for animal nutrition. The dried stems do not require sophisticated equipment for harvesting and can be used to produce solid biofuels with a calorific value of - 18.7 MJ / kg.

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THE INFLUENCE OF SEWAGE SLUDGE FERTILIZATION ON THE AGROBIOLOGICAL PECULIARITIES OF *POLYGONUM SACHALINENSE* SPECIES IN THE REPUBLIC OF MOLDOVA

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Abstract. The aim of the experiment was to investigate the effects of fertilization with sewage sludge obtained from the municipal wastewater treatment (geotub) on the plant productivity and fodder quality of giant knotweed, also called Sakhalin knotweed, *Polygonum sachalinense* species in the Republic of Moldova. The experiment involved the following treatments: the control version without fertilizer and the sewage sludge application in a dose of 50 t/ha. It was established that sewage sludge fertilization contributes to the fulfilment of the genetic potential of giant knotweed; the forage has an increased content of protein, fats and a diminished content of cellulose. The implementation of this species contribute to solving some acute problems – the storage and the use of sewage sludge, the provision of the animals with qualitative food, the production of renewable energy and the raise of living standards in rural areas.

Key words: sewage sludge, giant knotweed, *Polygonum sachalinense*, productivity, fodder quality, renewable energy.

INFLUENŢA FERTILIZĂRII CU NĂMOL ASUPRA PARTICULARITĂŢILOR AGROBOILOGICE A SPECIEI *POLYGONUM SACHALINENSE* ÎN REPUBLICA MOLDOVA

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Rezumat. Scopul cercetărilor a fost de a investiga efectul fertilizării cu nămol, obținut în rezultatul epurării apelor reziduale municipiale în geotub, asupra productivității plantelor și calității furajului de hrișcă de Sahalin, *Polygonum sachalinense*. Experiența a cuprins următoarele variante: martor fără fertilizare și fertilizare cu nămol orășănesc în doză de 50 t/ha. S-a stabilit că aplicarea nămolului orășănesc contribue la manifestarea potențialului genetic productiv a plantelor de hrișcă de Sahalin; furajul are un conținut mai înalt de proteină și grăsimi și mai diminuat de celuloză. Implementarea acestei specii poate contribii la rezolvarea unor probleme acute – stocarea și valorificarea nămolului orășănesc, asigurarea animalelor cu furaje calitative, producerea energiei renovabile și creșterea nivelului de trai în spațiul rural.

Cuvinte cheie: nămol orășănesc, hrișcă de Sahalin, *Polygonum sachalinense*, productivitate, calități furajere, energie renovabilă.

Introduction

It is well known that for a modern, sustainable agriculture, the animal husbandry is very important, since it ensures a balance between plant growing and livestock breeding. Currently, the revitalization of the animal husbandry sector continues to be a serious problem for the Republic of Moldova because of the livestock reduction, deplorable state of pastures and hayfields, decreasing areas of traditional fodder crops, failure to ensure a continuity of fodder, unbalanced ration of vegetable protein, which acts negatively on physiological condition and productivity of animals.

An important part in solving the above-mentioned problems is played by the herbaceous perennial plant species with intensive growth which, on the one hand, can ensure animals with vegetable food, and on the other hand, can become a source for obtaining biomass for the production of renewable energy. Some scientific researches were conducted in the Botanical Garden (Institute) of the ASM over decades with the purpose of mobilization, improvement and implementation of some new, non-traditional plant species in order to use effectively the land resources to obtain food with a high level of vegetable protein [9]. A promising introduced species is the Sakhalin knotweed or the giant knotweed *Polygonum sachalinense* Fr. Schmidt (*Fallopia sachalinensis, Reynoutria sachalinensis*), a herbaceous perennial plant of the family Polygonaceae, originating in the Far East, being investigated in many scientific centres and implemented as a plant with multiple use in different regions of the globe [4,8,9,12,14, 15]. In the conditions of Moldova, it is maintained on the same field more than 15 years, it reproduces vegetatively by pieces of rhizomes and rooted cuttings (about 25 thousands / ha), which can be planted in the deeply processed soil, both in autumn and early spring. The vegetation starts with the establishment of positive temperatures in late February - early March. This plant species is distinguished due to its intensive growth and development in spring and reaching the optimal harvesting period when annual fodder plants, such as maize, Sudan grass, sorghum and sunflower are in the early stages of development. It is a good fodder for cattle, sheep and goats used fresh, as well as silage and flour with a high content of proteins and vitamins.

It is highly resistant to low temperatures in winter. Although it is affected by spring frosts, it quickly recovers and continues its growth and development. It is also tolerant to drought and heat, diseases and pests. The leaves fall suddenly from the stem with the autumn frosts and, because their decomposition process is very fast, they can help to provide the soil with nutrients. The stems remain intact until spring, serving as shelter and food for wild animals, as a means of retaining snow, and as a source for bio fuel production.

For the formation of 10 tons of fresh mass the Sakhalin knotweed extracts from the soil 40-50 kg of nitrogen, 9-10 kg of phosphorus, 50-60 kg of potassium and 19-

23 kg of calcium. For the fulfilment of the genetic potential of the Polygonum sachalinense it is necessary to find fertilization methods and to form a positive balance of humus and biofile elements in the soil.

Nowadays, because of the surging prices of mineral fertilizers and the shortage of financial resources from the agricultural sector it is possible to use, at best, the mineral fertilization of some crops (wheat, sugar beet) only with nitrogen. Taking into consideration the above-mentioned facts, the organic fertilization is the main agro-technical practice which will positively influence the level of humus in soil. Fertilization with manure is applied very rarely due to lack of necessary stocks, so in 2010 the amount of organic fertilizer used in the whole country was about 162 700 tonnes. Animals are kept in private farms and the manure is often thrown out randomly and is a source of pollution.

Unconventional organic fertilizers (sludge and compost from wastewater treatment plants) can contribute to improving the situation regarding the preservation of soil humus, because they have variable nutrient content depending on their origin. The urban sewage sludge is a feasible source of phosphorus – one of the most important nutrients for crop production. The influence of urban sewage sludge on the chemical properties of the soil, forage productivity and quality were highlighted in numerous studies [1, 5, 6, 16].

Materials and methods

The object of study was the Sakhalin knotweed, *Polygonum sachalinense* Fr. Schmidt, the "Gigant" cultivar, created the Botanical Garden (Institute) of ASM and recorded in the catalogue of varieties approved in the Republic of Moldova in 2012. The research of the qualities of urban sewage sludge from the biological wastewater treatment station from "Apă Canal Chişinău" J.S.C. was conducted by the Republican Centre of Applied Soil Science. The experiment was performed on the land; the soil humus content was of 2.0 to 2.1%, with a pH of 7.5. The sludge from the geotube with the duration of storage of four months and the dose of incorporation of 50 t / ha was used as fertilizer. The plants grown on the plots without fertilization were the control version. Scientific researches on the growth, development and productivity of plants were performed according to the methodical indications [11], biochemical composition of harvested fresh mass [13], and calorific value [2].

Results and discussions

The studies conducted by the Republican Centre of Soil Science showed that the moisture of the sludge from the geotube with the duration of storage of four months constitutes 75.5%, and the pH=7.8. The content of organic substance in the dry matter is more than 80%, the quantity of valuable nutrients for plants amounts to 2.6%

nitrogen, 1.7% phosphorus, 0.22% potassium. The nitric nitrogen constitutes 95.5 mg/100g. The concentration of heavy metals usually represents a very important limiting factor for application on an agricultural land due to the potential negative effects on the plant biomass and their transfer to food and to the entire food chain. The heavy metals in the analyzed sludge don't exceed the Admissible Concentration Limit [7] with the values for lead of 310.75 mg / kg, cadmium - 30.8 mg / kg, chromium - 315.3 mg / kg, nickel - 86.3 mg / kg, copper 117.3 mg / kg, zinc 371.3 mg / kg.

As a result of the study of biological peculiarities in the first year of vegetation, we can mention that over 20 to 25 days after planting, the Sakhalin knotweed grows and develops slowly and forms 3-5 leaves in the aerial part. During the next period, the development of the stem with internodes begins. At the end of May, the plants reach a height of 47-63 cm and have 5-7 internodes with 19-23 cm long and 8-11 cm wide leaves. In the next decade, it was observed the branching of the central stem from the 7th internodes giving rise to first-degree shoots which form smaller leaves. In early July the plants grow over 125-133cm tall having developed the central stem with 12-15 internodes, begins the appearance of second-degree shoots which branch out and form third-degree shoots over a period of 25-32 days. At the end of August the plants reach 164-170cm, the first six internodes are already wooden; in the underground part of the stem, is observed the appearance of dormant buds which next year will contribute to formation of the bush. The stem branching continues until the end of vegetation forming shoots with leaves and internodes of the fourth and fifth degree. Essential differences regarding the growth and development of Sakhalin knotweed due to urban sewage sludge fertilization weren't observed in the first year of vegetation, but it was found that the complete leaf fall happens 10-12 days later than usual, a fact which shows that the plants which grow on fertilized soil are more resistant to hoar frost in autumn. The fresh mass productivity of plants which grew on fertilized soil was 2.59 kg / m^2 or with 10.4% more than the control.

In the second year of vegetation, the bud opening of Sakhalin knotweed in the variant with fertilized soil was more even, and the stem formation began 2-3 days earlier, a fact which influenced positively the growth, development and productivity of the plants. It was found, that in April, the plants from the control version represented a bush with 3-5 shoots, and the plants from the version with fertilized soil – 5-7 shoots. It was observed that the plants from the variant with fertilized soil have longer and thicker internodes and their leaves are larger, fine with a darker coloration.

The first harvest of the fodder was performed in the second half of May when the plants were over 2 m tall, having developed 14-16 leaves. The productivity of the harvested fresh mass, Table 1, of the plants which grew on fertilized soil was 6.45 kg/m^2 and 1.46 kg/m^2 of dry matter, the content of the leaves in the fodder constitutes 48% and at plants from the version without fertilization of the soil - 4.98 kg/m^2 and 1.23 kg/m^2 of dry matter respectively, with a satisfactory content of 43% of leaves in the fodder .

The restart of vegetation and revival of Polygonum sachalinense plants after the first harvest was different. The plants which grew on soil fertilized with sludge restarted their growth and began to develop new stems after 6-8 days, and the growth and development of the plants from the control variant was uneven and delayed. So, the plants grown on the plots fertilized with sewage sludge by the end of vegetation, in mid-October have developed strains with a height up to 2.08m and in the control - strains with smaller internodes, their height being of 1.49 m. The productivity of the plants from the version grown on the plots fertilized with sewage sludge was of 4.18 kg/m^2 fresh mass or 1.26 kg/m^2 dry matter, with a content of leaves in the fodder of 45%, and in the control version - 2.36 kg/m^2 fresh matter or 0.73 kg/m^2 dry matter.

The annual productivity of Sakhalin knotweed plants grown on the plots fertilized with sewage sludge has reached 10.63 kg/m^2 fresh mass and the plants from the control - 7.34 kg/m^2.

It can be mentioned that sewage sludge fertilization influences the biochemical composition of dry matter and nutritional quality of the fodder. Thus the plants fertilized with sludge, Table 1, are characterized by an increased content of raw protein, raw fat and nitrogen free extractive substances and an essentially decreased content of raw cellulose compared to the control version. 1 kg of natural fodder of Sakhalin knotweed at the first harvest on plots fertilized with sewage sludge contains 0.22 nutritive units, 45.03 g raw protein, including 27.47 g digestible protein and 8.46 raw fat, including 4.57 g digestible fat. The productive potential of plants fertilized with sewage sludge can reach 23 t nutritive units and 2.9 t digestible proteins per hectare and at the unfertilized (control) plants reaches 16 t and 1.7 t respectively.

Table 1

Biological characteristics, productivity and biochemical composition of Sakhalin knotweed plants,
***Polygonum sachalinense*, depending on fertilization (**second year of vegetation)

Indices	Control	Sewage sludge fertilization
Plant height at the first harvest, cm	219	238
Fresh mass productivity, kg/m^2	4,98	6,45
Dry matter content,%	24,65	22,63
The content of the leaves in the fodder,%	43	48
Plant height at the second harvest, cm	149	208
Fresh mass productivity, kg/m^2	2,36	4,18
Dry matter productivity, %	33,20	30,10
The content of the leaves in the fodder, %	37	45
Annual fresh mass productivity, kg/m^2	7,34	10,63
Annual dry matter productivity, kg/m^2	2,01	2,72
Biochemical composition of the dry matter:		

raw protein, %	15.90	19,90
raw fat, %	3,03	3,74
raw cellulose, %	32,37	25,63
nitrogen free extractive substances, %	42,38	44,05
minerals, %	6,32	6,68
1 kg of natural fodder at the first mowing has:		
nutritive units	0,21	0,22
dry matter, g	246,5	226,30
raw protein, g	39,19	45,03
digestible protein, g	23,90	27,47
raw fat, g	7,47	8,46
raw cellulose, g	79,79	58,00
nitrogen free extractive substances, g	104,48	99,69
minerals, g	15,57	15,12
digestible protein, g/ nutritive unit	114	125
metabolizable energy, MJ/kg dry matter	9,00	9,32

The dried stems of Sakhalin knotweed can be harvested in winter with the technical means of harvesting fodder and used to produce solid biofuel, to make briquettes and pellets. In autumn, with the establishment of negative temperatures, we found that the complete fall of the leaves and drying rate of the stems of fertilized plants is slower. The energy capacity of the harvested biomass does not change much depending of fertilization and is about 19.3 to 19.5 MJ / kg dry matter.

Conclusions

Sewage sludge fertilization helps to fulfil the genetic potential of Sakhalin knotweed plants; the obtained fodder has an increased content of protein and digestible fat, nitrogen free extractive substances and a decreased content of raw cellulose, which has a good influence on the physiological state and productivity of animals. The biomass of this species can become an effective source of renewable energy.

The implementation of the species Polygonum sachalinense can contribute to solving the acute problems: environmental – valuation of urban sewage sludge, food security - providing qualitative and early forage for animals, energy security - producing renewable energy which would contribute to raising the living standards in rural areas.

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AROMA OF FLOWERS

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Abstract. The article describes one of the main features of ornamental plants – the ability to emanate an aroma. The reader will learn why the smell of some plants is fine while the smell of others is acrid or persistent. We know ornamental plants that have a delicate smell and are successfully used in perfumery and cosmetics industry.

Key words: smell, ornamental plants, perfumery, cosmetics, soaps, aromatic oils.

AROMATUL FLORILOR

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Rezumat. Articolul deschide una din însuşirile principale ale plantelor decorative – capacitatea de a da miros. Cititorul află din ce cauză la unele plante mirosul este fin, iar la altele tăios sau pătrunzător. Cunoaştem plantele decorative care au un miros plăcut şi sunt cu succes folosite în industria de parfumerie şi cosmetică.

Cuvinte cheie: miros, plante decorative, parfumerie, cosmetică, săpunuri, ulei eteric.

Introduction

The plant world is amazing and mysterious. The humankind has been trying for a long time to discover the secrets of the nature. People have managed to find out some secrets but still are surprised and amazed by the various shapes, colours and aromas of ornamental plants. The aromatic flowers are one of the wonders of the nature. While smelling the exceptional scent of flowers, we often think why a plant smells fine, but another - unpleasantly or irritating. But, there are ornamental plants that do not smell at all. The humankind has never been indifferent to smells. Since the ancient times, people have known that the influence of different scents is various, some bring joy, spiritual strength, and others can depress and can be dangerous for the human health.

If we answer the question why the flowers smell, then it can be said that they contain special glands which secrete essential oil, which have one or another aroma. The essential oil glands are so small that they can be seen only with a microscope. The essential oils are some liquid organic substances, which give a strong flavour of ornamental plants. The range of containing essential oil in different ornamental plants is very varied. If in the flowers of rose, the essential oil content is of some tenths of percent, in the carnations (Dianthus) it is almost 20%.

Sometimes people ask: why do the plants need essential oils or fragrance. This question can have different answers; some people consider that the smell is necessary for the plants in order to attract the insects that pollinate the flowers, while others consider that the essential oils help to control the sweat. The air blended with essential oils covers the plant and, in this way, the plant is protected from both heat and cold at night. The smell of many plants scares or chases many insects. The strong smell of sage, wormwood, peppermint, lavender etc. scares the gnats, flies, moths, etc. If we plant in the garden marigolds, then the ground around them, it is clean of diseases and pests.

The essential oils usually gather in the green parts of the plants, but before the plants bloom, they move in the flowers. But, there are, also, some paradoxes, for example: the iris contains the most of the essential oil in the rhizomes and the violets – in the leaves. There are plants, like the wild bergamot, which contain essential oils in different parts of the plant. An interesting factor is that at some plants the essential oils gather more in the flowers and, especially, in the corolla (rose, tuberose) but, sometimes, they gather in the calyx (sage, pelargonium). The smell of lavender does not change from the seed germination until the early stage of the plant development, which means that the essential oil content does not change.

The pedoclimatic conditions of the locality also influence strongly the content of essential oils. For example, the ornamental plants from the temperate zone (wild bergamot, violet etc.) secrete more essential oil when the humidity is high and the air temperature is average, but the ornamental plants from hot and dry areas (sage, lavender, marigold, etc.) secrete more essential oil when the temperatures are high and the humidity is low.

Results and Discussions

Many harmful insects can't stand the Dalmatian Pyrethrum (*Pyrethrum cinerariaefolium*) smell.

Many ornamental plants (the iris - *Iris*, the sweet basil - *Ocimum basilicum*, the Mexican marigold - *Tagetes erecta*, the French marigold - *Tagetes patula*, the tuberose - *Polianthes tuberosa*, the basket of gold - *Alyssum saxatile*, the pot marigold - *Calendula officinalis*, the Lily of the Valley - *Convallaria majalis*, the saffron crocus - *Crocus*, the carnation - *Dianthus*, the common hyacinth - *Hyacinthus orientale,* the lily - *Lilium*, the Night-scented Stock - *Matthiola*, the common mignonette - *Rezeda odorata*, the rose – *Rosa,* the sage - *Salvia muscus*, the lilac - *Syringa*, the sweet

violet - *Viola odorata*, etc..) contain essential oils and are used in the preparation of soft drinks, fruit liqueurs, filling candies and cakes etc. At the present time, there are a lot of products of perfumery and cosmetics (perfumes, colognes, creams, means of fortifying the hair, deodorants) which have various smells and the plants serve as an invaluable material for the preparation of perfumes and hygienic products. We will present a short description of the most important ornamental plants used in perfumery and cosmetics.

Rose (*Rosa*). The rose flowers have been well known due to their beauty and aroma since the ancient times. The essential oils of rose were well known by the Eastern peoples who prepared pink water and pink butter. The oil-bearing rose species are shrubby in appearance and grow up to 150 cm tall. The old branches are gray and with thorns. The oil-bearing rose grows as an industrial culture in Bulgaria, France, Turkey, Morocco, Iran and India. At the present time the largest area where oil-bearing rose species are cultivated is in Bulgaria. In Russia the first variety of oil-bearing rose was obtained in the Nikita Botanical Garden and was named "Красная роза»; this variety was grown in Crimea, Moldova and in the region Krasnodar, on an area of 5000 ha and produced 9,000 kg of oil. Recently, the varieties Miciurinca and Festivalnaia have been obtained and produce 5-6 kg of oil per hectare. Under favourable conditions a rose can yield for 15-20 years. This plant loves humidity especially during the period of budding. The rose oil is the most fragrant if the petals are harvested at 4-8 o'clock in the morning. The rose oil is widely used in perfumery, cosmetics, and production of liquors, wines, which have healing properties. It is widely used in medicine, namely against headaches, cholecystitis, conjunctivitis, burns, maladies of the oral cavity and of the respiratory tract, etc.

Lavender (*Lavandula*). The genus lavender includes 48 species, and only two species namely the common lavender (*Lavandula officinalis*) and the spike lavender (*Lavandula latifolia*), which require more heat, are used in industry. In the culture, the common lavender is more prevalent, being cultivated in France, USA, Hungary, Germany, Bulgaria etc. In Russia, the varieties Stepnaia, Record and Voznesenscaia-34 are widespread. The varieties Krîmceanca and Pannea are cultivated nowadays. On a plant, there are up to 1000 of flowers and in each flower there is a drop of essential oil. 60-100 kg per hectare can be harvested. A bush can yield fruits for 6-10 years, depending on the mode of growth. The lavender oil is used in perfumery and cosmetics, it is a component of eau de cologne, lotions, crèmes, deodorants and lipsticks, and it helps to heal burns, nerve diseases, improves the brain-work and is also used in curing the epilepsy. The lavender oil is used against sweat, for cleaning the air in sections where it destroys the microbes.

Violet (*Viola*). The genus includes about 400 species; the most commonly used varieties are Yalta, Nikitscaia and the variety "Vesna" which has been recently obtained. The flowers are of 1 cm in size and their colour can be lilac or violet. The flowers and the leaves serve as material for extracting oil despite the fact that a small

amount of essential oil is found in the rhizomes. In the western countries, the species *Viola odorata* var. *parmensis* is commonly used for oil extraction. In Moldova the variety Krîmscaia is often used; depending on the age of the plant there are obtained 72-107 quintals of leaves per 1 hectare and there are harvested up to 13 kg of oil.

Lily (*Lilium*). The lilies are native to the temperate zone of Europe, Asia and North America. They have been used in culture for a long time and have been appreciated for their decorative qualities. In the ancient Greece, Rome and Egypt, the white lily embodied the beauty, the love and the moral purity. It was used as a cosmetic plant. Nowadays, over 3000 varieties are known. The most of the varieties have a pleasant smell. The most important species which have a pleasant aroma and are used in perfumery and cosmetics are the white lily (*Lilium candidum*) the royal lily (*L.regale*) the Japanese lily (*L. Speciosum*), the violet lily (*L. fialkovaia*) which smells like the violets, the martagon lily (*L. martagon*), etc. Because not only the flowers but also the bulbs have a pleasant aroma, they are widely used in food flavouring.

Iris (*Iris*). In Greek, the word Iris means rainbow. The iris is loved by many peoples, there have been written a lot of legends and poems about it. The smell of different varieties is varied but pleasant, so, the rhizomes of iris are widely used in perfumery. In order to obtain rhizomes which smell like violets three species are used namely: the Dalmatian iris (*Iris pallida*), the German iris (*I.germanica*) and the Florentine iris (*I.florentina*). This genus which includes about 300 species and grows on all the continents of the Northern Hemisphere contains essential oils. The iris is a plant which can be used in the leather industry, the production of liqueur and in the textile industry. By grinding the dried rhizomes a large amount of starch or iris flour which can be added to the toothpaste, high quality powder, confectionary products and various medicines are obtained.

Geraniums (*Pelargonium*). One of the richest species in essential oil is the rose geranium (*Pelargonium roseum*) which grows spontaneously in South Africa on the southern, dry slopes. It is a semi-shrub of 1.5 m high with a pleasant smell, which reminds us of the smell of the rose. This species reproduces well vegetatively. 30-35 cuttings can be obtained from a plant. On an area of a hectare it is necessary to plant 20,000 cuttings. This plant prefers light, sandy, clean of weeds and fertilized soils. From a hectare, 400 quintals of leaves can be harvested and 30-35 kg of essential oil can be obtained.

Basil (*Ocimum*). The genus includes about 200 species, which are widespread in spontaneous state in Southern Asia and tropical Africa. In the temperate zone, the plants are grown in order to extract essential oils. One of the richest plants in essential oil is the African basil (Ocimum gratissimum). It is a plant of 80-100 cm tall with 10-20 first-order branches, ending with compound inflorescences. The essential oil is found more on the flower calyx and on the opposite side of the leaves. The harvest on 1 ha of irrigated land is of 60 kg where the quantity of eugenol is of 40 kg. The basil is harvested when it blooms.

Marigold (*Tagetes*). These plants are native to Central America, especially Mexico, where about 26 species grow. In floriculture, three species are spread: the Mexican marigold (*Tagetes erecta*), the French marigold (*T. patula*) and the southern cone marigold (*T.minuta* or *T.tenuifolia*). The marigolds adorn country gardens and are used in various burial ceremonies. The most essential oil is extracted from the southern cone marigolds. The essential oil is extracted in small quantities in France, India, Australia and Russia. The essential oil is extracted during the mass flowering (August-September).

Lilac (*Syringa*). There are three zones where the lilac grows naturally: Eastern Asia, the Himalayas and the Balkan - Carpathian. In Eastern Asia, about 28 species and many forms grow. In the Balkan-Carpathian zone two species are met: the common lilac and the Hungarian lilac. The ordinary lilac alone has over 1500 varieties. In the Himalayan region, at the height of 6000 meters, two species of lilac grow: the Afghan and the Himalayan lilac. The lilac is used for extracting essential oil only for some decades. The essential oil is extracted from the inflorescences. The richest in essential oil is the common lilac which contains essential oil in its inflorescences of about 8.0 cm long, simple, of pink or violet colour. Up to 500 the inflorescences grow and bloom on a bush. From one hectare of common lilac, from 3.0 up to 11.0 kg of essential oil is obtained depending on the weather conditions.

There are many ornamental plants containing essential oils which still must be studied. Such plants are: the thyme (*Thymus*), the saffron crocus (*Crocus*), the common mignonette (*Rezeda odorata*), the yarrow (*Achillea millefolium*), the elecampane (*Inula helenium*), large-flowered tickseed (*Coreopsis grandiflora*), the wild bergamot (*Monarda fistulosa*), the tuberose (*Polianthes tuberosa*) etc.

Nowadays, it is necessary to widen the assortment of ornamental plants with a pleasant aroma whose essential oils can replace the ones imported across the border and less productive than the native ones. With the development of the science and technology, the devices that allow us to solve the mysteries of studying the essential oils produced by the ornamental plants are also improving.

Conclusions

1. The climatic conditions of the Republic of Moldova are favourable for the growth of aromatic plants.

2. The aromatic plants can be used in perfumery and cosmetics.

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BIOLOGICAL MEANS FOR FIGHTING PHYLLOXERA

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Abstract: A valuable source of genes which indicate the resistance to phylloxera was created, which may be involved in hybridization as a donor of this feature, as germ plasma, or as a maternal form. The widespread use of this source of genes will make possible to create a gene pool which will help to solve successfully the global challenge of combating phylloxera.

Key words: synthesis, hybrids resistant to phylloxera based on the species *Vitis rotundifolia* Michx.

METODE BIOLOGICE DE LUPTĂ CU FILOXERA

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Rezumat: A fost creată o sursă de gene care indică rezistența la filoxeră care poate fi implicată în procesul de hibridare în calitate de donor a acestui caracter sau ca plasmă embrionară sau ca formă parentală maternă. Utilizarea pe larg a acestei surse de gene va permite crearea unui genofond, astfel, soluționând cu succes problema mondială privind lupta cu filoxera.

Cuvinte cheie: sinteză, hibrizi rezistenți la filoxeră în baza speciei Vitis rotundifolia Michx.

The palm of supremacy in the creation of the first distant hybrids of vine belongs, undoubtedly, to A. P. Wylie (1868, 1871), who was the first who revealed, among hybrid seedlings true distant hybrids, intermediate between the varieties of the species *Vitis vinifera* L. (2n = 38, the subspecies *Euvitis*) and the American spontaneous species *V.rotundifolia* Michx. (2n = 40, the subspecies *Muscadinia*). Repeated interbreeding between these two species was carried out by C. Dearing (1917), L.R. Detjen (1919a, b) and, again, true distant hybrids were obtained. Also, in the U.S.A., by backcrossing the hybrids F_1 (N.C. 6-15, N.C. 6-16) with the European and local varieties, R. T. Dunstan (1962 a, b; 1964) created the derived from BC_1 or the distant hybrids F_2, the so-called DRX: DRX-58-5; DRX-55, etc.

This completes the preliminary (or pre-synthesis) stage preceding the real synthesis of the new vine genome. It resulted in the creation of two generations (F_1 and F_2) of distant hybrids containing in the somatic cells a diploid number of chromosomes 2n = 39 (Fig. 1), that is by one complete haploid set (n = 19 and n = 20) borrowed from the parental species *V. vinifera* L. and *V. rotundifolia* Michx., respectively.

Figure 1. A bunch of the distant hybrid DRX-55 obtained from backcrossing with *Muscadinia rotundifolia* (Planch.) Small.; its berries contain viable seeds

At this stage, any researcher in the world did not notice any change, character or symbol (genotypic, phenotypic or cytological) of the hybrids that would directly or indirectly indicate the beginning of the real synthesis of a new vine genome, because they did actually not exist.

According to the researches carried out by P.Viala and L.Ravaz (1895) from more than 70 analyzed wild and cultivated species of the genus Vitis L., only at *Vitis rotundifolia* Michx., which had an extremely thick and hard wood, as at the species *Cornus max* L., was observed the highest degree of resistance to phylloxera. Simultaneously, it was shown that at *V.vinifera* L., with a fragile wood with a low density, the degree of resistance to the pest is zero. Lacking the similar results, published only 27 years later, the outstanding professor Wylie (1868) visually identified exactly what and how needs to be interbred in order to start creating distant hybrids of vine and, at the same time, to lay the material foundation of the creation of vine resistant to phylloxera. Today, thanks to the classical works of the Russian scientists, it is assumed that only by distant hybridization between the species of the genera *Vitis* L. (2n = 38) and *Muscadinia* Planch. (2n = 40) forms that are resistant to phylloxera can be created. This method, is admittedly, the only one of the existing methods of interbreeding that allows to combine in the same genotype the quantity and the quality of the crop of the varieties *V.vinifera* L. with the resistance to phylloxera of the American spontaneous variety *V.rotundifolia = Muscadinia rotundifolia* (Planch.) Small (Карпеченко, 1935; Вавилов, 1935, 1965; Цицин, 1960, 1970, 1977; Негруль,1967; Жуковский,1971).

In this connection, the outstanding Russian geneticist Nikolai Vavilov mentioned that in solving practical issues, the attention should be paid firstly to distant hybridization. At the same time, he considered that the strategy of the genetics consists in researching new forms of synthesis.

Materials and Methods

Cytological studies were conducted in 2005 on the offspring of the hybrids F_5, F_4, F_3, namely: DRX-M_5-720 (104 seedlings), DRX-M_5-722 (135 seedlings), DRX-M_5-723 (60 seedlings), DRX-M_5-724 (504 seedlings), DRX-M_5-732 (30 seedlings), DRX-M_5-735(48 seedlings), DRX-M_5-740 (139 seedlings), DRX-M_5-741 (72 seedlings), DRX-M_5-745 (4 seedlings), DRX-M_5-757 (9 seedlings), DRX-M_3-11, 14, 28, 38, 39 (134 seedlings), DRX-M_4-542, 546, 604 (60 seedlings). As a method of determining the number of chromosomes in the somatic cells of the seedlings, temporary cytological preparations coloured with propion-lacmoid according to Kaptari (1967) were used. As well as other colouring liquids, it has the necessary properties for both fixing and colouring the material. Along with this, we widely used the *acet-lacmoid* according to Darlington and La Cour (1969), as well as our modification of production of temporary preparations, the so-called "squash" (Ş. Topală, 1983).

In order to assess the new genome of vine, in 2009, the inflorescences of two distant hybrids - DRX-M_5-757 and DRX-M_5-790 were isolated with sewn parchment bags to perform interbreeding with varieties with seeds and seedless varieties. At the same time, experiments on the cross-pollination and self-pollination of inflorescences were performed. In both cases, positive results were obtained, indicating ample opportunities of involving the distant hybrids in any combination as both maternal (♀), and paternal (♂) forms.

Results and Discussions

It is considered that the species of the genus *Vitis* L. interbreed relatively easily and produce fertile offspring. Consequently, the genomes of the most species of the genus *Vitis* L. are homologous and the interbreeding within this genus can be regarded as congruent (Karpechenko, 1935). Taking into account the theory of distant hybridization (G. D. Karpechenko 1935), the interbreeding between the species belonging to the genus *Vitis* L. (2n = 38) and *Muscadinia* Planch. (2n = 40), on the contrary, succeeds with great difficulty, and their hybrids can be regarded not only as interspecific, but also as distant or intergeneric. Such hybrids are usually completely sterile because of the differences in their structure and number of chromosomes, the absence of bivalent (pairwise) conjugation of chromosomes, that is, the non-homology of the genomes of the interbreeding species. In connection with this, the interbreeding between the representatives of the genus *Vitis* L. and *Muscadinia* Planch. is, of course, incongruent.

The absolute sterility of the male gametophyte and the high or partial sterility of the female gametophyte, characteristic of the distant hybrids F_1-F_2, remained unchanged. Because of this significant defect, they had not been interesting regarding the genetic improvement of vine and, in the most cases, were abandoned. The key problem *(the high sterility of the hybrids which prevented them from being involved in the process of hybridization for boosting the immunity*) has been solved (Ş. Topală, 1983,

1988) by applying intensive backcrosses under *in-situ* conditions with the obligatory use of freshly collected pollen at the time of interbreeding or collected in the previous years and conserved in liquid nitrogen at a temperature of 195 º C. Namely these backcrosses made strictly in certain hours of the morning, in hothouse conditions of the Moldavian Research Institute of Viticulture and Winemaking, gave a new impulse, that is to say, accelerated the biochemical reactions of the real synthesis of the new genome of vine, which had begun 115 years ago in the United States.

On the basis of the distant hybrid (the vegetable mule DRX-55) with 2n = 39, in 1982, in in-situ conditions, it was performed the first backcrossing with the interspecific hybrid *Aramon x V.riparia*. In this way, the first 32 local distant hybrids were created in Moldova. To the distant hybrids or "vegetable mules" with an unpaired number of chromosomes (2n = 39), except for DRX-55, also belong others are hybrids, containing sterile pollen in their anthers, which consist of some sterile pollen grains, half the size of normal grains. First of all, belong to this category: DRX-58-5; DRX-M_3-32; DRX-M_3-2 and many other derivatives from interbreeding with the wild species *Muscadinia rotundifolia* (Planch.) Small.

In 1984, 15 different combinations of backcrossing of DRX-55 with the complex French hybrids of the type Seyve Villard, original species, polyploid forms, different varieties of *Vitis vinifera* L. and 3 clones of *Muscadinia rotundifolia* (Planch.) Small (Fig. 1) were performed. As a result of the performed backcrosses, 412 local distant hybrids were created which constitute, conditionally, the F3 generation or the hybrid population BC_2. Therefore, in Moldova, by complex interbreeding and backcrosses, 444 distant hybrids involving *Vitis rotundifolia* Michx., have been created; they are marked as DRX-M-, where the letter M indicates the place of their creation (Moldova).

For the first time, the phenomenon of synthesis at the partially fructiferous hybrids F3 was discovered in 1987 (that is after 119 years after the first interbreeding). The appearance of pollen grains with a normal size and shape, such as those characteristic of the standard European varieties: Aligoté, Cabernet, Chasselas, etc. was observed at these hybrids. So, the similarity of the sterile pollen grains of the hybrids: F_3 – DRX-M_3-205 and F_2 - DRX-55, DRX-58-5 was clearly visible. At the same time, it was observed the similarity of the size and shape of the pollen grains of partially fructiferous distant hybrids, natural Linnaean species and synthetic species of vine, which is a cytological evidence of when and where the synthesis of the new genome began, how the synthesis took place and under what conditions and under the interaction of what factors this process was completed. Another type of sterility of pollen grains was observed at the distant hybrid NC 6-15. Its sterile pollen grains acquire a geometric shape of a circle, which is met again at the distant hybrids F_3 - DRX-M_3-32; DRX-M_3-40, etc.

This rare phenomenon indicated the occurrence of the process of regulation of the meiotic division in male and female gametophytes. At the same time it indicated the beginning of the normalization of meiosis in the male and female gametophytes,

which inevitably lead to the complete restoration of the fruitfulness (fertility) of the distant hybrids of vine. But the most reliable, accurate and incontestable cytological proof of the completion of the synthesis of the new vine genome is, undoubtedly, *the direct count of the number of chromosomes*. Making countless cytological preparations, which allowed monitoring of the synthesis in meiosis and mitosis, their careful analysis and the direct count of the number of chromosomes on more than 20 metaphase plates incontestably proved that *all the distant hybrids F5 contain 38 chromosomes in their somatic set*. The comparison of these hybrids with the sterile distant hybrids of vine of the American selection, (the vegetable mules, DRX-55 and DRX-58-5 with 2n = 39), proved another very important cytological fact. Namely, that the 39th unpaired chromosome, named the *tricky* chromosome (or *intriguer*, because it had caused all the perturbations in the meiosis, numerous disturbances of the normal course of the meiosis which lead to the almost absolute sterility) of the distant hybrids, was displaced irreversibly from the nucleus (karyotype) into the cytoplasm.

So, the synthesis of a new vine genome was originally performed in *in-situ* conditions in Moldavian Research Institute of Viticulture and Winemaking and in *ex-situ* conditions – in the Botanical Gardens (Institute) of ASM and in Moldavian Research Institute of Viticulture and Winemaking.

The presence in pollen sacs of normal pollen grains together with the sterile ones proves that in the given place and moment started and went on, in fact, *the final stage of the synthesis of the new genome*, including, under *ex-situ* conditions under the influence of internal and external factors. In 1987, the synthesis of the new genome of vine of the F3 hybrids, which had started to fructify, was greatly increased by performing, under ex-situ conditions, of two backcrossing combinations of freshly collected pollen from the hybrids Seyve Villard: DRX-M_3-90 x S.V. 20- 366 и DRX-M_3-232 x S.V.12-309.

Thus, as a result of these two very successful backcrossings, *a new population of distant hybrids was created*; with more than 200 plants, it constituted, conventionally, the generation F_4, or the offspring BC_3. After a limited number of backcrosses of F4 hybrids, as female plants, (♀) with a variety of European vines, as male plants, (♂) 80 plants were created (conditionally – the generation (F_5) or the offspring BC_4). In addition to these hybrids, here were included the seedlings obtained from seeds from cross-pollination of flowers, mainly forms whose fruitfulness (fertility) had been fully restored. The generation of mixed distant hybrids F5 was moved, in 2001, to the experimental plot of the Moldavian Research Institute of Viticulture and Winemaking on the natural background of contamination with the root form of phylloxera. In the first years of vegetation, phylloxera destroyed almost a half of distant hybrids F5. In the following years the death of hybrids because of the phylloxera continued, but not with the same intensity. In 2011 (after 10 years) almost half of the initial number

of planted hybrids with various degrees of resistance to the root form of phylloxera remained. The distant hybrids of the given generation blossomed in 2003 and, judging by the normal shape and size of their pollen grains, similar to the pollen of the bisexual standard varieties of vine, it can be concluded that their meiosis is correct, that is, the reduction division occurs without any disturbances and deviations from the norm, and the fertility of the hybrids F_5 is fully restored.

The experimental data is a conclusive evidence of the completion of the synthesis of a new vine genome of the hybrids F_5 under natural conditions and of the gradual disappearance of the complex biochemical reactions in the nuclei of the maternal cells of the pollen and ovules in the embryo sac under the interaction of internal and external factors.

Thus, for the first time in the world, the synthesis of a new genome of vine (n = 19) consisting of the chromosomes of the genome of the European vine species – *Vitis vinifera* L. and the chromosomes of the genome of the spontaneous species *Vitis rotundifolia* Michx. (n = 20). The value of the American native species is the absolute immunity to diseases and pests, including the phylloxera. Under the influence of backcrosses, which had been planned in advance and executed at a precisely fixed time in the morning, with freshly collected pollen happened the following: firstly, the process of creation of the synthetic species intensified (Zawadzki, 1968), and secondly, forced the displacement in the cytoplasm of the 39th unpaired chromosome (the so called tricky chromosome). It was the cause of all disturbances in the meiosis of both gametophytes - male and female, that is the complete sterility of pollen and the zero fertility of the hybrids F_1-F_2 and even more than ½ of F3, this is a fact which was proved for the first time in the world by our cytological investigations and direct counting of chromosomes in the metaphase plates (2n = 38 instead of 2n =39, determined for DRX- 55 and DRX-58-5).

By the way, R.T. Dunstan (1962), the American author of distant hybrids in the *"Some fertile hybrids of bunch and Muscadine grapes"* mentions mistakenly for DRX-55 and DRX-58-5 2n = 38. The author writes: *"Cytological analysis of four seedlings of N.C. 6-15 x Euvitis reveals the chromosome number of each (DRX-55, DRX 58-5, DRX 59-3, DRX 59-5)* to be 2n=38.

The first of these four, designated DRX-55 resulted from open-pollinated of N.C.6-15, being the one surviving plant from nine seeds produced in 1954. The vine is intermediate in certain characteristics, although some-what more Euvitis appearance. Although its flowers appear morphologically perfect its pollen is entirely sterile". Despite the high scientific authority of the American cytologists, Ș. G. Topală and N. I. Guzun (1980) emphasize once again: it was found that the somatic set of these hybrids contains a different number of chromosomes – 2n = 39.

Our cytological investigations of the meiosis of the distant hybrids F_2 - F_5 showed that the mechanism of displacement of the unpaired chromosome consisted in the implementation of intensive backcrosses. The latter were performed with freshly collected pollen, firstly under *in-situ* and then under *ex-situ* conditions, a fact which delayed the movement of the unpaired chromosome during the anaphase from the equator on the spindle fibres to one of the poles of the cell, forcing, in this way, its secretion into the cytoplasm. At the same time, it was the cause of the infertility and the inability to form under *ex-situ* and *in-situ* conditions of viable seeds in the hybrids F_1, F_2, as well as more than half of the distant hybrids F_3 and some of the hybrids F_4.

The hybrids with an unpaired number of chromosomes in their somatic cells are sterile, do not produce fruits, seeds and are named according to Goryunov (1960) *vegetable mules*, by analogy with the naturally appeared about 4,000 years ago and existed on the planet Earth sterile mules, i.e. *a hybrid between a horse and a donkey*. By the way, these hybrids were mentioned for the first time in the world in the Bible (see 1 Esd.: 2,66: *«Коней у них семьсот тридцать шесть, лошаков у них двести сорок пять»,* (c.442) or Ezr. *"Their horses were seven hundred and thirty six, their mules two hundred and forty-five"*, (p.406) or Esd. "*Leurs chevaux: 736; leurs mulets***:** 245", (p.1070) or Ezr. *„Aveau şapte sute treizeci şi şase de cai, două sute patruzeci şi cinci de catâri"(p.489)*.

It should be emphasized that the mules, throughout this long period, did not have offspring and did not reproduce themselves, because they are almost completely sterile, and yet their population in the world currently is more than 3.4 million individuals.

But the problem of sterility or infertility of these biological objects still drew the attention of people who could not discover the cause of this mysterious phenomenon. It was found that the real cause of the sterility of the mule (Ş. Topală, 2008), zebroid, vegetable mule is in the unpaired chromosomes (63, 53, 39) and mismatch, incompatibility or discrepancy between the chromosomes of the parental forms, in general, because of their different internal structure, leading to all kinds of disturbances. These irregularities have occurred because of absence in 1st meiotic prophase of the paired chromosomes conjugation, as was pointed out by Thompson (1940): "*Similarly in hybrids between the horse and zebra Drozophyla melanogaster and D.simulans sterility is due to premeiotic disturbances"* that is perturbations in the I prophase of the meiosis.

Although the sterility of the reproductive organs of mules was marked for millennia (4000 years), its true reason remained unknown and was revealed by Thompson (1940) and confirmed by us (Topală, 1983.1987) by distributing the data on the sterility of pollen and ovules from the vegetable mule to the real mule (Topală, 2008) that is by extrapolating data concerning the phenomenon of sterility between these different organisms of different genera, long before the establishment of the chromosome number in the genus Equus L. (Graphodatsky, Radjabli, 1988).

Figure 2. Bunches of the distant hybrids of vine F_5: DRX-M_5-723 (left); DRX-M_5-790 (right) or synthetic species - *Vitis vinifolia* and *V.cruceştiana*, respectively.

The synthesis of the new genome of vine (n = 19, 2n = 38) took a period of time equal to the XIX-XXI centuries (a relatively insignificant period). Today, we can say that the synthesis is apparently definitive, comprehensive and irreversible (Topală, Dadu, 2006, 2009; Topală, 2008, 2011). Thus, during the synthesis of the new genome of vine, it was recreated the new haploid set of chromosomes with localized genes in it (Rieger, Michaelis, 1967) - n = 19 from the chromosomes belonging to two species of the genus *Vitis* L.: the cultivated *V.vinifera* L. (n = 19) and the wild *V.rotundifolia* Michx. (n = 20). In the process of synthesis, the main stages of its course were outlined, namely:

firstly, the preliminary synthesis, carried out by the American scientists (Wylie, 1868, 1871; Dearing, 1917; Detjen, 1919a, b), which originates from the creation of the distant hybrids F1 by interbreeding *V.vinifera* L. x *V.rotundifolia* Michx.; this stage is completed by the creation of DRX-s (R. Dunstan, 1962, 1964a, b): DRX-58-5, DRX-55, DRX-60-24, etc., which contain in the somatic cells a diploid number of chromosomes equal to 2n = 39;

secondly, the actual synthesis of the new vine genome: it began by backcrossing *in-situ*: DRX-55 x (*Aramon x V.riparia*), then continued by backcrossing with the parental species, the French hybrids of the type Seyve Villard, polyploid forms and different varieties and clones of *Vitis vinifera* L. As a result, a new population of autochthonous distant hybrids (F3) was created, numbering 444 unique forms.

The process of synthesis of the new genome, along with the process of creation of the synthetic species (Fig. 2, 4, Zawadzki, 1968; Topală, 2008b) intensified under ex-situ conditions as a result of two backcrosses: DRX-M_4-90 x S.V. 20-366 and DRX-M_4-232 x S.V. 12-309. Thus, the 4th generation (F_4) was conditionally created, including a more than 200 autochthonous distant hybrids;

thirdly, on the basis of the hybrids F_4, about 80 hybrid plants - F_5 - were created, as a result of interbreeding for special purposes: (28 seedlings of *DRX-M_4-510 x Moldova*, 11 seedlings of *DRX-M_4-520 x GM 325-58*, 28 seedlings of *DRX-M_4-520* x Cristal), and seedlings grown from seeds obtained from cross-pollination of flowers and collected exclusively from the hybrids with completely restored fertility;

fourthly, the real formation, establishment and improvement of a new genome according to the ever-changing environmental conditions and modern techniques of cultivation of vine;

Figure 3. Bunches of the distant hybrids DRX-M5-724 (left) and DRX-M5-734 (right) or Vitis vinifolia

Figure 4. Bunches of distant hybrids of vine F_5: DRX-M_5-757 (left) and the DRX-M_4-631 (right), or the synthetic types *Vitis rotundifera* and *V.nigra,* respectively

fifthly, in the fifth generation (F_5) under ex-situ conditions, the most significant work was done in the long-lasting cycle of synthesis of a new genome and the creation of synthetic species of vine. Here, in the first place, was completed and gradually stopped the process of creation of synthetic species of vine (Zawadzki, 1968) under the influence of internal and external factors. At the given stage, the synthesis of a new genome of vine was completed (Topală, 2008 a, b; Topală, Dadu, M. Istrati 2005; Topală, Dadu, 2006; 2009) by a final and irreversible displacement into the cytoplasm of the unpaired 39th chromosome. Detailed cytological studies of the synthetic species and their seed progeny made by us on living and fixed material in mitosis (rootlets, points where sprouts grow) and meiosis (anthers) definitely proved that all of them contain in their somatic set a true diploid (2n = 38) and, respectively, haploid number of chromosomes equal to (n = 19). For the first time in the world these cytological data were obtained by direct counting of chromosomes in more than 20 ideal metaphase plates in which each chromosome is numbered. Besides, the Botanical Garden of ASM and the Moldavian Research Institute of Viticulture and Winemaking have a sufficient number of distant hybrids with 2n = 39 and distant hybrids with new the genome of vine - 2n = 38 in order to give the cytologists from any country the opportunity to test the validity of these scientific facts.

Of course, the distant hybrids created by us, both: with a somatic chromosome number of 2n = 39, and with the chromosome number 2n = 38 (i.e. with the new genome of vine) definitely needs a more profound study with the help of genetic, biochemical and molecular biological methods. It is also necessary to study the hybrid nature using molecular markers (preferably - AFLP-markers) regarding the resistance genes. In addition, the *microsatellite (SSR)* - and AFLP-*markers* can be used for the genetic analysis. But the already obtained cytological data provide indisputable evidence that the synthesis of a new grape genome is, certainly, one of the major scientific and practical achievements in selection of vine concerning its immunity. In fact, in support of our research, the selectionists - winegrowers of the Institute of Vine and Wine "Magarach" note that, nowadays, using modern molecular genetic studies, indisputable evidence of the presence in the structure of synthetic species *V.rotundifera, V.vinifolia, V.cruceştiana, V.nigra* and others, of the genes of *V.vinifera* L. *V.rotundifolia* Michx. as well as their share in the new genome can be brought (Volinkin, Likhovsky Oleinik, 2011).

And finally, at the hybrids F_5, the passing of meiosis without deviations in both gametophytes was recorded, as judged by the normal shape and size of the pollen grains, the formation of bunches of normal density with grapes and viable seeds in synthetic species of vine (Figure 5), like the bisexual varieties of Vitis vinifera L. Ultimately, here, the synthesis reached its climax – the fertility was restored (the basic properties of a plant are to produce fruits and to reproduce, which are shown by the restoration of the vital functions of the reproductive organs, which implies a mechanism of reproduction and conservation of their own species in a given environment).

Figure 5. Bunches of grapes of the distant hybrids F5: DRX-M5-740 (left) and DRX-M5-745 (right)

These are the stages of the radical transformation of the sterile hybrid of the American selection DRX-55, the vegetable mule with 2n = 39, into the synthetic species (Zawadzki, 1968; Topală, 2008, 2011; Topală, Dadu, 2006, 2009), which are the exponents of the new genome and harmoniously combine in a single genotype the high quality of the crop with the resistance to diseases and pests, including the phylloxera.

Conclusions

For the first time in the world, a new genome of vine (n = 19) from the chromosomes of the genome of the species *Vitis vinifera* L. (n = 19) and the chromosomes of the genome of the spontaneous American species, resistant to phylloxera, *V.rotundifolia* Michx. (n = 20) was synthesized, (according to the available data concerning the morphological structure, the chromosomes are in the ratio of 9vv +10 vr), but a more precise determination of the belonging of the chromosomes to a certain species could be established by means of differential colouring and other modern methods of colouring and studying the chromosomes.

For the first time in the world's science, the unpaired 39th chromosome (the so-called "scandalous intriguer" or "tricky chromosome" because it caused the sterility of the gametophytes and the low, practically zero, fruitfulness of the hybrids) was displaced from the nucleus (karyotype) in the cytoplasm by performing intensive backcrosses with the hybrids Seyve Villard. The process took place over three generations (F_3, F_4, F_5) as evidenced by the direct counting of chromosomes in the metaphase plates.

Among the hybrids of the 5th generation the artificial synthetic types of grapes were revealed: *Vitis viniolia, V.rotundifera, V.cruceştiana, V.nigra* and others that are new exponents of the genome of vine and successfully combine the quality and quantity of the crop of the species *Vitis vinifera* L. with the resistance of the species *V.rotundifolia* Michx. (Fig. 5).

On the basis of the hybrid DRX-55 a new plant species was synthesized, of the type *V.vinifera* L. (Fig. 6) similar to the own-rooted cultural grapes growing before the appearance of phylloxera in Europe (1868), but differs fundamentally from them by its hybrid origin and high resistance to phylloxera.

Figure 6. Bunches of grape of the distant hybrid (F_5) DRX-M_5-734, or the synthetic species *Vitis vinifolia*, which indicate the complete recovery of fertility.

The synthesis of the new genome of vine, providing high quality grapes and resistant vines, allowed, for the first time, to confirm, using the species Vitis vinifera L. as an example, the accuracy of the chromosomal hereditary theory of T. Morgan (1933).

Now, it is established that the synthesis of the new genome of vine from the chromosomes of the species *V. vinifera* L. and *V.rotundifolia* Michx. has been carried out in accordance with the latest international scientific researches, with the same object of study, as the paper: "The discovery of how chromosomes are protected by telomeres and the enzyme telomerase", done by the American scientists E.H.Blackburn, C.W.Greider, J.W.Szostak (2009).

For the first in the world, *a valuable source of genes which indicate the resistance to phylloxera was created*, which may be involved in hybridization as a donor of this feature, as germ plasma, or as a maternal form. The widespread use of this source of genes will make possible to create a gene pool of 10,000 seedlings and to solve successfully the global challenge of combating phylloxera.

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BIOLOGICAL AND CHEMICAL STUDY OF VOLATILE OIL FOR *PEROVSKIA ATRIPLICIFOLIA* BENTH SPECIES

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Abstract: The species *Perovskia atriplicifolia* is a perennial, herbaceous plant of the *Lamiaceae* family. In the Republic of Moldova, it is not investigated as an aromatic and medicinal plant. It has a high ecological plasticity, no special requirements to climatic factors. It is appreciated for its febrifuge properties and is used in medicine to reduce fever. It is also used as a culinary plant and with decorative or air purification purposes.

Key words: plant, volatile oil, components, chromatography, antioxidant compounds

STUDIUL BIOLOGIEI ŞI COMPOZIŢIEI CHIMICE ALE ULEIULUI VOLATIL LA SPECIA *PEROVSKIA ATRIPLICIFOLIA* BENTH

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Rezumat: Specia *Perovskia atripicifolia* se comportă ca plantă perenă, erbacee din familia *Lamiaceae*. În R. Moldova nu este cercetată ca plantă aromatică şi medicinală. Are o plasticitate ecologică mare, nu are cerinţe deosebite faţă de factorii climaterici. Este apreciată pentru proprietăţile sale febrifuge şi utilizată în medicină pentru scăderea febrei, prezintă şi utilizări culinare, dar şi decorative ori de purificare a aerului.

Cuvinte cheie: planta, ulei volatil, componenţi, cromatografie, compuşi antioxidanţi

Introduction

Perovskia sp. is a genus of flowering plants in the family *Lamiaceae*. The members of this genus are native to southwest and central Asia. It comprises nine species, including the species popularly called Russian sage (*Perovskia atriplicifolia*).

Perovschia atriplicifolia Benth. The seeds material was originally received from Russia. The plants were grown in the open, in ecological balance, on the general agro background without the use of fertilizers. In the Botanical Garden, it was grown from seeds received by the international exchange of seeds in 2006, to reflect the peculiarities of development and volatile oil content, highlighting the processes of reproduction, selection of some plant forms with a higher productivity of herba and volatile oil , development of primary processes of growth and cultivation, research of chemical composition of essential oil, experimentation of herba and volatile oil samples in perfume and cosmetics industry and in medicine.

Material and methods

Phenological observations were made on 25 plants, once in three days until the end of the flowering stage and once a week - until the end of the growing season, according to the program developed by the Botanical Garden of Moscow.

The biological research has been carried out according to the methodical requirements of Rabotnov (1950), supplemented later by Smirnova (1976, 1987); the essential oil was isolated by steam distillation of the aerial parts of the plant and the supercritical fluid extraction method (SFE); qualitative and quantitative phytochemical analysis of the plant was investigated for the determination of antioxidant compounds of polyphenols and flavonoids type; in order to obtain the volatile oil, it was used 50 grams of the dry plant material that has been hydrodistillated (Neo-Clevenger method) to obtain a volume of 1.5 ml/100 g, the resulting oil was subjected to the gas chromatographic method; the chemical composition of the essential oil was determined by gas chromatographic analysis coupled with mass spectrometry (GC-MS) using the gas chromatograph Agilent Technologies type 6890N coupled with the mass detector (MSD) type 5975 inert XL Mass Selective Detector.

Results and discussions

The results of the experimental studies have shown that the species *Perovskia atriplicifolia* is a perennial, herbaceous plant of the *Lamiaceae* family. In Moldova, it is not researched as an aromatic and medicinal plant. It has a high ecological plasticity, no special requirements to climatic factors. It develops a bush of 17-18 whitish stems and lobed leaves, deeply toothed gray- silver with a length of 5 cm and a width of 2.5 cm. The mature stems are woody at the base and the young stems are herbaceous and square in cross section. The stems and the leaves give off a pungent odour

of sage when crushed. Because of its characteristic smell, in England, this plant is called "Russian sage". In late August the plant blooms, producing inflorescences with tubular flowers of blue or lilac colour. The flowers can reach a length of 30 cm and last up to 3 months. The plant forms a bush, which in native conditions reaches a height of 1.5 m and a circumference of 60 cm, in our conditions it reaches 80-90 cm in height.

Chemical composition. It was analyzed the essential oil isolated by steam distillation of the aerial parts of the plant *Perovskia atriplicifolia* Benth. Of the 39 compounds identified, the major components were 1.8-cineole and limonene (40.13%), α-pinene (17.87%), δ-3-carene (9.13%), β-pinene (6.59%), camphene (6.17%) and camphor (5.36%).

In another study of the species *Perovskia atriplicifolia* Benth, the results of the supercritical fluid extraction method (SFE) were compared with those obtained by steam distillation. It was investigated the effect of various parameters, such as pressure, temperature, solvent type and volume of solvent on the yield of extraction of the essential oil. The extracts obtained by the two methods have very different composition. The main constituents of the oil obtained by steam distillation were: 1.8 - cineole, limonene, camphor , β - caryophyllene , α - pinene , camphene and α - humulene. On the other hand, the major components of the SFE extract were as follows: 1.8- cineole, limonene, camphor, β - caryophyllene, cadinene, γ , α - pinene and α - terpinyl acetate . The results showed that an increase in temperature from 35 ° C to 65 ° C (at a constant pressure of 100 atm) drastically reduced the number of extracted components. Also, the number of extracted constituents and the percentage of the key analytes increased when lower pressures were used. By using different solvents (e.g. methanol , ethanol , dichloromethane and hexanes) to extract the essential oil under reduced pressure (100 atm.) and a temperature of 35 ° C, it was shown that the hexane was more selective than other solvents (Pourmortazavi et al ., 2003).

In *Perovskia atriplicifolia* Benth collected from Pakistan, were 19 isolated volatile components. Of these, δ3-carene monoterpenes (22.3%) and 1.8-cineole (27.5%) is approx. 50% of the oil, and β-caryophyllene (10.8%) and α-humulene (5.7%) were the dominant sesquiterpene (Jassbi et al., 1999).

Action: It is appreciated for its febrifuge properties and used in medicine to reduce fever; it is also used as a culinary plant and with decorative or air purification purposes.

Qualitative and quantitative phytochemical analysis of the plants *Perovskia atriplicifolia* grown in the collection of the Botanical Garden were performed to determine the antioxidant compounds: polyphenol and flavonoids, specifically for the analysis of volatile oil.

The results reveal the biosynthetic capacity of the plant for the above mentioned compounds.

The analysis was performed by thin-layer chromatography (CCS), high performance liquid chromatography (HPLC) and by gas chromatography analysis combined with mass spectroscopy (GC-MS).

Two types of extracts were analyzed: methanol and ethanol made from dried plant material. For the extract, 2.5 grams were used, which were extracted 3 times with 30 ml methanol and ethanol alcohol, and the extract is brought to mark at 100 ml. These extracts were analyzed phytochemically.

To obtain the volatile oil was used 50 grams of dried plant material that was hydrodistllated (Neo-Clevenger method) to give a volume of 1.5 ml/100 g. The resultant oil was subjected to gas chromatographic method.

HPLC-UV is the separation on a high performance liquid chromatograph Agilent 1200 type equipped with a reverse phase column Eclipse XDB-C18 (150 mm x 4.6 mm, 5 □ m) UV-Vis detector MULTIDIODE (English diode-array detector).

The separation was carried out using a mobile phase (gradient concentration) consisting of solution of acetonitrile and 2 mM sodium acetate (adjusted to pH 3.5 with glacial acetic acid). (Table 1)

Table 1.

The concentration gradient used was as follows:

Time(minutes)	% CH_3COONa 2 mM (pH=3,5)	% CH_3CN
0	98	2
20	86	14
40	80	20
50	70	30
60	75	25
65	98	2
70	98	2

UV detection was performed at multiple wavelengths (220, 240, 260, 280**, 300, 320**, 350, 370 nm).

In order to identify the peaks, it was used the comparison of the values of retention time in the sample of chromatogram with the standards and the comparison of the absorption spectra for the obtained peaks with those of the standards analyzed in the same chromatographic conditions in both cases.

Thus, after the optimization of the chromatographic working conditions for the separation, standard solutions of gallic acid, chlorogenic acid, caffeic acid, salicylic acid, ferulic acid, p-coumaric, o-coumaric acid, rosmarinic acid, cinnamic acid, rutoside (quercetin-3-O-rutinozida), hyperoside (quercetin-3-O-galactoside), luteolin, luteolin-7-glucoside, apigenol, apigenin-7-glucoside, quercetol were injected.

Table 2.

The retention time for each standard substance

Nr.crt.	Standard Name	Retention time (min)
1	Gallic acid	3.92
2	Chlorogenic acid	11.46
3	Caffeic acid	14.85
4	Salicylic acid	17.61
5	P-coumaric acid	20.84
6	Ferulic acid	24.04
7	Rutoside	27.05
8	Hyperoside	27.35
9	Luteolin-7-glucoside	28.53
10	Rosmarinic acid	30.5
11	O-coumaric acid	29.68
12	Apigenol-7-glucoside	34.53
13	Cinnamic acid	45.24
14	Luteolin	47.93
15	Quercetol	48.14
16	Apigenol	53.01

For these standards, the absorption spectrum in UV was registered, a spectrum that was saved in the library of spectra. It has further been used for the identification of these compounds for various samples.

Figure 1 presents the chromatograms obtained for the standards, at a detection wavelength of 320 nm.

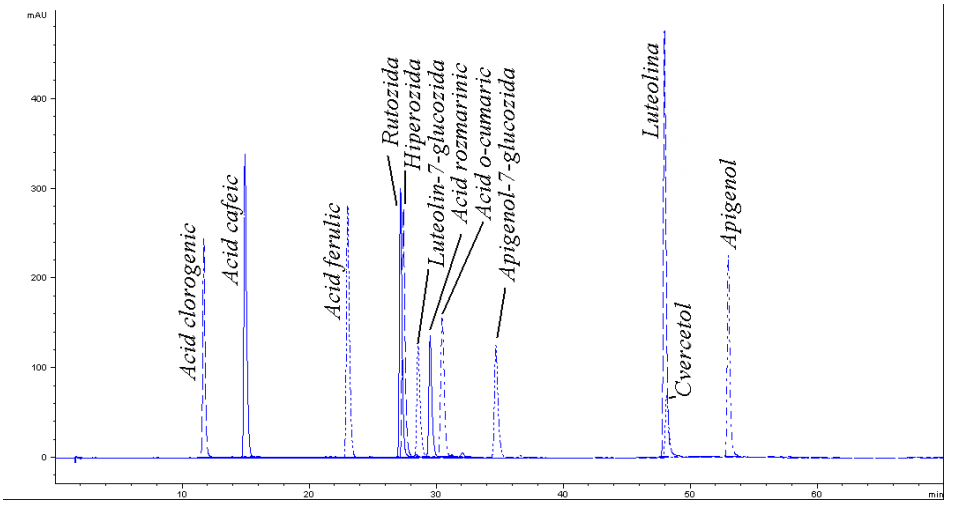
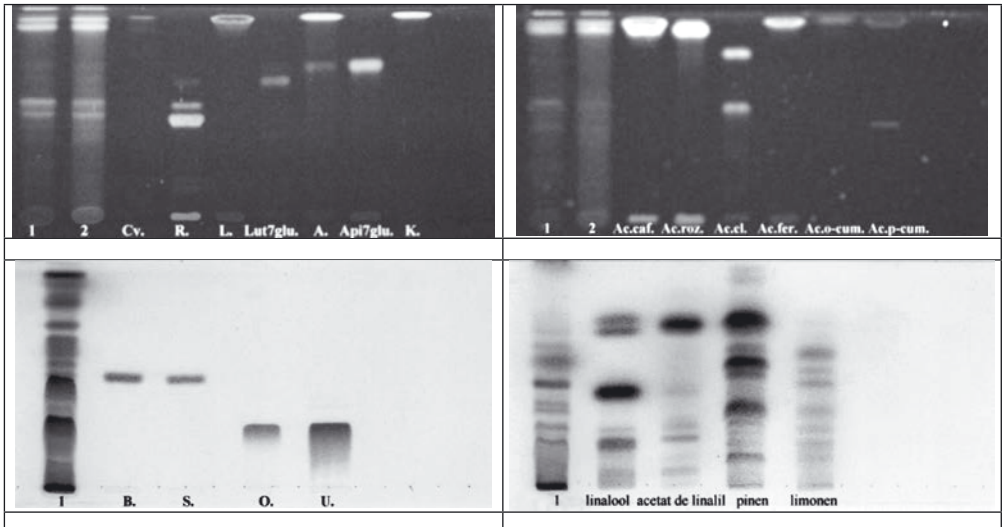


Figure 1. Chromatograms of the calibrators

For the analysis of the antioxidants of the samples it was done the following: alcoholic extracts were analyzed by HPLC under the same conditions. The obtained chromatograms were integrated. By comparing the relative retention time to the standards, but also by spectral comparison, it was revealed the presence / absence of the substances monitored in the samples analyzed.

Determination of the chemical composition of volatile oils and identification of the volatile terpene metabolites



Legend
Samples: **1**=*Perovskia atriciplifolia*, Chisinau-extract Me(OH); **2=** *Perovskia, atriciplifolia* Chisinau-extract Et(OH)70%;
Flavones standards: **Cv. = Cvrcetol, R. = rutoside, L. = luteolin, luteolin-7 Lut7glu =-O-glucoside, A. = apigenol, Api7glu. = Apigenol-7-O-glucoside, kaempferol K.**
Polyphenolcarboxylic standards: **Ac.caf. = Caffeic acid, Ac.roz. = Rosmarinic acid, chlorogenic acid Ac.cl. =, Ac.fer. = Ferulic acid, Ac.o-how. = O-coumaric acid, Ac. p-p-coumaric acid**
Triterpene standards: B. = β-sitosterol, stigmasterol S = O. = oleanolic acid, ursolic acid U
Oil standards: linalool, linalyl acetate, pinene, limonene

Figure 2. Qualitative phytochemical analysis by TLC

The chemical composition of the essential oil was determined by gas chromatographic analysis combined with mass spectrometry (GC-MS) using gas chromatograph Agilent Technologies 6890N type combined with mass detector (MSD) type 5975 inert XL Mass Selective Detector.

Chromatography conditions were as follows:

- HP 5MS column dim. ext. 30 m x 0.25 mm - dim.int. 0.25 μm (5% Phenyl-methylsiloxane);
- Mobile phase: Fibers - flow rate: 1 mL / min;
- Injector temperature: 250 ° C;
- Detector temperature: 250 ° C;
- Temperature regime: from 40 ° C initially (10 ° / min.) to 280 degrees (constant 5.5 min);
- Injection volume: 0.1-0.3 ¶;
- The rate of split-1: 100.

The appearance of chromatograms highlights, according to the standards, the flavonoids of the type rutoside (R.) and quercetol (cv.) and polyphenols, especially polyphenol acids such as rosmarinic acid (Ac. pink.), caffeic acid (Ac.caf.) and ferulic acid (Ac.fer.).

For triterpenes and phytosterols contained in dichloromethane extracts, standards have identified the presence of beta-sitosterol (B) and oleanolic acid (O) and ursolic acid (U). (Figure 2)

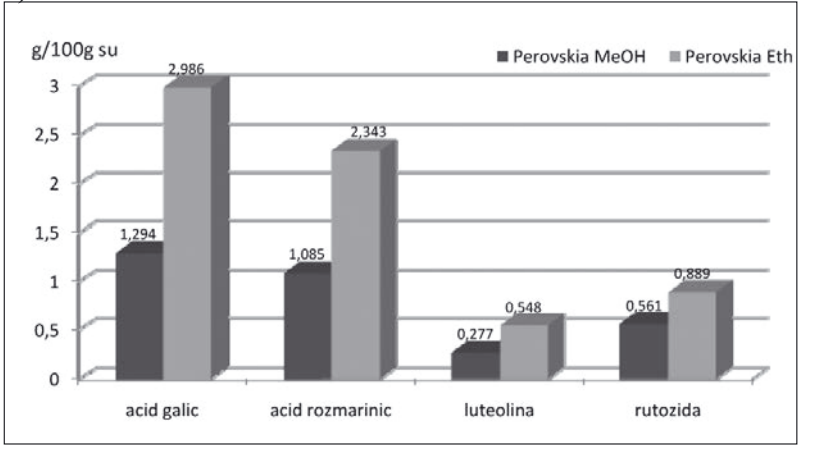
Table 3.

Quantitative phytochemical analysis by spectrophotometry / (a;b)

a)

Species/ Extract type	Polyphenols	Polyphenolic Acids rb.	Flavone	
	Gallic acid	Rosmarinic acid	Luteoline	**Rutoside**
	g/100g dm	g/100g dm	g/100g dm	**g/100g dm**
Perovskia MeOH	1.294	1.085	0.277	**0.561**
Perovskia Eth	**2.986**	**2.343**	**0.548**	**0.889**

b)



The amount of compounds with antioxidant action of polyphenolic and flavonoidic type is higher in the ethanolic extracts so that the gallic acid concentration is of 2.986 g/100g dm, and the rosmarinic acid is of 2.343 g/100g dm.

Concerning the flavonoids, the rutoside (0.889 g/100g dm) has higher values compared with the luteolin (0.548 g/100g dm).

The analysis of the volatile oil by gas chromatography combined with mass spectrometry (GC-MS) (Figure 4)

The volatile oil analysis is characterized by the presence of 28 compounds (Table 4) that have an area of above 0.5%; the others were detected with very low values, and their sum is found in the table at the *other compounds*.

The compounds identified with the highest values are: D-limonene - 21.47%, eucalyptol - 16.19%, α-pinene - 8.17, caryophyllene (α and β) - 11.91%, borneol and bornyl acetate (4.34% and 6.06%, respectively).

Table 4.

The compounds identified by GC-MS of volatile oil of Perovskia atriplicifolia Benth., of the plants selected from the collection of the Botanical Garden (I.) of Chişinău (2013)

Nr.crt.	Kovats Index	Retention Time	Compound	Area %
1	914	5.16	α-Thujene	0.20
2	923	5.29	**α-Pinene**	**8.17**
3	939	5.51	Camphene	3.87
4	969	5.94	β-Pinene	3.93
5	980	6.09	β-Myrcene	0.98
6	1011	6.53	Δ-3-carene	0.23
7	1020	6.66	*p*-Cymene	0.89
8	1028	6.76	**D-Limonene**	**21.47**
9	1031	6.80	**Eucalyptol**	**16.19**
10	1043	6.98	*cis*-β-Ocimene	0.23
11	1057	7.17	γ-Terpinen	0.55
12	1099	7.76	Linalool	0.53
13	1146	8.41	Sabinol	0.56
14	1176	8.84	**Borneol**	**4.34**
15	1187	8.99	4-Terpineol	0.51
16	1200	9.18	alfa-Terpineol	0.54
17	1299	10.56	**Bornyl acetate**	**6.06**
18	1360	11.41	α-Terpinyl acetate	3.08
19	1434	12.46	**β-Caryophyllene**	**6.20**
20	1466	12.91	**α-Caryophyllene**	**5.71**

21	1490	13.24	Germacrene D	0.36
22	1519	13.65	τ-Cadinene	0.91
23	1527	13.76	Calamenene / Cadina-1,3,5-triene	0.22
24	1586	14.59	Caryophyllene oxide	3.43
25	1599	14.77	α-Bisabolene epoxide	0.20
26	1612	14.95	Cubenol	0.64
27	1637	15.30	τ-Cadinol	3.77
28	1648	15.45	α-Eudesmol	1.24
			Other compounds	*4.99*

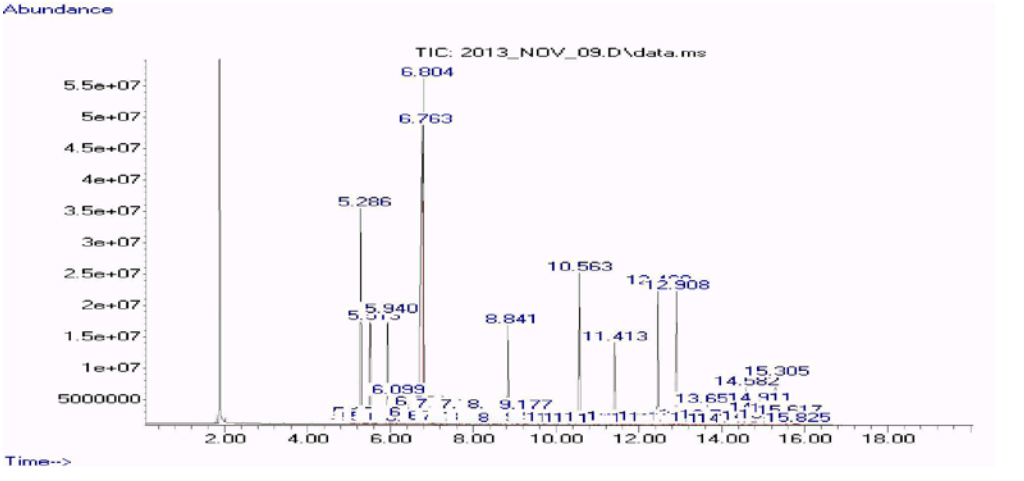


Figure 4. The GC-MS chromatogram of volatile oil of *Perovskia atriplicifolia* Benth. (Chisinau-2013)

Conclusions:

The results of the conducted studies are consistent with the results obtained in the doctoral thesis of C. Paduraru (2012) and with the project conducted by the Botanical Garden of Iasi - IDEI program project, code CNCSIS 2100, Theme 1040/2009.

In addition, our results correspond to those in the specialized literature, but there are some differences. So, I. Burzo and C. Toma (2012) have analyzed by GC -MS the volatile oil extracted from leaves and flowers of plants of Perovskia atriplicifolia Benth; we found that the volatile fractions correspond to those analyzed by us (except the camphor), but the quantitative share is different. The camphor and the limonene prevail in the leaves, and τ - cadinol, α - pinene and limonene in flowers. According I. Burzo and C. Toma (2012), the composition of the essential oil extracted from leaves has the following values: camphor - 15.16 % , limonene - 13.77 % , τ - cadinol - 10.13 % , eucalyptol - 9.03 % , β - caryophyllene - 6.20 % , α - caryophyllene - 5.71 % , δ

-3- caren - 5.65 % , α - pinene - 4% , α - terpenil acetate - 3.45 % , camphene - 3.4 %, borneol - 2.56 % cubenol - 2.22 % , β - pinene - 2.05 % bornyl acetate - 1.95 % , γ - cadinene - 1.5 % , 0.73 % α - eudesmol , myrcene - 0.62 % , α - terpineol - 0.5 % -0.41 % terpinolene , δ - cadinol - 0.34 % , γ -terpinene - 0.23 % , and other compounds at low concentrations.

The iranian researchers Pourmortazavi and colab. (2003), have analyzed by GC-MS the volatile oils from herba of Perovskia atriplicifolia Benth. They used two methods to obtain oil, namely the water vapour distillation and the supercritical fluid extraction (SFE). By classical method, distillation with water vapour, the volatile oil contains: eucalyptol, limonene, camphor, β-caryophyllene, α-pinene, camphene and α-humulene. By modern method, supercritical fluid extraction, the volatile oil obtained from Perovskia atriplicifolia plants originating from Iran contains: eucalyptol, limonene, camphor, β-caryophyllene, γ-cadinene, α-pinene and α-terpenil acetate. Also, in the volatile oil of Russian sage originating from of Iran, F. Sefldkon and colab. (1997)have identified 39 compounds of the dominant volatile fractions: limonene, eucalyptol + (40.13%), α-pinene (17.87%), δ-3 carene (9.13%), β-pinene (6.59%), camphene (6.17%) and camphor (5.36%).

The present compounds (especially the dominant ones: limonene, eucalyptol, α-pinene, α, β- caryophyllene) give to the volatile oil of Russian sage an amount of biological properties, namely: antibacterial, antiviral, anti-inflammatory and antitumor [James A. Duke, 1992].

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ONTOGENESIS AND QUALITY INDICES OF *CARTHAMUS TINCTORIUS* GROWN UNDER THE CONDITIONS OF IRRIGATION IN THE SOUTH OF UKRAINE

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Abstract: Results of the researches of ontogenesis and quality indices of the Carthamus tinctorius grown under the conditions of irrigation in South Ukraine are given in the article. Quality-quantitative changes of the biochemical composition of the oil of the variety “Sonachniy” from the studied factors are shown; it can be used in the pharmacological industry.
Key words: Carthamus tinctorius, irrigation, terms of sowing, quality-quantitative indices, oil content in seeds.

ОНТОГЕНЕЗ И КАЧЕСТВЕННЫЕ ПОКАЗАТЕЛИ *CARTHAMUS TINCTORIUS* ВЫРАЩИВАЕМЫЙ В УСЛОВИЯХ ОРОШЕНИЯ НА ЮГЕ УКРАИНЫ

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Резюме: В статье приведены результаты исследований онтогенеза и показателей качества Carthamus tinctorius, выращиваемый в условиях орошения на Юге Украины. Исходя из результатов качественно-количественных изменений биохимического состава масла формы «Sonachniy», то он может быть использован в фармацевтической промышленности.
Ключевые слова: *Carthamus tinctorius*, орошение, сроки сева, показателей качества-количественная, содержание масла в семенах.

The oil-bearing crops have a great economic value due to the various and wide application of the products of their processing in different industries of the national economy. The plants belonging to the group of oil-bearing have seeds and fruits containing a lot of fat (from 20 to 60%); they are the basic raw material for the obtaining

of oil. According to the prognosis of scientists-climatologists, the air temperature will rise by approximately 1-4°C on the territory of Europe in 2030. There is a forecast, concerning the amount of precipitations, showing the tendency to more droughty weather in summer and moist - in winter. Thus, the change of climate will affect the agro climatic conditions, namely lengthening of vegetation period, plants moisture providision, hydrothermal indices [1-3].

Adaptation of agricultural crops to the climate change will also be done through replacement of the grown crops with more heat-loving and drought-resisting ones. One of the perspective oil-bearing crops for growing under the droughty conditions of the South of Ukraine is *Carthamus tinctorius*, the morpho-biological features of which are adapted to the extreme conditions of South Steppe of Ukraine [4-6].

Taking it all into account, the task to study the ontogenesis and quality indices of the crop under the conditions of irrigation of the South of Ukraine was put.

The field and laboratory researches with safflower were conducted during 2010-2012 in the Institute of Rice of NAAS of Ukraine, located in Skadovsk District of Kherson Region. Different elements of technology were studied, including, terms of sowing of the crop under study.

The estimation of laboratory indices of the selected vegetable samples was conducted using the method of Ginzburgh [7]; determining of mass fraction of essential oil on the device of Clevenger with the subsequent count of dry mass equivalent. An essential oil was got through the method of hydro-distillation from fresh inflorescence in the period of the mass flowering of *Carthamus tinctorius*. The composition of essential oils was determined using the method of chromatography on Agilent Technology 6890N chromatograph with a mass-spectrometry detector 5973 N. The components of essential oils were identified with the help of the method of content-index and the method of addition of pure matters and mixtures of the known chemical composition. The content indices of components were calculated on the basis of results of the test analyses of essential oils [8].

The duration of the interphase periods of *Carthamus tinctorius* in experiments was different. The most protracted periods were «sprouting - budding» and «flowering – ripeness», less long – «budding – flowering» (figure 1). The influence of terms of sowing on the duration of both interphase periods, and vegetation on the whole was also defined – increase of the given indices when sown in early terms and, vice versa, decline at the late sowing.

In the years of conducting of researches, the productivity depended on the terms of sowing and fluctuated averagely – 1.89 t/ha at the early term of sowing (3^{d} ten-day period of March) to 1.24 t/ha at the late term of sowing in the 3^{d} ten-day period of April. The terms of sowing were corrected depending on the soil-climatic conditions of the year.

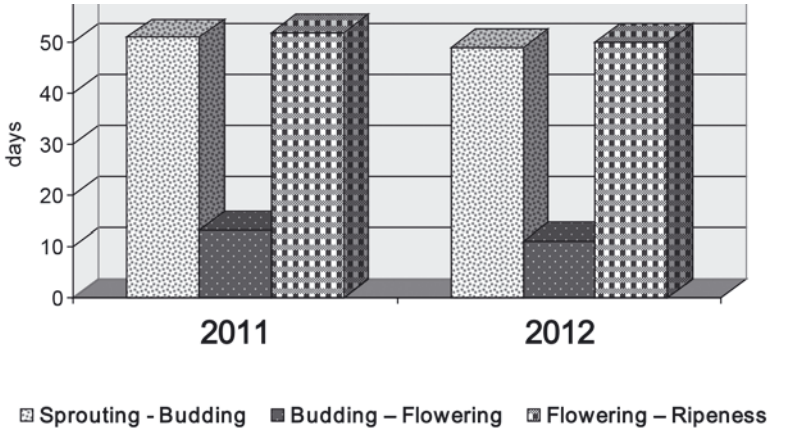

Figure 1. Duration of interphase periods of *Carthamus tinctorius* plants under the conditions of irrigation

Scientific researches established, that the seeds of *Carthamus tinctorius* contain about 15-37% of drying light yellow, fat oil, with its amount rising to 60% in kernel. The oil got from cleared seeds, does not yield in quality to the sunflower according to the most essential indices. It is used in food and for margarine production. It is applied for technical aims too, for the production of white paints and enamels, which cannot turn yellow in the course of time. The dyeing matter – cardamom, contained in flowers, is used for handicraft industry of carpets. In oriental cuisine it is used as a substitute of saffron. Achenes of *Carthamus tinctorius* are good forage for birds, and its pomace is suitable for cattle feeding in small doses because of its bitter taste [9].

The results of the biochemical analysis showed, that the oil of *Carthamus tinctorius* contains the heightened content of lanolin, oleic, palmitic and stearic acids the amount of which depended on the amount of harvest of seeds got as a result of different terms of sowing (figure 2).

The early terms of sowing excelled the middle and the late terms on productivity of oils seeds averagely on 15-32%, accordingly.

The value of polyunsaturated fatty acids in the body of a human being is very great. On the one hand, they hinder the development of atherosclerosis and bring down the level of cholesterol in blood, on the other hand – they have antiinflammatory action. These acids belong to omega-3 unsaturated fatty acids promoting quick transformation of cholesterol into bile acids and getting it out of a human body.

In addition, they strengthen the walls of blood vessels, promote their elasticity and bring down the risk of atherosclerosis development; take active part in the synthesis and exchange of vitamins B, B_6, choline; improve the conductivity of nervous impulses in the central and peripheral nervous system.

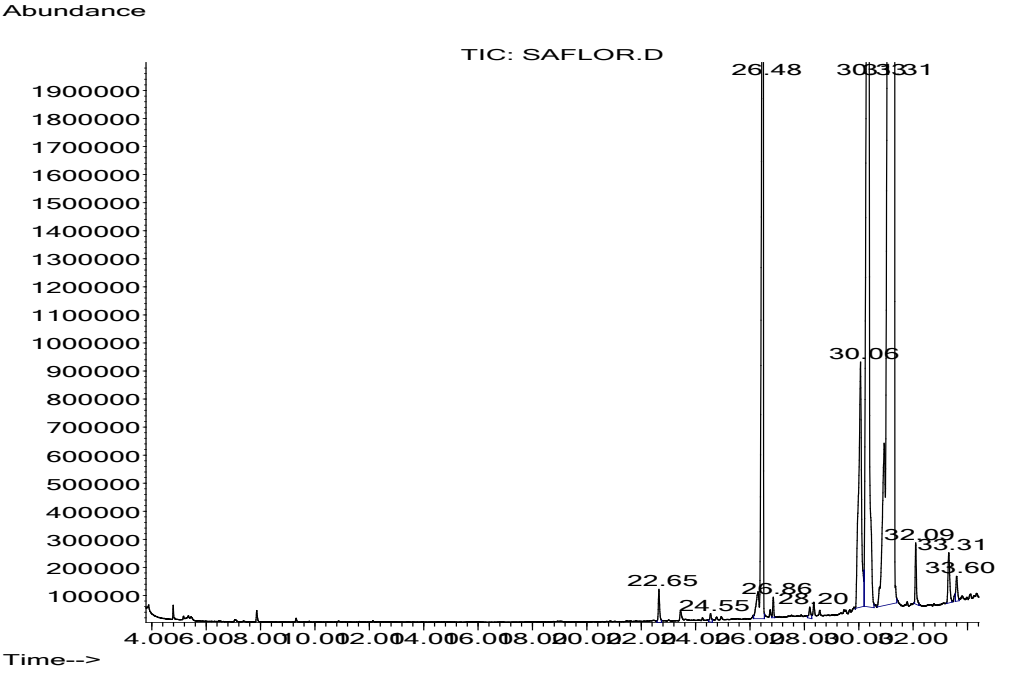

Figure 2. Content of fatty acids in the oil of *Carthamus tinctorius*

The unsaturated fatty acids are divided on mono-, di-, tri-, tetra-, penta-, hexane according to the number of double bonds. The given connections of unsaturated fatty acids possess double bonds and they are the structural elements of phospholipids of membranes in the human body. It should be noted that linolic, linolin, arachidonic and other irreplaceable fatty acids, enter into a body with food, and their high content in *Carthamus* oil allows it to be recommended for people and animals.

Thus, the study of terms of sowing of *Carthamus tinctorius* allows defining the features of plant ontogenesis, duration of interphase periods and vegetation of plants. The influence of natural and agrotechnical factors on productivity of plants and quality indices is set. According to the results of researches *Carthamus* is recommended to be sown in early spring period, which will allow getting high and stable harvest of oil seeds and high-quality oil. Its consumption will allow to bring down the content of cholesterol in blood and rise the vital tone.

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INFLUENCE OF AGROTECHNICAL METHODS ON THE QUALITATIVE INDICATORS OF ESSENTIAL OIL OF FENNEL IN THE ZONE OF SOUTHERN STEPPE OF UKRAINE

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Abstract: The article highlights the important aspect of introduction of the valuable aromatic plant, fennel, in the steppe area of southern Ukraine. It describes the effect of cultivation technology elements (the dates of sowing, the spaces between rows, mineral nutrition background) on the content of essential oil in fennel seeds.

Key words: *Foeniculum vulgare*, essential oil, fennel seeds, the dates of sowing, row spacing, mineral nutrition background, fertilization rates.

ВЛИЯНИЕ АГРОТЕХНИЧЕСКИХ ПРИЕМОВ НА КАЧЕСТВЕННЫЕ ПОКАЗАТЕЛИ ЭФИРНОГО МАСЛА ФЕНХЕЛЯ В ЮЖНЫХ СТЕПНЫХ ЗОНАХ УКРАИНЫ

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Резюме: В статье освещается важный аспект внедрения ценного ароматического растения, фенхеля, в степной зоне Юга Украины. Описывается эффекты элементов технологии выращивания (даты сева, промежутки между рядами, минерального питания фоне) на содержание эфирного масла в семена укропа.

Ключевые слова: *Foeniculum vulgare*, эфирное масло, семена укропа, даты посева, расстояние между рядами, минеральное питание фон, темпы оплодотворения.

Fennel (*Foeniculum vulgare Mill.*) – valuable essential-oil, officinal, spicy, vegetable, aroma, melliferous and ornamental plant. Fennel seeds and products of its processing are used in medicine, cooking, in a variety of industries, veterinary medicine, animal husbandry [1, 2]. The beneficial properties of plants are caused by the

essential oil and its major components – anethole (anis camphor) and fenchone [1, 2]. The demand for fennel oil is growing every year and requires searching for new ways to improve its yield and collection [3].

In Ukraine the traditional zones of cultivation of the crop are Western regions and the Crimea region [1]. In recent years, due to the rapid development of various branches of industry and increase of production capacity, there was a necessity of expansion of areas under *Foeniculum vulgare* and its introduction into rotation in new regions, in particular in the southern Steppe of Ukraine.

In 2011-2013 field experiments were established and conducted in the Kherson Regional State Centre for Expertise of Plants Varieties on dark chestnut soils, typical for the southern Steppe of Ukraine. The plan of the experience included such factors and their variants: Factor A – nutrition background: without fertilizers; N_{30}; N_{60}; N_{90}; Factor B – the sowing date: early (the 3rd ten-day period of March); middle (1st ten-day period of April); late (2nd ten-day period of April); Factor C – row spacing width, cm: 15; 30; 45; 60. The experiment is laid with the help of the method of split sections, replication – fourfold. The agrotechnics of fennel cultivation during the experiment was usual, except for the factors and options that have been studied. The essential oil content in seeds was determined using the method of Ginsburgh [4].

The results of the studies testify to the fact that the content of essential oil in the seeds of fennel depends on the effect of hydrothermal conditions and of the factors studied.

In 2011 the mass fraction of essential oil in the seeds of fennel, on average over the experiment, constituted 5.74, in 2012 and 2013 – 5.52 and 5.69%, respectively. The decrease of this indicator in 2012 in comparison with other years of research may be associated with the losses of essential oil under the influence of considerable amount of precipitation in the interphase period of fructification - ripeness.

The content of essential oil in the seeds of fennel, averagely for the years of research, changed according to the variants of experiment from 5.14 to 6.27% of absolutely dry matter. The most favourable conditions for accumulation of essential oil were observed in N_{60}, when sown in the early period with the width of the row spacing of 45 cm, the least favourable – without fertilizers control, sown in a later date with usual rows.

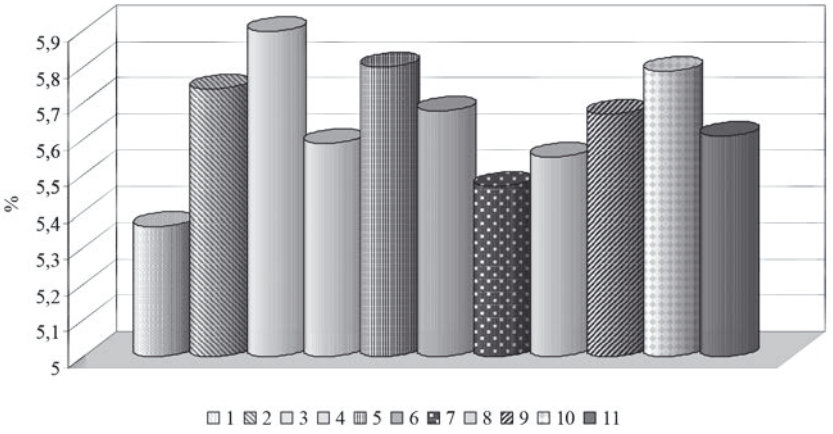
In average by factors of value of indicator in the variant without fertilizers was 5.36%, nitrogen fertilizers have increased it 1.04-1.10 times. The mass fraction of essential oil in the seeds of fennel under the background of N_{30}, N_{60} and N_{90} increased relative to the control test 0.38; 0.54 and 0.23%, respectively (Fig. 1). Under the background of the N_{90} there was a decrease of this characteristic in comparison with the variants of application of N_{30} and N_{60} on 0.15 and 0.31%, respectively.

The positive influence of fertilizers on the content of essential oil in the fennel seeds depended on the interaction with the other surveyed factors. Under the back-

ground of N_{60}, there was an increase of this characteristic relatively to the control test of 1.08 times on the sites of late term sowing with a row spacing of 15 and 60 cm; 1.13 times when sown in the third decade of March with row spacing of 45 cm.

The mass fraction of essential oil in the fennel seeds sown in the early period, in the third ten-day period of March constituted 5.80%. When seeds are sown 10 – 20 days later, the analyzed characteristic fell by 0.12 - 0.33 %, or 1.02-1.06 times.

Average factor, the highest concentration of essential oil in seeds of 5.79% was observed at sowing with aisle 45 cm. Changing the width of the aisle relatively 45 cm resulted in the decrease of this indicator by 0.12-0.24 %, or 1,02-1,04 times.

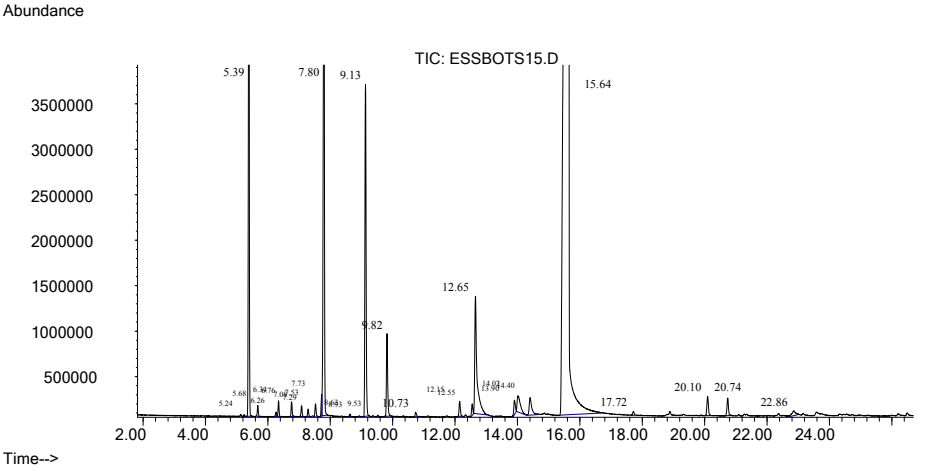


Factor A - background supply: 1 - without fertilizers; 2 - N_{30}; 3 - N_{60}; 4 - N_{90}.
Factor B - sowing date: 5 - early; 6 - medium; 7 - late
Factor C - row spacing: 8 - 15 cm; 9 - 30 cm; 10 - 45 cm; 11 - 60 cm

Fig. 1. The content of essential oil in the fennel seeds, on averagely due to the studied factors, % of absolutely dry matter

The chromatographic analysis of the composition of the fennel seeds essential oil revealed 28 chemical compounds, the main of which are trans-anethole, limonene, α-pinene, fenchone, estragole, linalool (Fig. 2).

In addition, the essential oil of fennel contains such components as: α-tuyen, camphene, sabinene, beta-pinene, beta-myrcene, beta-phellandrene, Delta-Karen, p-cymene, 1,8-cineole, gamma terpinene, trans-linalool oxide, α-terpinolene, camphor, terpinene-4-ol, p-COP-1-ene-8-ol, carvone, 4-methoxybenzaldehyde, 1-methoxy-4(prop-1-enyl)benzene, patience acetate, trans-caryophyllene, beta-bergamot, α- bisabolen.



1 - 5,246 (0,023% α- tuyen); **2** - 5,395 (7,691% α-pinene); **3** - 5,679 (0,151% camphene); **4** - 6,261 (to 0.055% sabinene); **5** - 6,346 (0,215% β-pinene); **6** - 6,761 (0,216% β-mircen); **7** - 7,086 (0,169% β-phellandrene); **8** - 7,289 (0.115% Delta-Karen); **9** - 7,524 (0,219% p-cymene); **10** - 7,727 (0,282% 1,8-cineole); **11** - 7,799 (8,770% limonene); **12** - 8,629 (0.040% γ-terpinene); **13** - 8,926 (0,015% trans linalool oxide); **14** - 9,129 (5,726% fenchone); **15** - 9,531 (0,027% α-terpinolene); **16** - 9,819 (1,435% linalool); **17** - 10,735 (0,090% camphor); **18** - 12,147 (0,279% terpinene-4-ol); **19** - 12,548 (0,203% p-COP-1-ene-8-ol); **20** - 12,652 (3,817% estragole); **21** - 13,901 (0,322% carvone); **22** - 14,014 (0,767% 4-methoxybenzaldehyde); **23** - 14,402 (0,654% 1-methoxy-4(prop-1-enyl)benzene); **24** - 15,647 (67,581% trans-anethole); **25** - 17,721 (0,084% patience acetate); **26** - 20,098 (0,422% trans-caryophyllene); **27** - 20,743 (0,369% β-bergamot); 28 - 22,858 (0,262% α-bisabolen).

Fig.2. Component composition of fennel seeds essential oil

Therefore, the most favourable conditions for the accumulation of essential oil in the seeds of fennel were observed under N_{60}, when carrying out the sowing in the early period (in the third ten-day period of March) with the width of the row-spacing of 45 cm.

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УДК 635.35..631.58..631.544.7 (477-292.485)

THE EFFECTIVENESS OF APPLICATION OF HYDROGEL AKVOD AND MULCHING OF SOIL WITH ORGANIC MATERIALS AT GROWING CAULIFLOWER IN THE CONDITIONS OF FOREST-STEPPE OF UKRAINE

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Abstract: The results of the investigations of the application of water-holding granules Akvod for growing seedlings of cauliflower in containers and mulching of soil with sawdust and straw in the conditions of Forest-Steppe of Ukraine are given. It was found, that the application of water-holding granules Akvod and mulching of soil are influencing the terms of approach of phenological phases and duration of interphase periods of cauliflower plants. A considerable influence of the studied methods on the biometric characteristics of plants and crop capacity of commercial product was marked. The biggest diameter of heads was marked in the variants with application of granules and mulching of soil with sawdust. The variants with mulching of soil with sawdust and straw differed in the mass of the head. The application of water-holding granules Akvod for growing seedlings of cauliflower in containers and mulching of soil with sawdust and straw are increasing the crop capacity by 2.6-10.2 t/ha, securing of higher qualitative characteristics of product and biggest share of first sort in comparison with the control variant.

Key words: cauliflower, sawdust, straw, water-holding granules Akvod, variety Unibotra, mulching of soil.

ЭФФЕКТИВНОСТЬ ПРИМЕНЕНИЯ ГИДРОГЕЛЯ АКВОД И МУЛЬЧИРОВАНИЯ ПОЧВЫ ОРГАНИЧЕСКИМИ МАТЕРИАЛАМИ ПРИ ВЫРАЩИВАНИИ КАПУСТЫ ЦВЕТНОЙ В УСЛОВИЯХ ЛЕСОСТЕПИ УКРАИНЫ

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Резюме: Приведены результаты исследований применения водоудерживающих гранул Аквод при выращивании рассады капусты цветной в кассетах и мульчирования почвы опилками и соломой в условиях Лесостепи Украины. Установлено, что применение водоудерживающих гранул Аквод

и мульчирование почвы влияют на сроки наступления фенологических фаз и продолжительность межфазных периодов растений капусты цветной. Отмечено значительное влияние изучаемых приемов на биометрические показатели растений и урожайность товарной продукции. Наибольший диаметр головки отмечено в вариантах с применением гранул и мульчированием почвы опилками. Варианты с мульчированием почвы опилками и соломой отличались массой головки. Применение водоудерживающих гранул Аквод при выращивании рассады капусты цветной в кассетах и мульчирование почвы опилками и соломой повышает урожайность на 2,6–10,2 т/га, обеспечивает высокие качественные показатели продукции, наибольшую массовую долю первого сорта по сравнению с контрольным вариантом.

Ключевые слова: капуста цветная, опилки, солома, водоудерживающие гранулы Аквод, сорт Униботра, мульчирование почвы.

For Ukrainian agriculture, mulching is a promising means of conservation of the soil moisture and prevention of the soil loss due to erosion. There is no scientific assessment of mulching on the soils of Ukraine and the researches in this area are very few. Mulching reduces the evaporation of soil moisture. Applying soil mulching under drought conditions reduces the nonproductive loss of moisture 1.7 times, and with sufficient moisture - 3 times. A significant positive impact of mulching on the moisture regime is determined to the depth of the root-inhabited layer - 50 cm. Mulching also improves the temperature regime and the agro-physical condition of the soil. Mulching significantly increases the effectiveness of the mineral fertilizers, especially in arid growing conditions (nitrogen - by 53-60%, phosphorus and potassium by 20-23 %). The productivity of agricultural crops increases by 20-25% as a result of soil mulching. Soil mulching is advisable in the zone of insufficient or unstable moistening [1].

Under the conditions of unstable moistening, which include the forest-steppe of Ukraine, an important issue in the technology of growing vegetables and, in particular, cauliflower is the preservation and rational use of moisture during the growing season of the plants. The requirements of cauliflower plants are caused by the biological characteristics of the species. By traditional technology the use of mechanized processing does not preclude the question of use of manual labour for loosening the soil in rows and destroying the weeds. In this regard, mulching can reduce the expenses of the manual labour to care for the plants and to reduce the evaporation from the soil surface.

Along with the mulching of the soil, the application of new superabsorbents enables the plants to make rational use of moisture during the growing season, reducing the variations of the soil moisture in the absence of rainfall during the short-term droughts that occur periodically in the forest-steppe zone. The hydrogel Akvod – is a new generation of materials that have the unique ability to absorb and retain at swelling to 4 litres of water in 10 g of granules. The hydrogel is not toxic, it retains its properties at high and low temperatures in the soil for up to 5 years. It saves the water at irrigation up to 50-60%. The preparation is presented in the form of granules. [2]

Materials and Methods

The purpose of the research is to study the influence of the water-holding granules Akvod on growing seedlings and mulching of the soil with sawdust and straw on plant productivity and crop quality of cauliflower.

The studies were conducted in 2011-2012, on the experimental field of Vinnitsa National Agrarian University. On the experimental field there was gray-forest, medium-loamy soil. The humus content - 2.4%, the reaction of the soil solution (pH) - 5.8, the total absorbed bases - 15.3 mg equivalent/100 g of soil, P_2O_5 - 21.2 mg/100 g of soil, K_2O - 9.2 mg/100 g of soil. The variety Unibotra of cauliflower was cultivated using the seedling method. The seedlings were grown in a greenhouse for seedlings in boxes with a cell size of 6x6 cm. The cultivation technology of the seedlings was common. During the cultivation of the seedlings, in the experiment, it was studied the variant with the use of Akvod hydrogel granules, which were added at a rate of 20 g of granules per 10 kg of soil mixture. In the control variant, the granules were not used. In the experiments, it was considered the variant with mulching of soil with sawdust and straw, and the control variant - without mulching. The seedlings were planted in early April, at the age of 60 days, in the soil prepared in accordance with the zonal recommendations. The plants were planted according to the scheme 70x30 cm, and then, the soil was mulched with sawdust and straw. At each tonne of straw and sawdust were added 5 kg of nitrogen in the form of ammonium nitrate to improve the nitrification and to prevent the nitrogen deficiency of plants. The methodology provided for phenological observations, biometric measurements and calculations. When the plants reached the technical maturity, the harvest was gathered and calculated. [3] The harvest was gathered as the heads were formed, in accordance with the applicable standard - «ДСТУ 3280–95. Капуста цвітна свіжа. Технічні умови» [4].

Results and Discussions

According to the average data, the soil temperature in the areas covered by sawdust, depending on the time of the measurements, was in the range of 5.7 to 22.2 ° C, in the variant with mulching with straw - from 5.7 to 23.1 ° C, in the control variant – from 5.4 to 23.5 ° C, that is with 0.3-1.3 ° C higher. The reduction of the soil temperature under the mulching materials of organic origin may be due to the fact that the light surface of straw and sawdust reflects a part of sunbeams unlike the dark surface of the soil, which absorbs them to a greater extent. During the years of research, in the period of planting of the seedlings, the lowest average temperature in the first ten days of April was observed in 2011 – 4.2 ° C, in 2012 the value of this index amounted to 6.3 ° C, that is with 2.1 ° C higher. The plants got a lower sum of effective temperatures in 2011 - 463 ° C, with 115 ° C less than in 2012. Taking into account the biological characteristics of cauliflower plants, very important for their

growth and development during the growing season is the relative humidity. It should be noted that during the years of research, the humidity indices were the lowest in 2011 - 62.0%, whereas in 2012 - 66.7%. Despite the low humidity during the growing season in 2011, the given index was within the optimal limits. According to the literature, the optimal humidity for growing cauliflower varies between 60-85% [5].

The optimal soil moisture for growing cauliflower is of 70-80% minimum moisture-holding capacity [5]. Concerning the years of the research, it should be noted that during the growing season of 2011, in the control variant, during 30 days, the soil moisture was below the optimum and fluctuated between 50.7-67.7% of the minimum moisture-holding capacity, in 2012, in a period of 50 days it was observed a decrease of the soil moisture below the optimal level. In the variants where mulching with straw had been used, in 2012, in a period of 30 days the soil moisture was below the optimal level – 64.4-69.8% minimum moisture-holding capacity, while in the variants where mulching with sawdust had been used, the soil moisture indices were within the optimal limits and above.

During the interphase period: transplanting the seedlings – formation of the heads, the variant with the mulching of soil with sawdust and use of granules was more notable: the period lasted 47 days, and in the control variant - 49 days, that is, by 2 days longer. The interphase period: formation of the heads – the technical maturity in the variant with mulching of the soil with sawdust was of 22 days and it was by 2 days shorter compared with the control variant. In addition, when the soil had been mulched with sawdust, it was also noted a simultaneous obtaining of production - 11-12 days, but when the soil had been mulched with straw, the production was obtained during 13 days, and in the control variant - 14 days.

In the phase of technical maturity, the greatest plant height was observed when the soil had been mulched with sawdust (53.2 and 57.6 cm) and straw (43.6 and 47.1 cm). In the control group, this index was 39.9 cm, that is by 13.3 cm and 17.7 and 3.7 and 7.2 cm lower (Table 1). Regarding the thickness of the stem, the best results were observed in the variants with mulching of the soil with sawdust - 13.5 and 14.3 mm and straw - 12.8 and 13.3 mm, in the control variant - 11.4 mm, so, the stems were by 2.1 and 2.9 and 1.4 and 1.9 mm thinner.

The analysis revealed a strong direct connection between the plant height and the stem thickness (r=0.93). In the phase of technical maturity, a larger diameter of the rosette of leaves had the plants in the versions with mulching of the soil with sawdust and with the use of granules - 60.2 cm and without the use of granules - 55.9 cm, and in the control variant - 44.7 cm, that is by 25.7 and 20.0% smaller. The analysis revealed a strong direct connection between the plant height and the diameter of the rosette (r=0.99). The leafiest plants in the given phase were in the variants with mulching of soil with sawdust - 16.8 and 17.6 pc./plant, and in the control variant - 14.4 pc./plant, that is by 2.4 and 3.2 pc./plant less.

An important index for assessing the condition of the plants is the leaf area and it was the highest in the variant with mulching of the soil with sawdust and using granules for growing the seedlings - 38.0 thousands m^2/ha and without the use of granules - 35.2 thousands m^2/ha and in the control variant - 24.2 thousands m^2/ha, that is by 13.8 and 11.0 thousands m^2/ha less.

Table 1

The biometric and physiological characteristics of cauliflower plants in the phase of technical maturity of heads after mulching the soil and using water-holding granules (average for 2011-2012)

Variant		Plant height, cm	Number of leaves, pieces.	Stem thick-ness, mm	Diameter of the rosette, cm	Leaf area, thousands m^2/ha	Net photosyn thetic-productivity g/m^2 per day
Mulching material	Use of granules						
Sawdust	Without granules	53,2	16,8	13,5	55,9	35,2	8,5
	With granules	57,6	17,6	14,3	60,2	38,0	9,1
Straw	Without granules	43,6	15,0	12,8	49,9	28,8	7,1
	With granules	47,1	15,7	13,3	51,7	31,1	7,6
Without mulching	Without granules (C)	39,9	14,4	11,4	44,7	24,2	6,4
	With granules	42,1	14,5	12,4	46,2	27,8	6,9

C – control

The analysis revealed a strong direct relationship between the leaf surface area and the thickness of the stem (r=0.97). According to the average data, high rates of net photosynthetic productivity of plants were observed in the variants with mulching of the soil with straw and with the use of granules - 9.1 g/m^2 per day and without the use of granules - 8.5 g/m^2 per day, and in the control variant - 6.4 g / m^2 per day, that is by 2.7 and 2.1 g/m^2 per day less. In all the variants with the use of water-retaining granules it was noted a higher net productivity by 0.4-0.5 g/m^2 per day in comparison with the variants without the use of granules. The analysis revealed a strong direct relationship between the leaf area and net photosynthetic capacity (r=0.99).

The most significant index characterizing the effectiveness of the studied methods is the crop yield (Table 2). The highest yield of marketable products of cauliflower was obtained in the variants with mulching of the soil with sawdust and with the use of granules - 29.6 t/ha and without granules - 27.0 t/ha, while in the control variant the crop yield was of 19.4 t/ha, that is by 10.2 and 7.6 t/ha less. The importance of this difference was confirmed by the results of the analysis of variance.

Table 2

The yield and quality parameters of cauliflower crop after mulching of the soil and application of water-holding granules

Variant		The average for 2011-2012.		Crop yield t/ha			±, to the control
Mulching material	Use of granules	diameter of the head, cm	weight of the head, g	2011 г.	2012 г.	average	
Sawdust	Without granules	14,9	566	28,5	25,4	27,0	+7,6
	With granules	15,6	621	31,2	27,9	29,6	+10,2
Straw	Without granules	13,1	462	23,7	20,3	22,0	+2,6
	With granules	13,4	501	25,2	22,5	23,9	+4,5
Without mulching	Without granules (C)	12,4	407	20,9	17,8	19,4	–
	With granules	12,9	449	23,1	19,6	21,4	+2,0
$НИР_{05}$	A	–		1,2	1,3	–	
	B			1,0	1,1		
	AB			1,7	1,9		

C – control

The crop yield was significantly higher, in comparison with the control, in the variants with mulching of the soil with straw and without mulching, with the use of granules. The analysis revealed a strong direct relationship between the leaf area and the crop yield (r=0.99), and a strong direct relationship between the crop yield and the net photosynthetic productivity (r=0.79). In the years of the researches, higher crop yields in all the investigated variants were obtained in 2011, which is due to the large

amounts of precipitation during the growing season in that year of research - 211.5 mm against 164.0 mm in 2012, that is by 47.5 mm less. The analysis revealed a strong direct relationship between the yield of cauliflower and the amount of precipitation (r=0.99).

The greatest diameter of the head was observed in the variants with mulching of the soil with sawdust and with the use of granules - 15.6 cm and without granules - 14.9 cm, and in the control variant - 12.4 cm, that is by 20.5 and 16.8% less. The analysis revealed a strong direct relationship between the crop yield and the diameter of the head (r=0.99).

The best results concerning the weight of the head was observed in the variants with mulching of the soil with sawdust - 566 and 621 g, and with straw - 462 and 501 g, and in the control variant - 407 g, that is by 159 and 214 and 55 and 94 g less. The analysis revealed a strong direct relationship between the crop yield and the mass of the head of cauliflower (r=0.99). It was established as well a strong direct relationship between the mass of the head and its diameter (r=0.99).

The largest share of the first-rate production in the total yield was observed in the variants with mulching of the soil with sawdust and with the use of water-holding granules - 26.9 t/ha and without the use of granules - 22.7 t/ha, and in the control variant - 14.2 t/ha, that is by 12.7 and 8.5 t/ha less (Table 3). The analysis revealed a strong direct relationship between the crop yield and the share of the first-rate production in the total yield (r=0.99). The smallest share of the second-rate production in the total yield was observed in the variants with mulching of the soil with sawdust and with the use of granules - 2.2 t/ha, and in the control variant - 5.2 t/ha, that is by 3.0 t/ha more. The analysis revealed a strong inverse relationship between the crop yield and the share of the second-rate production in the total yield (r=-0.87). It was also established a strong inverse relationship between the share of the first and second-rate production (r=-0.91).

Table 3

The structure of the cauliflower yield after the mulching of the soil and application of water-holding granules (the average for 2011-2012)

Variant		First-rate		Second-rate	
Mulching material	Use of granules	t/ha	%	t/ha	%
Sawdust	Without granules	22,7	84,0	4,3	16,0
	With granules	26,9	90,6	2,7	9,4
Straw	Without granules	16,8	75,9	5,3	24,1
	With granules	18,6	77,9	5,3	22,1
Without mulching	Without granules (control)	14,2	73,0	5,2	27,1
	With granules	16,4	76,7	5,0	23,4

Conclusions

So, as a result of the conducted research, it has been found that the use of mulching of the soil and the use of water-holding granules Akvod influence the timing of the phenological phases and the duration of the interphase periods of cauliflower plants. There was observed a significant influence of the studied techniques on the biometric characteristics of the plants and the yield of marketable products. The highest yield was provided by the variants with mulching of the soil with sawdust and with the use of granules - 29.6 t/ha and without granules 27.0 t/ha, which provided a significant increase in yield in comparison with the control variant - 10.2 and 7.6 t/ha. These variants also provided high quality indices of the production, especially, there were obtained high indices of the share of the first-rate production in the total yield - 26.9 t/ha (90.6%) and 22.7 t/ha (84.0%), respectively. The analysis revealed a strong direct relationship between the crop yield and the share of the first-rate production in the total yield (r=0.99). The smallest part of second-rate production in the total yield was observed in the variants with mulching of the soil with sawdust and with the use of granules - 2.2 t/ha, and in the control variant - 5.2 t/ha, that is by 3.0 t/ha more. The analysis revealed a strong inverse relationship between the crop yield and the share of the second-rate production in the total yield (r=-0.87) and it was also established a strong inverse relationship between the share of the first and second-rate production (r=-0.91).

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IV. LANDSCAPE ARCHITECTURE

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PERSPECTIVES OF USING *GINKGO BILOBA* L. IN LANDSCAPE-GARDENING

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Abstract: A brief survey of the history and some results of the introduction of *Ginkgo biloba* L. were given. The possibility of using the cultivars of this species in ornamental gardening in the Republic Moldova was presented. For the future, the mobilization of 27 new cultivars was planned.
Keywords: *Ginkgo biloba*, landscape-gardening, mobilization, the new cultivars.

PERSPECTIVE DE UTILIZARE A *GINKGO BILOBA* L. ÎN ARHITECTURA PEISAGISTICĂ

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Rezumat. În articol o succintă prezentare a istoriei și unele rezultate ale introducerii *Ginkgo biloba* L. au fost aduse. A fost luată în examinare posibilitatea de a folosi soiurile acestei specii în arhitectura de landșaft a Republicii Moldova. Mobilizarea a 27 de soiuri noi a fost planificată pe viitor.
Cuvinte-cheie: *Ginkgo biloba*, arhitectura de landşaft, mobilizare, cultivaruri noi.

Introduction

In the landscape-gardening of the cities and villages of Moldova, a significant place is occupied by the gymnosperms. However, the assortment of conifers, used in landscaping is not very rich; only except for a few of them which there are in abundance, because of what the originality of plantations suffers (*Picea pungens* 'Glauca',

Juniperus communis 'Stricta', *Pinus pallasiana*, *P. sylvestris*, *P. nigra*, *Picea abies*, *Platicladus orientalis*, *Juniperus sabina* etc.).

Low-growing species and cultivars of conifers are not commonly used, except for private gardens, particularly for designing the rocky hills and, are not always used in the recent years species which show stability when are exposed to unfavorable environmental conditions.

For the enrichment of the assortment of gymnosperms plants, useful in Moldova for ornamental horticulture, it is necessary to involve new species and cultivars, to study their bioecological peculiarities, and to develop optimal methods for their mass reproduction.

As one of the most interesting and unique species, at this stage, our attention was paid to the Mesozoic relict of the Japanese-Chinese origin, a single representative of the class *Ginkgopsida* – bilobate ginkgo (*Ginkgo biloba* L.), that has been preserved till our days and, being one of the most beautiful and long-lived and durable breed species.

Ginkgo biloba L. – a dioecious and deciduous tree, reaches 35-40 m in height and 4-4.5 m in diameter. The pyramidal crown, at the male plants is narrower, but at the female plants is wider. In natural conditions ginkgo grows in the mountain forests of South-Eastern China (Tien-Mu-Shan), together with coniferous and deciduous species, not higher than 1500 meters above the sea level in the warm and humid climates. It has been cultivated since ancient times. It has been cultivated since the XI century, in China, in all the regions with a temperate climate. Later, it was introduced in Japan and Korea.

The German botanist Engelbert Kaempfer, who visited Japan at the end of the XVII century, studied *Ginkgo biloba* L. and gave a description with illustrations of leaves and fruits in his book "Amoenitatum exoticarum" (1712), and called this original tree – ginkgo. Later, Carolus Linnaeus introduced this name in botanical literature.

In Europe, ginkgo, for the first time, was introduced in 1730 in the Botanical Garden of Utrecht (Netherlands), from 1754 Ginkgo was cultivated in the Royal Botanical Gardens, Kew (United Kingdom). In the Botanical Garden of Vienna (Austria), it was introduced approx. in 1786.

In North America from England, William Hamilton had brought gingko in 1784.

In the Ukraine, in culture, it was registered at the beginning of the XIX century (in 1809, in Krasnokutsk acclimatization garden, in 1811 in Kremenetsk botanical garden, and from 1818 in the Nikita Botanical Gardens).

Materials and methods

The objects of study were *Ginkgo biloba* L. plants growing in the Botanical Garden of the Academy of Sciences of Moldova, in the urban plantations, ancient parks, and also in private gardens. For researches, known methods, recommendations

to refine the taxonomic composition, determining the degree of hardness and drought resistance, reproductive ability, the level of adaptation and perspective of introduction were used [1, 2, 3, 4].

Results and discussions

In Moldova, *Ginkgo biloba* L. has been cultivated since the second half of XIX century. This is due to the landlords' laying of gardens and parks in the villages Mileshti (second half of XIX century) Ivancea (1880), Ginkauts (end of XIX century) Temeleuts (1903-1908).

The secular, the largest specimens are growing in the park. Mileshti, district Nisporeni. Here grows one female and one male specimens of *Ginkgo biloba* L. which have reached a height of 23 and 22 meters and 0.90 and 0.65 m in diameter. The female specimen differs by significant and regular fructification.

In Chisinau there are several fruit-bearing specimens of ginkgo.

In the Botanical Garden, Academy of Sciences of Moldova, there is a group of male and female specimens that reproduce and develop well.

The specimens of both sexes are growing in the park and near the building of the Government of the Republic of Moldova. The female specimen fructifies abundantly and makes viable seeds.

Several ginkgo trees grow in the dendrological garden of Chisinau.

Young specimens are often found in private gardens and parks, but, in gardening of cities and villages, ginkgo trees have not received a proper distribution. In Chisinau, the pollen of ginkgo matures in the end of April - the first half of May. The buds begin to blossom in April, the foliage ends in the second half of May. The leaves are fan-shaped, or dissected into two lobes, are leathery, glabrous, of bluish-green coloration. The growth of shoots finishes in July - early August. The annual growth in height is of 35-45 cm. In the Central Moldova conditions, the seeds mature in October. The fruits are round or oval, with a fleshy envelope reaching 1.9-2.8 cm in diameter, of amber yellow color, unpleasantly smelling due to the presence of succinic acid. The seeds are without a fleshy cover, woody, dihedral, tri- tetrahedral. The kernel is edible, sweetish toasted in the form in Japan and China is used in food. A mass of 1000 seeds with the fleshy envelope weighs 5.5-6.0 kg, without envelope - 1.5-2.0 kg.

Besides the basic and single *Ginkgo biloba* L. species in the ornamental horticulture of Moldova (mainly in private gardens and collections) were identified 10 cultivars:

'Anny's Dwarf' – dwarf form (with slow growth);

'Aurea' – is characterized by golden-yellow color of the leaves;

'Fastigiata' – differs in its columnar crown, the branches moving away from the trunk up;

'Korinek' – conical form;

'Mariken' – has a compact spherical crown;

'Saratoga' – cultivar with a narrow columnar and compact crown;

'Tit' – dwarf form of fancy crown;

'Troll' – compact form, with leaves from the normal to the rounded;

'Tubifolia' – trees with dense crown and dissected leaves on young shoots and leaves twisted in the tube on the older shoots;

'Variegata' – a shrubby form with variegated foliage.

Some ecological and biological peculiarities and the elaborated optimal methods for their propagation have been studied.

The reproduction of Ginkgo has a number of features. Fresh harvested seeds have high germination. After cleaning, the seeds were sown at once, at a depth of 3-5 cm, in the substrate (boxes) with sand, compost, peat and black earth in relation 1:1:1:1. Up to 30-35 seeds per linear meter can be sown. Groundwater seed germination was 90%. The annual ginkgo seedlings were planted into the containers for further growth.

Ginkgo is well reproduced by green cuttings. Cuttings were carried out in greenhouses with high pressure fogging system, in the period from spring to midsummer. Harvesting grafts, was performed by cutting shoots with a heel or cutting with a part of last year's wood. In order to enhance the root formation, cuttings were treated with chemicals. We used indolylacetic acid (IAA, 250 mg / l) and IBA acid (IBA-25 mg / l).

Cuttings were tied in bundles and placed in above-mentioned solutions for 16-24 hours, plunging the lower ends at 2-3 cm, and just before the planting, cuttings are dipped into the Kornevin powder.

Before planting the cuttings for the rooting in the substrate, which consisting of sieved river sand, was disinfected with a weak solution of potassium permanganate, but before the planting was heavily moistened with water. The cuttings were planted in straight rows, with a distance of 3-4 cm into a substrate depth of 1.5-2 cm. The substrate around the cuttings is slightly compacted and after planting abundantly watered and covered with foil.

Ginkgo is well reproduced by the cleft grafting method, in the splitting of the axial shoot, by copulation and inoculation. Inoculation in the cleft and shoot axial splitting was carried out before bud blossoming in early spring. The works on inoculation were performed in the cool greenhouse. As the rootstock were served 2-3-year-old seedlings, grown from seeds, collected from female plants in the park of the village Mileshti, district Nisporeni. As grafts macroblasts and microblasts served the cuttings of 3 cultivars of *Ginkgo biloba* L.('Mariken', 'Saratoga', 'Variegata') respectively collected from female and male specimens, in the same locality. Cuttings were harvested in February and kept in plastic bags in the refrigerator till the moment of inoculation. As strapping material for connecting grafted components, a special band for inoculation was used. The rooting rate inoculation at the female and the male constituted 85% and 80%, respectively. Inoculations of cultivars also gave a good

result – more than 70% rooting rate. The copulation and inoculation were performed in April and August, respectively. The highest rooting rate (75%) has been received in the spring by copulation. The great intraspecific biodiversity of *Ginkgo biloba* L. (130 of cultivars) [5, 6, 7, 8] creates greater opportunities for enriching the natural dendroflora by new ornamental plants.

In the future is planned the mobilization of cultivars with improved ornamental characteristics (male specimens):

'Barabit's Nana' – dwarf form, up to 2 m in height;

'Beijing Gold' – shrubby form, up to 4 m, yellow leaves in spring, and striped in summer;

'Chase Manhattan' – compact form, up to 1.5 m, small leaves, dark-green color;

'Chi-Chi' – with a bizarre shape of the crown;

'Chris Dwarf' – compact form (with slow growth);

'Elmwood Fastigiata' – has a columnar shape;

'Fairmount' – is characterized by a dense pyramidal crown;

'Globus' – has a spherical shape with large leaves;

'Heksenbezen Leiden' – compact, rounded, dwarf form, up to 3 m;

'Horizontalis' – trees with the broad crown, lower branches practically are horizontal relative to the trunk;

'Jade Butterfly' – semi-dwarf with slow growth;

'Louis' – unique sort of slow growth;

'Magyar' – has a vertically narrow pyramidal crown;

'Pendula' – cultivar which has a weeping form of the crown;

'Princeton Sentry' – a narrowly columnar form up to 30 m;

'Spring Grove' – dwarf variety, with a compact crown, up to 3 m in height, as well as female specimens with the larger seeds and diverse by form (as a fruit of culture):

'Epiphylla' – have fused ovule and leaf petioles, seeds seem to be attached to the leaves;

'Eastern Star' – Chinese cultivar with abundant fruiting, large seeds;

'Geisha' – Japanese cultivar with large fruits;

'King of Dongting Mountain' – Chinese cultivar exclusively for seed production;

'Long March' – upright growing trees, its fruits are large and tasty;

'Ohazuki' – Japanese variety with slower growth, and with several vertices;

'Salem Lady' – sort with large fruits, therapeutic in the baked form;

'Santa Cruz' – a compact umbrella-shaped form;

'Sanstream' – form with variegated foliage;

'Tremonia' – trees with a column-shaped crown expressive texture, and bluish green foliage;

'Umbrella' – the low-growing trees, with an umbrella shaped crown.

The majority of these cultivars can be purchased at ornamental nurseries and garden centers in Europe, with subsequent cultivation in nursering farms in Moldova.

Conclusions

The perspectivity of *Ginkgo biloba* L. in ornamental horticulture of Moldova is justified by many factors: it grows well, reproduces well, the generative reproduction, by cuttings, method of inoculation is characteristic of *Ginkgo biloba* L., it is not exigent to soil and climatic conditions, is resistant to winter frost and drought, dust and gases, to fungal and viral diseases, practically it is not affected by pests, its requirements for environmental factors are insignificant.

Ginkgo is light-demanding; exigent to soil moisture content (can't tolerate stagnant water and excessively dry soil). At young ages, there is the probability of damages of the roots by rodents.

Ginkgo can be used more often in urban landscaping and green building, in this case it is necessary to take into account his dioecious state, i. e. the fact that the fruits, at maturity, have an unpleasant strong smell, as well as the fact that the fruits which fall down may clog up the territory under the canopy of the crown.

That is why, in urban plantations, only male specimens should be used.

Ginkgo should be more widely used in the school arboretum with educational purposes as a relict plant of the ancient flora.

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STYLES OF ARRAGEMENT OF ESTATE PARKS (HISTORIC GARDENS) OF THE REPUBLIC OF MOLDOVA

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Abstract: This paper presents the styles of landscape architecture: regular style, landscape style and mixed style. Also, in the article are described the main characteristics of these styles and their trends. For each style were described models of parks from Europe and Republic of Moldova.

Key-words: style of landscape architecture, regular, landscape and mixed style.

STILURILE DE AMENAJARE ALE PARCURILOR MOŞIEREŞTI DIN REPUBLICA MOLDOVA

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Rezumat. În această lucrare sunt prezentate stilurile arhitecturii peisajere: regulat, iregulat şi mixt. De asemenea, sunt reliefate caracteristicile lor principale şi tendinţele contemporane. Pentru fiecare stil au fost expuse modele de parcuri din Europa şi Republica Moldova.

Cuvinte-cheie: stil de arhitectură peisajeră, stilul regulat, iregulat, mixt.

Introduction

Human beings have always sought to create and admire the beauty around them. One of the forms of man-made beauty is the landscape architecture. This art combines the beauty of plants and the human creativity. Although the first gardens were created for practical purposes - cultivation of medicinal plants and vegetables, over time they became decorative.

The art of arranging gardens is an old one; its origins date back to the ancient times [8]. Until nowadays we have evidence about the ancient gardens from old drawings and writings. The ancient Egyptians, Assyrians, Babylonians, Greeks, Romans and other peoples created their own gardens. In medieval times, more precisely at their

end, in the Renaissance era, and in the modern era there were shaped and formed the landscape architecture styles: regular (geometric), irregular (landscape) and mixed.

The estate parks in our republic were also created in regular, irregular and mixed styles. However, they have some distinct characteristics, due to the specific local culture and landscape.

The goals and the objectives of this research work are to highlight the styles of landscape architecture and to exemplify them by presenting models of European and Moldavian parks (gardens).

Materials and Methods

There are various estate parks in our country. A lot of estate parks are based on different species of trees, and fit into the category of dendrological parks. Most dendrological parks are concentrated in the areas of forest steppe in the North and in the areas of forest in centre of the Republic of Moldova, an area with fertile soils and rich in black earth. Fewer parks were created in the south of the country – a steppe area, unfortunately, poor in forests, [6].

The general condition, diversity and basic characteristics of the old estate parks were investigated at the initial stage. The factual materials were obtained by selecting and studying bibliographic sources and documents, plans and schemes of parks, by visiting and documenting objects, by comparing the estate parks of our country with those of Europe and other methods.

The analysis of the parks was performed on the base of landscape architecture styles: regular, irregular and mixed. Also, were presented models of parks in every style both of the whole European continent, as well as local ones.

Results and Discussions

It was mentioned above, that in the landscape architecture, three styles: regular, irregular and mixed had been created and shaped. The first two styles have distinctive features, and the mixed style is a combination of the regular and irregular ones.

The geometric (regular) style is characterized by straight alleys and strict location of the elements of the parks and the actual elements of architecture. Floral arrangements and water objects with regular shape are also typical. At the same time, the fountains and flower layers are shaped like geometric figures such as rectangle, circle, square and others. This style goes so far as to require a regular shape of trees and shrubs which is obtained by cleaning and pruning [16]. The same characteristics of this style can also be found in other works from this field [1, 3, 12]. Models of parks in regular style are: Versailles, Trianon, Vaux-le-Vicomte in France, Villa Torlonia, Villa Albani in Italy and others. In our country, there aren't parks arranged in regular style, but there are areas of parks designed according to the principles of this style, as mentioned in the paper.

Versailles. It is a palace-park ensemble located in the suburb Versailles of Paris, France. Initially Versailles was a hunting palace with a small parterre and a huge hunting park. It was rebuilt after 1662 by King Louis XIV's order. In the planning of the park, the natural conditions had been taken into account; the palace was built on a terrace with the facade facing west. The park was arranged in sectors.

The first sector, the official one, is placed between the palace and the cross-shaped channel. This section includes the main decorative elements (fig. 1): the parterre, the bushes, the water objects and the green carpet. The latter is a great lawn of 600x60m, surrounded by parallel alleys which emphasize the main axis of the park. The oval fountain of Apollo is the most beautiful creation in the park. The main sculpture depicts the god Apollo in a chariot drawn by horses, surrounded by tritons and dolphins.

Fig. 1. A view of the park of Versailles [7]

The second part of the park includes the cross-shaped channel, which looks like a forest-park. The decorative vegetation harmonizes with the channel of about 1500 m long and 65 m wide, and the sculptures harmonize perfectly with the vegetation, so that it doesn't influence negatively the perspective [16].

The specialist Liuben I. [16] describes in detail the link between the land relief and the landscape style preferred for arrangement of parks. So, a flat relief, such as a plain, is favourable for arranging parks in regular style. This is explained by the fact that plain landscapes allow drawing straight lines and regular shapes typical of this style and the slight bumps are not an impediment, because the architect is able to use them according to his own imagination. The uneven relief, with slopes, irregularities, on the contrary, is ideal for arranging parks in landscape style. This idea is partially true, so it can be explained the success of the regular style in eastern France, with a

plain relief, and the appearance of the landscape style in China and Japan, countries with an irregular relief.

As we can easily deduce, taking into consideration the relief of the Republic of Moldova and the idea previously exposed by the specialist Liuben I., the local parks were designed and decorated according to the principles of the irregular and mixed styles, but there are also some inclusions of the regular style. This fact was confirmed by the conducted researches, studied field literature, plans and schemes of local parks, field visits.

From the analysis of the schemes and plans of parks that we have today [12, 13, 14], it can be said that in planning the parks Mileşti, Nisporeni; Ivancea, Orhei (fig. 2); Mândâc and Miciurin (Ghica Voda) Drochia; Stolniceni, Edineţ and others, elements of the regular style are used.

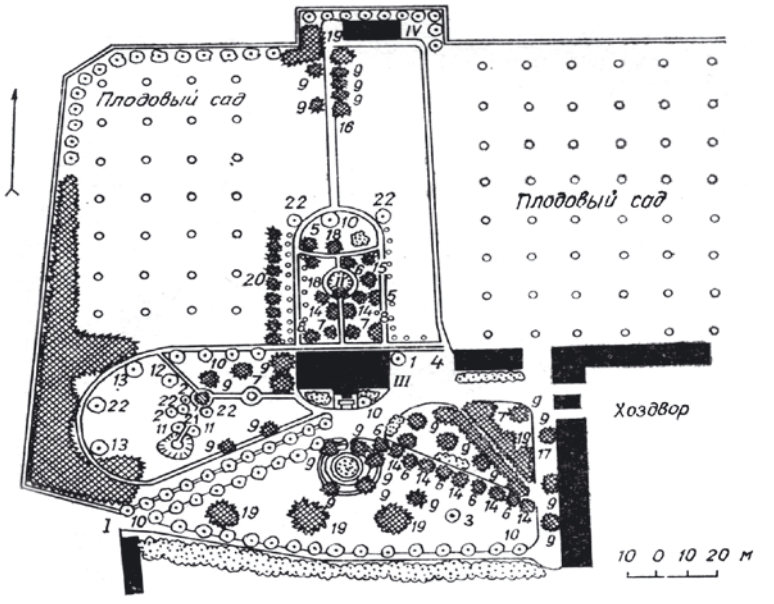

Fig. 2. Plan-scheme of the park Ivancea, v. Ivancea, d. Orhei [12]; I- Entry, II-IV - Construction of the mansion; 1-22 - Plant species.

The irregular (landscape) style. From its name we can deduce that the parks arranged according this style look like natural landscapes or are devoid of the strictness of the straight geometric shapes. The specialists in the field [1, 3, 4, 12, 16] say that typical of the landscape style is the free planning of alleys, but it is possible that some parks have straight portions, free and optical compositional axes having a subordinate role in the composition. The main alleys create compositional centres of the

park or garden and the dominant features of the design, while the adjacent alleys serve to unite the centres of the composition and the secondary alleys [16]. Some examples of parks arranged in landscape style are Kensington Gardens, Kent Park, Royal Botanic Gardens Kew in the United Kingdom, the Petit Trianon in France, Weimar park, the park of the palace Nymfenburg from Germany and others. Parks created in landscape style in our country are Țaul and Rediul-Mare Donduşeni, Pavlovca, Briceni, Pohrebeni, Orhei [13].

Royal Botanic Gardens Kew. It was created in London, United Kingdom. At first it was an exotic park and later it became the Royal Botanical Gardens. Even if the natural conditions of the place were monotonous, the architect succeeded to make them more cheerful using groups of trees as massive compositional forms and planning alleys. The pagoda of 150 m high is the dominant element of the park (Fig. 3); along with cedar trees it forms the Chinese corner. The complex also comprises a rock garden with about 200 species of plants, a rosary, a platform for composting (closed to the public), a Japanese corner and numerous collections of plants: aquatic, carnivorous, bonsai, orchids and others [10].

Fig. 3. Botanic Gardens Kew in winter with the Chinese pagoda rising to the sky and on the right a - glasshouse [10].

The Weimar Park, Germany, is also a model of a park designed in the landscape style. The planning of the park is in harmony with nature, with many deep perspectives and artistic impressions. Unlike the garden of Kew, the park abounds with architectural elements: some ruins, a building in ancient Roman style, a cliff with a portal, sculptures of Goethe and Pushkin, the park has a special emotional charge; being arranged by the writer Goethe, it even has a house on its territory which bears his name. The Ilm River in the northern part of the park is the main element of the composition, which enriches it. The park contains many glades, open spaces, which create deep perspectives [16].

The Hincăuţi Park of the village Hincăuţi, Edineț district, Moldova (fig. 4) is one of the best examples of landscape style in our country. There are no statues or

fountains here. In this park dominates the nature, even architectural buildings are simple [13], resembling the park of Kew. The planning of the alleys in the park is sinuous; they surround the main groups of trees and open areas where the natural grass vegetation grows. For this reason, it is created a unique ring-shaped alley which separates the main glade of the park in several sectors. The wide glades, the open and sunny spaces resemble those from Weimar Park. The park design radiates simplicity and naturalness, its charm comes from the majestic groups of plants and alternation of closed and open landscapes which creates a great comfort in the eyes of the observer [13].

Fig. 4 View of the park Hincăuţi from the village Hincăuţi, Briceni [2].

Mixed style. This style is a compromise between the classic, regular style and the landscape style; it appeared about the end of the eighteenth century and early nineteenth century. [12] Characteristics of this style are straight alleys with wide transversal profiles, able to receive a large number of people, which are used in the central parts of the composition, where the buildings are grouped. In more remote areas the alley becomes more sinuous; the vegetation is arranged in a similar way to what can be found in nature, thus creating quiet corners. In general, the perspective axes are shorter, but it is not suitable for an axis of perspective begun in geometric style to be continued in landscaping style [10]. Some parks arranged in mixed style are: Crystal Palace Park in London, United Kingdom, Monceau park, which contains too many elements and scenes, and Bagatelle park in Paris, France, which is considered one of the best parks in mixed style [5]. In our country, models of parks arranged in mixed style are: the parks of Ivancea, Orhei (fig. 2), Mileşti, district Nisporeni, the parks Mândâc and Miciurin (Ghica Voda) of the district Donduseni.

Bagatelle Park of the suburb of Paris, France, is arranged in mixed style. It belongs to the castle with the same name. The park covers an area of 24 ha. Its fame is brought by the immense rosaries containing over 9000 roses. Every year, the Festival of New Roses is organized (presentation of new species of roses) [11]. The castle is the central point of the park; the entire composition is dominated by it, although this fact is not obvious thanks to the craftsmanship of the creators. The northern part of the park is arranged in landscape style and includes a lake, groups of flowers and two large lawns; the centre includes the parterre arranged in regular style, the terrace along which are old fashioned decorations, the southern part of the composition comprises the rosary, a regular garden in Mauritanian style with fountains, a few hundred years old greenhouse and a vegetable plot. [9] From the above, we can conclude that the park is a set of smaller "gardens", but they all form an entity.

Ivancea park. It was created on the outskirts of the village Ivancea, district Orhei, Moldova, near C. A. Belioz's mansion, built in 1852. It is one of the most representative parks in mixed style. The park occupies a dominant position over the village and the surrounding area, drawing attention due to its unusual construction. An unusual feeling arises because of the multitude of tops of conifers, unusual for this land. Unlike many Bessarabian parks, Ivancea park abounds in romantic fountains like Bagatelle park, the fountains being elements of the landscape style. The park composition is set up of five well-defined sectors; the first three are closely related to the estate and form the festive part of the park, which is also similar to the park of Bagatelle castle. The other two are related to the auxiliary buildings and are simpler. Landscaping was done by straight alleys and sectors, smooth contours, characteristic of the regular parks and gardens, but also with specific elements of the landscape style – artificial hills, fountains and sinuous paths [13].

In some research works [4, 12], the modern gardens with subtypes are mentioned as rural gardens, weekend gardens and special gardens which include rock gardens, rosaries, bog (swamp) gardens and others. However, the contemporary era has given rise to a new individual style specific to the current reality.

Conclusions

1. Three landscape architecture styles: regular (geometric), irregular (landscape) and mixed were studied;

2. In the paper, were presented the main characteristics of every landscape architecture style;

3. Some models of European and Moldavian parks characteristic of each style were analyzed;

4. The regular style is characterized by straight lines, geometric shapes and abundance of statues, fountains and other elements;

5. For the landscape style, sinuous alleys and dominance of natural elements are typical;

6. The mixed style adopted some of the principles of its predecessor styles, being a mixture of geometric and landscape style;

7. The parks in geometric style are characteristic of the plain land, without irregularities, and the landscape and mixed style of parks are more suitable for lands with rugged relief, with slopes, like the relief of the Republic of Moldova.

8. The irregular landscape with slopes and irregularities of the Republic of Moldova, determined the arrangement of parks in mixed and landscape style. Parks in regular style were not created in our country.

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V. SCIENTIFIC CHRONIC

CZU: 57 (478) (092)

ACADEMICIAN VLADIMIR A. RÎBIN 120 YEARS FROM BIRTHDAY

I. Comanici

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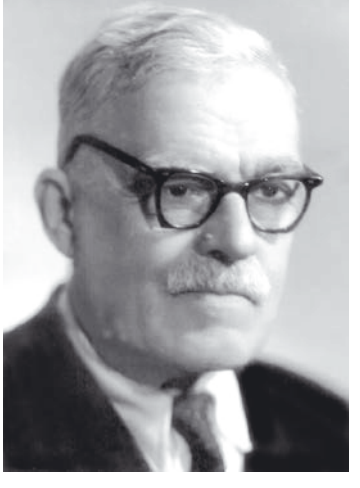

In November 2013, the Botanical Garden (Institute) of the ASM celebrated, with the participation of many scientists from the country, 120 years since the birthday of the Academician Vladimir A. Rîbin, a famous genetic scientist, botanist, disciple and collaborator of acad. N.I. Vavilov, author of the discovery "Provenance of the Domestic Plum".

V. A. Rîbin was born on November 26, 1893, in the city Saratov; he spend his school years in the city Stepanakert, Nagorno-Karabakh; went to secondary school in Tiflis (Tbilisi), and in 1912 entered the University of St. Petersburg, the faculty of natural sciences, speciality "plant physiology".

V. A. Rîbin lived and studied in a tumultuous time full of adversity and difficulties. His parents were forced to move from one city to another. Russia was shaken by historical and social events of great significance - the revolution of 1905-1907, the bourgeois-democratic revolution of February 1917, the World War I, the Civil War, the World War II, the Leningrad blockade and, also, the terror and persecutions of genetics and geneticists under the domination of Lysenkoism in biology.

Because of these events V. A. Rîbin graduated from university in 1922. Meanwhile (1918-1919) he had been working as assistant at the department of anatomy and physiology of the University of Tiflis, and in 1919-1921 – as Lecturer at the department of plant morphology and systematic of the Krasnodar Polytechnic Institute, between the years 1921-1922 – as assistant at the department of anatomy and physiology of the Leningrad Agricultural Institute. In 1923, he was hired at first as a probationer and then as laboratory assistant at the fruit growing and vegetable growing department of the State Institute of Experimental Agronomy, organized this year (1923). In 1924 V. A. Rîbin undergoes a traineeship at the laboratory of cytology of the famous cytologist G.G. Navaşin; in 1925, he transferred at the All-Union Research Institute of Applied Botany and New Crops organized in 1924 on the basis of the former Bureau of Applied Botany and Selection, institute headed by N.I. Vavilov. Here Rîbin V. A. worked as junior assistant at the fruit growing and vegetable growing department headed by Acad. Paşkevici at whose initiative V. A. Rîbin started to study the anatomical structure of the fruits of different varieties of apple (*Malus* Mill.) for the classification of the groups of varieties elaborated by Paşkevici. N.I. Vavilov, the director of the institute, reorganized the work of the department according to his views and the extensive program of multilateral study of crop plants and related forms, concepts which formed the basis of all the activity of the institute. V. A. Rîbin performed the cytological studies. Thus, he performed the cytological analysis of several species of apple (*Malus* Mill.) and established, for the first time in science, the diploid chromosome number $2n=2x^1=34$ at 10 species – *Malus angustifolia* Mich., *M. baccata* (L.) Borkh., *M. sylvestris* (L.) Mill., *M. domestica* Borkh., *M. fusca* Schneid., *M. prattii* C.K. Sneid., *M. prinifolia* (Willd.) Borkh., *M. pumila* Mill., *M. spectabilis* Borkh., *M. zumi* Rehd. He also discovered two tetraploid species (2n = 4x = 68) - *M. sargentii* Rehd. and *M. toringo* Sieb. Further, the cytological study of the seminal generation from the cross pollination of the varieties *"Parmen auriu de iarnă"* and *"Renet de hârtie"* revealed the triploid forms, and studying the meiosis of several cultivars, discovered a triploid variety – *"Renet de Canada"* and also explained the cause of the sterility of the pollen of the given variety.

These scientific data obtained by V. A. Rîbin constitute the main element of the cytogenetics of apple, a very important fruit crop.

Thus, beginning his scientific work under the guidance of acad. N.I. Vavilov on the theoretical bases of plant selection, V.A. Rîbin continued the researches in this scientific field his whole life, especially concerning the distant hybridization, allopolyploidy, evolution and selection. As part of the cytological research of the interspecific hybrids of tobacco (*Nicotiana tabacum* L.) and makhorka (*N. rustica* L.), V.A. Rîbin developed a method of creating autotetraploid forms. Further, the scientist used, among the first in the former Soviet Union, colchicine in artificial obtaining of autotetraploid forms of potato, flax, hemp, sunflower, broad bean (*Vicia faba*).

In 1929 V. A. Rîbin was confirmed as assistant, and in 1931 was appointed researcher and active member of the All-Union Institute of Plant Industry (ВИР) and until 1941 was a researcher in the laboratory of cytology of the department of physiology and head of the department of physiology of the generative processes. V. A. Rîbin carried out some researches on the karyological analysis of wild and cultivated species of potato; these researches formed the basis of the systematics of section *Tuberosum* of the genus *Solanum*. In honour of his scientific merit in this domain, one potato species was named *Solanum rybinii*. The great scientific discovery made by acad. V. A. Rîbin, that made him world renowned, was the *"resynthesis of the domestic plum"* – that is, the *artificial synthesis* of the species *P. domestica* L. In 1930, in London, it was held the Ninth International Congress of Horticulture, which was attended by Acad. N.I. Vavilov. At this congress, the British scientists M. B. Crane and W. J. C. Lawrence advanced the hypothesis that *P. domestica* L. would come from the spontaneous crossing of the species *Prunus cerasifera* Ehrh. (diploid 2n = 16) and *Prunus spinosa* L. (tetraploid 2n = 4x = 32). After returning from the congress, N. I. Vavilov thoroughly examined this issue with V. A. Rîbin and entrusted him to verify experimentally the hypothesis of Crane and Lawrence. Rîbin went immediately to Shuntuk not far from Maikop, where the "subdivision" of the All-Union Institute of Plant Industry (ВИР) had been recently organized. Here in the surroundings of the village Shuntuk grew together the cherry plum (*P. cerasifera*) and the blackthorn (*P. spinosa*) in forest stands and, therefore, there were favourable conditions for spontaneous hybridization. Researching these stands, V. A. Rîbin discovered some very low fruitful specimens which proved to be nothing else than triploid spontaneous hybrids between the two species – cherry plum and blackthorn (2n=3x=24). For artificial crosses, 6 trees of cherry plum and 16 - of blackthorn were chosen. In the spring of 1933, V. A. Rîbin performed crosses in different combinations of blackthorn and cherry plum and obtained 442 fruits (seeds) from which in the spring of 1934 grew 16 hybrid seedlings. The cytological analysis showed that 15 of them had by 2n=3x=24 chromosomes and one of the combination of blackthorn x cherry plum proved to be alohexaploid with 2n=6x=48 chromosomes, like *Prunus domestica* L. (Fig. 1). In this way, it was synthesized artificially the common plum (syn.: garden plum, domestic plum) *Prunus domestica* L. and experimentally confirmed the hypothesis of the scientists M.B. Crane and W.J.C. Lawrence about the origin of the species *P. domestica,* by spontaneous crossing between cherry plum and blackthorn; the hypothesis, being verified and confirmed experimentally, became *theory*, namely *the theory of the genesis of species* based on allopolyploidy. V. A. Rîbin named the newly artificially synthesized plum the *Prunus domart* (*P. domestica artificialis*). Acad. N. I. Vavilov ardently supported these works. The results were published in - Рыбин В.А. Опыт синтеза культурной сливы из родственных ей диких видов. in: Социалистическое растениеводство, 1935, 15; Рыбин В.А. Гибриды тёрна и

алычи и проблема происхождения культурной сливы. in: Тр. по прикладной ботанике, генетике и селекции, 1936, сер. 2, 10, с. 1-46. The news about the artificial synthesis of the common plum quickly spread in the specialized scientific community. This work was published in textbooks, in important editions, everywhere where *P. domestica* L. is mentioned. V.A. Rîbin is quoted by renowned scientists: M. Schmidt (1939), F. Kobel (1954), W. Chendler (1960), J. Endlich & H. Murawski (1962), M. Zwintzscher (1962); In: Pomologia României. 1965, vol. 4; In: Prunul. 1965 etc. The All-Union Institute of Plant Industry (ВИР) recorded important results internationally recognized. It seemed that the good times had been established. But, as we shall see below, it was not to be. As writes Acad. V. A. Rîbin himself, at first, the work on the synthesis of plum from the assumed related species aimed at solving a theoretical problem - *the genesis of the garden plum* (*P. domestica* L.). This purpose, as we mentioned, was achieved successfully by V.A. Rîbin. Further, the task of the scientist was to solve the practical problem – the transmission to the cultivated varieties of the frost resistance and of the immunity of their wild related species, using *the method of synthesis of amphidiploids*. In this regard, V. A. Rîbin took into account the experiences of R. Wellington, M. Crane and W. Lawrence etc. which had shown that the crosses between the varieties of *P. domestica* L. had not influenced significantly the frost resistance. Therefore it would be reasonable to apply the *method of synthesis of amphidiploids* and their crossing with the cultivated varieties. For this purpose, V. A. Rîbin chose to cross with P. spinosa L. (2n = 32) three diploid species (2n = 16) that are very resistant to winter frosts – the Canadian plum (*P. nigra*), American plum (*P. americana*) and Ussurian plum (*P. ussuriensis*), the latter being resistant even to temperatures of about - 55 ° C. In 1938 V. A. Rîbin managed to obtain a triploid hybrid 2n=3x=24 at the crossing of the blackthorn x Ussurian plum which, as was expected, in the F1 was sterile and its number of chromosomes had to be doubled in order to become fertile (2n=6x=48). But unfortunately, these wonderful works were interrupted because of the terror and the persecution of genetics and geneticists which took place in the All-Union Institute of Plant Industry (ВИР) and not only. The creative enthusiasm inspired by N. I. Vavilov and the astounding results obtained in the institute could not but disturb the envious, ignorant, careerists greedy for power. The fact that the "Institute of post-graduate education" of the Lenin All-Union Academy of Agricultural Sciences (ВАСХНИЛ), transferred in 1930 to the ВИР Institute, where the requirements were much higher, was inconvenient to some haughty and full of self-conceit young people. In this way, a certain opposition was formed in the institute. The signs of negative attitude towards N.I. Vavilov intensified, but the condition of maximum tension in the institute was created with the appearance of Trofim Lysenko – a sinister, very energetic man, very haughty, half-learned, endowed with the spirit of self-advertisement. Making an alliance with the scientist Prezent, Lysenko started fighting genetics and geneticists. It was declared that the genetic laws were reactionary

and that the geneticists hold reactionary, anti-scientific views. Lîsenko promoted strongly, by unscientific methods, new principles in genetics: the gene variability, the change of the gene in the determined direction, the possibility of directed mutations, the inheritance of acquired characteristics, the appropriate modifications, denial of the role of chromosomes etc. In this way, the "Michurinist biology" was "created". Lysenko, denying the role of chromosomes, said: "The heredity is possessed by everything living, every cell, every particle of the body, and not just the chromosomes, even juices are endowed with hereditary traits" * (the concluding remarks of acad. T. Lysenko at the Session of Lenin All-Union Academy of Agricultural Sciences (ВАСХНИЛ) of 31.VII-7.VIII. 1948). N.I. Vavilov, at the IV session of Lenin All-Union Academy of Agricultural Sciences (ВАСХНИЛ) from 19-27 of December, 1936, disproved these "principles" of the Lysenkoists, showing that the possibility of directed mutations and appropriate modifications, inheritance of acquired characteristics had not been proven experimentally by anyone and were in flagrant contradiction with the ideas and concepts of the modern genetics. "In order to reject the conceptions engraved in the geneticists' consciousness, we need precise experiments. We do not have such experiments." (N.I. Vavilov. Concluding remarks at the session). After the IV Session of the Lenin All-Union Academy of Agricultural Sciences (ВАСХНИЛ), a more difficult period started. The financing of The All-Union Institute of Plant Industry (ВИР) for 1937 was reduced; the publication the world-famous magazine "Труды по прикладной ботанике, генетике и селекции" stopped. While N.I. Vavilov and his colleagues worked with scientific methods, T. Lysenko and his supporters used administrative means, blackmail, denunciations. In 1938, T. Lysenko became President of the Lenin All-Union Academy of Agricultural Sciences (ВАСХНИЛ), The All-Union Institute of Plant Industry (ВИР) being subordinated to the Lenin All-Union Academy of Agricultural Sciences (ВАСХНИЛ). After all the successes achieved by The All-Union Institute of Plant Industry (ВИР) under the leadership of N.I. Vavilov, the report for 1938 presented at the meeting of the presidium of the Lenin All-Union Academy of Agricultural Sciences by N.I. Vavilov was not approved by T. Lysenko. In 1939, during a reception in Kremlin, T. Lysenko managed to provoke the discontent of I.V. Stalin towards N.I. Vavilov. The fate of N.I. Vavilov was predetermined. On the 6th of August 1940, during his expedition in the Western Ukraine, N.I. Vavilov was arrested, charged with high treason and sentenced to death. Many geneticists, followers of N.I. Vavilov, were forced to leave the institute. V. A. Rîbin transferred to the Peterhof Institute of Fruit and Vegetable, and in 1942, during the blockade of Leningrad; he was evacuated to Perm together with the Institute of Fruit and Vegetable. In 1946, he returned at the All-Union Institute of Plant Industry (ВИР), as senior scientific assistant in the department of anatomy and cytology, but in January 1948 he was forced again to leave the All-Union Institute of Plant Industry (ВИР) and transferred to the Main Botanical

Garden of the Academy of Sciences of U.S.S.R. in Moscow, but after the "famous" session of the Lenin All-Union Academy of Agricultural Sciences (ВАСХНИЛ) in August 1948, he was blamed for Morganism by a certain Teterev and dismissed from his post. He found a job as many other geneticists, cytologists in the remote areas of the country, for example, C.I. Pongalo in Tiraspol, V. A. Rîbin in Crimea, at the Branch of A.S. USSR, where he led the sector of botany and phytotechnology. Here, he had to study the lemon in the so-called trench crops also with "geographical sowings of tea and eucalyptus" that some scientists attempted to introduce in the region. In Crimea, V. A. Rîbin restarted informally to study the resynthesis of the plum. During these works, V. A. Rîbin established that the resynthesized plum did not cross with the species – cherry plum and blackthorn from which it originates, but it crossed easily with the cultivated varieties of plum and creates a hexaploid generation. In Simferopol, V. A. Rîbin has brought the 8 seedlings of such hybrids. In July 1956, V. A. Rîbin came in Moldova to work at the Botanical Garden of ASM. In order to continue his work, V. A. Rîbin has brought the 8 hexaploid hybrid seedlings mentioned above and the allotriploid hybrid, blackthorn x ussurian plum (2n=3x=24), sterile, obtained in 1938, which had been kept in the Pushkin town (subordinate to Leningrad) in the phytotechnology laboratory. This hybrid - with the participation of the Ussurian plum, the most resistant to frost - was particularly interesting. V. A. Rîbin tried many methods and procedures of processing with colchicine in order to double its number of chromosomes and to turn it into a fertile hexaploid hybrid. All these actions proved to be in vain. Then, V. A. Rîbin grafted the vigorous cherry plum in the Botanical Garden, on the old land. The calculation was: according to the probability theory, from thousands of flowers (the rootstock being vigorous) at least one fruit must be formed. Indeed, in 1959, the hybrid grafted on the vigorous cherry plum was blooming very abundantly, like the blackthorn and formed six fruits (seeds). In 1961 Prof. V. A. Rîbin organized on the basis of the cytology group the laboratory of distant hybridization of plants and soon became the head of the laboratory. From the 6 seeds collected from the allotriploid hybrid blackthorn x ussurian plum (2n=3x=24), six seedlings were grown. Two of these seedlings proved to be hexaploid (2n=6x=48). One of them manifested heterosis and in the 3rd year of vegetation (1962) blossomed (Fig. 4) and formed 25 fruits. In the second year of fruiting, that is, at the age of 4 years (1963), the hexaploid hybrid of blackthorn x Ussurian plum was pollinated (after isolation and castration) with the pollen of the variety violet Reine Claudes and fructified well. On the free branches, the fruitage was abundant. So, it turned out that the allohexaploid plum artificially obtained easily crosses with the cultivated varieties of *Prunus domestica* L. and the trispecific hybrids inherit the characteristics of the cultivated plants and some of the precious characteristics of the wild species. One of the trispecific hybrids yielded 56 kg of qualitative fruits. So it was practically demonstrated that the method of synthesis of amphidiploids could be successfully applied in selection.

V. A. Rîbin was completely devoted to science. Despite the problems during the tumultuous time full of revolutions, wars, terror and persecutions in which he lived, he was able to contribute efficiently to the study of the plant biology. During his professional activity, V. A. Rîbin was consistent with the scientific principles of N.I. Vavilov and the scientific truth. He was severe and demanding towards the research methods and towards the materials for publication. He permitted to publish only the information that was relevant for science. V. A. Rîbin published relatively few - 63 scientific works, including 2 methodical monographs on cytology and one brochure on nut-tree grafting because he was demanding towards himself. He had no aspirations or occupations except the science. Knowing German, English, French, to a certain extent, Latin, V. A. Rîbin translated several monographs: "The effects of cross-pollination and self-pollination" by Ch Darwin, "The selection of fruit trees" by N. Hansen, "The genetics of fruit and vegetable plants" by M. Crane and Lawrence W. (1936), "Fruit growing on physiological bases" by F. Kobel (1935, 1954), "Selection of rootstocks of fruit plants" by H. Tydeman. V. A. Rîbin was an honest, tactful and intelligent person, being educated in a family of intellectuals, his father was a judge, and his mother, Olga Speranskaia, originating from a family of doctors, was a learned woman, worked in scientific institutions, studied the transpiration in plants. In different periods of his activity, V. A. Rîbin had the happy opportunity to work under the leadership of the academicians – N. I. Vavilov, S. G. Navashin, N. A. Maximov, V.V. Pashkevici and with the world-class scientists G. D. Karpechenko, G. A. Levitskii, S.V. Bukashov etc. V. A. Rîbin, as we knew him, was a learned man with good manners, some said - aristocratic, he was modest, benevolent, always willing to share his vast knowledge; he had a spiritual and understanding attitude towards us, but allowed no familiarities. V. A. Rîbin died in 1979. He was buried at the Central Cemetery in Chişinău. In 1954, for scientific achievements, V. A. Rîbin was awarded the Order of Lenin, in 1965, he was elected academician of the ASM. We mention that one year after the official condemnation, the lysenkoism still was present in the ASM, so the candidacy of V. A. Rîbin for membership was not submitted, but at the last moment, the "submission" came from three academicians of the A.S. of the U.R.S.S. and then V. A. Rîbin was elected academician without going the step of corresponding member, like many others. As sign of gratitude and appreciation of his scientific merit, the bas-relief of Acad. V. A. Rîbin is placed at the entrance façade of the central block of the Botanical Garden of ASM.

TO 110 YEARS FROM THE BIRTHDAY OF THE ILLUSTRIOUS SWEDISH CYTOGENETIST, PROFESSOR ARNE MÜNTZING

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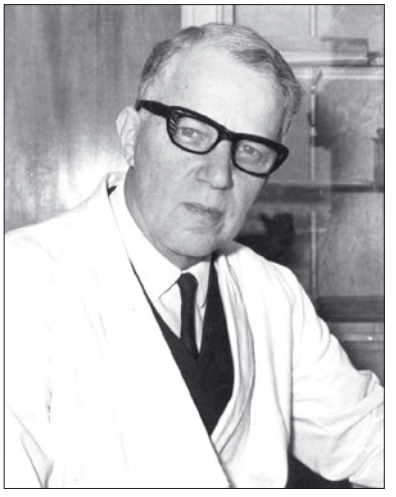

Forty-five years have passed since, for the first time, I met Professor Arne Müntzing, Director of the Institute of Genetics of University of Lund, Sweden. I still keep in my mind the impression which remained after our first meeting. He was an outstanding scientist, one of the most talented geneticists from the well known pleiad of Swedish scientists (H. Nilsson-Ehle, Å. Gustafsson, A. Levan, E. Åkerberg, A. Lundqvist, A. Hagberg, etc.).

Arne Müntzing was born on March 3, 1903, in the city of Göteborg (Sweden), where he graduated from primary school. He studied at university in Lund (1926) and obtained the scientific degree of Candidate of Philosophy (1929) and later - Doctor of Philosophy (1931). He was working as an assistant from 1925 till 1929, and in 1930-1938 was employed as Associate Professor at the Institute of Genetics of Lund. Since 1938, he cumulates the post of Director of the Institute of Genetics and of Professor. During his activity as Associate Professor, he gave lectures on cultivation of plants at the Station of sugar beet selection in Hilleshög (1929-1931); during 1931-1938 he worked as head of the Department of Cytogenetics of the Swedish Seed-Growing Union in Svalöv. Arne Müntzing was a member and vice president of the Swedish Research Council for Natural Sciences, and in 1946 - Member of Forest Research Foundation. In 1949 he was elected member of the State Institute of Human Genetics in Uppsala. During 1945-1954 he was a member and vice president of the Board of Directors of the Institute Alnarp (Institute of Agriculture and Gardening), and

from 1948 till 1950 - Member of the Institute of Animal Husbandry of Wiad. From 1929 till 1935, he was working as Secretary of the Department of the Union of Development of Scandinavian Researches in Agriculture. In the years 1936-1937, he was working as Secretary of the "Mendel" Society, in 1938 - vice president and from 1939 - president of this society.

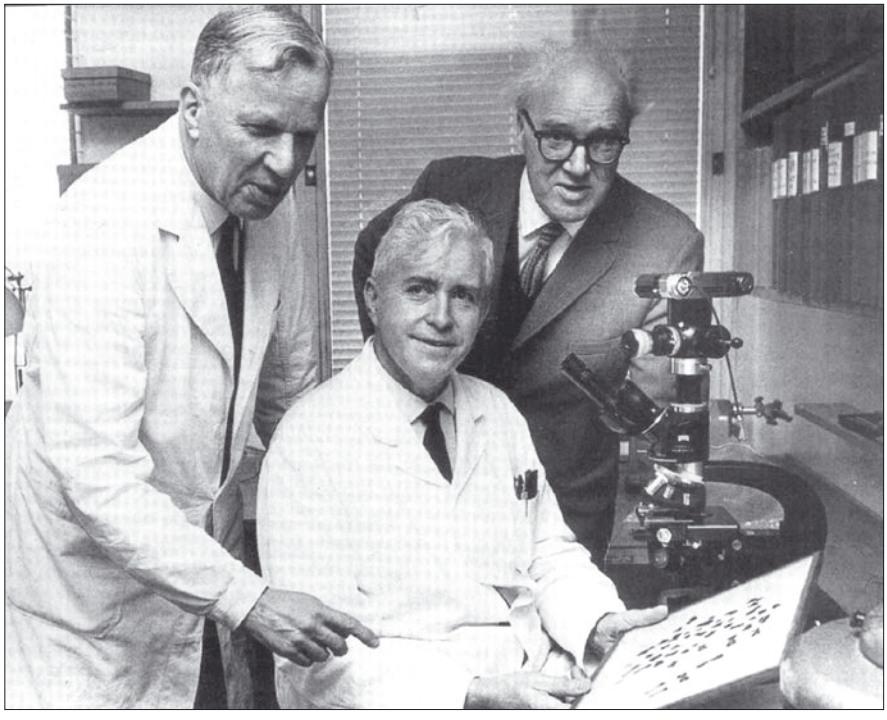

Fig. 2. Arne Müntzing, Albert Levan, Åke Gustafsson. ***In the center***: Professor Albert Levan - author of identification of chromosome karyotype

In 1948 he was vice president of the Organizing Committee of the Eighth International Congress of Genetics in Stockholm and archivist at the Genetics Section at the Eighth International Botanical Congress (Stockholm, 1950) and member of its Organizing Committee. In 1948-1954, he was working as president of the Chamber Music Society and of Symphony Orchestra Society of Lund.

He made study and research trips in Germany, Finland, England, Denmark, USA, where he worked as a research associate in Genetics Science over the years 1933-1934 (at the University of California, he collaborated with E.B. Babcock and Roy Clausen). He gave lectures at various American universities. He travelled repeatedly in Ecuador and other countries from South America, West Indies, with the purpose of researching the cultivation of the cacao tree.

He participated in several international conferences in the field of genetics and botany. He lectured on genetics and botany. He lectured: In 1937, at the International Scientific Congress in Biology (Berlin-Dahlem), at the Universities of Amsterdam, Utrecht, Vageningen, Wiirsburg, in 1939 – at Oslo University and the Institute of Agriculture of AS (Norway), in 1945 - at a conference in London which was organized by the Genetics Society of Great Britain, in 1946 - at Columbia University in New York, in 1948 - Agricultural Institute of Copenhagen, in 1950 – at the "Golden Jubilee of Genetics" in Columbus and Ohio . In 1951 he was invited by the Indian Government and the Indian Scientific Congress to participate in this conference and to lecture as a guest at several universities in India. In 1935 he lectured in England and Wales at the invitation of the British Council and in the same year – in Helsingfors, at the invitation of the local university. In 1956 he attended the International Symposia of Genetics in Tokyo and lectured at various Japanese universities. In 1958, at the invitation of the Genetics Society of Canada, in connection with the International Congress of Genetics in Montreal, he gave a lecture in memory of Huskin. Arne Müntzing was one of the vice presidents at this congress. In 1959, he gave a lecture at an annual meeting, referring to some aspects of the work of Ch Darwin. He also lectured at the universities of Texas, Austin, New York. In the same year, he lectured (as a member of the Delegation of Lund University) in Moscow and Leningrad.

He was elected member of the following academies - Academy of Sciences of Helsinki, Danish Academy of Sciences, Copenhagen, from 1936, he is a member of German Academy of Natural Research Leopoldina. He was also Honorary Member of the Botanical Society of Edinburgh and of the Genetic Science Society of Japan. He published more than 250 scientific papers and a compendium of genetics (published in Swedish in 1953, in German in 1957, in English in 1961).

One of the chief works – "Genetics: Basic and Applied" was published in translation in Russian (Изд-во М.: Мир, 1967). Many years Arne Müntzing was chief editor of the journal "Hereditas" which was edited by the Scandinavian countries in English and had great authority on all continents.

Is significant Arne Müntzing's addressing to the young generation of geneticists from the former Soviet Union, included in the Introduction to the latest edition of the manual of "Genetics" published in Russian (1967): "I wish you success and I am confident that your work in this direction, the key to contemporary biology, will be fruitful. A lot of your predecessors in your country were really outstanding geneticists and these great traditions of the past will guarantee your future success." The geneticists of the Institute with the selectionists from the Svalöv Station (Institute) created well-known varieties of winter wheat (Odin, Diana, Elve, Diamond, etc.), winter rye (Kungs-II, Varne - tetraploid rye), oat (Sull-II, Susbru, Blenda, Linda, Same, Tutys etc.), barley (Pallas, Mary, Edda-II), winter rape, alfalfa (Timofeevca, meadow fescue, orchard grass).

By the thirties of last century, at the Sector of Genetics (Head, Arne Müntzing) of the Svalöv Station started the first experiences in creating polyploid forms, continued later by Prof. A. Levan, E. Åkerberg, U. Lundquist, A. Hagberg.

Arne Müntzing with his colleagues A. Levan, E. Åkerberg, U. Lundquist, A. Hagberg studied the phenomenon of the instability of the triploid level and the low fertility of tetraploids. As a result of the investigations, were highlighted the polyploid forms of rye, red clover, kale, fodder beet, turnip and other crops.

Arne Müntzing initiated the cytogenetic researches of amphidiploid wheat x rye hybrids (triticale), which laid the foundations of the creation of amphidiploids and allopolydiploids of cereal crops. In 1967-1978, in the Cytogenetics Sector, led by Professor Arne Müntzing, was investigated the role of β-chromosomes (supernumerary chromosomes) of the species of the genera: *Secale, Poa, Triticum, Hordeum*. The problem of inbreeding (Inzucht) and apomixis was also investigated.

The focus was on the development of research methods of metabolic DNA in cancerigenic tissues chromosomes; cancerigenic cells were researched using cytogenetic and biochemical methods, the method of autoradiography with labeled atoms. It can be mentioned that the Institute of Genetics, in those years, was funded by the Rockefeller Foundation (USA) which was concerned with the cancer problem. In the Institute of Genetics of Lund, for the first time, in 1956, Prof. Albert Levan determined the number of chromosomes in the human karyotype.

Already working as director of the Botanical Garden of the Academy of Sciences of Moldova, and, at the same time, chief of the laboratory of cyto-embryology, in September 1967 we received a confirmation of our request from Professor Arne Müntzing to work as a trainee researcher at the Institute of Genetics.

My training course at the Institute of Genetics of the University of Lund was determined by the fact that the researches conducted by Professor Arne Müntzing – well-known geneticist and cytogeneticist – were related to the studies of crop plants from the point of view of cyto-embryology or karyology. The researches carried out by A. Müntzing and the collaborators of the institute, published in the popular magazine "Hereditas", raised the investigations of this institution at the European level.

I flew in Stockholm from Moscow by airplane TU-104 and I went from there by train to Lund (route Stockholm - Malmö). At the railway platform I was met by the Director of Genetics Institute - Arne Müntzing and Dr. T. Nurdqvist – my future scientific adviser. The first days I lived in "Grand Hotel" and then I was transferred, at our request, at Ph.D. candidates' hall of residence.

At the first meeting Prof. A. Müntzing asked if I am not a relative of Maria Cibotari, whom he knew personally – I learned from him a lot about her artistic biography – and whom he considered the most famous soprano from the first half of the twentieth century. He also knew about her tragic fate.

It was written in detail about the results achieved by Professor Arne Müntzing and the collaborators of the Institute of Genetics in the "report" presented by us, after finishing our training, at the Ministry of Foreign Affairs of the USSR (Moscow, 1968, 60 p.), and in the article „В гостях у шведских генетиков" (Ж., Генетика, Москва,1968, 4, 9).

After World War II and early ' 70, Swedish geneticists (A. Müntzing, Å. Gustafsson, A. Levan, E. Åkerberg, A. Lundquist, A. Hagberg, etc.) widely applied the methods of polyploidy and mutagenesis to a number of crop plants and studied the consequences, such as - the appearance of sterile and triploid forms, decreased fertility, the appearance of some anomalies etc. They also demonstrated that the doubling of the number of somatic chromosomes caused some cytogenetic changes in mitosis and meiosis. Finally, this research resulted in obtaining some tetraploid forms of rye, red clover, Jerusalem artichoke (turnip).

I remember that the meeting with Prof. A. Müntzing went with much esteem and attention when, the day after arrival, was reviewed the work program which included electron-microscopic research of the reproductive sphere of different subspecies of maize. The cytogenetic researches on meiosis, pachytene chromosomes, their chromomeric structure and organization, and the creation of cytological maps constituted a separate chapter. It can be mentioned that at the Institute of Genetics of Lund, in the late 50 ', A. Müntzing and A. Lima-de-Faria conducted the first researches on the chromomeric structure of pachytene chromosomes of rye.

On the same day (!) was resolved the problem of buying the materials, requisites and tools necessary for the application of electron microscopy methods, growing experimental material (in the greenhouses of the Institute), preparation of biological objects for electron microscopy, documentation, analysis of data, etc. (3 days all the ordered materials: tweezers, scalpels, grids, boxes for grids, photo-materials, reagents, clamps, special glass for microtome knives etc. were brought and placed at my disposal). Every day, I met Professor A. Müntzing at lunch, which the collaborators of the Institute take strictly at 12^{00} o'clock in the library of the Institute. Every time he asked how the research was going at the institute, he was interested in the progress of my work, asked if I didn't lack anything and he didn't forget to add that he was always ready to listen and help me whenever I needed.

I want to tell about one more fact. From the first day after my arrival in Lund, the director personally gave me three keys - to the entry to the institute, to the office of electronic microscopy and to the library of the institute. A significant fact was that the electron microscope (of Japanese production) was installed and tested 2-3 days before my coming to Lund. The first and subsequent electronographies with the electron microscope Hitachi 7 were made by us. I wrote about the achieved results in the monograph - "Эмбриология кукурузы" (Кишинев: Штиинца, 1972. 384 с.). Before

I took the train to Stockholm, I returned the keys to the above mentioned offices. At my departure, Prof. A. Müntzing, T. Nurdqwist and some collaborators who had become my friends led me to the railway station.

In the second year after our departure from Lund (1968) Prof. A. Müntzing, according to the age requirements, passed the post of Director of the Institute of Genetics of Lund to Professor Åke Gustafsson who had hold the post of director of the Institute of Forestry of Stockholm before that. I remember with great pleasure that at my first visit at the Swedish Academy (where I was awarded the Silver Medal Carl Linné, in connection with the celebration of 260 years from his birth) and the profile institute, Å. Gustafsson made me acquainted with the Climatronic researches of the Institute of Forestry of Stockholm.

By the ' 70 - ' 80, A. Müntzing published some scientific generalizations, I would say philosophical generalizations, stated in various lectures and communications on evolutionary genetics: ”The Stream of Life” – lecture given at the Conferment of Degrees, University of Lund, Sweden, May 22-d, 1968; ”Genetic signals in the evolution of organisms”. Print from “Biological Signals”, published by Kungl. Fysiografiska Sallsapet of Lund, Sweden, 1975.

Time flies. So many years have passed. As a delegate or guest, I attended several congresses and international scientific symposia. I remain loyal to the opinion - Sweden appears to be unique among the countries which can take pride in so many outstanding personalities in various fields. The portraits studded inside the hall of the Grand Assembly of the Royal Academy of Stockholm, which I will keep in my memory all my life, testify about Swedes’ care and solicitude for their great predecessors.

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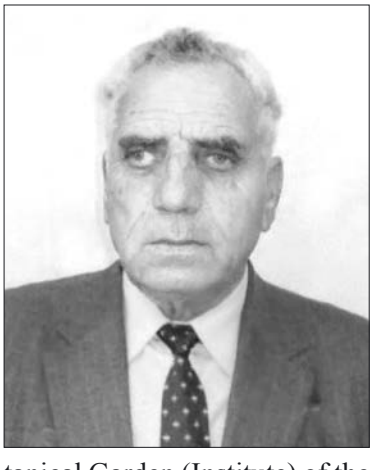
DR. HAB. VICTOR SAVA

(75th birthday and 50 years of scientific and educational work)



Victor SAVA was born on May 15, 1938 v. Cojuşna, d. Chişinău (now Străşeni). In 1959, he graduated from the College of Culture in Soroca and in 1964 graduated from the Faculty of Biology and Geography of the Tiraspol State Pedagogical Institute.

After graduating from the institute, he worked as biology and geography teacher in the General Education School in v. Recea, d. Străşeni.

During 1967-1970 he was studying for doctor's degree at the Botanical Garden (Institute) of the Academy of Sciences of Moldova. During his research activity, he advanced from junior researcher to head of the Laboratory of Floriculture of the Botanical Garden (Institute) of the Academy of Sciences of Moldova; currently he works as senior researcher of the same laboratory.

In 1972, he defended his doctoral thesis on the theme "*Влияние условий выращивания на морфологические признаки и семенную продуктивность декоративных растений"* (*Influence of growth conditions on the morphological characteristics and seed production of ornamental plants*).

As a result of the appreciation of the successful introduction of annual ornamental plants of diverse phytogeographical origin in the Republic of Moldova, he wrote "The range of ornamental plants" for landscaping urban and rural areas (1990).

In 1991, he defended the habilitation thesis in biology on the theme: "*Биологические основы интродукции однолетних декоративных растений в Республике Молдова*" (*Biological basis of the introduction of annual ornamental plants in the Republic of Moldova*).

He has a broad experience in ornamental plant breeding and their introduction into the national economy, having obtained 82 inventions and patents for new plant varieties: China aster**,** chrysanthemums, canna, peonies, dahlias and yellow lilies.

As a result of the introduction and breeding of ornamental plants in the Republic of Moldova, he created, at the Botanical Garden (Institute) of the ASM, the collection of annual ornamental plants with a wide range of uses: decorative flowering, decorative foliage mosaic, lianas, etc. He proposed technologies of production of planting material and seeds for about 40 species of introduced flowering plants.

In the period from 1998 until 2010, he continued the educational work at the higher education institutions: Institute of Real Sciences MSU, Free International University of Moldova, and State Agrarian University of Moldova with the course "Floriculture". He was scientific advisor to two PhD theses in biology and to 30 diploma papers.

Now, he is a member: of the Profile Scientific Seminary of the Botanical Garden (Institute) of the ASM, of the Specialized Scientific Council for defense of the doctoral and habilitation theses of the specialty "03.00.05-*botany*", of the Council for Seed Production of the Institute of Genetics and Plant Physiology of the ASM.

The scientific results of his researches have been published in about 400 scientific papers, including 10 monographs, two textbooks, two popular scientific books, 20 booklets.

In 2001, he was awarded the state decoration "Om Emerit" (Man Emeritus), in 2007 and 2010 was appreciated as the best university professor, in 2008 was awarded the scientific-educational title of "university professor" and in 2010 was awarded the medal "Dimitrie Cantemir" of the Academy of Sciences of Moldova.

DR. VALENTINA ŢÎMBALÎ

(The 60th birthday)

Valentina Ţîmbalî was born on July 15, 1953, v. Olăneşti, Ştefan-Vodă. In 1970, she graduated from the school in her native village and then continued her studies at the Faculty of Biology and Chemistry of the Tiraspol State Pedagogical Institute, from which she graduated in 1977 with a distinction. She started to work at the Botanical Garden of ASM immediately after graduation. During 1979-1982 she was studying for the doctor's degree at the ASM.

After getting a Ph.D., she was hired as a junior researcher in the Laboratory of Tropical Plants of the Botanical Garden (Institute) of ASM and she climbed the career ladder until she became the head of this laboratory. Since 1991 until now, she has been working as head of the Laboratory of Tropical Plants of the Botanical Garden of ASM.

Her doctoral thesis on the theme: "*Биология развития и особенности выращивания бромелиевых, перспективных для озеленения интерьеров" (Developmental biology and peculiarities of growing bromeliads promising for interior landscaping)* was successfully defended in 1985 and was created a collection of plants from the Bromeliaceae family, which contained about 130 taxa.

As a result of the researches, the developmental biology and the growth peculiarities of bromeliaceae were studied. It was developed an assortment of plants, used for the decoration of different rooms.

During the scientific work, she contributed and still contributes to the development of the researches on the breeding, study and introduction of plants on protected land in the Republic of Moldova.

The scientific results of the researches conducted by her were published in more than 80 scientific papers, including the monograph "*Тропические и субтропические растения в оранжереях Ботанического сада АН МССР"*, (*Tropical and subtropical plants in the greenhouses of the Botanical Garden of AS of the MSSR*) (1985);

"*Бромелиевые (биология, интродукция, агротехника*)" (*Bromeliads (biology, introduction, agrotechnics*)), (1989).

She actively contributes to the environmental education of population, especially of young people, both by lessons-excursions to the collections of tropical, subtropical and succulent plants and by speeches at the national radio and television channels.

For the results achieved, the creation and maintenance of the collections of plants on protected land over about 35 years in the Botanical Garden of ASM, Dr. Valentina Ţîmbalî was awarded the honorary title of "Om Emerit" and diplomas of the Academy of Sciences of Moldova.

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