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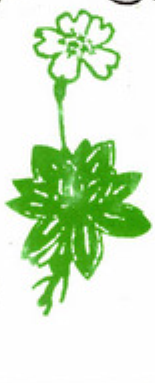
TOME 46

1 2001

janvier – juin

série de biologie végétale



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REVUE
ROUMAINE
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SÉRIE DE BIOLOGIE VÉGÉTALE

TOME 46, N° 1 janvier–juin 2001

SOMMAIRE

MORPHO-ANATOMICAL CHARACTERIZATION OF THE ENDEMIC SPECIES *DIANTHUS CALLIZONUS* SCHOTT ET KOTSCHY

ANCA AIFTIMIE-PĂUNESCU*, CONSTANTIN TOMA**

In this paper it is analyzed the structure of vegetative organs of the endemic species *Dianthus callizonus* for the first time. Our data offer more arguments for its systematic position, but also contradict the previous descriptions according to which this species is a glabrous one.

Key words: morpho-anatomy, endemic species, *Dianthus callizonus*.

INTRODUCTION

According to the "red list" of spermatophytes in Romania, the *Dianthus* genus is pretty well represented and comprises 31 species as follows: 12 with European area, 12 with area over the European limits and 9 are endemic for Romanian Flora. The endemic species of *Dianthus* genus in the Romanian flora are: *D. banaticus*., *D. callizonus, D. campestris* ssp. *serbanii, D. carthusianorum* ssp. *tenuifolius, D. glacialis* ssp. *gelidus, D. henteri, D. kladovanus, D. petraeus* ssp. *spiculifolius* and *D. racovitae.*

On the Jurassic age reefs limestone of Piatra Craiului Massif are growing 53 endemic species approximately. Among these, *Dianthus callizonus* is considered one of the rarest, being found in this area exclusively, that is why it is known as "the pink of Piatra Craiului". This species is the emblem of the Meridional Carpathians. It is very precious from the scientific point of view because it still makes botanists raise questions of genetics and taxonomy [4], so more studies concerning this plant are timely.

In consulted literature the information regarding this species is about chorology, ecology, taxonomic observation and botany description [4, 8, 9]. The histo-anatomical studies concern the *Caryophyllaceae* family [3, 5, 6, 7, 13, 14], other genus [11, 12], short data about this species, [8, 10], or other species of this genus [1, 2].

MATERIAL AND METHODS

The samples were collected from Piatra Craiului Massif ("Piatra Craiului Mare" Mountain, 1900 m altitude).

The fixing, coloring and analyzing methods are the customary ones made in the Phyto-anatomical Laboratory of the University of Iaşi.

The histo-anatomical analysis was made on permanent preparations which comprised superficial sections of the leaf epidermis, cross sections through the root, stem and leaf (sheath and lamina).

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The preparations were examined at Novex-K light microscope and the drawings were performed at MC1 projection microscope.

To examine the micromorphology of the leaves we have used the scanning electron microscopy methods: the leaf samples were dehydrated using physical methods, metallised and analysed at the Tesla BS-340 scanning electron microscope.

Microphotographs were taken from light microscope or from scanning electron microscope.

Photos were taken with the Nikon N6006 AF camera at original habitus (Piatra Craiului).

RESULTS

The **cross-sectioned root** (Plate I fig. 1, 2) has a circular outline.

The root structure is of secondary type being a result of cambium and phellogen activity. The phellogen produces many layers of suber, on the external part and less layers of phelloderm, on the internal one.

The cambium produces less phloem on external part and a thick zone of xylem, on the internal one. The secondary phloem is represented by leading elements on cambial face and by cells of starchy parenchyma, on the phellodermic face. The secondary xylem has vessels of different diameter, disposed in the zone of ground cellulosic parenchyma. Even if the structure is of secondary type, there are not visible elements of libriform.

The stem in cross-section from the basal node (usually subterranean) has an irregular outline. The epidermis has small isodiametric cells with thickened and cutinizated external walls. The bark is thin, made up of 3–4 layers of parenchymatous cells.

The central cylinder starts with a thick sclerenchymatous and lignified pericycle made up of tangentially elongated cells with thick walls, which is typical for the most of the *caryophyllaceus* species (Chermezon, 1910; Toma *et al.*, 1969, 1971; Metcalfe and Chalk, 1972; Simeanu and Corneanu, 1978).

The conducting tissue is made up of vascular bundles on a ring disposed, some of them being prominent in the pith. All the bundles have less xylem vessels with thick and lignified walls.

All the interfascicular, perifascicular and medullar parenchyma has moderated thick and lignified walls with numerous pits.

On some sections there are leaves basis glued to the stem epidermis, having a simple structure (homogeneous parenchyma with vascular bundles). The external epidermis has small tangentially elongated cells and the internal one has big radial elongated cells.

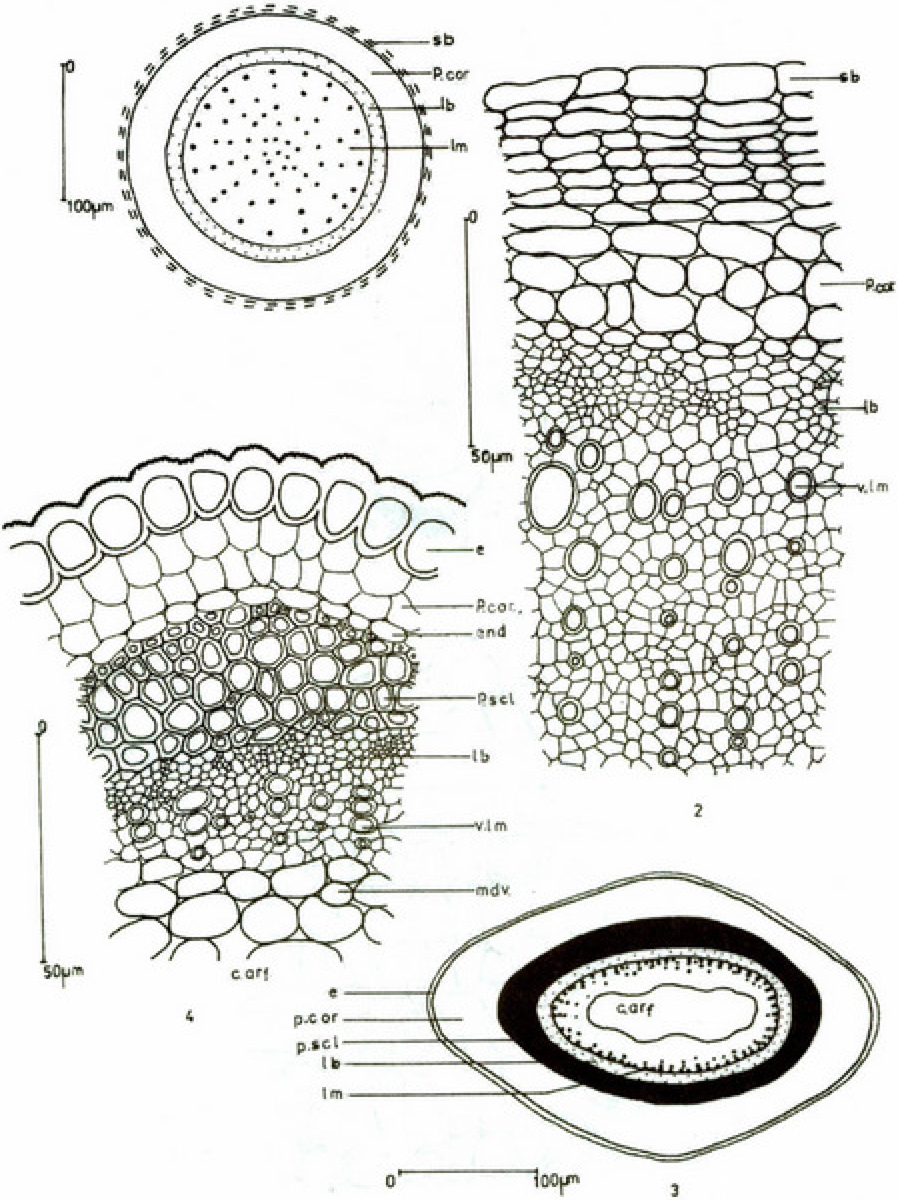


Plate I: Cross-section of the root: fig. 1 – diagram, fig. 2 – detail.
Cross-section of the stem (internode): fig. 3 – diagram, fig. 4 – detail.

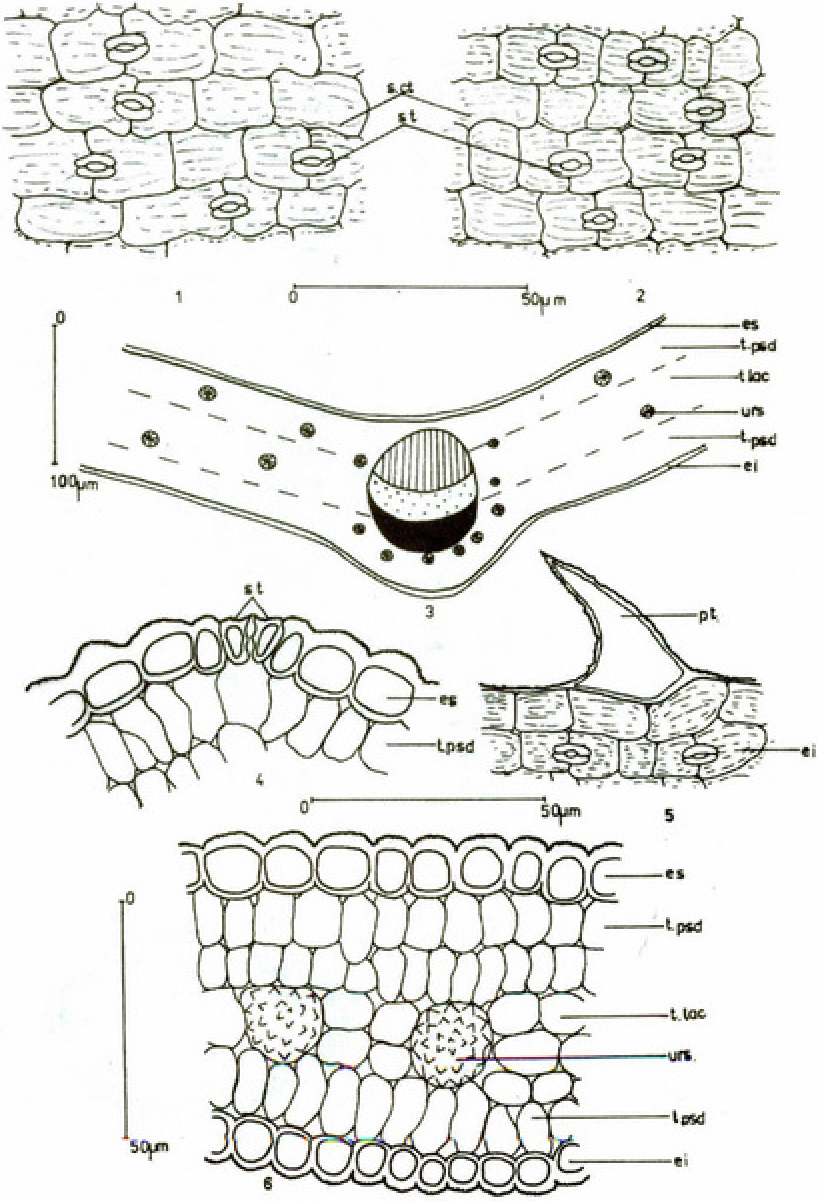


Plate II: Superficial section of the lamina: fig. 1 – upper epidermis – detail, fig. 2 – lower epidermis – detail. Cross-section of the lamina: fig. 3 – midvein – diagram, fig. 4 – stomata – detail, fig. 5 – covering hair – detail, fig. 6 – detail between the veins.

Photo 1 – ***Dianthus callizonus*** – original habit.

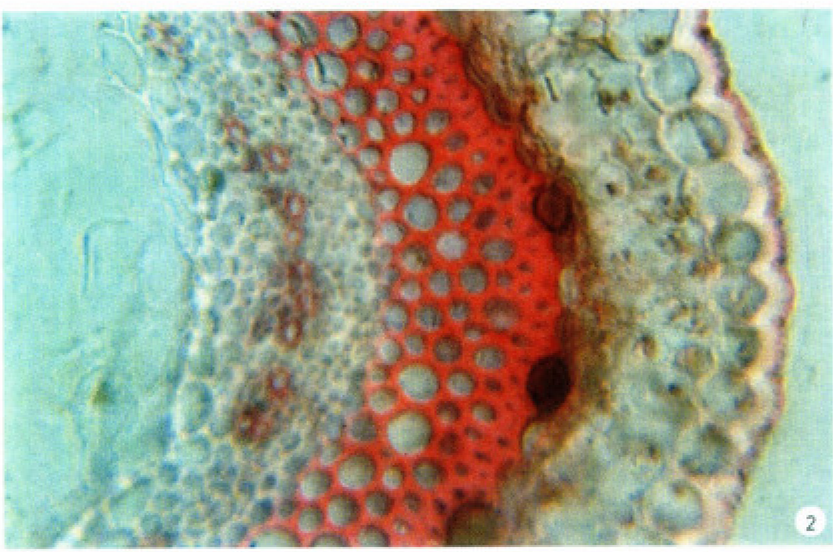


Photo 2 – Light microscopical view from cross-section of the stem (internode level).

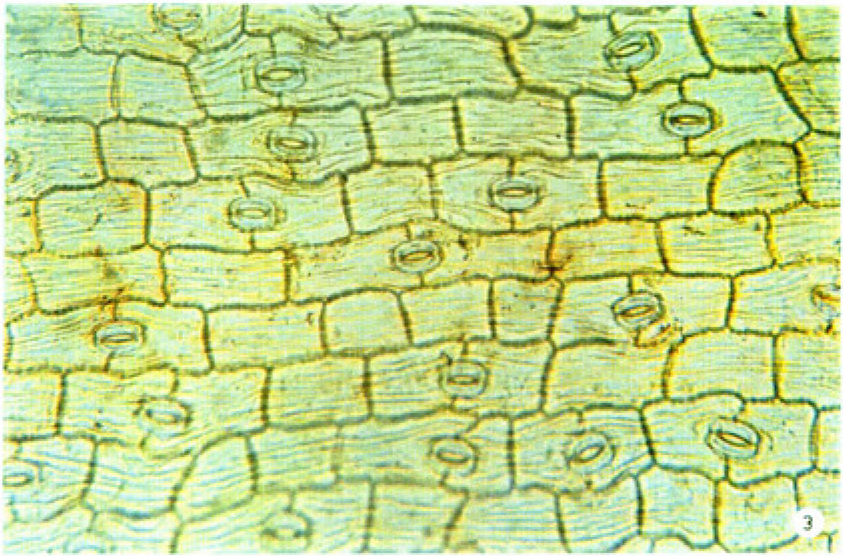


Photo 3 – Light microscopical view of epidermis (superficial section).

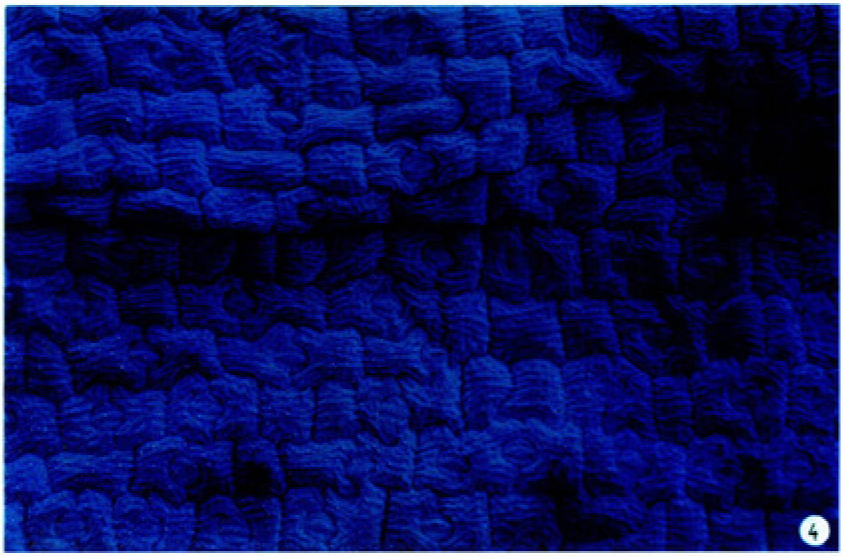


Photo 4 – Electron-scan microscopical view of the epidermis.

At the basal level, the internode has the following structural characteristics:

– epidermis made of isodiametric cells, with the external wall thickened and covered by a thick cuticle;

– cortical parenchyma of meatus type;

– as a result of phellogen activity, there is a thick zone of suber (10–12 layers of cells) on the external part and a thin one (3–4 layers of cells) of collenchymatous phelloderm on the internal part;

– the central cylinder is very thick, being limited at the external part by a sclerenchymatous pericycle 3–4 layered, made up of cells with very thick and lignified walls;

– the vascular tissue is made up of two concentric rings, the phloemic one being thinner and collenchymatised;

– pith is of meatus type made up of large parenchymatous cells, some of them with ursins of calcium oxalate;

– at the node level, the xylem ring is interrupted in two opposite zones, so it forms two bands face to face separated by two small bundles.

On the terminal internode level, (Plate I, fig. 3, 4, photo 2) the structure is of primary type:

– epidermis has tangentially elongated cells with very thick external and internal walls, the external one being covered by a thin cuticle;

– bark is thick, made up of 8–10 layers of cells, less collenchymatised;

– pericycle is sclerenchymatous, thick (3–5 layered) made up of cells with thick and lignified walls;

– vascular tissue is made up of two concentric rings: a phloemic collenchymatised external one and another xylemic with scattered vessels on the cellulosic parenchyma;

– pith is thick of parenchymatous-cellulosic type.

The cross-section between the last superior node has an elliptic outline.

Epidermis has big cells with very thick external wall covered by a crenated cuticle. The bark is thin with different size cells some of them with calcium oxalate ursins. The sclerenchymatous pericycle is very thick (5–8 layers of cells).

In the central cylinder, the vascular tissue has the same ring-like structure but the xylem is interrupted forming two opposite arches.

The pith is thick parenchymatous cellulosic of meatus type.

The leaf (Plate II, fig. 1–6, photos 3, 4). The base of lamina for the two opposite leaves forms a ring thicker in two poles because of the vascular bundles which are prominent at the external (lower) face; these bundles have a primary type structure with a sclerenchymatous sheath in the external part of the phloem. The epidermis presents big cells tangentially elongated with the external wall very thick and covered by a crenated cuticle. Between the lateral veins, the mesophyll is very thin (1–2 layers of cells) but it has big cells also with calcium oxalate ursins.

In the middle part of lamina, the epidermis in surface view is made of polygonal cells with straight lateral walls. On the surface of the external wall there are visible numerous cuticular streaks. From place to place there are stomata of diacitic (caryophyllaceous) type in both epidermes. On the edge of the lamina there are unicellular or bicellular short needle-shaped covering hairs supported by a stalk made by two epidermal cells.

In cross-section the lamina has a "V" letter outline with very distant branches. There are big tangentially elongated cells in both epidermes with very thick external wall and covered by a fine streaked and crenate cuticle. From place to place there are visible stomata in both epidermis and on the edge unicellular and bicellular covering hairs with very thick walls.

The midvein, in cross-section, is a little prominent on the lower face and comprises one main vascular bundle with a primary type structure, accompanied by sclerenchyma. In the fundamental parenchyma there are some big cells with calcium oxalate ursins.

The lateral secondary veins have the same structure with smaller bundles but not accompanied by sclerenchyma. The lateral tertiary veins have small bundles with 2–3 small xylem vessels, whole bundle being surrounded by a parenchymatous sheath.

The mesophyll is differentiated in palisade tissue towards both the epidermis and spongy tissue in the middle. The palisade tissue is often one-layered on the lower face and two-layered on the upper face. Some cells from the middle mesophyll are bigger and with calcium oxalate crystals.

DISCUSSION AND CONCLUSIONS

If we compare the results of our researches by *Dianthus callizonus* with those existing in the literature concerning *Caryophyllaceae* family, we can conclude that this species has some general structure characteristics common to the members of this family, but also some specific ones. Among the common ones, the most obvious are:

- on the root level
  – xylem composed mainly of parenchyma but including scattered vessels;
  – the absence of medullary rays;
  – even if the structure is of secondary type, there are no libriforms;
- on the stem level
  – cork of pericyclic origin;
  – thick sclerenchymatous and lignified pericycle;
  – ring-like disposed conducting tissue;
  – big parenchymatous cells with calcium oxalate ursins.

The arrangement of the vascular tissue appears to be characteristic for the species and therefore of diagnostic value.

- on the leaf level – stomata of diacitic type for both epidermes;
  – palisade tissue towards both faces.

Among the specific characteristics the most interesting seems to be those from the leaf level. On the surface view the epidermis has cells with the external wall covered by a streaked and crenate cuticle (Plate II, fig. 1, 2). The cuticular streaks are obvious for both light and scanning electron microscope (Photo 3, 4). We underline this feature because the data from consulted literature points out that the shape and the appearance of epidermis cells have taxonomic importance [6, 7, 9, 10].

The presence of the covering hairs on the leaf edge is recorded for the first time for this species. In the specific literature it is noticed that this species is glabrous.

Abbreviations: c.arf. – air cavity; **c.ep.** – epidermal cell; **c.st.** – stomatic cell; **col.** – collenchyma; **ep.** – epidermis (**i** – lower, **s** – upper); **f.c.** – vascular bundle; **lb.** – phloem; **lm.** – xylem; **p.cor.** – cortical parenchyma; **p.fd.** – fundamental parenchyma; **p.scl.** – sclerenchymatous pericycle; **sb.** – suber; **scl.** – sclerenchyma; **s.ct.** – cuticular streaks; **t.lac.** – spongy tissue; **t.psd.** – palisade parenchyma; **p.t.** – covering hair; **urs.** – calcium oxalate ursin; **v.lb.** – phloemic bundle; **v.lm.** – xylemic bundle.

REFERENCES

1. AIFTIMIE A., 1998, *Anatomical researches upon some endemic plants III* ***Dianthus tenuifolius*** *Schur*. An. Şt.Univ. "Al.I.Cuza" Iaşi, s. II a (Biol.veget.), **44**: 29–36.
2. AIFTIMIE A., 1999, *Anatomical researches upon some endemic plants IV* ***Dianthus petraeus*** *ssp.* ***spiculifolius***, ***Dianthus pontederae*** *ssp.* ***kladovanus***. An. Şt. Univ. "Al.I.Cuza" Iaşi, s. II a (Biol.veget.), **45**: 55–62.
3. DEYSSON G., 1965, *Eléments d'anatomie des plantes vasculaires*. Ed. Sedes, Paris.
4. DIHORU G., PÂRVU C., 1987, *Plante endemice în flora României*. Ed. Ceres, Bucureşti.
5. ESAU K., 1965, *Plant anatomy*. John Wiley and Sons, Inc., New York, London, Sydney.
6. METCALFE C.R., CHALK L., 1972, *Anatomy of the Dicotyledons*. **2**, Clarendon Press, Oxford.
7. NAPP-ZINN KL., 1973, 1974, *Anatomie des Blattes. II.Angiospermen*. In: *Handbuch der Pflanzenanatomie*, **8** 2A_{1-2}, Gebrüder Borntraeger, Berlin, Stuttgart.
8. SANDA V., 1968, *Câteva consideraţii critice asupra caracterelor anatomo-morfologice în taxonomia unor specii ale genului* ***Dianthus*** *L.* Stud. Cerc. Biol., ser. Biol. veget., **20** (4): 337–343.
9. SANDA V.,1969, *Contribuţii la corologia şi taxonomia subsecţiei* ***Alpinii*** *Vierharpper a genului* ***Dianthus*** *L. din flora României.* Stud. Cerc. Biol., ser. Biol. veget., **21** (1): 23–30.
10. SIMEANU V., CORNEANU G., 1978, *Studii morfo-anatomice asupra unor specii ale genului* ***Dianthus*** *L.* An. Univ. Craiova, **9** (19): 19–23.
11. TOMA C., 1969, *Structura anatomică a organelor vegetative de* ***Lychnis (Silene, Polyschemone) nivalis.*** Kit., endemism propriu Munţilor Rodnei. Com. de Bot. ale S.S.B. : 251–266.

12. TOMA C., RUGINĂ R., FLENCHEA G., TONIUC A., 1971, *Contribuţii la diferenţierea anatomică a speciilor de* ***Lychnis*** *din România.* Stud. Com. Muz. Şt. Nat. Bacău, 535–548.
13. TOMA C., RUGINĂ R., 1998, *Anatomia plantelor medicinale. Atlas.* Ed. Acad. Rom., Bucureşti.
14. VESQUE J., 1883, *Contribution à l'histologie systématique de la feuille des* **Caryophyllinées**. Ann. des Sci. Nat. Bot., sér. 6, **15**: 107–147.

Received March 10, 2001.

* *Institute of Biology, Splaiul Independenţei 296, Bucureşti, Romania*

** *"Al.I.Cuza" University, Faculty of Biology, Bd. Copou 20A, 6600, Iaşi, Romania*

THE ULTRASTRUCTURAL MESOPHYLL STUDY IN THE DESCENDANTS OF *NICOTIANA TABACUM* L. (cv. *Xanthi*) TRANSFORMED PLANTS

RODICA DUMITRESCU

In this experiment there were used leaves taken from the descendants obtained by germination of the seeds of the transformed plants by electrotransfection and co-cultivation methods, with the help of the binary vector p JIT 79, which contains markers of the gene npt II and gus.

The aim of this paper is the testing of the gene expression stability at the cellular level at the descendants of the plants genetically transformed, cultivated in natural conditions.

The morphometrical analysis of leaf chloroplasts at *Nicotiana tabacum* L. (cv. *Xanthi*) descendants showed a decrease of the measured parameters to the control. In the case of the descendants of the transformed plants obtained by co-cultivation with *Agrobacterium tumefaciens* containing the plasmid p JIT 79, on the medium without kanamycin chloroplasts of different organization have been noticed. The big number of thylakoid/chloroplasts (134,9) though smaller than control (239,60), is higher than that of the descendants "D". These modifications were caused by the disorganization of the internal thylakoidal system, as a result of the starch inclusion big size, which caused breaking of the stromatic thylakoids, which link the grana.

Key words: Nicotiana tabacum L. (cv. *Xanthi*), descendants, pJit 79, *Agrobacterium tumefaciens*, co-cultivation.

INTRODUCTION

A remarkable scientific progress led to the application of the genetic transformation methodology recently. In comparison with the plant improvement, the genetic transformation offers the possibility of introduction of only one character to a variety estimated as remarkable, and the transferred gene can come from any source that may extend the possibility of plant improvement.

The obtaining of the transformed plants by molecular biology techniques consists in the stable introduction and expression of some new genes, having the role of making the plants resistant to illness, pests, herbs, antibiotics and so on. This process goes through three stages:

a. The identification, the isolation and the cloning of the gene we are interested in.

b. The transfer of the interest gene made by genetically engineering techniques for the culture plants.

c. The plants selection which shows the transferred character and the gene expression stability test, in time, in natural conditions (1).

The *Agrobacterium* mediated transformation was studied at both the dicotyledonous plants (16) and monocotyledonous plants (5). At the monocotyledonous ones the gene transfer efficiency is low in comparison with dicotyledonous plants. The use of Ti plasmid of *Agrobacterium tumefaciens* as an effective vector to introduce foreign genes into cells of dicotyledonous plants has encouraged the wide application of these techniques for legume crops (13, 17).

Electroporation, i.e. electric field mediated membrane permeabilization (23, 19).

In this experiment there were used leaves taken from the descendants obtained by germination of the seeds of the transformed plants by electrotransfection and co-cultivation methods, with the help of the binary vector p JIT 79, which contains markers of the gene npt II and gus (3,11).

The information in the literature proves that the genetic transformed plants lead to their genetic diversity. The new features due to transfer can be transmitted to descendants in a mendelian manner, in a segregation ratio of 3/1 (7). Rovenska J. *et al.* (18), prove that the expression of genes in the transformed cells can modify several pathways of biosynthesis. Tepfer D. (20), Brainsberger E.S. *et al.* (4), Hlinkova E.and Ondrej M. (14), Hlinkova E. (15) confirmed the expression of main morphoregulatory genes in various combinations changed both the qualitative and quantitative content of soluble proteins which influenced the phenotype of the transformed plants. Protein modifications are associated with genetic message modifications.

The aim of this paper is the test of the gene expression stability at the cellular level at the descendants of the plants genetically transformed, cultivated in natural conditions.

MATERIAL AND METHODS

1. Plant material. This study was carried out at the level of descendants obtained in natural conditions from the seeds of the plants genetically transformed, of *Nicotiana tabacum L.* (cv. *Xanhti*). The plants were genetically transformed from petiole inocula by two methods: electrotransfection and co-cultivation.

2. Electroporation condition: the electroporation was made with rectangular electric pulses 0.125 kw/cm and 0.15 kw/cm, representing the best condition for electrotransfection (2).

Plasmid DNA. Ap BIN-19 derivate plasmid p JIT-79, containing the gus gene linked to 35 S promoter + Ca MV gene (19 S) ATE and chimeric 5' non – NPT II nos – 3' gene, conferring kanamycin (km) resistance was used.

3. The descendants obtained. After passing all the biotechnology stages genetically transformed plants were obtained. The plants that responded positively to the histochemical test gus and km were cultivated in soil. From the seeds of the transformed plants there were obtained descendants having different phenotypic aspects, which were denoted by D1, D2, and D3. (3).

4. Electronomicroscopical studies. To make electronomicroscopical studies, samples of foliate mesophyll of the descendants D1, D2, D3, obtained by germination of the seeds of the transformed plants by electrotransfection and co-cultivation, with p JIT 79 of *Nicotiana tabacum* L. (cv. *Xanthi*) have been fixed in 1% glutaraldehyde, postfixed in 1% OsO4, and embedded in the synthetic resin ME 6611. The ultrasections were stained with uranyl acetate and contrasted by Reynolds method and visualized on Tesla BS-500 electron microscope.

5. Morphometrical analysis. It was made by evaluation of some cellular parameters in 30 chloroplasts/variant.

6. Statistical analysis. For statistical analysis of the parameters evaluated t test was used.

RESULTS AND DISCUSSION

I. ULTRASTRUCTURAL ASPECTS OF THE MESOPHYLL CELLS IN THE DESCENDANTS OF PLANTS TRANSFORMED BY *NICOTIANA TABACUM* L. (cv. *Xanthi*)

– **Control, plant mesophyll** made evident normal parenchymatic cells with their big central vacuole and their cytoplasm rich in cellular organelles with characteristic structures (Pl. I, fig. 1).

– **The mesophyll of descendants D1,** resulted from electrotransfection-transformed plants, does not show significant modifications to control. The nucleus laid in the center has its chromatin granular fibrillar and its nuclear membrane intact and non-dilated. The hyaloplasma rich in cellular organelles with characteristic structures shows an incipient process of vacuolization (Pl. I, fig. 2) which intensifies in some cells. Some vacuoles contain electronoopaque formations (Pl. I, fig. 3). Multilamellar bodies have been found in some vacuoles (Pl. I, fig. 4) as a result of the membrane rest metabolisation.

Generally, the chloroplasts have a parietal position and show a well organized thylakoidal system (Pl. II, fig. 5). In some situations, the chloroplasts are grouped occupying the whole cellular space (Pl. II, fig. 6).

– **The mesophyll of the descendants D2** shows a rarefaction of the hyaloplasma and diminution of the cellular organelles number. The chloroplasts have their thylakoidal system disorganized because of the presence of the big size amiliphere inclusions (Pl. II, fig. 7). It can be noticed the intensification of the vacuolization processes. Mitochondria have dilated crista. Some chloroplasts are in different stages of differentiation (Pl. II, fig. 8), while the others have their thylakoidal system well represented (Pl. III, fig. 9).

– The mesophyll of the descendants D3 evidenced a clear plasmalemma detachment of the cellular wall (Pl. III, fig. 10) and plasmalemma vesicularisation (Pl. III, fig. 11). Atypical mitochondria and heavily modified chloroplasts are present among the cellular organelles (Pl. III, fig. 12).

– The mesophyll from descendants of the transformed plants by co-cultivation, with *Agrobacterium tumefaciens,* which contains plasmid p JIT 79.

The plasmalemma detachment of the cell wall and a hyaloplasm area decrease due to the vacuolization extension (PL. IV, fig.13) took place in some cells. Chloroplast structural modifications have been also noticed. Besides chloroplasts with a well represented thylakoidal system (Pl. IV, fig. 14), have been noticed ellipsoidal or spherical ones, with a thylakoidal system in different stages of disorganization and even vacuolization (Pl. IV, fig. 15).

The cell viability depends on the intact keeping of the cell membranes. The lipid bilayer represents a barrier through which the transmembrane transport control takes place (22). In an electroporation process a cell membrane electropermeability reversible induction took place. The degree of electropermeability is very important for the cell membrane stability or instability. Thus, the membranes or their lysis could be broken as a result of the chemical imbalance, caused by transport, through the transient pores (secondary process of electroporation). The chemical stress is associated with the molecular transport in the cell membrane. By electropermeability induction a membrane deformation and membrane material redistribution surface modifications show up. According to P. Stoeva quoted by Dumitrescu (11), by electrotransfection technique, biochemical reactions started, causing the appearance of cellular changes. The expression of these changes was strongly demonstrated in control plants in comparison with the transgenic plants. (10). The established differences in the reaction could be the influence of the stress conditions of co-cultivation, regeneration and selection of transgenic plants.

II. THE MORPHOMETRIC ANALYSIS OF THE *NICOTIANA TABACUM* L. (cv. *Xanthi*) DESCENDANTS CHLOROPLASTS

The morphometric analysis of leaf chloroplasts at *Nicotiana tabacum* L. (cv. *Xanthi*) descendants showed a decrease of the measured parameters as compared to the control (Table 1).

In the case of the descendants "D", grana nr./ chloroplast is between 14.62 (D3) and 21.51 (D2) while the descendants obtained by co-cultivation (23.05) showed a number close to that of the control (26). (Table I). The thylakoid number/grana of the descendants "D" is between 3.71(D3) – 5.94 (D1) and 9.21 for the control.

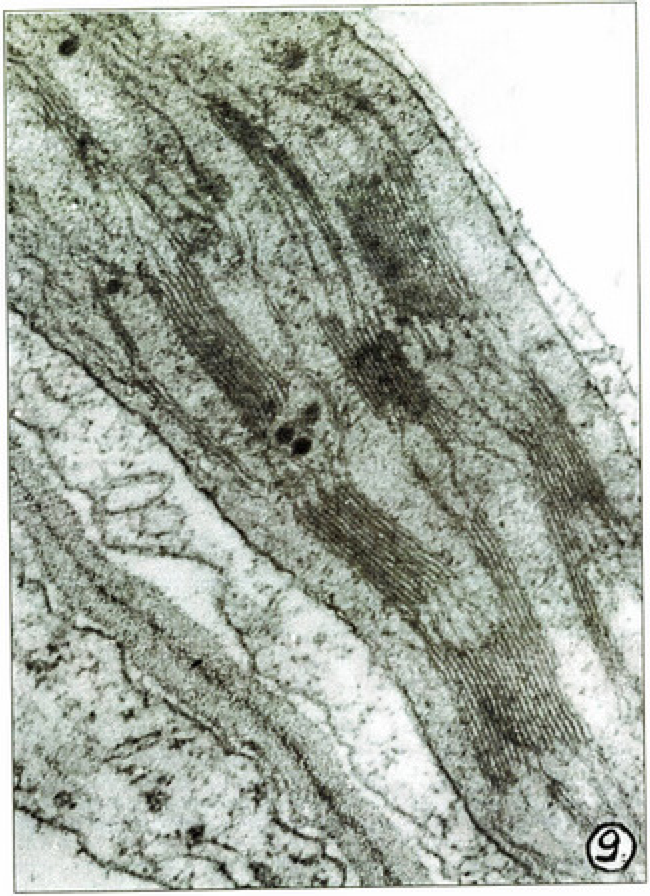
The morphometric analysis made evident differently organized chloroplasts, in the descendants of the transformed plants by co-cultivation with

PLATE I – The ultrastructural aspects of the mesophyll plant control.
Fig. 1 (× = 38356) – The ultrastructural aspects in the mesophyll of the descendants R_1, of the plants transformed by electrotransfection.
Fig. 2 (× = 20000), Fig. 3 (× = 27599), Fig. 4 (× = 32459).

PLATE I

PLATE II – The ultrastructural aspects in the mesophyll of the descendants R_1 of the plants transformed by electrotransfection.
Fig. 5 (× = 18000), Fig. 6 (× = 45000).
– The ultrastructural aspects in the mesophyll of the descendants R_2 of the plants transformed by electrotransfection.
Fig. 7 (× = 43765), Fig 8 (× = 13685).

PLATE II



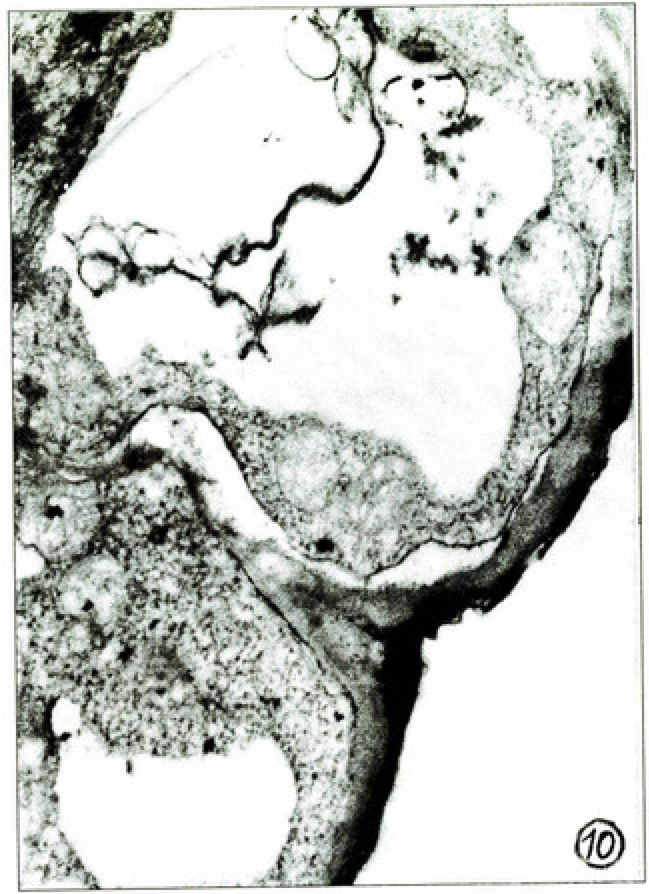


PLATE III – The ultrastructural aspects in the mesophyll of the descendants R_2, of the plants transformed by electrotransfection.
Fig. 9 (× = 11343) – The ultrastructural aspects in the mesophyll of the descendants R_3, of the plants transformed by electrotransfection.
Fig. 10 (× = 16798), Fig. 11 (× = 15300), Fig. 12 (× = 15600).

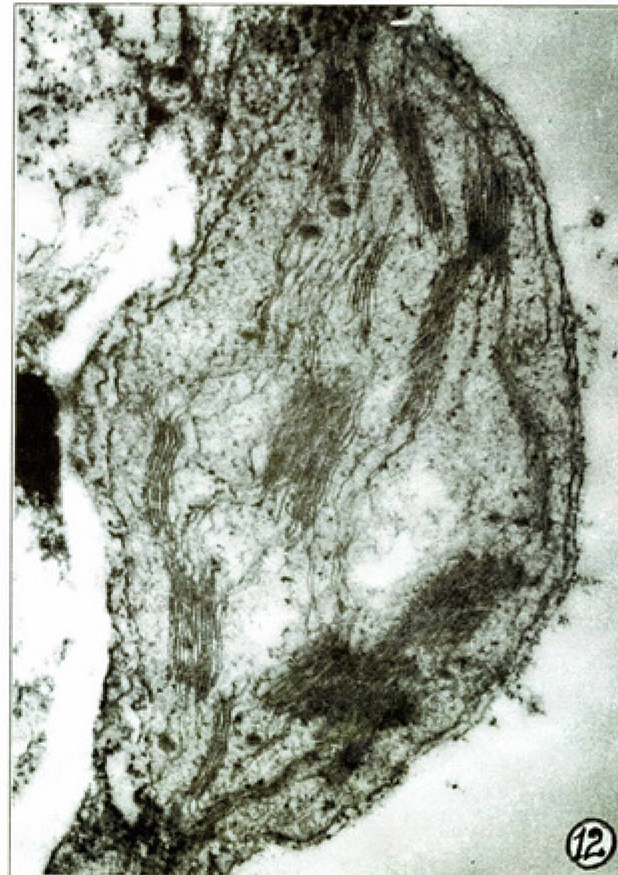


PLATE III

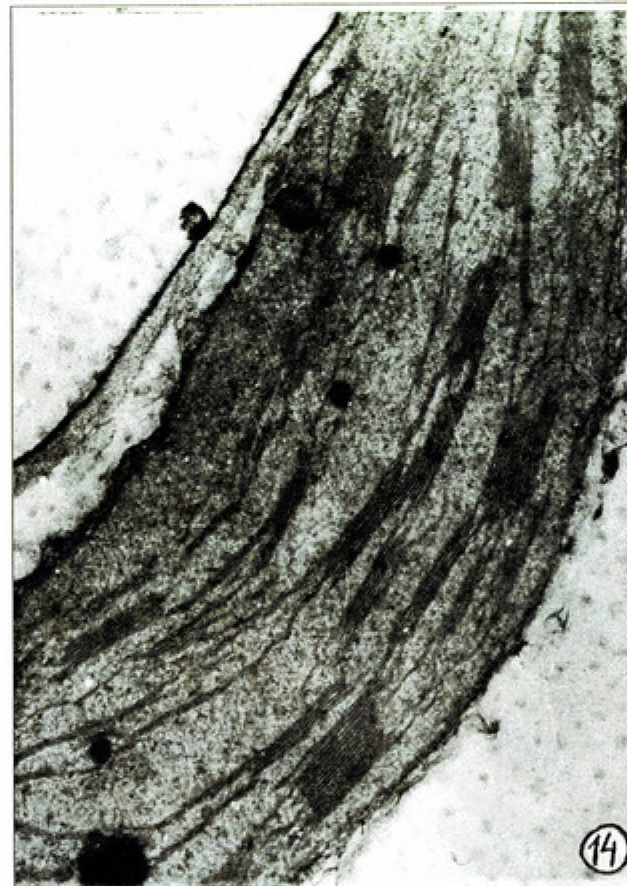


PLATE IV – The ultrastructural aspects in the mesophyll of the descendants of transformed plants by cocultivation with *Agrobacterium tumefaciens*.
Fig. 13 (× = 13200). Fig. 14 (× = 15230), Fig. 15 (× = 23200), Fig. 16 (× = 18200).

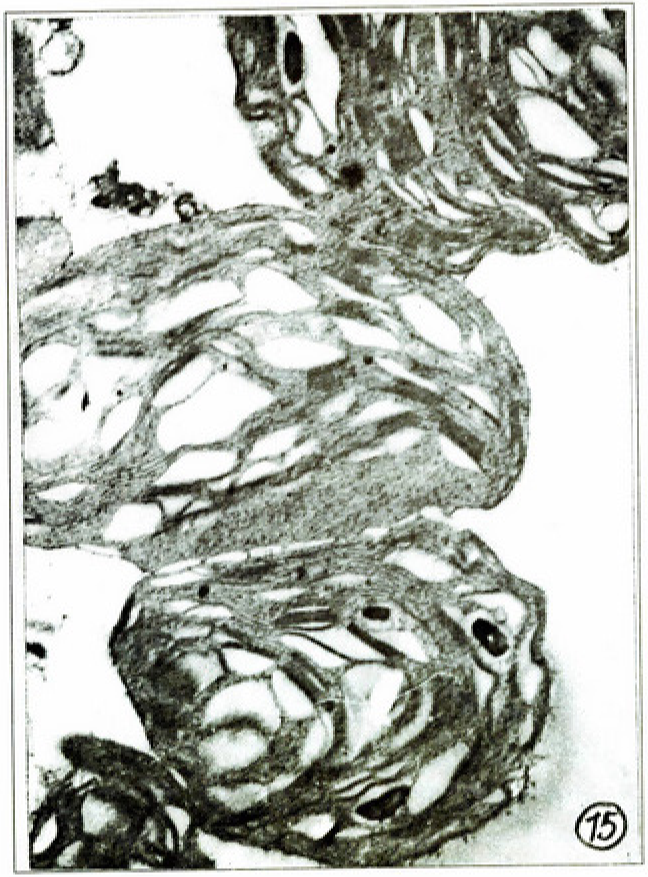


PLATE IV

Agrobacterium tumefaciens. The total thylakoid number/chloroplast, although decreased to the control, is increased to that of the descendants "D". These modifications were caused by the disorganization of the internal thylakoidal system, as a result of the starch inclusion big size, which caused breaking of the stromatic thylakoids, which link the grana.

Table 1

The morphometrical analysis of *Nicotiana tabacum* (cv. *Xanthi*) descendants chloroplasts

VARIANTS	G/CL	S	T/G.	S	TOTAL T/CL	S
Control	26 + 1.39	NS	9.26 + 1.05	NS	239.6 + 4.13	NS
D1	21.53 + 2.63	NS	5.94 + 1.04	NS	128.0 + 9.00	NS
D2	19.20 + 2.67	**	4.67 + 0.71	**	95.6 + 7.27	NS
D3	14.62 + 2.13	****	3.71 + 0.62	****	54.3 + 5.36	NS
Co-cultiv.	23.05 + 1.61	NS	5.85 + 0.68	*	134.9 + 5.24	NS

Legends: **D1**, **D2**, **D3** = electrotransfection descendants, **Co** = = cultivation descendants, **S** = significance
Biological significance: * $p < 0.05$, ** $p < 0.02$, *** $p < 0.01$, **** $p < 0.001$.

Crăciun C. *et al.* (8) estimated that the presence of specific genes causes modifications of the parenchymatic cell ultrastructure, mainly of their chloroplast. The histochemical studies made with Aurelia Brezeanu *et al.* (3) showed that the transient or stable gus gene expression was manifest on certain areas, by the appearance of blue spots, depending on the different susceptibility of a particular cell population for gene transfer by electroporation. According to Egnin M. (12), the lack or the slight expression of the gene gus in the electroporated tissue would be due to the gene methylation level. The studies made by Rodica Dumitrescu (10) on transformed plants of *Nicotiana tabacum* L. (cv.*Xanthi*), with p JIT 79 showed their phenotypical modifications caused by the different insertion of the gene. At the subcellular level, as a gene expression, modifications of cellular organelles membranes were noticed. The results of this study proved that the gene expressions are stable and manifest at the descendants' level of the transformed plants too.

III. THE STATISTICAL ANALYSIS

The statistical analysis showed the following:

Grana/chloroplasts number diminution to control regarding the descendants D3 of the transformed plants obtained by electrotransfection is very significant ($p < 0.001$).

Thylakoids/ grana number diminution regarding the descendants D2 of the transformed plants obtained by electrotransfection shows a high significance ($p < 0.01$) and at the descendants D3, the significance is higher ($p < 0.001$). Regarding the descendants of the transformed plants obtained by co-cultivation the thylakoids number/ grana diminution to control shows a very low significance ($p < 0.05$).

The total number of thylakoids/ chloroplasts does not show significant modifications (NS) to control.

CONCLUSIONS

1. The ultrastructural modifications noticed at the descendants of the transformed plants of *Nicotiana tabacum* L. (cv. *Xanthi*) are direct cause of the stress conditions during the electrotransfection, co-cultivation, regeneration and selection of the transgenic plants.

2. The ultrastructural modifications of the chloroplasts prove different variability and resistance to kanamycin of the descendants "D" of transformed plants.

In the case of the descendants of the transformed plants obtained by co-cultivation with *Agrobacterium tumefaciens* containing the plasmid p JIT 79, on the medium without kanamycin, chloroplasts of different organization have been noticed. The big number of thylakoid/chloroplasts (134.9), though smaller than control (239.60), is higher than that of the descendants "D".

REFERENCES

1. Badea Elena Marcela and Daniela Săndulescu, 2001, *Biotehnologii vegetale* , Fundaţia Biotech, Bucureşti, Romania.
2. Brezeanu Aurelia *et al.*, 1997, *Optimization of electrotransfection methods to Nicotiana tabacum L.* (cv. *Xanthi*) *intact tissue.* Proceedings of the Institute of Biology/Annual Scientific Session, Bucharest, **1**: 387–394.
3. Brezeanu Aurelia et al. 1999, *The expression of "gus" gene in Nicotiana tabacum L.* (cv.*Xanhti*), *electroporated intact seedling using p JIT – 79.* Proceedings of the Institute of Biology, **2**: 273–282.
4. Brainsberger E.S., Valcke R.L.M., Clijsters H.M., De Greef J., Van Onckelen H., 1991, *Effect of introduction of Agrobacterium tumefaciens T-dna, ipt gene in N.T.(L) Petit, Havana S.R.-1 plant cells.* Plant Cell Physiol., **32**:489–496.
5. Chan M.T., Chang H.H., Ho S.L. Tong W.F., Yu S.M., 1993, *Agrobacterium mediated production of transgenic rice plants expressing a chimerical-amylase promoter-glucuronidase gene.* Plant. Mol. Biol., **22**: 491–506.
6. Chowrira G.M., Akella V., Lurquin P.F., 1995, *Electroporation mediated gene transfer into intact nodal meristems in plants.* Mol. Biotechnol., **3**: 17 –23.
7. Cornea Călina Petruţa, I. Vătafu, Andreea Barbu, 1998, *Elemente de inginerie genetică.* Edit. All Educational, Bucureşti.

8. Crăciun C *et al.*, 1997, *The ms-15 and a genes effect on ultrastructural features of the leaf in Lycopersicum esculentum Mill.* Current Problems in Cellular and Molecular Biology, **II**: 470–477.
9. Dumitrescu Rodica, Aurelia Brezeanu, Dan Fologea, 1997, *The influence of the porator electric field on the ultrastructure and survival rate of the cells in Nicotiana tabacum* L. (cv. *Xanthi*). Current Problems in Cellular and Molecular Biology, **II:** 528–534.
10. Dumitrescu Rodica, 1998, *The electronomicroscopy analysis of the regenerants obtained by electrotransfection (p JIT 79) from Nicotiana tabacum L.* (cv. *Xanthi*). Current Problems in Cellular and Molecular Biology, **III**: 476–482.
11. Dumitrescu Rodica, 1999, *The quantitative modifications of chloroplasts in the cells of the Nicotiana tabacum* L.(cv. *Xanthi) plants with p JIT 79.* Current Problems in Cellular and Molecular Biology. Papers presented at 17th Annual Meeting of Romanian Society for Cell Biology, **IV**: 580–583.
12. Egnin M., Prakash C.S., 1990, *Genetic transformation and regeneration of transgenic sweet potato plants.* Hort. Sci., **30** (3) : 435.
13. Hall T.C. De Rose R.T., 1988, *Transformation of plant cells.* In: Bock. G.Marsh J.(ed). Applications of Plant Cell and Tissue Culture, 123–137, John Wiley & Sons, New York.
14. Hlikova E., 1994, *Protein paterns of transgenic soybean carrying different morphoregulatory genes from T-DNA p TIC 58.* Cell Biol. Int., **18**: 542.
15. Hlikova Elena, B.Obert, D.Filip, 1998, *Phenotypes of tobacco plants expressing genes for the synthesis of growth regulators.* Biol. Plantarum **41** (1) 25–37.
16. Klek H., Horsch R., ROGERS S., 1987, *Agrobacterium mediated plant transformation and its further applications in plant biology.* Annu. Rev. Plant Physiol., **38**: 763–764.
17. Kung S.D., Wu R.(ed), 1993, *Transgenic plants.* **2:** 27–31, Present Status and Social and Economic Impacts. Academic Press, San Diego.
18. Rovenska J. Kutacek M., Opatrny Z., Eder J., 1988, *Agrobacterium T-DNA encoded IAA biosynthesis.* In: Kutacek M., Bandurski R.S., Krekule J.(ed): *Physiology and Biochemistry of Auxin in Plant*, 409–414, Academia Prague.
19. Saker M.M. α T. Kuhne, 1998, *Production of transgenic kidney bean shoots by electroporation of intact cells*. Biologia Plantarum, **40**, (4) 507–514.
20. Tepfer D.,1984, *Transformation of several species of higher plants by Agrobacterium rhizogenes several transmission of transformed genotypic and phenotype.* Cell, **47**, 959–967.
21. Townsend J.A., Akella V., Lurquin P.F., 1995, *Electroporation mediated gene transfer into intact nodal meristems in plants.* Mol. Biotechnol., **3**: 17–23.
22. Ty Tsong, 1991, *Electroporation of cell membranes.* Biophysics J., **60**: 297–306.
23. Van Wert α Sunders A.I., 1991, *Electrofusion and electroporation of plants.* Plant Physiol., **99**: 365–367.
24. Zambyski P., 1988, *Basic processes underlying Agrobacterium-mdiated DNA transfer to plant cells.* Ann.Rev. Genet., **22**: 1–30.

Received March 15, 2001.

* *Institute of Biology, Romanian Academy*
Splaiul Independenței N° 296, Bucharest, Romania

THE INFLUENCE OF ACIDITY ON THE GROWTH AND ACTIVITY OF ACIDOPHILIC HETEROTROPHIC BACTERIA, ISOLATED FROM MINING EFFLUENTS

CARMEN CIŞMAŞIU

The growth and multiplication of acidophilic bacteria are the results of a series of co-ordinated metabolic reactions, which can take place only in certain acidic conditions. The influence of acidity, in various experimental conditions, on the growth and activity of acidophilic bacteria, isolated from the mines situated in two different areas of the country (Baia – Tulcea dept. and Roşia Poieni – Alba dept.) are studied in this work.

The experiments prove a strong relationship between the acidity of the medium and the behavior of the acidophilic heterotrophic bacteria. Heterotrophic bacteria, in optimal developmental conditions – at pH 2.5 and temperatures 28 °C – may have an active contribution to the processes of biosorption of metallic ions that have polluted different environments.

Key words: acidophilic bacteria, metallic ions, biosorption.

INTRODUCTION

The structural and physiological characterization of the species from microbial communities may lead to the discovery of new species of microorganisms, which could play an important role in bioremediation processes, due to their metabolic activity in environments polluted with metabolic ions (1, 6).

The development of the biotechnological processes, based on the activity of microorganisms, demonstrated their efficiency in recovering the metals from the sulfuric ores and mining effluents and in the bioremediation of environments polluted with residual inorganic substances, such as metallic ions and their compounds (4). The acidophilic heterotrophic bacteria may be used in biotechnological applications (biomining and biosorption) for recovering the metals, controlling the pollution and improvement of fossil fuel (6).

Bioremediation of environments polluted with metallic ions has gained importance, with the development of the mining industry, as an ecological process as compared to conventional methods. The adaptive ability of these microorganisms to various environmental conditions is very important for their use in the biosorption of metals from acidic mining effluents. This feature offers new perspectives for their use in commercial biotechnological processes (1, 3, 4, 7).

The study of mining effluents having high concentrations of metallic ions evidenced the presence of certain groups of microorganisms, adapted to the environmental conditions. Besides the *Thiobacillus* genus study, in the last years similar studies have been made with the acidophilic chemoorganotrophic bacteria, especially in the case of the *Acidiphilium* genus.

The preferences of acidophilic bacteria for the low pH level are the result of their adaptation to the environment under the influence of essential factors of their natural habitat. The temperature level plays an important role in the adaptation too (2, 3, 4).

The activity of bacteria is influenced by the acidity levels and temperature levels, although these microorganisms are adapted to grow in conditions of low pH. This paper presents our results about the influence of the acidity and temperature on the growth and activity of acidophilic heterotrophic bacteria, in various experimental conditions.

MATERIALS AND METHODS

1. PHYSIOLOGICAL GROUPS OF MICROORGANISMS

The populations of acidophilic heterotrophic bacteria that have been studied were isolated from acidic mining effluents from two different sites (Baia – Tulcea Dept. and Roşia Poieni – Alba Dept.), both containing high concentrations of metallic ions.

Heterotrophic acidophilic bacteria have been represented by populations of *Acidiphilium rubrum* (A_B), isolated from Baia and *Acidiphilium* sp. (A_{VS}), isolated from Roşia Poieni (plate 1). The strains belonging to *Acidiphilium* genus have been isolated on selective Manning liquid medium, having glucose as source of carbon and pH 2.5 (5).

2. EXPERIMENTAL CONDITIONS

The bacterial cultures have been incubated, for 20 days, in various experimental conditions:

- temperature: 28 °C and 37 °C;
- acidity: pH – 2.0, 2.5, 3.0, 3.5;
- static and stirring (150 rpm) conditions.

The bacterial cultures have been grown in 100 ml Erlenmeyer glasses containing 30 ml growth medium and 3 ml inoculum (bacterial culture 7 days old).

The growth of *Acidiphilium* cultures has been quantified by measuring the optical density (OD) at the wavelength of 660 nm (spectrophotometer UV), while the activity has been appreciated through the decrease of the pH (pH meter type PHM 26) of the culture solutions every 4 days.

RESULTS AND DISCUSSION

A. The influence of acidity on the growth and activity of acidophilic heterotrophic bacteria at 28 °C (Figs. 1 and 2).

The experiments have indicated that the influence of acidity on bacterial growth and activity is stronger for the population isolated from Baia than for the population isolated from Roşia Poieni, both in static and dynamic conditions. We have also found that the optimal pH for the bacterial growth and activity is 2.5, though a variation of pH within the range 2–3.5 keeps these two parameters within 70–80% of the maximum values (Fig. 1 and Fig. 2).

Comparative analysis indicates that these two populations of acidophilic heterotrophic bacteria have similar optimal conditions for growth and activity, but *Acidiphilium* sp. isolated from Baia is more efficient in sulfur oxidation evidenced by stronger decreasing the final pH value of culture medium than *Acidiphilium* sp. isolated from Rosia Poieni. We have also observed a direct correlation between the sulfur oxidation activity of the A_B population and growth evolution of bacterial culture (Fig. 1 and Fig. 2).

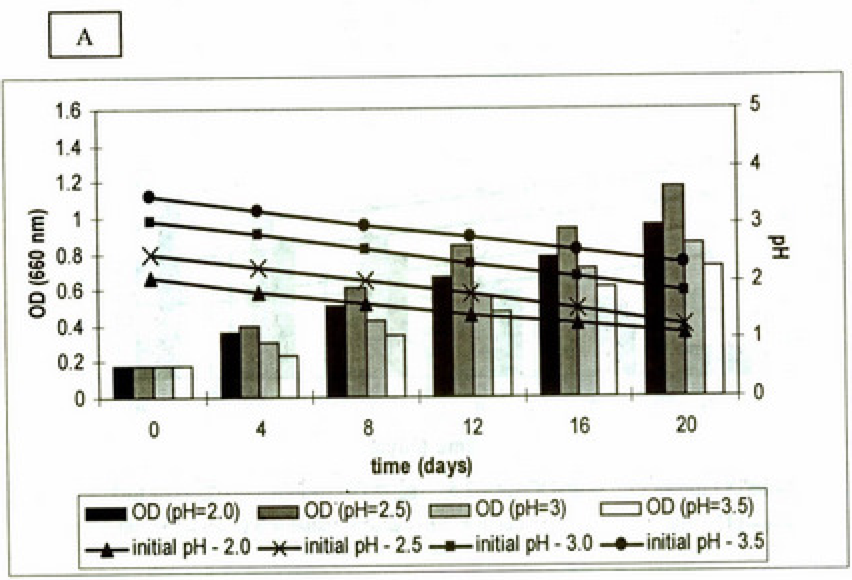


Fig. 1

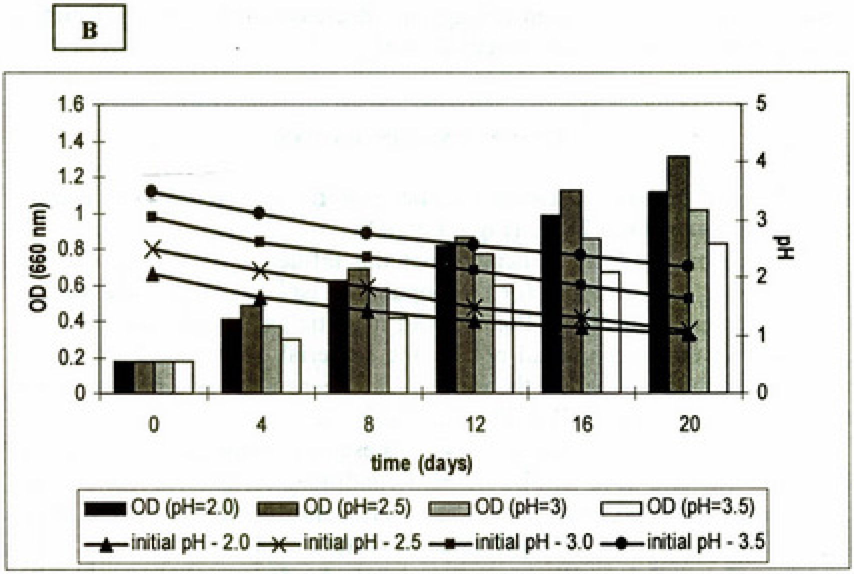


Fig. 1 – The dynamics of the growth and activity of acidophilic heterotrophic bacteria isolated from Baia at 28 °C:
A – static conditions; B – stirring conditions.

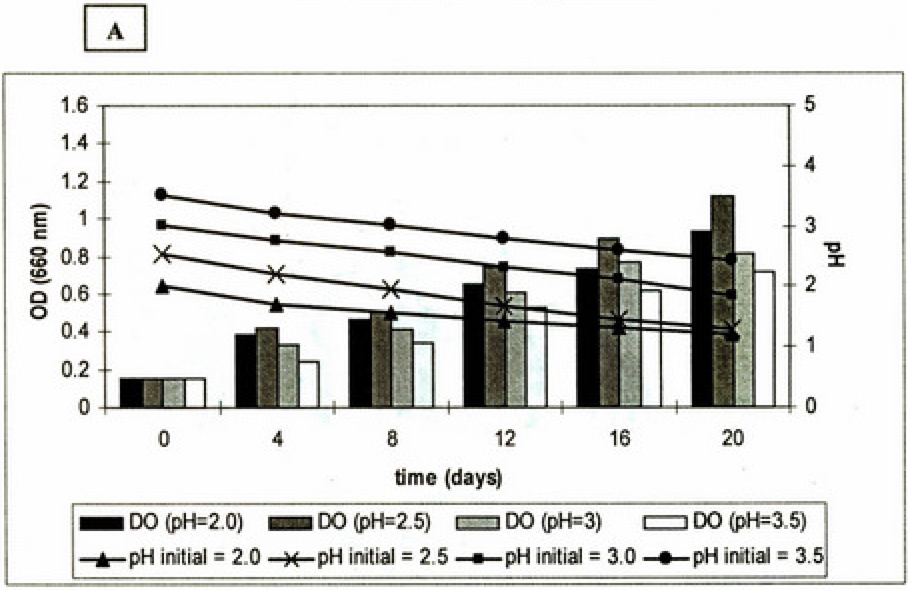


Fig. 2

B

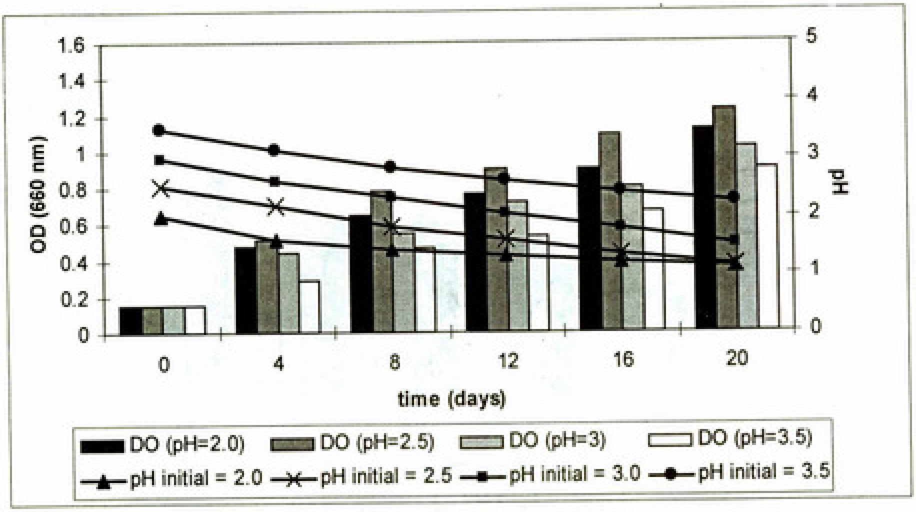


Fig. 2 – The dynamics of the growth and activity of acidophilic heterotrophic bacteria isolated from Valea Şesei at 28 °C: A – static conditions; B – stirring conditions.

B. The influence of acidity on the growth and metabolic activity of acidophilic heterotrophic bacteria at 37 °C (Figs. 3 and 4).

A

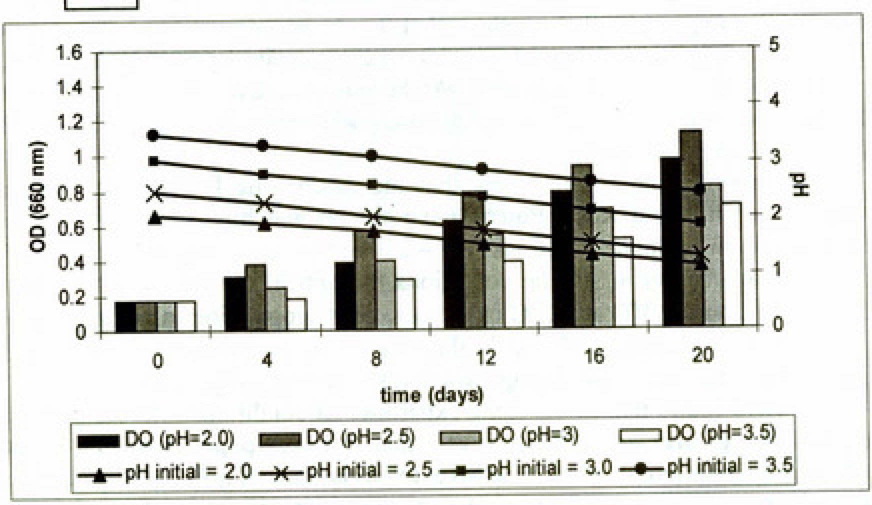


Fig. 3

B

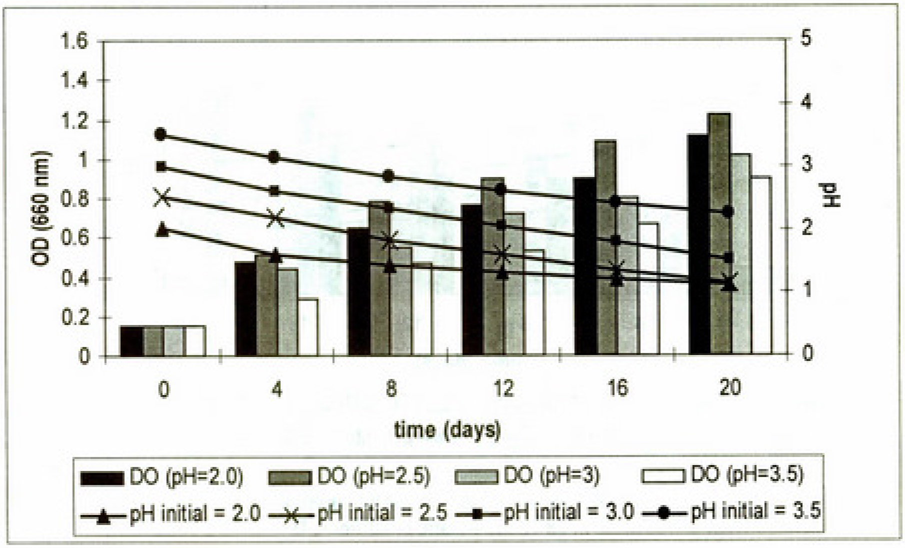


Fig. 3 – The dynamics of the growth and activity of acidophilic heterotrophic bacteria isolated from Baia at 37 °C:
A – static conditions; B – stirring conditions.

The comparative studies, on populations of acidophilic heterotrophic bacteria of *Acidiphilium* sp. (A_B and A_{VS}), show that the A_B population has a better growth (OD = 1.308) than the A_{VS} population (OD = 1.201) in similar conditions (pH – 2.5, temperature – 28 °C, stirring conditions). At the temperature of 37 °C there is a smaller difference between the growth levels of the two populations: A_B (OD = 1.233) and A_{VS} (OD = 1.163) (Figs. 3 and 4).

The experimental results show that the acidophilic heterotrophic bacteria, isolated from Baia and Roşia Poieni, have similar and higher growth levels and activity levels in optimal conditions (pH – 2.5 and temperature 28 °C). Thus, after 20 days of incubation in stirring conditions the optical density has the values of 1.233 at 37 °C and 1.302 at 28 °C in the case of A_B population in comparison with 1.163 at 37 °C and 1.201 at 28 °C in the case of A_{VS} population. The temperature has not influenced very much the growth and the activity (Figs. 1 and 3).

We have found that in the same experimental conditions , the A_B population shows higher growth and activity levels than the A_{VS} population. On the other hand, this population is more sensitive to variations of pH level, because the bacterial growth has a stronger decrease when the pH varies from 2.5 to 3.5 (Figs. 1–4).

Comparative results regarding the influence of acidity on bacterial growth and activity, in the same experimental conditions, indicate that the pH decreasing of culture medium is more accentuated during the A_B population growing. Thus, at 28 °C the pH varies from 2.50 to 1.07 in the case of A_B population as compared to variation from 2.5 to 1.16 in the case of A_{VS} population (Figs. 2 and 4). Increasing the temperature and the pH of the solution furthermore negatively influences the growth and metabolic activity of heterotrophic bacteria, in both static and stirring conditions. These results could help us to improve the growing conditions for the acidophilic heterotrophic bacteria cultures (Figs. 3 and 4).

The experiments comparing the effect of acidity on acidophilic heterotrophic bacteria, isolated from mining effluents and tailings from Baia and Roşia Poieni, offered strong evidence that the stirring conditions are more adequate for growing these bacterial strains in lab (Figs. 1 and 3).

The results of this study are consistent with previous observations about populations of chemolithotrophic sulfoxidizing bacteria, such as *Thiobacillus thiooxidans* and *Thiobacillus intermedius*, which illustrate a good growth level and sulfur oxidation level at a pH between 2.0 and 3.5.

CONCLUSIONS

1. There is a strong relationship between the acidity of the medium and the development of the acidophilic heterotrophic bacteria. Therefore, a high acidity of medium indicates a high developmental and activity level for these microorganisms.

2. The optimal temperature for the development and metabolic activity of acidophilic heteroptrophic bacteria is 28 °C. Our results show that a continuous exposure to higher temperatures, about 37 °C, may induce a certain degree of adaptation of bacterial populations to the thermal conditions.

3. The level of growth and metabolic activity of acidophilic heterotrophic bacteria is maximum at a pH – 2.5, though a variation of pH within the range 2.0 – 3.5 affects bacterial growth with less than 30%.

4. The experimental results indicate a higher activity level of bacterial populations, correlated with a higher growth level, in stirring conditions, as compared to static conditions. The acidophilic heterotrophic bacteria, in optimal developmental conditions, at pH – 2.5 and temperature – 28 °C, may have an active contribution to the processes of biosorption of metal ions in the presence of high concentrations of metallic ions.

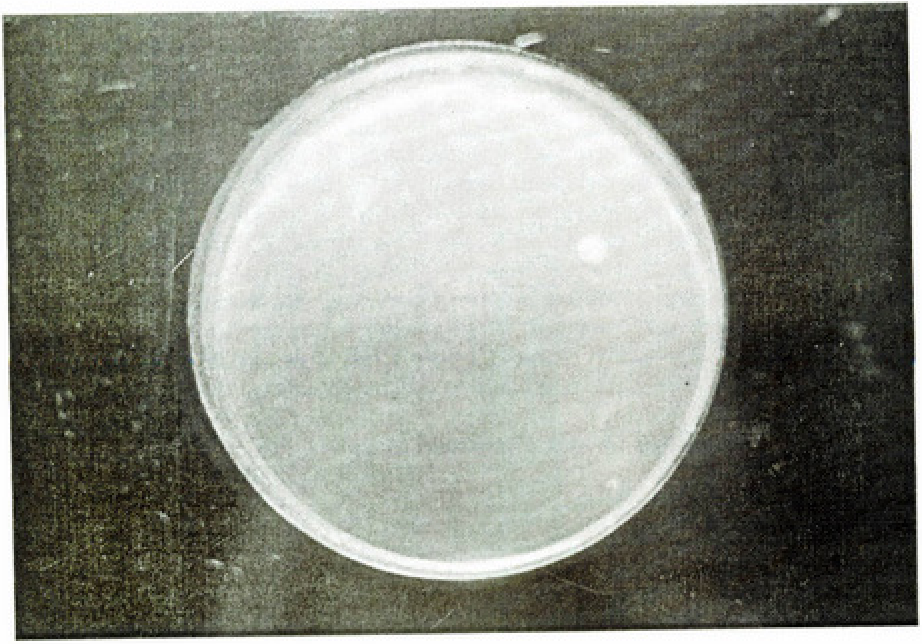


Plate 1 – *Acidiphilium* sp. (white colony) and yeast (yellow colony) isolated from mining effluents.

REFERENCES

1. Hallberg, K. B., Kolmert, A. K., Johnston, D. B., Williams, D. A., 1999, *A novel metabolite phenotype among acidophilic bacteria : aromatic degradation and the potential use of these organisms for the treatment of waste water containing organic and inorganic pollutants*. In : Proceedings of the International Biohydrometallurgy Symposium IBS'99, Madrid, Spain, June 20–23, edited by R. Amils and A. Ballester, Elsevier, 719–727.
2. Harrison, A. P. Jr., 1984, *The acidophilic thiobacili and other acidophilic bacteria that share their habitat*, Ann. Rev. Microbiol., 38: 265–292.
3. Hawksworth, D. L., 1992, *Biodiversity of microorganisms and its role in ecosystem function*. In: "Biodiversity and Global Change", Eds. Solbvig, O. T., Van Emden, H. M., Van Oordt, Paris, Int. Union of Biological Sciences, 88–93.
4. Johnson, D.B., 1999, *Importance of microbial ecology in the development of new mineral technologies*. In: "Biohydrometallurgy and the Environment Toward the Mining of the 21st Century" – Part B, Proceedings of the International Biohydrometallurgy Symposium IBS'99, edited by R. Amils and A. Ballester, Madrid, Spain, 645–656.
5. Karavaiko, G. I., 1988, *Methods of isolation, evaluation and studying of microorganisms*. In: "Biotechnology of Metals Manual" Center of Int. Projects GKNT, Moscow, Eds. G. I. Karaivaiko, G. Rossi, D. A. Agate, N. S. Groudev and A. J. Avakyan, 47–86.
6. Rawlings, D.E., 1999, *The molecular genetics of mesophilic, acidophilic, chemolithotrophic, iron – or sulfur – oxidizing microorganisms*. In: "Biohydrometallurgy and the Environment Toward the Mining of the 21st Century" – Part B, Proceedings of the International Biohydrometallurgy Symposium IBS'99, edited by R. Amils and A. Ballester, Madrid, Spain, 1–20.
7. Wichlacz, P. L., Unz, R. F., 1981, *Acidophilic heterotrophic bacteria of acidic mine waters*, Appl. Environm. Microbiol., 41: 1254–1261.

ASPECTS OF THE SAXICOLOUS VEGETATION IN THE COZIA MOUNTAINS

MARILENA ONETE

The paper presents an important saxicolous community growing on the rocks of the Cozia mountains described as *Daphno blagayanae – Silenetum lerchenfeldianae* ass. nova (holotypus hoc loco: Tab. 1, n° 7). Within the association, the Carpathic species account for 13.95%, while the Balkan-Carpathic species account for 34.88% (*Silene lerchenfeldiana* Baumg inclusively var. *lotriensis* (Grec.) Borza, *Daphne blagayana* Freyer, *Dianthus henteri* Heuff, *Symphyandra wanneri* (Roch.) Heuff.; *Viola declinata* W. et K., *Bruckenthalia spiculifolia* (Salisb.) Rchb., *Rosa coziae* Nyár., *Campanula kladniana* (Schur.) Witasek. Both the relict character of the associations and the overall vegetal landscape give a particular value to the National Park of Cozia Mountains, within the system of National Parks of the Romanian Carpathians.

Key words: tertiary relicts, saxicolous vegetal associations, floristic disjunction.

INTRODUCTION

With the high ridge of Făgăraş massif, to which they belong, as north boundary Cozia Mountains are located east of the Olt river gorges bearing the same name (N. Popescu, 1968). Consisting almost exclusively of injection gnais, known as "Cozia gnais", the rocky steep slopes of Cozia Ridge are particularly favorable to the development of saxicolous vegetation (V. Mihăilescu, 1963). However, due to the resistance of gnais to the process of cracking due to gelifraction, the floristic diversity of the coenoses established on these rocks is lower than that of the coenoses developing on calcareous rocks or even on mica shists.

Despite the remarkable interest of the flora and vegetation of Cozia massif, the floristic and phyto-sociological investigations are quite scarce (D. Tătăranu, 1949; E. I. Nyárády, 1955; G. Coldea şi A. Pop, 1988).

RESULTS AND DISCUSSION

The phyto-sociological investigations conducted in 2001, in the central area of Cozia massif, showed the relict characters of the vegetal groups populating the rocks. This relict character is accounted for by the location of the massif in a periglaciar area that was not affected by the selective effects of the glaciation from the Pleistocen.

REV. ROUM. BIOL.– BIOL. VÉGÉT., TOME 46, N° 1, P. 29–34, BUCAREST, 2001

The saxicolous coenoses from the central area of Cozia massif belong to the Dacian-Balkan association *Silenion lerchenfeldianae* Simon 1957, considered as a vicariant of the alliance *Androsacion multiflorae* Br.-Bl. In Br.-Bl. et Jenny 1926, which includes saxicolous coenoses growing on acid (siliceous) rocks in the mountains of Central Europe (Grabherr G., Mucina L., 1993). Both alliances belong to the order *Androsacetalia multiflorae* Br.-Bl. in Meier et Br.-Bl. 1934. Despite the fact that our study only investigated the rocks from the central area of the massif, we could determine a novel association within which *Daphne blagayana* plays the role of regional differentiation.

Ass. *Daphno blagayanae-Silenetum lerchenfeldianae* ass. nova.

Holotypus hoc loco: Tab. 1, n° 7.

The nomenclature type is described from a gnais rock formation located under Ciuha Mică peak at an altitude of 1625 m. The most representative species for recognizing the alliance *Silenion lerchenfeldianae* have been identified in the investigated coenosis: *Silene lerchenfeldiana* Baung (including var. *lotriensis* (Grec.) Borza), *Symphyandra wanneri* (Roch.) Heuff., *Dianthus henteri* Heuff. and *Daphne blagayana* Freyer.

The chorologic spectrum of analyzed coenoses shows that the Carpathian endemits (including the Dacian ones) account for 13.95%, while the geoelements with Carpathic-Balkan components account for 34.88 % (Fig. 1).

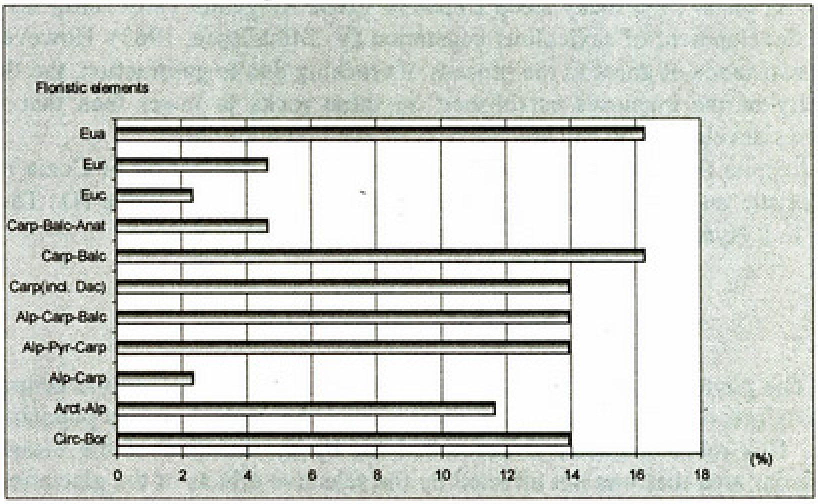


Fig. 1 – Spectrum of geoelements of ass. *Daphno blagayanae-Silenetum lerchenfeldianae*.

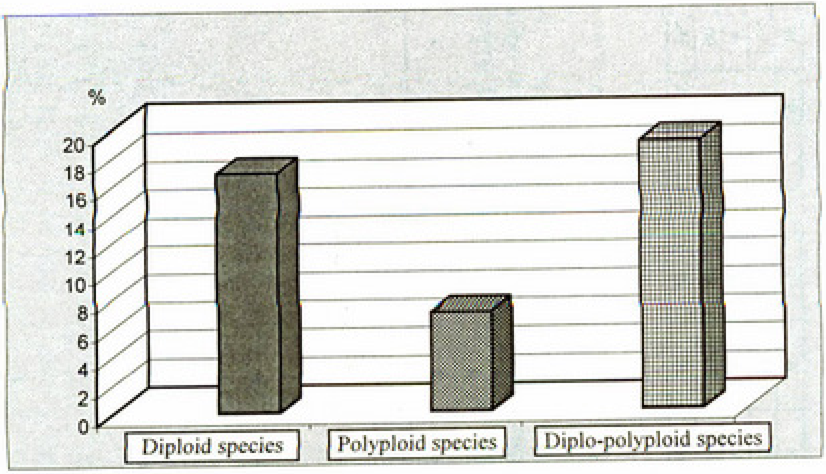


Fig. 2 – Chorologic spectrum of ass. *Daphno blagayanae-Silenetum lerchenfeldianae.*

Besides the components of the association, on the investigated territory we have also identified *Veronica bachofenii* Heuff., an old tertiary species whose disjunctive area is in a ratio of schisovariance with *Veronica dahurica* from the region of the Amur River in Oriental Asia (Meusel H., 1978).

Within the overall analyzed coenoses, the diploid species account for 67.27% while the diploidy index determined with the formula of Pignatti (1960) shows a value of 2.05. Both the chorologic spectrum and the diploidy index reveal the relict character of the association, which has an expressive regional color for the Southern Carpathians.

Code Corine: 62. 42

Code Eunice: H 3.1/P-62. 42

CONCLUSION

According to the information available so far, the most representative and numerous coenoses with *Silene lerchenfeldiana* from the Southern Carpathians grow in the Cozia mountains. The low possibilities for migration of the Dacian-Balkan components of these coenoses show their relict character. The coenopopulations of *Daphne blagayana*, integrated within different associations, still are quite spread in this massif.

Both the scientific importance of the saxicolous flora growing in the Cozia mountains and the overall landscape value of these mountains entrust a particular value to the National Park of the Cozia Mountains, within the National Park system.

As. *Daphno blagayanae – Silenetum lerchenfeldianae* ass. nova

Geoelements*	x**	2n	Relevé	1	2	3	4	5	6	7	8	9	10	K
			Altitude m s.m.	1570	1570	1550	1570	1625	1625	1625	1667	1450	1450	
			Exposition	E	E	E	V	V	V	V	E	E	V	
			Slope (°)	85	90	85	80	90	70	80	90	90	90	
			Recording area (m^2)	4	4	4	16	16	16	25	4	16	16	
	Silenion lerchenfeldianae													
Carp-Balc			*Silene lerchenfeldiana* Baum*** [incl. var *lotriensis* (Grec.) Borza]	3	2	3	2	1	1	2	2	+	2	V
Carp-Balc	9	18	(D) *Daphne blagayana* Freyer	1	.	+	+	.	.	+	.	+	.	III
Dac	15	30	*Dianthus henteri* Heuff.	.	.	.	+	1	1	1	1	+	+	IV
Carp-Balc-Anat	17	34	*Symphyandra wanneri* (Roch.) Heuff.	.	.	.	.	+	1	+	.	1	+	III
	Androsacetalia multiflorae													
Circ-Bor		144	*Asplenium septentrionale* (L.) Hoffm.	.	.	.	.	.	.	.	.	+	.	I
	Asplenietea trichomanis													
Eua	7	28	*Saxifraga paniculata* Miller	.	.	.	.	+	+	.	.	.	.	I
Circ-Bor		148	*Polypodium vulgare* L.	.	.	.	.	.	+	.	.	.	.	I
	Variae syntaxa													
Carp			*Festuca rupicola* Heuff. ssp. *saxatilis* (Schur.) Rauschert	.	+	+	+	+	1	3	2	2	+	V
Circ-Bor			*Deschampsia flexuosa* (L.) Trin.	+	+	+	.	+	.	.	.	+	+	III
Carp-Balc	7	28	*Thymus balcanus* Borb.	.	+	.	.	.	.	.	.	+	.	I
Euc	7	28	*Saxifraga cuneifolia* L.	+	.	.	.	.	+	.	.	.	.	I
Arct-Alp	5	30	*Juncus trifidus* L.	.	.	.	+	+	+	.	+	.	+	III

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Alp-Carp-Balc	18	36	*Cerastium eriophorum* Kit.	.	.	.	.	+	1	.	2	.	.	II
Carp-Alp			*Anthemis carpatica* Willd.	.	.	.	.	+	+	+	1	1	+	III
Circ-Bor	9	18	*Solidago virgaurea* L.	.	.	.	.	.	.	+	.	+	.	I
Alp-Carp-Balc			*Laserpitium krapfii* Crantz	.	.	.	.	.	.	+	.	+	.	I
Arct-Alp	11	22	*Sedum annuum* L.	.	.	.	.	.	.	+	.	.	+	I
Alp-Carp-Balc			*Phyteuma confusum* Kern.	.	.	.	.	+	+	.	+	.	+	II
Carp-Balc			*Viola declinata* W. et K.	.	.	.	.	.	.	.	+	+	.	I
Arct-Alp	9	27	*Hieracium alpinum* L.	.	.	.	.	+	.	.	.	.	+	I
Carp-Balc-Anat	9	27	*Bruckenthalia spiculifolia* (Salisb.) Rchb.	.	.	.	.	+	.	.	.	.	+	I
Carp-Balc-Alp			*Primula minima* L.	.	.	.	.	.	2	.	.	.	1	I

Species only in a relevé: *Allium ericetorum* Thore 7: +, *Calamagrostis arundinacea* (L.) Roth. 4: + , *Campanula kladniana* (Schur) Witasek 9: + , *Chamaecytisus ciliatus* (Wahl.) Rothm. 3: +, *Cortusa matthioli* L. 9: +, *Cotoneaster integerrima* Medicus 7: +, *Genista tinctoria* L. ssp. *oligosperma* (Andrae) Prod. 3: +, *Hypericum maculatum* Crantz 8: +, *Iris ruthenica* Ker-Gawler 9: +, *Jovibarba sobolifera* (J. Sims.) Opiz 8: +, *Juniperus communis* L. ssp. *alpina* (Suter) Èelak 10: +, *Potentilla aurea* L. ssp. *chrysocraspeda* Lehm. 8: +, *Rosa coziae* Nyár. 7: +, *Rumex arifolius* All. 8: +, *Silene pussila* W. et K. 9: +, *Trisetum macrotrichum* Hackel 8: +, *Vaccinium gaultheroides* Big. 10: +, *Vaccinium myrtillus* L. 10: +, *Vaccinium vitis-idaea* L. 10, +.

Site and data of relevé: 1, 2 – behind of Silvic department Călimănești house (14.08.2001); 3, 4 – under peak Durducului (15.08.2001); 5-7 – under peak Ciuha Mică (16.08.2001); 8, 9 – under peak Cozia; 10 – at rocks "La Foarfeci" (16.08.2001).

* Geoelements were established according to Popescu A., Sanda V., 1998.

** Basic number (x) and chromosome number (2n) were established according to Löve Á., Löve D., 1961 and Fedorov An. A., 1969.

*** Nomenclature of taxons was adopted according to "Standardliste der Farn-und Blüten-pflanzen der Budesrepublik Deutschland, 1993."

REFERENCES

1. * * *, 1993, Standardliste der Farn-und Blüten-pflanzen der Budesnepublik Deutschland Floristische Rundleriefe Beiheft, Verlag E. Goltze, Göttingen, **3**:1–478.
2. Boşcaiu N., 1971, *Flora şi vegetaţia Munţilor Ţarcu, Godeanu şi Cernei*, Edit. Academiei, Bucureşti.
3. Boşcaiu N., Täber F., 1985, Die zönologischen Verhältnisse der dazischen und dazisch-balkanischen Arten aus dem rumanischen Karpaten-raum, Vegetatio, **59**:185–192.
4. Coldea G., 1991, Prodrome des associations végétales des Carpates du sud-est (Carpates roumaines), Documents Phytosociologiques, **13**:317–539.
5. Coldea G., Pop Adriana, 1988, Cercetări fitocenologice în muntele Cozia, Contrib. Bot., Cluj-Napoca, 51–65.
6. Fedorov An. A., 1969, *Hromosomyie cisla ţsvetkovyh rostenii*, Nauk, Leningrad.
7. Grabherr G., Mucina L., 1993, *Die Pflanzengesellschaften Österreichs*, Teil II: *Natürliche waldfreie Vegetation*, Gustav Fischer Verlag, Jene-Stuttgart-New York.
8. Horvat I, 1938, Pflanzensoziologische Waldeduntersuchungen in Kroatien, Glas šum. Pok., Zageb, 6: 127–279.
9. Horvat I., Glavač V., Ellenberg H., 1974, *Vegetation Südesteuropas*, Gustav Fischer Verlag, Jena.
10. Löve Á., Löve D., 1961, Chromosome numbers of Central and Northwest European plant species, Opera Botanica (Lund), **5**:1–581.
11. Meusel H., 1978, *Vergleichende Chorologie der Zentral-europäischen Flora*, Band I-II, Gustav Fischer Verlag, Jena.
12. Mihăilescu V., 1963, *Carpaţii sud-estici de pe teritoriul R.P.Române*, Edit. Ştiinţifică, Bucureşti.
13. Nyárády E. I., 1955, *Vegetaţia muntelui Cozia şi câteva plante noi pentru flora Olteniei, Moldovei şi Transilvaniei,* Bul. Şt., Secţ. şt. biol., agron., geol. geogr., VII (2):209–246.
14. Pignatti S., 1960, Il significo delle specie poliplidi nelle associazioni vegetali, Atti Inst. Veneto si Sc., Lett. Arti, 118:75–98.
15. Popescu A., Sanda V., 1998, *Conspectul Florei Cormofitelor spontane din România*, Acta Botanica Horti Bucurestiensis, 3–336.
16. Popescu N., 1968, *Munţii Căpăţinii şi Coziei*, Edit. Cons. Naţ. Ed. Fizică şi Sport, Bucureşti.

Received May 30, 2001.

Institute of Biology
Spl. Independenţei 296
79651 Bucharest

THE VEGETATION OF SAINT ANNA ROCKS OF PRAHOVEAN SIDE OF BUCEGI MASSIF

CLAUDIA D. BIȚĂ

The paper presents the coenosis of the following associations: *Asperulo capitatae-Seslerietum rigidae* (Zólyómy 1939) Coldea 1991, *Asplenio-Cystopteridetum fragilis* Obed.(1939) 1949, *Dryopteridetum robertianae* (Kuhn 1937) Tx. 1937 (*Gymnocarpietum robertianae Boşcaiu 1971 nom. mut. propos*), *Thymetum comosi* Pop et Hodişan 1963 from the Saint Ana Rocks (Bucegi Mountains), which are characteristic of the debris and rocky vegetation.

Key words: rocky vegetation, plants community.

INTRODUCTION

The Saint Anna Rocks are placed over Peleş Valley, between Sinaia and Poiana Țapului, and due to their picturesque landscape they are a touristic attraction.

Although they are near Sinaia, their flora and vegetation are less investigated. The only scientific paper about the vegetal communities of these rocks was published by V. Sanda, A. Popescu and Gabriela Fişteag (10).

However, for their picturesque interest and for their rich flora, these rocks had been put in Prahovean Massif of Bucegi protected area. As a part of National Park of Bucegi, this massif had been protected on 3740.0 ha area through the Decision of Council of Ministers no. 965/1945.

The calcareous rocks form the klipps of Sf. Ana and they are populated by the vestiges of some eastern massifs of tectonic contact between Absian Barremian in wilt-flysch facies and Absian Barremian of some synchrony and heterotic flysch areas from superior Jurassic period.

The investigations from summer 2000 emphasize four communities, some of them had been published by V. Sanda, A. Popescu and Gabriela Fişteag (10).

MATERIAL AND METHODS

The studies had been effectuated based on field observations, on the consultation of literature and of the herbarium from the Institute of Biology. The main method of study was the most frequent and appreciable of Montpellier School, Braun-Blanquet method. We composed the phytocoenotical tables for each association and we framed it in the appropriate class.

REV. ROUM. BIOL.– BIOL. VÉGÉT., TOME 46, N° 1, P. 35–46, BUCAREST, 2001

RESULTS AND DISCUSSIONS

Syntaxonomical system of investigated plant associations:

Cl. *Asplenietea rupestria* Br.-Bl. 1934
Ord. *Potentilletalia caulescentis* Br.-Bl. 1926
All Cystopteridion (Nordh.) J.L. Rich. 1972
As. *Asplenio-Cystopteridetum fragilis* Obed.(1939) 1949
Cl. *Thlaspietea rotundifolii* Br.-Bl. 1926
Ord. *Thlaspietalia rotundifolii* Br.-Bl. 1926
All. *Stipion calamagrostis* Br.-Bl. 1918 (*Achnatherion calamagrostis* Boşcaiu 1971 nom. mut. propos)
As. *Dryopteridetum robertianae* (Kuhn 1937) Tx. 1937 (*Gymnocarpietum robertianae* Boşcaiu 1971 nom. mut. propos)
As. *Thymetum comosi* Pop et Hodişan 1963
Cl. *Seslerietea albicantis* Br.-Bl. 1926 em Oberd. 1978
Ord. *Seslerietalia albicantis* Br.-Bl. 1926
All. *Seslerion rigidae* Zólyómy 1939
As. *Asperulo capitatae-Seslerietum rigidae* (Zólyómy 1939) Coldea 1991

The spreading character of saxicolous coenoses reduces both the intrapopulational pressure and the interspecific pressure and, in this way, it favours the lasting of some old relict species. The primary colonizations of these rocky areas constitute the *Asplenio-Cystopteridetum* association with regional subassociation *campanuletosum carpeticae* (Sanda, Popescu et Doltu 1977). We emphasize the presence of differential species own to the Curvature of Carpathians (*Campanula carpatica, Saxifraga cuneifolia, Valeriana montana*). Due to humic accumulations from fissure of rocks, the transgressive species of *Seslerietea albicantis* class have been infiltrated. (Table 1).

These species take the places of the association with groups of this class in proportion as their domination increases.

We confirm the presence of *Melampyrum saxosum* species in the flora of Bucegi Massif. This plant species is rare for the Romanian flora and it was noticed for the first time by Grecescu (1898) in these mountains, too.

The mobile or various conditions of fastening calcarous debris are populated by coenoses of *Thlaspietea rotundifolii* Br.-Bl. 1926.

Although these coenoses have a breaking new character due to primary colonisations, they cannot develop continuity as long as the debris remain standing into active mobility condition to permit other populations colonization. For this reason, M. Guinochet (1955) called these colonizations as the permanent pioneer stages.

These debries are populated by *Gymnocarpium robertianum* in association with various nemoral species from the beech forest in shady areas. The most prevalent among these sciaphilous communites is *Dryopteridetum robertianae* (Kuhn 1937) Tx. 1937 (*Gymnocarpietum robertianae* Boşcaiu 1971 nom. mut. propos).

Table 1

Asplenio -Cystopteridetum fragilis Oberd. (1939) 1949

Bioform	Geoelement	x	2n	Number of survey. Exposure Slope (degrees) Area (sq.m.)	1 East 45 4	2 East 45 4	3 East 30 4	4 East 85 4	5 East 85 4	K
1	2	3	4	5	6	7	8	9	10	11
				Cystopteridion et Potentilletalia						
H	Circ	9	72	*Asplenium viride*	2	2	+	1	-	IV
H	Cosm	7	168	*Cystopteris fragilis*	+	-	+	2	+	IV
Ch	Euc	7	28	*Saxifraga cuneifolia*	+	-	+	-	+	III
H	Euc	12	24	*Moehringia muscosa*	-	-	+	-	1	I
Th	Med	7	14	*Sedum hispanicum*	+	-	-	-	-	I
H	Carp	17	34	*Campanula carpatica*	+	+	-	-	+	III
				Asplenietalia						
H	Cosm	9	72	*Asplenium trichomonas*	+	-	-	+	1	III
G	Circ	37	148	*Polypodium vulgare*	-	-	-	-	+	I
H	Circ	7	28	*Poa nemoralis*	3	-	2	3	-	III
H	Euc	8	16	*Valeriana tripteris*	-	+	-	-	-	I
				Variae syntaxa						
H	Euc	9	18	*Veronica urticifolia*	+	-	+	-	-	II
G	Euc	11	44	*Galium schultesii*	+	-	-	-	-	I

Table 1 (continued)

1	2	3	4	5	6	7	8	9	10	11
Th	Med	-	-	*Cnidium silaifolium*	+	+	-	-	-	II
Th	Cosm	8	64	*Geranium robertianum*	+	-	+	+	-	III
H	Balc-Carp	9	18	*Hieracium rotundatum*	+	-	-	-	-	I
H	Eua	7	14	*Fragaria vesca*	-	+	-	-	-	I
H	Eur	8	32	*Valeriana montana*	-	-	+	-	-	I
H	Euc	8	56	*Valeriana sambucifolia*	+	-	+	+	+	IV
H	Eua	8	16	*Actaea spicata*	-	-	+	-	-	I
H	Eur	7	28	*Hordelymus europaeus*	-	-	-	+	-	I
G	Cosm	15	90	*Botrychium lunaria*	-	+	+	-	-	II
H	Eua	8	16	*Campanula persicifolia*	+	-	+	-	+	III

The place and date of surveys: Sinaia, Saint Anna, 09.07.2000

Table 2

Dryopterideturn robertianae (Kuhn 1937) Tx.1937

Bioform	Geoelement	x	2n	Number of survey Exposure Slope (degrees) Area (sq.m.)	1 East 45 4	2 East 45 4	3 East 45 4	4 East 45 4	5 East 45 4	K
1	2	3	4	5	6	7	8	9	10	11
				Stipion calamagrostis et Thlaspietalio						

Table 2 (continued)

1	2	3	4	5	6	7	8	9	10	11
G	Circ	10	160	*Gymnocarpium robertianum*	3	3	3	3	3	V
Th	Alp	10	20	*Senecio squalidus*	+	-	-	-	-	II
Th	Cosm	8	64	*Geranium robertianum*	+	-	-	-	-	I
				Asplenietea						
H	Circ	9	72	*Asplenium viride*	-	+	-	-	-	I
H	Cosm	7	168	*Cystopteris fragilis*	+	-	-	-	-	I
Ch	Euc	7	28	*Saxifraga cuneifolia*	-	-	-	-	+	I
H	Circ	7	28	*Poa nemoralis*	+	-	+	+	-	III
				Variae syntaxa						
H	Eur	8	32	*Valeriana montana*	2	2	3	+	2	V
H	Eua	7	14	*Fragaria vesca*	+	+	-	-	-	II
Ch	Eur	9	18	*Euphorbia amygdaloides*	+	-	+	+	-	III
H	Euc	9	18	*Veronica urticifolia*	+	+	+	+	-	IV
G	Eua	10	60	*Petasites albus*	+	-	-	-	-	I
Th	Carp	-	-	*Melampyrum saxosum*	1	1	2	-	+	IV
H	Eur	6	12	*Luzula luzuloides ssp. cuprina*	+	+	-	-	+	III
H	Euc	8	56	*Valeriana sambucifolia*	+	-	+	-	-	II
H	Eua	8	16	*Campanula persicifolia*	+	-	-	-	-	I
H	Euc	7	14	*Astrantia major*	+	-	-	-	-	I
G	Eua	10	40	*Epipactis atrorubens*	+	+	-	1	-	III
H	Euc	17	34	*Cirsium erysithales*	+	-	+	+	-	III

Table 2 (continued)

1	2	3	4	5	6	7	8	9	10	11
Th	Med	-	-	*Cnidium silaifolia*	+	-	-	-	+	I
H	Carp	9	18	*Hieracium rotundatum*	+	-	-	+	1	III
H	Alp	9	54	*Achillea stricta*	-	+	-	-	-	I
H	Alp	9	54	*Achillea stricta*	-	+	-	-	-	I
Th	Eur	8	16	*Linum catharticum*	-	+	-	-	-	I
G	Euc	11	44	*Galium schultesii*	-	+	-	-	-	I
H	Eur	7	42	*Mercurialis perennis*	-	-	-	2	-	I

The place and date of surveys: Sinaia, Saint Anna, 09.07.2000

Table 3

Thymetum comosi Pop et Hodisan 1963

Bioform	**Geoelement**	**x**	**2n**	**Number of survey** **Exposure** **Slope (degrees)** **Area (sq.m.)**	**1** **East** **45** **4**	**2** **East** **45** **4**	**3** **East** **45** **25**	**4** **East** **45** **25**	**5** **East** **45** **25**	**K**
1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**
				Stipion calamagrostis et Thlaspietalia						
Ch	Carp	7	28	*Thymus comosus*	2	3	3	1	3	V
H	Eur	11	44	*Galium album*	1	3	3	2	2	V

Table 3 (continued)

1	2	3	4	5	6	7	8	9	10	11
H	Eur	11	22	*Vincetoxicum hirudinaria*	3	3	2	2	+	V
H	Eua	15	30	*Origanum vulgare*	+	1	+	-	-	III
				Variae syntaxa						
H	Eua	7	14	*Sedum maximum*	+	-	+	-	-	II
H	Eur	8	16	*Scabiosa columbaria*	+	+	+	+	-	IV
H	Euc	6	24	*Coronilla varia*	+	+	+	+	+	V
H	Euc	17	34	*Cirsium erysithales*	+	+	+	+	-	IV
H	Eua	8	16	*Salvia glutinosa*	+	-	-	-	-	I
H	Eua	7	28	*Bromus benekenii*	+	-	+	-	-	II
H	Alp	-	-	*Bupleurum falcatum ssp cernuum*	+	+	+	-	-	III
Th	Med	-	-	*Cnidium silaifolium*	2	+	-	+	+	IV
H	Euc	11	44	*Galium schultesii*	+	-	-	-	-	I
H	Carp	-	-	*Melampyrum saxosum*	+	-	-	-	-	I
H	Euc	9	36	*Lamiastrum galeobdolon*	+	-	-	-	-	I
H	Euc	9	18	*Veronica urticifolia*	+	-	+	-	-	II
H	Carp	11	22	*Primula elatior ssp. carpatica*	+	-	-	-	-	I
H	Carp	-	-	*Trisetum fuscum*	-	+	-	-	-	I
Th	Cosm	8	64	*Geranium robertianum*	-	+	-	-	-	I
Ch	Eur	9	18	*Euphorbia amigdaloides*	-	+	-	-	-	I

Table 3 (continued)

1	2	3	4	5	6	7	8	9	10	11
H	Eua	8	32	*Hypericum perforatum*	-	-	-	+	-	I
H	Euc	9	18	*Melica ciliata*	-	-	-	-	+	I

The place and date of surveys: Sinaia, Saint Anna, 09.07.2000

Table 4

Asperulo capitatae-Seslerietum rigidae (Zòlyòmy 1939) Coldea 1991

Bioform	**Geoelement**	**x**	**2n**	**Number of survey**	**1**	**2**	**3**	**4**	**5**	**K**
				Exposure	**East**	**East**	**East**	**East**	**East**	
				Slope (degrees)	**70**	**80**	**70**	**75**	**75**	
				Area (m.p.)	**16**	**4**	**4**	**16**	**16**	
1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**
				Seslerion rigidae						
H	Carp	11	22	*Asperula capitata*	1	+	+	1	+	V
H	Carp	-	-	*Sesleria rigida*	4	2	4	3	2	V
Ch	Carp	7	28	*Thymus comosus*	2	-	2	+	+	IV
H	Alp	-	-	*Bupleurum falcatum ssp. cernuum*	+	+	-	+	+	IV
H	Carp	15	60	*Dianthus spiculifolius*	2	2	+	+	2	V
TH	Carp	-	-	*Erysimum witmannii ssp. transsilvanicum*	-	+	-	-	+	I

Table 4 (continued)

1	2	3	4	5	6	7	8	9	10	11
				Seslerietalia						
H	Euc	9	18	*Biscutella laevigata*	+	+	-	-	+	III
H	Euc	8	24	*Minuartia verna*	+	+	+	+	+	V
H	Euc	9	18	*Acinos alpinum*	-	-	+	-	+	II
Ch	Alp	12	24	*Helianthemum alpestre*	-	-	+	-	-	I
H	Alp	17	34	*Polygala alpestris*	-	-	+	-	-	I
H	Eur	8	16	*Scabiosa columbaria*	-	-	-	-	+	I
				Asplenietalia						
H	Circ	9	72	*Asplenium ruta-muraria*	+	2	-	+	+	IV
H	Circ	7	28	*Poa nemoralis*	-	+	+	2	+	IV
Ch	Eua	7	28	*Saxifraga paniculata*	-	-	-	+	-	I
H	Eua	7	14	*Sedum maximum*	+	-	-	-	-	II
				Variae syntaxa						
Th	Med	-	-	*Cnidium silaifolium*	+	+	+	+	+	V
H	Eur	11	22	*Vincetoxicum hirundinaria*	+	-	+	-	-	II
H	Euc	9	18	*Melica ciliata*	+	-	-	-	-	I
Th	Med	7	14	*Sedum hispanicum*	-	-	+	-	-	I
G	Eua	-	-	*Orobanche alba*	-	-	-	-	+	I
H	Eur	11	44	*Galium album*	+	-	-	-	+	II

The place and date of surveys: Sinaia, Saint Anna, 09.07.2000

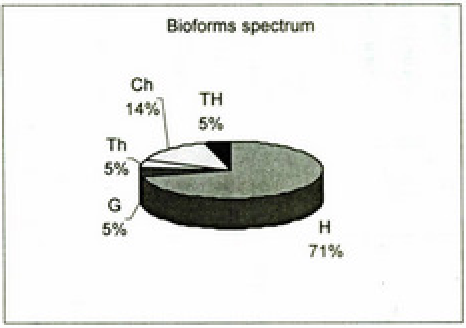
We emphasize the presence of the *Melampyrum saxosum* dissemination (Table 2).

In sunny ecotopes, on mobile or fixed debries the *Thymetum comosi* Pop et Hodişan 1963 association grows up. This association is endemic and it has a large dispersion in south-eastern Carpathians (Table 3).

As a result of these debris stabilization humic accumulations are produced that lead to some coenoses from the class *Seslerietea albicantis* Br.-Bl. 1926 em Oberd. 1978.

On the different state of erosion and chemical alteration rocks that favor rich in skeleton fragments rendzinic accumulations grow up communities from the alliance *Seslerion rigidae* Zólyómy 1939. In the studied territory, on the rocks the association *Asperulo capitatae-Seslerietum rigidae* Zólyómy 1939 is dominated (Table 4).

The diploid statute of *Biscutella laevigata* (I. Tarnavschi 1948) disseminations and the presence of other diploid saxicolous species indicate these rocks were not under the glaciers but in as periglaciar area during the last glaciar period. At that time, the physical and chemical structural processes were very active. This was the way that favoured the lasting of a remarkable contingent of saxicolous relicts.



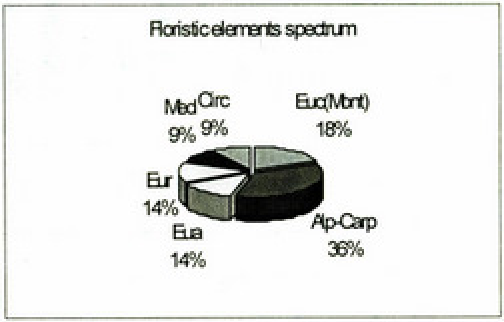
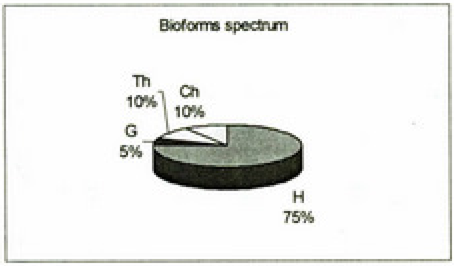


Fig. 1 – Ass. *Asplenio Cystopteridetum fragilis.*



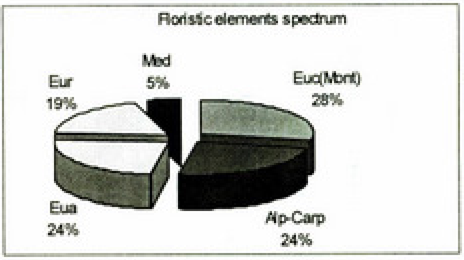


Fig. 2 – Ass. *Dryopteridetum robertianae.*

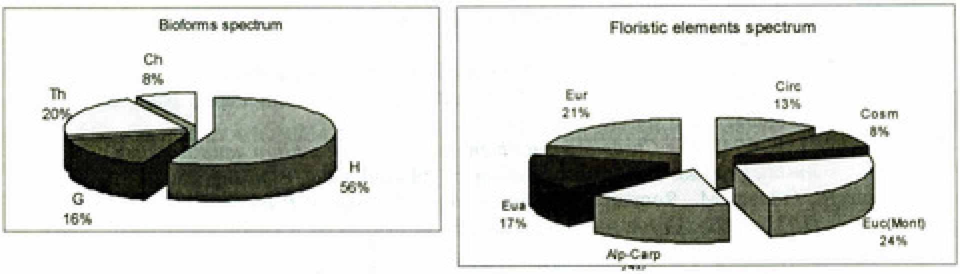


Fig. 3 – Ass. *Thymetum comosi.*

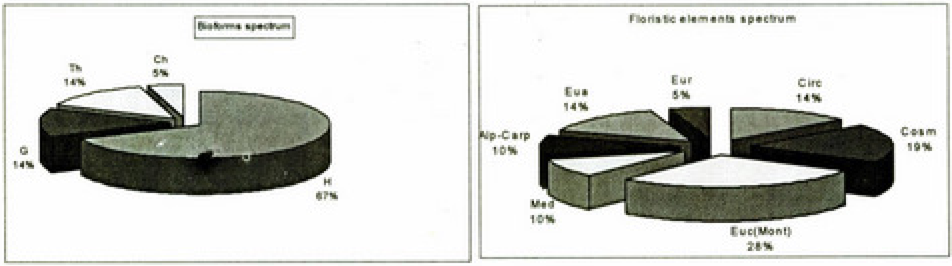


Fig. 4 – Ass. *Asplenio capitatae - Seslerietum rigide.*

CONCLUSIONS

The presence of calcareous rocks within the reservation Saint Anna Rocks favorizes the survey of many relict phytocoenoses and preserves the diversity of flora and fauna in the reservation.

A long-time monitoring has to be implemented to observe the habitat evolution and allow the intervention in case of need.

REFERENCES

1. Beldie A., 1956, *Rezervaţia naturală Bucegi*, Ocrot. Nat., **56**(2):31.
2. Beldie A., 1967, *Flora şi vegetaţia munţilor Bucegi*. Edit. Acad. R.P.R. Bucureşti.
3. Boşcaiu N., 1971, *Flora şi vegetaţia munţilor Ţarcu, Godeanu şi Cernei*. Acad. R.S.R. Bucureşti.
4. Coldea G., 1991, *Prodrome des Associations Végétales des Carpates du Sud-Est (Carpates Roumaines)*, Documents Phytosociologiques, vol. XIII, Università degli Studi, Camerino.
5. Guinochet M., 1954, *Sur les fondements statistiques de la phytosociologie et quelques-unes des leurs conséquences,* Aktuelle Probleme der Pflanzensoziologie, Veröff. d. Geobot. Inst. Rübel in Zürich, fasc. 29, Bern.
6. Mihăilescu S., 2001, *Flora şi vegetaţia masivului Piatra Craiului*, Edit. Vergiliu, Bucureşti.
7. Mutihac V., Ionesi L., 1974, *Geologia României*, Ed. Tehnică, Bucureşti.

8. Popescu A., Sanda,V., 1998, *Conspectul florei cormofitelor spontane din Romania*. Acta Bot. Horti Buc., 336 p.
9. Puşcaru D. & al., 1956, *Păşunile alpine din munţii Bucegi*. Edit. Acad. RPR, Bucureşti.
10. Sanda V., Popescu A., Fişteag G., 1996, Aspecte de vegetaţie din Parcul Naţional Bucegi. Naturalia. Studii şi Cerc. **2–3**: 193–204.
11. Tarnavschi I. T., 1948, *Die Chromosomenzahlen der Anthophyten-Flora von Rumänien mit einem Ausblick auf das Polyploidie Problem*. Suppl., Bul. Grad. Bot. Cluj, 28.
12. Toniuc N., Oltean M., Romanca G., Zamfir M., 1992, *List of Protected Areas in Romania (1932–1991)*, Ocrot. nat. **36**(1): 23–33.

Received April 17, 2001.

Institute of Biology
Spl. Independenţei 296,
E-mail: claudia.bita@ibiol.ro

THE IMPACT OF THE URBAN WASTE WATERS OF THE PITEŞTI TOWN UPON MICROBIAL COMMUNITIES FROM GOLEŞTI LAKE (ARGEŞ RIVER) IN 1999–2000 PERIOD

DOINA IONICĂ, CRISTINA SANDU, SIMONA SIMIONESCU

The structural and functional state parameters of the microbial communities, under the action of command factors (especially external), are analysed, in the artificial lakes on the Argeş river, in 1999–2000 period. The considerable input of organic matter and nutrients, carried by the effluents of urban waste waters of Piteşti town, in the Goleşti artificial lake, is well illustrated through the changes of microbial communities parameters. The numerical density increased 1.5 times, the production 6 times and the organic matter destruction rate 2 times.

Key words: microbial communities, organic matter input, microbial biomass and production, microbial matter destruction rate.

INTRODUCTION

The problem is one of larger themes within the framework of the Research-Development National Program "Horizon 2000", a theme initiated since 1999 by ANŞTI.

The accumulated knowledge about the urban pollution impact (with special reference to urban waste waters) upon biocenotic structures from aquatic ecosystems reveals the followings:

1) in our country, much of research focused on the anthropic impact upon aquatic ecosystems, even concerning the Argeş river;

2) the state parameters specific to abiotic components and several ones which characterize the qualitative structural (taxonomical diversity, bioindicators) and quantitative (density, biomass) aspects of some traditional biotic components, such as phytoplankton and zooplankton, were frequently considered.

The study of a less considered biotic component, namely microorganisms, constitutes the newness of our researches. We investigated mostly the heterotrophic bacteria, whose major functional role in an ecosystem is to DECOMPOSE the dead organisms. The microbial communities realize this role by the degradation of autochthonous and allochthonous organic matter, by the release of mineral compounds needed for the primary producer development, thus ensuring a major trophic base for the consumers and detrivorous organisms.

The aim of investigation was to evaluate the structural (number, biomass) and functional (productivity, destruction rate) parameters of microbial communities in Goleşti lake, depending on the organic matter and nutrients from urban waste waters of Piteşti town.

MATERIAL AND METHODS

The sampling was done in the following stations: Curtea de Argeş lake–dam: surface and bottom (I); Goleşti–lake tail: surface (II) and Goleşti–dam: surface and bottom (III) (Fig. 1), on water column, with Patalas type device. Field observations and determinations on some physical and chemical parameters (depth, transparency, pH, dissolved oxygen) were effected. Water samples for chemical and microbiological laboratory analyses were collected.

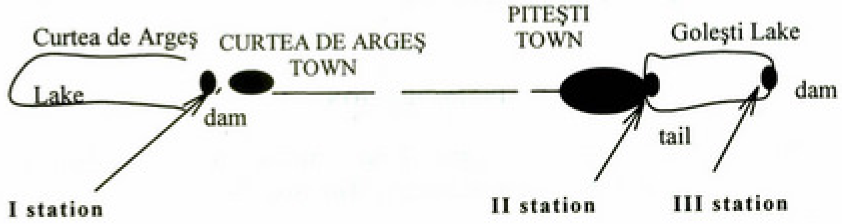


Fig. 1 – Water sampling stations.

The total organic matter present in the water was determined by the method of chemical consumption of oxygen, by using $KMnO_4$, in 1999 year, and in the next year, the same method by using CCOCr instead of $KMnO_4$. All nitrogen and phosphorus mineral forms, present in water, were also determined.

The numerical abundance of heterotrophic bacteria, including both vegetative and sporulated forms, was determined by the gelose culture method (4). The assessment of phosphate quantity in phospholipids was used for microbial biomass determination (1). The oxygen consumption method helped to determine the microbial production and destruction rate, by "in situ" tests (4).

RESULTS AND DISCUSSIONS

The position "in cascade" of the two lakes enabled us to demonstrate the existence of an allochthonous organic matter and nutrient input in Goleşti "lake–tail" area. We noticed an increase of the trophic level of the Goleşti lake, affected by the cumulative action of all upstream destructive factors. This situation was compared to a reference ecosystem, respectively the Curtea de Argeş lake, situated upstream against to majority of pollution sources (2).

The aquatic microbial communities represent a biotic component which, owing to the metabolic particularities, rapidly react to environment changes. Consequently, the mentioned structural and functional parameters can be valuable indices of allochthonous input of organic matter from urban waste waters of the Piteşti town, and, also resulted from the development and senescence of organism populations (algae especially) within ecosystem (3) (Fig. 2).

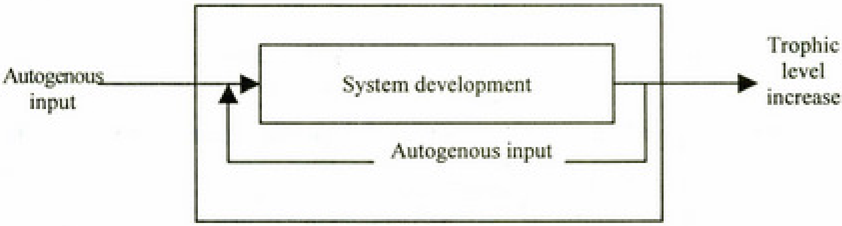
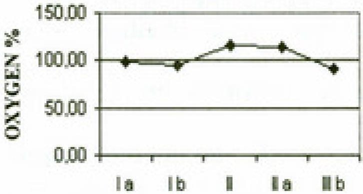
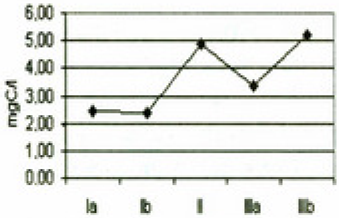


Fig. 2 – The general scheme of the ecosystem development (after D. Nicolescu).

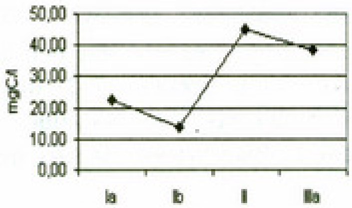
The maximum values of the state parameters of abiotic components (Fig. 3) were found in "lake tail" area (III st.) compared to the other observation sites. As for the total organic matter in water, the values registered here show that this area belongs to the second quality category. Also, the nutrients represented by the total mineral nitrogen and phosphorus have the highest values. The dominant nitrogen form is represented by nitrates, that of phosphorus by organic phosphorus. The organic matter, nitrates and phosphorus are components of the more or less purified waste waters.



Consumption of oxygen (%)



TOM- CCO-Mn method/1999



TOM-CCO Cr method /2000

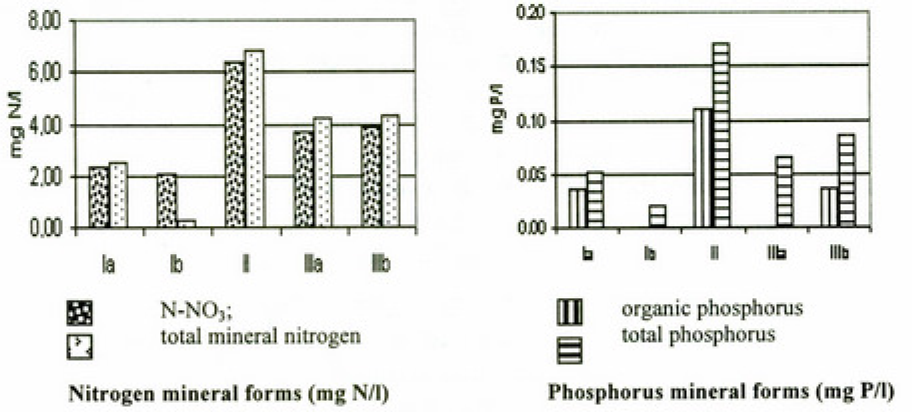


Fig. 3 – Chemical characteristics in observation points.
Ia – Curtea de Argeş-dam – surface; Ib – Curtea de Argeş-dam – bottom; II – Goleşti – lake tail; IIIa – Goleşti-dam – surface; IIIb – Goleşti-dam – bottom.

The urban waste waters determine the luxuriant growth of heterotrophic bacterial communities in Goleşti lake through the major input of organic substrate, a nutrient for the microbial communities. This fact is well illustrated by values of some microbiological parameters. Thus, the numerical density of spore forming bacteria doubles in "lake-tail" area against the dam one, and the microbial biomass, as well as the matter destruction rate, have values two times higher than in the dam area (Fig. 4).

Also the dam functional regime, the alternation of the stagnant water periods with those of water output from lakes, determine spatial dynamics of microbial communities as follows:

– the microorganisms are drawn into the sediments of fine particles, in the stagnant water periods;

– in the output water periods, the microorganisms are drawn along with the detritus and biocenotic components in the water horizontal streams, resulting the increase of the trophic level of Goleşti lake, situated upstream to Curtea de Argeş lake.

These processes are well illustrated by physical, chemical and microbiological parameters of the two lakes in the dam area Fig. 5. Both organic matter and nutrients increase 2–3 times. Consequently, several microbiological parameters reach especially high values: the numerical density increased 1.5 times, the production 6 times and the organic matter destruction rate 2 times, respectively.

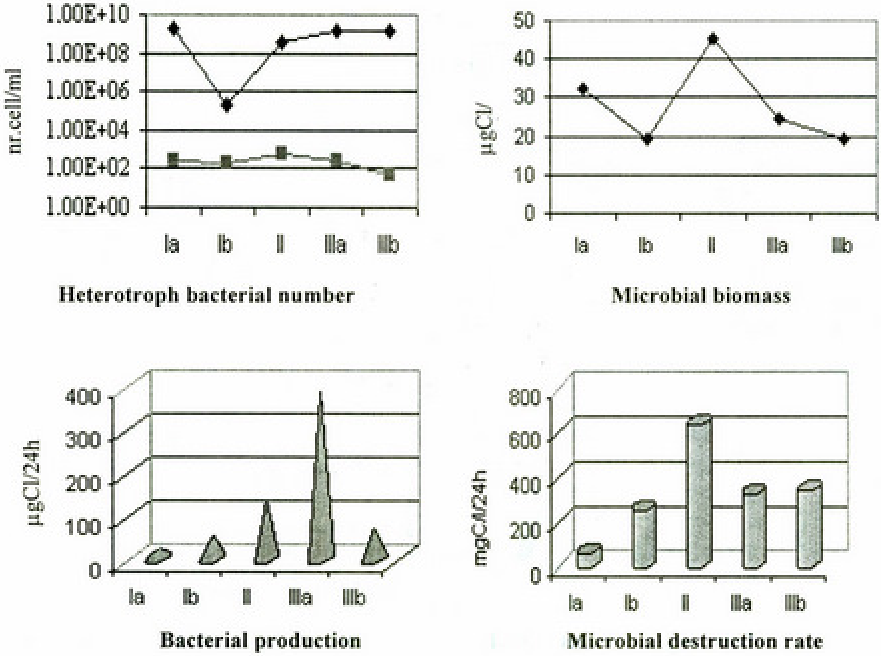


Fig. 4 – Microbial characteristics in observation points.
Ia – Curtea de Argeş-dam – surface; Ib – Curtea de Argeş-dam – bottom; II – Goleşti – lake tail; IIIa – Goleşti-dam – surface; IIIb – Goleşti-dam – bottom.

The microorganisms, by their capacity of decomposing organic matter, firstly consume the dissolved organic matter, which in this case represents 30% of total organic matter (in 2000 year) (Table 1).

Table 1

Physical and chemical water characteristics in observation points

Station / Parameter	Curtea de Argeş		Goleşti tail	Goleşti dam	
	Surface	**Bottom**	**Surface**	**Surface**	**Bottom**
1	**2**	**3**	**4**	**5**	**6**
Depth (m)	6	-	1,5	7	-
Transparency (m)	0.525	-	0,05	0.4	-
T/D index	0.0915	-	0.1	0.055	-
Temperature (°C)	11.4	10.9	18.5	22	25
pH	7.62	7.78	7.68	8.075	7.76
Oxygen (mg/l)	10.67	10.419	11.05	9.7835	7.468
Saturation (%)	97.9	94.9	116.2	113.6	91.3

Table 1 (continued)

1	2	3	4	5	6
TOM (mg.C/l) $KMNO_4$ meth.	2.426	2.37	4.893	3.345	5.201
TOM (mg.C/l) CCO-Cr meth.	22.5	13.87	45	38.62	-
$N\text{-}NO_2^-$ (mgN/l)	0.013	0.0155	0.055	0.0465	0.018
$N\text{-}NO_3^-$ (mgN/l)	2.35	2.1	6.4	3.7	3.9
$N\text{-}NH_4^+$ (mgN/l)	0.16	0.155	0.365	0.455	0.37
tot min. N (mgN/l)	2.52	0.2705	6.817	4.195	4.288
$P\text{-}PO_4^{-3}$ (mg.P/l)	0.016	0.006	0.035	0.0465	0.050
Organic P (mg.P/l)	0.035	0.014	0.109	0.0185	0.035
Total P (mg.P/l)	0.051	0.02	0.171	0.065	0.085

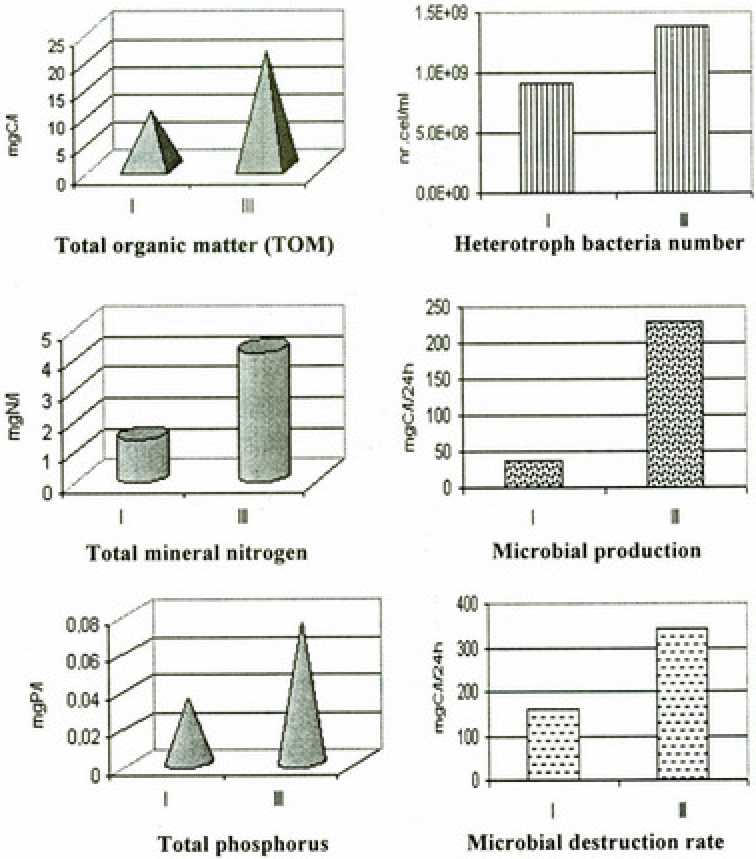


Fig. 5 – Chemical and microbial parameters in dam area of the two lakes.
I – Curtea de Argeş Lake; III – Goleşti Lake.

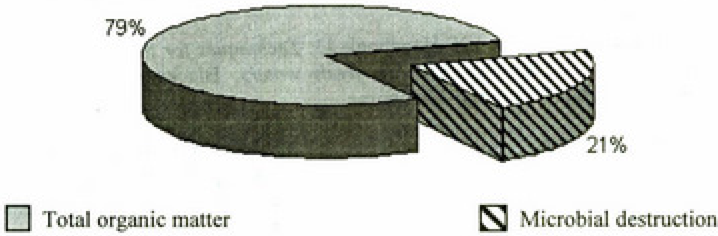


Fig. 6 – The ratio between accumulated organic matter and microbial destruction.

By reporting the organic matter subjected to microbial destruction, against the total quantity of organic carbon, a ratio of 1:4 results (Fig. 6).

This means that the organic substance accumulation exceeds the capacity to recycle organic matter by microbial decomposing.

CONCLUSIONS

1. The Curtea de Argeş artificial lake could be considered a reference ecosystem, the anthropic impact having an unsignificant effect, according to values of the analyzed parameters;

2. The Goleşti artificial lake, situated upstream the Piteşti town, is characterized by the evolution of the trophic level from meso- to eutrophic, the structural and functional parameters of microbial communities reaching maximum values in the lake tail area;

3. The location of the maximum values of the structural and functional parameters of microbial communities from Goleşti artificial lake, in the "lake tail" area, shows a direct input of organic matter and nutrients, resulted from the output effluent of urban waste waters of Piteşti town;

4. The gradual increase of microbial parameter values, in lakes of the Argeş river, from upstream to downstream, is well reflected in the increase of the trophic level of Goleşti artificial lake.

REFERENCES

1. Findley R.H. *et al.*, 1979, *Efficacy of phospholipid analysis in determining microbial biomass in sediment.* Appl. Env. Microb., **55**, 11, 2888–2893.
2. Doina Ionică *et al.*, 1999, *Research on the effects of urban pollution reflected in structure and functionality of main biocenotic components of the anthropized aquatic ecosystems of Argeş river*. Proc. Instit. Biol., Bucharest, p. 91–98.

3. Dorina Nicolescu *et al.*, 1998 – *Semnificaţia parametrilor structurali ai comunităţilor microbiene în modelarea proceselor biologice din acumulările râului Argeş*. Seminar ARDI. Vol.II (under press).
4. Sorokin Y. I and Katota H., 1972, IBP Handbook 23, *Techniques for the assessment of microbial production and decomposition in fresh waters*. Blackwell scientific publications, Oxford-London-Edinburgh-Melbourne.

Received May 20, 2001.

Institute of Biology
Spl. Independenţei 296 – 79651
PO-Box 56–53, Bucureşti
email: dioni@ibiol.ro

DIAGNOSIS OF IONIC EQUILIBRIUM IN THE BIOGEOCHEMICAL CYCLES OF TERRESTRIAL ECOSYSTEMS

ALEXANDRA VASU

As an image of a sequence in time in the ecosystems dynamic equilibrium, an original methodology of ionic equilibrium research in the biogeochemical cycles, as part of an ecosystemic integrated interdisciplinary methodology, is presented and illustrated by a case study in *Pinus silvestris* ecosystems with different stability, respectively productivity levels. The research results prove the peculiarity of the substances cycle and the different reactions of the entire ecosystem expressed by different ionisation levels, ionic activities and ratios, with consequences in the ecosystem stability and productivity level.

Key words: ecosystemic approach, interdisciplinary methodology, ionic equilibrium, biogeochemical cycles, ecosystem stability, ecosystem productivity.

INTRODUCTION

The increasing social-economic requirements for biomass production often lead to an ecological disbalance between the vegetation needs and the biotope supply, with consequences in the ecosystem stability, associated mostly with lowering of productivity.

The sustainable development strategies of the terrestrial ecosystems are necessary to harmonise the social-economic needs with the optimisation of the biomass production.

The ecological equilibrated bioproductivity is conditioned by the limiting of the land use to the compatibility range, with the persistence in time in a restricted variation interval of the soil fertility and its resistance to the disturbance factors induced by the intensive land use. It is possible to be achieved by the ensuring, even by anthropogenic intervention, of an advanced correspondence between the ecological needs of the biocenosis and the support ability of the soil, respectively of the whole biotope. Therefore, the elaboration of sustainable development strategies may be achieved only by the substantiation of the production technologies on the systemic – integrated knowledge of the ecosystem functions, by the structural and functional knowledge of the producer-biotope relationship. It is essential depending on the inorganic compounds dynamics, for the valuation of the dynamic equilibrium state at the research moment and for the estimation of the temporo-spatial evolution direction.

REV. ROUM. BIOL.– BIOL. VÉGÉT., TOME 46, N° 1, P. 55–68, BUCAREST, 2001

OBJECTIVE

The aims of the paper are to present an original systemic diagnosis methodology of inorganic compounds species in dynamic equilibrium in the biogeochemical cycles of the terrestrial ecosystem at the equilibrium status at the research moment and its application for the estimation of terrestrial ecosystem stability and actual effective productivity level, as well as for the disfunctions identification in the ecosystem equilibrium and evolution and also for the diagnoses of disturbance processes and their mechanisms. The importance of the knowledge of the ionic equilibrium in the biogeochemical cycles of the terrestrial ecosystems is illustrated with case studies in two *Pinus sylvestris* ecosystems with different stability, respectively productivity levels.

METHODOLOGY

RESEARCH APPROACH

The studies were carried out in an ecosystemic approach (Chirita 1974, Vasu 1972, 1984, 1985, 1988, 1989, 1989a, 1994, 1994a,b, Vasu et all. 1995–1996) considering that in the terrestrial ecosystems, the soil – as edaphotop – is the subsystem which achieves the systemic unity of life with the environment by the multitude of functional connections with the other subsystems of the biotope and with the biocenosis, in forming and evolution processes, as well as in the nutritional processes. In the soil, the focus of the main connections is the soil solution (Lindsay 1979, Vasu 1985, 1988, 1989a, Vasu 1994, 1994a, 1997, 1997a, 1998, Vasu *et al.* 1995–1996, Seceleanu *et al.* 1995).

The specific ion activities of the soil solution, directly, easy available to the plant roots (effective soil fertility), in dynamic equilibrium with the exchangeable adsorbed ions, potentially available to the plant (potential soil fertility), constitute the *mobile systemic structure* of the soil, which acts as ecological factors in the ecosystem functions and determines straightway the actual (effective) ecosystem stability and productivity. The mobile systemic structure of the soil is in dynamic equilibrium with the constitutive elements of the mineral, organic, organo-mineral and organo-metallic compounds – *basic systemic structure* – of the soil and with the other biotope subsystems and processes which influence the nutrients availability, respectively act as ecological determinants in the ecosystem functions and induce indirect influences on the ecosystem stability and productivity.

The terrestrial ecosystems are dynamic systems where the aqueous solution is the nutrient conveyer and therefore the pH, Eh, temperature, moisture and potential ionisable in soil solution, adsorbed ions (evaluated through SB -sum of soil exchangeable cations, Te -effective total exchange capacity at the soil pH) are considered *thermodynamic stability conditions* parameters of the soil system equilibrium and *moving conditions* parameters of the nutrients in the rock-soil-plant-animal systems (Vasu 1985, 1988, 1989, 1989a, 1994, 1994a, 1996, Vasu et all. 1995–1996 etc.) and in the human body probably too.

The *potential productivity* is appreciated by the trophicity index (Chirita 1974 modified Vasu 1987, Vasu 1984, 1987, 1997) and the mesoclimate influences.

The *effective (actual) productivity* (Filip 1976, Vasu 1997) is estimated at the research moment by the simultaneous study of the nutrients regime (Vasu, Filip 1989) (equilibrium composition-quantity and speciation of the nutrients in the biogeochemical cycles) with the ecoclimatic regimes (humidity regime and energetic regimes) of the ecosystems. The actual ecosystems productivity is estimated as compared to the potential one to render evident the ecosystems stability. The ecosystem disturbances induce effective productivity lowering as compared with the potential productivity with negative influences in the ecosystem stability.

METHODS

The inorganic compounds dynamics study methodology is part of an original interdisciplinary systemic diagnosis methodology in ecosystemic approach (Chirita 1974), which studies the soil ecosystemic integrated (partially published Vasu 1972, 1985, 1994, 1996, Vasu *et al.* 1995–1996, Vasu in Paucă-Comănescu *et al.* 1989, Vasu in Seceleanu *et al.* 1995). The interdisciplinary methodology connects multidisciplinary methods, in which each discipline work with its own specific methods, adapted to the ecosystemic purposes, but very important all at the same profoundly analysed level, for making possible the interrelated interpretation (by logic analysis or multi-variable analysis) of the investigation results (Vasu 1985, 1994b, 1998, Vasu *et al.* 1996, Vasu in Seceleanu *et al.* 1995, Zanelli, Vasu 1976).

The methodology is verified in more than 550 ecosystems, out of which more than 140 with a different kind of human impact. It was verified on a large geographical scale, about 26° latitude, 35° longitude and 1500 m altitude differences, in Romania, Spain, France, Germany, Sweden and Russia. It was applied even in two additional EU "PECO" Projects, CT 910043 and CT920141.

In the interdisciplinary ecosystemic research, the work of the entire team must be done in the similar ecosystemic research meaning.

The scientific and psychological compatibility of the team memberships is very important.

For concluding results it is very important the selection of the representative ecosystem for the studied phenomenon, as well as of the equilibrated comparison ecosystem.

The research area for each elementary ecosystem unit is considered 20 m/20 m around the main producer tree.

The soil profile is placed under the crown of the main producer tree.

The soil system analysis is only partially published (Vasu 1972, 1984, 1985, 1994, 1997 a,b, 1998, Vasu *et al.* 1995–1996, Vasu in Pauca-Comanescu *et al.* 1989 and in Seceleanu *et al.* 1995).

The dynamics of inorganic compounds in soil is differently studied depending on the special purposes, for soil pedogenetic evolution level and *direction identification (soil system as natural or transformed body), for the soil* function as edaphotop or for the soil use as production means.

The ionic equilibrium is biochemically and geochemically influenced by the chemical composition of the vegetation and by the mineralogical composition of the parent material.

The analysis to assure the data for the estimation of the ionic activity in dynamic equilibrium in the terrestrial ecosystems involves the analysis of total cations and SiO_2, C, N, P contents in vegetation (roots, stems, leaves and fruits of the primary producers, as well as of the main accompanying species, especially ecological indicative species for the biopedoclimatic belt), the analysis of total and aqua soluble or hydrolysed cations and SiO_2 , C, N, P contents in litter (Ol, Of, Oh horizons), seed and parent material and finally the analysis of the main component of the biogeochemical cycle, the soil (subsystem which achieves the systemic unity), consisting in the analysis of total and aqua soluble C, N, P contents, mobile (exchangeable adsorbed ions) and aqua soluble (ionizate and hydrolysed) cations and SiO_2.

The total C in vegetation, litter, seed and soil, as well as the C of different fractions of humic substances, separated from the soil by the selective extraction with water, dilute acids or $Na_4P_2O_7$ solution (Kononova, Belcicova, Ponomareva mod.Vasu 1972) were determined. In the vegetal material it is better to determine total C by the modified Anne-Anstette method (Vasu 1972). All C forms were finally determined titrimetrically after oxidation with a sulphuric-chromic mixture, using as indicator phenylanthranilic acid (Walkley Black mod. Vasu 1972).

Total N has been determined by the distillation method and total P spectrophotometrically as molybdenum blue from sulphuric-perchloric wet mineralizer (Ginsburg mod. Vasu 1972).

The total content of macro and micro elements, as well as SiO_2, were determined in HCl dissolved ash, after SiO_2 separation and gravimetrical measurement, by atomic absorption spectrophotometry.

The organo-metallic compounds were determined in remade pyrophosphate extracts, respectively organo-mineral SiO_2 gravimetrically and macro and microelements by atomic absorption spectrometry.

The ionic equilibrium in the systemic mobile structure of the soil (the most expressive in the biogeochemical cycle) was established (Vasu unpublished) at the research moment (as a moment in the dynamic equilibrium of the whole ecosystem). There were determined the aqua soluble (soil/deionizate water 1/2.5, the same ratio and the same deionizate water as by the potentiometric pH and Eh measurement) content of $C_{(a)}$, $N.NO_3$, P, H, macro and micro elements and hydrolysed SiO_2 and the mobile ions adsorbed on the colloidal complex, excepting the two first elements, which were related to the total content. The first four elements were determined straight in the water extract (each one in 10 ml extract), $C_{(a)}$, and H were determined titrimetrically, $N.NO_3$ potentiometrically and P spectrophotometrically as molybdenum blue. The macro and micro elements were measured, by atomic absorption spectrophotometry, in the remade extract (minimum 50 ml extract evaporate, oxidate with 1 ml from the mixture 1/1/2 perchloric/sulphuric/nitric acids and then chlorinated, with SiO_2 separation) and SiO_2 gravimetrically. The mobile-exchangeable adsorbed ions from the soil (Vasu 1972) were extracted in dilute HCl solution (0.05M) and straight in the extract measured, P spectrophotometrically as molybdenum blue and macro and micro elements by atomic absorption spectrophotometry. The effective acidity, H and Al were titrimetrically determined in KCl solution extract and the potential active acidity at pH 8.3 also titrimetrically in CH_3-COOK solution extract (Vasu 1972).

As geochemical ionic source, there were, qualitatively and quantitatively determined the main mineral compounds (expandable minerals, illite, chlorite, kaolinite, mixed layers) in the clay fraction under 0.002 mm.

The thermodynamic stability and moving parameters were determined: pH and Eh potentiometrically in deionisate water suspension (1/2.5 soil/water ratio), SB titrimetrically in the same HCl solution in which were determined the individual ions and all the analysis at the same temperature about 20 °C on the air dry soil materials and vegetal materials stabilised at 70 °C, with humidity correction at 105 °C for all the materials.

For the individual characterisation of the results, interpretation scales were established, which allow also the evaluation of the ecological significance of the results as ecological factor or determinant.

Since the ionic equilibrium analysis uses conventional, relative methods, excepting the total analysis, for a realistic diagnosis of the ionic equilibrium in the biogeochemical cycle the data interpretation is better to be made comparatively with those of an equilibrate ecosystem in similar ecopedoclimatic conditions without the presumptive disturbing factors, in which the actual, effective productivity of the ecosystem is in concordance with the potential trophicity of the soil and with the production capacity of the biotope.

CASE STUDY

As application of the above presented methodology a case study is presented in a disturbed *Pinus silvestris* ecosystem Vindeln (E4), with low stability and productivity, invaded with *Vaccinium myrthilus* as compared with another one Jadraas (E3), with high stability and productivity with *Vaccinium vitis-idaea*, both in northern Sweden. The studies were carried out with exclusive UE financial support in an Additional Project to "VAMOS" Project, co-ordinated by M.M.Couteaux, CNRS-CEFE Montpellier, France, 1995–1996.

The Ecosystem E4 Vindeln is localised in Sweden, Vasterboten region, Norrland province, 9 Km N-NE from the town Vindeln, in a transition climate between cold temperate and subarctic, in coniferous belt, at their northern limit.

Situated in Norrland Tableland the subunit of Sweden Tableland, with 200–500 m altitude, located on the alluvial plain of rivulet Kryclan, tributary of river Vindeln, at 175 m elevation and about 25 m relative altitude, the studied ecosystem is on an alluvial terrace (2–5 m relative altitude), with very undulating microrelief with many microdepressions and ridges.

The main producer of the ecosystem is the cultivated Scots pine (*Pinus sylvestris*), in a pluriaged forest, 65 years old, with composition 9 Pi+1 *Piceea excelsa, Betula verrucosa, Salix caprea*, with consistence 0.7 and low productivity (cl.IV). *Juniperus communes* is about 5% represented.

The ecological conditions assure a vigorous growth of *Vaccinium myrthillus* (70 %) in the ecosystem. *Vaccinium vitis-idaea* is less abundant and owns only 10 %.

Calluna vulgaris and *Empetrum nigrum* are two dwarf shrubs, moderately represented (10 % and 5 % respectively). With low representation (1 %) *Lycopodium complanatum* is present.

The inferior layer of the vegetation is represented by bryophytes and lichens. *Pleurozium schreberi*, *Dicranum polysetum* and less *Polytrichum commune* are the main moss species registered. The "reindeer lichens" *Cladonia rengiferina* and *Cladonia stelaris* (sin. *C. alpestris*) appear between the trees and dwarf shrubs, in the small sunny areas, covering 10 % of the mosses surface.

At 64°13" Northern latitude (schematic Map of the terrestrial world 1:150000000) the ecosystem belongs to the temperate, cold macro climate. The physico-geographical position of the ecosystem, on alluvial terrace easy winding, with 175 m altitude and about 80 Km from the Baltic Sea coast and more than 500 km from the Scandinavian Mountains, as well as the biodiversity, determine the temperate cold mesoclimate transitional to continental subarctic climate, cool boreal climate, with a 6 months biologically active period. The mesoclimate is strongly influenced by the Golfstream, otherwise it would be much colder. According to the specific structure of the phytocenosis the ecoclimate is in spring from little active to suboptimum (only at the soil surface optimum) and at the litter fall from negative (–2 °C) to suboptimum, as compared with the measurement in meteorological standard conditions, negative (–4 °C) to optimum in spring and negative (–4°) to suboptimum at the litter fall. The hydric soil regime is in spring variable from suboptimum to suprapessimum in Bs and in the parent material strata pessimum to suprapessimum and at the litter fall dominant inactive, excepting Bs active.

The soil type in FAO/UNESCO Classification is a Haplic podzol (podzol aluminium illuvial). The thin soil (Bs up to 21 cm), with 5 parental materials strata (till 220 cm), relatively poor drained, has the rooting depth up to 60 cm in the Cog horizon at the limit of $Cgor_1$ horizon.

The soil has a thin Ol horizon (0.5 cm) consisting of undestroyed needles, leaves, pine cones and branches and subjacent a thick (7 cm) green moss very well developed, an Of horizon (7 cm) consisting of fermented and undestroyed needles, leaves, with traces of a peat forming process and a very weakly developed Oh horizon (0.5 cm), with moderate mineralisation (white spots of SiO_2). The mineral horizons are relatively thin, Au (2.5 cm), medium loamy sand, Es (4.5 cm), medium loamy sand and Bs (14 cm), medium sand; C_1 (31 cm), medium loamy sand and the other parent material strata all fine sand.

The thermodynamic stability conditions are characterised by the variation of the energetical-thermal parameters, active humidity index, pH and Eh values, measured in spring and at the litter fall and the adsorption complex parameters.

The thermal variations are in spring pedothermal inactive in the main rooting depth and little active in the upper part of the soil profile and phytothermal negative only near to the soil surface and dominant inactive, with a suboptimum daily maximum. At the litter fall they are pedothermal little active to active at the day maximum, only below 100 cm inactive and phytothermal negative to very active at the day maximum.

The soil is hydric in the spring in the proper soil variable from suboptimum in Au to suprapessimum in Bs and in the parent material strata pessimum to suprapessimum. At the litter fall it is dominantly inactive, excepting Bs horizon which is active.

The thermodynamic stability conditions are physico-chemically characterised in spring by extremely strong acidity at the soil surface (in Au pH 3.13 and in Es 3.60) and high oxidising level (Eh 533–526 mV) and in the other horizons by strong acidity (pH 4.50–5.07) and also high oxidising level (502–535 mV). At the litter fall they are characterised by extremely strong acidity pH 3.53–3.62) and an extremely high oxidising level (574–569 mV) at the surface, by strong acidity (pH 4.20) and high oxidising level (524 mV) in Bs horizon and buffered to moderate acidity (pH 5.18–5.62), but also high oxidising level (514–536 mV) in all the parent material strata. The exchange characteristics of the poor clayey-humic-amorphous material complex are defined by a very low effective cation exchange capacity (Te 5.3–9.7 me/100g soil), only in Au 25.3 and Es 16.4 me/100g soil, increased by acid components, due to the extremely acid bioaccumulation. The exchange capacity has an extremely low base content (1–3 me/100 g soil), only in the bottom part of the profile a little greater (4.6–6.6 me/100 g soil and in the Au horizon increased (3.75 me) by the extremely acid bioaccumulation. The soil is generally oligobasic, extreme oligobasic in the Es horizon and saturated to oligomesobasic in the C horizons at both acidity levels.

The vegetation, with very strong to strong acidity (pH 3.33–4.52) and moderate to high oxidising (Eh 434–532 mV) movement conditions, assures a moderate active nutrition, with some accumulation of K, Al, and Mn in *Pinus sylvestris*, of K, Ca, Mg, Mn in *Piceea excelsa*, K, Ca, Mg, Al and Mn in *Betula verrucosa*, K, Ca, Mg, Fe, Al, and especially Na in *Vaccinium myrthillus* roots and Mn in leaves.

The litter as the main link between the vegetation and the mineral soil, where occur the main transformations of the organic matter to humificated specific soil organic matter is characterised, on a surface of 40×40 cm, by a high accumulation of organic matter in the fermentation Of layer (7 cm, with 44 % Ct); it is 108g in Ol, 270g in Of and 37g in Oh.

The bioaccumulation of the litter, relative moderate in the Ol layer, is of raw organic matter to moder type humification (C/N: 58 in Ol, 53 in Of and 38 in Oh). The bioaccumulation of the raw organic matter is limited in the soil in the thin Au horizon.

In the soil, the total humus content is very-very high for sandy soil, especially in the thin Au horizon (Ct: 11300 mg) and surely very low in the fine sand parent material strata (40–60 mg). In the weathered Au horizon, part of the organic matter is mutual colloidal protected by amorphous SiO_2 resulted from the weathering process which occurs and a large part of the organic matter is complexed with metallic ions, resulting from the relative high mineralization in Oh, from the weathering in Au and hydrolysis in C_1.

The litter mineralization degree (SiO_2/Ct) is in Ol:0.01, in Of:0.3 and in Oh:4.7, with corresponding relative high release of CO_2 in the atmosphere and of N, K in the soil, but blocking of P in the Ol layer and low recycling of Ca, Mg, blocked in the needles. The high acidity and the active weathering mobilise in the humid ecoclimate a high quantity of Fe, Al, Mn and even relative high quantities of Pb, Zn, and Cu. The nitrification degree (N.NO_3/Nt) is very low in Oh (0.0004), but moderate in Au and Es (0.0020–0024).

The mineral bioaccumulative influences are mostly in the weathered thin Au horizon on the increase of the ionic activity of N, P, K, Ca, Mg, Al, Mn, Zn and Cu in the extremely strong acid and very oxidising soil solution.

The ionic equilibrium in the soil is also influenced by the mineralogical composition and the main solidification processes. The clay mineral composition is very much influenced by the extreme strong acid and oxidising litter (pH 3.2 and Eh 524 mV in Oh), which induce extremely strong acid hydrolysis (H^+ potential active 20 ppm). Thus, in Au and Es horizons, extremely strong acid, pH 3.5–3.6 (potential active H^+ 3.3–9 ppm) and very oxidising (500–535 mV), in the humid ecoclimate, an intense weathering process occurs, with high illite smectisation (on illitic substratum), unusual for this soil type. The new formed smectite is less expanded than normal (1.6–1.65 nm with EG). The mineralogical composition in the ratio smectite/chlorite/illite/kaolinite is: in Au 42/25/30/3, in Es 67/25/7/1, in Bs 0/69/21/10 and in C_2 0/24/67/9.

The extremely high active acidity and potential active acidity (H^+(a) 20-3.3-9-1 ppm in Oh, Au, Es, Bs), activated by the high proportion (70%) of acidophile *Vaccinium myrthillus*, determines the high intensity of the weathering processes specific for spodosols, as well as a non specific very intensive illite smectisation (67% smectite and 7% illite in Es, where H^+ is 9 ppm), with SiO_2(om) and especially high Al, Fe and K release in the soil solution.

In the deeper part of profile, in C_1 and C_2 horizons, with a relative higher clay content (9.5, respectively 5.3%), as compared with traces in the lower strata, where a moderate acid hydrolysis occurs (potential active H^+ increases from 0.2 ppm in Bs to 0.9–1.1 in C_1, C_2) the soil solution is enriched in ionised Fe, Al and in hydrolysed SiO_2(a) 140–100 ppm.

The extremely high active and potentially active acidity determines the enriching in Al(a), becoming dominant in the soil solution, with toxic effect for the vegetation. Thus, the disturbance of the nutrients ratios in the soil solution is a limitative factor for the ecosystems main producer, explaining their low stability, productivity and low biological quality.

As control for the analysed ecosystem an equilibrated ecosystem is presented concisely. It is the Ecosystem E3 Jadraas, Sweden from the same Project, also with *Pinus sylvestris*. The ecosystem is situated in Gavleborg region, Gostricland province, 3 km SE from town Jadraas.

The researched ecosystem is in an equally aged Scots pine forest, with cultivated *Pinus sylvestris*, 70 years old, as main producer, with 10 Pi disseminated *Piceea excelsa* and deciduous composition (*Acer platanoides, Betula verrucosa, Salix caprea*), 0.6 consistence and high stability and productivity (cl.II). Among the accompanying species, *Vaccinium vitis-idaea* dominates (75%), *Vaccinium-myrthillus* is only rarely scattered (3%). *Calluna vulgaris* grows in small clusters and owns 20% from the accompanying shrubs. 90% from soil surface is covered with mosses which belong to the species *Pleurozium shreberi* and *Dicranium scoparium*. The other 10% from the soil surface is covered with lichens such as "reindeer lichens" *Cladonia rangiferina* and *Cladonia stelaris*, well developed in sunny places.

Macroclimatic the ecosystem area, at 180 m altitude, on the winding moraine plateau, belongs to the temperate macroclimate. Near the Baltic Sea and at a relatively great distance (over 400 km) from the Scandinavian Mountains the specific mesoclimate is transition moderate temperate to oceanic. The ecoclimate is obviously warmer than E4. In spring pedothermic is little active to active in the main rooting depth and only below 100 cm inactive. As compared E4 is pedothermic inactive in the main rooting depth and only at the surface little active. Phytothermic E3 is inactive in the bottom part (till 60 cm) and in the upper part of the ecosystem (>13 m) and in the rest little active to very active-suboptimum. As compared E4 is negative near to the soil surface and in rest dominant inactive. At the litter fall E3 is pedothermic dominant active to very active and phytothermic negative in the upper part (>13 m), respectively inactive to suboptimum in the bottom part, while E4 is at the litter fall pedothermic little active to active at the day maximum, only below 100 cm inactive and phytothermic negative to very active at the day maximum.

On fluvio glaciar deposits resulted after the transport of the moraine deposits by the dense river network, formed of thick strata of sands with fine grovels insertions, the soil parent material consisted of alluvial and lacustrine deposits originated from Quaternary moraines.

The ecosystem area is situated in Svealand Tableland, the subunit of Sweden Tableland on the alluvial plain of Jadraan river, at 180 m elevation. The microrelief is high alluvial plane, former delta 8000 years before, very slowly undulated.

In FAO/UNESCO Classification the soil is Haplic podzol (iron humus podzol). The thin soil is a little thicker than in E4, respectively Bs is up to 30 cm and continued with a BC horizon up to 43 cm, with frequent rooting depth up to 50 cm and relatively frequent between 80–100 cm, whereas in E4 Bs is up to 21 cm and the rooting depth up to 60 cm and rarely up to 70 cm. Between 43–150 cm three, moderately drained, parent material C strata are developed.

The soil has also a large litter stratum (19 cm), with 6 cm moss between Ol (1.5 cm) with more than 85 % new undestroyed needles and a very thick fermented Of (11 cm), followed by a very small, advanced mineralised Oh (0.5 cm).

The soil is hydric in spring very active and at the litter fall active -very active, whereas in E4 the soil is in spring from suboptimum to suprapessimum and at the litter fall dominant inactive.

The physico-chemical thermodynamic stability is characterised in spring by extremely strong acidity in Au, Es, Bhs (pH 3.42–3.72), very strong acidity in Bs (pH 4.31) and buffered to strong acidity in BC, C horizons (pH 4.75–5.09), by very high oxidising levels (dominant 540–560 mV and 574 in Au). At the litter fall very strong acidity in Au, Es, Bhs (pH 5.39–4.30 mV), moderate acidity in Bs and BC (pH 4.95) and buffered to the low limit of weak acidity in C horizons (pH 5.75–5.87) and high, respectively very high oxidising levels (dominant 500–530 mV).

The exchange characteristics of the very poor clayey-humic-amorphous material complex are: extremely low effective cation exchange capacity (Te 1.5–5.1 me/100g soil), excepting the thin (1 cm) Au horizon with acid bioaccumulation to 7.3 me, with extremely low exchangeable bases content (SB 0.4–1.4 me/100g soil), effective extreme oligobasic buffered to oligobasic in C_3 strata. As compared in E4 are obviously higher, Te 5.3–25.3 me, with SB 1–6.6 me /100 g soil.

With uniform coarse texture the soil is very poor in clay fraction (0.6–1.3%), only in thin (3 cm) Bhs horizon is 3.2 %; below 30 cm the sandy stratified C has only clay traces. Above 30 cm are identified chlorite-like minerals (28–46 %), expandable minerals (31–43 %), illite (9–24%) and kaolinite, excepting Bs in which dominant are the expandable minerals. Below this depth the chlorite-like minerals content becomes very high (63–77 %) and the illite and kaolinite contents are comparable (18–21 %, respectively 16–24 %).

The total humus content is relatively high for such a coarse sandy soil, but with a low humification degree to moder dominated by fulvic acids.

The vegetation, with relatively the same biodiversity like in E4 biodiversity, has the appropriate strong to very strong acidity (pH 3.68–4.65) and moderate to high oxidising (Eh 430–506 mV) nutrients moving conditions, but it has an active influencing nutrients turnover through extremely strong acid (pH 3.82–3.32) and high oxidising litter (Eh 512/529/524 mV), with high accumulation (as macro nutrients reservoir) in the thick Of (11 cm), weak mineralised (SiO_2/Ct 0.15) and with a thin Oh (0.5 cm) advanced mineralised (14.66), as compared with E4 where the litter is similar acid, but a little less oxidising (Eh 494/484/524 mV) and totally differently mineralised, extremely weak mineralized in Of (0.01) and only moderate in Oh (4.7). As a consequence, the buffering capacity of the bioaccumulation against the unusual aggressive weathering process in E4 is much lower than in E3. Thus, in Oh-Au-Es-Bs-C_1 horizon sequence, in E4/E3, the aqua soluble (ionisate or hydrolysate) are: H^+(a) ppm 20.0/0.5-3.3/1.0-9.0/0.8-1.0/0.4-0.5/0.3; SiO_2(a) ppm 168/58-94/35-61/50-61/51-140/45; Al^{+3}(a) ppm 45/5-13/5-26/10-10/4-41/4; Fe^{+3}(a) ppm 13/2-14/3-13/3-13/2-39/3; Na^+(a) ppm 36/1-5/8-5/1-1/1-10/2; Ca^{2+}(a) ppm 36/8-19/6-16/8-5/4-7/2; N.NO_3^- (a) ppm 1.3/4.0-7.5/4.0-6.0/0.2-0.2/0.2-0.9/0.2; P(a) ppm 12.2/1.6-3.4/1.1-1.2/1.4-1.1/0.3-0.3/0.6; K^+(a) ppm 38/2-34/9-4/7-6/5-6/1; Mg^{+2}(a) ppm 13/1-7/1-4/1-3/1-6/1; Mn(a) ppm 4.5/0-1.7/0-0.7/0.2-0.9/0-0.5/0.3; Zn^{2+}(a) ppm 0.9/0.7-0.9/0.7-0.9/1-1/0.8-0.9/0. Pb, Cu, Cd are practically absent in both soil profiles, excepting a very small activity in the soil solution of Cu 0.4 ppm in Oh of E3 and 0.2 ppm in Au of E4, whereas in both are present as mobile forms Cu and Pb. The ionisation level in the soil solution as compared with the ionic adsorbed form is extremely variable, from 0.1% to more than 70% from element to element, soil profile to soil profile and horizon to horizon. It depends on the variable relationships of the 5 thermodynamic stability and moving conditions parameters, on the bioaccumulation effect variation, on the intensity of the weathering processes, inducing mineralogical composition changes like those presented.

The ionic equilibrium in the soil solution of E3 is influenced by the bioaccumulation, especially of Ca, Mg, N, P, K, Na, but also by the actual active solidification processes like the hydrolysis, or weathering one, evidentiated by the SiO_2(a) quantity and Al, Fe abundance (fixed as mobile exchangeable forms, or mostly as organo-metallic complexes). The processes are more aggressive in E4 ecosystem, with the extremely high, illite smectitisation and H^+, Al, Fe, Mg, K, Mn, Ca, P release in the soil solution, due to the high abundance of acidifiant *Vaccinium myrthillus*.

The equilibrated composition, with a low concentration (ionic activity) of the soil solution (N, P, K, Ca, Mg, Al, Fe, Mn, Zn, Cu, SiO_2, C(a) etc) in E3, in the active rooting depth, available for the vegetation in the strong acid, oxidising and active ecopedoclimatic moving conditions, ensuring a uniform nutrition, concordant with the extractings of the cultivated *Pinus sylvestris*, explain the high productivity (cII) and stability of the forest in E3, as compared with those in E4, with higher concentration, but unbalanced, of ionizate in soil solution and mobile nutrients form, with dominance of toxic Al, and the other weathering products.

CONCLUSIONS

1. The human impact acting, ignoring the ionic equilibrium in the biogeochemical cycles, determines the main disturbances in the ecosystems functions with stability and productivity lowering;

2. The presented case study reveals that only the ionic equilibrium is able to point out the real life and the deep real causes of stability and productivity lowering in E4, as well as to acknowledge the acting processes, their mechanisms and their intensity;

3. The biodiversity is appropriate in the two ecosystems, but the stability and the productivity are totally different, probably because of the nutritional compatibility differences of the accompanying species with the main producer. The deep study of the nutrients cycling emphasises the much larger compatibility of *Pinus sylvestris* with *Vaccinium vitis-idaea*, than with *Vaccinium-myrthillus*, antagonistic from nutritional point of view. *Vaccinium-myrthillus* induces, by its aggressive acidification, with weathering intensity increasing, the disbalance in the ionic ratio in the soil solution, expressed by high H^+ and toxic Al^{3+} activity in the nutritive soil solution.

4. The usual characterisation of the two analysed ecosystems is not able to explain the higher stability and productivity of E3, with a smaller clay content and a very poor clayey-humic-active amorphous material colloidal complex, with a lower exchange capacity and mobile-exchangeable potential nutrients than E4;

5. The ecosystem chemistry, focused on the soil solution chemistry and on the ionic equilibrium in the biogeochemical cycle, is essentially involved in the terrestrial ecosystems stability achievement, as well as in emphasising the disturbance level and its reversibility;

6. Only the knowledge of the ecosystem disturbance level and causality allow the correct substantiation of the sustainable land use and soil resources management.

REFERENCES

1. Chiriţa C.D., 1974, *Ecopedologie cu baze de pedologie generală*, Edit. Ceres, Bucureşti, 590 p.
2. Filip F., 1976, *Despre productivitatea primară a ecosistemelor terestre*. Travaux de station "Stejarul", Ecologie Terrestre et Genetique, p. 217–222.
3. Lindsay W.L., 1979, *Chemical equilibria in soils*, ed. Wiley-Interscience, New York, 449 p.
4. Paucă-Comănescu M., Almăşan H., Arion C., Bândiu C., Caracaş V., Doniţă N., Falcă M., Honciuc V., Oromolu-Vasiliu L., Popescu A., Sanda V., Tacina A., Vasu A., 1989, *Făgetele din Romania – Cercetări ecologice*, Edit. Acad. R.S.R., Bucureşti, 262 p.
5. Seceleanu I., Vasu A., Filip F., Matei M., Matei S., Crăciun C., Popescu I., Carcea I., Stefănescu D., Chivulete S., 1995, *Dessertification risk assessment and land use planning in Mediterranean coastal area*, EU Project **CT 910043**, 59 p. + 15 tables, 34 figures & 6 sheets.
6. Vasu A., 1972, *Principalii constituenţi din soluri acide forestiere. Metode de separare şi dozare*. Referat doctorat, arhiva Fac. de Chimie, Universitatea Bucureşti, 213 p.
7. Vasu A., 1984, *Extracţia selectivă a unor elemente din soluri cu agenţi de complexare*. Teză de doctorat, arhiva Fac. de Chimie, Universitatea Politehnică, Bucureşti, 150 p.
8. Vasu A., 1985, *Cercetarea analitică a solului ca component al ecosistemului*. **Vol. jubiliar 100 de ani de învăţământ silvic**, Universitatea Braşov 1983, p. 75–80.
9. Vasu A., 1988, *Sistemul sol-edafotop-mijloc de producţie*. Analele ICPA, **XLIX**, p. 127–144, Bucureşti.
10. Vasu A., 1989, *Componenţi în echilibru în circuite biogeochimice ale ecosistemelor terestre*. Analele ICPA, **L**, p. 175–198, Bucureşti.
11. Vasu A., 1989a, *Componenţi în echilibru dinamic în circuitul sol-plantă-animal*, Edit. Acad. RSR, St. cerc. biol., Seria biol.veget., **41**, *1*, p. 33–40, Bucureşti.
12. Vasu A., 1994, *The humification quality and the actual bioproductivity of the terrestrial ecosystem*, in: Senssi N., Miano T.M. (ed), *Humic substances in the global environment*, Elsevier, Amsterdam, p. 713–718.
13. Vasu A., 1994a, *The soil chemistry, ecosystemically integrated into the environmental research*. Proceedings 15th World Congress of Soil Science, Acapulco, Mexico, p. 56–57.
14. Vasu A., 1994b, *Fundamentarea ecosistemică a reconstrucţiei ecologice a ecosistemelor terestre*, Edit. Acad. Române, Memoriile Academiei Române, **seria IV**, tomul 17, p. 209–218.
15. Vasu A., 1997, *Soil Chemistry and Ecosystem Productivity*, Mitteilungen der Deutschen Bodenkundlichen Gesellschaft, **85**, *1*, p. 381–384.
16. Vasu A., 1997a, *Diagnosis of terrestrial ecosystems disfunctions for the substantiation of sustainable use of the soil resources*, Publ. SNRSS, 1997, **vol. 29 D**, p. 263–269.

17. Vasu A., 1998, *Inorganic compounds dynamics in the soil ecosystemic integrated*, Proceedings 16[th] World Congress of Soil Science, **CD–ROM** Symposium nr. 6/24 August, Scientific registration 691, Montpellier, France, 6 p.
18. Vasu A., Filip F., 1989, *Contribuţii la cunoaşterea regimului de substanţe nutritive al ecosistemelor terestre*. Publ. SNRSS Piteşti 1988, **vol. 26 A**, p. 131–136.
19. Vasu A., Seceleanu I., Matei G.M., Matei S., Crăciun C., Taina I., Ştefănescu D., Chivulete S., Vasilescu P. 1995, 1996a, *Variation du réservoir de matière organique du sol*. Additional EU Project **CT 920141**, 127 p. + 25 tables, 40 figures & 10 sheets.
20. Zanelli S., Vasu A., 1976, *Some specific chemical methods for investigating acid forest soils*. Vth Congress of Jugoslav Society of Soil Science, Sarajevo, p. 81–89.

Received April 17, 2001.

Institute of Pedology
R.I.S.S.A.
B-dul Mărăşti 61, Bucharest

STRUCTURAL DIVERSITY OF TREE LAYER IN FÂNTÂNELE FOREST (COMANA FOREST DEPARTMENT)

MIHAELA PAUCĂ-COMĂNESCU*, CONSTANTIN BÂNDIU**

There are presented the tree layer structures of 12 forest types identified in Fântânele forest, a very diverse area including many habitats, from wet and marshy to meso-xeric and xeric conditions. Richness of wood species as well as density of trees, their diameter and height, are very different according to habitat and silvicultural management. Both causes determined a high diversity within the same forest type and between different types. Old forests (more 120 years) and several younger ones (60 years) are described, some of them even-age and the others pluri-age structured. Data regarding tree size and their frequency distribution in these forest types are registered.

Key words: forest, tree layer structure, height, diameter.

Comana forest located in the Romanian Plain, south of Bucharest, is well known due to its large dimensions (about 10 000 ha), which actually is quite rare in the plains from Romania and Europe in general. It has a degree of nature reservation close to the optimal, the status of preservation being provided during the latest years by the forest management.

Being at the contact area between silvosteppe and forest plain dominated by Quercus species, and on the lower terraces, wet, sometimes even marshy, of the Neajlov river, habitats with particular microrelief and microclimate developed, which differentiate rather homogeneous and specific units within the forest massif, such as Fântânele forest. This forest is located at the lowest altitude and very close to the confluence of the Câlniştea and Neajlov rivers, which provides a pedological and hydric regimen privileged compared to the rest of the area.

The botanical investigations started at the beginning of the century and continued until the present day showed a wide floristic diversity with over 1 200 species (Panţu, 1900–1912, 1935; Negrean, 1968; Tarnavschi *et al.*, 1974) of which 330 are present in Fântânele forest (Paucă-Comănescu *et al.*, 2000).

The surveyed area, although of small dimensions (200 ha), is noticed not only by the richness of species, and by the interesting and very different ecosystem structures in which the trees are the backbone of the ecosystem; they differ not only by their taxonomic affiliation, but also by the manner of association, by the numeric or biomass proportion, as well as by age or individual size.

MATERIAL AND METHOD

Fântânele forest was first "decomposed" according to the main dominant forms of microrelief, then we have identified the main types of forest; some of them could be framed phyto-sociologically, others just reflected the current stage of anthropization due to forest management interventions in time.

The elementary investigation units were circular in shape, with areas of 500 sq.m, while the number of replicates differed from 2 to 5 according to the surface covered by the type of surveyed forest. The species mentioned in tables are only those observed on the surveyed plots.

The size of the trees was determined by measuring the diameter and height with a ruler for circumference and with the Bitterlich relascope for height. The volumes were determined with the dendrometric tables (Giurgiu *et al.*, 1972).

RESULTS AND DISCUSSION

Fântânele forest (Comana forest department, UP I Călugăreni, u.a. 76–85), by its position, in Neajlow flood plain and in the bordering plains (Găvanu-Burdea and Burnas), on coordinates 26°00' E longitude and 44°10' N latitude, is noticed due to several habitats, some wet ones – in the temporary and permanent pools as well as on the banks of the slow flowing river, others on low terraces – crossed by the 22 springs (the "fântânele" from the area), to adjacent kneaded slopes and, finally, to plateau habitats, which are in fact prolongation of the local plain. The difference in altitude is 44 m (between 46 and 90 m).

The types of forest identified in the mentioned habitats are shown in Table 1. We mention that the forests developed by meso-xerophilous species cover the high land (75–90 m), where the temperate-continental plain climate is strongly felt (Fig. 1). The composition and structure of the tree layer change on the slopes (60–80 m), next to the plain mixed deciduous forests with oak (*Quercus robur*) being observed plain mixed deciduous forests with common oak (*Q. polycarpa*), which are usually met at higher altitudes in Romania, starting with the undercarpathian hills, this being the lower limit of their ecological expansion. Most frequently, however, are lime forests, which are forests derived from oak forests-plain mixed deciduous forests and from plain mixed deciduous forests, following the preferential cut of *Quercus* trees by the local forest management. These forests are noticed by the presence of many ash species (4 of the 5 existing in the country), some of them in high numbers. *Fraxinus angustifolia* is dominant due to flood plain conditions, but sporadic individuals of *F. palliasiae* were observed, particularly on the low areas and at the foot of the slopes, where they alternate with *F. coriariifolia*. In the higher part, the plain mixed deciduous forests with common oak, *F. excelsior* replaces almost completely the other ash species. At the foot of the slope, besides ash forests, typical oak forests form due to a surplus of humidity;

on the temporary brooks and particularly on the long-time deserted meanders from the first terrace, poplar groves and ash forests form, while in the puddle areas with almost permanent water, willow galleries developed.

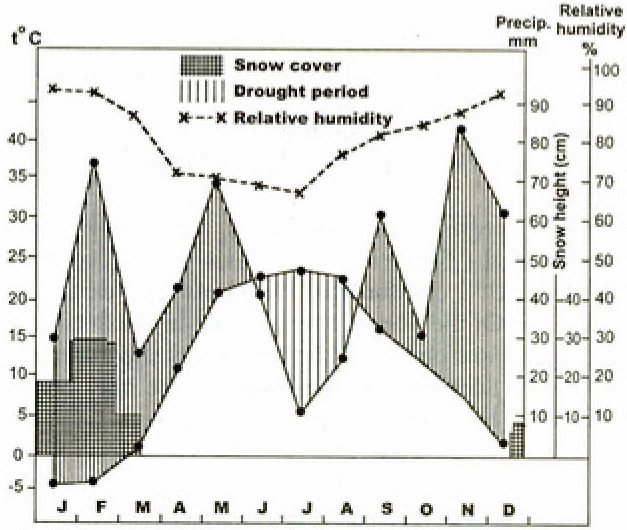


Fig. 1 – Climadiagram type Gaussen (synthetic data for Comana climate).

According to the climatic diagram of Gaussen (Fig. 1), the regional macroclimate is characterized by a steep rain misbalance, with a long end extremely severe draught period (about 3 months, from May till August), with a deficit that may reach 40 mm/month during the summer, which means a lack of almost 1 ton of water for the soil down to the depth of 1 m. The graph also shows a strong Mediterranean influence manifested by the existence of two periods of high rains, almost equal, one in late spring and one in late autumn. The draughty character of the climate is also revealed by the low value of De Martonne's index of aridity, equal to 26 (average annual rainfall 560 mm, average annual temperature 11.2 °C). The general climatic influences are reflected in the presence of species and species groups with more or less xerothermal character, particularly at the limit of the forest massif, where besides the basic species, *Quercus robur*, other species were observed, such as *Q. cerris, Q. frainetto, Fraxinus ornus* or *Ruscus aculeaetus*, etc. The bottom line is that we have there a mild continental climate with Mediterranean influence, with a trend towards excessive, characteristic to the boundary area between forest plain and silvosteppe.

Compared to the general climate, the presence of the pools and of Neajlov branches changed both the air humidity and the water supply to the ground, even during the periods of high draught (as it was in 2000), when the permanent water pooles dried out and turned into moist land which you could step on. This additional humidity explains, on the one hand, the presence of mesophyll tree species and, on the other hand, the large size of the trees and their longevity, except some species, particularly the poplar and the oak.

Soil distribution in the surveyed forests reflects the same water regimen determined by the proximity of the water table and by the microclimatic changes, distinguishing 2 main directions of evolution: a) the zonal one, from argiluvic and cambic classes, with deep reddish brown soil, eubasic and mesobasic, semigleic or gleic and b) the hydromorphous one, with humid-phreatic soil, large edaphic, with long annual floods, up to submerged gley soil (corresponding to the sediment of the pools).

The structure of forests, seen horizontally, shows the existence of 5 main categories, which cover the following areas: plain mixed deciduous forests 25%, secondary lime forests 45%, oak forests 15%, poplar groves and ash forests 14%, willow forests only 1%. Of the 40 tree species cited in the botanic surveys (Paucă-Comănescu *et al.*, 2000), we observed 26 tree species and 14 brush species, but in the elementary plots of dendrometric measurement we only observed 20 species (Table 1), the other ones having fewer, sporadic specimens.

Table 1

Biometrical data of constitutive species in different forest types

Species	Age (year)		Freq.	Diameter (cm)		Height (m)	
	I	II	%	Mean	Max.	Mean	Max.
1	2	3	4	5	6	7	8
1. Typical mixed deciduous plain forest							
Quercus robur	160	60	2.0	90.0	90	29.0	29
Fraxinus angustifolia	80	-	6.1	28.7	30	20.2	22
Ulmus minor	-	50	14.3	11.4	26	9.0	19
Tilia tomentosa	-	60	14.3	8.0	25	2.5	15
Acer campestre	-	50	49.0	9.5	25	6.0	13
Carpinus betulus	-	60	10.2	8.0	16	5.5	10
Malus sylvestris	-	50	4.1	6.5	7	5.0	8
2. Plain mixed deciduous forest including common oak							
Quercus polycarpa	140	80	4.0	61.0	61	24.5	25
Tilia tomentosa	100	60	36.0	42.8	48	21.5	22
Carpinus betulus	-	40	4.0	11.0	16	7.0	12
Acer campestre	-	30	36.0	5.7	8	5.1	9
Sorbus torminalis	-	40	16.0	3.8	7	3.5	6
Malus sylvestris	-	40	4.0	8.0	10	6.0	8
3. Plain mixed deciduous forest enriched in lime							
Quercus robur	110	50	2.4	34.0	34	21.5	22
Fraxinus angustifolia	100	50	4.7	43.0	48	24.0	25
Tilia tomentosa	100	50	2.4	44.3	47	24.0	25
Carpinus betulus	100	40	42.9	23.3	45	17.0	24
Acer campestre	-	40	2.4	8.0	10	6.5	8

Table 1 (continued)

1	2	3	4	5	6	7	8
4. Secondary lime forest including deciduous species							
Quercus robur	130	-	2.2	59.0	60	27.5	28
Fraxinus angustifolia	120	-	2.2	58.0	60	27.5	
Tilia tomentosa	100	60	84.5	18.8	46	16.0	
Carpinus betulus	-	60	2.2	27.0	30		
Acer campestre	-	60	8.9	14.8	17		
5. High plain secondary lime forest							
Quercus robur	130	70	2.6	53.0	54	26.5	27
Fraxinus pallisiae	110	60	2.6	49.0	50	25.3	26
Tilia plathyphyllos	100	60	79.0	18.4	61	15.0	27
Acer platanoides	-	40	2.6	14.0	15	12.5	15
Acer campestre	-	40	10.6	5.8	8	5.0	7
Carpinus betulus	-	60	2.6	32.0	20	17.5	20
6. Secondary lime forest on slope							
Quercus robur	120	65	2.9	58.0	58	20.0	21
Tilia plathyphyllos	90	60	47.1	27.1	55	16.0	23
Ulmus minor	-	35	2.9	4.0	6	5.0	7
Acer campestre	-	30	47.1	5.1	8	4.8	8
7. Intermediate oak-mixed deciduous forest							
Quercus robur	125	70	13.5	50.6	68	25.5	29
Fraxinus angustifolia	120	70	7.1	30.3	36	21.0	23
Fraxinus pallisiae	-	70	2.7	21.5	22	17.5	19
Tilia tomentosa	-	65	13.5	15.8	26	14.0	20
Carpinus betulus	-	65	10.8	19.3	31	12.0	17
Acer campestre	-	60	46.0	7.9	18	6.0	14
Acer tataricum	-	60	5.4	18.0	21	10.0	11
8. Plain oak forest in ground of slope site							
Quercus robur	-	60	52.6	22.1	35	17.5	21
Fraxinus angustifolia	-	60	10.5	19.0	39	18.6	23
Fraxinus pallisiae	-	60	2.7	4.0	5	4.0	6
Tilia tomentosa	-	50	5.3	5.0	6	3.5	5
Ulmus minor	-	60	2.6	7.5	9	5.5	8
Carpinus betulus	-	60	10.5	13.8	23	9.0	14
Acer campestre	-	40	13.2	10.2	16	7.0	11
Pyrus pyraster	110	-	2.6	46.0	46	21.0	21
9. Poplar forest including deciduous species on low terrace							
Populus alba	120	70	46.8	48.2	90	33	
Fraxinus angustifolia	-	40	17.0	6.4	16	15	120
Ulmus procera	-	40	31.9	8.9	22	18	120
Tilia cordata	-	4–	2.2	7.0	8	9	
Malus sylvestris	-	60	2.2	15.0	15	12	
10. Poplar forest in depression site							
Populus alba	140	80	38.2	60.5	108	30.0	34
Fraxinus angustifolia	-	50	11.8	9.5	26	8.5	20
Tilia plathyphyllos	-	50	8.8	10.3	14	6.0	8
Carpinus betulus	-	50	5.9	6.5	8	5.0	7
Acer campestre	-	40	5.9	4.0	5	4.0	5

Table 1 (continued)

1	2	3	4	5	6	7	8
Ulmus procera	-	60	23.5	10.8	30	8.2	21
Ulmus laevis	-	40	5.9	14.0	10	9.0	10
11. Ash forest in depression site							
Fraxinus excelsior	100	60	7.8	38.0	46	24.0	27
Fraxinus angustifolia	-	60	66.7	21.7	38	17.0	24
Fraxinus pallisiae	100	60	17.6	17.8	46	14.5	26
Ulmus procera	-	40	5.9	6.7	9	5.0	7
Acer tataricum	-	40	2.0	8.0	9	5.0	6
12. Willow galleries in pool site							
Salix alba	-	35	100	30.0	58	10.5	15

The mentioned species grow in a mosaic and form intimate mixtures in most cases. Only seldom we observed their aggregation in small size biogroups, usually in phytocoenoses dominated by a single tree species.

Unlike the spatial distribution of the species, which is homogeneous and quite uniform, the distribution of individuals within the same species was largely grouped, the main cause being the origin, almost generalized, from shoots. In average, from a single stump (primordial individual), resulted 6 lime trees (with a peak of 26), 1.7 ash trees, 1.4 oaks, very seldom the shoots being separated up to the basis. The highest shoot frequency was observed in the lime, in all 3 species that were observed, ranging between 40 and 65%, while the lowest, below 30%, was observed in the other tree species.

The tree layer has a cover of 85–100% according to canopy projection and (0.6) 0.7–0.8 (0.9) by trunk projection. The high cover of the tree layer is due mainly to the oak – with its large canopy (15–35 m in diameter and 500–1500 cb. m canopy volume), then to the lime, ash and common maple and hornbeam, which despite the small canopies compensate by the large number (80–400 individuals/ha). In the wet area, the monodominant poplar, ash or willow forests give the general aspect, the other components playing a minor role.

The most obvious difference in the horizontal structure of the forest was observed in the variation of trunk diameter, essentially due to species composition and, within the same species, by the age and vigorousness (Table 2). As it can be observed, in the plain mixed deciduous forests and in oak forests, the main, basic species cover the diameter classes of 25–50 cm and over 50 cm, the codominant species cover the middle classes, while the middle and small size species present in all phytocoenoses of mixed forest, displayed the smallest diameters, 11–25 cm, even under 10 cm. This situation was also observed in the monodominant forests of poplar or ash, as well as in the willow galleries, where this single species covers all the diameter categories.

The structure of the forest – by the age categories that were determined by the arrangement, and by their correlation with the diameter categories – is very different between habitats and by the characteristics of the species or of the forest management.

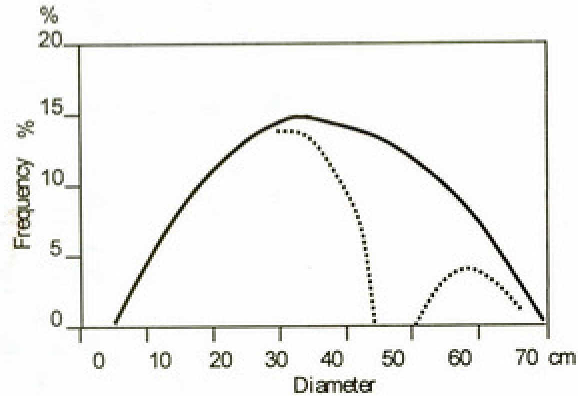


Fig. 2 – Structure by diameter of trees in a willow gallery.

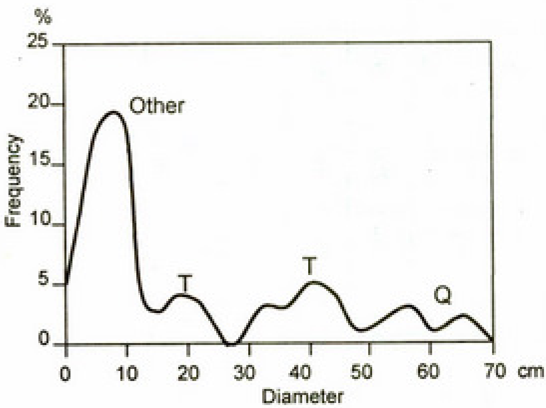


Fig. 3 – Structure by diameter of trees in a mixed deciduous plain forest. (T = *Tilia tomentosa*, Q = *Quercus robur*), other = the other associated species).

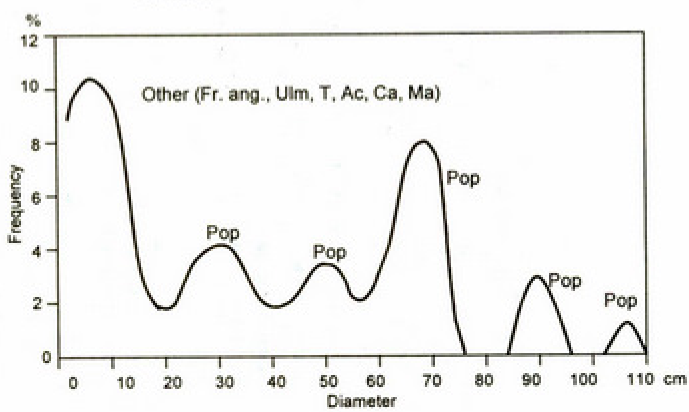


Fig. 4 – Structure by diameter of trees in a poplar forest in depression site (Ulm = *Ulmus div.*, T = *Tilia tomentosa*, Ac = *Acer campestre*, Ca = *Carpinus betulus*, Pop = *Populus alba*.

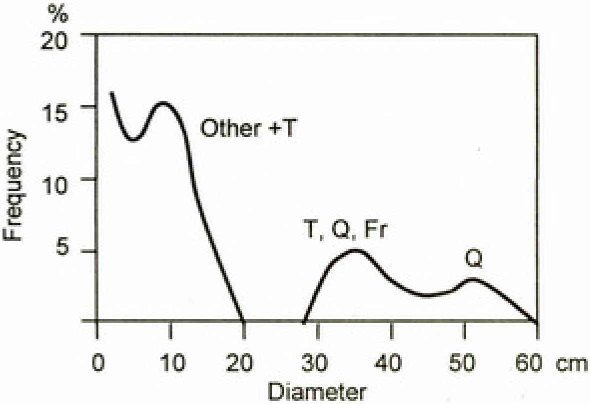


Fig. 5 – Structure by diameter of trees in a mixed deciduous plain enriched in Tilia.

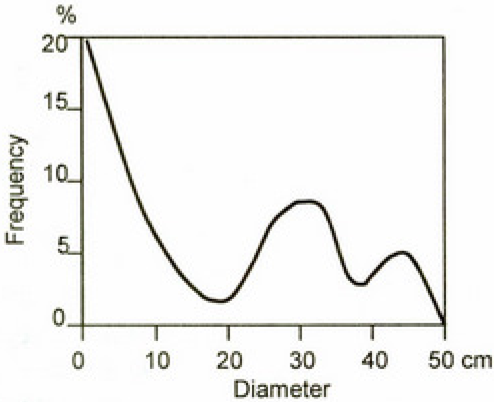


Fig. 6 – Structure by diameter of trees in an ash forest in depression site.

In the case of willow forests, usually present in monodominant phytocoenoses, we determined an even-age structure – with a single age category, differentiated biometrically according to the individual growth conditions (Fig. 2). The structure was much more complex in the other types of forest.

The absolutely dominant structure in most surveyed brush stands was the pluriage one, consisting of several tree generations. This structure, typical pluri-age was observed in the plain mixed deciduous forests, in the oak forests and in the secondary lime forests – the most spread phytocoenoses in Fântânele (Fig. 3). We determined 5–6 successive tree generations, similar to waves arranged in decreasing steps, each generation being represented by 1–2 dominant species; the codominant or dominated species (common maple, hornbeam, lime) are located at the beginning of the curve, while the oak and ash are located at the end of the curve; the lime usually takes the central position, but was spread in all age categories belonging to all diameter categories.

A characteristic of Fântânele forest is that lime is a common element within the basic forest types, the ecological unit that provides the largest numerical and biomass proportion in the tree layer, with imposing individuals, reaching together with the oak and ash the top layer, but also with many individuals of modest dimensions growing together with the trees of the second layer, where the common maple and the hornbeam are dominant.

Two patterns of pluri-age structure can be differentiated: a) – the one including very old and numerous generations of trees (about 6 generations), with exceptional development, for example a plain mixed deciduous forest, an oak forest, a poplar forest (Fig. 4), and b) – the one including several young generations (about 3–4), in the plain mixed deciduous forests with a lot of lime and in the secondary lime forests, secondary structures developed by the intentional cut of *Quercus* trees and even of the older lime (Fig. 5).

In parallel with the general types of structure by age, there are at Fântânele cases of transition, with intermediary aspect, quasipluri-age (in ash forests) or quasi even-age (in the young oak forests or in the plain mixed deciduous forests with many oak individuals).

Table 3 shows the data on biomass accumulation expressed volumetrically.

We noted thus that species with low frequency, below 5%, as the oak, ash, lime sometimes, may form up to 75% of the volume of wood accumulated in the different types of forests that were surveyed. This reverse situation accounted by the large individual dimensions (up to 9 cubic meters/tree, in average 2.8 cubic meters/tree) is balanced by their low number, which is known in the structure of all populations. The largest dimensions were determined in the white poplar with a peak of 12.5 cubic meters/tree, the average being 3.5 cubic meters/tree.

The volume of wood accumulated in this forest does not differ too much from one population to another, ranging between 20–300 cubic meters/ha in oak (in plain mixed deciduous forests, lime forests and oak forests), 50–100 cubic meters in ash, 10–400 cubic meters in lime forests (in plain mixed deciduous forests), 500 cubic meters in ash forests, 400 cubic meters in willow forests and as exception, 1150–1250 cubic meters in poplar forests. These exceptional values of the poplar in the monodominant poplar forests is accounted not only by their old age, but also by the optimal habitat, fertile soil and permanent water supply, which provided a great development of the volume and a high density of the individuals. Because the poplar wood is light, spongy, with very low apparent density (g/cmc), the accumulated biomass was much lower compared to the volume and thus it is closer quantitatively to the accumulation recorded in other species.

Table 3

Tree volume of different species in different analyzed forest types

Forest type	Size units	Oak	Lime	*F. pallisiae*	*F. coriariifolia*	Elm	Maple	Hornbeam	Common maple	Wild pear	Wild service	*Fr. ornus*	Poplar	Total Mean
Typical mixed deciduous plain forest	m^3/tree	3.400	0.524	-	-	0.002	-	0.568	0.026	-	-	-	-	0.676
	m^3/ha	272.0	272.6	-	-	0.2	-	34.1	2.1	-	-	-	-	581.0
	%	46.8	46.9	-	-	0	-	5.9	0.4	-	-	-	-	100
Typical mixed deciduous plain forest -Young-	m^3/tree	0.237	0.076	0.278	0.225	-	-	0.035	-	0.115	-	-	0.345	0.101
	m^3/ha	46.6	86.6	14.8	7.4	-	-	1.4	-	1.4	-	-	6.9	165.1
	%	28.2	52.5	8.9	4.5	-	-	0.9	-	0.8	-	-	4.2	100
Secondary lime plain forest with flower ash	m^3/tree	-	0.371	-	-	-	0.030	0.375	0.078	-	0.003	0.114	-	0.308
	m^3/ha	-	222.3	-	-	-	1.2	45.0	7.8	-	0.6	6.3	-	283.2
	%	-	78.5	-	-	-	0.4	15.9	2.8	-	0.2	2.2	-	100
Lime-mixed deciduous plain forest	m^3/tree	1.893	0.462	1.892*	-	-	-	0.010	0.008	-	-	-	-	0.575
	m^3/ha	39.7	256.7	30.1	-	-	-	0.4	2.8	-	-	-	-	329.7
	%	12.0	77.8	9.3	-	-	-	0.1	0.8	-	-	-	-	100

It is well known that a large number of ages, thus a pluri-age forest, results in a strong vertical organization of the canopy on several levels, which means the amplification of the diversity of structure. The frequency curves from Figs. 3–5 and the data from Tables 1–2 show the high degree of diversification of this forest; the strong vertical undulation of the heights, with several "age waves" is obvious particularly in the plain mixed deciduous forests, in oak forests and in the secondary lime forests, being less obvious in poplar forests, ash forests, lacking all together in the willow forests. The tables also show a clear differentiation of the species by height. The highest canopies belong to *Quercus* forests, ash forests, partly to lime forests, and in the poplar forests to the dominant species, the other species forming the lower layers.

Most trees forming the canopy are 5–15 m high, the dominant and predominant species, lower in number, reach 14–21 m, while the vertical differentiation peaked in the dominant species with 21–29 m.

The genetic differences between the species, obvious in the surveyed cases, based on site differences (climatic and edaphic) resulted in a particular structural biodiversity, rarely observed in the Romanian Plain (Table 2).

CONCLUSIONS

The structural diversity of the tree layer in Fântânele forest peaked in the region due to the topic diversity. It is illustrated both by the rich specific composition resulted from the development in this area of wooden species originating from very different areas, and from the spatial structure of the trees (as location, proportion, both horizontally and vertically). We noted thus a wide vertical expansion of the layers, followed by the differentiation of the species by eco- and coenofunctional categories, the strong undulation of the canopy, the occupation and maximal utilization of the ecological niches with biologically compatible species; an optimal ecosystemic integration of the mesophilous species from hilly areas and from sub mountain areas (*Quercus polycarpa, Fraxinus excelsior*), with some xero-mesophilous and xerothermal species, sometimes with the replacement of the basic species (*Quercus robur* and *Fraxinus angustifolia*) by some species from eastern areas *(Fraxinus coriariifolia*) or southern *areas (Fraxinus ornus, Q. cerris, Q. frainetto*), in variable proportions, appropriate to the habitat.

REFERENCES

1. Bândiu C., 2000, *Estetică forestieră*, Edit. Timpul.
2. Doniță N, Chiriță C., Stănescu V., 1990, *Tipuri de ecosisteme forestiere din Romania*, Ed. Centrului de Propagandă Agricolă, București.

3. Giurgiu V., Decei I., Armăşescu S., 1972, *Biometria arborilor şi arboretelor din România*. Ed. Ceres, Bucureşti.
4. Negrean G., 1968, *Contribuţii la flora României*, Stud. Cercet. Biol., Ser. Bot., **20**(4): 333–336
5. Paucă-Comănescu Mihaela, G. Negrean, Maria Paspaleva, M.Tălpeanu, N. Doniţă, C. Bândiu, Marilena Onete, 2000, *Pădurea Fântânele (Oc. Silvic Comana) – arie de conservare a biodiversităţii native a pădurilor şi zonelor umede din Câmpia Română*, Ocrot. Nat., 44, in press.
6. Paucă-Comănescu Mihaela, Bândiu C., Doniţă, G.Negrean, Tutunaru V., Onete Marilena, 2000, *Stabilirea categoriei de rezervaţie a ariilor protejate constituite la Călugăreni, Padina Tătarului şi Comana (Ocolul Silvic Comana) în vederea realizarii intervenţiilor corespunzatoare în arboretele respective*, Contract 334/2000, Direcţia Silvică Târgovişte.
7. Tarnavschi I., Paucă A., Andrei M., Cristurean I., Ionescu V., Lungu L., Nedelcu G.A., Petria E., Popescu A., Rădulescu D., Mitroi N., Şerbănescu-Jitariu G., 1973, *La flore du complexe de Comana*, Acta. Bot. Horti Bucurestiensis, p. 239–286, Bucureşti.

Received May 15, 2000.

* *Institute of Biology*
Splaiul Independenţei, 296
79651 Bucharest

** *ICAS*
Şos. Ştefăneşti, 128
Bucharest

"EUROCOMPETENCES TRANSFER IN VOCATIONAL GUIDANCE FOR YOUNG SPECIALISTS IN BIOSCIENCE FIELD" is a three years European pilot project, funded under the European Leonardo da Vinci program, the promoting organisation being University POLITEHNICA Bucharest / Human Resources Training Centre (HRTC). The partners are putting together their experience and expertise, combining different points of view and contributions as they are from *diverse institutional contexts*: **Universities** from Romania, the United Kingdom, Germany and Slovakia, which bring their capacity in building-up performant teaching materials; a professional Society / **NGO** (Romanian Society for Biotechnology and Bioengineering), a Foundation / **NGO** (University Foundation CERA), and a **Research Institute** (Research Institute for Chemistry) which bring the points of view of *project users and the needs of the target group*; finally, a **SME** (Pluri Consultants), specialised in human resources & career consulting.

The members of the project consortium are preparing:

– A *transnational information network* for labour force demand monitoring in the companies acting in the bioscience field, at European and regional level, in line with qualitative and quantitative criteria;

– *An identification methodology*, elaborated in accordance with the European models used for vocational training and guidance, for the expected and available competencies in the above mentioned field;

– *Curricula and training modules* for young people, who need professional knowledge to be applied in the bioscience companies (printed and multimedia courses, open distance learning systems);

– *Reference work instruments* (word glossary, characterisation of needed professional competences, guide of demanded jobs) for the domain of interest.

The project seeks to develop new and innovative models with an European dimension for vocational training and guidance dedicated to the bioscience domain, which will improve the quality of training in order to anticipate the future qualification requirements. (For more information the Project web site is `www.cpru.pub.ro/leonardo`).

Melania-Liliana Arsene

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La «Revue roumaine de biologie – Série de biologie végétale» publie des articles originaux dans les domaines suivants de la biologie végétale: biologie moléculaire, cytologie, cytogénétique, morphologie, physiologie, génétique, microbiologie, systématique, chorologie, géobotanique, écologie végétale et phytopathologie. Le sommaire est complété par les rubriques: 1. La vie scientifique, qui traite des manifestations scientifiques dans le domaine de la biologie (symposiums, conférences, etc.); 2. Comptes rendus des plus récentes parutions dans la littérature.

Les auteurs sont priés de présenter leurs articles en double exemplaire et espacés à double interligne. Le contenu des articles sera introduit sur des disquettes dans un langage connu, préférablement Word 6.0. La composition et la mise en vedette seront faites selon l'usage de la revue: caractères de 11/13 points pour le texte, de 12/14 points pour le titre de l'article et de 9/11 pour les annexes (tableaux, bibliographie, explication des figures, notes, etc.) et le résumé en anglais de 10 lignes au maximum, qui sera placé au début de l'article. Il est obligatoire de spécifier sur les disquettes le nom des fichiers ainsi que le programme utilisé.

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La responsabilité pour le contenu des articles revient exclusivement aux auteurs.

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CONSTANTIN TOMA, RODICA RUGINĂ, *Anatomia plantelor medicinale. Atlas (Anatomie des plantes médicinales. Atlas)*, Editura Academiei Române, 1998, 320 p., 155 pl., ISBN 973-27-0601-5

ISSN 0250-5517

REV. ROUM. BIOL.–BIOL. VÉGÉT., TOME 46, N° 1, P. 1 – 84, BUCAREST, 2001