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BIOLOGIE VEGETALĂ

BIOLOGIE ANIMALĂ

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THE BIODIVERSITY OF CERAMBICIDS (INSECTA, COLEOPTERA, CERAMBYCIDAE), FROM DOFTEANA DENDROLOGICAL PARK-NEMIRA MOUNTAINS

Gabriela GURĂU*

Key words. cerambicids, biodiversity, Simpson biodiversity index, Shannon-Wiener biodiversity index, theoretical diversity, equitability.

Rezumat. Lucrarea prezintă rezultatele cercetărilor pe care le-am efectuat în anii 2001-2002 în Parcul Dendrologic Dofteana, care este situat în "Rezervaţia Naturală Nemira". Parcul Dendrologic Dofteana, este situat în nord-estul Munţilor Nemira, suprafaţa sa este de 34 ha (din care 24 ha sunt ocupate de arboretum) iar zona sa centrală are o importantă colecţie de plante exotice.

Pentru o analiză a faunei de cerambicide din Parcul Dendrologic Dofteana din punct de vedere al biodiversităţii, am calculat indicii de diversitate Simpson şi Shannon-Wiener, diversitatea teoretică şi echitabilitatea.

Numărul de exemplare colectate este 173, grupate în 11 specii, 6 genuri şi două subfamilii ale familiei *Cerambycidae (Cerambycinae şi Lamiinae).*

Valorile indicilor de diversitate calculaţi pentru cele 11 specii colectate în staţia Parcul Dendrologic Dofteana sunt următoarele: indicele de diversitate Simpson D = 3,35, indicele de diversitate Shannon-Wiener H (S) = 2,24, valoare căreia conform tabelului nr. III îi corespunde valoarea 7 (numărul teoretic de specii din biocenoza analizată). Diversitatea teoretică este H(S)max = 3,46. Echitabilitatea din această staţie de colectare este 63%.

Introduction

In the paper, we present the results of our studies made in Nemira Mountains between 2001-2002 as part of the doctoral thesis "The biodiversity of cerambicids (*Coleoptera, Cerambycidae*) from Nemira Mountains and their ecological significance". The studied area is part of Nemira Mountains and is named Dofteana Dendrological Park (figure no.1). For a more accurate analysis of the fauna of cerambicids from Nemira Mountains, from the biodiversity point of view, we calculated Simpson and Shannon-Wiener biodiversity indices, theoretical diversity and the equitability.

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Figure no.1 Aspects from Dofteana Dendrological Park

Nemira Mountains are part of the central group of Eastern Carpathians (Trotuşului Mountains), limiting their Southern part. With an area of 700 km², Nemira Mountains are situated in Bacău county (cca 90% of their surface), Covasna and Harghita county (10% of their surface).

Nemira Mountains are limited in the northern part by Uz river, in their eastern part by Trotuş river, Oituz river, Breţcu and Lemnia are southern limits and Bărzăuţa spring and Repatului Mountains are western limits of the studied area. Dofteana Dendrological Park is situated in north eastern part of Nemira Mountains.

The surface of Dofteana Dendrological Park is 34 ha (24 ha of it is occupied by the arboretum). In the centre area, the park has a important collection of exotic plants like: *Picea omorica* Purck., *Pinus nigra* Arn., *Pinus branksiana* Lamb., *Juniperus virginiana* L..

Material and methods

The method used in order to collect the material is directly from the plants. From the beginning of our study, in order to apply this method, we have took in count the next principle: the collecting conditions must be the same for each sample in order to compare the dates and results. So, in our field expeditions, we have the same parameters for each expedition: 5-7 days of collecting activity, 6 hours/day, and a team of 6 persons.

In order to find the biodiversity state fauna of cerambicids from Nemira Mountains, the synthetical relations between the number of species

and number of specimens, we considered necessary to use the biodiversity indices: Simpson and Shannon-Wiener, the theoretical diversity and the equitability.

Results and discutions

Analising the presence of cerambicids in this area, it comes out that two species are dominant *Stenurella melanura* and, *Stenurella bifasciata* species which cumulate 64,16% (38,15% respectively 26,01%). The other species are present in proportions less than 15% (graph.no. 1).

The number of specimens collected from Dofteana Dendrological Park in the entire period of study (2001-2002) is 173. After the analysis, realized in the Entomological Laboratory of Natural History Museum "I. Borcea" Bacău, we have found a number of 11 species part of 5 genera, from 2 subfamilies of *Cerambicydae* family (*Cerambycinae, Lamiinae).*

Out of the table no.I, is obvious that the number of species from Dofteana Dendrological Park area is small, situation justified by the presence of a different vegetation in this collecting area. Species like *Stenurella melanura* (represented by 66 specimens) *and Stenurella bifasciata* (45 specimens) are dominant species. Species like *Leptura maculata, Pseudovadonia livida and Judolia sexmaculata* are represented only by one specimen.

In order to find out about the biodiversity state of fauna of cerambicids from Dofteana Dendrological Park, according with table no.II, we calculated the Simpson and Shannon-Wiener indices, the theoretical diversity and the equitability. So, the Simpson index has a 3,35 value, Shannon-Wiener index =2,24, theoretical diversity =3,46 and equitability =63%.

Analyzing these values, is obvious that for a 2,24 value of Shannon-Wiener index, (according with the table III published by Lloyd and Ghelardi in 1974) the theoretical number of species is 7, the real number of species being 11 (the one found in the field).

Conclusions

After we have analyzed the 173 specimens of cerambicids collected from Dofteana Dendrological Park between 2001-2002, it comes out that are belonging to 11 species and 5 genera from 2 subfamilies of *Cerambycidae*

family (*Cerambycinae* and *Lamiinae*). To Dofteana Dendrological Park area correspond a small number of species; situation justified by the presence of different vegetation in this collecting area.

The Simpson index has a 3,35 value, Shannon-Wiener index =2,24, theoretical diversity =3,46 and equitability =63%. Analyzing the values, it comes out that for a 2,24 value of Shannon-Wiener index, (according with the table published by Lloyd and Ghelardi in 1974) the theoretical number of species is 7, the real number of species found in the field being 11.

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Table no. I The species of cerambicids collected from Dofteana Dendrological Park (2001-2002).

No.	Species	2001	2002	Total
1.	*Stenurella melanura*	28	38	66
2.	*Stenurella bifasciata*	13	32	45
3.	*Strangalia attenuata*	8	29	37
4.	*Stenurella septempunctata*	1	10	11
5.	*Chlorophorus sartor*	-	5	5
6.	*Stenurella nigra*	1	1	2
7.	*Chlorophorus varius*	-	2	2
8.	*Stenopterus rufus*	1	1	2

9.	*Pseudovadonia livida*	1	-	1
10.	*Judolia sexmaculata*	-	1	1
11.	*Leptura maculata*	-	-	1
TOTAL	53	120	173	

Graph. no. 1 Abundance of the cerambicids in Dofteana Dendrological Park

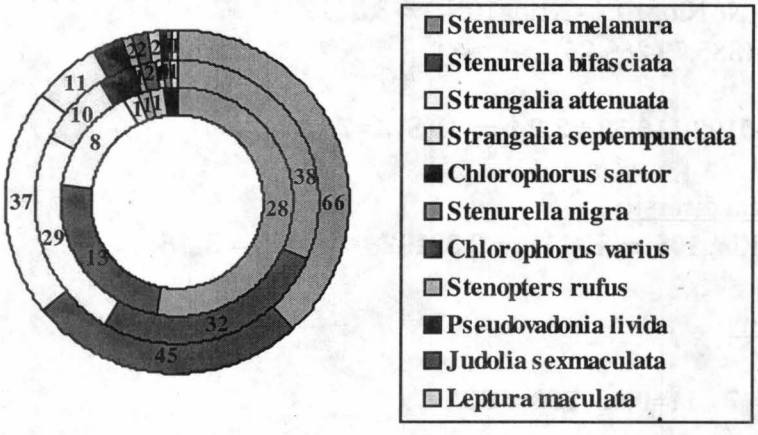

Table no. II Diversity indices for the species of cerambicids collected in Dofteana Dendrological Park (2001-2002).

No.	Species	N	N-1	N(N-1)	Log10N	Nlog10N
	Stenurella melanura	66	65	4920	1,8195	120,087
	Stenurella bifasciata	45	44	1980	1,6532	74,394
	Strangalia attenuata	37	36	1332	1,5682	58,0234
	Stenurella septempunctata	11	10	110	1,0413	11,4543
	Chlorophorus sartor	5	4	20	0,6989	3,4945
	Stenurella nigra	2	1	2	0,3010	0,6020
	Stenopterus rufus	2	1	2	0,3010	0,6020
	Chlorophorus sartor	2	1	2	0,3010	0,6020
	Pseudovadonia livida	1	0	0	0	0
.	*Judolia sexmaculata*	1	0	0	0	0
.	*Leptura maculata*	1	0	0	0	0
TOTAL		**173**	**162**	**8368**	**2,23**	**269,2592**

Simpson diversity index

$D = N(N-1) / \sum N(N-1)$

$D = 173 \cdot 162 / 8368 = 28026 / 8368 = \mathbf{3,35}$

Shannon-Wiener diversity index

$H(S) = K/N(N\log10N - Nr\log10Nr) = 3,321928 / 173 \cdot (173 \cdot 2,23 - 269,2592) =$
$= 0,0192 \cdot (385,79 - 269,26)$

$H(S) = 0,0192 \cdot 116,53 = \mathbf{2,24} \Rightarrow \mathbf{H(S)_{max}=7}$

Theoretical diversity

$\mathbf{H(S)_{max}} = K\log10S \Rightarrow H(11)_{max} = 3,321928 \cdot 1,0413 = \mathbf{3,46}$

Equitability

$E = S / S = 7 / 11 = 0,63 \cdot 100 = \mathbf{63}$

Table no. III Theoretical diversity M (S) corresponding to numerical values of S species (*after Lloyd and Ghelardi 1974*).

S	M(S)	S	M(S)	S	M(S)	S	M(S)	S	M(S)
1	0,0000	41	4,7861	81	5,7506	142	6,5521	255	7,3915
2	0,8114	42	4,8200	82	5,7681	144	6,5721	260	7,4194
3	1,2997	43	4,8532	83	5,7853	146	6,5919	265	7,4468
4	1,6556	44	4,8856	84	5,8024	148	6,6114	270	7,4736
5	1,8170	45	4,9173	85	5,8192	150	6,6306	275	7,5000
6	2,1713	46	4,9483	86	5,8359	152	6,6495	280	7,5259
7	2,3714	47	4,9787	87	5,8524	154	6,6683	285	7,5513
8	2,5465	48	5,0084	88	5,8687	156	6,6867	290	7,5763
9	2,7022	49	5,0375	89	5,8848	158	6,7050	295	7,6008
10	2,8425	50	5,0661	90	5,9007	160	6,7230	300	7,6250

11	2,9701	51	5,0941	91	5,9164	162	6,7408	310	7,6721
12	3,0872	52	5,1215	92	5,9320	164	6,7584	320	7,7174
13	3,1954	53	5,1485	93	5,9474	166	6,7757	330	7,7620
14	3,2960	54	5,1749	94	5,9627	168	6,7929	340	7,8049
15	3,3899	55	5,2009	95	5,9778	170	6,8099	350	7,8465
16	3,4780	56	5,2264	96	5,9927	172	6,8266	360	7,8870
17	3,5611	57	5,2515	97	6,0221	174	6,8432	370	7,9264
18	3,6395	58	5,2761	98	6,0221	176	6,8596	380	7,9648
19	3,7139	59	5,3004	99	6,0366	178	6,8758	390	8,0022
20	3,7846	60	5,3242	100	6,0510	180	6,8918	400	8,0386
21	3,8520	61	5,3476	102	6,0792	182	6,9076	410	8,0741
22	3,8520	62	5,3707	104	6,1064	184	6,9293	420	8,1087
23	3,9779	63	5,3934	106	6,1341	186	6,9388	430	8,1426
24	4,0369	64	5,4157	108	6,1608	188	6,9541	440	8,1757
25	4,0937	65	5,4378	110	6,1870	190	6,9693	450	8,2080
26	4,1482	66	5,4594	112	6,2128	192	6,9843	460	8,2396
27	4,2008	67	5,4808	114	6,2380	194	6,9992	470	8,2706
28	4,2515	68	5,5018	116	6,2629	196	7,0139	480	8,3009
29	4,3004	69	5,2260	118	6,2873	198	7,0284	490	8,3305
30	4,3478	70	5,5430	120	6,3118	200	7,0429	500	8,3596
31	4,3936	71	5,5632	122	6,3350	205	7,0788	550	8,4968
32	4,4381	72	5,5830	124	6,3582	210	7,1128	600	8,6220
33	4,4832	73	5,6027	126	6,3811	215	7,1466	650	8,7373
34	4,5230	74	5,6220	128	6,4036	220	7,1796	700	8,8440
35	4,5637	75	5,6411	130	6,4258	225	7,2118	750	8,9434
36	4,6032	76	5,6599	132	6,4176	230	7,2434	800	9,0343
37	4,6417	77	5,6785	134	6,4691	235	7,2743	850	9,1236
38	4,6792	78	5,6969	136	6,4903	240	7,3045	900	9,2060
39	4,7157	79	5,7150	138	6,5112	245	7,3341	950	9,2839

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STUDY ON RESINOUS FOREST SCOLYTIDAE STEM PEST IPS TYPOGRAPHUS

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Summary. Present paper sets out a study on resinous forest stem insects pest - Ips typographus - carried out in Negoiu region from the Făgăraş Mountains. The work includes morphologic , biologic and ecologic data concerning Ips typographus species, a representative species for Scolytidae family which includes extremely harmful species for resinous forest. A thorough comprehension of the biology and ecology of this insect is vital for the application of the methods to fight against.

Key words. Ips typographus, stem insects pest, population density, Scolytidae family, biotic factors, abiotic factors, beetle, gallery system.

Introduction

The multiplication of resinous stem insects pest was possible due to a complex of factors of abiotic and biotic nature. Among the important abiotic factors that create the proper conditions for insect maturation are to be mentioned the climatic ones .[1,3] The wind and the snow were the most important factors which being over critical limits had calamitated in great proportions the resinous forests. Among the harmful factors should be also mentioned: the excess moisture or the lack of it, extreme temperature, low atmospheric humidy, pollution by industrial noxae, some particularities of the resinous forests, the inadequate practical administration of the resinous forests along the years – multiple monoculture - as well as the insects' attack (like Lymantria monacha L.) on spruce fir tree which had intensely multiplied and in this waythey contributed to the physiological weakness of the trees and consequently they were attacked by the stem insect pest.

The distructive action of the wind and the snow was one of the most important factors that favoured the appearance and multiplication of the resinous forest stem insects pest.[3]

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The broken resinous trees are attacked by stem instects only in the fitst year, then they die and are no longer adequate for infestation, while the uprooted trees are infested in the following year too.

Infestation of the weakened trees.

Once the resinous trees are weakened physiologically, they were exposed to the insect pest which gets their complete dryness.

The most harmful insects are those that attach the stem of the weakned trees. There are more phases to be noticed in the proces of infecting the trees exposed to pollution.

In spring, at the beginning of the vegetation season, when the Scolytidae' flight occurs the weakened trees are vulnerable at infestation and at the insects attack. The spruce fir is infected by insects like Ips typographus, Ips amitinius on thicker areas of the trunk; on wetter zones species like Hylurgops and Ttypodendron lineatum, can appear too and Pytiogenes chalcographus, Chriphalus abies can be found on less thinck zones and on the branches.[3]

Phytogeographical characterization of the collecting zone:

The material was gathered in the central zone of the Carpathians in the Făgăraş Mountains. The vegetation conditions are favourable especially for spruce fir. The zone was chosen so that the study would cover an extended area. The material was collected from the trees growing at the edge of touristic path, on the sunny zone, particularly on the felled trees by storm, grouped or isolated. The trees attacked by isects from Scolytidae family can be identified after the entrance and exit holes made by the beetles. Those holes can be noticed on the area of the trunk exposed to the sun. The infested trees can be identified due to the redness of the upper branches of the conifers.(Fig. No. 1)

Characterization of Ips typographus:

In Scolytidae family are small and very small beetles (1-9 mm); about 90 species. Ips typographus (Fig. No.2) is an Eurosiberian species. In our country there exists a common species. The size of the beetle is 4,2-5,5 mm.

Its colour is dark brown , almost black and a little glowing. Its anntenaes and legs are yellow –redish. The body of the beetle is coverd with hairs.

Fig. No 1

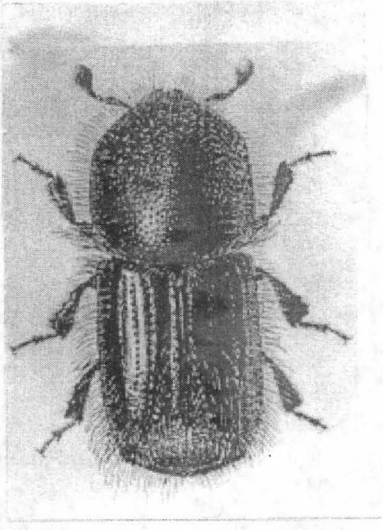
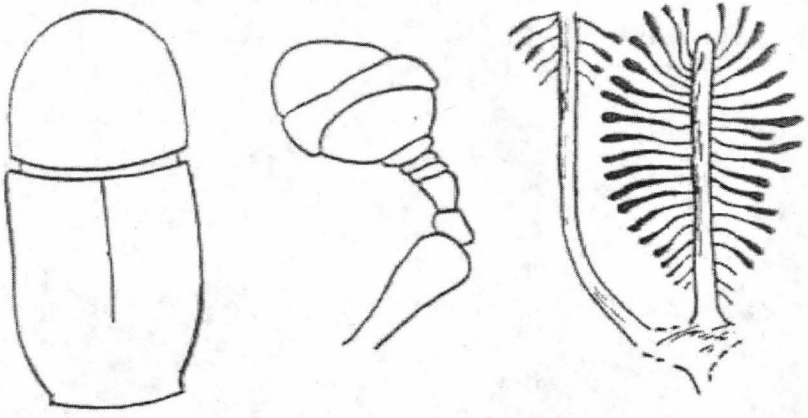


Fig no.2

Ips typographus attacs spruce fir (Picea abies) aged 80-100 years old and seldom under 50 years old. It is an insect specific to the spruce fir tree (Picea abies) but the pine (Pinus silvestris) can also be attacked. – Pinus nigra and Larix decidua – are also attacked when they grow in a forest with spruce fir trees. The most of that zone vegetation consists of spruce fir trees (Picea abies) and some of them show older attacks made by Ips typographus. A recent attack can be noticed in the touristic path zone .

The attacked trees are different from the healthy ones as their top branches are redish and they start to get dried from top to bottom. The beetles of Ips typographus like the other beetles belonging to Scolytidae family attack, as a rule, only the weakened trees either by other beetles' attack or phungi or the trees felled by wind and snow. If the environmental conditions are favourable their number increases and they will attack the healthy trees too, especially those that have physiological problems during the dry weather.[1,2,3,5]



a. b. c.

Fig. No.3: a. Body form b. Anntenaes form c. Gallery system

The attacked trees bark can be easly peeled. Inbetween the bark and wood the system of galleries, holes carved in the wood can be noticed. These prints bear the charactheristics that define the species being an important clue for the identification of the species.The system of galleries starts with the so colled "mother gallery" which are vertical with two arms and seldom with one or more arms. These "mother galleries", have the width 3-3,5 mm and the lenght 6-15 cm. Sometimes they also have a kind of "airing holes". On the side walls of the gallery the female cuts small "niches" at 2 to 10 mm distance where it deposits 20-100 eggs (the emergence of larvae depends on temperature and takes place after 10-15 days from deposition). The galleries cut by larvae are, at the beginning, narrow and perpendicular on the "mother gallery" then their shape become winding. Once the larvae grow the galleries enlarge too. The mature larva cuts at the end of the gallery a " pupate room". The duration of the larva and pupa stage depends on temperature and humidy. On average the larva stage lasts 24 days and the pupae one stage lasts 12 days.[1,2,3,6]

As a rule, Ips typographus has two generations a year. The first flight happens in spring, very early when the temperature of the air is 20°C, namely at the latter half of April. The second flight takes place at the beginning of July, the second flight is unimportant. The pairing and the deposition of eggs are under the bark. Ips typographus, is a polygame species and the male,

pairs more females (2-12). In this special occasion, the male cuts an entrance hole where the burk is thinner or it enters under the "scales" of the bark, in this case the hols can hardly be observed.[3,6]

Material and method

We collected the material and the data about the density of the population of ips typographus in May and end of Oct. 2000. As we have already mentioned above, we were interested in observing the phenomenon on the spruce fir (Picea abies) from the touristic path zone on the Serbota Valley. The spruce fir trees were found down the ground, probably felled by strong winds, that helped us easly collect the biological material and we were able to observe the shape of gallery better. To ease the understanding of explanations we noted the spruce fir with " a, b, c, d,". In the following cases: a, b, and d, we had in view the in and out access holes of the beetles (insects). Their number was a clue for us to identify the density of the population. We observed the in and out access holes of the betlees on a surface of 10 cm, the samples being collected every 30 cm on the hight. From the data shown in Table no. 1 we drew the conclusion that the density of the population is bigger between 2-5 m which is characteristic to Ips typographus. Samples (individuals) from this species were not found on the spurce fir noted with "a" and "d" which indicates thatthe attack was long ago and the "individuals" from young generation had migrated to another tree. Not having any beetles, we guessed that there might have been an attack of Ips typographus studying the shape of the left gallery.

In case of the spruce fir – tree "c" found at a distance of 10 m from the group of trees "a, b, d" we also study the betlees' in and out access galleries on a surface of 10 cm, every 50 cm on its length. The data are shown in Table no. 1. Like in the other cases the density of the grooves was bigger between 3-5 m at the bottom of the tree. The bark of the spurce fir – tree "c", which was easily peeled, we found a great number of individuals (600) from species Ips typographus.

The beetles were collected with a special instrument, or with the twerzer, then we put them in test tubes with chloroform. Later we identified them in the laboratory. In case of spurce fir – tree "c", we peeld some burk and we put fragments in a devise called "tulgren". After the beetles had left the bark they were collected in plastic bags. Periodically we observed the beetles emergence and we drew the following figure (nr. 4); the emergence

time was rather long 21 days. This emergence, if graduated, has an adjusting character and is genetically printed. Thus, if the flight of all beetles took place at the some time and the environmental conditions were unfavourable, all the individuals of this species would die and the species perpetuation wouldn't be achieved .

Table no. 1

No. samples	Spurce fir a		Spurce fir b		Spurce fir c		Spurce fir d	
	No. holes 10 cm	height m	No. holes 10 cm	height m	No. holes 10 cm	height m	No. holes 10 cm	height m
1	25	1,5	4	0,5	10	0,5	6	0,5
2	21	2	10	1	12	1	9	1
3	21	2,5	4	1,5	20	1,5	12	1,5
4	30	3	14	2	24	2	16	2
5	27	3,5	16	2,5	25	2,5	19	2,5
6	41	4	20	3	32	3	13	3
7	27	4,5	16	3,5	15	3,5	11	3,5
8	40	5	11	4	12	4	7	4
9	10	5,5	10	4,5	27	4,5	7	4,5
10	27	6	7	5			11	5
11	7	6,5	7	5,5			4	5,5
12	14	7	4	6				

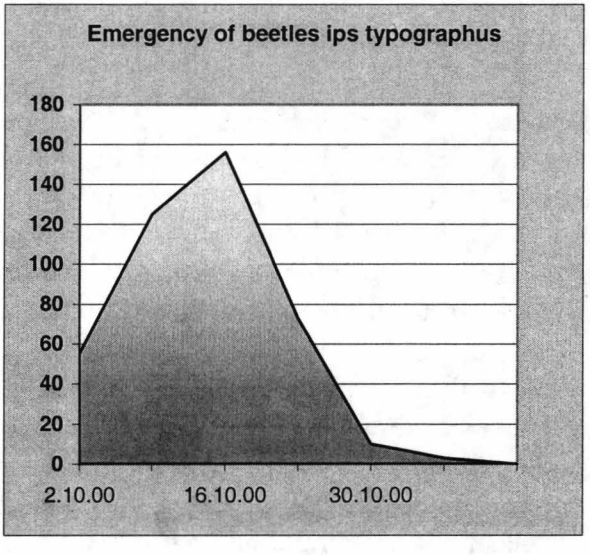


Fig no. 4

We also noticed on that zone weakened conifers due to the insects or fungi attacks- these trees would have been perfect hosts for beetles from Scolytidae family .

Methods to combat Ips typographus

Preventive measures. Prophilactic and indirect preventive measures aim at preview the emergency and mass multiplication of these insects pest and ensure the forest vegetation better developing conditions :

a. Phytosanity hygiene measures
b. Phytosanity control
c. Phytosanity quarantine measures

Mechanic combat measures. Among the efficient combat methods we can mention the mechanic ones like : trap poles and trap trees which after attracting the insects they can be burnt.[7]

Chemical combat . insecticides classified according to their action

a. Ingest insecticides
b. Contact insecticides
c. Organic insecticides

In the perioud 1980-1985 the pheromone technique was tested and applied on a large scale and proved to be efficient as specificity, intensity and action duration in the detection, attack prevention and control of spruce bark beetle, ips typographus. At the beginning of the beetle flight, the pheromone plays the part of a sexual aggregation attractant, which attracts both males and females and in summer the sexual attraction increases during the reproductionperiod, mainly females being caught.[4].

Conclusions

The material was collected from Negoi zone from the Făgăraş Mountains from two different collecting centres .

We collected 885 samples of beetles from species Ips typographus and they were examined in the laboratory.

We studied the species morphology having in view thouse charecteristics of taxonomical interest.

We carried out a research on the biology and ecology of the species both on open air and in the laboratory .The data were completed using the bibliography.

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BIOLOGIE VEGETALĂ

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STUDY OF SULPHATE-REDUCING BACTERIA IN AN ECOSYSTEM FROM MANGALIA

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Abstract. Sediment and water samples from the cave Movile, the lake Kara Oban and the spring Hagieni (Mangalia ecosystem), were analyzed microbiologically. The number of desulfofiers was counted using two bacterial culture media (Van Delden and Herbert). No sulphate reducing activity in any of the water samples analyzed was detected. The desulfofiers were detected in all the sediments, in both the culture medium used, and their number had an order of magnitude of 10^2-10^3 bacteria/g sediment dry matter. The number of the desulfofiers registered different values, depending on sampling site, consistence of the sediment and culture medium used. The mean values of the two methods applied showed the following descending order of the three sediments studied, as regard the number of sulphate reducing bacteria: the cave Movile (1870 desulfofiers/g sediment dry matter) > Kara Oban (1740 desulfofiers/g sediment dry matter) > Hagieni (757 desulfofiers/g sediment dry matter). The lack of significant differences between the values obtained using the two bacterial culture media showed that both the methods applied are proper for this kind of determinations.

Key words. microorganisms, sulphate reduction, desulfofiers, sediments

Introduction

Sulphur is an ordinary element, wide spread in nature, in soils, waters, atmosphere, and it is a fundamental component of the biochemical structures of all the organisms. The assembly of the natural transformations

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of the element is defined by the syntagm natural cycling of the sulphur. The most important inorganic forms of the sulphur are: elemental deposits (S^0), H_2S and metalic sulphides (reduced forms), as well as the oxidized forms: SO_2, SO_3, $S_2O_3^{2-}$, $S_4O_6^{2-}$, SO_4^{2-}, H_2SO_4. Sulphur is also present in organic compounds, especialy like component of some amino acids, like cysteine and methionine.

The microbial processes of sulphur oxidation as well as those of assimilatory and dissimilatory reduction of the sulphates, are revised by Kiss (1972) and Zarnea (1994). Big amounts of sulphur oxidized forms (sulphates), present in soils, stones, fresh or salt waters, can be mobilized by bacteria and subsequently used in biosynthesis (assimilatory reduction). Dissimilatory reduction of sulphates is accomplished by some sulphate-reducing, strictly anaerobic bacteria: Desulfovibrio, Desulfotomaculum, Desulfomonas. Only few quantities of sulphur released are used for the biosynthesis of the proper organic substances of these bacteria. The majority is released in the environment as H_2S. The sulphate-reducing bacteria (desulfofiers) can use sulphate as the terminal electron acceptor in their respiration. As H donors they can use either its molecular form (chemolithotrophy), or organic sources (chemoorganotrophy). These bacteria can assimilate C from CO_2 (autotrophy), or from organic compounds (heterotrophy).

We presents in this paper our studies on the bacterial sulphate reduction activity in an ecosystem from Mangalia (Constanța county, Romania). The ecosystem is situated at NV of Mangalia. Lascu et al. (1994) makes a detailed geological and geographical description of the site.

Materials and methods

The number of desulfofier bacteria was determined in the following terrestrial and subterranean waters and sediments: the cave Movile, the lake Kara Oban and the spring Hagieni. The samples were collected in September 2003. In order to count the number of desulfofiers we used two bacterial mediums: Van Delden (Allen, 1957), with the following composition: 0.2 g asparagine, 0.1 g K_2HPO_4, 0.15 g $MgSO_4 \cdot 7H_2O$, 0.5 g Ca SO_4, 0,5g Na lactate, traces of $FeSO_4 \cdot 7H_20$, 100 ml and distilled water; pH 7.0; and the Herbert medium (1976), with the following composition: 0.05 g KH_2PO_4, 0.1 g NH_4Cl, 0.1 g $CaSO_4$, 0.2 g $MgSO_4 \cdot 7H_2O$, 0.02 g $FeSO_4 \cdot 7H_2O$, 0.35 g Na lactate, 0.1 g yeast extract, 0.01 g ascorbic acid, 0.01 g tioglicolic acid, 100 ml distilled water; pH 7.0. The medium distributed in tubes (15ml/tube) was autoclaved

at 105şC, 30 minute daily, three consecutive days. Successive dilutions (10^{-1}–10^{-6}) of the samples were made and series of 5 tubes containing the culture medium were inoculated with 1 ml of each dilution (10^{-1}–10^{-6} for sediments and 10^{-1}–10^{-4} for water samples). Incubation was carried out 10 days at 30şC. The cultures were examined in order to detect the presence of H_2S produced as a result of the sulphate reducing activity of the desulfofiers presents in the samples. We used paper bands imbued in a 10% solution of lead acetate. The presence of a black pot on the paper (PbS), after boiling the medium in acid medium (HCl) indicate the sulphate reduction and the formation of H_2S, as a result of the bacterial activity (desulfofiers) in the samples analyzed. The dry matter of the sediments was also measured, so that the number of the bacteria was expressed in cells/g sediment dry matter.

Results and discussions

We could not detect the presence of a sulphate reducing activity in any of the water samples analyzed, either in medium Van Delden, or in medium Herbert. The desulfofiers were detected in all the sediments, in both the culture medium used. The results are presented in Table 1 and Fig. 1. As we can see, the number of the desulfofiers registered different values, depending on sampling site, consistence of the sediment and culture medium used. The results were interpreted according to the Alexander (1965) table, taking into account the 10^{-2}-10^{-4} dilutions.

The sediment from the cave Movile was the most consistent (72.2% dry matter), closely followed by that from the lake Kara Oban (71.84% dry matter). The sediment from Hagieni contained more water (52.84% dry matter). The lowest level of the sulphate reducing activity was registered in this last sediment, using both the Van Delden (625 desulfofiers/g sediment dry matter), and the Herbert (889 desulfofiers/g sediment dry matter) culture medium. It seems like the Herbert medium, which contains more nutrients, stimulated the development of the suplhate reducing bacteria. The observation is also sustained by the results obtained in sediment from the lake Kara Oban. Thus, 1531 desulfofiers/g sediment dry matter were counted when used the Van Delden medium and 1949 desulfofiers/g sediment dry matter when used the Herbert medium.

However, this observation is not sustained by the results obtained in the sediments from the cave Movile. Thus, the number of sulphate reducing bacteria cultivated in the Van Delden medium was 1939 desulfofiers/g

sediment dry matter, higher than the number registered in the Herbert medium (1801 desulfofiers/g sediment dry matter). It means that, even cheaper, the Van Delden medium is quite proper for this kind of determinations.

Table 1 Number of desulfofiers in the three sediments analyzed.

Sampling site	Dry matter (%)	Van Delden medium		Herbert medium	
		Cells/g wet sediment	Cells/g sediment (dry matter)	Cells/g wet sediment	Cells/g sediment (dry matter)
Cave Movile	72.2	1400	1939	1300	1801
Kara Oban	71.84	1100	1531	1400	1949
Hagieni	52.84	330	625	470	889

The mean values of the two methods applied show the following descending order of the sulphate reducing activity in the three sediments analyzed: cave Movile (1870 desulfofiers/g sediment dry matter) > Kara Oban (1740 desulfofiers/g sediment dry matter) > Hagieni (1801 desulfofiers/g sediment dry matter) (Fig. 1).

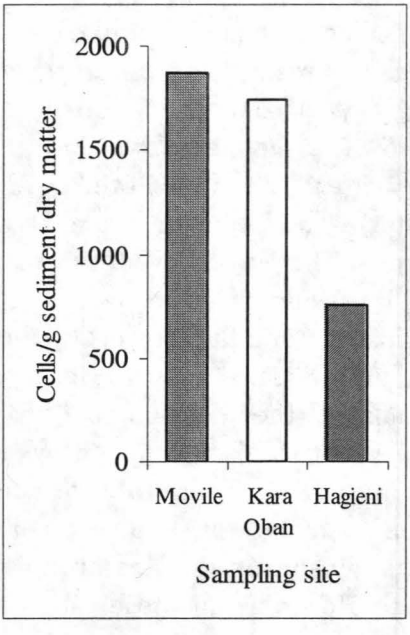

Fig.1. Mean values of the desulfofiers counted in the three sediments analyzed.

Many researchers, Romanians or from other countries, have studied the dissimilatory sulphate reduction in different habitats. King and Klug (1980) study the sulfhydrolase activity in sediments of Wintergreen lake (Michigan). The experiments show, for one thing, the enzymatic hydrolysis of the sulphuric acid esters in sediments, and, on the other hand, the use of resulting sulphate by the sulphate-reducing microorganisms, which rapidly convert it to H_2S. The dissimilatory sulphate reduction in marine environment is more active, as compared to the assimilatory reduction (Nedwell, 1982).

The situation differs in the fresh waters, where only few quantities of sulphate are available. Because the sulphate reduction needs reducing anaerobic conditions, the process is carried out only in sediments. The highest level of the sulphate reduction is registered in the upper layer of the aerobic zone of the sediments. Here, the concentrations of the H donors and acceptors, both provided by the upper layer of the sediment, are the highest. The sulphate reduction rate decrease with the depth of the sediment. The same decrease is followed by the number of the sulphate reducing bacteria. Oren (1988) consider that in the hypersaline environments, the most important precursors of methanogenesis are substrates like methionine, methanol or methylamines, rather than hydrogen or acetate. The explanation offered by the author is that the hypersaline environments are generally very rich in sulphates, as compared to the ocean water and thus, the sulphate reducing bacteria efficiently compete with the methanogenic ones for the H_2 and acetate present in the environment.

The presence of the desulfofiers is registered in sediments of some salt lakes riverain to the Black Sea (Costineşti and Techirghiol lakes). The number of desulfofiers was of an order of 10^4 bacteria/g dry sediment (Muntean, 1996). Manolache (2001) reports the presence of desulfofiers in sediments sampled from three Romanian caves (Tăuşoare, Valea Firii and Vântului. The number of desulfofiers in the cave sediments is lower (10^2 cells/g dry matter) than in the salt lake sediments. As we can see, our result as regard the number of the sulphate reducing bacteria are in good agreement to those registered by other researchers in sediments from Romanian lakes or caves.

Conclusions

The sulphate reducing activity was registered in all the three sediments studied. We could not detect the presence of desulfofiers in any of the water samples analyzed. The level of the sulphate reducing activity in sediments differed depending on the sampling site, consistence of the sediment (dry matter), and the culture medium used.

In the case of sediments from Kara Oban and Hagieni, the highest number of desulfofiers was registered when the Herbert culture medium was used, while in the case of sediment from the cave Movile, we counted more desulfofiers/g sediment in medium Van Delden. The differences are not significant, indicating that both the methods applied are proper for this kind of determinations.

The mean values of the two methods applied showed the following descending order of the three sediments studied, as regard the number of sulphate reducing bacteria: cave Movile (1870 desulfofiers/g sediment dry matter) > Kara Oban (1740 desulfofiers/g sediment dry matter) > Hagieni (757 desulfofiers/g sediment dry matter). Thus, the number of desulfofiers in sediment from Hagieni is only 40% from that registered in sediment from the cave Movile, while the number of desulfofiers in sediment from Kara Oban is lower only by 7% as compared with the most active sediment analyzed.

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Studiul bacteriilor sulfat-reducătoare din ecosistemul Mangalia. Au fost analizate microbiologic probe de sediment şi apă prelevate din peştera Movile, lacul Kara Oban şi izvorul Hagieni (ecosisteme din zona Mangalia). A fost stabilit numărul numărul bacteriilor desufoficatoare prin utilizarea a două medii de cultură (Van Delden şi Herbert).În probele de apă analizate nu s-a putut detecta activitatea de reducere bacteriană a sulfaţilor. Desulfoficatoarele au fost puse în evidenţă în cele trei sedimente, în ambele medii de cultură, numărul lor fiind cuprins între 10^2-10^3 bacterii /g substanţa uscată a sedimentului.Numărul desulfoficatoarelor a înregistrat valori diferite în funcţie de locul de prelevare, consisenţa sedimentului şi a mediului de cultură folosit.Valorile medii obţinute prn aplicarea celor două metode prezintă următoarea ordine descrescătoare a celor trei sedimente studiate privind numărul bacteriilor desulfoficatoare:peşteraMovile (1870 desulfoficatoare /g substanţa uscată a sedimentului) > Kara Oban (1740 desulfoficatoare /g substanţa uscată a sedimentului) > Hagieni (757 desulfoficatoare /g substanţa uscată a sedimentului).Prin utlizarea celor două medii de cultură nu s-au înregistrat diferenţe semnificative a nummărului bacteriilor desulfoficatoare, ambele fiind corespunzătoare evidenţierii acestui grup ecofiziologic de bacterii.

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MICROBIOTA OF SOME MINERAL WATERS FROM TRANSILVANIA (LĂPUŞ DEPRESSION, MARAMUREŞ COUNTY)

M. DRĂGAN-BULARDA*, C. IUŞAN**

Rezumat. Microbiota prezentă în apele minerale este strâns legată de condiţiile ecologice, fizico-chimice, hidrogeologice ale zăcământului, precum şi de starea sanitară a zăcământului, respectiv a instalaţiilor de exploatare. Au fost analizate din punct de vedere microbiologic 9 izvoare de apă minerală din depresiunea Lăpuşului, judeţul Maramureş. S-a determinat prezenţa bacteriilor din următoarele grupe fiziologice (ecologice): sulf-oxidante, sulfatreducătoare, desulfoficatoare, amonificatoare, nitritbacterii, nitratbacterii, denitrificatoare, fier-reducătoare, heterotrofe aerobe. Pentru testarea parametrilor igienico-sanitari s-a determinat numărul de coliformi totali, coliformii fecali şi enterococii fecali. Rezultatele obţinute au arătat că bacteriile heterotrofe aerobe au fost prezente în toate izvoarele analizate, dar în număr redus, întrucât apele minerale studiate sunt oligotrofe, oferind o cantitate foarte mică de substanţă organică. Bacteriile sulf-oxidante au fost evidenţiate din aproape toate izvoarele, la fel şi cele sulfat-reducătoare, confirmând rolul acestora în circuitul biogeochimic al sulfului, având implicaţii în chimismul apelor. Fier-reducătoarele au fost evidenţiate în număr mic, la fel şi cele din ciclul biogeochimic al azotului. Indicatorii bacteriologici ai stării sanitare nu au fost evidenţiaţi, demonstrând calitatea sanitară a apelor din aceste izvoare.

Cuvinte cheie. Microbiota, mineral watwrs, Transilvania, Rumania

Mineral waters have been used as a factor of cure from ancient times and nowdays the procedures of treatment offered by nature medicine are more and more emphasized and as a result, the possibilities offered by natural therapeutic factors (mineral waters, therapeutic silts) are being reevaluated. The using of such natural resources offer multiple socio-economic advantages regarding the reduction of hospitalisation periods, giving up drug treatment, which can have some undesirable secondary effects etc.

The mineral springs considered in this study are situated in the Depression of Lăpuş, Maramureş county: Stoiceni, Rogoz I (At the River

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spring), Rogoz II (Across the River spring), Rogoz III (At the Well spring), Borcut I (Near the Valley), Borcut II (On the Grassland), Măgureni I (BabeiValley), Măgureni II (Babei Valley - basin), Măgureni III (The Stone Well). The mineral water samples were taken on the 28th of September 2003, in sterile conditions.

Researching the microbiology of the mineral waters in this zone requires the evidentiation of some ecological groups of bacteria: sulphur-oxidizing, sulphate-reducing, desulphuring, ammonifying, nitrifying, denitrifying, iron-reducing, mesophilic heterotrophic. Another purpose of the study was to establish the hygienico-sanitary parameters: mesophilic heterotrophic bacteria, total numbers of coliform bacteria, faecal coliform bacteria, faecal enterococcus. The interpretation of the microbiological data was done according to the hydrochemical analysis of these mineral waters and the geology of the area. These microbiological analyses are the first of this kind for these springs.

The chemical analysis of the mineral water of Stoiceni shows that it is carbon-gaseous, bicarbonated, magnesian water, sodium water, calcicum water with a high metaboric acid content, hypotonic with a total mineralization of 9.2 g/l and 1.7 g/l free CO_2 content, and as a result of the microbiological analyses it turned out to be sulphurous, which confers it therapeutic qualities. At Borcut on the Drăgoiasa Valley, there are some carbogaseous mineral water springs. At Măgureni three spring are known: two on Babei Valley, of sulphurous nature, carbonated, calcic, magnesian, sodic with a total mineralisation of 1.8 g/l and 0.005 g/l H_2S. Between 1869 and 1951 there was a small balneary station here. The third spring, The Stone Well, is of carbogaseous nature. The mineral springs at Rogoz have not been studied hydrochemically.

Materials and methods.

The samples were prelevated in conditions of sterility, from the 4 places in the Depression of Lăpuş, situated around Târgu-Lăpuş: Stoiceni (1 spring), Rogoz (3 springs), Borcut (2 springs), Măgureni (3 springs). For taking the water samples, the following materials were used: flat bottomed flusk which have been previously sterilised, in the autoclave at 121°C, 1 h.

Media specific for each ecologic group of bacteria were used as folows:

♦ Sulphur-oxidizing bacteria

1. Enrichment medium: 0.1 g K_2HPO_4; 0.05 g $MgSO_4x7H_2O$; 0.1 g NH_4NO_3; 1 g suphur powder; 1 g $CaCO_3$; 100 ml distilled water.
2. Basal medium: 0.05 g K_2HPO_4; 0.05 g $(NH_4)_2SO_4$; 0.01 g $CaCl_2$; 0.01 g Na_2CO_3; 0.1 g natrium silicate; 1 ml of microelements solution; 100 ml distilled water.
3. Solution of microelements: 5 mg EDTA; 2.2 g $ZnSO_4x7H_2O$; 0.544 g $CaCl_2$; 0.506 g $MnCl_2x4H_2O$; 0.499 g $FeSO_4x7H_2O$; 0.11 g $(NH_4)_6Mo_7O_2xH_2O$; 0.15 g $CuSO_4x5H_2O$; 0.161 g $CoCl_2x6H_2O$; 100 ml distilled water; and pH is ajusted to 6 with KOH. The solution is sterilised at 105 °C, for 30 minutes, in 3 consecutive days.
4. S medium (Aaronson, 1970): 1 g S; 0.3 g $(NH_4)_2SO_4$; 0.3 g KH_2PO_4; 0.025 g $CaCl_2$; 0.05 $MgSO_4x7H_2O$; 100 ml distilled water; 1 ml solution of microelements. The medium is sterilised at 105 °C, 30 minutes in 3 consecutive days.
5. R medium (Aaronson, 1970): 0.05 g NH_4Cl; 0.05 g $MgCl_2x6H_2O$; 0.2 g KH_2PO_4; 0.5 g $Na_2S_2O_3x5H_2O$; 0.2 g KNO_3; 0.1 g $NaHCO_3$; 1 ml solution of microelements; 100 ml distilled water. Sterilisation is done in the autoclave at 105 °C for 30 minutes in 3 consecutive days.

♦ Sulphate-reducing bacteria:

1. Van Delden medium: 2 g asparagine; 1 g K_2HPO_4; 1 g $CaSO_4$; 1.5 g $MgSO_4$; 5 g natrium lactate; 0.02 g $FeSO_4$; 1000 ml distilled water, pH is ajusted at 7. Sterilisation is done at 105 °C, 30 minutes in 3 consecutive days.

♦ Desulphuring bacteria

1. The nutritive medium broth with lead acetate imprignated strips $(CH_3COO)_2Pb$.

♦ Iron-reducing bacteria

Ottow medium 1968, Pârvu 1977: 0.8 g K_2HPO_4; 0.8 g KH_2PO_4; 0.2 g $MgSO_4x7H_2O$; 0.2 g KCl; 0.5 g yeast extract; 20 g glucose; 5 g peptone; 0.05 g $Fe_2O_3x3H_2O$; 0.1 g $MnSO_4$; 1000 ml distilled water, pH is ajusted to 7, sterilisation by autoclavation at 105 °C, for one hour a day, in 3 consecutive days.

♦ Nitrite bacteria

1. Alef medium, 1995: 1 g $(NH_4)_2SO_4$; 0.5 g K_2HPO_4; 2 g NaCl; 0.2 g $MgSO_4x7H_2O$; 0.05 g $FeSO_4x7H_2O$; 6 g $CaCO_3$; 10 ml

solution of microelements; 800 ml distilled water. The medium is sterilised at 120 °C, 20 minutes.

2. The solution of microelements: 1000 mg Fe III-citrat, 10 mg $MNCl_2x4H_2O$; 5 mg $ZnCl_2$; 5 mg LiCl; 2.5 mg KBr; 5 mg $CuSO_4$; 1000 mg $CaCl_2$; 1 mg $Na_2MoO_4x2H_2O$; 5 mg $CoCl_2$; 5 mg $SnCl_2xH_2O$; 2.5 g KI; 5 mg $BaCl_2$; 1 mg $AlCl_3$; 10 mg H_3BO_3; 20 mg EDTA; 1000 ml distilled water. Microelements solutions is sterilised in autoclave at 112 °C, 20 minutes.

- Nitrate bacteria

  Growing medium: 0.2 g $NaNO_2$; 6 g $CaCO_3$; 0.5 g K_2HPO_4; 0.2 g $MgSO_4x7H_2O$; 0.005 g $FeSO_4x7H_2O$; 0.5 g NaCl; 800 ml distilled water, pH is ajusted at 7.6 and is sterilised in autoclav at 120 °C, 20 minutes.

- Mesophilic heterotrophic bacteria

  Simple gelose medium: 1000 ml nutritive broth; 20 g agar-agar. It is sterilised at 120 °C for 30 minutes.

- Denitrifying bacteria

  Mineral medium: 10 mg natrium acetat; 20 mg KNO_3; 0.5 g K_2PO_4; 0.2 g $MgSO_4x7H_2O$; 10 ml Vinogradski solution; 800 ml distilled water, pH is ajusted at 7 and is sterilised in autoclave at 115 °C, 20 minutes.

- Ammonifying bacteria:

  Nutrient medium: 10 g bactopeptone; 5 g NaCl; 1000 ml distilled water; pH 9; sterilisation at 121°C, 30 minutes.

- The hygienico-sanitary indicators

  1. For the coliform germs the presumption test was done, which required the lauryl sulphate broth medium and double lauryl sulphate broth. The lauryl sulphate broth medium has the folowing composition: 20 g peptone; 2.75 g Na_2HPO_4; 2.75 g NaH_2PO_4; 5 g NaCl; 5 g lactose; 0.1 g lauryl natrium sulphate; 1000 ml distilled water. The doubles concentrated lauryl sulphate broth medium has the following composition: 40 g peptone; 5.50 g Na_2HPO_2; 5.50 g NaH_2PO_4; 10 g NaCl; 10 g lactose; 0.2 g lauryl natrium sulphate; 1000 ml distilled water, pH is ajusted at 6.8 and is sterilised at 115 °C, 20 minutes.
  2. For the faecal coliforms the MacConkey medium was used.

3. For faecal enterococcus, the sodium azide (simple and double concentrate) media and the sodium purple bromcresol azide media have been used.

After the prelevation of samples, decimal dilution were prepared, between 10^{-1} and 10^{-2}. These dilution were inoculated into 2 Petri dishes or 5 test tubes containing the specific nutritive medium for each ecological group of bacteria. The inoculation volume was: 1 ml. The incubation of the media lasted for 7 days at 28 °C for the physiological groups of bacteria and for 24 hours at 37 °C for the hygienico-sanitary indicators. For establishing the number of bacteria Alexander`s table was used. In the case of the mesophilic heterotrofic bacteria cultured in Petri dishes the bacterial colonies wich apperead on the surface of the medium after incubation were counted, according to the principle that each colony is generated by one bacterium. For the hygienico-sanitary indicators, the tables stipulated by the STAS method were used.

The cultures of sulphur-oxidizing bacteria were examined by establishing the opalescence of the medium and the pH modification after incubation. The presence of desulphurizer bacteria was observed by indicating the H_2S by the method of lead acetat stripes. The nitritbacteria were examined by producing NO_2^- . Nitrate bacteria were examined by producing the NO_3^- .The Fe^{2+} ions produced by the Fe reducing bacteria were evidentiated by adding the α,α – dipiridyl reagent. The ammonifying bacteria were evidentiated by means of the Nessler reagent.

Results and discussions.

The results of the bacteriological analyses are presented in tables 1-3 and in fig.1.

Table 1. Sulphur-oxidizing bacteria in studied mineral water springs.

Water source	Spring	Nutrient medium	Initial pH	Final pH	Bacteria number (no./ml)
Stoiceni	La casă	M	7.5	7.5	4380
Stoiceni	La casă	R	7.5	7.5	
Stoiceni	La casă	S	7.5	6.0	
Rogoz I	Râu	M	7.5	7.5	0
Rogoz I	Râu	R	7.5	7.5	
Rogoz I	Râu	S	7.5	7.5	
Rogoz II	Peste râu	M	7.5	7.5	3600
Rogoz II	Peste râu	R	7.5	6.5	
Rogoz II	Peste râu	S	7.5	7.0	
Rogoz III	La fântână	M	7.5	7.5	2800
Rogoz III	La fântână	R	7.5	6.5	
Rogoz III	La fântână	S	7.5	7.5	
Borcut I	Lângă Vale	M	7.5	7.5	6000
Borcut I	Lângă Vale	R	7.5	6.5	
Borcut I	Lângă Vale	S	7.5	7.5	
Borcut II	Pe pășune	M	7.5	7.5	2100
Borcut II	Pe pășune	R	7.5	7.0	
Borcut II	Pe pășune	S	7.5	7.5	
Măgureni I	Valea Babei	M	7.5	7.5	
Măgureni I	Valea Babei	R	7.5	6.5	

Măgureni I	Valea Babei	S	7.5	6.5	6800
Măgureni II	V. Babei bazin	M	7.5	7.5	9300
Măgureni II	V. Babei bazin	R	7.5	5.5	
Măgureni II	V. Babei bazin	S	7.5	6.5	
Măgureni III	Fântâna de piatră	M	7.5	7.5	6400
Măgureni III	Fântâna de piatră	R	7.5	7.0	
Măgureni III	Fântâna de piatră	S	7.5	7.0	

M- control medium, R- R medium, S- S medium.

We can see that important multiplications of the occurred medium R at the Rogoz II, Rogoz III, Borcut I, Măgureni I, II, III springs and in medium S modifications appeared in the case of the Stoiceni, Rogoz II, III and Măgureni II and III springs.

The data regarding the number of bacteria from different ecological group are presented in the following table:

Table 2. Number of different bacteria type in mineral spring water of Lăpuş Depression

Source	Stoi.	Rog. I	Rog. II	Rog. III	Borcut I	Borcut II	Măg. I	Măg. II	Măg. III
Spring	La casă	Râu	Peste râu	La fântâ-nă	Lângă vale	Pe păşu-ne	Valea Babei	V. Ba-bei bazin	Fântân a piatră
Aerobe heter. B	1200	2100	16000	29000	17000	11000	95000	21000	9000
Amon.b	28	45	54	350	920	54	16000	16000	31000
Nitrite-b.	6	11	9	14	14	9	20	27	290
Nitrate-b.	7	10	7	12	13	11	22	28	350
Denitr.b	110	95	24	39	39	39	140	33	40
Desulph. b	22	32	160	160	72	140	64	280	350
Ironred b.	14	62	350	45	350	32	38	64	60

Stoi. - Stoiceni, Rog. – Rogoz, Măg. – Măgureni.

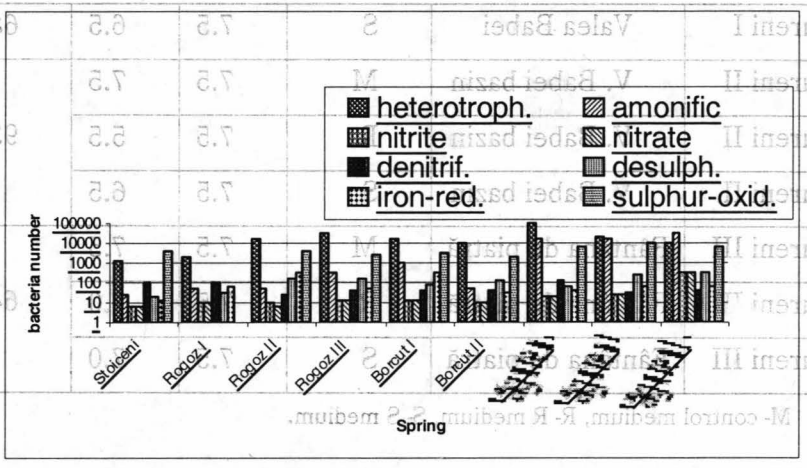

Fig. 1. Ecological groups of bacteria in mineral water springs.

From this table we can see that mesophilic heterotrophic bacteria are present in a greater number in the springs Babei Valley, Babei Valley basin and La fântână, which shows there are rainwater infiltration and the other springs do not offer favorable conditions for the development of this group of microorganisms.

The bacteria concerned in the sulphur cycle along side the sulphur-oxidizing bacteria, the presence of sulphate-reducing bacteria was present in all of the springs, but without establishing their number. The desulphuring bacteria are present in the springs at Măgureni. The iron-reducing bacteria were found in relatively small number in all the researched springs.

The data regarding the hygienic-sanitary parameters are given in the following table:

Table 3. Hygienico-sanitary parameters

Water source	Spring	Mesophylic heterotrophic bacteria No./100 ml	Total coliforms bacteria No./100 ml	Faecal oliforms bacteria No./100 ml	Faecal nterococcus No./100 ml
Stoiceni	La casă	12000	2	-	-
Rogoz I	Râu	21000	3	-	-

Rogoz II	Peste râu	130000	1	-	-
Rogoz III	La fântână	120000	2	1	-
Borcut I	Lângă vale	150000	3	4	-
Borcut II	Pe păşune	120000	6	3	-
Măgureni I	Valea abei	500000	10	7	-
Măgureni II	V. Babei-bazin	240000	10	5	-
Măgureni III	Fântâna de piatră	90000	11	3	-

The number of total coliform bacteria is reduced, being admitted by STAS. The faecal coliform bacteria indicate a recent faecal pollution are not present in the first 3 springs and are present in small numbers in the following 6. The presence of total coliform bacteria in these 6 springs can be explained, the samples were taken in a period rich in a raing period and there were infiltrations into these springs from the surface. Faecal enterococcus have not been found, so the mineral waters can be consumed without an epidemiological danger.

Conclusions.

The microbiological analysis of the 9 springs in the Depression of Lăpuş emphasize a rich microbiota: sulphur-oxidizing, sulphate-reducing, desulphurizing, ammonifyier, denitrifyier, nitrifyier, iron-reducing, mesophilic heterotrophic bacteria. The presence of these groups of bacteria is closely linked to the ecologic and physico-chemical conditions of the mineral deposit, and the hygienic-sanitary characteristics of the extraction facilities.

The mineral springs at Măgureni present a significant microbiota, these springs being of sulphurous nature, however, a high number of mesophilic heterotrophic bacteria were found, due to the fact that these springs are not taken care of very good, two of the spring (Babei Valley and Babei Valley basin) being left to waste, although they have remarkable qualities.

The mesophilic heterotrophic bacteria present in small numbers prove that the studied mineral waters are oligotrophic. The spring at Stoiceni has a small microbiota, this spring being a new one appeared in the basement a house. The springs situated south of this, have been mentioned in balneology since 1834. Among the mineral waters in Romania which have high salinity and contain chlorids the percentage of CO_2 which contributes to total mineralisation exceeds 26% only at Covasna while at Stoiceni it exceeds 23%.

The hygienic-sanitary parameters are between natural limits measures for improving the exploitation facilities should be taken in the case of the springs at Măgureni, wich are left to waste.

The presence of physiological groups of bacteria and absence of faecal enterococci represents an eloquent proof of the absence of toxic substances from these mineral water springs.

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THE COMPARATIVE STUDY ABOUT HYGIENIC-SANITARY STATE OF TWO DAM RESERVOIR FROM SOMEŞUL MIC UPPER BASIN

Manuela-Claudia CURTICĂPEAN*, Mihail DRĂGAN-BULARDA**

Rezumat. . Ţinând seama de utilizarea complexă a lacurilor de acumulare Gilău şi Tarniţa, apa şi sedimentul acestora au fost studiate din punct de vedere bacteriologic, prin determinarea a trei indicatori igienico-sanitari: număr probabil de coliformi totali, coliformi fecali şi enterococi fecali. Probele de apă şi sediment au fost prelevate sezonier, în anul 2003. Comparând rezultatele obţinute cu valorile maxime din Normativul privind obiectivele de referinţă pentru clasificarea calităţii apelor de suprafaţă, elaborat în 2002 (ce înlocuieşte STAS 3001-91) s-a observat faptul că atât pentru lacul Gilău, cât şi pentru lacul Tarniţa, majoritatea secţiunilor s-au încadrat în categoria I de calitate. În cazul lacului Tarniţa s-a constatat faptul că prezenţa plajelor şi a docurilor pentru bărci, în zonele periferice ale lacului, are o influenţă negativă asupra lui, aici înregistrându-se o densitate mai ridicată a bacteriilor coliforme decât în zonele centrale ale lacului, fenomen accentuat şi de activitatea recreaţională. Deoarece, atât în cazul lacului Gilău, cât şi în cazul lacului Tarniţa, valorile rapoartelor CF/EF au fost mai mari decât 4 (în secţiunile unde enterococii fecali au fost prezenţi), indică prezenţa unei surse umane de impurificare a apelor şi sedimentelor celor două lacuri.

The Gilău and Tarniţa dam reservoirs, are positioned in the Someşul Mic upper basin that has an area of 3804 km^2 and a length of 167 km (··· 1992). The Someşul Mic upper basin is a part of the Someş-Tisa hydrographic basin, which extend on 22.380 km^2, including the main water courses from intracarpathian space - Tisa, Someş, Crasna and Tur - water courses which form and convey in transit the state border.

In upper basin of the Someşul Mic river have been build a dam reservoirs succession (big dam reservoires disposed in a „water fall" system), most important one Fântânele-Tarniţa-Someşul Cald-Gilău.

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The dam of the *Tarniţa* reservoir, positioned upstream the Gilău lake, has been placed into the Someşul Cald valley's narrowing sector. The reservoir depresion is sculptured in crystalline schists covered with quaternary sands and gravels. The dam of the *Gilău* reservoir was build into the Someşul Cald valley's narrowing sector at the out going from the mount space. The reservoir depresion was build into the erosion basin outlined by the Someşul Cald river with Someşul Rece river confluence (Şerban, 1999).

The *Tarniţa* artificial lake, that was opened for use in 1973, represent one of the most severe anthropic effect on the natural course of the Someşul Mic river. The reservoir, with a capacity of 77,4 million m^3 water (table 1), has the following functions: mainly- energetics, the averting of the floods, to attenuate the floods waves and, in the future, supply with drinking water for Cluj-Napoca city (...).

The *Gilău* artificial lake, positioned downstream the Tarniţa lake is the first reservoir opened for use in 1972. The reservoir, with a capacity of 4,1 million m^3 water (table 1), has the following functions: mainly- to supply with drinking and industrial water for Cluj-Napoca city and the nearby vilages (the Gherla city and the Aghireş area), energetics, the averting of the floods, to attenuate the floods waves and to suplly with water the Gilău troutdes (...).

Table 1 The characteristics of the two dam reservoirs from the Someşul Mic upper basin (Şerban, 1999)

Water course	Dam reservoir	Surface (ha)	Length (km)	Height of the dam (m)	Capacity (mill. m^3)
Someşul Cald	Tarniţa	220	8	97	77,4
Someşul Mic	Gilău	72	2	23	4,1

The bacteriological studies of the water and sediments from Tarniţa and Gilău dam reservoirs have a special signifiance, the studied bacteriological indicators can serve as appreciation criteria for the evolution of water quality, especially in some critical situations (the discharge of the accumulation lakes, floods, drought), but also as basis for the decision in case of bringing to normal measures the aquatic ecosystems (Cuşa, 1996).

The sediments constitute a key link in the biogeochemical cycle of the elements in the aquatic systems. There are finalized the mineralisation processes of the organic substances that were not degraded in the water

column. The bacteriological indicators have, in sediments, constancy and bigger stability degree, being less influenced by some changes of the environmental conditions (comparative with waters) and, therefore, reflect in time the evolution of water quality. The enzymological researches on the sediment of the accumulation lake, followed the knowledge completion about the complex processes that happen in these habitates, with a special semnification, especially for the ecosystems with an ecological value, like the dam reservoir that suplly with drinking water, needs an increased exigency for bacteriological water quality (Cuşa, 1996).

Materials and methods

The bacteriological analyses were performed in 2003 on water and sediment samples collected as follows: Gilău lake - in March, May and October; Tarniţa lake - in April, September and October.

The water samples were taken seasonal, from surface but also from different depths in a vertical profile of the lake's water. Also, it was taken into account the areas of lakes: dam, middle and tail of the lake. For the *Gilău* lake have been established 8 sampling sites of the water samples: Dam - 0 m, Dam - 3 m, Dam - 6 m, Dam - bottom, Middle lake - 0 m, Middle lake -bottom, Tail lake - 0 m, Tail lake - bottom. For the *Tarniţa* dam reservoir, sampling sites of the water samples were 15: Dam - 0 m, Dam - 5 m, Dam - 10 m, Dam - 15 m, Dam - right border 0 m, Dam - left border 0 m, Middle lake - 0 m, Middle lake - 5 m, Middle lake - 10 m, Middle lake - beach I right border, Middle lake - beach I left border, Middle lake - beach II right border a and b, Tail lake - right border and Tail lake - left border.

The sediment samples were taken also seasonal, in the same time with water samples at 0-10 cm depth. For the *Gilău* dam reservoir have been choosed 7 sampling sites of the sediment: Dam -right border, Dam - left border, Dam - middle, Middle lake - right border, Middle lake - left border, Middle lake - middle and Tail lake - middle. For the *Tarniţa* dam reservoir have been choosed 10 sampling sites of the sediment: Dam - right border, Dam - left border, Middle lake - beach I right border, Middle lake - beach I left border, Middle lake - beach II right border a and b, Tail lake - right border a and b and Tail lake - left border a and b.

The bacteriological analyses were aimed for the determination of three hygienic-sanitary indicators, namely: the probable number of the total coliform bacteria, faecal coliform bacteria and faecal enterococcus.

In order to determine the bacteriological indicators from water samples we used the multiple tubes methods according to *Drăgan-Bularda* (2001). In order to determine the hygienic-sanitary indicators from sediment samples we used the methods according to *Cuşa* (1996). Due to the fact that the sediment categories may have a variable water content, that can influence the expression of the microbian charge reporting to the sediment weight, the humidity of each sediment sample was established during their preparation for the analyses (Cuşa, 1996).

The results obtained were compared with the values from „The normative regarding the reference objectives for clasification of the surface water quality", issued in 2002 (this replaces STAS 3001-91) that suppose analyses of two indicators (total coliform bacteria and faecal coliform bacteria) in order to microbiologically characterize the water.

In order to establish the nature of lake's water faecal pollution we used an index which represent the ratio between the number of faecal coliform bacteria (FC) and faecal enterococcus (FE) (Barbato et al., 1990; Cuşa, 1996).

Results and discussion

The water's sanitary quality was estimated based on presence or absence of pathogenic microorganisms or that ones indicating the possibility of their presence. The water may be dangerous for health and life, as a pathogenic microorganism potential carrier. The main pollution bacteriological indicators from waters and sediments are: the group of total and faecal coliform bacteria (as a major indicator of faecal water contamination) and faecal enterococcus. Depending on the isolation and association of these bacteria groups and on their quantity variation, one can appreciate the hygienico-sanitary state of the water and sediment.

The generic term „coliform bacteria" designate a category of bacteria which belong to the *Enterobacteriaceae* group, characterized by a common series of properties, from which one essential for their definition is the capacity to produce fermentation of lactose at 35-37°C with acid and gas production in 24-48 hours and secandly is the capacity to grow in the

presence of the biliary salts and other agents with surface action. The coliform bacteria that present these metabolic caracteristics also at 44°C are „faecal coliforms" or „thermotolerants". Although, the coliform bacteria have not an exclusive faecal origin, there are increased quantities in human and animals with warm blood faecal, fact to allow decelation of this group even at a considerable dilution. Their presence in the natural environments indicate a recent faecal contamination (Diudea et al., 1986; Madigan et al., 2000; Millea, 2001; Kenneth, 2003).

In accordance with morphological and biochemical caracteristics, the coliform group is formed by bacteria belonging to the following species: *Escherichia, Citrobacter, Klebsiella, Erwinia, Enterobacter, Serratia, Yersinia.* All these species involve Gram-negative, unsporulated, aerobic and optional anaerobic bacteria (Diudea et al., 1986; Millea, 2001).

The species from the coliform bacteria group are not patogen, strictly speaking, but, in some conditions, can often induce infections of the urinary tract. Therefore, they are considered opportunist patogens. Due to this fact, the coliform germs are considered as indicator organisms of high importance. As higher as is their number in the natural environment this increases the probability of the presence for patogen microorganisms in the environment.

Table 2 The results of the bacteriological analyses performed on water and sediment of the Gilău dam reservoir (average values for 2003)

Sampling sites	Total coliform bacteria (no/100ml) (no/g.d.s.)	Quality class (according to the normative)	Faecal coliform bacteria (no/100ml) (no/g.d.s.)	Quality class (according to the normative)	Faecal enteroc occus (no/100 ml) (no/g.d.s.)	**FC/FE ratio**
Water samples						
1. Dam- 0 m	753	I	748	I	6	124.6
2. Dam- 3 m	1559	I	1119	I	12	93.2
3. Dam- 6 m	5726	I	816	I	2	408
4. Dam- bottom	30741	II	7393	II	3	2464.3
5. Middle- 0 m	606	I	390	I	0	-
6. Middle-bottom	809	I	614	I	2	307
7. Tail- 0 m	416	I	603	I	0	-
8. Tail-bottom	1417	I	1369	I	4	342.2
Sediment samples						
1. Dam- middle	86	-	59	-	0	-
2. Dam-right border	372	-	281	-	21	13.3

3. Dam left border	217	-	155	-	13	11.9
4. Middle-middle	89	-	61	-	0	-
5. Middle-right border	206	-	163	-	27	6
6. Middle-left border						
7. Tail- middle	152	-	121	-	13	9.3
	140	-	95	-	0	-

g.d.s. = g of dry sediment

The enterococcus are Gram-positive, unsporulated, optional anaerobic germs, they have not ciliums and have form like coccis and bacillus. The normal habitat of these germs is the intestinaly tract of human and warm blooded animals. Also, their presence in water indicate its faecal contamination. They are more resistant than *Escherichia coli* to environmental influences.

The Gram-positive pathogen germs (*Staphylococcus aureus, Streptococcus pyogenes* and *Streptococcus pneumoniae*) and the Gram-negative pathogen germs (*Neisseria gonorrhoeae* and *Neisseria meningitidis*) are part of enterococcus group. Have been estimated that these germs challenge one third from all the human bacterian infections, including pneumonia, different types of septic shock, gonorrhoea and meningitidis (Madigan et al., 2000; Kenneth, 2003).

In the water of Gilău and Tarnița dam reservoirs have been established that there is a vertical quantitatively distribution of total and faecal coliform bacteria, their number being higher with depths, because of their accesibility to organic matter (tables 2 and 3) (Ailiesei and Jâpa, 1995; Ailiesei et al., 1998; Jâpa and Ailiesei, 1999). The increase of the bacteria number with the depths may be determined by the effect of the sedimentation of solid particles (fig. 1 and 3). Through sedimentation, the solid particles draw with them the bacteria to the deeper layers, their number therefore decreasing in the surface zones (Millea, 2001). The values of the faecal enterococcus indicator has not been detectable in all the water samples. In Gilău and Tarnița dam reservoir have been registered detectable values but low ones, especially in water sampled from high depths and peripherical zones (Tarnița dam reservoir - in two beaches).

Table 3 The results of the bacteriological analyses performed in water and sediment of the Tarnița dam reservoir (average values for 2003)

Sampling sites	Total coliform bacteria (no/100 ml) (no/g.d.s.)	Quality class (according to the normative)	Faecal coliform bacteria (no/100ml) (no/g.d.s.)	Quality class (according to the normative)	Faecal enterococcus (no/100 ml) (no/g.d.s.)	FC/FE ratio
Water samples						
1. Dam- 0 m	19	I	19	I	0	-
2. Dam- 5 m	1477	I	1163	I	0	-
3. Dam- 10 m	2007	I	1403	I	7	200.4
4. Dam- 15 m	3067	I	2310	II	12	192.5
5. Dam-right border 0 m	28	I	28	I	0	-
6. Dam-left border 0 m	67	I	67	I	0	-
7. Middle- 0 m	62	I	52	I	0	-
8. Middle- 5 m	1350	I	955	I	0	-
9. Middle- 10 m	1400	I	1200	I	0	-
10. Middle- beach I right border	12333	II	6200	II	78	79.4
11. Middle- beach I left border	865	I	635	I	0	-
12. Middle- beach II right border a	8200	I	5350	II	50	107
13. Middle- beach II right border b	7100	I	4100	II	55	74.54
14. Tail – right border	2933	I	2067	II	0	-
15. Tail – left border	2433	I	2000	II	0	-
Sediment samples						
1. Dam-right border	1295	-	974	-	0	-
2. Dam- left border	2184	-	1604	-	0	-
3. Middle- beach I right border	19718	-	11147	-	0	-
4. Middle- beach I left border	2485	-	2060	-	0	-
5. Middle- beach II right border a	6930	-	5410	-	0	-
6. Middle- beach II right border b	7188	-	5668	-	0	-
7. Tail – right border a	2031	-	1614	-	0	-
8. Tail – right border b	2163	-	1761	-	0	-
9. Tail – left border a	2498	-	2027	-	0	-
10. Tail – left border b	2610	-	2250	-	0	-

Concerning the seasonal distribution of the coliform bacteria and faecal enterococcus it has been established that there are many oscillations of bacterial number, recording the minimum values in the cold season and maximum values in the warm season (Millea et al., 1993).

The bacteriological analyses of the sediment samples have confirmed the results obtained for the water samples. Thus, the maximum values of total and faecal coliform bacteria have been established in the peripherical zones of reservoires (tables 2 and 3). For Tarnița dam reservoir (fig. 4) have been established an accented increase of the values of these indicators, in the beach areas, where are human establishments and summer, the holiday activities are enhanced.

Unlike the peripherical zones of the Gilău dam reservoir (fig. 2), where have been recorded small values of the faecal enterococcus in the sediment samples, in the Tarnița dam reservoir the faecal enterococcus missed from the sediment samples.

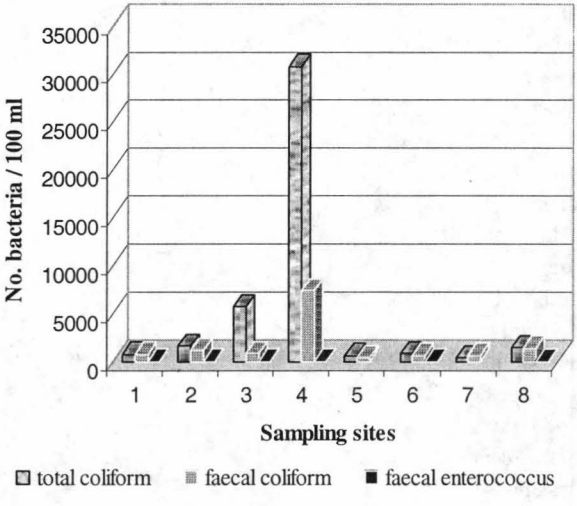

Fig.1 Bacterial average values from the Gilău dam reservoir water samples.

By comparison of the obtained results with the maximum values from „The normative regarding the reference obictives for clasification of the surface water quality", issued in 2002

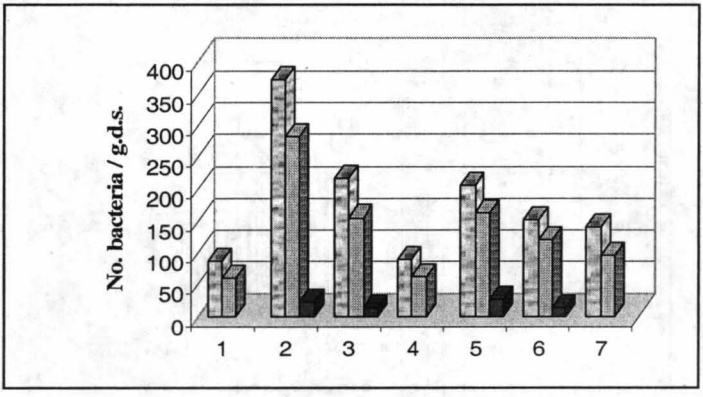

Fig. 2 Bacterial average values from the Gilău dam reservoir sediment samples

(this replaces STAS 3001-91) it has been observed that for each dam reservoir Gilău and Tarnița, the majority of the sections have been framed in the Ist quality category (tables 2 and 3). For the Gilău dam reservoir (fig. 1), there was an exception, namely Dam -bottom section, which has been framed in the IInd quality category for total and faecal coliform bacteria. For the Tarnița dam reservoir (fig. 3), Middle -beach I right border section has been framed in the IInd quality category for total coliform bacteria and Dam - 15 m, Middle - beach I right border, Middle - beach II right border a and b, Tail - right border and Tail -left border sections have been framed in the IInd quality category too, for the faecal coliform bacteria.

In the peripherical zones of the Tarnița dam reservoir it has been established that the presence of the beaches and docks for boats have a negative influence, there being recorded an increased density of the coliform bacteria as in the middle zones of the reservoir; this phenomenon being enhanced by the holiday activity too. More, it has been determined an increased density of the coliform bacteria in sediment samples, in comparison with water samples of the reservoir (An et al., 2002), possible because of bacteria's skill to form mixtures with the sediment particles and to use some present substances in that enviroment (Dean-Ross et al., 2002).

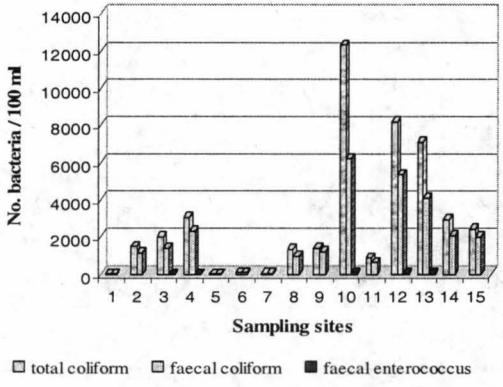

Fig. 3 Bacterial average values from the Tarnița dam reservoir water samples.

To establish the nature of the faecal pollution of water and sediment for the studiated reservoirs, we used an index which perform the ratio between number of faecal coliform bacteria (FC) and faecal enterococus (FE). A value of the ratio higher than 4 (FC/FE > 4) emphasize an human source of pollution. When the ratio is between 2 and 4 (2 < FC/FE < 4), it is a mixed pollution prevailing the human source, but when the ratio is between 0,7 and 1 (0,7 < FC/FE < 1), it is a mixed pollution prevailing the animal source. The pollution with domestic animals dejections is charaterized by a lower value of the ratio, namely 0,7 (FC/FE < 0,7) (Barbato et al., 1990; Cuşa, 1996).

Because, for the Gilău dam reservoir and Tarnița dam reservoir too, the values of the CF/EF ratios were higher than 4 (table 2 and 3), indicate the presence of the human source of impurificaton in the water and sediment.

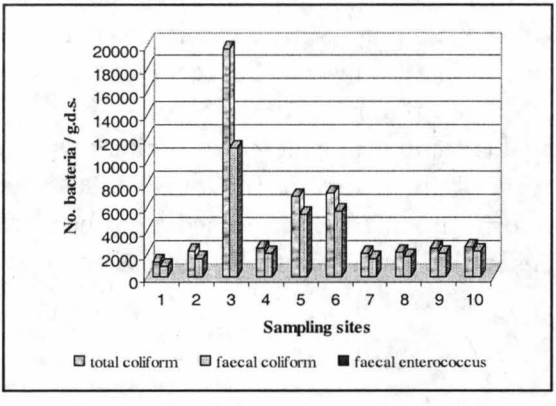

Fig. 4 Bacterial average values from the Tarnița dam reservoir sediment samples.

Conclusions

1. In the water of the Tarniţa dam resevoir and Gilău dam reservoir too, it has been establised that there is a vertical quantitative distribution of the total and faecal coliform bacteria, their numbers being higher with the depth, because of their accessibility at organic matters. For the Gilău and Tarniţa dam reservoirs, low values of the faecal enterococcs have been recorded especially for the deep water samples and for the peripherical areas (Tarniţa dam reservoir - in the beaches areas).

2. In the Gilău and Tarniţa dam reservoir it has been recorded the maximum values of the total and faecal coliform bacteria in the peripherical areas; for the Tarniţa dam reservoir has been established an increased values of these indicators, in the two beaches areas, along human establishments and where, summer, holiday activities are enhanced.

3. By comparison of the obtained results with the maximum values from „The normative regarding the reference obictives for clasification of the surface water quality", issued in 2002 (this replaces STAS 3001-91) it has been observed the fact that for each dam reservoir Gilău and Tarniţa, the majority of the sections have been framed in the I^{a} quality category. In the peripherical zones of the Tarniţa dam reservoir have been established that the presence of the beaches and docks for boats have a negative influence, there being recorded an increased density of the coliform bacteria as in the middle zones of the reservoir; this phenomenon being enhanced by the holiday activity too.

4. Because, for the Gilău and Tarniţa dam reservoirs, the values of the CF/EF ratios were higher than 4, indicates the presence of the human source of impurificaton of the water and sediment.

5. The results of the bacteriological analyses on the water and sediment of the Gilău and Tarniţa dam reservoirs, indicated the fact that these two lakes have a good hygienic-sanitary state but, taking into consideration the potential infectious of these germs and the danger what they constitute for the health imposes the application of measures for the reservoir water quality protection, considering also its complex use.

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BACTERIOLOGICAL AND ENZYMOLOGICAL RESEARCHES ON THE ACTIVATED AND ANAEROBIC MESOPHILIC DIGESTED SLUDGE FROM BISTRIŢA WASTEWATER TREATMENT PLANT

Andreea MÎNDRUŢ*, Mihail DRĂGAN-BULARDA**

Rezumat: Cercetări bacteriologice şi enzimologice asupra nămolului activ şi fermentat de la Staţia de Epurare Bistriţa. Nămolul activ reprezintă o comunitate microbiană complexă, a cărei structură depinde de sănătatea populaţiei şi parametrii operaţionali ai staţiei de epurare. Studiul bacteriologic şi enzimologic al nămolului activ şi fermentat de la Staţia de Epurare Bistriţa s-a focalizat pe compoziţia şi dinamica microbiotei în cursul procesului de tratare a apei uzate municipale. Probele au fost recoltate din decantorul secundar şi, respectiv, din metantanc. Densităţile bacteriilor nitrificatoare şi denitrificatoare au fost determinate în probele de nămol activ. Celelalte analize bacteriologice au implicat determinarea a 4 indicatori în nămolul activ şi nămolul fermentat: bacterii heterotrofe mezofile, coliformi totali, coliformi fecali şi enterococi fecali. Pe baza densităţilor s-a calculat gradul de îndepărtare prin fermentare anaerobă mezofilă a fiecărui indicator. Analizele enzimologice le-au completat pe cele bacteriologice cu scopul aprecierii potenţialului enzimatic al nămolului activ şi fermentat. În ambele tipuri de nămol s-au determinat activitatea dehidrogenazică actuală şi potenţială, activitatea fosfatazică şi activitatea catalitică totală. Pe baza valorilor relative ale activităţilor enzimatice s-au calculat indicatorii enzimatici ai calităţii nămolului activ şi fermentat. Valorile acestor indicatori au fost cuprinse între 0,262 şi 0,377 pentru nămolul activ şi 0,277-0,400 pentru nămolul fermentat, ceea ce indică un potenţial enzimatic mediu.

Introduction

Activated sludge, a common biological treatment method for municipal and industrial wastewater represent a complex microbial community. This biological treatment may reduce the number of microorganisms, especially the number of pathogens, in the wastewater and the sewage sludge by creating adverse conditions for pathogens survival[4].

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Due to intricate interactions within the microbial community, process control of wastewater treatment plants can be difficult. Population shifts within the microbial community may result from changes in the plant operating conditions and cause sludge quality problems such as poor sludge settling, compaction and dewatering.

The bacteriological studies of the activated and digested sludge from Bistrița wastewater treatment plant focused on the broader effects of the treatment on sewage sludge microbiota, highlighting the reduction rates of the bacteriological indicators in sludge through anaerobic mesophilic digestion.

Monitoring the levels of indicator species from the digested sludge has a special significance in the case of land application of biosolids. The indicator and representative organisms studied are ones that have been found to respond to treatment process and environmental conditions in a manner similar to other organisms, therefore they provides information about the survival of the larger group[5].

The enzymological researches yielded informations about the complex processes that happen in the activated sludge and the fertilisation potential of the digested sludge.

In the last two decades in our country many enzymological investigations were performed on activated sludges from industrial treatment plants and on soils fertilized with digested and dried sludge witch exhibited enzyme activities[3, 7, 10]. There are no microbiological and enzymological analyses concerning the activated and anaerobic digested sludge from Bistrița wastewater treatment plant.

Materials and methods

The bacteriological and enzymological analyses were performed in March, August and November 2003 on activated sludge samples and anaerobic mesophilic digested sludge samples collected from Bistrița wastewater treatment plant.

The activated and digested sludge samples were taken from the secondary settling tank and the anaerobic digestion tank, respectivelly.

There have been determined two physiological groups from activated sludge, namely the nitrifying and dentrifying bacteria, and four hygienico-sanitary indicators from both, activated and digested sludge, namely: the total

number of the mesophilic heterotrophic bacteria (37°C)(CFU=colony- forming units), the probable number of the toatal coliform bacteria, the probable number of the faecal coliform bacteria and the probable number of the faecal enterococcus.

In order to determine the nitrifying bacteria we used casein broth with potassium nitrite supplement and the presence of the nitrifiers were studied after dyphenilamine addition to the inoculated medium. For the denitrifying bacteria determination we used tryptic soy broth(Merck) according to Tiedje[14].

The bacteriological indicators were determined using the next media: the gelose for mesophilic heterotrophic bacteria, the lauryl sulphate broth and Geam-Levine media for coliform bacteria, the lauryl sulphate broth for faecal coliform, the sodium azide media and the sodium and purple bromcresol azide media for faecal enterococcus[15].

The enzymological analyses were performed on the both sludges and we determined the actual and potential dehydrogenase activity according to Thalmann[13], the phosphatase activity according to Tabatabai and Bremner[12] and the total catalytic activity according to Drăgan-Bularda[6]. We calculated the enzymatic indicators of the sludge quality using the relative values of the enzymatic activities[9].

Results and discussion

The polarized relationship between the nitrifying and denitrifying bacteria is a problem in the sludge testing because each of this two physiological groups either produces or uses the nitrate. That's why is so important to study comparatively this two bacterial groups in activated sludge. The densities of the nitrifying and denitrifying bacteria are presented in table 1.

The results indicate the maximum values of the number of nitrifying and denitrifying bacteria in August, with a higher density for the nitrifying bacteria(fig. 1). This increase in the nitrifying bacteria number may be due to the effects of the increasing temperature, the sludge's age and the excessive nutrient levels in primary effluent. At the same time, the excessive development of oxygen consumer microorganisms in summer increases the nitrate concentrations and creates the anoxic zones in the interior parts of the sludge flocs, where starts nitrate reduction by the denitrifying bacteria.

Less dense flocs are characterized by a lower denitrification rate due to the lack of anoxic zones. Floc flotation, caused by N_2 gas bubbles resulting from the dissimilatory reduction of nitrate is one of the multiple aspects of the phenomenon of bulking sludge, frequently identified to Bistrița wastewater treatment plant.

The bacteriological indicators have been determined in all the activated and digested sludge samples. The densities of indicator bacteria in activated and digested sludge are presented in table 2. The values drop in the case of the digested sludge for all the indicators. In activated sludge the maximum values of all the indicators (mesophilic bacteria: $148x10^6$ CFU/ml; total coliforms: $23x10^7$/100 ml; faecal coliforms: $4.9x10^7$/100 ml and faecal enterococcus: 2000/100 ml) have been reported in August. For digested sludge the maximum values also detected in August were: mesophilic bacteria-$105x10^6$ CFU/ml; total coliforms-$6.3x10^6$/100 ml; faecal coliforms-$2.6x10^6$/100 ml and faecal enterococcus-400/100 ml.

The reduction rates of the bacteriological indicators in sludge show the sludge treatment efficiency[11]. Due to this fact we determined the percentage and logarithmic reduction of the indicator bacteria through anaerobic mesophilic digestion. The results are presented in table 3

Table 1. Numerical evolution of the nitrifying and denitrifying bacteria in activated sludge.

Analyses period	Nitrifying bacteria MPN*/100 ml	Denitrifying bacteria MPN/ml
April	$11x10^6$	$5.4x10^3$
August	$26x10^6$	$6.4x10^3$
November	$9x10^6$	$5.6x10^3$

* MPN = most probable number

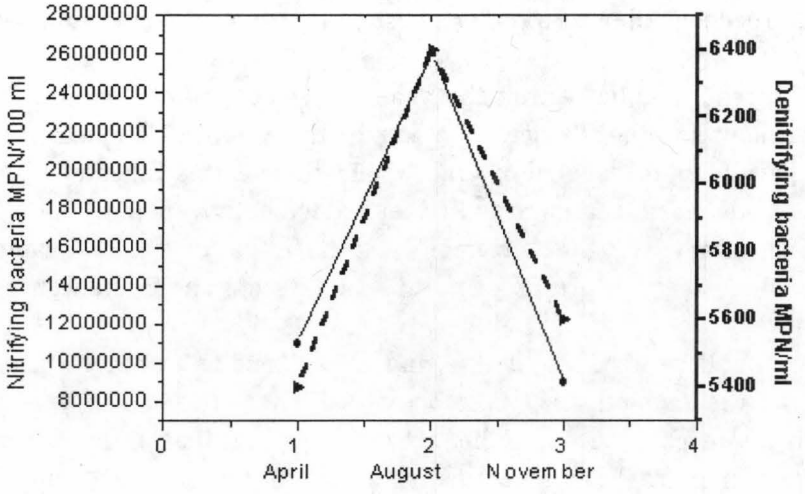

Fig. 1: Comparative evolution of the nitrifying and denitrifying

Table 2. Hygienic-sanitary state of activated and anaerobic digested sludge. **bacteria densities in the activated sludge.**

Analyses period	Sludge type	Hygienic bacteriological parameters			
		Mesophilic heterotrophic bacteria (CFU/ml)	Total coliform bacteria (MPN/100 ml)	Faecal coliform bacteria (MPN/100 ml)	Faecal enterococcus (MPN/100 ml)
April	Activated	$74.5x10^6$	$16x10^7$	$2.2x10^7$	800
	Digested	$56x10^6$	$2.5x10^6$	$1.7x10^6$	200
August	Activated	$148x10^6$	$23x10^7$	$4.9x10^7$	2000
	Digested	$105x10^6$	$6.3x10^6$	$2.6x10^6$	400
November	Activated	$91x10^6$	$14x10^7$	$1.75x10^7$	200
	Digested	$71x10^6$	$2.7x10^6$	$1.4x10^6$	< 200

In November the percentage and logarithmic reduction couldn't be calculated for faecal enterococcus because of the broad field of most probable number value in the digested sludge. The reduction rates indicate that the

total coliforms and the faecal coliforms are removed more than 90% and the faecal enterococcus about 75%. The mesophilic bacteria have less reduction rates.

The results indicate that the anaerobic mesophilic digestion is not a good treatment for the sludge disinfecting, the remove of faecal indicator bacteria having a low level. For land application of this digested sludge is required an advanced treatment to further reduce pathogens[5,8].

The results of the enzymological analyses of the activated and digested sludge samples are presented in the tables 4 and 5 and figures 2, 3, 4 and 5.

In the both type of sludge and in all sludge samples have been detected all the studied enzymatic activities. The potential dehydrogenase activity is more intense than the actual one, fact that reflects that the glucose(carbon source) has a stimulating action on the enzymes synthesis by the microorganisms. This influence of the added glucose is more striking on activated sludge samples in November (1.550 mg triphenilformazan/g.d.s.)(g.d.s. = gram dried sludge). The weak stimulating effect of the added glucose on he dehydrogenase activity from the others sludge samples(activated and digested sludge) is due to the accumulation in sludge of high amounts of organic matter from primary effluent, which allow the sludge to be used as a soil conditioner and, on the other hand, provide a good development of microorganisms with a relative intense actual dehydrogenase activity[1,9].

The phosphatase activity has a higher level in the activated sludge than in the digested one. Maximum values were obtained in August: 0.120 mg p-nitrophenol/g.d.s. for activated sludge and 0.113 mg p-nitrophenol/g.d.s. for digested sludge.

We observed that the catalityc activity is more intense in the digested sludge with a maximum level in August (31.22 mg H_2O_2/g.d.s.) than in activated sludge where the activity is 28,17 mg H_2O_2/g.d.s..

The values of the enzymatic indicator of the quality are the expression of the sludge's enzymatic potential. Theoretically the enzymatic indicator may exhibit values between 0 (no activity in the samples) and 1 (all the real individual values are equal to the maximum theoretical individual values of all activities) [9]. Both activated and digested sludge have a similar enzymatic potential with maximum values in November for the activated sludge (0.337)

Table 3. Percentage(%) and logarithmic(log) reduction of the indicator bacteria through anaerobic mesophilic digestion.

and in August for the digested sludge (0.400). The anaerobic digested sludge exhibited a better potential than the activated sludge.

Analyses period	Reduction rates							
	Mesophilic heterotrophic bacteria		Total coliform bacteria (MPN/100 ml)		Faecal coliform bacteria (MPN/100 ml)		Faecal enterococcus (MPN/100 ml)	
	%	log	%	log	%	log	%	log
April	24.83	1.40	98.43	8.19	92.27	7.30	75.00	2.77
August	29.05	1.46	97.26	8.34	94.69	7.66	80.00	3.20
November	21.97	1.35	98.06	8.13	92.00	7.20	-	-

Table 4. Evolution of the enzymatic activities in the activated sludge.

Analyses period	Activity exprima-tion	Dehydrogenase activity (mg triphenilformazan/g.d.s.)		Phosphatase activity(mg p-nitrophenol/ g.d.s.)	Total catalytic activity (mg H_2O_2/g.d.s.)	EISQ *
		actual	potential			
April	a.v.**	0.152	0.261	0.116	28.05	0.262
	r.v.***	49.51%	16.83%	96.66%	99.57%	
August	a.v.	0.307	0.579	0.120	28.17	0.337
	r.v.	100%	37.35%	100%	100%	
November	a.v.	0.261	1.550	0.108	27.95	0.377
	r.v.	85.01%	100%	93.10%	99.21%	

* EISQ= enzymatic indicator of sludge quality; ** a.v.= absolute value; *** r.v.= relative value.

Table 5. Enzymatic activity analyses performed on the anaerobic mesophilic digested sludge.

Analyses period	Activity exprima-tion	Dehydrogenase activity(mg triphenilformazan/g.d.s.)		Phosphatase activity(mg p-nitrophenol/ g.d.s.)	Total catalytic activity (mg H_2O_2/g.d.s.)	EISQ
		actual	potential			
April	a.v.	0.127	0.195	0.108	31.03	0.277
	r.v.	44.87%	38.08%	95.57%	99.39%	
August	a.v.	0.283	0.512	0.113	31.22	0.400
	r.v.	100%	100%	100%	100%	
November	a.v.	0.221	0.302	0.093	30.84	0.318
	r.v.	78.09%	58.98%	82.30%	98.78%	

It is difficult to establish if the enzymatic activities are less intensely because of one or more of the next parameters: effects of heavy metals(Zn, Cr, Pb) detected in the Bistrița digested sludge, nature of the sludge, activity of microbiota or enzymes synthesis[2].

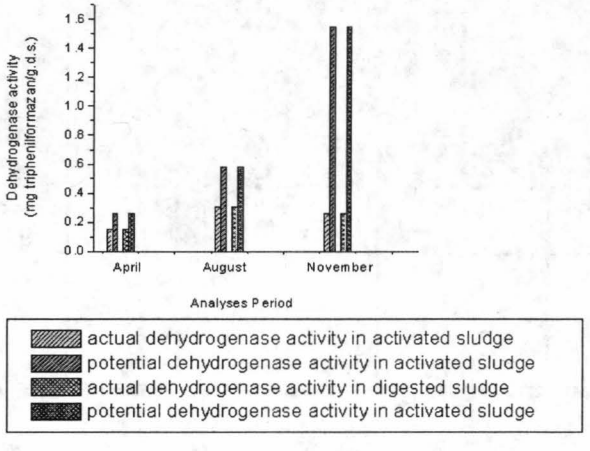

Fig. 2: Actual and potential dehydrogenase activity in the activated and digested sludge

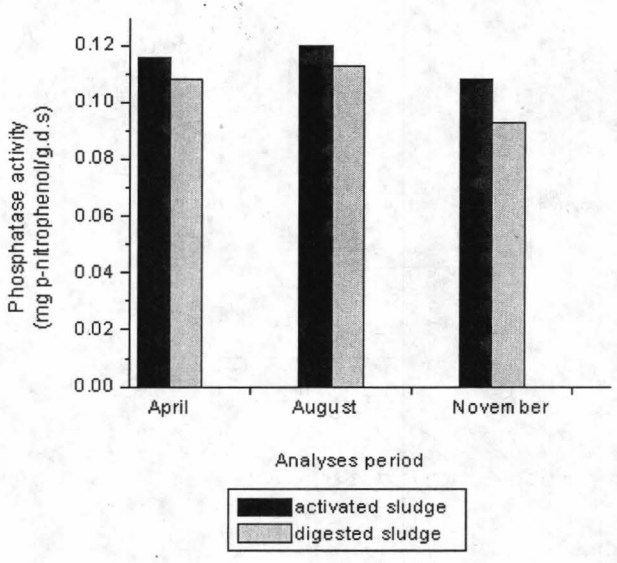

Fig. 3: Phosphatase activity in the activated and digested sludge

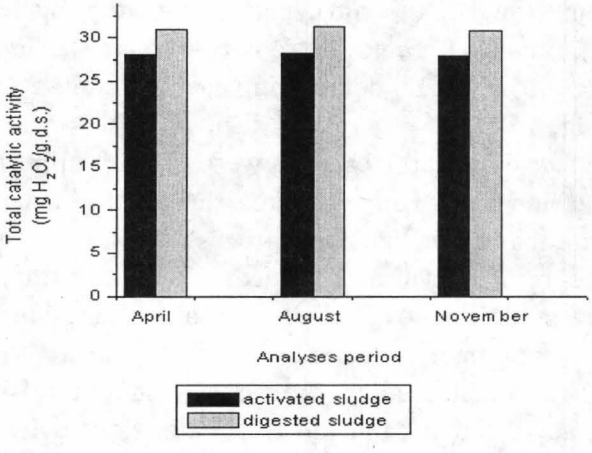

Fig. 4: Total catalytic activity in the activated and digested sludge.

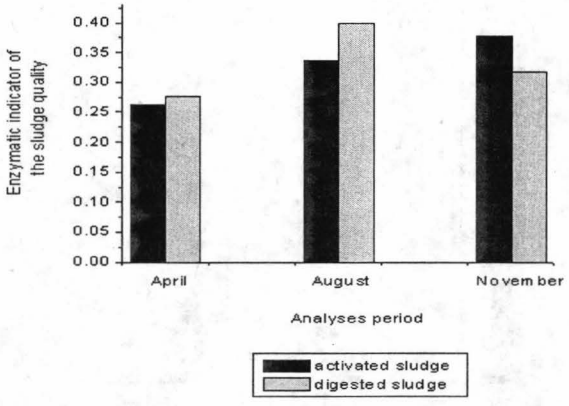

Fig. 5: Enzymatic indicators of the activated and digested sludge quality

Conclusions

In the absence of the third step of wastewater treatment which involves nitrification/denitrification and phosphorus removal, it is required the parallel monitoring of the nitrifying and denitrifying bacteria for their capacity of NH_4^+ and NO_3^- removal from the treated effluent. The results indicate that the nitrifying and denitrifying bacteria keep a numerical balance in activated sludge.

The faecal indicator bacteria were present at normal levels in the activated sludge and their removal rates throuh the anaerobic mesophilic digestion indicate that if this biological process meets the required resistence times and temperatures typically reduces bacterial pathogens by 90% or more. The digested sludge applied as soil conditioner needs further treatments for the pathogen reduction

There have been studied the actual and potential dehydrogenase activity, phosphatase activity and the total catalytic activity of the activated and digested sludge. The enzymatic indicator of the sludge quality has been determined at the maximum value of 0.400 in the digested sludge samples, fact that indicate a moderate enzymatic potential of this type of sludge.

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THE TAXONOMIC AND ECOLOGICAL DATA REGARDING THE MACROLICHENS FROM PIATRA CRAIULUI NATIONAL PARK (I)

Corina-Neli MARCOCI*

Abstract. Lucrarea face referiri la taxonomia şi ecologia macrolichenilor din Parcul Naţional Piatra Craiului. Materialul lichenologic a fost colectat în lunile de vară din anii 2000-2002 din jumătatea nordică a zonei de studiu. Au fost identificate 62 specii care aparţin la 10 familii şi 4 ordine, din care 42 specii nu au mai fost identificate în zonă.
Key words. Piatra Craiului National Park, taxonomy, ecology

Introduction

This paperwork presents the results regarding the taxonomy and ecology of macrolichens from Piatra Craiului National Park.

The main part of Piatra Craiului National Park is the Massif Piatra Craiului, placed in Meridional Carpathians, having a lenght of 25 km, with the highest peak of 2238 m altitude (Baciului Peak).

Materials and metods

The lichenological material was collected in summer month of 2000-2002 from the following areas: Crăpătura, Diana, Plaiul Foii-Spirlea, Padina Popii, Piatra Craiului Mică Peak, Piatra Mare - Ascutit-Turn Peak, Zărnesti Gorges, Curmătura Zănoaga, trail to Vlădusca pasture (up to 1800 m alt.), La Zaplaz, Marele Grohotis. Foliouse and fruticulouse lichens collected from studied region are caracterized by corticolouse (dominant), tericolouse, lignicolouse and saxicolous.

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Results and discussions

In this region were identified 214 taxons. The author identified 62 species, belonging to 10 families and 4 orders, out of which 42 species have not been previously quoted in the researched zone (table 1).
Abbreviation: H.E.Pa - Parmelia type; H.E.Us - Usnea type; H.E.Pe - Peltigera type; Ch.Ce. - Cetraria type; Ch.Cl. - Cladonia type; H.E.So. - soredious type; H.E.Co. - Collema type; H.E.Ra. - Ramalina type; H.E.ex. - externalcrustos type; L - light; T - temperature; U - moisture; R - chemical reaction of the substratum.

Table 1. The macrolichens from Piatra Craiului National Park

Nr. ctr.	L	T	U	R	Species	Bioform	Substratum	Places
1.	4	5	4	3	*Chrysothrix candelaris* (J).R.Laundon	H.So	corticolous	La Om Peak, La Zaplaz, Plaiul Foii-Spirlea , Crapaturii Valley
2.	7	4	6	3	*Bryoria fuscescens* (Gyeln)Brodo & D.Hawksw	H.E.Us	corticolous (*Picea abies*)	Ascutit Peak , Padina Popii Peak
3.	8	0	0	8	*Toninia sedifolia* (Scop)Timdal		saxicolous	La Om Peak, Diana Brana Caprelor, La Zaplaz
4.	5	0	0	4	*Cladonia coniocraea* auct.	Ch.Cl.	terricolous, lignicolous	Magura village, Diana-Brana Caprelor, trail towards Diana, Plaiul Foii Spirlea, Curmatura Zanoaga, trail towards Vladusca pasture (1800 m), Ascutit Peak - Turn, Piatra Craiului Mica, Padina (at the chain)
5.	7	5	0	4	*Cladonia fimbriata* (L.)Fr.	Ch.Cl	terricolous, lignicolous	Magura village, Diana-Brana Caprelor, La Zaplaz, trail towards Diana, Plaiul Foii - Spirlea, Curmatura Zanoaga, trail towards Vladusca pasture (1800 m), Ascutit Peak Turn, Piatra Craiului - Mica, Padina at the chain
6.	7	0	0	0	*Cladonia pyxidata* (L.)Hoffm	Ch.Cl.	terricolous, lignicolous	Brana Caprelor, Marele Grohotis, Ref.Grind -La Om Peak, La Zaplaz, trail towards Diana, Plaiul Foii Sirlea, Curmatura Zanoaga, Curmatura-Piatra Mica
7.	7	0	0	0	*Cladonia pyxidata* ssp. *chlorphaea* (Sommerf.)V.W irth.	Ch.Cl	terricolous, lignicolous	Magura village, trail towards Diana, Plaiul Foii Spirlea, Cabana Curmatura, Zarnestiului Gorges
8.	8	0	0	0	*Cladonia arbuscula*	Ch.Cl.	terricolous	Plaiul Foii Spirlea, Piatra Craiului Mica Peak, Crapatura (1600 m)

Nr. ctr.	L	T	U	R	Species	Bioform	Substratum	Places
					(Wallr.)Flot			
9.	5	4	0	2	*Cladonia digitata* (L.)Hoffm	Ch.Cl.	lignicolous	Trail towards Vladusca pasture
10.	7	4	0	3	*Cladonia pleurota* (Florke.)Schaer.	Ch.Cl.	lignicolous	Crapaturii Valley (canyon)
11.	7	6	0	0	*Cladonia cervicornis* ssp. *verticilata* (Hoffm)Ahti	Ch.Cl	terricolous	Trail towards Crapatura
12.	6	4	6	2	*Cladonia cenotea* (Ach.)Schaer	Ch.Cl.	terricolous	Trail towards Diana, Curmatura Zanoaga
13.	6	5	0	4	*Cladonia furcata* (Huds.)Schrad	Ch.Cl	terricolous	Diana-Brana Caprelor, Crapatura Valley
14.	6	4	5	0	*Cladonia rangiferina* (L.)Weber ex F.H.Wigg	Ch.Cl	terricolous	Trail towards Crapatura, Piatra Craiului Mica Peak
15.					*Cladonia coccifera* (L.)Willd	Ch.Cl	terricolous	Plaiul Foii Spirlea
16.	8	0	5	2	*Pycnotelia papillaria* Dufour	Ch.Cl	terricolous	La Om Peak, Curmatura Zanoaga
17.	4	6	5	8	*Collema auriforme* (With.)Coppins & J.R.Laundon	H.E.Co	saxicolous	Trail towards Diana, Diana Valley, Plaiul Foii Spirlea, Crapatura Padina Popii
18.	7	0	0	8	*Collema tenax* (Sw.)Ach.em Degel	H.E.Co	saxicolous	Trail towards Diana, Diana Valley, Plaiul Foii Spirlea, Piatra Mare Creast –Ascutit Peak Turn, Piatra Mica Peak
19.	5	4	7	6	*Collema flaccidum* (With.)Coppins & J.R.laundon	H.E.Co	saxicolous	Diana Valley, Piatra Mare Creast - Ascutit Peak Turn, Curmatura Zanoaga
20.	9	0	0	8	*Lecanora muralis* (Schreb.) Rabenh	H.E.ex	saxicolous	Brana Caprelor, Marele Grohotis, Diana-Brana Caprelor, Zarnestiului Gorges, Piatra Mare Creast - Ascutit Turn Peak
21.	5	5	4	5	*Lecanora argentata* (Ach.) Malm	H.E.ex	corticolous (*Fagus sylvatica*)	Plaiul Foii Spirlea, Curmătura Zănoaga
22.	6	5	3	6	*Lecidella elaeochroma* (Ach.)M.Choisy	H.E.ex	corticolous (*Fagus sylvatica*)	Ref-Grind, La Om Peak, Plaiul Foii - Spirlea
23.	8	0	5	0	*Cetraria islandica* (L.)Ach	Ch.Ce	terricolous	Marele Grohotis, Ref.Grind-La Om Peak, Zarnestiului Gorges

Nr. ctr.	L	T	U	R	Species	Bioform	Substratum	Places
24.	9	1	8	2	*Flavocetraria nivalis* (L.)Karnef & Thell	Ch.Ce	terricolous	Ref Grind, La Om Peak
25.	6	3	7	2	*Vulpicida juniperinus* (L.)Mattson & M. J. Lai	Ch.Ce	corticolous (*Pinus mugo*)	Marele Grohotis, Ref. Grind - La Om Peak, Diana-Brana Caprelor, La Zaplaz
26.	9	2	8	2	*Flavocetraria cucullata* (Bellardi)Karnef & Thell	Ch.Ce	corticolous	Piatra Craiului Mică,
27.	8	2	8	3	*Melanelia hepatizon* (Ach.)Thell		terricolous	Diana-Brana Caprelor, Curmatura Zanoaga
28.	7	3	7	3	*Evernia divaricata* (L.)Ach.		corticolous (*Picea abies*)	Brana Caprelor, Plaiul Foii-Spirlea
29.	7	5	3	3	*Evernia prunastri* (L.)Ach.	H.E.Pa	corticolous (*Picea abies*)	Trail towards Crapatura, trail towards Diana, Plaiul Foii-Spirlea
30.	7	0	3	3	*Hypogymnia physodes* (L.)Nyl.	H.E.Pa	corticolous (*Picea abies*)	Trail towards Diana, Diana Valley, Plaiul Foii-Spirlea
31.	7	4	3	3	*Hypogymnia tubulosa* (Schaer.)Hav	H.E.Pa	corticolous (*Picea abies*)	Magura village, trail towards Crapatura, trail towards Diana, Diana Valley, Plaiul Foii-Spirlea, Padina Popii, Crapatura Valley
32.	6	3	7	3	*Hypogymnia vitatta* (Ach.) Parr.	H.E.Pa	corticolous (*Picea abies*)	Trail towards Vladusca pasture
33.	5	4	7	4	*Menegazzia terebrata* (Hoffm.)A.Massal	H.E.Pa	corticolous (*Picea abies*)	Trail towards Curmatura (up to Zarnesti Gorges)
34.	6	6	4	4	*Flavoparmelia caperata* (L.)Hale.	H.E.Pa	corticolous (*Fagus sylvatica*))	Plaiul Foii-Spirlea, Crapatura Valley
35.	7	0	3	5	*Parmelia sulcata* Taylor	H.E.Pa	corticolous (*Fagus sylvatica*))	Trail towards Crapatura, trail towards Diana, Plaiul Foii Spirlea
36.	6	4	5	2	*Parmeliopsis ambigua* (Wulfen)Nyl	H.E.Pa	corticolous (*Picea abies*)	Plaiul Foii-Spirlea, Diana (1600 m)
37.	6	3	7	2	*Parmeliopsis hyperopta* (Ach.)Arnold	H.E.Pa	corticolous (*Picea abies*)	Plaiul Foii, Spirlea
38.	7	5	3	5	*Melanelia exasperatula* (Nyl.)Essl	H.E.Pa	corticolous (*Picea abies*)	Plaiul Foii-Spirlea, Diana
39.	7	4	5	2	*Platismatia glauca* (L.)W.Culb.&C. F.Culb	H.E.Pa	corticolous (*Fagus sylvatica*)	Trail towards Crapatura, Diana Valley, Plaiul Foii – Spirlea

Nr. ctr.	L	T	U	R	Species	Bioform	Substratum	Places
40.	8	4	3	2	*Pseudevernia furfuracea* (L.)Zopf.	H.E.Pa	corticolous (*Picea abies*)	Magura Village, Brana Caprelor, Diana Brana-Caprelor, La Zaplaz, trail towards Crapatura, trail towards Diana, Diana Valley, Plaiul Foii Spirlea, Padina Popii, Curmatura Zanoaga, Curmatura, Piatra Craiului Mica Peak
41.	7	5	7	5	*Usnea florida* (L.)Weber ex F.H.Wigg	H.E.Us	corticolous (*Picea abies*)	Trail towards Crapatura, trail towards Diana, Diana Valley, Plaiul Foii - Spirlea, spre Padina Popii
42.					*Usnea articulata* (L.)Hoffm	H.E.Us	corticolous (*Picea abies*)	Plaiul Foii – Spirlea
43.	7	4	5	3	*Usnea hirta* (L.)Weber ex F.H.Wigg	H.E.Us	corticolous (*Picea abies*)	Trail towards Diana, Plaiul Foii – Spirlea, Padina Popii
44.	7	5	3	6	*Physcia stellaris* (L.)Nyl	H.E.Pa	corticolous (*Acer sp.*)	Plaiul Foii – Spirlea
45.	7	4	3	7	*Physcia aipolia* (Ehrh ex Humb)Furnr	H.E.Pa	corticolous (*Acer sp.*)	Plaiul Foii Spirlea
46.	7	5	6	6	*Ramalina fastigiata* (Ach.	H.E.Ra	corticolous (*Picea abies*)	Trail towards Crapatura, Plaiul Foii Spirlea
47.	7	5	5	6	*Ramalina fraxinea* (L.)Ach.	H.E.Ra	corticolous (*Picea abies*)	Trail towards Diana
48.	8	1	8	2	*Thamnolia vermicularis* (Sw.)Schaer.	Ch.Cl	terricolous	Ref. Grind – La Om Peak, Diana Brana Caprelor
49.	5	0	5	3	*Baeomyces rufus* (Huds.)Rebenh	H.Ba	terricolous	La Zaplaz
50.	5	4	7	5	*Lobaria pulmonaria* (L.)Hoffm	H.E.Pa	corticolous (*Acer sp.*)	Spirlea Valley, Marele Grohotis, Plaiul Foii Spirlea
51.	5	4	7	5	*Nephroma laevigatum* Ach.	H.E.Pe	terricolous	Spirlea Valley
52.	6	0	6	8	*Solorina saccata* (L.)Ach.	H.E.Pe	terricolous	Brana Caprelor, Marele Grohotis, La Zaplaz, Piatra Mare Creast – Vf. Ascutit Peak – Turn
53.	6	5	5	6	*Peltigera canina* (L.)Willd	H.E.Pe	terricolous	Magura village, Brana Caprelor, Diana, trail towards Crapatura, Plaiul Foii – Spirlea, Zarnesti Gorges, Curmatura Zanoaga
54.	8	0	3	8	*Peltigera rufescens* (Weiss.)Humb	H.E.Pa	terricolous	Brana Caprelor,
55.	5	4	6	5	*Peltigera horizontalis* (Huds.)Baumg	H.E.Pe	terricolous	Trail towards Crapatura, trail towards Diana, Plaiul Foii-Spirlea, Zarnesti Gorges
56.	6	5	5	5	*Peltigera polydactylon* (Neck)Hoffm	H.E.Pe	terricolous	Trail towards Crapatura, Plaiul Foii – Spirlea, Zarnesti Gorges, Curmatura Zanoaga, Piatra Craiului Mica Peak,

Nr. ctr.	L	T	U	R	Species	Bioform	Substratum	Places
								Ascutit Peak - Turn, Cabana Curmatura, trail towards Vladusca pasture
57.	8	4	0	3	*Peltigera malacea* (Ach.)Funk	H.E.Pe	terricolous	Trail towards Crapatura, trail towards Diana, Plaiul Foii – Spirlea, Piatra Mare Creast – Ascutit Peak -Turn, Curmatura Zanoaga
58.	5	5	5	5	*Peltigera praetextata* (Florke. ex Sommerf)Zopf.	H.E.Pe	terricolous	La Zaplaz, Plaiul Foii – Spirlea, Zarnesti Gorges, Piatra Mare Creast – Ascutit Peak– Turn
59.	7	5	3	7	*Xanthoria parietina* (L.) Th.Fr	H.E.Pa	corticolous	Plaiul Foii – Spirlea
60.	9	0	0	8	*Xanthoria elegans* (Link.)Th.Fr.	H.E.Pa	saxicolous	Ref. Grind, La Om Peak, Diana – Brana Caprelor, Plaiul Foii Spirlea, trail towards Padina Popii
61.	7	5	3	6	*Xanthoria polycarpa* (Hoffm.)Rieber.	H.E.Pa	corticolous	Plaiul Foii Spirlea
62.					*Xanthoria sorediata* (Vain.)Poelt	H.So	corticolous	Plaiul Foii – Spirlea, trail towards Padina Popii

Out of figure 1 it can be seen, that in the studied area prevail the epiphytic hemicriptophytic lichens like Parmelia type (54,83%) followed by, in equal procent (12,9%), Usnea and Ramalina type. In spurce fir forests (*Picea abies*) prevail *Hypogymnia physodes*, *H. tubulosa*, *Pseudevernia furfuracea*, *Evernia prunastri* and in beech forests or mixture, species like: *Parmelia sulcata*, *Flavoparmelia caperata*, *Chrysotrix candelaris* and *Xanthoria parietina*.

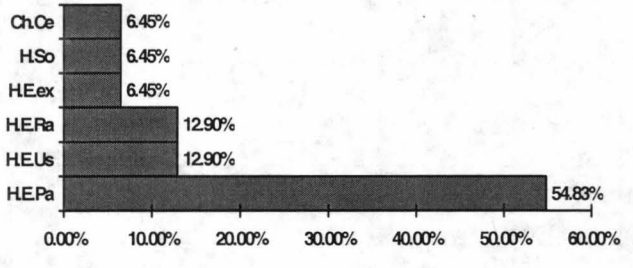

Fig. 1 Corticolous lichens bioforms distribution in Piatra Craiului National Park

Figure 2 refers at the repartition of corticolous lichens depending on the principal ecological elements. According to this, prevailed the following forms: temperate photophylous, mesotermophilous, xeromesophilous and acidophilous (pH = 4,1 – 4,0) species.

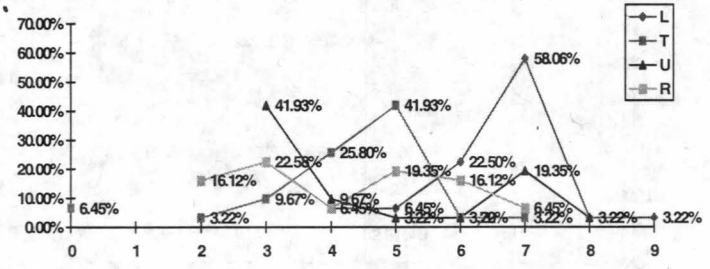

Fig.2 The repartition of corticolous lichens depending on the principal ecological elements

The repartition of terricolous lichens depend on their biological form, is presented in the figure 3 and it shows the prevail of chamephytic lichens Cladonia type folowed by Peltigera lichen type. In studied zone was recorded a great diversity of species belonging to Cladonia and Peltigera genera with very well developed specimens, due to the ecological life conditions.

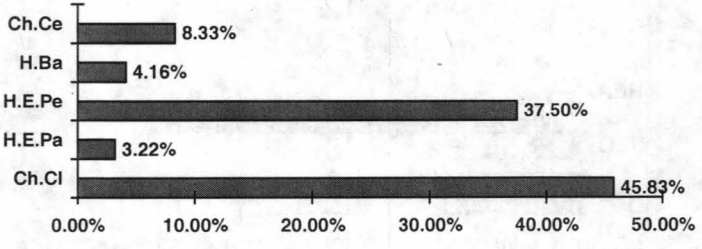

Fig. 3 Terricolous lichens bioforms distribution in Piatra Craiului National Park

In the figure 4 is presented the repartition of terricolous lichens, depending on main ecological elements. It emphazise following repartition: fotophylous lichens (33,33%), followed by fotoschiaphylous – temperate

fotophylous (25%) and fotoschiaphylous (20%). Depending on temperature factor prevailed equaly micromezoterme and eurithermic species (33,33%). According tohumidity factor prevail eurihygrous species (37,50%) followed by xeromezophylous – mezohygrophylous (33,33%).

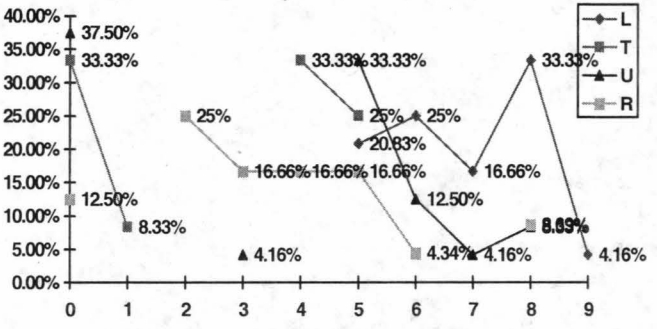

Fig.4 The repartition of terricolous lichens depending on the principal ecological elements

On the territory of researched area is a saxicolous lichens rich both from the number of species point of view and, also, from the number of specimens. Here prevailed epiphytic hemicriptophytic lichens like Peltigera type followed by species like Collema type (21,42%) but also chamephytic lichens like Cladonia type (14,28%) (fig. 5)

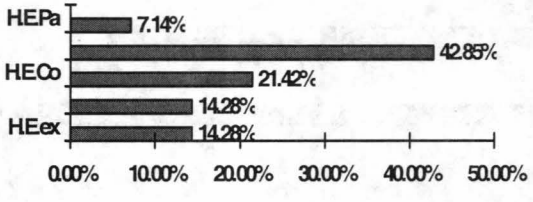

Fig. 5 Saxicolous lichens bioforms distribution in Piatra Craiului National Park

Regarding the repartition depending ecological factors (fig. 6) prevailed fotoschiaphylous species (28,50%), eurithermic (42,85%), eurihygrous (50%) and temperate acidophylous (21,42%) lichens.

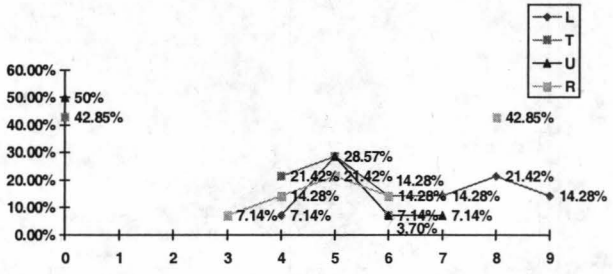

Fig.6 The repartition of saxicolous lichens depending on the principal ecological elements

Regarding the bioforms aspect (fig. 7) prevail chamephytic forms like Cladonia type (43,75%) followed by epiphytic hemicriptophytic lichens like Parmelia type (*Platismatia glauca, Physcia tenella, Hypogymnia tubulosa, Pseudevernia furfuracea*) (37,50%).

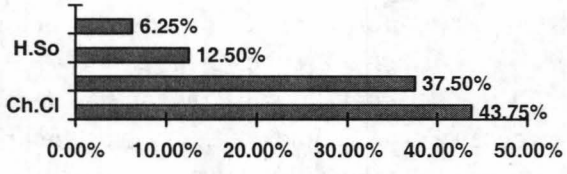

Fig. 7 Corticolous lichens bioforms distribution in Piatra Craiului National Park

In the lignicolous lichens category enter the species wich is developing on rotten trunk. Their growth and distribution is linked with the pH of the substratum and the climatic conditions (light, moisture and teperature). Prevail the temperate photophylous species (62,50%), micromezothermic and mezothermic (31,25%), eurihygrous (43,75%) and acidophylous (43,75%) (fig 8).

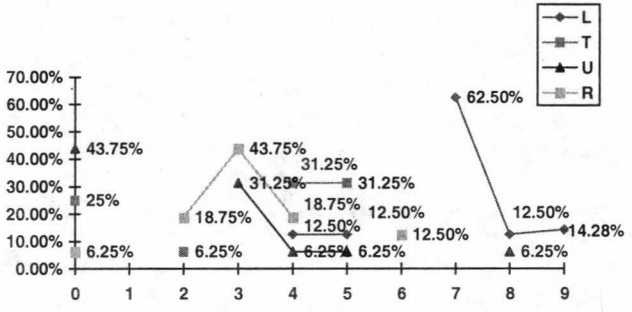

Fig.8 The repartition of lignicolous lichens depending on the principal ecological elements

Conclusions

This paperwork presents the results regarding the taxonomy and ecology of macrolichens from Piatra Craiului National Park. Foliouse and fruticulouse lichens colected from studied region are caracterized by corticolouse terricolouse, lignicolouse and saxicolous elements. In this region were identified 214 taxons. The autor identified 62 species, belonging to 10 families and 4 orders, out of which 42 species have not been previously quoted in the researched zone. Generally speaking prevailed macrolichens like Cladonia type followed by Parmelia and Peltigera type. Also, depending on the substratum, it emphazise following repartition: corticolous lichens, terricolous species, saxicolous lichens and lignicolous lichens.

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BIODIVERSITATEA MACROMICETELOR DIN BAZINUL SĂRĂRIEI-MUNŢII NEMIRA

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Abstract. The mycologycal material was collected in 1999 and 2003. In this region I identified 101 species (7 Ascomycetes and 94 Basidiomycetes), belonging to 25 families, 9 orders and 3 clases. The identified species are common for researched forests. The author identified 101 muschrooms taxa, out of which 99 species have not been previously quoted in the researched zone.
Cuvinte cheie: Bazinul Sărăriei, macromicete, bioforme, spectru ecologic

Introducere

Munţii Nemira fac parte din grupa centrală a Carpaţilor Orientali (Munţii Trotuşului), delimitând extremitatea ei sudică şi se încadrează în zona munţilor sedimentari sudici, întinzîndu-se pe o suprafaţă de 700 km² în judeţul Bacău, Covasna şi Harghita.

Zona studiată se află în centrul Munţilor Nemira . Limita de E-este constituită din pârâurile Doftenița şi Ciunget; la S-E este prezentă locaţia "La Cascada", iar limita de S e constituită de Culmea Căprioarei. Limita V este constituită de câteva masive Nemira Mică (1627m), Şaua Şandrului şi Şandru Mare (1640m). Cel mai important curs de apă aflat pe suprafaţa acestei zone îl constitue Dofteana, care izvorăşte de sub Muntele Şandru Mare, îşi uneşte apele cu pârâul Ţiganca, se strecoară prin cheile şi cascada care-i poartă numele.

Pădurile prezente pe aceste masive sunt făgete precum şi molideto-făgete şi brădeto-făgete . Flora mai cuprinde plante ca*: Filipendula vulgaris* Moench., *Leucanthemum vulgare* Lam., *Achillea milleifolium* L., alături de care se regăsesc unele specii rare şi endemice cum este *Saxifraga cymbalaria* L.

Consideraţii privind metoda de lucru şi conspectul macromicetelor

Conspectul macromicetelor din Bazinul Sărăriei a fost elaborat pe baza cercetărilor proprii, efectuate în cursul anilor 1999 şi 2003, precum şi a

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informațiilor bibliografice, luând în considerare studiile efectuate de Papp, C. (1957). Am identificat 101 specii care aparțin la 25 familii, 9 ordine, 3 clase și două subîncrengături. Din acestea, *Xylaria hypoxylon* și *Russula emetica* au mai fost citate de Papp, C. Autorul a mai identificat 11 specii de macromicete pe care nu le-am regăsit în zonă. Prezența fiecărei specii este însoțită de date privind: substratul pe care s-a întâlnit la colectare, data colectării, apartenența la diferite grupe de bioforme și categorii ecologice. Nomenclatura de clasare a speciilor în forme superioare de organizare s-a realizat după J. Webster , 1993.

Abrevieri: Categorii ecologice: S – saprofite,; Sl – saprofite lignicole; St – saprofite tericole; Sp – saprofite praticole; Sh – saprofite humicole; Sf – saprofte foliicole; Mr – simbionte; SPl – saproparazite lignicole. Bioforme: Gs – saprobionta; Gm – mycorrhiza; Epx – epixilobionta; Epbr – epibriobionta; Ex – endoxilobionta; Th – mycetotherobyonta.

CONSPECTUL MACROMICETELOR DIN BAZINUL SĂRĂRIEI- MUNȚII NEMIRA

Subîncrg. **ASCOMYCOTINA**

Cls. **Pyrenomycetes**

Ord. **Sphaeriales**

Fam. **Xylariaceae**

Hypoxylon rutilu Tul. – pe ramuri uscate de fag; (23.06.2003), Sl., Px.; *Xylaria hypoxylon* (L.: Fr) Grev. – pe cioate și trunchiuri în putrefacție; (25. 06. 2003) , Sl., Epx.;

*Xylaria polymorpha (*Pers.: Fr.) Grev. – pe lemn putred de foioase; (25. 06. 2003), Sl., Epx;

Xylaria longipes Nitschke – pe lemn putred de foioase; (25. 06. 2003), Sl., Epx;

Fam **Helvellaceae**

Helvella macropus (Pers.: Fr.) P. Karst. – în păduri de conifere; (26. 06. 2003), St., Gs.;

Leptopodia elastica (Bull.) Bond. – pe sol, prin păduri de amestec; (26. 08. 2003), St., Gs.;

Ord. **Helotiales**

Fam. **Helotiaceae**

Chlorosplenium aeruginascens (Nyl.) Karst. – pe lemn putred de foioase ;(26. 08. 2003) Sl., Epx.;

Subîncrg. **BASIDIOMYCOTINA**

Cls. **Hymenomycetes**

Ord. **Auriculariales**

Fam. **Auriculariceae**

Auricularia mesenterica (Discks. :S.F. Gray) Pers. – pe lemn de foioase; (26. 06. 2003), Sl.Epx., Co.;

Ord. **Aphyllophorales**

Fam. **Stereaceae**

Hymenochaete mougeotii (Fr.) Cke. – pe lemn de conifere; (25. 06 2003), Sl. Epx.;

Hymenochaete rubiginosa (Dicks. :Fr.) Lev. – pe lemn putred de gorun ;(25. 06. 2003), Sl., Epx.;

Stereum hirsutum (Willd. : Fr.) S. F. Gray. – pe trunchiuri putrede de mesteacăn, (26. 07. 1999), Sl, Epx ;

Fam. **Meruliaceae**

Merulius tremellosus Schrod. : Fr. – pe trunchiuri vii sau moarte; (24. 06. 2003) Sl., Epx.;

Fam.**Thelephoraceae**

Thelephora terrestris Ehrh.: Fr.- pe sol , prin păduri de foioase şi răşinoase ,(24. 06 2003), St., Gm.;

Thelephora caryophyllea (Sch. : Fr.) Fr. – pe sol, prin păduri de foioase şi conifere , (25. 06. 2003), St., Gm.;

Fam. **Cantharellaceae**

Cantharellus cibarius (Fr.: Fr.) Fr. – pe sol, prin păduri de amestec, (29.06.2003), Mr., Gm., Cooo;

Fam. **Clavariaceae**

Ramaria apiculata (Fr.) Donk. – pe lemn degradat, în păduri de conifere, (23.08.2003), St., Epx.;

Fam. **Hydnaceae**

Creolophus cirrhatus (Pers.: Fr.) P. Karst. – prin păduri de amestec, (23.06.2000),

Hericium coralloides (Fr.) Pers. – pe trunchiuri vii şi moarte de foioase, (24.06.2003), SPl, Ex-EPx, Co.;

Hydnum repandum Fr. – pe sol, prin păduri de foiase şi răşinoase, (23.06.2003), Mr., Gm., Co.;

Fam. **Polyporaceae**

Polyporus varius var. *elegans* (Fr.) Gill. et Lucand. – pe lemn de foioase, (24.07.2003), SPl., Ex.-EPx.;

Polyporus varius var. *nummularius* Bull.: Fr. – pe lemn de foioase, (24.07.2003), SPl., EPx.;

Daedalea quercina L.:Fr. – pe lemn de stejar, (26.06.2003), Sl., Ex.-EPx.;

Heterobasidion annosum (Fr.: Fr.) Bref. – pe trunchiuri în degradare, (26.03.2003), SPl., EPx.;

Trametes cinnabarina (Jack.) Fr. – pe trunchiuri vii şi moarte de foiose, (24.06.2003), SPl., Ex-EPx.;

Bjerkandera adusta (Willd.: Fr.) P. Karst. – pe trunchiuri vii şi moarte de carpen şi fag., (25.06.2003), SPl., Ex.-EPx.;

Laetiporus sulphureus (Bull.: Fr.) Murr. – pe trunchiuri vii şi moarte de salcie, (25.06.2003), SPl., Ex.-EPx.;

Ganoderma applanatum (Pers.) Pat. – la baza copacilor, (25.06.2003), SPl., Ex.-EPx.;

Fam. **Fistulinaceae**

Fistulina hepatica Schff.: Fr. – pe lemn de stejar, (26.06.2003), SPl., Ex.-EPx., Co.;

Fam. **Boletaceae**

Boletus crocipodius (Let.) Kuhn. – prin păduri de foioase, (26.06.2003), Mr., Gm.;

Boletus felleus Bull.: Fr. – pe sol, în păduri de amestec, (26.06.2003), Mr., Gm.;

Boletus luridus Schaeff.: Fr. – pe sol, făgete şi brădete, (26.06.2003), Mr., Gm., Co.;

Xerocomus chrysenteron Bull. (Quel) – pe sol, în păduri de foioase şi conifere, (26.07.2003), Mr., Gm., Co.;

Xerocomus subtomentosus L.: Fr. – pe sol, în păduri de foioase şi conifere, (24.06.2003), Mr., Gm., Co.;

Fam. **Pleurotaceae**

Lentinus tigrinus (Bull.: Fr.) Fr. – pe trunchiuri de salcie, (26.06.2003), Sl., EPx.;

Panus conchatus (Bull.: Fr.) Sing. – pe lemn de foioase, (27.06.2003), Sl., Ex.-EPx.;

Pleurotus ostreatus var. *pulmonarius* (Fr.) – pe trunchiuri de fag, (26.06.2003), Sl., Ex-EPx.;

Schizophyllum commune L.: Fr. – pe lemn viu şi mort de foioase şi răşinoase, (25.06.2003), SPl., Ex-EPx.;

Fam. **Tricholomataceae**

Armillariella mellea (Vahl. In. Fl. Dan.: Fr.) Karst. – în jurul arborilor, pe rădăcini şi în jurul cioatelor, (26.08.2003), SPl., Ex-EPx., Coo.;

Clitocybe gibba (Pers.: Fr.) Kumm. – pe sol, în păduri de foioase şi conifere, (26.07.2003), St., Gs., Co.;

Clitopilus prunulus (Scop.: Fr.) Kumm. – pe sol, prin păduri, (26.07.2003), St., Gs.;

Laccaria amethystina (Bolt.: Hook) Murr. – prin păduri de foioase şi răşinoase, (26.06.2003), St., Gs., Coo.;

Lepista inversa (Scop.) Pat. – prin păduri de conifere, (1.10.2003), St., Gs.;

Panellus mitis (Pers.: Fr.) Kuhn. – pe lemn de răşinoase, (27.06.2003), Ex-EPx.;

Pseudoclitocybe cyathyformis (Bull.:Fr.) – pe sol, pe muşchi, (13.10.2003), St., Gs-EPx.;

Resupinatus applicatus (Batsch.: Fr.) S.F.Gray – pe lemn degradat de foioase şi răşinoase, (26.06.2003), Sl., Epx.;

Oudemansiella radicata (Relh.: Fr.) Sing. – pe sol şi pe cioate putrede dar şi pe rădăcini, în păduri de foioase şi conifere, (26.06.2003), SPl-St., Ex-Gs., Co.;

Collybia fusipes (Bull.: Fr.) Quel. – la baza trunchiurilor vii şi pe cioate, în gorunete, (29.06.2003), SPl., Ex-EPx.;

Collybia longipes (Bull.) Quel. – pe sol, în brădete, (29.06.2003), Sl., Ex-EPx.;

Collybia velutipes (Curt.: Fr.) Kumm. – pe trunchiuri de arbori, (26.06.2003), Sl., Gs.;

Marasmius bulliardi Quel – pe frunze, în păduri, (24.06.2003), Sl., Gs.;

Marasmius cohaerens (Pers.: Fr.) Cooke et Quel. – pe frunze şi trunchiuri putrede în făgete şi brădete, (24.06.2003), Sf.-Sl., Gs.-EPx.;

Marasmius rotula (Scop.: Fr.) Fr. – pe ramuri şi rămurele în putrefacţie şi pe frunze în putrefacţie, în gorunete şi făgete, (24.06.2003), Sf.-Sl., Gs-EPx.;

Marasmius wynnei Bk. & Br. – prin păduri, pe frunze în putrefacţie, (24.06.2003), Sf., Gs.;

Marasmiellus longuidus (Lasch.) Sing. – pe strat ierbos în putrefacţie, (24.06.2003), Sf., Gs.;

Micromphale foetidum (Sow.: Fr.) Sing. – pe lemn putred, (24.06.2003), Sl., EPx.;

Megacollybia platyphylla (Pers.: Fr.) Kolt. Et Ponz. – pe lemn putred în păduri de amestec, (28.06.2003), Sl., EPx., Co.;

Mycena atrocyanea (Batsch.: Fr.) Gill. – pe sol, prin păduri de răşinoase, (24.06.2003), Sf., Gs.;

Mycena aurantiomarginata (Fr.: Fr.) Quel – în păduri de conifere, (24.06.2003), Sf., Gs.;

Mycena mucor (Batsch.: Fr.) Giel. – pe frunze în putrefacţie, prin păduri, (24.06.2003), Sf., Gs.;

Mycena pura var. *rosea* (Bull.) Gill. – pe frunze căzute, (28.06.2003), St., Gs.;

Mycena renati Quel – pe lemn în degradare, (28.06.2003), St., EPx.;

Mycena sanguinolenta (A. et S.: Fr.) Kumm. – pe frunze căzute în pădure, (28.06.2003), St., Gs.;

Omphalina ericetorum (Pers.: Fr.) Lge. – pe muşchi, prin turbării, (28.06.2003), St., Gs.;

Omphalina sphagnicola (Berk.) Mos. – pe muşchi de turbă, (28.06.2003), St., EPbr.;

Fam. **Rhodophyllaceae**

Entoloma helodes (Fr.: Fr.) Kumm. – în păduri, (28.06.2003),

Fam. **Agaricaceae**

Agaricus silvaticus Schff.: Fr. – pe sol, în păduri de foioase şi conifere, (26.06.2003), St., Gs., Cooo.;

Lepiota cristata (Alb. et Schm.: Fr.) Kumm. – pe sol, prin păduri, (26.07.1999), St., Gs.;

Macrolepiota rachodes (Vitt.) Sing. – pe sol, în molidiş şi brădete, (26.08.2003), St., Gs., Cooo.;

Fam. **Amanitaceae**

Amanita pantherina (D.C.: Fr.) Krombch. – pe sol, în păduri de foioase şi conifere, (26.08.2003), Mr., Gm., Tox.;

Amanita rubescens Pers.: Fr. – pe sol, prin păduri de foioase şi conifere, (26.08.2003), Mr., Gm., Coo.;

Amanita vaginata (Bull.: Fr.) Vitt. – pe sol, în păduri de foioase şi conifere, (26.08.2003), Mr., Gm., Tox.;

Fam. **Coprinaceae**

Coprinus micaceus (Bull.: Fr.) Fr. – pe sol şi pe trunchiuri putrede, (26.08.1999), St.-Sl., Th.-EPx.;

Coprinus plicatilis (Curt.: Fr.) Fr. – pe sol, în gorunete şi făgete, (26.08.1999), St., Gs.;

Panaeolus papilonaceus (Bull.: Fr.) Quel – în pajişti, în locuri gunoite, (26.08.1999), St., Th.;

Panaeolus sphinctrinus (Fr.) Quel – în pajişti, pe bălegar, (26.08.1999), St., Th., Tox.;

Fam. **Cortinariaceae**

Cortinarius venetus (Fr.) Fr. – pe sol, în brădete, (26.08.2003), Mr., Gm.;

Fam. **Strophariaceae**

Hypholoma capnoides (Fr.: Fr.) – pe trunchiuri căzute de brad, (26.08.1999), Sl., EPx.;

Hypholoma fasciculare (Huds.: Fr.) Kumm. – pe trunchiuri căzute şi cioate, în păduri de foioase şi conifere, (26.06.2003), Sl., EPx., Tox.;

Hypholoma sublateritium (Fr.) Quel. – pe trunchiuri tăiate de foioase, în păduri, (21.06.2003), Sl., Epx.;

Psilocybe coprophila (Bull.: Fr.) Quel. – pe bălegar, prin pajişti, (21.06.2003), St., Gs.;

Psilocybe montana (Pers.: Fr.) Kumm. – prin pajişti, pe muşchi, (21.06.2003), St., Gs.;

Fam. **Russulaceae**

Russula aeruginea Lindbl. – pe sol, prin păduri de molid, (26.08.1999), Mr., Gm.;

Russula delica Fr. – pe sol, în brădete şi făgete, (26.08.1999), Mr., Gm., Coo.;

Russula decolorans Fr. – pe sol, prin păduri de conifere, (27.08.1999), Mr., Gm.;

Russula emetica (Schff.: Fr.) Pers.- pe sol, în gorunete, brădete şi făgete, (27.08.1999), Mr., Gm., Tox.;

Russula farinipes Rom. – pe sol, prin păduri de foioase, (27.08.1999), Mr., Gm.;

Russula laricina Vel. – pe sol, în păduri de conifere, (27.08.1999), Mr., Gm.;

Russula mustelina Fr. – pe sol, în gorunete, (26.08.2003), Mr., Gm., Co.;

Russula sanguinea (Bull.) Fr. – pe sol, în păduri de foioase şi conifere, (27.08.1999), Mr., Gm.;

Russula vesca Fr. – pe sol, în păduri de foiose şi conifere, (27.08.1999), Mr., Gm., Cooo.;

Lactarius blennius (Fr.: Fr.) Fr. – pe sol, în făgete, (27.08.1999), Mr., Gm., Tox.;

Lactarius fluens Boud. – pe sol, în păduri de foioase, (27.08.1999), Mr., Gm.;

Lactarius rufus (Scop.:Fr.) Fr. – pe sol, în brădete, (27.08.1999), Mr., Gm., Co.;

Lactarius salmonicolor Heim.: Lacl. – pe sol, în păduri de molid, (27.08.2003), Mr., Gm.;

Lactarius scrobiculatus (Scop.: Fr.) Fr. – pe sol, în făgete şi brădete, (27.08.2003), Mr., Gm., Tox.;

Cls. **Gasteromycetes**

Ord. **Lycoperdales**

Fam. **Lycoperdaceae**

Bovista plumbea Pers.: Pers. – pe sol, în poieni, (27.08.2003), Sp., Gs., Co.;

Calvatia candida (Rostk.) Holl. – pe sol, prin pajişti, (27.08.2003), Sp., Gs.;

Lycoperdon perlatum Pers. – pe sol, în păduri de foioase şi conifere, (27.08.2003), Sh., Gs., Co.;

Lycoperdon pyriforme Schff.: Pers. – pe cioate şi trunchiuri putrede, în gorunete, făgete şi brădete, (27.08.2003), Sl., Epx., Co.;

Fam. **Astraeaceae**

Astraeus hygrometricus (Pers.) Morg. – pe soluri argiloase, (27.08.2003), St., Gm.;

Ord. **Nidulariales**

Fam. **Nidulariaceae**

Cyathus striatus (Huds.: Pers.) Wild. – pe lemnputred, frunze căzute, prin păduri de foioase şi răşinoase, (27.08.2003), Sh-St., Gs.-EPx.;

Ord. **Phallales**

Fam. **Phallaceae**

Phallus impudicus L.: Pers. – pe sol, în gorunete şi făgete, (27.08.2003), Sh., Gs.;

Mutinus caninus (Huds.: Pers.) Fr. – pe sol, în păduri de amestec, (27.08.2003), Sh., Gs.;

Rezultate şi discuţii

Analiza taxonomică scoate în evidenţă următoarele: subîncrengătura ASCOMYCOTINA totalizează un număr redus de specii (7 sp.) aparţinând la 3 familii, 2 ordine şi 1 clasă (Pyrenomycetes). Cea mai bine reprezentată din punct de vedere al numărului de specii, este subâncrengătura BASIDIOMYCOTINA cu 94 specii aparţinând la 21 familii, 7 ordine şi 2 clase (Hymenomycetes, Gasteromycetes)

Analizând spectrul ecologic (fig. 1), se constată următoarele: cel mai bine reprezentate sunt speciile **saprofite** (S) = 63,86% şi anume: **saprofite lignicole** (Sl) = 25,75% (fam. Helvellaceae, Xylariaceae, Stereaceae), **saprofite tericole** (St) = 24,72% (fam. Tricholomataceae), urmate de speciile **simbionte** (Mr) = 25,75% (fam. Boletaceae, Russulaceae); **saproparazite** (Sp) = 15,45% şi anume **saproparazite lignicole** (SPl) = 13,39% (fam. Polyporaceae).

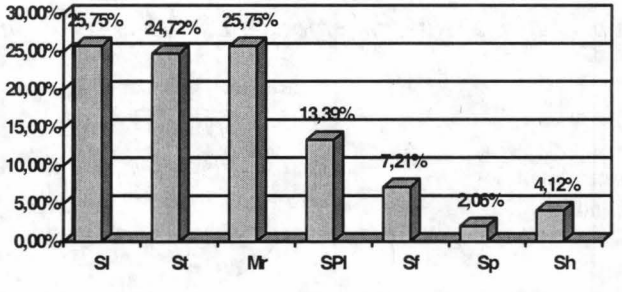

Fig. 1 Categorii ecologice la macromicete din Bazinul Sărariei

Din analiza bioformelor (fig. 2), rezultă că pondereea cea mai mare a speciilor de macromicete din Bazinul Sărăriei aparţin bioformei MYCETOGEOBIONTA (59,74%) şi anume: **saprobionta** (Gs) = 30,90% (fam. Helvellaceae, Agaricaceae, Tricholomataceae), **mycorrhiza** (Gm) = 28,84% (fam. Boletaceae, Russulaceae). Semnificative sunt şi speciile care aparţin formei biologice MYCETOEPIBIONTA (35,75%) : **epixilobionta** (EPx) = 24,72% (fam. Xylariaceae, Stereaceae, Meruliaceae); **epibryobionta** (EPbr) = 1,03% (fam. Tricholomataceae). De o pondere mai mică sunt speciile ce aparţin bioformei MYCETOENDOBIONTA (15,45%) şi anume **endoxilobionta** (Ex) = 15,45% (fam. Polyporaceae, Pleurotaceae). Puţin reprezentate sunt speciile ce aparţin bioformei MYCETOTHEROBIONTA (Th) = 2,06% (fam Coprinaceae).

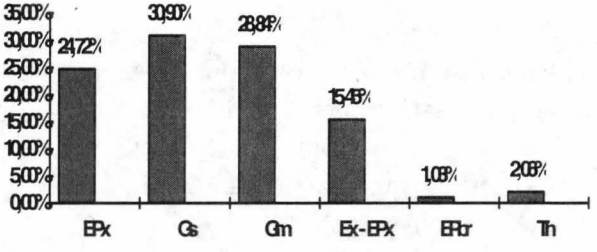

Fig.2 Spectrul bioformelor la macromicetele din Bazinul Sărariei

Un studiu important este şi cel al inventarierii speciilor de macromicete comestibile, toxice şi necomestibile. Din analiza pe teren în zona bazinului Sărăriei reies următoarele (fig. 3): 23,23% specii sunt **comestibile:** *Agaricus sylvaticus*, *Macrolepiota rachodes*, *Amanita rubescens*, *Russula delica*; 71,71% specii **necomestibile**; 7,07% specii **toxice:** *Amanita pantherina*, *Russula emetica*, *Lactarius scrobiculatus*.

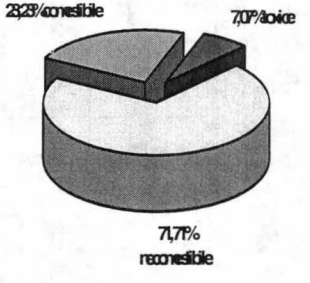

Fig. 3 Repartiţia macromicetelor din Bazinul Sărăriei în funcţie de valoarea lor nutritivă

Concluzii

Materialul micologic a fost colectat din bazinul Sărăriei, în lunile iunie – octombrie a anilor 1999 şi 2003. Am identificat 101 specii, încadrate sistematic în 2 subîncrengături, 3 clase, 9 ordine şi 25 familii. Sub aspectul bioformelor se evidenţiază speciile de tip MYCETOGEOBIONTA (59,74%). Analiza spectrului ecologic arată o pondere a speciilor **saprofite** (S) – 63,86%.

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THE UTILIZATION OF HISTOCHEMICAL AND HISTOENZIMOLOGICAL METHODS IN THE HEPATIC TOXICOSE INDUCED BY ENVIRONMENTAL XENOBIOTICS

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Abstract: Prezenţa în ambientul uman a xenobioticelor şi a radicalilor liberi impune găsirea unei metodologii adecvate pentru depistarea efectelor lor nocive, la nivelul unor organe ţintă; unul dintre acestea este ficatul. Metodele histochimice şi histoenzimologice permit localizarea activităţilor enzimatice "*in situ*", putându-se evidenţia astfel zonele tisulare afectate prioritar de xenobiotice şi radicali liberi. Astfel am determinat activităţile LDH, SDH, CyOx,, ATP-aza şi G6P-aza în cazul animalelor intoxicate cu CCl_4, nitrozamine şi alcool etilic.

Histochemistry, in general, and histoenzimology, in particular, (as an important component of histochemistry) are parts of the ensemble of the biological sciences. Histochemistry is a border discipline that combines the histo-morphological methodology with the chemical methodology (LISON, 1960; CHAYEN, BITENSKY, 1991; STOWARD and PEARSE, 1991).

In order to understand the biological functions, it is of capital importance to define the links between structure and function, that means the possibility to refer to a given structure with its chemical charge, or to a stage of metabolism that takes place at its level (MIHAIL and RUSU, 1973). The presence of some chemical substances, organic or inorganic, in certain biological structures, as well as their dynamics, are important, too, in the study of the correlation between structure and function. No matter the method used or the issue of the examination of a biological object, it is always interesting to know its chemism, and its structure. Thus, the chemical processes in an organism are not anymore seen as if they were taking place in a lab-tube, but we can make the connection between function and its structural substrate. All the same, any histological structure must be

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connected with its functional and dynamical aspects. Therefore, from an incomplete descriptive stage we move to causal-explicative expositions.

The histochemical research must satisfy two kinds of exigencies. Some of them are morphological and require the exact conservation of the localization of the investigated substance, therefore requiring the respect for the cellular and tissue form (architecture). The other exigencies are chemical, requiring the specific and adequate evidence of the investigated substance.

Histochemistry, with very few exceptions, does not identify chemical entities, but only chemical functions or free radicals (LISON, 1960).
In many cases, this is easily accomplished with the help of histochemistry. Thus, we can easily make visible the catecholamines in the adrenal medulla, the glycogen in liver only at the hepatocytic level or the alkaline phosphatase in the intestinal villi epithelium, but not in the intestinal crypts. As the biochemistry is concerned, this localization would be very difficult to accomplish. Histochemistry is useful and superior where a localization of certain substances or metabolic activities is required. The biochemist does not use tissue sections, but homogenates that, analyzed, give average values of the different components, and it is very hard to observe the link between a structure and an enzyme, for example.

Therefore, microchemistry and/or biochemistry have a preponderant analytical character, and histochemistry has, above all, a synthetic character. While chemical and biochemical analyses undone or destroy the morphological integrity of cells and tissues and obtain an average value of the investigated chemical compounds, histochemistry does not destroy the biological structures, but it saves them more or less, together with the chemical constituents that they consist.

Histochemistry has as target the detection, analyze and localization of those chemical constituents or enzymatic activities, at the level of certain well-defined structures of cells and tissues, therefore it is in fact a "Topochemistry".

Histochemistry was metaphorically compared by LISON (1960) with a palace, in which rooms you wander and you can see of what is it composed. But microchemistry (biochemistry) must demolish it in order to find its composition. The beginning of histochemistry can be considered the determination of glycogen with iodine, made by the great physiologist

CLAUDE BERNARD (1854), and it continues its progress ever since. The entire XX-th century was witness to its development and affirmation.

Imunohistochemistry represents the modernization and adaptation to the new scientific requests, but the "classic" histochemistry has not finished her resources yet.

Histochemistry addresses very successfully especially to those chemical substances that have a macromolecular structure and are more or less bound to the cellular and subcellular structures, and less to those substances that have a small, soluble, more mobile molecule.

Histochemistry must fulfill two fundamental necessities, which are: *the morphological necessities* – that means the storage in good conditions of subcellular, cellular, and tissue structures, and *the chemical necessities* – that means the correct identification of the investigated compound or of the metabolic activity, where it is normally localized.

The foundation of the histochemical reactions is the formation of a colored compound "chromophore" or "chromogen" that can be observed with the optic microscope and can be even quantified, though the "classic" histochemistry was more qualitative. On this regard, there is a conventional system, internationally recognized, for the estimation of the differences found against a control. We present downwards the BARKA (1961) type international units system that expresses a qualitative evaluation upon the color intensity and upon the frequency of the colored granules:

0 = lack of histochemical activity
1 = weak reaction
2 = moderated reaction
3 = medium reaction
4 = intense reaction
5 = very intense reaction

The precision degree of histochemistry is similar to that of analytic microchemistry, but it depends upon the resolution of the optic microscope. A granule under 0.3 μ doesn't show and the reaction may seem negative. The problem of "artifacts" or of the histochemical false positive or negative reactions is rather complex, but it can be fairly resolved.

Histochemistry can help in the determination of many compounds, chemical constituents, or enzymatic activities, such as:

carbohydrates
proteins

lipids
nucleic acids (DNA and RNA)
vitamins
hormones
mineral elements
enzymes (mostly enzymatic activities), etc.

Histochemistry can be successfully utilized for reasons of fundamental or applied research in medicine and biology, in plants, animals, and human, accordingly.

Histoenzimology

Investigation of enzymes represents a very dynamic field of the modern histochemistry, considering its importance in the cell's economy, and it involved a rapid development (VAN NOORDEN and FREDERIKS, 1992; VAN NOORDEN and JONES, 1995). The studies show that enzymes can be directly detected only in very few cases. Deoxyribonuclease and ribonuclease could be very easily identified through imunohistochemical methods.

In most of the situations, only the enzymatic activity can be visualized, thus we indirectly make visible the enzyme. An enzyme is a chemical well-defined entity, but different enzymes, as the chemical individuality is concerned, can be active for the same substrate. For example, glucoso-6-phosphate is hydrolyzed by glucoso-6-phosphatase and by acidic phosphatase. Therefore, in this case, the activity of glucoso-6-phosphatase is not the same as the enzyme glucoso-6-phosphatase. All the same, the same enzyme, which is located in the same morphological site, can have different enzymatic activities. For example, acidic phosphatase hydrolyses gucoso-6-phosphate, glucoso-1-phosphate, and β-glycerophosphate. Thus, a subcellular site that presents three different enzymatic activities can contain only one enzyme.

Therefore, we have to consider two moments:

proving the enzymatic activity
identifying the enzyme responsible for this activity.

Providing the enzymatic activity requires two steps:

tissue preparation
the visualization of enzymatic activity

Tissue preparation

In this stage it is advisable to preserve the enzyme, not only its activity, but also its exact location. The enzyme immobilization represents a complex issue. We must consider the fact that enzymes are present in two forms in cells: lioenzymes, which are soluble in water or in hydro-alcoholic mixture, and desmoenzymes, which are attached to the insoluble elements of the cytoplasm. The definitive immobilization of a lioenzyme is rather problematical. It seems that only desmoenzymes are satisfactory suitable for the histoenzymatic methods. The immobilization of these enzymes can be done with chemical fixatives or with physical agents (cryofixation).

Conservation with chemical fixatives

Conservation, preservation of enzymatic activity: some enzymes resist the histological fixation and the embedding in paraffin (peroxidases, for ex.), and others can be visualized only on live, unfixed, frozen, cryotome sliced tissue (for ex. Cytochrome-oxidase, succinate-dehydrogenase, lactate-dehydrogenase, adenosine-triphosphatase, etc.). Most of the enzymes do not suit with the usual histological fixation processes. But some enzymes do resist the common fixatives. In this case, the most used histological fixatives are: acetone, absolute alcohol, saline or calcic 4% cold formalin (0-4°C), buffered osmium tetroxide (pH=7), cold when used, etc (HOPWOOD, 1985). Of course, after fixation follows the embedding in paraffin.

The slicing operations

The cryotome – is the apparatus with which fresh, cryo-fixated sections of tissue or fragments of organs are sliced. Surely, the slicing is performed at low temperature, as we will see. The temperature in the working-chamber can be between -1°C and -70°C. The fragments of organs, frozen on cork-pieces, are pulled from the plastic boxes, in which they were stored in the container with liquid nitrogen, and are fixated on the wet specimen-holder. They are introduced in the cryotome, where they freeze and tie the fragment of organ on the specimen-holder. The fragments of organs are sliced in the cryotome by a special knife or by razors (preferable) into sections of 5-7μ thick in general. The sections that are too thin, or too thick,

are not suitable. The slicing is done manually or even automatically, by the modern cryotomes. A plastic plate in front of the knife, named the antiroling plate, determines the sections to remain on the knife's blade, or on the blade from where they are cropped on microscope slides or on coverslips. The adherence of the slices onto the slides or the coverslips is possible because of the existing differences of temperatures.

The incubation of the slices

The slices on the slides or on the coverslips are immersed into an incubation medium, after a short drying (5 minutes). This medium of incubation contains, generally, the enzyme's specific substrate, a buffer solution with a pH that allows the catalysis to be carried on at maximum values, activators, etc. The incubation medium is made up to determine the staining of the product formed over the action of enzyme (chromophore or chromogen) or to create such a colored product, which has to be insoluble, in order for us to mark with great precision the location of the enzymatic activity. The dishes used to store the incubation media are called Laveran-type dishes and they have partitions for slides (5-10-12). The slides with the sections do not reach to one another, so we can avoid the damaging of the sections. The laverans have adequate lids that hinder the evaporation of the incubation medium. Generally, the chemicals that are consisted by the incubation medium are expansive and imported from abroad, from well-known companies.

Time of incubation

It is very different and depends on the enzymatic activity that is going to be examined, and also on the organ (tissue) in which it is located. Thus, there are organs with an important enzymatic content, such as the liver or the kidney, and others in which the enzymatic activity is harder to be visualized (lung, spleen, brain, etc.). The incubation time can differ from 10-15 minutes for the visualization of lactate-dehydrogenase from liver, to 1-2 hours for the visualization of adenosine-triphosphatase and cytochrome-oxidase from brain. For liver, the same enzymes cannot be incubated more than 30-60 minutes. The optimum incubation time is the period in which the investigated enzymatic activity is the utmost. A shorter incubation time

cannot make visible the enzymatic activity, giving the impression of a false negative reaction. A too long incubation time can cause the diffusion of the enzyme from the native site, that causes the apparition of false positive images (artifacts) in zones different from the ones in which the enzyme is normally located.
We can say, with maximum of certainty that the average incubation time can be considered from 30 to 60 minutes.

Temperature of incubation

Usually, sections from organs of homothermal animals are incubated at 37°C. But there are enzymes that can be visualized even at temperatures of 18-20°C (the so called room temperature). In poikilothermal animals and in plants, the enzymatic activities can be visualized under 37°C, too (usually between 20-30°C).

The enzyme identification

Some methods can lead to false positive reactions because of the unspecific absorption of the products of spontaneous degradation of the substrate. However, these unspecific reactions will seldom give serious errors, because all methods have control trials in which the enzymatic activity has been suppressed using an adequate method. This allows a separation between the false and the real enzymatic reactions. The enzyme identification is based, generally, on several conditions, such as:

activity on different substrates, adequately chosen;
utilization of some specific inhibitors or activators;
comparison between the histochemical and the biochemical results.

Enzymes that can be visualized using histochemical methods

Hydrolytic enzymes
Phosphatases
Carboxylic esterases
Aminopeptidases
Glucuronidases
Sulphatases

Carboanhydrases
Phosphorilases
Deoxyribonucleases

Enzymes that catalyze reactions of oxidation-reduction
Oxidases
Aerobe dehydrogenases
Anaerobe dehydrogenases
Peroxidases

Possibilities and limits of histochemistry

First of all, some fields are not easy to access for the histochemical *in situ* analysis. We are talking about the histochemical investigation of some substances existing in the tissue medium, that are dissolved and are not connected in one way or another to the cellular constituents. These substances can be visualized with appropriate reagents, but there isn't any certainty about their exact histological localization. Therefore, the morphological condition cannot be fulfilled for an exact histochemical reaction. The errors are directly linked to the degree of diffusibility of the investigated substance. The diffusibility is maxim for crystalloid substances with low molecular weight, such as: chlorides, sulfates, soluble phosphates, glucose, urea, etc. But, if the substances are in colloidal solution, they can be satisfactory fixated, as for glycogen, for example.

The real field of *in situ* histochemistry is that of the macromolecular substances from cells. Most of the histochemical research is done on tissue sections, and the existing elements are represented by the macromolecular cellular and tissue skeleton, without non-macromolecular soluble elements. *In vitro* microchemical studies are conducted on non-macromolecular cellular elements, and the directing principle here is that of analytical extraction. On the opposite, the histochemical methods are synthetic, reaching for the preservation of the macromolecular skeleton of the cells.

The sensibility of histochemical reactions is a very important matter. Histochemical methods can be as sensible as or more sensible than those used by microchemistry. For example, microchemistry is able to visualize up to 0.002μ. In histochemistry, one granule of Prussia blue of 1μ in diameter, easy to observe with an optic microscope, corresponds to an amount of Fe of

aprox. $2.5 \times 10^{-7}\mu$. Thus, the histochemical reaction for Fe proved to be more sensible than the same reaction performed with microchemical methods. The sensibility limits of histochemical reactions are rather low, but their visualization depends on the power of resolution of the optic microscope.
Histochemistry is (was) a science generally qualitative, because, when examining at the optic microscope, we can compare the color of a control tissue section with one of another section that was subjected to certain influences. But histochemistry is able to become semi-quantitative, or even quantitative, too. There are two methods used on this matter:

histophotometry (cytophotometry) – that allows the quantitative expression, on objective criteria, of the color intensity of a tissue section, appeared following a histochemical reaction. Adequately, there are: photometry in visible, in UV, etc.

the elution method – the dye on the section is washed with an appropriate solvent, and the resulted solution is assessed with a spectrophotometer. The amount of dye deposited on the histological structures is proportional to the investigated substance or enzymatic activity.
As the research accuracy is concerned, we can also use the microchemical (biochemical) dosage for the comparison with the histochemical results.

Histochemistry can be considered, as we showed in the beginning, not only a complex of methods, but a border science that uses efficiently, adequately and trustworthy a methodology that can be of great help for the researchers in the experimental morphology field, where the biochemical, microchemical and physiological influences are needed and welcomed (MIHAIL and RUSU, 1973).

The presence of xenobiotics and free radicals in human environment requires the finding of an adequate methodology for tracking down (discovering) their noxious effects, at the level of some "target" organs. One of these organs is the liver, because most of the xenobiotics are metabolized by the liver, and one of the appropriate methodologies consists of the utilization of histochemical and histoenzymological techniques.

In our experiments we used toxic substances that are present in human environment, such as: ethanol, nitrosamines (dimethylnitrosamine), and CCl_4. All these xenobiotics have an obvious hepatic tropism. It was discovered a mitochondrial hyperproduction of free radicals after an alcoholic overdosage, which produced lipid peroxidation of unsaturated fatty acids, especially in mitochondria.

CCl_4 is bioconverted into the CCl_3^- free radical, under the action of P4502E1 Cytochrome. CCl_3^- bounds covalently to the cellular macromolecules (FAROON and col., 1994), leading to massive enzymatic unbalances. The nitrosamines are metabolized in microsomes, under the MFO action, resulting into an alkyl radical that methylates DNA. At cytostructural level, the ethanol attacks the cellular membranes first, and then the mitochondria, where it produces megamitochondria. CCl_4 attacks the cellular membranes, the endoplasmic reticulum, and the mitochondria. The nitrosamines damage the cellular nucleus firstly.

The histoenzymological methods that consist of the assay of lactat-dehydrogenase (LDH), succinat-dehydrogenase (SDH), cytochrome-oxidase (CyOx) activities (oxido-reductive enzymes), and the assay of ATP-ase and glucoso-6-phosphatase (G-6-P) activities (hydrolytic enzymes) allowed the evidence of some "marker" enzymes, that are typical for the action of certain toxic substances. In the case of CCl_4 – induced toxicose, these enzymes are lactat-dehydrogenase and glucoso-6-phosphatase.

The histoenzymological methods allow the "in situ" localization of enzymatic activities, permitting the evidence of the tissue zones firstly affected by xenobiotics and free radicals.

Conclusions

The utilization of some histochemical and histoenzymatical methods proved to be necessary in the assay of hepatic toxic action produced by some xenobiotics present in the human environment. Thus, we evidenced enzymatic activities that are tipical for certain toxic substances.

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CORRELATIONS BETWEEN THE ACTUAL HILLY AND PLAIN VEGETATION FROM TRANSYLVANIA AND RECENT – SUB - RECENT PALYNOLOGICAL SPECTRA

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Rezumat. Corelaţii între vegetaţia actuală din zone colinare şi de câmpie din Transilvania şi spectrele palinologice recente şi subrecente. Vegetaţia actuală este influenţată de relieful variat şi condiţiile climatice, care variază în funcţie de altitudine, determinând etajarea vegetaţiei. Structura actuală a pădurilor este supusă însă şi presiunii antropice puternice, care produce modificări însemnate. Această influenţă se poate urmări foarte bine în spectrele polinice recente şi subrecente, care evidenţiază dinamica evoluţiei vegetaţiei pe măsură ce coborâm scara temporală.

În lucrarea de faţă ne-am propus să analizăm vegetaţia din zonele colinare şi de câmpie din Transilvania, stabilind anumite corelaţii între vegetaţia actuală şi cea de acum câteva zeci sau sute de ani, cu referire în special la formaţiunile forestiere, de querceto-cărpinete. Se poate constata o tendinţă generală de reducere a suprafeţei ocupate de aceste asociaţii, atât raportat la vegetaţia lemnoasă, cât şi la cea totală. Actualmente, querceto-cărpinetele reprezintă astăzi sub 40% din suprafaţa împădurită a Transilvaniei, iar procentul mediu de împădurire a zonelor joase şi colinare din Transilvania este de 10,88%.

Prin reducerea suprafeţei pădurilor sau modificarea spectrului esenţelor lemnoase sunt periclitate balanţa hidrică şi sursa de oxigen pe care o reprezintă aceste păduri, compoziţia şi stabilitatea terenurilor împădurite. Reîmpădurirea terenurilor degradate (cca. 250.000 ha) din zona colinară a Transilvaniei se impune ca o necesitate de maximă urgenţă, pentru a preîntâmpina declanşarea unor dezechilibre ecologice şi a opri deşertizarea. În efectuarea activităţilor de reîmpădurire în zonele colinare din Transilvania este esenţială, în lumina datelor fitoistorice existente, respectarea structurii iniţiale a pădurilor.

Cuvinte cheie: *vegetaţie, querceto-cărpinete, palinologie, Holocen, Transilvania*

Data regarding the actual hilly and plain vegetation from Transylvania

The actual vegetation of Transylvania is directly linked to the

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extremely variable relief and climatic conditions that considerably vary along the altitudinal gradient. This leads to a vertical arrangement of the vegetation. Our analysis refers to the vegetation from the low relief units i.e. plains and hills. The plain of Transylvania, the Târnavelor low plateau and the Secaşelor plateau are covered by sylvosteppe. The zone delimited by these relief units is surrounded by higher lands and passes radially to the regions of the forest zone, having a vegetation characteristic to the common oak and durmast oak vegetation belt.

Through the openings offered by Mureşului passage and the hills of Somes region some southern termophilous species entered Transylvanian Basin (exp. *Quercus frainetto, Q. cerris, Fraxinus ornus, Chrysopogon gryllus, Danthonia calycina,* etc.). These species represent moesian and iliric influences, noticeable from the Mureşului passage to the Secaşelor plateau and from the north-western part of the Someşean platform to Almaşului and Borşa regions (Csürös, 1963).

On the shadow and part-shadow hills from the Transylvanian plateau and the Transylvanian sub-Carpathians and, to a lesser degree, on the western piedmonts of the Apuseni Mountains, forests of durmast oak and hornbeam of the association *Lathyro hallersteinii-Carpinetum* Coldea 75 can be found. The characteristic and differential species for the association are: *Lathyrus hallersteinii, Helleborus purpurascens* and *Festuca drymeia.* From the derived phytocoenoses that settle in these sites after the clearance of the *Quercus petraea - Carpinus* forests we can mention those belonging to the associations *Festuco rubrae-Agrostietum capillaris* Horv. (51) 52, *Anthoxantho-Agrostietum capillaris* Sillinger 33, and *Agrosteto-Festucetum valesiacae* Ardelean 83 (on the sunny slopes).

The oak and hornbeam forests, ranged in the association *Melampyro bihariensi-Carpinetum* (Borza 41) Soó 64 em. Coldea 75, are spreading, at the present, on big surfaces inside the Hârtibaciului and Târnavelor plateaus, where they populate the light slopes of the hills, terraces and depressions. The characteristic species for the association is *Melampyrum bihariense.* Besides this, there can be found other geographical differentials like: *Lathyrus transsilvanicus, Hepatica transsilvanica, Aconitum moldavicum* and *Euphorbia carniolica.* The derived lawns, secondary installed after the clearance of the forest, are those belonging to *Festucetum pratensis* Soó 38 and *Alopecuretum pratensis* Now. 28 associations.

The common oak forests with durmast oak and tartaric maple tree (*Aceri tatarico-Quercetum petrae-roboris* (Soó 51) em. Zólyomi 57 association) considered as specific for the Transylvanian plain, are presently found in the area only fragmentary and on small surfaces. These types of phytocoenoses have been described in the surrounding of Cluj, Turda and Aiud and also Blaj and Alba Iulia, as adjacent zones. The recognition and edification species for this sintaxonomic unit is *Quercus petraea.* Hornbeam and beech are sub-dominant or co-dominant. After the clearance of the forests, this association was replaced by meso-xerophytic and xerophytic phytocoenoses ranged in *Carici humilis-Brahipodietum pinnati* Soó 42 and *Danthonio-Stipetum tirsae* Ghişa 41 associations.

The sunny slopes of the Someşan plateau are populated with forests of durmast oak and cerris belonging to the *Quercetum petraeae-cerris* Soó 57 association. The recognition and edification species for the association are: *Quercus cerris* and *Quercus petraea.* Besides these, the differential species *Tilia tomentosa* may sporadically appear in the association. The derived phytocoenotic units that secondary set on these lands, after the clearance of the *Quercus cerris - Q. petraea* forests, belong to the *Agrosteto-Festucetum valesiacae* Ardelean 83 and *Botriochloetum ischaemi* (Krist. 37) Pop 77 meso-xerophitic associations.

The hygrophilous phycoenoses of the *Scirpo-Phragmitetum* Koch 26 association, largely spread throughout Transylvania, are dependent to the pools around the springs and streams. The recognition and edification species for the association are: *Phragmites australis, Typha angustifolia, T. latifolia* and *Scirpus lacustris.* In a similar ecological environment vegetate the associations *Typhetum angustifoliae* Pign. 53 and *Phalaridetum arundinaceae* (Koch 26) Libb. 31, but these are spread on smaller areas. From the group of tall bulrush, the coenoses of the association *Caricetum rostratate* Rübel 12 are frequent in the region. The phytocoenoses of the associations *Caricetum acutiformis-ripariae* Soó (27) 30 şi *Caricetum gracilis* Tx. 37 have a smaller spreading (Csürös and colab. 1963).

The ligneous phytocoenoses edified by *Alnus glutinosa* from the *Stellario nemori-Alnetum glutinosae* (Kästner 38) Lohm. 57 association are frequently spread on the meadows of the valleys from the hilly and sub-montaneous belts, in different regions of Transylvania. The recognition species for the association are *Stellaria nemorum* and *Alnus glutinosa.*

The pollen spectra and the relative chronology of the vegetation's dynamics during the Holocene

The actual palynological data, from the specialty literature, regarding the plain and hillocky regions from Transylvania are not so numerous. The explanation resides in the conservation possibilities of fossil pollen, which are more reduced. As it is known, the pollen is preferentially preserved in oligotrophic peat-bogs, which develop at higher altitudes, usually in the mountain belt. In the hillock belt and on plains the developed bogs were the eu- and mesotrophic ones, which offer poorer conservation conditions for the fossil pollen.

Another potential source of quaternary fossil pollen, at medium and low altitudes, is represented by lacustrine sediments. In some of these sediments, the advanced degree of corrosion of the pollen's exine, caused by the oxidative processes that take place over time, is a major disadvantage. Many times although, these plain lakes offer to the palynologist better conditions than that of glacial lakes, which have, in majority, a rocky substrate.

Reviewing the sites palynologically analysed, from the plain and hillocky regions of Transylvania, from their relative small number we had to select only those that were possessing recent and sub-recent palynological spectra in order to fulfill our proposed objective i.e. to establish correlations between these palynological data and the actual vegetation from the targeted regions.

Because of multiple factors (surface recent peat, too lax, non-decompose; burned surfaces; peat-formations, floating reed islet type; high risk of sample contamination, etc.) many of the studied palynological sequences start at considerable depths, chronologically speaking. Ten or more centimeters may represent, on a temporal scale, hundreds of years. We know that the last postglacial climatic period, the Subatlantic (based on the climatic schemes of Blytt and Sernander, 1876, 1890) started approximately 3.000 years ago (Orombelli and Ravazzi, 1996). Based on some sequences, ^{14}C dated, the step, or the sedimentation rate of the peat was calculated taking into consideration the age of the levels and their depth.

There are, of course, correction factors that have to be taken into consideration in doing the calculations, when the ^{14}C data are lacking (and this is occurring quite often, unfortunately). It is known that the sedimentation

rate of the Subatlantic peat, also know as "young peat", is higher than that of "old peat", especially at the beginning of the Postglacial (Holocene), when the climatic conditions were different to those of the Subatlantic and nowadays.

Preboreal climate, which started approximately 10.000 years ago, was a passing one between the harsh conditions of the final Tardiglacial(recent Dryas) (cold and dryness) and the optimum ones (warmth and humidity) of the Boreal and the Atlantic. As a consequence, a progressive accumulation of the peat in time, with some oscillations, can be admitted. For example, the sedimentation step of the peat in the Atlantic, a moist and warm period of the Holocen, was bigger than that of the warm but drier Subboreal that followed.

Stagnations in the peat genesis process, or the so called "sedimentary gaps", can also be noted, even during the same climatic period, due to some local conditions or short termed regional climatic oscillations, but these stagnations can be detected on the pollen diagrams, following the dynamics of the main pollen taxa.

Because most of the palynologically analysed sequences does not have, due to objective reasons, an absolute chronology (^{14}C dating – calibrated or non-calibrated), the palynological analyse in itself delivers a relative chronology, based on the succession of the pollen spectra, whose graphical representation are pollen diagrams. Thus, the dynamics of the pollen taxa allows the rangement of the pollen spectra in the classical climatic schemes, wellknown from the palynological literature, from which the most used is that elaborated by Blytt and Sernander (1876, 1890) but we will not discuss it here as it is very well known from the palynological papers.

Based on this relative chronology and knowing the depths corresponding to the pollen spectra, from the analysed sequences, the sedimentation rate and the approximate age of one level from the studied sequence can be established. Applying this method we can approximately know the structure of the vegetation around the studied site, at one moment in its evolution.

Correlations between the actual vegetation and the analysed pollen spectra

The palynological analysis that we are dealing in were made on sequences located in Alba (Colțești), Bistrița-Năsăud (Zagra), Cluj (Pădureni-

Ţop, Sălicea, Valea Morii), Mureş (Deda, Dracăşviz), Sălaj (Hereclean) and Sibiu (Arpaşul de Sus, Avrig) counties (Fig. 1).

There is no perfect chronological correlation between these sites concerning the sedimentation rate. On the contrary, between some sites there are some major alterings (like Avrig and Valea Morii for example). That is why we have to take into consideration the proper sedimentation rate for each palynologically analysed sequence, not only to establish correlations between the recent and sub-recent spectra and the actual vegetation around the studied site or located nearby, but also to follow the evolution in time (positive or negative) and the respective vegetation dynamic trends, in favor or against some taxa.

There is no uniformity in the percentage calculation method for these sites, also. In older papers, the percentage ratio of all the taxa was done related to to the sum of the trees pollen (and eventually bushes - Σ A.P.), while in the modern ones, in our country or abroad, this percentage ratio is done to the sum of all the taxa pollen, both woody and herbaceous (Σ A.P. + Σ N.A.P.). In recent publications we used both methods, in order to allow a comparison of the palynological data, older or newer, obtained in different sites. In the present study, in order to avoid confusions, we will specify the way in which the *Quercus – Carpinus* forest ratio (related to to the woody vegetation or the total vegetation) was done for each discussed site.

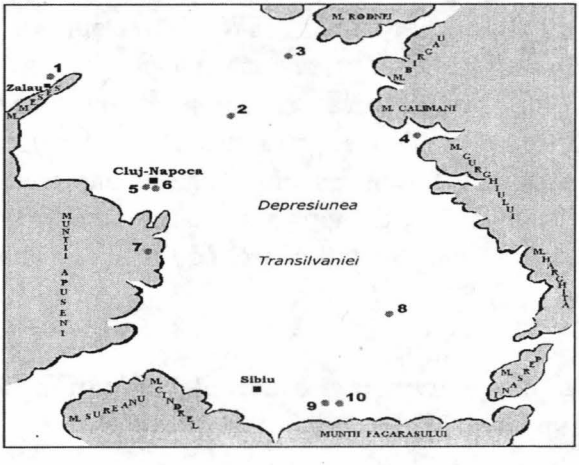

Fig. The location of the studied sites (1. Hereclean 2. Pădureni-Ţop 3. Zagra 4. Deda 5. Valea Morii 6. Sălicea 7. Colţeşti 8. Dracăşviz 9. Avrig 10. Arpaşul de Sus)

We have tried to establish the weight had by the *Quercus – Carpinus* forest in the Cluj region, in older times, based on palynological data obtained in Valea Morii (630 m.s.m.), Cluj County. Thus, considering the thickness of the analysed peat layers from the two sequences, I and II, belonging to the Subatlantic, which was of 120-130 cm, and taking into consideration the fact that the Subatlantic began 2800-3000 years ago, according to different chronologies, it results that the last analysed level from the surface, at 15 cm deep, has an age of approximately 300 years. It is plausible if we consider the scarcer peat genesis conditions offered by that biotope, that led to a low sedimentation rate.

From the performed palynological analysis results that ± 300 years ago, **the forests were covering 66.53% from all the vegetation of the region.** The forests from the hillocky zone, ***Quercus - Carpinus*** **respectively, were covering ± 44, 50% from the wooded surface and ± 29,61% from all the vegetation.** These *Quercus – Carpinus* forests were mainly composed by diverse species of *Quercus* (common oak, durmast oak and cerris), at least 30% altogether, by hornbeam with values of approx. 27% and by other wood essences.

Today, the *Quercus – Carpinus* forests cover 42,56% from all the wooded surface of the Cluj County; the average percentage of the forest cover in the low and hillocky zones of the Cluj County, with different wood essences, **is only 8,72%.** Thus, **a decrease of more than 20%** of the surfaces covered by the *Quercus – Carpinus* forests can be noted nowadays in comparison to 300 years ago when they were occupying ± 29,61%. **Similarly to the situation in Cluj County, the *Quercus – Carpinus* forests represent today less than 40% of the wooded surface of Transylvania. For the low and hillocky zones from Transylvania the average percentage of wooded surfaces is of 10,88%.**

In the last **two decades**, the surface of the hillocky forests of *Quercus sp.* (common oak with durmast oak, common oak with hornbeam, durmast oak with hornbeam, durmast oak with cerris) from Transylvania **has reduced by 2,5%**, because of the pest attack and the abusive clearance, **a rate that is more accelerated than the average one over tow decades, established by the palynological analysis, which is of 1,33-1,5%**; this is an alarm signal for the situation of the forests.

Although, the palynological analyses reveal an uninterrupted presence of the hillocky forests in Transylvania, starting from the Boreal till now, even they are decreasing.

Palynological analyses realized in the other mentioned sites point out the uninterrupted presence of the low and medium altitude forests from Transylvania, better represented than today by the *Quercus* and *Quercus – Carpinus* forests that had their maximum coverage in the warm periods of the Postglacial, in the Boreal-Atlantic and Subboreal respectively. They self-maintained at these low altitudes in the Subatlantic too, despite the wet and cold climate that favored beech development in the mountainous belt. These analyses strikingly point out the human impact upon the decidous forest, which is more increased nowadays.

Thus, at Colţeşti (600 m alt., Alba County), although the phase of the mixed oak with hazel tree and spruce fir was only lacunary found, for a thickness of 30 cm (Diaconeasa and colab., 1985), and that of the hornbeam was lacking, because of the sedimentary gaps, in the beech phase, occurred during the Subatlantic age, the mixed oak percentages related to to the other trees remain remarkable: maximum 48%. Inside these, the common oak prevails with a maximum of 38%.

On the contrary, up to the surface (the 5 cm level, having an age of less than 100 years), a decrease to half of these percentages is recorded. Regarding the hornbeam, even if it's maximum percentage in the sequence does not exceed 10% (because of the sedimentary gaps), up to the surface it's percentages drop to 1-2%. This phenomenon of the decidous forest percentage decrease, because of the clearance, is reflected in the pollen spectra starting from 50 cm deep, 500 years ago therefore. The percentages of the *Quercus – Carpinus* forest, from all the forests from Alba County are just 19,5% today.

In Bistriţa-Năsăud County, at Zagra (420 m alt.), V. Lupşa (1972) observed a substantial presence of the mixed oak in its own phase, that developed during the Boreal-Atlantic period (45% related to to the total summ of trees and shrubs). Inside these forests the elm dominated, with 36%, followed by the common oak with 13%.

The hornbeam phase is very expressively reflected, with a maximum of 33% hornbeam. At 10 cm level, with an approximately age of 150 years, the percentage of mixed oak decreases to half (22%) related to its absolute maximum, while that of the hornbeam decreases more drastically, at 1%.

Regarding the mixed oaks related to the total vegetation, in the 10 cm level they have a weight of approx. 10,13 % (6,16% common oak), plus 0,44% of the hornbeam, thus a total of 10,5% of the ± 150 years ago *Quercus-Carpinus* forests from the studied region.

Quercus-Carpinus forests cover today 22,3% from all the forests in Bistrița-Năsăud County. This percentage is almost identical to that recorded ± 150 years ago. So, we can notice that, although the surface of the forest has decreased very much today, the percentage of the *Quercus-Carpinus* forest did not modify in the structure of the forests from the county.

In Cluj County, besides the sequences from Valea Morii we mention those from Pădureni-Țop and Sălicea.

Diaconeasa has published in 1985 the pollen diagram from Pădureni-Țop (460 m alt.). The last analysed level from the surface is that of 25 cm deep, being, according to the estimations, almost 300 years old. At this depth a remarkable presence of the mixed oak can still be noticed inside the forests, of approx. 45%, from which the common oak has the biggest weight, with approx. 40%. Related to the mixed oak maximum found at the beginning of the Subatlantic (57% and 54%, respectively) a percentage decrease can however be observed as a consequence of the human impact.

The situation is similar for the hornbeam, too. Although the studied sequence was probably deposited in the second part of the Subboreal, the maximum of the hornbeam found in its own phase is significant (40%), while in the level from the surface (25 cm) its values decrease very much to approx. 15%.

Regarding the mixed oak presence in the total vegetation, at the 25 cm level, it covers approx. 20% (common oak 18%), at which approx. 6,66% is added by the hornbeam, thus a total of approx. 26,6% for the ± 300 years ago *Quercus-Carpinus* forests. The situation is extremely similar to that encountered ± 300 years ago on Valea Morii (29,61%).

At Sălicea (700 m alt.), Lupșa (1981) analysed a sequence of 440 cm, of atlantic age. This is how the situation of the mixed oak and hornbeam looks like (related to all the wooden vegetation in the period of maximum percentage of these wooden essences, found in the pollen diagram, in comparison to the situation from ± 200 years ago, i.e. the level of 10 cm depth): the mixed oak 52,5%, related to only 12%; the common oak 16,25% related to 10%, and **the hornbeam 81,3% related to to 6%!**

In the sequence from Deda, Mures County (Diaconeasa and Șuteu, 1980), the authors found extremely low percentages of the mixed oak and

hornbeam in the sub-recent level from 20 cm deep: 8% *Quercetum mixtum* from which common oak 5% (*Quercus*) and 1% hornbeam (*Carpinus*) related to all the trees pollen. These percentages are visibly higher at the depth of 50 cm: 14% *Quercetum mixtum*, 8% common oak and 3% hornbeam.

Also in the Mureş County, Tanţău and Fărcaş have recently (1997) done the palynological analysis of the Dracăşviz bog (420 m alt.). A characteristic of this sequence is the persistence of the mixed oak in the pollen spectra, having the Subatlantic age values (16-24% related to the trees pollen and 6,89-9,99% related to the total vegetation) similar to those from Boreal-Atlantic ages.

In the last analysed level from the surface, at 15 cm, that is probably quite old (200-300 years) because of the low sedimentation rate, the mixed oak percentages do not appear sensibly modified. Instead, the hornbeam has lower values (± 5%) in the 15 cm level related to its maximum in the sequence, of 42% (related to other trees). At the present, in Mures County *Quercus-Carpinus* forests have a coverage of 19% from all the wooden vegetation, situation that is similar to that encountered in Alba and Bistriţa-Năsăud County.

In Sălaj County, Şuteu and colab. (1978) performed a palynological analysis of the swamps ("taurile") from Hereclean, also recording the surrounding vegetation from that time. The diagram of the sequence, only 1 m long, is dominated by mixed oak whose percentage values (related to Σ A.P.) oscilates between 28-45%, although around the studied site the durmast oak forest was already missing at that time.

Concerning the hornbeam, it does not impress with big values in this sequence, but a progressive decrease of its values to the upper level (30 cm) can be observed. Here, the smallest percentage value, of approx. 2% is noticed. Today, in Salaj County a good situation of the *Quercus-Carpinus* forests is recorded related to the wooden vegetation and that is of 72,1% - a percentage that exceeds by far the average percentage value of the *Quercus-Carpinus* forests in Transylvania (less than 40%). Instead, the wooded rate of the county is very small, so the *Quercus-Carpinus* forests are few spreaded in reality.

In Sibiu County we have the palynological analysis recently done at Arpaşul de Sus (Fărcaş and colab., 2004 – in press) and Avrig (Tanţău and colab., 2004 – in press).

Concerning the presence of the *Quercus-Carpinus* forests in the palynological analyses at Avrig, the situation is like this: in the 5 cm level pollen spectrum, we found 15,67% mixed oak (related to all the vegetation) and 6,55% hornbeam. Therefore, concerning the *Quercus-Carpinus* forests we have a total of 22,22%, related to all the vegetation and 27,75%, related to all the wooden vegetation ± 150 years ago.

At Arpaşul de Sus (520 m alt.), the beech phase was quantified for a thickness of ± 285 cm. The 5 cm deep level would have an age of approx. 60 years. In the pollen spectra for this level the mixed oak has a percentage of 15,73%, related to the wooden vegetation, and only 5,59%, related to all the vegetation. We do not know, for the moment, which was its maximum distribution in its own phase (future research will clarify this aspect), but during the phase of the hornbeam and beech it did not have substantial values (maximum 15,38% and 10,71% respectively, related to all the vegetation).

Instead, the hornbeam, which in the up to surface level (5 cm) has only 3,93% from the wooden vegetation and 1,4% from all the vegetation, was much better represented before, in its own phase (the maximum = 58,98% and 53,54% respectively) and also in that of the beech (the maximum = 45,06% and 38,91% respectively).

At 5 cm deep we therefore notice a value of the *Quercus-Carpinus* forests of 19,66% from the total of forests inside the zone and 6,99% respectively, from all the vegetation, while their maximum representation during the Subatlantic age was much higher (i.e. 49,4% and 42,66% respectively, related to all the vegetation at 125 cm deep; 65,42% and 59,38% respectively at 355 cm etc.). Today, in Sibiu County there are values of 34,8% of the *Quercus-Carpinus* forests from all the wooden vegetation.

4. Conclusions

The question that rises is: what replaced the *Quercus-Carpinus* forests from Transylvania, being in decline because of the human activities. As the pollen diagrams show there are two main trends: one belongs to the wooden vegetation and the other to the herbaceous one. The herbaceous associations, secondary installed instead of the broken up *Quercus-Carpinus* forests, were mentioned when the actual vegetation was described. Concerning the wooden vegetation, the following aspects can be observed: in the sunny places, with lesser acidophilous soils, hazel trees generally

develop, belonging to the association *Corylo-Populetum tremuli,* while on the shaded slopes with acidic substrate, the birch groves with poplar prevail, belonging to the association *Populo-Betuletum pendulae* Coldea 1972.

The recent and sub-recent pollen spectra generally reveal with accuracy the reduction, or at least the decreasing trend of the dynamics of the wooden taxa that are specific to plain and hillocky zones. The biggest threat for the perpetuation of these natural forests is represented today not by the climatic deterioration, but rather by the human intervention, by clearance and grazing, activities that risk to destroy the natural balance in this ecosystem. Through the decrease of the forests surfaces or the modification of the wooden essences spectra, the hydric balance and the source of oxygen, represented by these forests, are endangered and also the composition and the stability of the wooded terrains.

The afforestation of the altered terrains (approx. 250.000 ha) from the Transylvanian hillocky zone is of uttermost importance for the prevention of some ecological lack of balance and for stopping the desertification process. In the light of phytohistorical data, respecting the initial structure of the forests is essential for the afforestation in the Transylvanian hillocky zones.

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L'HISTOIRE DE LA VÉGÉTATION TARDIGLACIAIRE ET POSTGLACIAIRE DU SUD DE LA ROUMANIE, REFLÉTÉE PAR LES ANALYSES PALYNOLOGIQUES

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Rezumat. Istoria vegetaţiei tardiglaciare şi postglaciare din sudul României, reflectată prin analizele palinologice. Lucrarea prezintă câteva date privind particularităţile vegetaţiei tardiglaciare şi holocene din sudul, sud-vestul şi sud-estul României, aşa cum acestea au fost evidenţiate prin analizele palinologice. În aceste regiuni, arborii foioşi termofili au apărut mult mai timpuriu decât în restul României. Analizele de polen efectuate în aceste regiuni demonstrează, chiar şi în lipsa unei cronologii absolute, existenţa refugiilor glaciare pentru elementele stejărişului amestecat (stejar, ulm, tei).

Contribuţii noi sunt aduse prin realizarea unor analize palinologice într-o secvenţă de guano, obţinută din Peştera Liliecilor de la Gura Dobrogei, situată în Dobrogea centrală. Rezultatele palinologice obţinute par să indice, în lipsa unor datări precise C^{14}, Subatlanticul (posibil Subborealul târziu) pentru constituirea depozitului de guano analizat. În diagrama obţinută pentru arbori şi arbuşti se remarcă lipsa polenului de conifere (în principal pin, molid şi brad) şi participarea foarte scăzută a celui de fag, spectrele polinice fiind dominate de elementele stejărişului amestecat şi de carpen, care atestă continuitatea acestor păduri în Dobrogea.

Din raportul A.P./N.A.P. reiese subordonarea permanentă a polenului de arbori, faţă de cel de ierboase, dintre care cel mai bine reprezentat este cel de graminee. Spre suprafaţă se înregistrează creşterea şi amplificarea taxonilor ierboşi cu semnificaţie antropică.

Cuvinte cheie: palinologie, fitoistorie, Tardiglaciar, Holocen, sudul României, guano, Peştera Liliecilor

I. Introduction

Les analyses palynologiques effectuées en Roumanie pendant plus de sept décennies ont établi l'histoire de la végétation tardi- et postglaciaire, c'est á dire la succession sylvestre, avec les phases typiques bien connues.

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Les différences qui apparaissent dans la chronologie et la composition des associations sporo-polliniques, caracteristiques á chaque phase sylvestre sont provoquées par l'altitude et les coordonnées géographiques (latitude et longitude) différentes de stations palynologiques.

La conservation du pollen et des spores fossiles demande certaines conditions, qui sont bien accomplies par les marais de tourbe, surtout les oligotrophes. Par conséquence, la plupart des analyses palynologiques effectuées en Roumanie ont envisagé les stations de haute altitude, qui ont permis la mise en place des tourbières oligotrophes, et moins l'étage collinaire ou les zones de plaine (tourbières eutrophes, lacs, paléosols, loess, dépôts de caverne etc.). Les résultats palynologiques obtenus dans ces types de sédiment permettent plus difficilement le rangement dans les phases sylvestres classiques, préétablies, à cause du degrée faible de conservation du pollen fossile et de la densité très réduite de celui-ci dans le matériel étudié.

II. L'histoire de recherches palynologiques dans le sud, sud-ouest et sud-est de la Roumanie

On faisant la révision chronologique des données palynologiques postglaciaires, obtenues dans les zones d'altitude basse et moyenne de la région envisagée, on constate qu'elles sont insuffisantes. Les datations C^{14}, absolument necéssaires pour établir une chronologie absolue manquent. A cause de la faible densité du pollen fossile conservé dans ces sédiments, de la domination du pollen des hérbacées par rapport au celui des arbres (à quelques exceptions), de l' aspect corrodé de l'exine du pollen et du degrée différent de décomposition de diverses types de pollen, dans certaines situations il est difficile à établir même la chronologie rélative, appuyée sur les phases sylvestres.

Le premier étude palynologique quaternaire effectué dans cette région appartient au Pop. Il a présenté en 1956 un exposé intitulé "Analyses de pollen dans des régions de plaine" (publié en 1957), dont il a fait une analyse critique de la tourbière eutrophe de Craiovița (110 m d'altitude). L'auteur arrive à des conclusions très intéressantes concernant le problème de la steppe, du hêtre et de l'âge de la chênaie mixte. Pop considère que la sédimentation de la turbe eutrophe de Craiovița a commencé pendant le Postglaciaire, pendant la domination de la chênaie mixte (*Quercetum mixtum*). Le marais se trouvait dans la zone du chêne (*Quercus*), et les

conditions climatiques actuelles sont encore favorables à la végétation sylvestre.

L'auteur refuse catégoriquement l'hypothèse de la descendance directe de ces chênaies à partir de celles tertiaires, mais il n'exclu pas l'existence des possibles refuges glaciaire pour les feuillus dans la région d'Olténie, hypothèse favorisée par les taux élevés du pollen des chęnaies obtenus à Craioviţa (le maximum du chêne = 72%, le maximum de la chênaie mixte = 82%). En ce qui concerne le hêtre (*Fagus*), les résultats palynologiques obtenus à Craioviţa ont déterminé Pop à penser que les hêtres d'autour de Craiova et les communautées plus nordiques ne sont pas des reliques tertiaires, ni au-moins postglaciaires. "Elles représentent des infiltrations récentes, des dernières deux-trois milles d'ans...".

En 1964, Iliescu et Cioflica ont publié "Ètude palynologique sur les carrières de Pantelimon", qui représente une synthèse des données obtenues dans 8 carrière. Les auteurs ont décelé 4 phases climatiques: froide, de transition, optime et plus froide, auxquelles correspondent des associations végétales de toundra (conifères, bouleau, aune et herbacées), de passage vers la steppe (la prédominance des herbacées, l'apparition les arbres thermophiles, par exemple le tilleul), de steppe avec *Ericaceae* et plus de tilleul (*Tilia*), ensuite de steppe avec *Chenopodiaceae, Asteraceae, Poaceae.*

Le même année, Iliescu et Ghenea ont étudié les dépôts loessoides de la Plaine d'Oltenie, plus précisément de Rogova, sur la terrasse supérieure du Danube (la terrasse Băileşti, à 36 m d'altitude). Ici, de même, ont été mises en évidence les mêmes 4 phases climatiques, soutenues par la présence des associations de toundra, avec 85% conifères, avec bouleau (*Betula*), aune (*Alnus*), saule (*Salix*), charme (*Carpinus betula*), charme d'Orient (*Carpinus orientalis*) et diverses familles d'herbacées, puis des associations steppiques avec des arbres et de la sylvosteppe avec des nombreux éléments thermophiles. Finalement, la dernière phase est caractérisée par la réduction des éléments thermophiles en faveur des conifères et par le développement des herbacées.

Des aspects palynologiques sont présents aussi dans l'ouvrage de Grumăzescu et Stăncescu-Grumăzescu (1967), concernant la signification paléogeographique de certains dépôts quaternaires du bord danubien de la Dobroudja de nord.

La même année, Boşcaiu et Lupşa ont publié les résultats des analyses palynologiques effectuées dans le sud-ouest de la Roumanie ("La

Grotte des Haïdouks" et "La Grotte de Veterani"), analyses favorisées par l'intense circulation des courants d'air dans ces grottes, qui ont transporté et ont sédimenté de grandes quantités de pollen atmosphérique.

Les études archéologiques effectuées antérieurement par Nicolăescu-Plopşor et ses collaborateurs à Grotte des Haïdouks ont décelé dans la structure du sédiment une couche supérieure attribuée au Néolithique, avec des vestiges des périodes de transition a l'Âge du Bronze et du Fer, la période Daco - Romaine et Féodale ancienne, et une couche inférieure qui porte des vestiges du Mésolithique.

L'existence de ces deux couches a été confirmée aussi par les analyses palynologiques. Les feuillus dominent dans la majorité des spectres polliniques de la couche supérieure. On constate la domination du pollen de hêtre (*Fagus* = maximum 40%), ce qui correspond au Subatlantique, pendant que dans les niveaux les plus superficiels ses fréquences diminuent beaucoup, à cause des déforestations de la période historique.

La couche inférieure de la séquence analysée met en évidence la domination du pollen de *Pinus* type d*iploxylon*. Les auteurs considèrent qu'il appartienne au type megatherm *Pinus nigra*, représenté aujourd'hui dans la vallée de Cerna par *P. nigra ssp. pallasiana*. Dans le niveau basal (140 cm de profondeur), le pollen de pin augmente jusqu'à 90%, dont la plupart appartienne à *P. sylvestris*, à caractère microthermique. La base de la séquence est attribuée au Boréal.

L'analyse palynologique des niveaux inférieurs de la séquence de La Grotte de Veterani a relevé expressivement la phase du pin (300 – 430 cm de profondeur), avec un maximum de 96% du pollen de *Pinus*, attribué par les auteurs aux types *Pinus sylvestris* et *Pinus nigra*. Le dernier est représenté aujourd'hui au Défilé du Danube par *Pinus nigra ssp. pallasiana*. Les auteurs supposent que les pinèdes existantes pendant la dernière glaciation dans cette région, aussi comme dans les Monts Cernei, ont été représentés par des groupements de *Pinus sylvestris*, mais avec "des enclaves de populations préglaciaires de *P. pallasiana*".

La continuité de la courbe du pollen de sapin (*Abies*) représente un intérêt phytogéographique à part dans ces spectres polliniques. Cela confirme la supposition de l'existence des refuges pour les populations d'*Abies*, pendant la glaciation, aux environs. Les chênaies mixtes (*Quercetum mixtum*) aussi sont bien représentées dans les spectres polliniques, grâce au climat plus doux de la région. Dans l'opinion des auteurs, ce climat "a assuré

leur survivance pendant la dernière glaciation, sur les rochers abrités de Défilé du Danube".

Les spectres polliniques de la Grotte de Veterani ne surprennent pas la transition de la phase de pin vers la phase de la domination des essences thermophiles. De toute façon, l'expansion de ces dernières dans le Défilé du Danube a été vertigineuse grâce à l'échauffement du climat postglaciaire. La discontinuité du diagramme pollinique ne permet pas l'enregistrement du moment de l'apparition du hêtre (*Fagus*). L'opinion des auteurs est que l'apparition du hêtre dans le Défilé du Danube doit être beaucoup plus précoce que dans les autres régions du pays, peut être même en début de Subboréal. Il est à signaler la présence d'une très courte phase de charme (*Carpinus betula* et *C. orientalis*), interposée entre la phase de la chênaie mixte et la phase de hêtre, dont les pourcentages de charme atteint un maximum de 25%.

En général toutes ces suppositions, concernant l'évolution de la végétation de Monts Cernei et de Défilé du Danube sont confirmeés par les analyses effectuées ultérieurement de mêmes auteurs, dans les grottes Cuina Turcului-Dubova (1970) et la Grotte de Climente (1971), situées aussi près de la localité Dubova, mais à une altitude plus élevée (270 m).

En 1968, Boşcaiu et colab. ont publié des données concernant l'histoire des forêts de Monts Cernei, suivies en 1971 par la monographie de Boşcaiu au sujet de Monts Ţarcu, Godeanu et Cernei, avec un chapitre d'histoire de la végétation de cette région. Ici sont, de même, présentées les données palynologiques obtenues dans les Monts Cernei (La Grotte des Haïdouks, Topenia).

En 1970, Conea a publié "Formations quaternaires en Dobroudja - des loess et des paléosols", dans laquelle Iliescu et Cioflică présentent les résultats de l'analyse palynologique effectuée dans la section nr. 3, Ovidiu. Ces résultats montrent l'existence de deux formations végétales caractéristiques: pour le sol actuel et son matériel parenteral une végétation de steppe et pour les sols sylvestres une végétation forestière. Les spectres sporo-pollinique du matériel loessoid et du cernosium actuel ne sont pas dominés par les graminées, ce sont les *Asteraceae* et les *Chenopodiaceae* qui dominent. Le N.A.P. total est 74-89%. Les auteures supposent que la steppe développée pendant la formation du loess était différente de celle développée pendant la formation du cernosium, parce qu'elle était dominée par *Chenopodiaceae* et des herbes xérophiles, tandis que le pollen des arbres (8-

22%) dans le cernosium actuel et son matériel parenteral a été apporté par le vent.

Dans les sols sylvestres étudiés, différents comme âge (l'un interstadial et deux interglaciaires), les pourcentages A.P. sont 80 - 89%. Les forêts reflétées par ces pourcentages étaient composées par: orme (*Ulmus*), chêne (*Quercus*), charme (*Carpinus*), rarement érable (*Acer*) et tilleul (*Tilia*). La fréquence du pollen de noisetier (*Corylus*) et de hêtre (*Fagus*) est très faible. La composition de la végétation herbeuse de ces sols est différente de celle steppique: la famille d'*Asteraceae* domine, tandis que les *Poaceae* sont beaucoup plus frécquentes que les *Chenopodiaceae*.

En 1971, Ban et Alexandru ont publié un très intéressant ouvrage, dans lequel ils montrent la correspondance entre les spectres sporo-polliniques actuels et les conditions physico-géographiques dans la partie sud-est de la Roumanie. Les prémisses de départ pour les auteurs sont: la productions de pollen d'une certaine espèce est différente dans des régions différentes et le type de sédiment, par les conditions caractéristiques de conservation offertes, influencent la composition des spectres polliniques.

Les auteurs ont étudié trois zones physico-géographiques: les forêts du nord de Dobroudja, la sylvosteppe de Dobroudja et la steppe de Dobroudja et Bărăgan. Ils ont constaté que la participation de la groupe A.P. se réduit de 79,4% (le maximum de la zone des forêts) à 24,1% dans la silvosteppe et 15,6% dans la steppe.

La même annee, Boşcaiu présente l'évolution postwürmienne de la végétation du Défilé du Danube, à l'occasion de la troisième conférence internationale de palynologie de Novosibirsk, pendant que Lupşa et Măgălie publient les résultats des recherches palynologiques effectuées dans le Plateau Mehedinţi (Dépression Balta).

En 1972, il est apparu l'ouvrage de Ban et Alexandru, concernant le pollen de conifères dans les spectres sporo-polliniques du sud-est de la Roumanie, et celui de Conea et Roman, qui envisage des aspects concernat l'évolution de quelques sols de la Plaine Roumaine.

En 1975 ils ont apparu aussi des travaux sur les analyses palynologiques effectuées au sud de la Roumanie, dont nous pouvons citer celles de Alexandru, à Snagov Gruiu, respectivement de Apostol et Lerkovskaia, de la région Codreni (Ilfov).

En 1977, Diaconeasa a publié l'ouvrage "La valeur documentaire phytohistorique de la tourbière de Mangalia-Herghelie (département

Constanța)", d'une grande valeur phytohistorique, dont on va revenir dans les pages suivantes. La même année, Alexandru a publié des aspects concernant le pollen des hérbacées, ouvrage suivi en 1980 par l'analyse palynologique de la séquence Greaca et par les analyses palynologiques effectuées sur quelques dépôts caractéristiques du Delta du Danube.

En 1982, Boşcaiu et colab. ont publié les résultats de l'analyse palynologique effectuée sur les sédiments obtenus de la tourbière Le Beau Lac-Mosoroasa, près de Băile Olăneşti (les Subcarpates d'Olténie). La succession sylvestre reconstituée par les auteurs a mis en évidence le fin de la phase de l'épicéa avec de la chênaie mixte (Atlantique), la phase du charme avec du hêtre (Subboréal) et la phase du hêtre de Subatlantique.

Diaconeasa et Mitroescu ont effectué en 1988 l'analyse palynologique de quelques lacs de la Plaine Roumaine: Balta Albă (dept. Buzău), Lacul Sărat et Iazu-Movila Miresii (dept. Brăila). Les résultats obtenus ont relevé la présence du pollen de hêtre (*Fagus*), avec des pourcentages élevés (le maximum = 40,24%), fait qui suggère l'âge subatlantique pour les bous lacustres analysés. La phase de la chênaie mixte avec du hêtre, caractéristique pour la zone de plaine pendant le Subatlantique, est confirmée par les pourcentages élevés du pollen de la chênaie mixte (*Quercetum mixtum*), trouvés dans les spectres analysés.

Le pollen de hêtre (*Fagus*), d'épicéa (*Picea*), du pin (*Pinus*) et du sapin (*Abies*) identifié dans ces spectres, est considéré par les auteurs comme transporté par les courants d'air, de forêts de Monts Vrancei. Le pollen des herbacées (sourtout les *Chenopodiaceae, Poaceae, Artemisia*) dominent les spectres polliniques. Cela indique le développement d'une végétation forte halophile autour des lacs, ainsi que le caractère steppique de la végétation induit par l'impact humain.

Les mêmes auteurs ont étudié du point de vue palynologique le lac Techirghiol (publié en 1991-1992), conformément aux 6 forages géologiques. Ils ont encadré les résultats obtenus dans les phases sylvestres spécifiques pour les zones de plaine: la phase de la chênaie mixte avec du noisetier, la phase de la chênaie mixte avec du charme et la phase de la chênaie mixte avec du hêtre. Les auteurs ont constaté, comme à Mangalia-Herghelie, la présence des forêts de feuillus, parmi les forêts de pin, à la fin du Tardiglaciaire. Ces forêts de feuillus contenaient des espèces de chêne (*Quercus*), orme (*Ulmus*), tilleul (*Tilia*), érable (*Acer*), charme (*Carpinus*), mais pas du hêtre (*Fagus*).

Très récemment (2000), Tomescu a publié une évaluation des spectres polliniques holocènes de la Plaine Roumaine. L'étude critique de 15 séquences holocènes (deux sédiments naturels, 10 dépôts anthropiques et deux mixtes) lui ont permis la reconstitution de la végétation holocène, dans une succession générale de steppe – forêt – steppe. Toutefois, l'auteur remarque que les spectres polliniques qui proviennent du lœss, de la tourbe alcaline ou des stations archéologiques sont moins crédibles pour les reconstitutions de la végétation, et il considère les spectres des sédiments lacustres en reflétant plus fidèle l'histoire de la végétation de la région.

III. L'évolution de la végétation tardi- et postglaciaire de Roumanie par rapport à celle de Balkans

On connaît maintenant assez bien la succession des associations sporo-polliniques tardiglaciaires et holocènes des stations de Roumanie dont on dispose des datations C^{14}. Celles-ci constituent points de repère dans la chronologie des séquences et rendent plus vraisemblable les comparaisons. De cette analyse on peut tirer des conclusions intéressantes concernant l'évolution d'ensemble de la végétation de Roumanie, en contexte européen, mais aussi sur l'évolution de la végétation dans le sud, sud-est et sud-ouest de la Roumanie.

Les diagrammes sporo-polliniques récentes (de dernières 5 années), accompagnés par des de datations C^{14} proviennent (chronologiquement) de Iezerul Căliman (Monts Căliman), Tăul Zănoguţii (Monts Retezatului) (Fărcaş et. colab. 1999, 2003), Monts Semenicului (Rösch et Fischer 2000, Fărcaş et colab. 2003), Preluca Ţiganului (Wohlfarth et colab. 2001, Björkman et colab. 2002) et Şteregoiu (Monts Gutâiului) (Feurdean et colab. 2001, Björkman et colab. 2002, 2003, Fărcaş et colab. 2003), Ic Ponor, Padiş, Cimitir, Stână (Bodnariuc et colab. 2002, Fărcaş et colab. 2003) et Căpăţâna (Monts Apuseni) (Fărcaş et colab. 2003), Mohoş (Tanţău et colab. 2003, Fărcaş et colab. 2003) et Lucs (Monts Harghitei) (Tanţău et colab. 2003), Bisoca (Subcarpates de Courbure) (Tanţău et colab. 2003 – sous presse), Poiana Ştiol (Monts Rodnei) (Tanţău et colab. 2003 – sous presse) et Avrig (Dépression du Făgăraş) (Tanţău 2003 – sous presse).

En ce qui concerne la présence des éléments de la chênaie mixte dans le Tardiglaciaire, elle a été confirmée palynologiquement dans des séquences datée de Roumanie (Iezerul Căliman, Mohoş, Tăul Zănoguţii etc.), mais en

faibles quantités, qui ne justifient pas leur présence "in situ", mais le transport du pollen de grande distance.

La situation est différente dans le sud-est de notre pays, illustrée d'une manière suggestive par les analyses palynologiques effectuées à Mangalia (Diaconeasa 1977). Ici, dans les spectres basals attribués au Tardiglaciaire, âge démontré par les pourcentages très élevés de pin (75,33%) mais sans avoir des datations C^{14}, il s'enregistrait déjà des pourcentages substantiels (14,6%) de la chênaie mixte, et pas des valeurs basses et disparates, telles quelles apparaissent dans le Tardiglaciaire des spectres polliniques de séquences datées de Căliman, Retezat et Mohoş.

Dans la vision de l'auteur "ces données palynologiques démontrent, contrairement aux celles de Transylvanie, que dans le sud et le sud-est du pays, pendant le Tardiglaciaire ont coexisté les pinèdes et les chênaies mixtes dans des écosystèmes bien développés, avec la différence que les pinèdes, aujourd'hui disparus de Dobroudja, étaient 4-5 fois plus répandus que les chênaies-mixtes" (Diaconeasa 1977).

S'ils existaient dans les zones basses, de plaine, des pinèdes déjà mélangés aux chênaies mixtes dans cette période froide du Tardiglaciaire, il faut admettre l'hypothèse de l'existence des refuges glaciaires assez près des sites étudiés, aux altitudes moyennes, où les éléments thermophiles ont trouvé des niches écologiques avec des conditions propices pour survivre aux rigueurs climatiques de la glaciation.

A Mangalia, dans le niveau situé en dessus du sédiment argileux basal et attribué à l'Alleröd, apparaît un spectre pollinique avec 51,33% pollen de pin et 35,33% pollen de la chênaie mixte. Même dans les spectres appartenant au Dryas récent il y a du pollen de la chênaie mixte (17%), avec le chêne comme élément dominant.

Ces pourcentages élevés du pollen de la chênaie mixte des régions de sud-est de la Roumanie, pouvaient être reflétés par le transport lointain dans les spectres polliniques des Carpates attribués au Tardiglaciaire, fait qui suggère l'existence d'une migration de ces éléments thermophiles vers le nord et nord-ouest, au fur et à mesure de l'échauffement du climat.

Les informations palynologiques fournies par le profil de Mangalia montrent la réalité, si on tienne compte aussi des résultats palynologiques obtenus à Gramousti et à Rezina, au nord-ouest de la Grèce, dans les Monts Pinde (Willis 1992 a, b, c). Ces informations présentent des similitudes avec celles de Ioannina (Bottema 1974).

On a suggéré depuis longtemps que les taxons ligneux ont survecu dans les régions montagneuses de Grèce et de Balkans pendant les stades froids du Quaternaire (Lang 1970, van der Hammen et colab. 1971, Bottema 1974, West 1980, Huntley et Birks 1983). La survie de la flore tempérée dans ces régions a été directement liée aux conditions climatiques des stades froids, la plupart de ces régions n'étant pas couverte par glace (Denton et Hughes 1981). L'idée de la migration des taxons tempérés des refuges glaciaires de Balkans vers le nord de l'Europe dans l'Holocène précoce est bien établie et documentée (Huntley et Birks 1983, Bennett et colab. 1991, Willis 1992a, b, c, 1994, Tzedakis 1993).

En Bulgarie aussi, près de Mangalia, dans les spectres polliniques des lacs Durankulak (Bozilova et Tonkov 1985, Bozilova et Atanassova 1990) et Varna (Tschakalova et Bozilova 1984, Bozilova et Ivanov 1985, Bozilova 1986, Bozilova et Filipova 1986), au bord de la Mer Noire, même si le Tardiglaciaire n'est pas enregistré, le chêne était déjà présent dans l'Holocène précoce, il y a 9.000 d'ans (attésté à la base du profil de Varna par 20%). Pendant l'Holocène précoce, au bord de la Mer Noire il poussait une végétation steppique, avec des *Poaceae*, *Chenopodiaceae, Asteraceae* et *Artemisia*, parsemée des groupements d'arbres et d'arbustes, parmi lesquels le chêne, l'orme, le tilleul, le noisetier et le bouleau (Bozilova et colab. 1996).

Dans les profils d'âge tardiglaciaire des monts Rila et Rodopi, à Sucho Ezero (Bozilova et Smit 1979, Bozilova et colab. 1990) et à Kupena (Bozilova et colab. 1989), les palynologues bulgaires ont trouvé du chêne, même avec plus de 20% (à Sucho Ezero, où il est accompagné aussi par le noisetier, avec les même pourcentages élevés), preuve de sa présence tardiglaciaire dans la région.

Même si on ne peut pas affirmer avec cértitude, mais on suppose **la présence des refuges glaciaires de la chênaie mixte en Roumanie aussi**, aux altitudes au moins de 300 – 400 m des régions sud – sud-est (par exemple Dobroudja), il est plus difficile à admettre l'existence des refuges glaciaires pour le charme, le hêtre et le sapin, qui généralement manquent dans les spectres polliniques tardiglaciaires datés avec C^{14} des Carpates roumains. Ces taxons sont attéstés par leur pollen plus tard.

Dans le spectre de Mangalia (Diaconeasa 1977), l'auteur signale la présence du pollen de sapin (4%) et de charme (0,6%), avec des pourcentages réduites, pendant l'Alleröd. Malheureusement, il manque à Mangalia les informations phytohistoriques de transition, caractéristiques au Préboreal,

qui auraient pu donner des indices sur le moment de l'expansion du charme et du hêtre. La présence des chênaies – charmaies, mise en évidence à Mangalia et Techirghiol (Diaconeasa et Mitroescu 1991-1992) pendant le Postglaciaire précoce, avec des valeurs significatives, reste un moment de référence pour l'histoire des forêts de Roumanie, et elle indique probablement **l'affirmation du charme beaucoup plus précoce dans le sud-est (Dobroudja), par rapport au reste de la Roumanie** (Diaconeasa et Fărcaş 1998).

Conformément aux ces données concernant l'apparition et l'expansion des taxons ligneux, on peut tirer des conclusions concernant la chronologie, la vitesse et les routes de migration de ces taxons, si on tient compte aussi des mentions apparues dans la littérature de spécialité, dans ou sur les pays balkaniques, comme centres de migration de ces taxons des refuges glaciaires.

En Bulgarie aussi, le sapin et le charme ont été signalés dans les monts Rila et Pirin (Bozilova 1975) pendant l'Alleröd. L'auteure suppose qu'r ce temps-là il existait même un étagement de la végétation, dont les forêts de hêtre ne manquaient pas. À proximité du littoral de la Mer Noire, à Durankulak (Bozilova et Tonkov 1985, Bozilova et Atanassova 1990), le charme est attésté d'il y a ± 8.500 ans, et le hêtre d'il y a ± 7.000 d'ans. À Shabla-Ezeretz, Filipova (1985) a confirmé aussi avec C^{14} la présence du hêtre d'il y a ± 7.000 d'ans, prèsque simultanément avec l'apparition du sapin et du hêtre dans les diagrammes des monts Stara Planina et Sredna Gora (Filipovich 1977, Petrov et Filipovich 1987).

Les diagrammes de Bulgarie montrent une phase de la maxime extension du charme pendant l'Holocène précoce, tandis que pour le hêtre ils suggèrent une expansion tardive (Bozilova et Beug 1992, Bozilova et colab. 1996).

En Grèce, la présence tardiglaciaire du sapin (Rezina), du charme (Kopais) ou du hêtre (Ioannina) est attéstée par C^{14} dans des diverses diagrammes (Allen 1986, 1990, Bottema 1974, Willis 1992), preuve de leur présence dans des refuges glaciaires de région, mais le moment de leur expansion est aussi différencié. Ce moment, aussi bien que la date de la mise en place de ces forêts qui peut être déduite dans les diagrammes sporo-polliniques tardi- et postglaciares, est aussi important.

En Bulgarie, à la différence de Roumanie, les forêts de sapin s'installent beaucoup plus vite et marquent leur affirmation maximale

pendant le Subboréal, tel que le diagramme de Monts Konjavska, à Tschokljovo (Tonkov 1988, Tschakalova et colab. 1990, Tonkov et Bozilova 1992) le montre. Quant au charme, le moment de son expansion dans la région du littoral bulgaire de la Mer Noire, s'est passé il y a ± 7.000 d'ans, et la phase de sa maxime extension s'est déployée il y a ± 5.800 d'ans, donc pendant la derniére partie de l'Atlantique (Bozilova et colab. 1996).

Toutes ces données palynologiques montrent qu'en Dobroudja, ou les forets de charme sont beaucoup plus vieilles que celles de Transylvanie et de Carpates roumaines, le charme est arrivé du sud de la Péninsule Balkanique et puis il a migré au nord-nord-ouest, tant à l'extérieur qu'á l'intérieur de la chaîne carpatique roumaine (Diaconeasa et Fărcaş 1998). Les taxons ligneux qui ont formé leurs phases plus vite qu'en Roumanie, c'est à dire le sapin en Bulgarie, respectivement le sapin et le hêtre en Slovénie, ont immigré probablement de ces centres vers la Roumanie, où ils ont développé leur phases seulement pendant le Subatlantique.

À part des datations C^{14} dont on dispose pour diverses stations de Transylvanie et des Carpates roumains, on a effectué très récemment et en première de telles datations sur le dépôt de guano de la Grotte d'Adam (295 m altitudine), sur la vallée de Cerna, près de la station Băile Herculane. Les résultats ont été publiés par Carbonnel et colab. (1996, 1999). L'âge obtenu pour la base du dépôt (250 cm), indique le Boréal pour la mise en place du dépôt. Les deux échantillons de guano, de 120 et 250 cm de profondeur ont fourni les datations C^{14} suivantes: 910-810 ans B.C. et 6.470-6.360 ans B.C. (âge calibré), respectivement 2.740 ± 60 ans B.P. et 7.600 ± 80 ans B.P. (âge conventionnel).

La datation du dépôt de guano de la Grotte d'Adam repésente la première tentative de datation de ce type de matériel organique dans l'Europe, d'une importance remarquable. Elle donne des repères chronologiques certes pour l'analyses palynologique effectuée sur ce dépôt par Girard et Bui Thi Mai (manuscrit). On peut interpréter le diagramme sporo-pollinique rédigé par ceux-ci, par le filtre des informations palinologiques obtenues par Boşcaiu et colab. (1967) dans les grottes de Monts Cernei et de Défilé du Danube. Malheureusement, l'analyse palynologique du guano de la Grotte d'Adam s'arrête à environ 165 cm de profondeur.

Si on tient compte du pas de sédimentation différent des niveaux soidits "supérieurs", au- dessus de 120 cm de profondeur et les "inférieurs"

(120 - 250 cm de profondeur), nous pouvons éxtrapoler l'âge approximatif du niveau 165 cm, d'où l'analyse palynologique commence. La datation C^{14} du niveau 120 cm reflète, conformément au schéma chronologique élaboré par Orombelli et Ravazzi (1996), le moment de la transition de Subboréal au Subatlantique, qui a commencé il y a ± 2.700 d'ans.

D'après nos calculs, confirmés par l'échelle chronologique du sédiment de guano, rédigée par Carbonnel et colab. (1999), le niveau 165 cm pourrait corréspondre á l'âge approximatif de 4.500 d'ans, c'est à dire au Subboréal précoce, donc le diagramme pollinique élaboré par Girard et Bui Thi Mai reflète l'histoire de la végétation à l'alentours de la grotte à partir de cette période-là. Malheureusement, ils manquent les indications très importantes concernant les périodes plus anciennes.

IV. L'analyse palynologique du dépôt de guano de La Grotte des Chauves-Souris de "Gura Dobrogei"

Pour apporter des données supplémentaires concernant l'histoire de la végétation du sud de la Roumanie nous avons effectué récemment l'étude d'un dépôt de guano de La Grotte des Chauves-Souris de "Gura Dobrogei" ("La Bouche de Dobroudja"), située en Dobroudja centrale, au nord de la localité Târguşor et près du village Gura Dobrogei (Fig. 1).

On a récolté une séquence de guano, à l'aide d'un carottier russe modifié, jusqu'à 120 cm de profondeur. La séquence a été échantillonée aux distances égales (tous les 2,5 cm) et les échantillons obtenus ont été soumis à la préparation chimique, dans le Laboratoire d'Écologie Terrestre de l'Université Paul Sabatier de Toulouse, France. On a obtenu 48 préparations microscopiques qui ont été sommairement examinées au microscope, pour une première sélection. Une partie de ces préparations se sont avérées stériles, des autres extrêmement pauvres en pollen, ainsi qu'on a sélectionné seulement 9 préparations. Même dans certaines de ces préparations, la densité du pollen des arbres et des arbustes (A.P.) est très réduite par rapport à celle du pollen des herbacées (N.A.P.), ce qui donne aux tableaux et aux diagrammes obtenus seulement une valeur rélative.

Par la représentation graphique des spectres polliniques des 9 préparations analysées, nous avons obtenu le diagramme des arbres et des arbustes (Fig. 2), et le diagramme des familles de herbacées, des taxons herbeux et des spores (Fig. 3), dont les pourcentages de chaque taxon,

respectivement chaque famille, ont été obtenus par rapport à la somme totale.

Dans le diagramme des arbres et des arbustes (Fig. 2), on observe d'abord l'absence du pollen de conifères, à l'exception de quelques apparitions extrêmement isolées du pollen de genévrier (*Juniperus*). Le pin (*Pinus*), l'épicéa (*Picea*) et le sapin (*Abies*) manquent complètement.

Si on regarde le diagramme de La Grotte d'Adam (Girard et Bui Thi Mai, manuscrit), on constate aussi la diminution marquée du pollen de conifères, en plein Subatlantique, à partir du niveau 80 cm vers la surface. La situation des conifères est semblable dans les séquences analysées par Boşcaiu et colab. (1967) dans La Grotte de Veterani et dans La Grotte des Haïdouks, surtout si on tient compte que les auteurs ont effectué les calculs des pourcentages seulement par rapport à la somme A.P. (Σ A.P.). Cette méthode donne des taux des taxons plus élevés que par rapport à la somme totale (Σ A.P. + Σ N.A.P.).

Les diagrammes obtenus par nous, de même que celui de La Grotte d'Adam, sont réalisés par rapport à la somme totale, donc les taxons apparaissent avec des taux plus réduits. Même dans ces conditions, dans le diagramme de La Grotte des Haïdouks, pendant le Subatlantique, les taux des conifères abaissent fortement, au-dessous de 10%, à partir de 100 cm de profondeur vers la surface.

Dans La Grotte de Veterani, à partir de 100 cm de profondeur vers la surface, le pollen de pin (*Pinus*) et de l'épicéa (*Picea*) abaissent aux valeurs souvent subunitaires, et seulement le sapin (*Abies*) se maintient jusqu'à la surface, avec des valeurs situées au-dessous de 10%. Si on parle au sujet des conifères, il est à prendre en considération l'aspect de l'emplacement géographique des sites recherchés. Les grottes du Defilé du Danube et surtout celles de la vallée de Cerna se trouvent en pleine zone montagneuse. Le pollen de conifères est transporté de hautes altitudes, des montagnes environnantes (les monts Cernei, Mehedinţi, Almăjului, Semenicului, Godeanu, etc.).

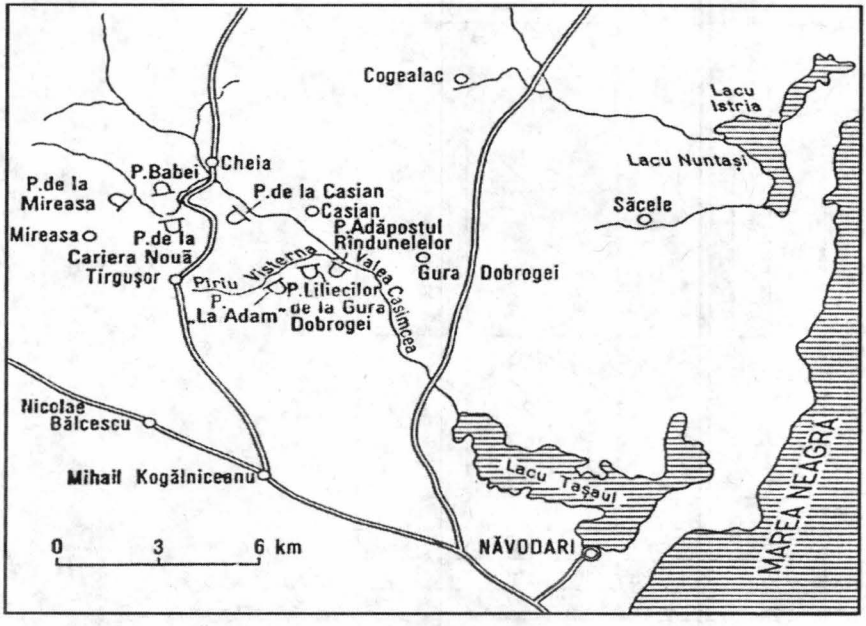

Fig.1. Localisation de la Grotte des Chauves-Souris (Peştera Liliecilor).

La Grotte des Chauves-Souris de Dobroudja centrale est située loin des influences des montagnes, le pollen de conifères devant parcourir presque 200 km en ligne aérienne, s'il provient de forêts des Carpates de Courbure. Ce n'est pas impossible, parce que le pollen de conifères est facilement transporté par les courants d'air, mais c'est normal que son incidence dans les spectres polliniques subatlantiques de Dobroudja soit très réduite. Lorsque la fréquence pollinique dans les préparations microscopiques et aussi très réduite, la probabilité que le pollen de conifères apparaisse dans ces spectres polliniques est presque nulle.

La situation du hêtre (*Fagus*) est semblable. Aujourd'hui on ne le trouve en Dobroudja que dans la réserve "La Forêt La Vallée des Hêtres", à côté de la localité Luncavița, en Dobroudja de nord. Dans les spectres polliniques de La Grotte des Chauves-Souris il apparaît sporadiquement et avec des valeurs très réduites. Pendant le Subatlantique, dans les spectres polliniques de la zone de plaine, le hêtre apparaît toujours subordonné aux chênaies mixtes (*Quercetum mixtum*), et même au charme (*Carpinus*), comme résultat du transport de son pollen à grande distance.

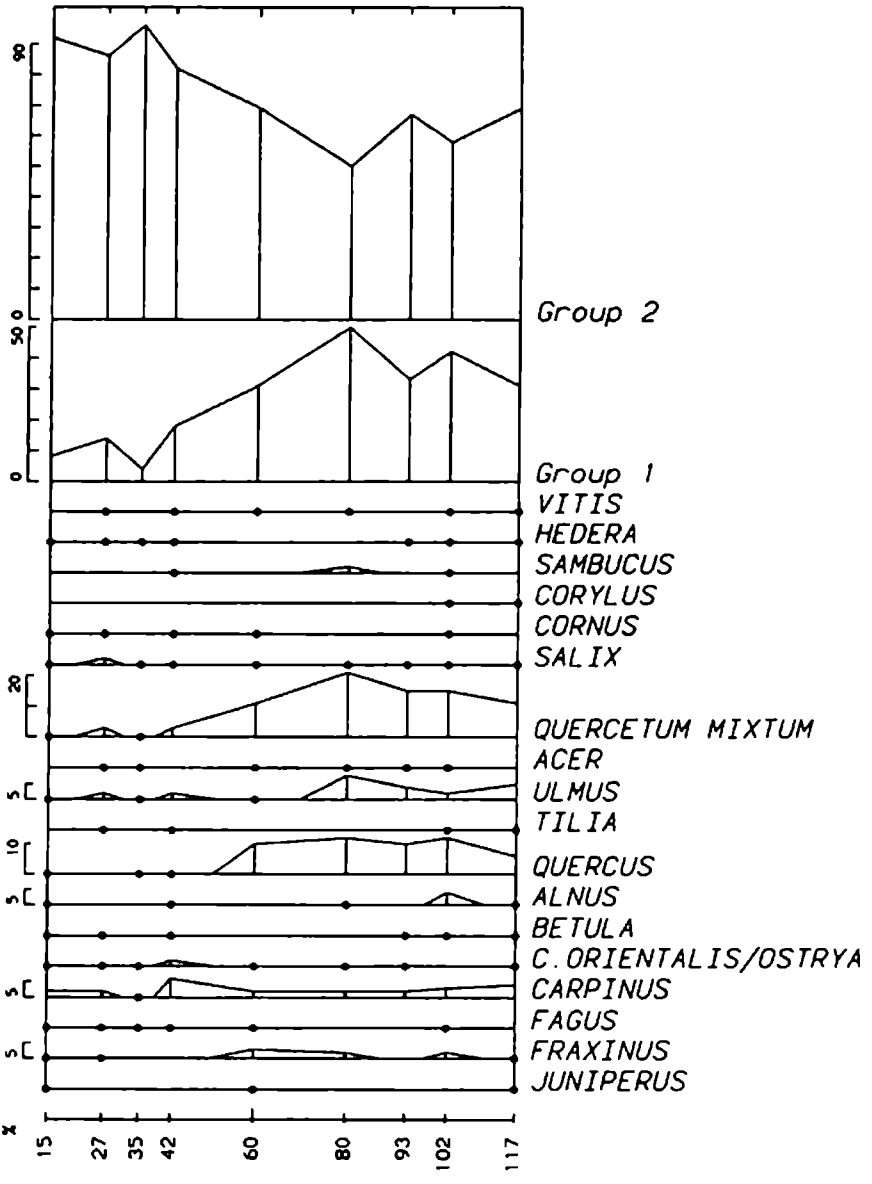

Fig. 2 Diagramme pollinique des arbres et des arbustes.

L'optimum climatique du hêtre, qui a favorisé son expansion a été assuré pendant le Subatlantique, plus frais et plus humid que le Subboréal qui l'a précédé. Pourtant, les nécessités écologiques de l'espèce ont déterminé la mise en place des forêts de hêtre dans l'étage montagneux. D'ici, leur pollen

a été transporté par les courants d'air aux altitudes plus basses, de plaine. Ça explique les différences des taux polliniques entre le diagramme de La Grotte des Chauves-Souris et celui de La Grotte d'Adam, située à une altitude plus élevée et dans une région montagneuse, où le maximum pollinique du hêtre pendant le Subatlantique a abouti à environ 20%.

Quant à la présence du genre *Carpinus* dans le diagramme pollinique de La Grotte des Chauves-Souris, son pollen appartient à deux espèces, le charme (*Carpinus betulus*) et le charme d'Orient (*Carpinus orientalis*). On trouve même aujourd'hui ces espèces en Dobroudja, dans les associations des forêts mésophiles de feuillus (l'unité F_{43}), respectivement des forêts xérothermes de feuillus (les unités G_{20} et G_{40}), conformément à "La végétation de la Roumanie" (1992). Le pollen de charme se maintient constamment dans les spectres polliniques (Tab. 1 et 2), avec les maximums du charme de 40% (respectivement 6,22% par rapport à la somme totale), et du charme d'Orient de 15% (respectivement 2,28%).

Le genre *Carpinus* ne montre pas des valeurs plus élevées, pendant le Subatlantique, ni dans la séquence de La Grotte d'Adam. Son apparition étrangement tardive, dans les spectres polliniques subboréales de cette séquence est due, probablement, à son "obscurcissement" par le pin, et à l'existence de certains niveaux, en base de la séquence, avec des fréquences polliniques extrêmement réduites, où même avec des hiatus de sédimentation, comme on l'a montré antérieurement.

Faute de données palynologiques similaires, sur le guano, en Dobroudja, nous ne pouvons pas savoir si les niveaux situés en base de la séquence de La Grotte des Chauves-Souris ne reflètent pas la fin du Subboréal. Seulement de datations C^{14} ultérieures, effectuées sur ce dépôt, pourraient préciser son âge.

En ce qui concerne les chênaies mixtes (*Quercetum mixtum*), elles ont enregistré dans cette séquence les valeurs les plus élevées parmi les A.P. À partir de la base de la séquence (117 cm de profondeur), les valeurs du chêne (*Quercus*= 30,27% par rapport à la Σ A.P., respectivement 6,72% par rapport à la Σ A.P.+ Σ N.A.P) et de l'orme (*Ulmus*= 23,85% par rapport à la Σ A.P., respectivement 5,29% par rapport à la Σ A.P.+ Σ N.A.P) surpassent les valeurs de tous les autres taxons ligneux. Les maximums absolus de séquence aboutissent à 55% chêne (respectivement 14,90% par rapport à la somme totale), 3,66% tilleul (respectivement 0,81%), 27,95% orme (respectivement 10,19%), 10% érable (respectivement 1,17%), et 77,50% chênaie mixte (26,26%), valeurs assez importantes, pourtant subordonnées

en permanence aux celles des herbacées (N.A.P.). Ces valeurs des chênaies mixtes sont semblables aux celles obtenues dans La Grotte d'Adam. Sporadiquement et avec des valeurs modestes, le pollen de frêne (*Fraxinus*), appartenent aux diverses espèces est associé au pollen des chênaies mixtes.

Même aujourd'hui, les associations de sylvosteppe avec du chêne pédonculiflore (*Quercus pedunculiflora*), du chêne pubescent (*Q. pubescens*) et de l'érable tatar (*Acer tataricum*), spécifiques à l'unité de végétation L_9, ou avec du chêne pédonculé et pédonculiflore (*Quercus robur, Q. pedunculiflora*), des frênes (*Fraxinus angustifolia, F. pallisae*) et des peupliers blancs, des trembles et des grisards (*Populus alba, P. tremula, P. canescens*), spécifiques à l'unité de végétation L_{12}. ("La végétation de la Roumanie", 1992), sont présentes dans la végétation de la Dobroudja de nord et centrale.

Ils sont présentes aussi les associations caractéristiques au forêts xérothermes de feuillus: des forêts ouest-pontiques de chêne pubescent (*Quercus pubescens*), de charme d'Orient (*Carpinus orientalis*) et de frêne (*Fraxinus ornus*) (l'unité G_{40}), et des forêts balkaniques de chêne chevelu (*Quercus cerris*) et de chêne pubescent (*Quercus pubescens*), avec du charme d'Orient (*Carpinus orientalis*), du frêne (*Fraxinus ornus*) et du sumac (*Cotinus coggyria*) (l'unité G_{20}). L'unité F_{43} comprend les associations caractéristiques aux forêts mésophiles de feuillus, moesique-ouest-pontiques, de chêne (*Quercus polycarpa, Q. dalechampii*), rouvre (*Q. petraea*), charme (Carpinus betulus), tilleul argenté (Tilia tomentosa), dans des ensembles avec des forêts xérothermes (conformemart à „La vegetation de la Roumanie", 1992).

La continuité de ces chênaies mixtes sur le territoire de la Dobroudja est attestée aussi par les analyses polliniques. Les courbes des chênaies mixtes sont visiblement marquées, dans les niveaux plus superficiels, par l'intervention anthropique, reflétée de même par la forte augmentation de certains taux de N.A.P. ("non arborum pollen"). Dans le diagramme pollinique des arbres et des arbustes de La Grotte des Chauves-Souris (Fig. 2), il apparaît en outre représenté, avec des taux modestes et sporadiquement, le pollen de bouleau (*Betula*), d'aune (*Alnus*), de saule (*Salix*), de cornouiller (*Cornus*), de sureau (*Sambucus*), de lierre (*Hedera*) et de vigne (*Vitis*), sans une valeur phytohistorique spéciale.

Le diagramme pollinique des herbacées de La Grotte des Chauves-Souris (Fig. 3) est caractérisé par une grande diversité, même si certains taxons apparaissent subunitaires ou sporadiquement. Le rapport A.P./N.A.P. (Fig. 2) reflète la subordination permanente du pollen des arbres et des

arbustes, aux celui des herbacées. Le maximum des taux de l'A.P. est seulement 36,47%, tandis que le minimum abaisse jusqu'à 3,34%. Ces valeurs qui abaissent surtout dans la moitié supérieure de la séquence, reflètent une activité anthropique de plus en plus intense vers nos jours.

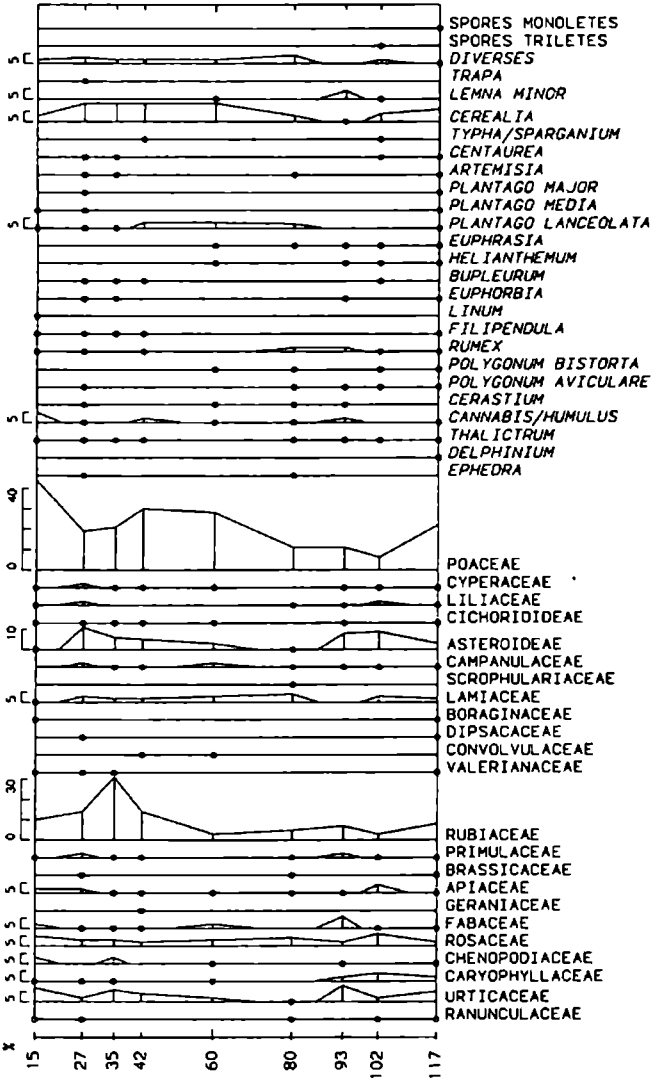

Figure 3. Diagramme pollinique des herbacées.

Les familles des herbacées avec la meilleure représentation des taux dans le diagramme sont les *Urticaceae, Caryophyllaceae, Rosaceae, Fabaceae, Apiaceae, Rubiaceae, Lamiaceae, Asteraceae* et *Poaceae*, et les taxons les plus significatifs sont *Cannabis type, Rumex, Plantago lanceolata* et *Cerealia tip*, qui suggèrent clairement le procès d'anthropisation. Parmi les herbacées, le maximum absolut des pourcentages appartient à la famille de *Poaceae*, qui dans le niveau de 15 cm a marquée 580,64% par rapport à la Σ A.P., respectivement 44,44% par rapport à la ΣA.P.+ N.A.P.

V. Conclusions

Les données palynologiques tardi- et postglaciaires obtenu dans le sud de la Roumanie sont insuffisantes et dépourvues de datations C^{14}. Au parcours de sept décennies d'études palynologiques effectués en Roumanie, seulement 30 travaux scientifiques ont envisagé dépôts où sédiments tardiglaciaires et holoccnes du sud, sud-est ou sud-ouest de la Roumanie. Ce nombre est très réduit par rapport aux études qui ont envisagé les Carpates où la Transylvanie.

On se connaît assez exactement la succession des associations sporo-polliniques tardiglaciaires et holocènes, dans les stations de Roumanie dont on dispose de datations C^{14}. Ces datations constituent des repères qui soutiennent la chronologie de ces séquences et font les comparaisons plus croyables. De cette analyse on peut tirer plusieurs conclusions intéressantes, concernant tant l'évolution d'ensemble de la végétation de la Roumanie en contexte européen, que l'évolution de la végétation du sud de la Roumanie.

Dans ces régions sudiques, les feuillus thermophiles se font l'apparition beaucoup plus vite que dans le reste de la Roumanie. Les analyses palynologiques effectuées dans ces régions semblent indiquer l'existence des refuges glaciaires pour les éléments de la chênaie mixte (chêne, orme, tilleul) au sud de la Roumanie.

Des données complémentaires pour l'histoire de la végétation du sud-est de la Roumanie sont apportées par l'analyse palynologique d'une séquence de guano, provenant de La Grotte des Chauves-Souris de "Gura Dobrogei", située en Dobroudja centrale. Les résultats palynologiques obtenus semblent indiquer le Subatlantique (possible le Subboréal tardif) pour la mise en place du dépôt de guano analysé.

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BEMERKUNGEN ÜBER DIE SAMENKEIMUNG UND DAS VEGETATIVE WACHSTUM BEI *ANGELICA ARCHANGELICA* L.

Martin KEUL*, Dana BATHORY*, Dan VÂRBAN**

Zusammenfassung. In vorliegender Arbeit wurden Untersuchungen über die Samenkeimung und das frühe vegetative Wachstum bei *Angelica archangelica* L. unter verschiedenen Bedingungen auf unterschiedlichen Substraten im Labor, im Gewächshaus und im Versuchsfeld durchgeführt. Unter Laborbedingungen unterblieb die Keimung innerhalb von 2 Monaten bei konstanten (23° C, 25° C şi 30° C) oder abwechselnden Temperaturen nach einer Kältevorbehandlung (1° C) bzw. nach einer Gibberellin-Vorbehandlung. Nur unter Licht wurde eine geringe Keimrate von unter 10% festgestellt. Nach Aussaat in Gartenerde dauerte das Erscheinen der Pflänzchen im Gewächshaus 30-45 Tage und erreichte im Mittel etwa 12%, bei den in Blumentöpfen im Freien gekeimten Samen betrug das Erscheinen der Keimlinge nach 2 Monaten etwa 50%. Der Wachstumsrhythmus der Keimpflanzen verläuft in den frühen Wachstumsphasen je nach den gegebenen Bedingungen sehr langsam (um 1 mm/Tag), wird jedoch mit dem Übergang in die aktive Wachstumsphase nach Ausbildung des 2. und der nachfolgenden Blätter und mit der Entwicklung des Wurzelsystems und des Photosynthese-Apparates, insbesonders bei den unmittelbar auf dem Versuchsfeld entwickelten Pflanzen, bis auf etwa 4 mm/Tag am Ende der Beobachtungszeit beschleunigt.
Stichwörter: Engelwurz, Samenkeimung, vegetatives Wachstum

Rezumat. Observaţii asupra germinaţiei seminţelor şi creşterii vegetative la *Angelica archangelica* L. Cercetările au urmărit germinţia seminţelor şi creşterea vegetativă în fazele timpurii de dezvoltare la *Angelica archangelica* L. pe diferite substrate şi în diferite condiţii de laborator, în seră şi în câmp experimental. În condiţii de laborator, seminţele nu au germinat în decurs de 2 luni la temperaturi constante ((23° C, 25° C şi 30° C) sau alternante după un pretratament la temperaturi scăzute ((4° C) sau cu giberelină. Un procent scăzut de germinaţie de sub 10% s-a înregistrat doar în condiţii de iluminare. La însămânţare în sol de grădină, în condiţii de seră, răsărirea plantulelor realizează un procent de cca. 12% după un intercal de 2 luni. Variantele efectuate în ghivece pe sol şi amplasate în

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condiţii naturale răsărirea a fost de cca. 50% după 2 luni. În primele faze de dezvoltare cotiledonală, ritmul de creştere al plantulelor a fost deosebit de lent (sub 1mm/zi), care se accelerează însă cu formarea celei de a doua frunze odată cu dezvoltarea sistemului radicular şi a aparatului fotosintetic până la 4 mm/zi în ultimul interval de măsurare.
Cuvinte cheie: angelică, germinaţia seminţelor, creştere vegetativă

Einleitung

In den letzten Jahrzehnten besteht ein weltweit zunehmendes Interesse für die Verwertung von Heil- und Gewürzpflanzen (Laza und Rácz, 1975; Crăciun und Mitarb., 1977; Mihalea, 1986, Muntean, 1990) und ihres mannigfaltigen Wirk- und Aromastoffgehaltes (Ciulei und Mitarb., 1993) in der Phytotherapie als heute anerkanntes Naturheilverfahren (Reichling, 1994) und als Rohstoffe zur Herstellung hochwirksamer Arzneimittel in der pharmazeutischen Industrie (Duke, 1993). Nach Farnworth und Billigs (1977, zitiert nach Duke, 1993) enthalten etwa 25% der zugelassenen Arneimittel wenigstens eine therapeutisch wirksame Komponente pflanzlicher Herkunft, wovon etwa 10% reine Extrakte darstellen.

Zur Deckung der steigenden Bedürfnisse an Heilpflanzen ist das Einsammeln aus der spontanen Flora heute nicht mehr ausreichend und aus verschiedenen Gründen auch nicht mehr zu vertreten, so dass Arzneipflanzen in kontrollierten Kulturen angebaut werden sollten. Denn einerseits haben Wildpflanzen eine schwankende quantitative und qualitative Zusammensetzung und therapeutische Wirksamkeit, die die erforderliche Standardisierung der Qualität der Phytopharmaka erschwert (Reichling, 1994), andererseits sind in letzter Zeit viele wertvolle spontane Heil- und Aromapflanzen durch anthropische Einflüsse, wie zunehmende Zerstörung und starke Umweltverschmutzung ihrer Habitate, und nicht zuletzt auch durch ihr über lange Zeiträume erfolgtes exzessives Einsammeln in ihrer Existenz bedroht und wurden deshalb in vielen Ländern Europas in Rote Listen als seltene und bedrohte Arten aufgenommen oder werden durch besondere nationale Gesetze geschützt (Rácz und Rácz, 1975; McNeely und Thorsele, 1991; Boşcaiu und Mitarb., 1994; Dihoru und Dihoru, 1994; Bundesamt f. Naturschutz, Bonn-Bad Godesberg, 1996).

Für den kontrollierten Anbau von Wildpflanzen sind genaue Kenntnisse ihrer ökologischen Ansprüche und biologischen Merkmale erforderlich, die in manchen Fällen noch weitgehend lückenhaft sind (Meyer-Berge, 1991). Das trifft z. B. auf die geschätzte Gemüse-, Heil- und

Gewürzpflanze *Angelica archangelica* L. zu, die in Westeuropa seit dem 16. Jh. angebaut wird (Heeger, 1956). Wiederholte Ansätze zum Anbau der Engelwurz oder Angelika hat es in den letzten Jahrzehnten auch in Rumänien gegeben (Zitti und Mitarb., 1958; Laza und Heltmann, 1970; Lăzurca, 1995). Trotz einiger eingehender physiologischer Untersuchungen (Taylor, 1949; Bomme und Mitarb., 1982; Ojala, 1985, 1986; Cseresnyes und Băleanu, 1986) sind viele Aspekte hinsichtlich der Biologie der Samenkeimung, der Wachstums- und Entwicklungsprozesse der Engelwurz noch nicht zufriedenstellend geklärt (Păun und Mitarb., 1986, 1988).

Die vorliegende Arbeit befasst sich mit einigen Aspekten zur Problematik der Samenkeimung und des vegetativen Wachstums bei *Angelica archangelica* L. unter Laborbedingungen, im Gewächshaus und auf dem Versuchsfeld.

Material und Arbeitsmethoden

Die „Samen" (eigentlich Achänen) von *Angelica archangelica* L. stammen aus Kulturen (Garten der pharmazeutischen Fakultät und Versuchsfeld der Universität für Landwirtschaft, Cluj-Napoca, Ernten 2002). Die Samen wurden nach der Ernte bei Zimmertemperatur etwa 2 Wochen hindurch zur Trocknung auf Papierunterlage ausgebreitet.

Keimversuche wurden in den Monaten März-Mai unter verschiedenen Laborbedingungen in mehreren Varianten durchgeführt. In einer der Versuchsvarianten wurden die Samen in Linhardt- und Petri-Schalen mit destilliertem Wasser oder auf Filterpapierunterlagen, ohne oder nach einer Vorbehandlung mit einer Gibberellinsäure-Lösung (GA_3 1mg/l) für die Dauer von 24 und 48 Stunden zur Keimung angesetzt. In einer anderen Versuchsvariante wurden luftgetrocknete oder mit destilliertem Wasser 24 Stunden vorgequollene Samen abwechselnd niederen (4° C) und höheren ((23° C, 25° C şi 30° C) Temperaturen ausgesetzt und anschließend im Thermostaten bei 25° C im Dunkeln oder bei Zimmertemperatur (20-22° C) unter Licht (Fluoreszenzlampen, Quantenstromdichte 100 μmoli/m²/s) ausgesät.

Für die Keimversuche auf einer Mischung aus Gartenerde und gewaschenem Fluss-Sand (1:1) als Substrat wurden Blumentöpfe verwendet und die Samen in etwa 1-2 cm Tiefe ausgesät. Die Keimung erfolgte entweder unter kontrollierten Laborbedingungen bei Zimmertemperatur (20-22° C)

oder im Freien unter Plastfolien bei normalem diurnalem Temperaturwechsel im Frühjahr (mit gelegentlich niederen nächtlichen Minustemperaturen von -3-5° C im April und Tageshöchsttemperaturen um 10-15 °C Anfang Mai bis 25°C um die Monatsmitte). Die Keimergebnisse wurden in Abständen von 5-10 Tagen beobachtet und Wachstumsmessungen durchgeführt. Während der Versuche wurden die Ansätze regelmäßig mit Wasser versorgt.

Bei den Versuchen im Gewächshaus wurden je 3-4 Samen in 1-2 cm Tiefe in Gartenerde in Plastbecher ausgesät. Keimung und Keimlingswachstum erfolgten unter diffusen Lichtverhältnissen und Gewächshaustemperaturen (Tageshöchsttemperaturen um 15-18° C). Die Auszählung der gekeimten Samen (das Erscheinen der Kotyledonen) wurde periodisch an 380 Ansätzen durchgeführt und das Wachstum an 50 Keimlingen durch Längenmessungen der Kotyledonen, des 1. und 2. Blattes verfolgt. Nach Ausbildung des 2. Blattes wurden die Pflänzchen in Abständen von 20-30 cm ins Versuchsfeld auf im Herbst mit Stalldung gedüngte Gartenerde verpflanzt. Außerdem wurden Samen im Frühjahr unmittelbar auf dem Versuchsfeld ausgesät.

Die Ergebnisse der Keimversuche und der durchgeführten Wachstumsmessungen wurden statistisch verarbeitet.

Ergebnisse und Diskussion

Die Engelwurz *Angelica archangelica* L. ist eine monokarpische, 2-3jährige, sehr robuste, 1-1,5 m (oft bis 2 m) hohe krautige Pflanze mit dickem, verweigtem Wurzelstock, die im ersten Jahr nur basale Blätter bildet und im 2. Jahr aus dem Wurzelstock einen hohen, dicken und verzweigten Blütenstängel mit grünlich-weißen Doldenblüten entwickelt (Flora R.P.R, Bd. VI, 1958). Die eurasiatische Art hat eine kurze Vegetationsperiode und ist von Sibirien bis Island in Zonen mit jährlichen Niederschlagsmengen von 500-1300 mm und Bodentemperaturen zwischen 5 und 19° C (Simon und Mitarb., 1984) verbreitet. Die in Rumänien unter Naturschutz stehende Engelwurz bevorzugt feuchte Standorte an Bächen, Mooren und Waldrändern in der montan-subalpinen Stufe auf nährstoffreichen, illuvialen Böden mit einem pH von 4,1-7,3 und Bodentemperaturen von 5-15° C (Păun, 1986). Für die in Europa verbreiteten *Angelica archangelica*-Populationen wurde einheitlich eine Chromosomenzahl von 2n=22 (Laza und Racz, 1975; Ojala,

1986) bestimmt. Durch ihre besonders in Wurzeln und Früchten vorkommenden reichen Gehalte an verschiedenen Inhaltsstoffen, insbesondere an wertvollen ätherischen Ölen, Furanocoumarinen, Coumarinen u.a. (Zobel und Brown, 1991; Ojala, 2001; Ciulei und Mitarb, 1993; Lăzurca, 1995), wird die Pflanze als Gemüse-, Heil- und Gewürzpflanze sehr geschätzt. Die Vermehrung der Pflanze erfolgt nur durch Samen, weshalb sich die meisten Autoren mit der Problematik der Samenkeimung und Samenlagerung befasst haben (Ojala, 1985).

Die in vorliegender Arbeit unter Laborbedingungen bei konstanten (23°C, 25°C şi 30°C) oder abwechselnden Temperaturen (Kältevorbehandlung trockener oder vorgeqollenen Samen bei 4°C für die Dauer von 7 Tagen und anschließender Keimtemperatur von 25 ° C), mit oder ohne Gibberellin-Vorbehandlung, im Licht oder im Dunkeln angesetzten Keimversuche ergaben meist negative Ergebnisse, wobei die Samen zwar zur Quellungsphase übergingen, aber innerhalb einer relativ langen Zeitspanne von etwa 2 Monaten keine Keimung zeigten und auf destilliertem Wasser und besonders auf Filterpapier-Unterlage einen betonten Pilzbefall aufwiesen. Nur unter Licht war eine geringe Keimrate von unter 10 % bei einigen der Ansätze festzustellen.

Diese Ergebnisse zeigen, dass die Keimprozesse bei der Engelwurz nach relativ langen Dauern induziert werden, wobei unter Laborbedingungen ein starker kryptogamischer Befall begünstigt wird, der wahrscheinlich zur Schädigung der Embryonen führt (Ojala, 1985; Lazurca, 1995). Bei den in Gartenerde durchgeführten Keimversuchen war der Pilzbefall nicht so stark ausgeprägt.

Im Falle der im Gewächshaus in Gartenerde durchgeführten Keimversuche erfolgte das Erscheinen der Kotyledonen mit großen Schwankungen zwischen den einzelnen Ansätzen nach etwa 35-40 Tagen und betrug im Mittel etwa 12%.

Bei den im Freien in Blumentöpfe in eine Mischung aus Gartenerde und Fluss-Sand (1:1) ausgesäten Samen dauerte das Erscheinen der ersten Kotyledonen unter normalen Frühjahrstemperaturschwankungen (gelegentliche nächtliche Temperaturen von bis zu -3-5° C im April und bei Tageshöchsttemperaturen um 10-15° C Anfang Mai bis 25°C um die Monatsmitte) etwa 45 Tage und erreichte innerhalb mehrerer Wochen schließlich Werte von ca. 50%. Das Keimlingswachstum im Kotyledonalstadium (graphisch nicht dargestellt) erreichte einen Zuwachs

von 15,67 ± 1,15 mm nach 6 Tagen und nach dem Erscheinen der ersten Blätter einen Zuwachs von 17,83 ± 3,92 mm nach 14 Tagen bzw. von 27 ±9,24 nach 21 Tagen entsprechend mittleren täglichen Zuwachsraten von 0,27 bzw. 1,31 mm. Daraus ist ersichtlich, dass die Wachstumsgeschwindigkeit in der ersten Entwicklungsphase sehr langsam verläuft und nach der Ausbildung der ersten Blätter zunehmend beschleunigt wird.

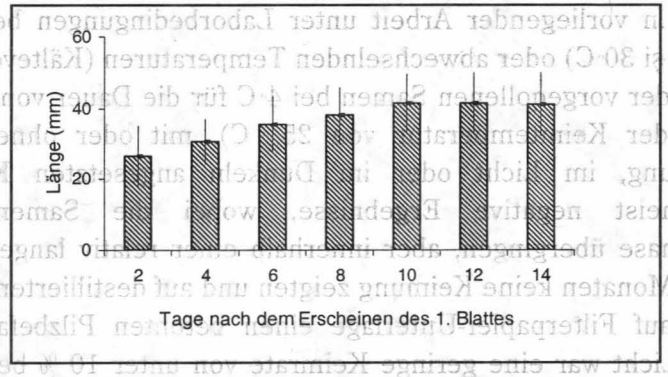

Abb. 1. Wachstumsdynamik des 1. Blattes bei Angelica archangelica L. (Blumentöpfe, Gartenerde+Sand, 1:1). Messungen im Abstand von 2 Tagen (M±D.S. n=10).

In Abb. 1 sind die Wachstumsabläufe der Keimpflanzen in Blumentöpfen im Freien nach Ausbildung des 1. Blattes dargestellt. Es wird festgestellt, dass die Keimpflanzen nach dem Erscheinen des ersten Blattes im Verlauf von 13 Tagen ein mittleres Wachstum von 1,13 mm/Tag aufweisen, wobei die anfängliche Wachstumsgeschwindigkeit von etwa 2 mm/Tag bis auf 0,1 mm/Tag während des allmählichen Absterbens des 1. Blattes und der Ausbildung des 2. Blattes abnimmt.

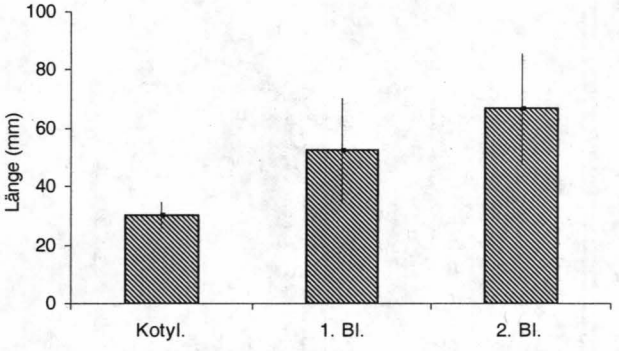

Abb. 2. Wachstum von Angelica archangelica L.-Keimpflanzen im Gewächshaus (M±DS, n=50)

In Abb. 2 ist die Wachstumsdynamik der Keimpflanzen unter Gewächshausbedingungen dargestellt. Unmittelbar nach ihrem Erscheinen zeigen die Pflänzchen im Kotyledonalstadium ein sehr langsames Wachstums von etwa 1 mm/Tag, das mit der Ausbildung des 1. Blattes bis auf etwa 4 mm/Tag zunimmt.

Ein Vergleich des Wachstums der unter verschiedenen Bedingungen kultivierten Angelica-Pflänzchen zeigt, dass die Keimpflanzen im Freien zwar eine etwas höhere Wachstumsgeschwindigkeit (1,13 mm/Tag) gegenüber den im Gewächshaus angezogenen Pflänzchen (unter 1 mm/Tag) aufweisen, aber allgemein zarter ausgebildet sind. Wahrscheinlich beruhen diese Differenzen auf homogeneren Temperaturbedingungen im Gewächhaus und auf unterschiedlichen Nährstoffangeboten der verwendeten Gartenerden als Substrate.

Bei den aus dem Gewächshaus ins Versuchsfeld umgesetzten Pflanzen (Abb. 3) beträgt die Wachstumszunahme innerhalb einer Zeitspanne von 13 Tagen 1,39 cm, entsprechend einer mittleren Wachstumsgeschwindigkeit von 1,07 mm/Tag.

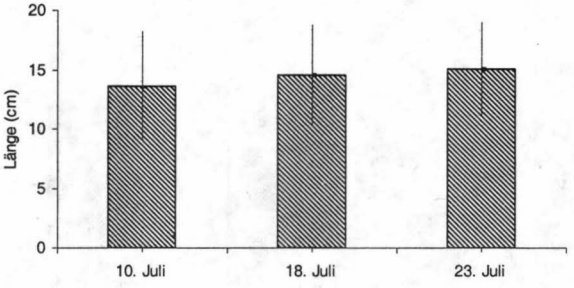

Abb. 3. Das Wachstum von Angelica archangelica L. nach Verpflanzung aus dem Gewächshaus ins Versuchsfeld (M±D.S. n=30)

In Abb. 4 ist das Wachstum bei den nach unmittelbarer Aussaat ins Versuchsfeld entwickelten Pflanzen dargestellt. In diesem Fall wurden im Vergleich zu den aus dem Gewächshaus ins Versuchsfeld umgesetzten Pflanzen in derselben Periode die höchsten Wachstumswerte mit mittleren täglichen Zuwachsraten von 4,04 mm/Tag erzielt.

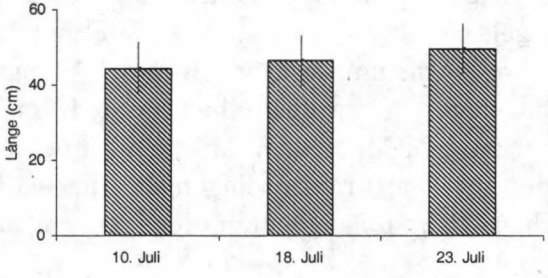

Abb. 4. Wachstumsverlauf der Blätter von Angelica archangelica L. nach Aussaat und Entwicklung der Pflanzen unmittelbar auf dem Versuchsfeld (M±D.S. n=30)

Die vorliegenden Untersuchungen über die Samenkeimung bei *Angelica archangelica* bestätigen die Befunde anderer Autoren (Ojala, 1985; Lăzurca, 1995), wonach die Einleitung der Keimprozesse länger dauert als bei anderen kultivierten Heilpflanzen und von der Herkunft, dem Samenalter, den Lagerbedingungen, dem Substrat und den Licht- und Temperaturbedingungen im Laufe der Keimung abhängig ist. Unter unseren Versuchsbedingungen mit Samen aus dem Vorjahr wurden auf destilliertem

Wasser nur unter Licht schwache Keimraten von unter 10% festgestellt, wobei eine GA-Vorbehandlung der Samen die Lichtwirkung nicht ersetzen kann (Ojala, 1985). Bei den im Gewächshaus oder im Freien in 1-2 cm Tiefe in den Boden ausgesäten Samen betrug die Keimrate nach einer Keimdauer von 30 bis 45 Tagen etwa 12 bzw. 50%.

Auf Grund der hier durchgeführten Keimversuche können keine näheren Erklärungen für diese erheblichen Variationen der Keimrate der Samen derselben Herkunft bei gleichem Alter unter denselben Aufbewahrungsbedingungen gemacht werden. Die Keimfähigkeit und die Keimrate werden von vielen Faktoren beeinflusst, u. a. durch die Größe der Früchte und ihrer Herkunft von Dolden verschiedener Ordnung (Ojala, 1986), das Reifestadium und die Lebensfähigkeit der Embryonen (Robinson, 1954, zitiert nach Ojala, 1985) oder durch die Induktion der sekundären Dormanz im Dunkeln, die durch Stratifikation und Licht gebrochen werden kann (Kivilaan, 1975). Bei den von uns im Freien angesetzten Keimversuchen könnte die festgestellte höhere Keimrate von etwa 50% eventuell durch diese Stratifikationswirkung niederer Temperaturen während der Keimung erklärt werden.

Die physiologischen Grundlagen der Samenkeimung bei *Angelica archangelica* sind trotz einiger eingehender Untersuchungen (Taylor, 1949; Bomme und Mitarb., 1982; Ojala, 1985; Lăzurca, 1995) noch nicht endgültig geklärt. Es ist allgemein bekannt, dass die Keimfähigkeit und die Lebensdauer der *Angelica*-Samen nach der Reife rasch abnehmen (Coiciu und Racz, 1962; Păun und Mitarb., 1986), weshalb die meisten Autoren empfehlen, die optimale Keimfähigkeit nach der Ernte der Samen durch Aussaat noch im Herbst desselben Jahres vor dem Eintreten der Keimruhe zu nutzen (Mihalea, 1986; Păun und Mitarb., 1986, 1988; Bayerische Landesanstalt für Bodenkultur und Pflanzenbau, 2001). Bei entsprechend kühler Lagerung der Samen unter Luftabschluss bleibt die Keimfähigkeit der Samen jedoch einige Monate ohne Verlust erhalten (Taylor, 1949; Bomme und Mitarb., 1982). Bei älterem Samengut kann die Keimung durch eine Kältevorbehandlung zur Brechung der Samenruhe vor der Aussaat induziert werden (Bayerische Landesanstalt für Bodenkultur und Pflanzenbau, 2001). Für die günstigste Wirkung einer Stratifikation bei niederen Temperaturen (4-10° C) werden von verschiedenen Autoren unterschiedliche Stratifikationsdauern angegeben, die von 6 Stunden (Cseresnyes und Băleanu, 1986), über 1-4 Wochen (Taylor, 1949; Lăzurca, 1995) bis zu

maximal 14 Wochen (Ojala, 1985) reichen. Allerdings scheint die günstigste Dauer der Stratifikation vom Genotyp (Ojala, 1985) und den Temperaturbedingungen während der Reife der Samen (Barton, 1965) und anderen Faktoren abzuhängen.

Zur Klärung einiger Fragen der Samenkeimung bei *Angelica archangelica* wurden von uns weitere Keimversuche mit Samen der Ernte 2003 von Wild- und kultivierten Pflanzen auf unterschiedlichen Keimsubstraten (destilliertes Wasser, Gartenerde, Bodenextrakte verschiedener Konzentration und auf Bodenrückstand), unter Dauerlicht oder im Dunkeln, ohne oder nach einer Kühllagerung trockener bzw. vorgeqollener Samen (Stratifikation) für die Dauer von 2 Wochen durchgeführt (Keul und Mitarb., 2004). Die erzielten Ergebnisse konnten zeigen, dass innerhalb von 5-7 Monaten nach der Ernte relativ hohe Keimwerte von 50-75% auf allen Substraten auch ohne Stratifikation erzielt werden, wenn die Samen auf dem Substrat unter Dauerlicht keimen. Aus diesen Ergebnissen scheint die besondere Bedeutung von Dauerlicht für die Induktion der Samenkeimung bei *Angelica archangelica* hervorzugehen. Eine Stratifikation fördert die Samenkeimung, ist aber wahrscheinlich erst nach dem Eintreten der Samenruhe notwendig, denn hohe Keimraten wurden in unseren Versuche innerhalb einer Lagerdauer von 5-7 Monaten nach der Ernte auch nach einer Vorquellung der Samen bei Zimmertemperatur ohne Kältevorbehandlung unter Dauerbelichtung induziert.

Nach dem Erscheinen der Keimblätter erfolgt das Wachstum der Keimlinge bis zur Ausbildung des Wurzelsystems sehr langsam (Lăzurca, 1995). Die Wachstumsintensität ist im Kotyledonalstadium von unter 1 mm/Tag am kleinsten, nimmt dann aber nach dem Erscheinen des 1. und insbesondere des 2. und der nachfolgenden Blätter mit Ausbildung des Photosynthese-Apparates bis auf ca. 4 mm/Tag am letzten Tag der Messung zu. Diese Angaben zur Wachstumsdynamik in den aufeinanderfolgenden Entwicklungsphasen (Kotyledonalphase, Phase der Blattbildung) haben unter den jeweils gegebenen Bedingungen im Labor, im Gewächshaus und im Freien oder im Versuchsfeld nur orientierende Werte, da zu erwarten ist, dass die unterschiedlichen Temperatur- und Lichtbedingungen, sowie das Nähstoffangebot auf verschiedenen Substraten in zeitlich etwas gegeneinader verschobenen Perioden einen unterschiedlichen Einfluss auf die Wachstumsprozesse ausüben. Bisher fehlen eingehende Untersuchungen über den Einfluss von Licht, Temperatur und anderen Faktoren auf

Wachstum und Entwicklung, sowie auf die Synthese und die Anreicherung von Inhaltsstoffen bei der Engelwurz (Păun und Mitarb., 1986). Im Verbreitungsgebiet der Engelwurz scheinen die Licht- und Temperaturbedingungen optimale Wachstums- und Entwicklungsprozesse zu gewährleisten, so dass genaue Kenntnisse über die ökologischen Ansprüche dieser Art auch für den Anbau dieser Heil- und Gewürzpflanze von besonderer praktischer Bedeutung sind.

Schlussfolgerungen

Die in vorliegender Arbeit unter Laborbedingungen bei konstanten (23°C, 25°C şi 30°C) oder abwechselnden Temperaturen (Kältevorbehandlung trockener oder vorgeqollenen Samen bei 4°C für die Dauer von 7 Tagen und anschließender Keimtemperatur von 25 °C), mit oder ohne Gibberellin-Vorbehandlung, im Licht oder im Dunkeln auf verschiedenen Substraten angesetzten Keimversuche ergaben meist negative Ergebnisse. Nur unter Belichtung wurde eine Keimrate von unter 10% festgestellt.

Bei den in Gartenerde im Gewächshaus ausgesäten Samen dauerte das Erscheinen der Pflänzchen 30-45 Tage und erreichte im Mittel etwa 12%, bei den in Blumentöpfen im Freien gekeimten Samen betrug das Erscheinen der Keimlinge nach cca. 2 Monaten etwa 50%.

Die durchgeführten Wachstumsmessungen zeigen, dass der Wachstumsrhythmus der Keimpflanzen in den frühen Wachstumsphasen je nach den gegebenen Bedingungen sehr langsam verläuft (um 1 mm/Tag). Mit dem Übergang in die aktive Wachstumsphase nach Ausbildung des 2. und der nachfolgenden Blätter entsprechend der Entwicklung des Wurzelsystems und des Photosynthese-Apparates wird das Wachstum im Zeitraum der durchgeführten Beobachtungen bis auf etwa 4 mm/Tag beschleunigt.

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PRELIMINARY RESEARCH ON THE VIOLA GENUS IN THE CĂPĂŢÂNII MOUNTAINS

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Introduction

The territory under research is located in the southern catena of the Carpathians between the Jiu and the Olt Rivers, that is the mountains region known as the Căpăţânii Mountains. The research on the field was carried out from 1997 to 2003, with planned itineraries. For the identification of the taxa, we have used the Romanian Flora, vol. III, and Flora Europaea, vol. I. Regarding the nomenclature, we have adopted the classified list solutions which are considered correct, according to the International Code of Botanic Nomenclature.

Results and discussions

The **VIOLA** genus is part of the **VIOLACEAE** Family and is represented, in the European Flora, by 123 species, among wich 24 species are to be found in the Romanian Flora. Following the research carried out in the Căpăţânii Mountains, we have identified 13 species of the Viola Genus, to which we can add 2 ssp and 3 forms.

1. ***Viola alba*** Besser; (*Viola virescens* Jord.) – White Violets. Querco-Fagetea, Alliarion; 2n=20; (white violets). They are frequently met in the lower mountain subfloor, in the forests, forest skirts, and bushes. In the territory under research, this species was found in the Luncavăţ, Olăneşti and Bistriţa-Vâlcea Basins. In the Luncavăţ Basin, near the town of Horezu, two forms of this species were identified: f. ***variegata*** and ***subcolina*** Querco-Fagetea, Alliarion H, Med-Ec; 2n=20; U,T$_{4}$R.

2. ***V. alpina*** Jacq. Alpine Violets. Rarely met in the alpine and subalpine floor. They were mentioned in the Buila Mountain – Vioreanu Peak

and in Stâna Comarniei by Buia and Păun, in 1956. H, Papavero-Thimion pulcherrimae, Caricion curvulae; rare; 2n=22; Alp-Carp; U.T.R.

3. ***V. arvensis*** Murray – Filed Violets. They vegetate in cultivated and ruderal areas, meadows and bushes. It is found in the Horezu Depression. Aperetalia; Th, Eua; 2n=34, D; U.T.R.

4. ***V. biflora*** L.-Yellow Violets. They are frequently met in the mountain and alpine floor, in moisten places, on the stream banks. To be found in the Luncavăț Basin, in the Valleys of Polovrăgeni, Curpeni, Blaj, Căpățânii, Balota, in the Olteț, Cerna, Bistrița-Vâlcea, Olăneşti Basins, and the Buila Mountain. Car. Adenostyletali; H, Cp; 2n=12; D; U.T.R.

5. ***V. canina*** L. – Wild Violets. The are frequently met in the mountain floor, in the meadows and in the forests. The species was identified in the Bistrița-Vâlcea and Râmeți Basins, on Dobra Hillock. Car. Nardo-Galion, Molinion, Violion caninae, Sedo-Scleranthetlia, Nardo-Callunetea; H, Eua; 2n=40, P; U.T.R.

6. ***V. declinata*** Waldst. Et Kit. – Bird's nail. It is frequently met in the upper mountain floor and the subalpine floor; in the Luncavăț Basin, Funicelul Saddle, Balota and Dârjala Mountains, Red Rocks, in the Buila Mountain, on Vioreanu and Vânturărița Peaks, in the Olăneşti and Bistrița-Vâlcea Basins. Potentillo-Nardion; H, Carp-B; 2n=20,26, P; U.T.R.

7. ***V. hirta*** L. – Car. Origanetalia; - Scentless Violets. They are frequently met in the lower mountain floor, in the forests, forest skirts, and bushes. Theyare to be found in the Luncavăț Basin and Horezu Depression. We also identifies an infrataxa of this species near the town of Horezu: f.fraterna. H, Eua; 2n=20; P; U.T.R.

8. ***V. odorata*** L. – Violets. They are frequently met in the lower mountain floor, in the forests, forests skirts, and bushes and riverside coppice. They were found in the Luncavăț, Olăneşti, Otăsău, Cerna and Bistrița-Vâlcea Basins; Querco-Fagetea, Aliarion, Prunetalia; H, Alt-Med; U.T.R.

9. ***V. reichenbachiana*** Jord. (*V.sylvestris* Lam. pro parte) - Wood Violet. It is frequently met in the mountain floor, in the forests. To be found in the Luncavăț, Olănești, Otăsău, Cerna and Bistrița-Vâlcea Basins. - Car. Fagetalia, Symphyto-Fgion; H, Eua; 2n=20, P; U.T.R..

10. ***V. riviniana*** Reichenb. - It is rarely met in the lower mountain floor, in the forests and forests skirts. It was identified in the Luncavăț and Otăsău basins, Costești Gorges, Horezu Valley. Fagetalia H, E; 2n=40,45, P; U.T.R.

11. ***V. rupestris*** F.W. Schmidt; (*Viola livida* Kit.). This species is rarely found in the territory under researche, vegetating on rocky coasts and detritus. It was identified by Buia and Păun in the Olteț basin. It is also found in the Luncavăț basin, near the locality of Vaideeni. Festucetalia vaginatea, Festucetali valesiacea, Car. Dicrano-Pinion; H, Circ; 2n=20; U.T.R..

12. ***V. suavis*** Bieb.; (*Viola cyanea* C. El., *Viola austriaca* A. Et j. Kern., *Viola pontica* W. Becker., *Viola ignobilis* G. Grin*). - Violets. They are frequently met in the lower mountain subfloor, in the forests and in the bushes. The species was identified in the Luncavăț Basin, at Urșani and Râmeți, in the Otăsău Basin. Aceri-Quercion; H, Eua, 2n=40; U.T.R.

13. ***V. tricolor*** L. S.l. - Three spotted brothers. This species is frequently found in the mountain floor, in the meadows and cultivated areas. One can see it in the Luncavăț Cerna, Olteț, Bistrița-Vâlcea, Olănești Basins, the Buila Mountain and at Bulzu. Molinio-Arrhenatheretea, Asplenio-Festucion, Seslerio-Festucion, Sedo-Scleranthetea, Th-H, Eua; 2n=26; D; U.T.R.

- ssp. ***tricolor***, it is frequently found in the lower mountain sublevel, in the meadows, and cultivated areas Th-H, Eua; U.T.R.

- ssp. ***subalpina*** Gaudin (*Viola saxatilis* F.W. Schmidt, *Viola tricolor* var.*luteola* Schur); It is rarely found in the upper mountain subfloor and the subalpine floor, in the meadows. Molinio - Arrhenatheretea, Asplenio - Festucion et Seslerio - Festucion, Sedo - Scleranthetea; TH, Th-H, Eua; 2n=26, D; U.T.R.

Conclusions

Following the botanical research carried out between 1997 and 2003 in the Căpățânii Mountains, there were identified 13 species, 2 subspecies and 3 forms of **Viola** genus. This genus is well represented in the Căpățânii Mountains, the 13 identified species representing 46,15% of the total number of species which vegetate in Romania. Some of them can be frequently met in the territory under research, while others are rarely met.

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AN ADDITION OF AL. BORZA'S ETHNOBOTANIC DICTIONARY

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Abstract: In this paper are enumerated 490 romanian names of plants which are't reported in the „Dicţionar etnobotanic" of Al. Borza (1968). Those names of plants are gathered by author from several localities and books. If we add these names which are already known (Al. Borza, 1968, C. Drăgulescu, 2003) we reach the conclusion that the romanian botanic vocabulary includes more than 16.000 phytonyms.
Key words: vernacular Romanian names of plants.

Rezumat: După o activitate etnobotanică de o viaţă Al. Borza a publicat în 1968 cel mai vast dicţionar etnobotanic românesc ce însumează 10.906 denumiri populare româneşti pentru 2.095 specii vegetale. În anul 2003 am editat un supliment la acest dicţionar ridicând numărul fitonimelor româneşti la 15.528 iar cel al plantelor numite la 2840. Între timp strângând din teren alte nume de plante şi alăturându-le celor găsite prin diverse lucrări botanice am înlocuit lista pe care o prezentăm în lucrarea de faţă. Sunt în continuare convins că poporul român a avut şi are în vocabularul său cel puţin 25.000 de fitonime, cele mai multe de origine latină, slavă traco-dacă dar şi maghiară, greacă şi neogreacă, franceză, turcă (inclusiv cumană şi pecenegă), germană (mai ales săsească), engleză, italiană, cu siguranţă şi celtă, scito-sarmată, pre-indoeuropeană ş.a.

Am inclus în această listă şi o serie de denumiri româneşti aparţinând unor specii exotice, denumiri selectate majoritatea din manuale. Ele circulă tot mai mult printre conaţionali şi aparţin mai ales unor arbori cu lemn preţios sau cu fructe comercializate şi în ţara noastră, unor plante ornamentale ori cu alte utilităţi. Aceste denumiri sunt, deci, livreşti, fără specificitate regională şi de aceea nu am mai precizat sursa bibliografică.

Denumirile ştiinţifice scrise cu litere groase sunt noi, necuprinse în cele două dicţionare amintite mai sus. Pentru ele am specificat autorii care le-au descris prima dată, pentru celelalte specii nu am mai citat autorii, aceştia putând fi găsiţi în Dicţionarul etnobotanic al lui Al. Borza.
Cuvinte cheie: Nume populare româneşti de plante.

After a lifetime etnobotanical activity Alexandru Borza published in 1968 the greatest Romanian ethnobotanical dictionary which brings together 10.906 Romanian popular denominations for 2095 vegetal species.

In 2003 I published a supplement to this dictionary increasing the Romanian phytonimes number to 15,528 and plants number to 2,840 species.

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In the meantime I made the list, that I present in this work, after I gatheride other plant names and adding them to name which I Found in various botanical studies.

I am further on convinced that the Romanian people had and has at least 25,000 phytonimes in its vocabulary, most of them of Latin, Slav, Thraco-Dacian origin, but also Hungarian, Greek and Neogreek, French, Turkish (Cumanian and Petcheneg origin included), German (especially Transylvanian Saxon), English, Italian origin, also Celtic, Scytho-Sarmatian, Preindo-European origin and so on.

I also included in this list several Romanian plant names which belong to some exotic species, most of them being selected from the manuals. They circulate more and more lately and belong mainly to some valuable trees or exotic fruits, to some ornamental plants or with other usefulnesses. So, these denominations are bookish, without regional specificity and that „why I didn't" mention the bibliographical sources.

The bold scientific namea are new, they are still not included in the above-mentioned dictionaries. That's why I mentioned the authors who described them for the first time, and for the other species I didn't mentioned the authors because they can be found in Al. Borza's „Dicționar etnobotanic".

Abies alba – Pieptenele lui Sân-Petru (2)
Achillea millefolium – Prâsnel (7), Şureşână (10)
Agropyron repens – Pipirău (7)
Agrostis canina – Cătuşă (Făgăraş)
Althaea rosea – Barba împărtului (Daia Română – Alba)
Ambrosia artemisiifolia – Ambrozie
Anemone nemorosa – Păsărele (Sălişte – Sibiu)
Anemone ranunculoides – Păşcuță (7)
Anredera baselloides (Kunth) Baill. – Telegraf (9)
Arctium lppa – Bruscan (10)
Aristolochia clematitis – Lingură (9), Ruşeță (5)
Artemisia absinthium – Bermet (10)
Artemisia dracunculus – Sagna calului (7)
Asarum europaeum – Copita măgarului (Mârşa-Sibiu), Copite (Făgăraş), Lingură (9)
Asparagus officinalis – Coada iepii (Sibiu)

Astrodaucus littoralis (Bieb.) Drude – Morcov de mare (Agigea)
Avena nuda L. - Ovăs golaş (15)
Avena sterilis – Ovăs sălbatic (15)
Bertholletia excelsa Humb. Et Bompl. – Nuc(ă) american(ă) (9)
Beta vulgaris – Gurguni (11), Pangică (11), Rudari (11), Săclă (7), Ţucuran (11)
Bidens cernua – Cârligătoare (Ardan – Bistriţa-Năsăud)
Bifora radians – Pucioagnă (Ardan – Bistriţa-Năsăud)
Brachychiton sp. – Arborele-butoi
Briza medie – Iarba miresii (Ardan – Bistriţa-Năsăud), Semincioara ierbii (15)
Bromus secalinus – Osic (Negreni-Olt)
Bryophyllum laxiflorum Baker – Mama cu copiii (Cluj-Napoca)
Buchloe dactyloides (Nutt.) Engelm. – Iarba bizonului
Bunias orientalis – Macareţ (8)
Calamagrostis epigeios – Firuţă dungată (6)
Calamus rotang L. – Trestie de mare (9), Rotang
Callistephus chinensis – Ruşi de toamnă (7)
Caltha laeta – Scrântitoare (5), Zlat (7)
Campanula rapunculus – Bănişori (9)
Cannabis sativa – (cânepa de sămânţă): durdălani, dârdălani, hăldani, hâldani (11)
Capsicum annuum: fitonime pentru ardei iute: doboş (11), piparcă usturoaie (11), pipearcă (11), poprici usturoi (11), puţoi (11), răcuşor (11), ţăpuran (11), ţăpurel (11), ţâr (11), ţâţă (11)
Carex arenaria – Pir roşu (3)
Carex brizoides L. – Târsău (Oaş)
Carex humilis – Rogoz mărunt (6), Rogoz pitic (6)
Carlina acaulis – Scaiul dracului de munte (2), Sita Ielelor (2)
Carum carvi – Carul câmpului (Lugoj)
Ceiba pentandra (L.) Gaertn. – (Arborele de) Capoc
Centaurea cyanus – Corabatică (9)
Centauria moschata – Peşmă (9)
Centaurea phrygia – Măseaua calului (Şirnea-Braşov)
Centaurea suaveolens – Peşmă (9)
Cerasus avium – soiuri: californe (11), cu grumaz (11), grase (11), iepureşti (11)
Chaiturus marrubiastrum – Poala Sfintei Marii (7), Talpa ursului (7)
Chelidonium majus – Buruiană crudă (Sibiu), Pleoscăriţă (7)

Chrysanthemum corymbosum – Ochiul boului a ferigei (7)
Cichorium intybus – Ciucoare (9)
Cinchona officinalis L. – China (3)
Cinchona pubescens Vahl – China roşie (3)
Circaea alpina – Perişor (9)
Citrus bergamia Risso et Poit. – Bergamotă (3)
Coccinia indica – Tătarcă (9)
Cola nitida (Vent.) Schott et Endl. şi sp. – Cola
Consolida regalis – Nemerişori (7)
Convolvulus arvensis – Cupa Maicii Domnului (2)
Corylus avellana – Amentul mascul: ciubucea (15), mătârlă (15)
Crataegus monogyna – Malaiel (10)
Crocus variegatus – Brânduşă albă (9), Brânduşă mică (9), Şofran vărgat (9)
Croton sp. – Croton
Cucumis melo – soiul Godină (9,14)
Cynodon dactylon – Iarba căţelei (Horezu – Vâlcea)
Cyperus papyrus L. – Papirus
Dalbergia latifolia Roxb. Ex DC. şi sp. – Palisandru
Daphne cneorum – Roşioare (8)
Datura stramonium ? Bolondicăi (Corund – Satu Mare), Borciu (7), buruiană înveninată (10), Faie (7)
Daucus carota – Moroc (7)
Ecbalium elaterium – Prăsitoare (Constanţa)
Echinochloa crus-galli – Mohor negru (9), Moreaţă (Popii-Slăviteşti-Teleorman)
Epilobium hirsutum – Răscoace (Sibiu)
Equisetum arvense – Barbă (Ardan-Bistriţa-Năsăud)
Equisetum palustre – Barbă (Ardan-Bistriţa-Năsăud)
Erodium cicutarium – Pliscul cocoşului (Ardan-Bistriţa-Năsăud)
Eryngium campestre – Salata dracului (Luduş-Mureş)
Eryngium planum – Samcă (7), Scai nerău (Corund şi Racova-Satu Mare)
Eupatorium canabinum – Cânepoală (9)
Festuca porcii Kack. – Păiuş arămiu (15)
Festuca pseudovina – Iarba oii (6)
Festuca rupicola – Fâşcă (6)
Festuca valesiaca Schleich – Fâşcă (6), Păiuş de câmpie (6)
Ganoderma lucidum – Lingura Maicii Domnului (9), Lingura Maicii Preceşte (9), urechiuşă (9)

Gentiana lutea – Ochimea (9)
Gleditschia triacanthos – Mucii babii (fructul) (Sibiu)
Garcinia mangostana L. –Mangustan
Guaiacum officinale L. – Guaiac (3)
Haematoxylum campechianum – Băcan negru (3)
Helleborus odorus – Strecurătoare (7)
Helleborus purpurascens – Bozută (9), Spânc (Corund-Satu Mare)
Hevea brasiliensis (Willd. ex A. Juss.) Mull. Arg. – Arbore de cauciuc
Hordeum hexastichon – Orz mucher(i) (9)
Hordeum vulgare – Soiurile Orz românesc (9), Orz moldovenesc (9)
Humulus lupulus – Comblău (Corund, Racova-Satu Mare)
Iles paraguayensis A. St. – Hill. – Mate
Illicium anisatum L. – Anason stelat (3)
Impatiens noli-tangere – Brei (10)
Impatiens sultani – Sporul casei
Indigofera tinctoria L. – Indigo
Inula salicina – Cioroinic (9)
Iris ruthenica – Custurice (Tălmaciu-Sibiu)
Juniperus communis – Tămâier (7)
Khaya sp. – Acaju
Juniperus communis – Tămâier (14)
Juniperus oxycedrus L. –Cad (3)
Krameria lappacea (Dombey) Burdet et B.B. – Ratania (3)
Laburnum anagyroides – Măgrin (10)
Lathyrus tuberosus ? – Sângele Voinei (2)
Leeythis zabucayo Aubl. – Nucul Paradisului
Lemna minor – Lintea raței (Ocna Sibiului)
Linum catharticum – Mărgeluță (9)
Liparis loeselii – Molișoare (9) nu Moşişoare ca în Borza
Lolium temulentum – Sălbatic (15)
Lunaria rediviva – Banul scârțarului (Bistrița-Vâlcea)
Luzula campestris – Pcita dracului (Năsăud)
Lychnis flos-cuculi –Opățel (6)
Lycium barbarum –Cătină de curte (Ocna Sibiului)
Lycopersicon esculentum: Borodici (11)
Lysimachia nummularia – Coardele la alea tari (5)

Malus domestica – soiuri: botane (11), buciumene (11), buicăne (11), dăneşti (11), de-ale aurului (11), de-ale untului (11), hâncâieşti (11), mălăieţe (11), murăşeneşti (11), posmogene (11), puhave (11), sămânţâi sau de sămânţă (11), scorţoboase (11), stănile (11), şiculane (11), şovarne (11), ţâţa-vacii (11), ţâţoase (11), ursăneşti (11), viorele (11)
Matthiola incana – Vioale în crăci (Avrig-Sibiu)
Mauritia vinifera – Palmier de vin
Melampyrum sp. – Grâu ţâganului (Ţichindeal-Sibiu)
Melandrium rubrum – Licurici (Tilişca-Sibiu)
Mentha aquatica – Minta lingorii (7)
Mercurialis perennis – Breiul cânelui (Veştem-Sibiu)
Mespilus germanica – Gorun (7), Scoruş nemţesc (9)
Metrosideros sp. – Arborele de fier
Metroxylon sagu Rottb. – Sago(tier) (3)
Monotropa hypopytis – Sparangă de brădet (7)
Myroxylon balsamum (L.) Harms – Arbore de balsam
Nicotiana tabacum – Buruiana leacului (2), Iarba dracului (2), Tămâia dracului (2)
Ochroma pyramidale (Cav. Ex Lam.) Urb. – Lemn de Balsa
Ocimum basilicum – Afinic (2)
Opuntia vulgaris – Nopal (9)
Orchis morio – Gheruţa cucului (Câlnic-Alba), Mugurucean (Ardan-Bistriţa-Năsăud)
Origanum vulgare – Busuioc(u) la ali tari (5), Şuvavâr (5)
Orobanche sp. – Sudoare (Ceru-Băcăinţi-Hunedoara)
Oxalis acetosella – Macrici iepuresc (Racova-Satu Mare)
Paeonia officinalis – Bujămac (10)
Panax ginseng C.A. Mey – Ginseng, Jenşen
Passiflora sp. – Floarea pătimirii (2)
Pastinaca sativa – Buruiana vântului (5)
Pennisetum purpureum Schumach. – Iarba elefanţilor
Persea americana Mill. – Avocado
Pharbitis purpurea – Haragică (2), Hărăgică (2), Telegraf (2), Zambele (2)
Phyteuma tetramerum şi sp. – Ghiara dracului (Sibiu)
Phyteuma wagneri – Spinuţă (9), Vătăjel (9)
Pimpinella anisum – Anij (1)
Pinus mugo – Brad rozmolin (10)

Piper cubeba – Cubeb (9)
Pistacia lentiscus L. – Arbore de mastic (3), Arbore de sacâz (3)
Pistacia terebinthus L. – Terebint (9)
Pistia stratiotes L. – Varză de apă
Plantago sp. – Cucuruz de minciună (Racova-Satu Mare), Paclagină (10)
Pogostemon patschouli – Paciulie (9)
Polygonatum sp. – Buiede cu boboloaşe roşii (10), Buedea vinii (10), Buedea vinului (10)
Polypodium vulgare – Niricică (5)
Portulaca grandiflora – Bună dimineaţa (Sibiu)
Potentilla sp. – Buruiana scrântului (10), Palma Maicii Domnului (2), Târâtoare (5)
Pringlea antiscorbutica – Varza marinarilor
Prunus domestica – Soiul: avrame (11), batoşe (11), boboloaşe (11), boateşe (11), cachii (11), cacopoua (11), ciorane (11), ciorăci (11), câineşti (11), cloţui (Băiculeşti-Argeş), cloşuţe (11), coadeşe (11), coadreşe (11), colduşe (11), cu gât (11), curcudele (11), dăgene (11), de uscat (11), drepte (11), dulci (11), foarteşe (11), gălbenioare (11), gogoneţe (11), marghite (11), mistrele (11), motrogane (11), motrune (11), moţate (11), murăşeneşti (11), narangii (11), negre (11), negre (11), nemţeşti (11), oarzăne (11), oltoane (11), pepenei (9), pişoalce (11), pârţâguşe (11), rânduri (11), sticloase (11), ştire (11), târnosive (11), zdronţ (11).
Prunus (?insititia) – Toltuş (14)
Psidium guajava L. – Guava
Pterocarpus indicus Willd. – Santal roşu (3)
Pterocarpus santalinum L.f. – Santal roşu (3)
Pulmonaria officinalis – Pulmon (Ardan-Bistriţa-Năsăud)
Pulsatilla sp. – Găgăţele (7)
Pyrus sativa – soiuri: acavile (11), de-ale fânului (11), de pârci (11), gălbioare (11), iepureşti (11), lucii (11), mânânţele (11), rocilate (11), roşii-gălbui (11), ursăneşti (11)
Quercus sp. – Gala: galeşă (7)
Quillaja saponaria Molina – Lemn de Panama (3)
Raphia farinifera (Gaertn.) Hyl. – Rafie
Raphia vinifera P. Beauv. – Rafie de vin
Rhamnus cathartica – Baţachină (9)

Rhamnus frangula – Bațachină (9), Buruiană de roșeață (10)
Rhododendron kotschyi – Ruja munților (9)
Rhus radicans L. – Sumac (3)
Rhus toxicodendron L. – Sumac veninos (3)
Ribes grossularia - ? Borșari (14); Fructul: ocroașă (14)
Ribes rubrum – Răbiză (14)
Robinia pseudacacia – Băgran (10), Begrin (10), Brăghin (10), Breagăn (10), Cârtac (9), Copaci (10)
Rosa canina și sp. – Oglej (14); Fructele: căcăzdări (Ardan-Bistrița-Năsăud)
Rubia tinctorum – Bațachină (9)
Rubus idaeus – Runcan (14)
Ruta graveolens – Pigan (4)
Saccharomyces cerevisiae – Țaică (14)
Salvia nutans L. – Jelitoare (Mândra-Sibiu)
Salvia officinalis ? – Alifacsă (7)
Sanguisorba officinalis – Crestățea (Tălmaciu-Sibiu), Cruciță (Orăștie)
Sanicula europaea – Omag (7)
Saponaria officinalis – Mărarul calului (7)
Sassafras albidum (Nutt.) Nees – Sasafras (3)
Scabiosa ochroleuca – Mușcata dracului (6)
Scolymus hispanicus L. – Anghinare sălbatică (Constanța)
Scrophularia nodosa – Bubărică (10), Frunză de bubă rea (9)
Sempervivum tectorum – Chicățăl (5), Licherniță (5), Troscățăl (5)
Serratula tinctoria – Pălămida boiangiilor (9)
Seseli osseum Crantz – Smeoaie (6)
Seseli pallasii Besser (Seseli varium Trev) – Smeoaie (6)
Shorea robusta C.F. Gaertn. – Sal
Sisymbrium officinale – Bruncuță (9), Bruncruț (9), Burnicruț (9), Sărncuță (9)
Smilax china – China noduroasă (3)
Solanum nigrum – Vindecătoare (Racova-Satu Mare), Zărnucă (Corund-Satu Mare)
Solanum tuberosum – Barabușcă (9), Bramburi (10), Cărăboi (11), Columpiri (11), Compiri (11), Corompei (11), Cucule (9), Râpe (11)
Solidago virgaurea – Salata muntelui (Victoria-Brașov)
?Stachys germanica – Sască (Cernetu-Teleorman)
Stachys lanata – Ureceha ursului (Moțăței-Dolj)
Stipa capillata – Mălură (7)

Stipa pennata – Păniță (9)
Stipa stenophylla – Colie (Păuca-Sibiu)
Strelitzia sp. – Floarea paradisului, Pasărea paradisului
?Succisa pratensis – Roien alb (16)
Symphytum officinale – Iarba lui Itate, Tat, Tatan (Ardan-Bistrița-Năsăud)
Tagetes sp. – Buzăiană (10)
Tamarindus indica – Tamarind (3)
Tanacetum balsamita – Golopăr (9)
Taraxacum officinale – Pilug (7)
Tectona grandis L.f. – (Arbore de) Tic, (Arbore de) Tec
Thalictrum flavum – Coada mielului (5)
Thymus comosus – Cimbru netot (Robeşti-Vâlcea)
Tragopogon orientalis – Cuci (8)
Tribulus terrestris – Păducherniță (9)
Triticum aestivum – soiurile Grâu cârnău de vară sau văratic, grâu cârnău mustăcios (9); firele de grâu răsărite târziu se numesc poghirc(ă) (14)
Triticum durum – Grâu sticlos, grâu orzesc (15)
Triticum monococcum – Grâu de un bob (15)
Triticum spelta – Grâu colus, grâu coluz (9)
Tussilago farfara – Ropan (7), Ropanior (7)
Typha sp. – Bocsău (10)
Vallisneria spiralis – Sârma apei
Veratrum album – Bozățel (9)
Veronica beccabunga – Bobovnic (1)
Vicia sativa (sp.) – Bicău (10), Bichiş (10), Bichiță (10), Boarşă (10)
Victoria amazonica (Poepp.) Sowerby – Nufăr de Amazon
Viola tricolor – Floarea Sfintei Treimi (2)
Vitis vinifera – Ciocot (9); soiuri: apătoşi (11), balaban (11), boabă (11), boboasă (11), bordeancă (11), bordo(u) (11), bordun (11,14), buschet (11), buşetă (11), cardonai (11), ceasla napoleon (11), coarbă (11), codargă (11), corb (11), corbească (11), crăpătoare (11), epurească (11), ferdinară (11), godarcă (11), gudarcă (11), însărcinată (11), jachet (11), jichet (11), mieluşei (11), mişchet (11), negru-moale (11), noah (11), nohan(ă) (11), novac (11), ochiul-oii (11), parmac (11), pârnău (14), plesnitoare (11), purcel (11), seibel (11), seiber (11), şardone(t) (11), teras (11), teraz (11), țigancă (11), țâță albă (11), țâță mare (11), țâță mică (11)

Zea mays – Păpuşai (Racova-Satu Mare); Tulpinile: bâlii (11), ciocani (11), coceni (11), fire (11), foi (11), hluji, hlujani (11), paie (11), strujani, strujeni (11), tulei, tuleni, tulheni (11), turjeni (11), vejii (11), ştiuleţii slab dezvoltaţi: ocioc (14), stilele (mătasea): cică (Ardan-Bistriţa-Năsăud); Soiuri: alburiu (9), măselat (9), morânglav (9), turcesc (9)

Ziziphus jujuba Mill. – Jujuba

The Romanian popular names which I couldn't establish species that they belong are:

- Biliora (10) floare asemănătoare cu muşcata
- Bârcaci (10) un burete de cioată
- Boierel (14) plantă cu flori galbene
- Bombir (10)
- Bosâiocul cânelui (10)
- Budeancă (14) o floare (probabil Tagetes)
- Buiedea bubei (10) se pune pe bube
- Buiezi d-ăle cu lapte, lăptaş (10)
- Buiezi de mâlcaviţă (10) contra reumatismului
- Buiezi de urât (10)
- Buiedzile datului (10)
- Buiezile popii (10) se pune pe răni
- Buledea gârlei (10)
- Buruiană de gânduri negre (2)
- Buruiană de năduşală (5)
- Buruiană de venin (5)
- Buşicei (14)
- Cicic (14) o floare roşie
- Coada mielului (5)
- Coconeţ (14) o ciupercă
- Frâna cocoşului (Ardan-Bistriţa-Năsăud)
- Fus (16) utilizată la arsuri
- Handră (14) iarbă ce creşte prin grâu
- Mateuş (14)
- Pieptănuş (14)
- Porcuţ (1) băi contra podagrei
- Rocă (14)
- Rogojel (14)

- Roien negru (16) folosită contra reumatismului
- Rusalin (14) o floare
- Şareş (16) cu rădăcina se fac fumigaţii contra fricii
- Vătajel (14)

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MICROBIAL COMMUNITIES AND ENZYMATIC ACTIVITIES IN SOME TRANSILVANIAN CAVES

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Rezumat: Microorganismele sunt cele mai numeroase şi cele mai răspândite organisme în mediul subteran dar în acelaşi timp şi cele mai puţin cunoscute. În această lucrare propunem să evidenţiem prezenţa microorganismelor în câteva peşteri transilvănene. Pentru aceasta am folosit atât metode microbiologice cât şi enzimatologice. În urma analizelor au putut fi evidenţiate microorganisme de diferite tipuri: oligotrofe, fier-reducătoare, amonificatoare, în toate peşterile, dar şi activităţi enzimatice precum activitatea dehidrogenazică, activitatea catalazică şi activitatea fosfatazică.

Microorganisms are a heterogeneous group, present in almost every environment; therefore they should be present in caves as well. This environment, the underground environment, makes the subject of our study. Very little studies have been made on this topic in the country as well as abroad. This is due to the methodological difficulties present at any stage of the study. In the beginning these studies have been carried out by hygienist, who were concerned only about the pathogen germs present in subterranean waters. But the bacteria that live fixed on solid surface are more abundant than the ones in the water (GOUNOT, 1994). Another major problem is to determine weather the bacteria are autochthonous or allochthonuos. JAMES (1994) states that many bacteria from caves, even those of deep caves, are present here accidentally and they develop only if they find favorable conditions.

Bacteria from caves have been studied for their potential role in the formation and dissolution of rocks. *CANAVERAS et. al.* (1999) suggested that bacteria present in caves may play a role in the formation of moonmilk deposits, as microbial communities predominantly composed of different species of the genus Streptomyces were found in association with hydromagnesite and needle-fiber aragonite deposits in the Altamira cave

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(Spain). Guadalupe Mountain (USA) caves contain a number of examples of possible interactions between microorganisms and speleothems. In particular, Lechuguilla and Cottonwood caves contain speleothems that have been referred to as „biothems" by *CUNNINGHAM et. al.* (1995) – features such as webulites and u-loops that appear to be calcified filamentous microorganisms. The discovery of „snottites" in Cueva de Villa Luz (*HOSE & PISAROWICS* 1999: *HOSE et. al.* 2000), a cave with active hydrogen sulfide vents, allows researches to speculate that such bacterial structures could become lithified later in the evolution of the cave, producting the u-loops that we see today. *DAVIS et. al* (1990) demonstrated the presence of filaments in the cores of the „rusticles", an iron oxide speleothem in Lechuguilla Cave.

A large variety of microorganisms were isolated from caves. Species from groups like actinobacteria, cyanobacteria, even archaebacteria were found to be living in caves. *GOUNOU* (1967) and *SEMIKOLENNYKH* (1997) isolated a series of actinobacteria in different caves from France and Rusia. *CANAVERAS et. al.* (1999) attributes an important role in the moonmilk formation to actinobacteria like: *Streptomyces roseoviridis, S. tuirus, S. xanthophaeus, S. flavogriseus, S. flavotricini.*

MENNE (1997) discovered in Rettenbachhole (Germany) species like *Myxococcus fulvus* and *Corallococcus coralloides* belonging to mixobacteria.

GRUIA (1964) describes a new species of cyanobacteria in Ialomiţa Cave (Romania) which he names *Ialomitzia cavernicola. JONES* (1995) considers cyanobacteria *Gleitleria calcarea* to be having an important role in the process of calcification in some caves from Cayman Islands (USA).

NORTHUP et. al. (1998, 2000) discovered in Lechuguilla Cave (USA) a new type of archaebacteria, of low temperature, which they named *Crenarchaeota.*

MOORE AND SULLIVAN (1997) discuss the possibility of using actinobacteria in medicine. It is known that moonmilk has been used for wound treatment since 16 century. And it is also known that soils and moonmilk from caves contain actinobacteria, which produce antibiotics, so the authors are enthusiastic about the possibility of discovering new and more efficient antibiotics.

Our goal in this study is to determine weather microorganisms are present in the samples we extracted from four transilvanian caves. The presence of microorganisms was tested using culturable and non culturable methods.

Materials and methods

Samples were collected from four caves Darninii, Humpleu, Huda lui Paparî and Ungurului. Some, were aseptically sampled using a flame-sterilized spatula and then they were tested using culturable methods. Others, which were not aseptically sampled, were used only to determine the enzymatic activity of the soils. Samples varied from soils to moonmilk and even guano. They were taken from different parts of the caves as you can see in Figures 1 to 4.

As microbiological methods we used selective media for oligotrophic bacteria, for iron-reducing bacteria, amonifieing bacteria and heterotrophic bacteria.

The media for oligotrophic bacteria, that we used, was described by *HATTORI & HATTORI* (1980) and it consists of: 10 g peptone, 10 g beef extract and 5 g NaCl. This solution is diluted in the moment of use by 10, 100 and 1000 times. The media was distributed in tubes (10ml/tube) and then sterilized at 120°C for one hour.

For iron-reducing bacteria we used a media consisting of: 3 g K_2HPO_4, 0,8 g KH_2PO_4, 0,2 g KCl, 0,2 g $MgSO_4 \cdot 7H_2O$, 0,5 g yeast extract, 5 g peptone, 20 g glucose, 1 g $Fe_2O_3 \cdot 3H_2O$. These are dissolved in 1000 ml distilled water, parted in tubes (7 ml/tube) and sterilized at 105°C for one hour (*OTTOW*, 1968).

Amonifieing bacteria were determined on a media consisting of 2 g peptone, 0,5 g NaCl and 100 ml distilled water; pH=7,0. The media, parted in tubes (10 ml/tube) was sterilized at 120°C for one hour.

These media were inoculated with the samples took from caves than incubated at 28°C for one week. On the media for oligotrophic bacteria we checked for the presence of microbial mats, for the other two media the presence of bacteria was determined using a color reaction.

As enzymatic activities we tested the dehydrogenase activity, the catalase activity and the phosphatase activity.

Catalase activity was determined using a technique based on *KAPPEN*'s method (1913). We took 3 g of material, active or inactivated at 120°C, added 10 ml of distilled water and 2 ml of H_2O_2 3%. This mixture was incubated at 20°C for one hour. The catalase activity is calculated from the difference between the active samples and the inctivated ones and is expressed in mg of H_2O_2.

Phosphatase activity was determined using mixturea of 5 g soiul and 10 ml of disodic fenilphosphate solution 0,5%. Because incubation took more than 2 hours we added 2 ml of benzene. Incubation took place at 37°C for 3 days. After incubation we added alaun solution, borax tampon and Gibbs reactive and we obtain a blue color. The intensity of the color shows the intensity of the reaction. Phosphatase activity is measured in mg of phenol/5 g of soil (*DRĂGAN-BULARDA*, 2000).

Dehydrogenase activity was determined using *CASIDA et. al.* (1964) method. The mixtures consisted of 3 g of material, 0,5 ml TTC 3% and 2 ml distilled water for the actual activity. For the potential activity instead of 2 ml of water we used only one and added another one of glucose 3%. Incubation took place at 37°C for 72 hours. The activity is measured in mg of trifenilformazan.

For each activity we also prepared witness samples without substrate or without soil.

Results

On the media for oligotrophic bacteria we obtain microbial communities which we than analyzed using photon microscopy. Samples examined in this way came from Darninii Cave. From Table 1 we can see that in this samples we had all kinds of bacteria like cocci, bacilli, bacterial spores, even yeast.

Table 1

Microscopic results obtained on oligotrophic media for samples from Darninii Cave.

Sample no.	Dilution	Morphologic groups	Gram Coloration	Dimensions (μm)
1	Not diluted	Cocci	Gram+	1
		Yeast	Gram+	2-3,5
		Bacilli	Gram-	1
		Yeast	Gram+	1-3
		Bacilli	Gram+	3-5
		Bacterial spores	Gram+	1,5-2
	Dilution 1/10	Bacilli	Gram-	≤1
		Bacterial spores	Gram-	1,5

	Dilution 1/100	-		
2	Not diluted	Yeast Bacilli	Gram+ Gram-	1-3 2
3	Not diluted Dilution 1/10; 1/100	-		
4	Not diluted	Bacilli Bacilli Bacilli	Gram+ Gram+ Gram-	3-5 5-10 1-3
	Dilution 1/10; 1/100	-		
5	Not diluted Dilution 1/10; 1/100	-		
6	Not diluted Dilution 1/10; 1/100	-		

Same samples inoculated on heterotrophic media did not grow bacterial cultures.

Samples from Humpleu Cave and Huda lui Paparî Cave were inoculated on media for iron-reducing bacteria. The results were taken after 4 days and then after 20 days because the reactive has a slow activity and we wanted to see if this influences our results. And as we can see in Table 2 it does. After 4 days only samples 2 and 4 from Humpleu Cave and samples 2 and 3 from Huda lui Paparî Cave showed the presence of iron-reducing bacteria, but, after 20 days all samples except sample 1 from Humpleu Cave showed the presence of iron-reducing bacteria.

Table 2

The presence of iron-reducing bacteria in samples from Humpleu Cave and Huda lui Papară Cave.

Origin of samples	Sample no.	Results	
		After 4 days	After 20 days
Humpleu Cave	1	-	-
	2	+	+
	3	-	+

	4	+	+
Huda lui Papară	1	-	+
	2	+	+
	3	+	+
	4	-	+
	5	-	+

For samples from Ungurului Cave besides iron-reducing bacteria we also inoculated media for amonifieing bacteria. The inoculus used for iron-reducing media was diluted 10, 100 and 1000 times and we can see that the amount of iron-reducin bacteria that grew on the media was proportional with the dilution. So far the first dilution, which was only 10 times we have the greatest number of bacteria and for the other two dilutions 100 and 1000 times the number decreases. As for the amonifieing bacteria we could reveal their presence in all three samples analyzed (Table 3).

Table 3

The presence of iron-reducing bacteria and amonifieing bacteria in samples from Ungurului Cave.

	Iron-reducing bacteria			Amonifieing bacteria
Dilution Sample no.	0,1	0,01	0,001	0,1
2	+	+-	-	+
6	+++	+	+	+++
9	+	+	+	+++

Now, for the enzymatic analyses we tested the dehydrogenase activity in all four caves and for all the samples. As we can see from the Table 4 and 5 the real dehydrogenase activity is very low, almost inexistent, it could only be measured in samples 2,3,4,5 from Darninii Cave, sample 5 from Huda lui Papară Cave and samples 4 and 10 from Ungurului Cave. But all the samples showed a potential dehydrogenase activity, except sample 6 from Darninii Cave.

Table 4

Dehydrogenase activity in samples from Darninii, Humpleu and Huda lui Papară Cave.

Origin of samples	Sample no.	Dehydrogenase activity	
		Actual	Potential
Darninii Cave	1	-	+
	2	+	+
	3	+	+
	4	+	+
	5	+	+
	6	-	-
Humpleu Cave	1	-	+
	2	-	+
	3	-	+
	4	-	+
Huda lui Papară	1	-	+
	2	-	+
	3	-	+
	4	-	+
	5	+	+

For samples collected from Ungurului Cave we also tested the catalase activity and the phosphatase activity of the soils. This results, along with the dehydrogenase results are shown in Table 5.

Table 5

Three enzymatic activities in Ungurului Cave: dehydrogenase, catalase and phosphatase activity.

Sample no.	Dehydrogenase activity		Phosphatase activity	Catalase activity
	actual	potential		
1	0	0,120	0,021	4,25
2	0	0,040	0,018	7,735
3	0,073	0,140	0,020	5,27
4	0	0,055	0,025	0,765

5	0	0,005	0,020	8,5
6	0	0,150	0,019	5,355
7	0,064	0,152	0,024	7,48
8	0	0,151	0,021	1,53

Discussions

The results show that microorganisms are present in cave soils. An interesting thing is that those microorganisms could develop on a low nutrient media, but they were not able to do that on a normal one, for heterotrophic bacteria. This thing leads us to the conclusion that microorganisms from caves are oligotrophs, this may be due to the media in which they grow, the underground environment, which is very poor in organic mather. Iron-reducing bacteria were present in almost all samples. This is quite what we expected to see because in caves we can find many iron oxides, which could be an alternative source of energy for the microorganisms. The presence of amonifieing bacteria cannot be explained, we expected to find these bacteria in guano but it seems that they also exist in other parts of the cave, not necessary related to the presence of bats.

We also tested the presence of certain enzymes in the soils. These enzymes accumulated on organo-mineral particles are the results of thousands of generation of microbial communities demonstrating not only the actual presence of microorganisms but also their succesion during time. So if we have enzymatic activitis in the samples it means that in those samples we have and had microbial communities.

Conclusions

1. Microorganisms are present in all the studies caves and in all kinds of samples: moonmilk, guano, limestone.
2. The microorganisms that could be revealed by cultural methods are: oligotroph bacteria, iron-reducing bacteria and amonifieing bacteria, showing a high metabolic diversity.
3. We can also observe a high diversity of shapes: bacilli, cocci, filaments.
4. Because microbial cultures grew only on oligotrophic media and not on heterotrophic media we can conclude that these microorganisms

are oligotrophs, they are not able to develop on a high nutrient media and they might be adapted to the cave environment which has low nutrient values.

5. Because the bacteria that have been found have certain properties like they are oligotrophs or iron-reducing bacteria we are tempted to say that they are autochthonous.
6. The presence of iron-reducing bacteria is associated with the presence of iron oxides in the cave.
7. The presence of amonifieing bacteria in all samples examined cannot be explained.
8. The presence of some enzymatic activities in caves demonstrates the persistence of microorganisms in this environment.
9. The presence of dehydrogenase activity is a proof of the existence of bacteria because this enzyme is specific only for live organisms.

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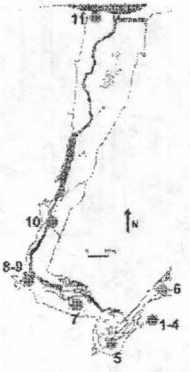
Fig. 1 Huda lui Papară Cave; 1-5 sampling points.
Fig. 2 Ungurului Cave; 1-11 sampling points.



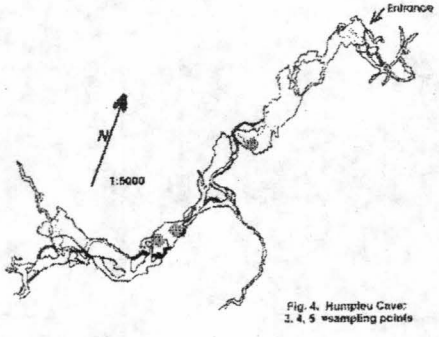


Fig. 4. Humpleu Cave:
2, 4, 5 =sampling points

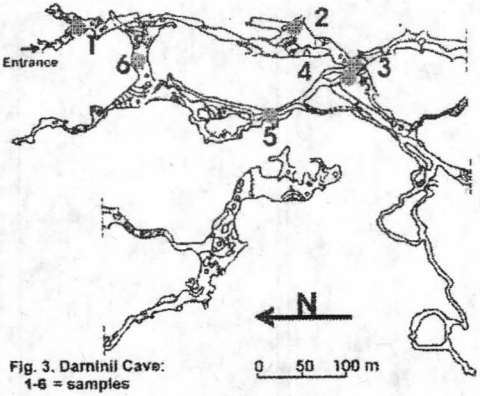


Fig. 3. Darninii Cave:
1-6 = samples

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