

784-გ. B
2005



ISSN 1512-2123

საქართველოს მეცნიერებათა აკადემიის

მაცნე

ბიოლოგიის სერია **B**
Biological series

2005
No. 4
Vol. 3

PROCEEDINGS
of the Georgian Academy of Sciences



ISSN 1512 – 2123

საქართველოს მეცნიერებათა აკადემიის

მაცნე

ბიოლოგიის სერია **B**
Biological series

2005
No. 4
Vol. 3

PROCEEDINGS

of the Georgian Academy of Sciences

Tbilisi, Georgia
2005

EDITOR IN CHIEF: Malkhaz M. Zaalishvili

EDITORIAL BOARD:

Aleksidze N.

Beridze T.

Chanishvili T.

Eliava I.

Grigolava M. (Executive Secretary)

Kajaia G.

Khurashvili B.

Kvesitadze G.

Kvinikhidze G.

Lezhava T.

Mchedlidze G.

Nakhutsrishvili G.

Sanadze G.

Shatilova I.

Tumanishvili G. (Associate Editor)

Ugrekheldidze D.

Zaalishvili T.

GRAPHIC AND COMPUTER DESIGN:

Devishvili T.

Devdariani M.

To order your copies, please send to:

Georgian Academy of Sciences,
Department of Biology
52, Rustaveli Avenue, Tbilisi, 0108, Georgia
Tel. +995-32 93-58-92
Fax: +995-32 93-58-92
E-mail: bio@gw.acnet.ge
www.acnet.ge/matsne/biology

Journal founded in 2001

ISSN 1512 – 2123

CONTENTS

Biochemistry	
Bliadze G., Chubinidze V. Qualitative and Quantitative Analyses of Anthocyanins in the Fruits of Different Peach (<i>Persica vulgaris</i> . Mill.) Cultivars.	1
Biophysics	
Mardaleishvili M., Migineishvili N., Sultanishvili N., Gamrekeli D. Adsorption Kinetics and Immobilization of Acid Phosphatase on Diatomite.	5
Botany	
Djugeli T. List of Representatives of Genus <i>Serratula</i> L. (Compositae) of the Caucasus Flora.	10
Lachashvili N., Khachidze M. The Vegetation of Argillaceous Badlands of Iori Plateau (East Georgia).	14
Cell Biology	
Atanelishvili I., Jimshitashvili N., Ratiani K., Tabatadze M., Gordeziani M. Stability of 11 ³⁽¹⁾ <i>Y Nocardiophasis Dessonvillei</i> Cells under the Influence of Different Agents.	22
Cytology	
Chiladze N., Mchedlidze T., Modebadze I., Tumanishvili T., Dzidziguri D. The Morphological Study of the Acidophilic Cells Within the Digestive Gland Tissue of <i>Helix Lucorum</i> .	26
Ecology	
Dzotsenidze N. For Study of Biology of Seed Germination of the Caucasus Fir (<i>Abias nordmanniana</i>).	31
Genetics	
Tabatadze N., Dadunashvili E., Jokhadze T., Zosidze N., Lezhava T. Functional Characteristics of Chromosomes in Patients with Alzheimer's Disease and Vascular Dementia.	35
Tadumadze N., Bablshvili N., Nibladze N., Sulaberidze S., Buadze T., Monaselidze J. Effect of Mercury Chloride on Conformational Changes of Chromatin.	40
Hydrobiology	
Tsiskarishvili L., Tsiskarishvili M. Chemical Composition of the Water and Plankton's Primary Production of the Lake Kartsakhi (Khozapini).	43

Immunology

- Nagervadze M., Diasamidze A., Akhvlediani L., Gogitidze T., Dumbadze G., Khalvashi N. **Correlation between Blood Rh Systems Group Antigens with Pulmonary Tuberculosis.** 47

Microbiology

- Antia I., Natroshvili N., Balanchivadze M., Batiashvili E. **Study of Avian Pathogenic *Escherichia coli*.** 52
- Laskhishvili M., Kutateladze L., Zakhariashvili N., Glonti N., Khokhashvili I., Jobava M. **Isolation and Identification of Microscopic Fungi from Salty Soils.** 55
- Lomtadidze Z., Kotia N. **Peculiarities of *Nocardia dassonvillei* Polysaccharide Metabolism.** 60

Palynology

- Connor, S.E., Kvavadze, E.V. **Climatic and Human Influences on Vegetation Dynamics around Tbilisi over The Past 6000 Years.** 64

Plant Physiology

- Aleksidze G., Tevzadze N., Gabatashvili B., Gigolashvili G. **Study of Lectin Activity in the Nuclear Protein Fraction of the Wheat (*T. Aestivum* V. *Hostianum*) Leaves.** 77

Zoology

- Palavandishvili N., Imnadze V. **The Seasonal Changes of Chemical Contents of Black Sea Mussels (*Rapana Thomasiana* Crosse) Bodies.** 83

QUALITATIVE AND QUANTITATIVE ANALYSES OF ANTHOCYANINS IN THE FRUITS OF DIFFERENT PEACH (*PERSICA VULGARIS.MILL.*) CULTIVARS

BLIADZE G.¹, CHUBINIDZE V.²

¹Research Institute of Gardening, Viticulture and Winemaking, Georgian Academy of Agriculture
²S.Durmishidze Institute of Biochemistry and Biotechnology, Georgian Academy of Sciences

(Received March 15, 2005)

Abstract

Qualitative and quantitative analyses of anthocyanins in the peels and pulps of fruits of different peach cultivars were studied. The fruits of cultivars with intensive coloration are distinguished by high content of anthocyanins. The ripe fruit peels are especially rich in anthocyanins, as pigments concentrate just in the peels. Different cultivars, as well as peels and pulps of the same cultivar differ by quantitative content of anthocyanins.

Introduction

Among natural polyphenol compounds anthocyanins pigments take significant place. Due to their high biological activity and wide distribution in plants, studies on anthocyanins intensively were carried out. Natural variety of coloration is induced just by anthocyanins. Also they take important role in vital processes of plants. Anthocyanins occur in different organs of plant: leaves, flowers, stems, roots and especially colored pulps. Colors range of anthocyanins depends on the amount and position of functional groups, as well as on pH of cell juice. Along with other flavonoid compounds anthocyanins are characterized with high P vitamin activity [Bridle & Timberlake, 1997; Cao et al., 2001; Chandra et al., 2001; Lee, 2002].

Anthocyanins always occur as glycoside, free aglycon was not found. Their great number is caused by the nature of glycated sugar, quantity, position of bond with aglycon, kind of bond etc. They occur as monoglicoside and diglicoside, as well as bioside [Gao & Mazza, 1994]. Usually in nature simple anthocyanins (glycosides) occur, although there are complex, acylated anthocyanins.

Materials and Methods

Researched material was collected on experimental plot of the Institute of Gardening, Viticulture and Winemaking, in Skra.

For quantitative and qualitative analyses peels and pulps of the fruit were separated and extraction in 1% hydrochloric acid methanol was carried out at room temperature in the dark [Tsiklauri, 1975]. Extracts were combined, filtrated and evaporated to small volumes on rotational evaporator in vacuum. Received samples were drip on chromatographic paper. Chromatography was carried out in different solvent systems: 1) butanol : acetic acid : water - 4:1:5 and 2) 5% acetic acid (Table1).



2378

Chromatograms were dried and the quantity of received anthocyanins was verified under treatment with ammonia vapor and without (during the treatment with ammonia vapor at existence of O-hydroxyl groups red color becomes violet). For quantitative studies of anthocyanins spectrophotometrical method was used [Harborne, 1958].

Exact weights of studied materials (peel and pulp of the fruit) were extracted by acidified methanol till complete extraction; extractions were put in rotational vacuum evaporator in cuvette. Intensity of absorption was conducted on spectrophotometer (CΦ-26, 515-533 nm). Control curve was plot by crystal cyanidin - 3 - monoglicoside. Data of analysis are presented in Table 2.

Results and discussion

As is seen from Table 1. studied cultivars qualitative contents more or less differ from each other (amount of spots). Fruits of cultivar "Bel" don't contain anthocyanins at all. According to obtained data (chromatographical, spectral, changes of coloration of anthocyanins in ammonia medium) it was established that peach fruits anthocyanins mainly are presented by monoglicosides of peonidin and cyanidin .

As is seen from Table 2. different cultivars, as well as peels and pulps of the same cultivar differ by quantitative content of anthocyanins. The fruit peels are especially rich in anthocyanins, as pigments mainly concentrate just in the peels.

Table 1. Anthocyanins of different cultivars of peach fruits (*Persica vulgaris*. Mill.) Solvents systems: I. butanol : acetic acid : water - 4: 1: 5. II. 5 % acetic acid.

cultivars	analysed material	Solvents and R _F meanings										Spots amount
		I	II	I	II	I	II	I	II	I	II	
Krimchaki	Peels	0,54	0,67	0,27	0,25	0,51	0,49	0,55	0,26	0,11	0,12	5
	Pulps	0,38	0,18	0,04	0,23	0,08	0,23	0,09	0,38	0,45	0,28	5
Alberta	Peels	0,55	0,70	0,31	0,31	0,63	0,57	0,59	0,34	0,19	0,17	5
	Pulps	0,51	0,62	0,03	0,03	0,09	0,25	0,09	0,31	0,47	0,32	5
Bestavashvili	Peels	0,78	0,80	0,39	0,39	0,55	0,32	0,55	0,57	0,25	0,29	5
	Pulps	0,72	0,85	0,31	0,31	0,41	0,49	0,49	0,63	0,38	0,35	5
Bel	Peels	0,49	0,51	0,18	0,18	0,37	0,38	*	0,35	0,17	*	3
	Pulps	-	-	-	-	-	-	-	-	-	-	-
Geoqchai	Peels	0,45	0,32	0,24	0,24	0,44	0,43	0,33	0,21	-	-	4
	Pulps	0,57	0,35	0,32	0,32	0,09	0,26	0,09	0,34	-	0,10	3
Vajuri	Peels	0,65	0,71	0,31	0,31	0,71	0,67	0,72	0,57	0,25	0,39	5
	Pulps	0,41	0,37	0,03	0,03	0,03	0,30	0,05	0,21	-	-	4
Eristavis vardisperi	Peels	0,83	0,95	0,75	0,75	0,85	0,80	0,88	0,59	0,65	0,58	5
	Pulps	0,54	0,71	0,58	0,58	0,45	0,72	0,57	0,50	0,70	0,45	5

* Traces

Thus studied peach cultivars by qualitative and quantitative content of anthocyanins apparently differ from each other. Content of anthocyanins always dominates in the fruit peels. The fruits of those cultivars which have visible intensive coloration are distinguished by high content of anthocyanins.



Table 2. Absorption maximums of anthocyanins and total amount of compounds (calculated on 1 g of row material) in the different cultivars of peach fruits (*Persica vulgaris*. Mill.)

cultivars	analysed materials	N _{max} (nm)	total amounts of compounds (mg)
Krimchaki	Peels	525 -533	3,5
	Pulps	525 - 533	2,9
Alberta	Peels	515 - 533	1,1
	Pulps	515 - 530	0,9
Bestavashvili	Peels	525 - 533	2,0
	Pulps	525 - 533	2,1
Bel	Peels	520 - 530	1,5
	Pulps	-	-
Geoqchai	Peels	525 - 530	0,5
	Pulps	520 - 530	0,3
Vajuri	Peels	525 - 533	1,9
	Pulps	520 - 533	0,4
Eristavis vardisperi	Peels	525 - 533	3,7
	Pulps	525 - 533	2,5

References:

- Bridle P., Timberlake C.F. *Anthocyanins as natural food colours - selected aspects*. Food Chem., **58**, 103-109, 1997.
- Cao G., et al. *Anthocyanins are absorbed in glycosylated forms in elderly women: a pharmacokinetic study*. Am.J.Clin.Nutr., **73**, 920-926, 2001.
- Chandra A., et al. *Separation, identification, quantification method validation of anthocyanins in botanical supplement raw materials by HPLC and HPLC-MS*. J.Agric. Food Chem., **49**, 3515-21, 2001.
- Gao L., Mazza G. *Quantitation and distribution of simple and acylated anthocyanins and other phenolics in blueberries*. J.Food Sci., **59**, 1057-59, 1994.
- Harborne J.D. *Quantitative Studies of anthocyanins by method of spectrophotometry*. Biochem.J. **70**, 1, 1958.
- Lee H.S. *Characterization of major anthocyanins and the color of red-fleshed Budd Blood orange (Citrus sinensis)*. J.Agric.Food Chem., **50**, 1243-46, 2002.
- Tsiklauri G., *Antocyanins of the cherry-laurel medicinal fruits*. Bulletin of the Academy of Sciences of the Georgian SSR, **79**, 1, 1975.

ანტოციანების თვისობრივი და რაოდენობრივი ანალიზი ატმის სხვადასხვა ჯიშის ნაყოფებში.

ბლიაძე ვ.¹, ნუბინიძე ვ.²

¹მებაღეობის, მვენახეობისა და მეღვინეობის სამეცნიერო კვლევითი ინსტიტუტი,
საქართველოს სოფლის მეურნეობის აკადემია.

²ს. დურმიშიძის სახელობის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი,
საქართველოს მეცნიერებათა აკადემია.

(მიღებულია 15.03.05)

რეზიუმე

შესწავლილია ატმის სხვადასხვა ჯიშების ნაყოფებში ანტოციანების თვისობრივი და რაოდენობრივი შემცველობა კანსა და რბილობში ცალ-ცალკე ანტოციანიდების მაღალი შემცველობით გამოირჩევიან იმ ჯიშის ნაყოფები, რომელთა შეფერილობაც ინტენსიურია. ანტოციანიდებით განსაკუთრებით მდიდარია მწიფე ნაყოფის კანი, რადგან ძირითადად სწორედ მასშია კონცენტრირებული პიგმენტები. ანტოციანების რაოდენობრივი შემცველობით განსხვავდებიან არა მარტო ცალკეული ჯიშები, არამედ ერთი და იგივე ჯიშის კანი და რბილობი.

ADSORPTION KINETICS AND IMMOBILIZATION OF ACID PHOSPHATASE ON DIATOMITE

MARDALEISHVILI M.^{*}, MIGINEISHVILI N., SULTANISHVILI N., GAMREKELI D.

Department of Biophysics, Iv. Javakishvili Tbilisi State University

(Received July 1, 2005)

Abstract

Acid phosphatase adsorption kinetics and immobilization on diatomite has been investigated. The process develops as a one-step course with rapid adsorption of enzyme on the external surface of the carrier and its free penetration into carrier's wide pores. Activity of acid phosphatase immobilized on diatomite was measured. It appeared that specific activity of acid phosphatase immobilized on diatomite was by 21,4% higher than that of acid phosphatase adsorbed on silica gel.

Key words: biocatalyst, immobilization, specific activity, acid phosphatase.

Introduction

Development of new technologies requires introduction of new approaches in enzyme stabilization methods. Low stability of biocatalysts is one of the limiting factors for their industrial application. Hence, growing attention has been attracted to the investigation of protein denaturation processes. Immobilization of enzymes on solid carriers is one of the promising techniques for enzyme stabilization [Poltorak et.al 1987, Khokhlova, 2002].

The objectives of our investigation were examination of acid phosphatase adsorption kinetics on diatomite and the development of a new biocatalyst.

Materials and Methods

Experiments were carried out on a wheat sprouts acid phosphatase from FLUKA. Immobilization was performed on solid carriers: commercial hydrothermal and wide pore silica gel as well as on a natural diatomite. Protein amount was measured according to Lowry [Lowry et al., 1951], acid phosphatase activity was assessed quantitatively (the amount of inorganic phosphorus cleaved off the substrate) [De Duve et al., 1955]. Immobilization on solid carrier was registered as indicated by Poltorak [Poltorak, Vorobyova, 1966].

Adsorption of acid phosphatase on diatomite was conducted in cylindrical weighing bottles at room temperature. 1 ml of acid phosphatase in acetate buffer (0,5 mg/ml, pH 5.0) was

^{*} Corresponding author. E-mail: medea54@hotmail.com

added to 100 mg purified, dry carrier (granule size 0,8-1,2 mm). Adsorption was judged according to the decrease of the protein amount in the contact solution at certain time intervals.

Results and Discussion

Kinetic curve of acid phosphatase adsorption on diatomite is illustrated on Fig. 1. Kinetic curve of acid phosphatase adsorption on diatomite is represented in coordinates of linear equation [Poltorak et al 1982].

$$\frac{1}{E_{abs}} = \frac{1}{E_{max}} + \frac{1}{E_{abs} \cdot K \cdot E_0} \cdot \frac{1}{t} \quad (1)$$

where: E_0 - enzyme initial concentration; E_{abs} - amount of adsorbed enzyme; E_{max} - liminal adsorption value of monolayer filling; K - bimolecular constant of adsorption rate; t - time.

Fig. 2 represents kinetic curve of acid phosphatase on diatomite in linear equation coordinates. As indicated on the figure, the process of adsorption reveals a one-step feature. Enzyme molecules are rapidly adsorbed on the outer surface of diatomite easily penetrating into wide pores of the carrier. Calculated maximal adsorption value E_{max} equals 0.2mg/100mg of diatomite and adsorption rate constant K - $177 \text{ M}^{-1} \text{ sec}^{-1}$.

Study of acid phosphatase adsorption kinetics on diatomite of different dispersity revealed that the process does not depend on the carrier granule size (Fig.3). At the same time adsorption kinetics of acid phosphatase of various concentrations on diatomite of identical dispersity (0.8mm-1.2mm) has been studied. The respective calculated kinetic parameters of adsorption are given in Table 1.

Table 1. Kinetic parameters of adsorption of acid phosphatase of different concentrations on diatomite of identical dispersity (0.8mm-1.2mm)

N	acid phosphatase concentration, mg/ml	K, $\text{M}^{-1} \text{sec}^{-1}$	E_{max} , mg/100mg of diatomite
1	0,4	38,6	0,2
2	0,5	177,0	0,2
3	0,6	300,6	0,2

Table 1 clearly demonstrates that E_{max} does not depend on the enzyme concentration. Data are in good accordance with our assumption of easy adsorption of enzyme molecules on outer as well as inner surface of diatomite.

The immobilization of acid phosphatase on solid carriers (both of granules 0,8-1,2 mm) was achieved through 24 hour incubation of samples at room temperature. Finally, the activity of acid phosphatase adsorbed on silica gel as well as on diatomite has been measured. The values of specific activity of acid phosphatase is shown on Fig. 4. It is obvious that acid phosphatase adsorbed on diatomite reveals considerably higher activity (by 21.4%) than that of the enzyme immobilized on silica gel.

In conclusion, the possibility of development of a new biocatalyst of acid phosphatase immobilized on natural diatomite has been proved.

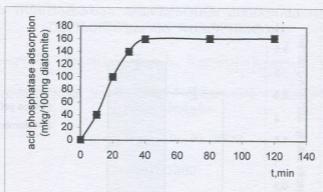


Fig. 1. Kinetic curve of acid phosphatase adsorption on diatomite.

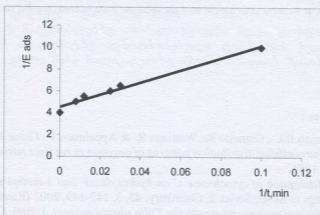


Fig. 2. Kinetic curve of Fig.1 in the coordinates of equation (1).

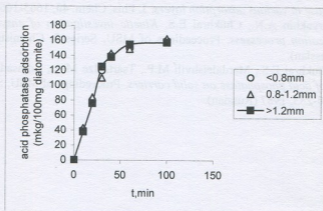


Fig. 3. Kinetic curve of acid phosphatase adsorption on diatomite of different dispersity.

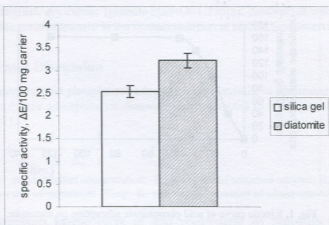


Fig. 4. Relative activities of acid phosphatase immobilized on solid carriers.

References :

- De Duve C., Pressman B.C., Gianetto R., Wattiaux R. & Appelmans F. *Tissue fractionation studies. Intracellular distribution patterns of enzymes in rat liver tissue.* Biochem. J., **60**: 604-617, 1955.
- Khokhlova T.D. *Adsorption of cytochrome C on hydroxylized and 3-methyl silylized silica gels.* Proceedings of MSU, Series 2, Chemistry, **43**, 3, 147-149, 2002 (Russian).
- Lowry O.H et al. *Protein measurement with the Folin phenol reagent.* J. Biol. Chem., **193**, 265-275, 1951.
- Poltorak O.M., Vorobyova E.S. *Adsorption modelling of biomembranes and activation of alkaline phosphatase in various adsorption layers.* J. Phys. Chem. **40**, 1665-1669, 1966 (Russian).
- Poltorak O.M., Pryakhin A.N., Chukhrai E.S. *Kinetic investigation of enzyme adsorption and immobilization processes.* Proceedings of MSU, Series 2, Chemistry, **23**, 6, 527-543, 1982 (Russian).
- Poltorak O.M., Chukhrai E.S., Mardaleishvili M.P., Tsartsidze M.A., Lomsadze B.A. *Adsorption kinetics of acid phosphatase on solid carriers.* Proceedings of MSU, Series 2, Chemistry, **28**, 3, 230-233, 1987 (Russian).

მშავა ფოსფატაზას ადსორბციის კინეტიკა და მისი იმობილიზაცია დიატომიტზე

მარდალეიშვილი მ., მიგინეიშვილი ნ., სულთანაშვილი ნ., გამრეკელი დ.

ბიოფიზიკის კათედრა, ივ. ჯავახიშვილის სახელობის თბილისის სახელმწიფო
უნივერსიტეტი

(მიღებულია 01. 07. 2005)

რეზიუმე

შესწავლილია მშავა ფოსფატაზას ადსორბციის კინეტიკა და მისი იმობილიზაცია დიატომიტზე. ადსორბცია მიმდინარეობს როგორც ერთსტადიური პროცესი, ფერმენტი სწრაფად ადსორბირდება დიატომიტის ზედაპირზე. თავისუფლად შეაღწევს და განთავსდება მატარებლის ფართო ფორებში. გაზომილია მყარ მატარებლებზე იმობილიზებული მშავა ფოსფატაზას აქტივობა. ნაჩვენებია, რომ დიატომიტზე ადსორბირებული ფერმენტის ხვედრითი აქტივობა 21,4%-ით მეტია, ვიდრე მისი სილიკატულზე იმობილიზაციის შემთხვევაში.

LIST OF REPRESENTATIVES OF GENUS *SERRATULA* L. (COMPOSITAE) OF THE CAUCASUS FLORA

DJUGELI T.*

Institute of Botany of the Georgian Academy of Sciences

(Received June 20, 2005)

Abstract

The species survey of genus *Serratula* L. of the Caucasus is presented. The list is composed by 8 species and 2 subspecies, among them one species - *S caucasica* is endemic for the Caucasus. Taxonomic classification, synonyms, chromosome number, distribution, and ecology of each species are given.

Key words : ecology, chromosome number, endemic, *Serratula*, systematics.

Introduction

The genus of *Serratula* comprises of above 50 species spreading in Europe, Asia and North Africa [Canto, 1984]. According to the literature data for the Caucasus flora there are 9 species of this genus [Borisova, 1963]. We have discovered 8 species and 2 subspecies [Djugeli, 2004]. One species: *S caucasica* is endemic for the Caucasus. While studying the problems of flora, biology and ecology of species, exact identification of taxons plays the decisive role.

Materials and Methods

During the investigation it was taken into consideration the whole morphological characteristics and geographical development. The private collection and also a material of different Botanical organizations (LE, TBI, TGM, TB, EKE, BAK, Grozno, Stavropol and Makhachkala Herbariums) were used. The species were listed according to system of A.G. Borisova [Borisova, 1963]. Species distribution are given as it is stated in the "Flora of Georgia" (1971–2003). Chromosome numbers are given according to "Chromosome number of flowering plants of the flora of USSR" [1990] and "Chromosome number in representatives of some families of vascular plants in the flora of Novosibirsk region.1" [Krasnikov & Lomonosova, 1990].

Results and Discussion

Gen. *Serratula* L.

Sect. 1. **Mastrucium** (Cass.) DC. 1837, 6:667. – *Mastrucium* Cass. 1825, Dict. Sc. Nat., 35 :173; 1826, 41:521.

* Corresponding author: e.mail: 15Djugeli@rambler.ru

1. *S. coronata* L. 1763, Sp.Pl. ed 2, 2:1144. – *S. wolfii Andrae* 1855, Bot. Zeit., 13:321. – *Mastrucium pinnatifidum* Cass. 1825, Dict. Sc. Nat., 35:173.

E c o l o g y : grows in the lower and middle mountain belts at height 1400 – 1800 m a.s.l., on steppe meadows, at edge of forests, in bushes and on water meadows, alkali soiled meadows and sedge marshes. VII – IX. $2n=22, 30$.

C a u c a s u s : Fore-Caucasus: Western, Central, East; Trans-Caucasus : Armenia.

G e n e r a l d i s t r i b u t i o n : Europe, Asia, Far East.

S e c t. 2. Piptochaete Boiss. 1875, Fl. Or., 3:590.

2. *S. erucifolia* (L.) Boriss. 1963, Flora of USSR, 28:270. – *S. xeranthemoides* Bieb. 1808, Fl. Taur.-Cauc., 2:265. – *Xeranthemum erucifolium* L. 1753, Sp. Pl., 2:858.

E c o l o g y : grows from the lower up to upper mountain belts at height 800 - 2100 m a.s.l., on dry stony and clay slopes, in worm woods semi-desert, phrygana and mountain steppes, in ruderal places, on fields.

G e o r g i a : 14 – Gare Kacheti.

C a u c a s u s : Fore-Caucasus: Western, Central, East; Trans-Caucasus: Azerbaijan, Armenia.

G e n e r a l d i s t r i b u t i o n : East Europe, Minor Asia, Western Siberia.

S e c t. 3. Klasea (Cass.) DC. 1837, Prodr., 6:668. – Klasea Cass. 1825, Dict., 35:173; 1826, 41:521.

R a n k 1. Centauroides Boriss.

3. *S. radiata* (Waldst. et Kit.) Bieb. 1819, Fl. Taur.-Cauc., 3:544. – *S. biebersteiniana* (Iljin) Takht. 1945, Takht. and Fed. Fl. Erev., 323. – *Carduus radiatus* Waldst. et Kit. 1802, Descr. et icon pl. rar. Hung., 1:9.

E c o l o g y : grows from the lower up to subalpine belts at height 800 - 2900 m a.s.l., on dry stony or clay slopes, mountains steeps, in sparse grown juniper and oak brushwood, on glades and at edge of forests, in subalpine highbushes. VII – VIII. $2n=60$.

G e o r g i a : 9 – Kartli, 14 – Gare Kacheti, 13 – Kiziki, 17 – Kvemo Kartli, 18 – Javakheti.

C a u c a s u s : Fore-Caucasus: Western, Central, East ; Trans-Caucasus: Azerbaijan, Armenia.

G e n e r a l d i s t r i b u t i o n : Europe, Minor Asia.

R a n k 2. Nitidae Boriss.

4. *S. nudicaulis* (L.) DC. 1806, Lam. et DC., Fl. Fr. ed 3,4:86.

4.1. subsp. **nudicaulis**. – *Centaurea nudicaulis* L. 1759, Syst. Nat. ed 10.2:1232. In Caucasus does not grow.

4.2. subsp. **transcaucasica** (Bornm.) Djugeli 2004, Notulae syst. ac geogr. Inst. Bot. Thbil., Fasc. 44-45:187 – *S. haussknechtii* Boiss. 1875, Fl. Or., 3:589 p.p. – *S. nudicaulis* subsp. *haussknechtii* var. *transcaucasica* Bornm. 1914, Bull. of Tifl. Bot.Gard., 32:2.–*S.transcaucasica* (Bornm.) Sosn. 1934, Grossg. Flora of Cauc., 4:194.

E c o l o g y : grows from the lower up to upper mountain belts at height 600 - 2300 m a.s.l., on dry slopes, in oak, oak – hornbeam forests, in cracks of rocks, at edge of forests, in brushwood, various kinds of bushes. Y – VIII.

G e o r g i a : 9 – Kartli.

C a u c a s u s : Fore-Caucasus: East; Trans-Caucasus: Azerbaijan, Armenia.

G e n e r a l d i s t r i b u t i o n : Minor Asia, Iraq, Iran.

R a n k 3. Coriacea Boriss.

5. *S. coriacea* Fisch. et C.A.Mey. 1837, DC. Prodr., 6:667. – *Centaurea strictissima* Boiss. et Buhse, 1860, Nouv. Mem. Soc. Nat. Mosc., 12:130.

E c o l o g y: grows in the lower, middle and upper mountain belts at height 1100 – 2100 m a.s.l., on the dry stony slopes, on detritus, stony and gypsiferous slopes, gum steppes and phrygana. YII- X. $2n=26$.

C a u c a s u s: Trans-Caucasus: Azerbaijan, Armenia.

G e n e r a l d i s t r i b u t i o n: Minor Asia, Iran.

R a n k 4. *Quinquefolia* Boriss.

6. *S. quinquefolia* Bieb. ex Willd. 1803, Sp. Pl., 3 : 1639. – *S. quinquefolia* M.B. 1808, Fl. Taur.-Cauc., 2:264; 3:544. – *Klasea quinquefolia* (MB.) Cass. 1825, Dict. Sc. Nat., 35:173.

E c o l o g y: grows at the height from 300 up to 2300 m a.s.l., in oak, oak-hornbeam forests, in cracks of rocks, at edge of forests, in bushes. YI-X.

G e o r g i a: 1 – Abkhasia, 2 – Svanetia, 4 – Samegrelo, 5 – Imereti, 7 – Ajaria, 8 – Shiga Kartli, 9 – Kartli, 12 – Kakheti, 14 – Gare Kakheti, 16 – Trialeti.

C a u c a s u s: Fore-Caucasus: Western, Central, East; Trans-Caucasus: Azerbaijan, Armenia.

G e n e r a l d i s t r i b u t i o n: Europe, Minor Asia, Northern Iraq.

S e c t. 5. *Leuzeopsis* Boriss. 1963, Fl. of the USSR, Ad., 28:607.

7. *S. serratuloides* (Fisch. et C.A. Mey.) Takht. 1945, Takht. and Fed., Fl of Erev., 323. – *Leuzea serratuloides* Fisch. et C.A. Mey. 1837, DC. Prodr., 6:666.

E c o l o g y: grows in the lower, middle and upper mountain, subalpine, alpine and subnival belts at height of 600 - 2900 m a.s.l., on dry detritus, stony and gypsiferous slopes, rocks, on marly lime, sandy slots, in sparse grown juniper. YII. $2n=28$.

C a u c a s u s: Trans-Caucasus: Azerbaijan, Armenia.

G e n e r a l d i s t r i b u t i o n: Minor Asia, Iran.

S e c t. 6. *Demetria* Boriss. 1963, Flora of USSR, Ad., 28:607.

8. *S. caucasica* Boiss. 1875, Fl. Or., 6:570 p.p.- *Centaurea cichoraceae* Stev. 1813, Mem. Nat. Mosc., 4:66.

E c o l o g y: grows in the upper, subalpine and alpine belts at height 2700 m a.s.l., on dry slopes. YII– VIII. $2n=30$.

C a u c a s u s: Fore-Caucasus: East; Trans-Caucasus: Azerbaijan.

G e n e r a l d i s t r i b u t i o n: Iran.

References:

- Borisova A.G. *Genus Serratula*. In: Flora of USSR, Acad.Sci.USSR, M.-L., 28, 258-301, 1963.
- Canto P. *Revision del genero Serratula L. (Asteraceae) en la Peninsula Iberica* Lazaroa, 6, 7-80, 1984.
- Chromosome numbers of flowering plants of the flora of USSR* (Guide-Book). Leningrad, 1990. B
- Djugeli T. *Conspectus Genera Serratula L. (Compositae) Florae Caucasus*. Notule systematicae ac geographicae Instituti Botanici Tbilissiensis, 44-45, 187-190, 2004.
- Krasnikov A.A., Lomonosova A.A. *Chromosome numbers in representatives of some families of vascular plants in the flora of the Novosibirsk Region. I*. Bot. Journ., 75, 1990.

**გვარ *Serratula* L. (Compositae) კავკასიის ფლორის
წარმომადგენელთა სია**

ჯუღელი თ.

საქართველოს მეცნიერებათა აკადემიის ნ. კეცხოველის სახ. ბოტანიკის
ინსტიტუტი

(მიღებულია 20.06. 2005)

რეზიუმე

მოცემულია გვარ *Serratula* L.-ს კავკასიაში გავრცელებულ სახეობათა სისტემა: სია მოიცავს 8 სახეობასა და 2 ქვესახეობას, რომლებიც მოთავსებულია 6 სექციაში ა. ბორისოვის (1963) სისტემის მიხედვით, მათგან *Serratula caucasica* Boiss. კავკასიის ენდემია. მოყვანილია სახეობების ნომენკლატურა, გავრცელება, ქრომოსომთა რიცხვი, ეკოლოგია.

THE VEGETATION OF ARGILLACEOUS BADLANDS OF IORI PLATEAU (EAST GEORGIA)

LACHASHVILI N.* , KHACHIDZE M.

N. Ketskoveli Institute of Botany of the Georgian Academy of Sciences

(Received June 10, 2005)

Abstract

Vegetation of argillaceous badland ecosystems of Iori plateau were studied. There are developed mainly phryganoid and foothill desert vegetations. Phryganoid vegetation is represented by phytocenoses of *Reaumuria alternifolia*, *Atriplex spinosa* and *Caragana grandiflora* formations (*Reaumurieta alternifoliae*, *Atriplexieta spinosae* and *Caraganieta grandiflorae*), whereas foothill desert vegetation – by phytocenoses of *Salsola nodulosa*, *Artemisia lerchiana*, *Gamanthus pilosus* and *Atriplex cana* formations (*Salsoletieta nodulosae*, *Artemisieta lerchianae*, *Gamanthetieta pilosae* and *Atriplexetieta canae*). Transitioned phytocenoses are frequent among them. Besides the mentioned phytocenoses those of *Stipa caspia* (*Stipetum caspiae*) are developed there. Their structure is very close to that of the above designated vegetation. 25 phytocenoses are distinguished. Distribution area and short diagnostic description of each phytocenosis are given in this paper.

Key words: argillaceous badlands, ecosystem, arid region, phytocenosis, characteristic species.

Introduction

In the South Caucasus ecosystems of argillaceous badlands mostly occur in arid and semiarid regions. On the one hand they are widespread in the central and eastern parts of the South Caucasus and on the other hand in the southern part of the Caucasus. In the Central and Eastern South Caucasus the above mentioned ecosystems are mostly represented in the southern part of Iori plateau (East Georgia), Sheki plateau (Azerbaijan) and Kabistan foothills (Azerbaijan) [Mikheef, 1929; Grossheim and Sakhokia, 1931; Sakhokia, 1931; Prilipko, 1970, 1980;]. Argillaceous badland ecosystems in these regions are of common origin and they constitute the whole system in the central and eastern parts of South Caucasus.

Ecosystems of argillaceous badlands of the central and eastern part of the South Caucasus have direct contact with ecosystems of arid forests (*Junipereta*, *J. foetidissima*, *J. polycarpus*; *Pistacieta*, *P. mutica*) and steppes (*Bothriochloeta*, *B. ischaemum*; *Stipeta*, *S. lessingiana*, *S. capillata*, *S. pulcherrima*) as well as desert ecosystems of Mtkvari-Arax lowland. They are characterized by, lack of real soil cover and presence of eroded and partitioned steep slopes. Lithologically stratifications of sea clays, claysand and easily crumbling sandstone of Apsheron and Aghchagil age are presented within the mentioned badland ecosystems [Sakhokia, 1931; Tsereteli, 1971]. The substrate is salinized and often contains gypsum. Average annual temperature

* - Corresponding author. E-mail: botanins@gw.acnet.ge

is 14°, annual precipitation is about 250-400 mm, evaporation 1000 mm, moistening coefficient 0,3-0,4 [Atlas of Azerbaijan SSR, 1963; Atlas of Georgian SSR, 1964]. Ecosystems of similar origin and structure in the central part of the South Caucasus also occur in Middle Kartli (environs of Kaspi, East Georgia).

Phytocenological structure of the argillaceous badlands of these regions of central and eastern parts of the South Caucasus was poorly studied till now [Grossheim and Sakhokia, 1931; Sakhokia, 1931; Lachashvili, 2004a, 2004b, 2005; Lachashvili, Khachidze, 2005].

The aim of our research was to determine the typological composition of vegetation of the designated ecosystems in the southern part of the Iori plateau, which is the main distribution range of the argillaceous badland ecosystems in Georgia, to define principal syntaxa and phytocenoses and study their phytocenological structure.

Materials and Methods

Geobotanical data were collected during long period (1984-1991 and 2003-2004) by route methods. Geobotanical descriptions were made on different areas, depending on topographic-edaphic conditions. Separate geobotanical descriptions were summarized in tables, which became a basis for determination of phytocenological structure and definitions of syntaxa and phytocenoses. These definitions are made using traditional geobotanical methods [Shennikov, 1964; Rabotnov 1983; Vasilevich, 1985]. The names of the plants correspond to Czerepanov and Gagnidze [Czerepanov, 1995; Gagnidze, 2005].

Results and discussion

Ecosystems of argillaceous badlands of Iori plateau are widespread in the south-eastern part of the area. They are distributed on Kotsakhura, Chobandaghi, Kaladara, "Patara chrdili", "Didi chrdili", Kumro, DUSDAGHI and other monoclinic low ranges, in Vashlovani depression and on massifs of Mijnskure and Usakhelo-Mta.

As a result of geobotanical investigation of these regions it was determined that in the argillaceous badland ecosystems vegetation coverage is formed fragmentarily. Quite often wide "dead" plots without vegetation are met. Mosaic distribution of the vegetation is caused by the complex topographic-edaphic conditions (partitioned relief, steep and eroded slopes, salinized substrate etc.). Phytocenoses of phryganoid vegetation and foothill desert occur in the main. Phytocenoses of transitioned structure are often met which makes difficult to strictly separate them.

Phryganoid vegetation is represented by phytocenoses of 3 main formations. These are: *Reaumurieta alternifoliae*, *Atraphaxieta spinosae* and *Caraganeta grandiflorae*. Among them phytocenoses of *Reaumurieta alternifolia* formation are the most fragment, whereas phytocenoses of *Atraphaxis spinosa* and *Caragana grandiflora* formations are rare.

Structure of *Reaumurieta alternifoliae* phytocenoses is very xerophyllous. According to their phytocenological structure they are very close to foothill desert vegetation. We refer them to the South Caucasus variant of the Irano-Turanian phryganoid vegetation. On Iori plateau phytocenoses of *Reaumuria alternifolia* are fragmentarily distributed almost on the whole area of argillaceous badland ecosystems. We have picked out some phytocenoses of the formation [Lachashvili, 2004a, 2005]

1. *Reaumuria alternifolia* + *Agropyron pectinatum*. These phytocenoses are distributed in Pantishara gorge. They are developed on South- and South-East-facing badlands. Relief is partitioned. Slope inclination is 35-40°. Slopes are covered with loamy, claysand and sandstone scree. General projective coverage of the phytocenoses is 25-30%. Projective coverage of

Reaumuria alternifolia varies from 7-8% to 15%, but that of *Agropyron pectinatum* is 6-8%. The characteristic species are: *Aellenia glauca*, *Amberboa glauca*, *Torularia eldarica*, *Astrodaucus orientalis*, *Lappula barbata* (frequency of occurrence of each species is 100%), *Artemisia lerchiana*, *Caccinia rauwolfii*, *Stachys fruticulosa* (80-80%). The total number of the recorded species is 25.

2. *Reaumuria alternifolia* + *Artemisia lerchiana* + *Stipa caspia*. These phytocenoses are distributed in Vashlovani depression and in Kaladara gorge. Phytocenoses are developed on argillaceous badlands, though the substrate also contains sandstone. Slope inclination is 40-45°. The slopes are South- and South-West-facing. The slopes surface is cracked. General projective coverage of the phytocenoses is 20-25%. The projective coverage of *Reaumuria alternifolia* is 8-12%, that of *Artemisia lerchiana* – 6-8%. The characteristic species are: *Stipa caspia*, *Amberboa glauca*, *Astrodaucus orientalis* (frequency of occurrence of each species is 100%), *Stachys fruticulosa* (75%). The total number of the recorded species is 22.

3. *Reaumuria alternifolia* + *Aellenia glauca*. These phytocenoses are distributed in Pantishara gorge. They are developed on argillaceous badlands. Rarely substrate contains claysand and sandstone. Slopes inclination is 30-45°. The slopes are South-facing. General projective coverage of the phytocenoses varies from 10-15% to 30%. Projective coverage of *Reaumuria alternifolia* is 10-15%, *Aellenia glauca* – 1-3%. The characteristic species are: *Amberboa glauca*, *Torularia eldarica* (frequency of occurrence of each species is 100%), *Caccinia rauwolfii*, *Capparis herbacea*, *Zygophyllum fabago* (80-80%), *Astrodaucus orientalis*, *Lappula barbata* (60-60%). The total number of the recorded species is 24.

4. *Reaumuria alternifolia* + *Stachys fruticulosa*. These phytocenoses are distributed in Pantishara gorge. They can be found on badlands of different aspect, inclination is 15-30°. They are developed on weathering crust of claysand and sandstone and occur on small plots (5-20 m²). General projective coverage of phytocenoses is 20-30%. Projective coverage of *Reaumuria alternifolia* is 15-20% and that of *Stachys fruticulosa* – 5-10%. Other species are not present (or occur very seldom). There are a lot of *Reaumuria alternifolia* seedlings.

5. *Reaumuria alternifolia* + *Salsola nodulosa*. These phytocenoses are distributed in Mijniskure (river Alazani gorge). They are developed on argillaceous badlands containing gypsum. The slopes are of 30-40° inclination and the aspect is North- and North-West-facing. General projective coverage of the phytocenoses is 15-30%. Only two species make up the phytocenoses: *Reaumuria alternifolia* (projective coverage varies from 10-15% to 20-30%) and *Salsola nodulosa* (from 5-7% to 10-15%).

6. *Reaumuria alternifolia* + *Artemisia lerchiana* + *Agropyron pectinatum* + *Ferula szowitsiana*. These phytocenoses are distributed on anticlinic low ranges, which directly border the Eldari lowland (part of the Mtkvari-Arax lowland). Phytocenoses are developed on North- and North-West-facing slopes. Slope inclination is from 15-20° to 35-40°. The substrate is argillaceous and salinized, contains gypsum. General projective coverage of the phytocenoses varies from 40-45% to 55-65% (the most part of the coverage falls on an ephemeral plant *Trachymia distachya* – from 10% to 25%). Projective coverage of *Reaumuria alternifolia* is 20-35%. The characteristic species are: *Artemisia lerchiana* (projective coverage – 8-10%), *Agropyron pectinatum* (5-8%), *Ferula szowitsiana* (5-8%), also *Salsola nodulosa*, *Centaurea ovina* (frequency of occurrence of each species is 100%), *Eremurus spectabilis* (83%), *Prangos ferulacea*, *Astrodaucus orientalis* (66-66%). The total number of the recorded species is 18.

7. *Reaumuria alternifolia* + *Artemisia fragrans*. These phytocenoses are distributed on anticlinic low ranges, which directly border the Eldari lowland. Phytocenoses are developed on South-facing badlands. The relief is very partitioned and eroded. Slope inclination is 25-45°. Substrate is argillaceous and salinized, contains gypsum. General projective coverage of the phytocenoses is 30-40%. Projective coverage of *Reaumuria alternifolia* is 18-25%, that of

Artemisia lerchiana – 7-12%. The characteristic species are: *Salsola nodulosa*, *Caccinia reuwolfii*, *Centaurea ovina*, *Trachynia distachya* (frequency of occurrence of each species is 100%). The total number of the recorded species is 11.

8. *Reaumuria alternifolia* + *Salsola nodulosa* + *Artemisia lerchiana* + *Agropyron pectinatum*. These phytocenoses are distributed on Mt. Kajiri massif, in the surroundings of Chatma and Chachuna. The phytocenoses are developed on North-facing badlands (Sometimes they occur on hills and hillocks relief). Slope inclination is 30-40°. Substrate is argillaceous and argillo-arenaceous, contains gypsum and sandstone scree. General projective coverage of the phytocenoses is 25-35%. Projective coverage of *Reaumuria alternifolia* is 12-18%. The characteristic species are: *Salsola nodulosa* (projective coverage – 4-5%), *Artemisia lerchiana* (4-5%), *Agropyron pectinatum* (3-4%), also *Stachys fruticulosa*, *Iris iberica*, *Tulipa eichleri* (frequency of occurrence of each species is 80%). The total number of the recorded species is 28.

The phytocenoses of *Atraphaxis spinosa* formation (*Atraphaxieta spinosae*) rarely occur on argillaceous badlands of Iori plateau. They are spread fragmentarily in a form on small plots mainly in Vashlovani depression and Mijniskure. Their phytocenological structure is very xerophilous. We have distinguished 3 phytocenoses.

1. *Atraphaxis spinosa* + *Stipa caspia*. These phytocenoses are distributed in Vaslovani depression. They are developed on argillo-arenaceous and argillaceous badlands. Sometimes the substrate contains conglomerates. Slope inclination is 35-45° and the aspect is South-facing. General projective coverage of phytocenoses varies from 15-20% to 35%. Projective coverage of *Atraphaxis spinosa* is 12-20%, that of *Stipa caspia* – 7-15%. The characteristic species are: *Scutellaria orientalis*, *Silene chlorifolia*, *Teucrium polium*, *Amberboa glauca* (frequency of occurrence of each species is 100%), *Stachys fruticulosa* (80%), *Artemisia lerchiana*, *Agropyron pectinatum*, *Rubia transcaucasica* (60-60%). The total number of the recorded species is 32.

2. *Atraphaxis spinosa* + *Artemisia lerchiana* + *Reaumuria alternifolia* + *Salsola nodulosa*. These phytocenoses are distributed in Mijniskure. They are developed on argillaceous badlands containing gypsum and having different aspects (South-, North-, North-East-facing). Slope inclination is 35-45°. General projective coverage of the phytocenoses is 20-30%. Projective coverage of *Atraphaxis spinosa* is 10-20%. The characteristic species are: *Artemisia lerchiana* (projective coverage 2-5%), *Reaumuria alternifolia* (2-6%), *Salsola nodulosa* (2-5%), also *Stipa caspia* (frequency 70%). The total number of the recorded species is 23.

3. *Atraphaxis spinosa* + *Caragana grandiflora*. These phytocenoses are distributed in Pantishara gorge, Vaslovani depression, Kumro and Mijniskure. Phytocenoses are developed on badlands of different aspects. Slope inclination is 30-55°. Substrate consists of clay, claysand and easily crumbling sandstones. General projective coverage of phytocenoses varies from 20-25% to 30-40%. Projective coverage of *Atraphaxis spinosa* is 10-15%, that of *Caragana grandiflora* – 6-10%. The characteristic species are: *Reaumuria alternifolia* (projective coverage – 2-6%), *Stachys fruticulosa*, *Artemisia lerchiana*, *Stipa caspia*. The total number of the recorded species is 35.

Phytocenoses of *Caragana grandiflora* formation (*Caraganeta grandiflorae*) are fragmentarily widespread on argillaceous and loamy soils of hill and hillock relief. However they are rarely met on argillaceous and argillo-arenaceous badlands. Phytocenological structure of *Caragana grandiflora* phytocenoses is less xerophilous than that of phytocenoses of *Reaumuria alternifolia* and *Atraphaxis spinosa*. In such an ecosystem we have distinguished only phytocenosis.

1. *Caragana grandiflora* + *Atraphaxis spinosa* + *Salsola nodulosa* + *Artemisia lerchiana*. These phytocenoses are distributed in Mijniskure. They are developed on South-, North- and North-East-facing badlands. The relief is very partitioned. Slope inclination is 25-50°. Substrate is argillaceous, contains scree claysand and sandstone. General projective coverage of phytocenoses is 30-40%. Projective coverage of *Caragana grandiflora* is 20-25%. The

საქართველოს
საბუნებისმეტყველო
მეცნიერებათა
აкадеმიის
ბიბლიოთეკა

characteristic species are: *Atraphaxis spinosa*, *Salsola nodulosa*, *Artemisia lerchiana*, also *Stipa caspia* and *Agropyron pectinatum*. The total number of the recorded species is 32. The above mentioned phytocenosis is one of the most xerophilous in the *Caragana grandiflora* formation.

Foothill desert vegetation of Iori plateau argillaceous badlands is mainly represented by phytocenoses of *Salsola nodulosa* and *Artemisia lerchiana* formations (*Salsoleta nodulosae* and *Artemisieta lerchianae*). Besides, phytocenoses of *Atriplex cana* (*Atriplexetum canae*) and *Gamanthus pilosus* (*Gamanthetum pilosae*) are rarely found. Structure of *Artemisia lerchiana* and *Salsola nodulosa* phytocenoses formed on badlands is rather different from that of the phytocenoses that occurring on plain relief (e.g. Eldari lowland, Lekistskali, Bughamoedani and Kumro plain places). First of all, the difference is in the development of ephemeral-ephemeroïd synusia – on badlands, in contrast to plain relief, the ephemeral synusia is absent, or is weakly formed and comprises 1 or 2 species (*Trachynia distacya*, *Eremopyron orientale*, *E. distans* and e.c.). In parallel the number of semi-shrubs, dwarf semi-shrubs and perennial herbs as well as their cenotic importance is increased (including those of characteristic species of the phryganoid vegetation).

In the area of the argillaceous badland ecosystems of Iori plateau **phytocenoses of *Salsola nodulosa*** are mainly distributed on low ranges, which directly border the Mtkvari-Arax lowland, on massifs of Mijnskure and in Kumroschevi. We have distinguished the following phytocenoses:

1. ***Salsola nodulosa* + *Reaumuria alternifolia***. These phytocenoses are distributed in Mijnskure. They are developed on North-, North-East-facing badlands. Slope inclination is 30-40°. Substrate is argillaceous, very salinized and contains a large amount of gypsum. General projective coverage of phytocenoses is 15-30%. Only two species make up the phytocenoses: *Salsola nodulosa* (projective coverage 10-20%) and *Reaumuria alternifolia* (5-10%).

2. ***Salsola nodulosa* + *Artemisia lerchiana* + *Reaumuria alternifolia* + *Ferula szowitsiana***. These phytocenoses are distributed in Kumro gorge and on low ranges adjacent to the Eldari lowland. Phytocenoses are developed on North-, North-East-facing badlands. Slope inclination varies from 10-15° to 30-40°. Substrate is argillaceous, very salinized and contains a large amount of gypsum. General projective coverage of the phytocenoses is 35-50%. Projective coverage of *Salsola nodulosa* is 12-15%. The characteristic species are: *Artemisia lerchiana* (projective coverage 6-8%), *Reaumuria alternifolia* (4-6%), *Ferula szowitsiana* (3-5%), also *Suaeda dendroides* (1-2%), *Eremopyron orientale*, *E. distans* (coverage of each species 5%), *Scorzonera cana*, *Tulipa eichleri*, *Prangos ferulacea*, *Bupleurum wittmannii*. The total number of the recorded species is 25.

3. ***Salsola nodulosa* + *Artemisia lerchiana***. These phytocenoses are distributed in Kumro. They are developed on the North and North-East-facing badlands. Slope inclination is 20-40°. Substrate is argillaceous, salinized and contains a large amount of gypsum. General projective coverage of phytocenoses is 30-40%. Projective coverage of *Salsola nodulosa* is 20-25%, that of *Artemisia fragrans* – 8-10%. The characteristic species are: *Reaumuria alternifolia*, *Suaeda dendroides*, *S. microphylla*, *Prangos ferulacea*, *Scorzonera cana*, *Eremopyron orientale*, *Gamanthus pilosus*. The total number of the recorded species is 29.

4. ***Salsola nodulosa* + *Atriplex cana* + *Artemisia lerchiana***. These phytocenoses are very rare. They are distributed in Vashlovani depression. The phytocenoses are developed on South and South-West-facing argillaceous badlands. The relief is very partitioned and eroded. Slope inclination is 35-45°. Substrate is very salinized and contains a large amount of gypsum. General projective coverage of phytocenoses is 20-25%. Projective coverage of *Salsola nodulosa* is 10-15%, *Atriplex cana* – 5%, *Artemisia lerchiana* – 4-5%. The characteristic species are: *Reaumuria alternifolia*, *Amberboa glauca*, *Agropyron pectinatum*. The total number of the recorded species is 23.



Phytocenoses of *Artemisia lerchiana* formation (*Artemisieta lerchiana*) are fragmentarily distributed almost on the whole area argillaceous badland ecosystems of Iori plateau. Its phytocenological structure is often close to that of the phryganoid vegetation. The following phytocenoses of the ecosystem are presented:

1. *Artemisia lerchiana* + *Reaumuria alternifolia* + *Atraphaxis spinosa*. These phytocenoses are distributed in Vashlovani depression on argillaceous badlands. Slope inclination is 35-45°. Slopes are South-facing. Substrate is salinized. General projective coverage of phytocenoses is 25-35%. Projective coverage of *Artemisia lerchiana* is 15-20%, that *Reaumuria alternifolia* and *Atraphaxis spinosa* – 4-5% (each). The characteristic species are: *Astrodaucus orientalis*, *Bupleurum wittmannii* (Projective coverage of each – 1-3%), *Zygophyllum falago*, *Suaeda dendroides*, *Stachys fruticulosa*. The total number of the recorded species is 22.

2. *Artemisia lerchiana* + *Salsola nodulosa* + *Reaumuria alternifolia*. These phytocenoses are distributed in Mijnickure. They are developed on North-facing badlands. Slope inclination is 20-35°. Substrate is argillaceous, salinized and contains gypsum. General projective coverage of phytocenoses is 20-25%. Projective coverage of *Artemisia lerchiana* is 18-22%. The characteristic species are: *Salsola nodulosa* and *Reaumuria alternifolia*, also *Petrosimonia brachiata*. The total number of the recorded species is 4.

3. *Artemisia lerchiana* + *Stipa caspia*. These phytocenoses are distributed in Pantishara gorge and Vashlovani depression. They are rare. The phytocenoses are developed on argillaceous and argillaceous badlands. Slope inclination is 25-30°. The slopes are South-facing. General projective coverage of phytocenoses is 30%. Projective coverage of *Artemisia lerchiana* is 15-20%, that of *Stipa caspia* – 5-8%. The characteristic species are: *Salsola nodulosa*, *Reaumuria alternifolia*, *Capparis herbacea*, *Astragalus stevenianus*, *Euphorbia seguierana*, *Stachys fruticulosa*. The total number of the recorded species is 33.

4. *Artemisia lerchiana* + *Agropyron pectinatum*. These phytocenoses are rare. They are distributed in Vashlovani depression. The phytocenoses are developed on argillaceous badlands. Slope inclination is 35-45°. The slopes are South-facing. Substrate is salinized and contains gypsum. General projective coverage of phytocenoses is 30-35%. Projective coverage of *Artemisia lerchiana* is 15-20%, that of *Agropyron pectinatum* – 15%. The characteristic species is *Salsola nodulosa*. The total number of the recorded species is 15.

Atriplex cana has limited distribution on the Iori plateau. It occurs only in Vashlovani depression. Accordingly its phytocenoses (*Atriplexetum canae*) are rarely found only in this locality in a form of small plots. The phytocenoses are formed on argillaceous badlands. Slope inclination is 35-45°. The slopes are South- and South-West-facing. Substrate is very salinized and contains gypsum. General projective coverage of phytocenoses is 15-20%. Projective coverage of *Atriplex cana* is around 15%. The characteristic species are: *Salsola nodulosa*, *Artemisia lerchiana*, *Stachys fruticulosa*, *Reaumuria alternifolia*, *Agropyron pectinatum*, *Amberboa glauca*. The total number of the recorded species is 27.

Gamanthus pilosus phytocenoses (*Gamanthetum pilosae*) are distributed almost on the whole area of the argillaceous badland ecosystems of Iori plateau. They are developed on argillaceous badlands of different aspect. Slope inclination is 20-45°. Substrate is very salinized. General projective coverage of phytocenoses is around 20-50%. In spring *Eremopyron orientale* and *E. distans* constitute the phytocenoses along with seedlings of *Gamanthus pilosus*. But in autumn only *Gamanthus pilosus* is present.

Besides the above mentioned phytocenoses of phryganoid and foothill desert vegetation, phytocenoses of *Stipa caspia* (*Stipetum caspiae*) are also found. They are fragmentarily distributed in a form of small plots almost on the whole area of the argillaceous badland ecosystems of Iori plateau. These phytocenoses are very close to foothill desert and phryganoid vegetation by their

phytocenological structure. However, their floristic composition is less xerophilous. In addition they are completely different from steppe vegetation. We have distinguished 3 phytocenoses:

1. *Stipa caspia* + *Reaumuria alternifolia* + *Salsola nodulosa*. These phytocenoses are distributed in Mijnskure. They are rare. The phytocenoses are developed on South-facing badlands. Slope inclination is 25-30°. Substrate is argillo-arenaceous and argillaceous, it contains sandstone scree and gypsum. General projective coverage of phytocenoses is 20-30%. Projective coverage of *Stipa caspia* is 15-20%, that of *Reaumuria alternifolia* and *Salsola nodulosa* – 5-7% (of each). The characteristic species are: *Artemisia lerchiana*, *Stachys fruticulosa*, *Astragalus stevenianus*, *Zygophyllum fabago*. The total number of the recorded species is 15.

2. *Stipa caspia* + *Reaumuria alternifolia* + *Artemisia lerchiana* + *Agropyron pectinatum*. These phytocenoses are distributed on an anticlinic low ranges, which directly border the Eldary lowland. They are rare. The phytocenoses are developed on North- and North-West-facing badlands. Slope inclination is 20-27°. Substrate is argillaceous and argillo-arenaceous. General projective coverage is 30-35%. Projective coverage of *Stipa caspia* is around 15%. Projective coverage of *Reaumuria alternifolia* is 6-8%, that of *Artemisia lerchiana* and *Agropyron pectinatum* – 4-6% (of each). The characteristic species are: *Gallium verum*, *Ferula szowitsiana*, *Eremurus spectabilis*, *Scutellaria orientalis*, *Crinitaria villosa*, *Trachyana distachya*. The total number of the recorded species is 15.

3. *Stipa caspia* + *Artemisia lerchiana*. These phytocenoses are distributed in Vashlovani depression, Kumro and Mijnskure. The phytocenoses are developed on argillo-arenaceous and argillaceous badlands. Substrate contains sandstone scree. The relief is partitioned. Slope inclination is 30-35°. The slopes are South- and South-West-facing. General projective coverage of phytocenoses is around 30%. Projective coverage of *Stipa caspia* is 18-20%, that of *Artemisia lerchiana* – 5-10%. The characteristic species are: *Astragalus stevenianus*, *Euphorbia seguierana*, *Galium verum*, *Amberboa glauca*. The total number of the recorded species is – 31.

References:

- Atlas of Azerbaijan SSR*. Baku-Moscow. p. 49, 1963 (in Russian).
Atlas of Georgian SSR. Tbilisi-Moscow. p. 269, 1964 (in Russian).
 Czerepanov S. K. *Vascular Plants of Russia and adjacent states (former USSR)*. Cambridge, p. 516, 1995.
 Gagnidze R. *Vascular plants of Georgia. A nomenclatural checklist*. Tbilisi, p. 247, 2005.
 Grossheim A. A. *Vegetation cover of the Caucasus*. Moscow, p. 265, 1946 (in Russian).
 Grossheim A. A. and Sakhokia M.F. *Description of the vegetation of Kabristan. Work on geobotanical investigation of winter pastures of Azerbaijan SSR*. Ser. A. Winter pastures. Fascicle 7. Baku, p.102, 1931 (in Russian).
 Lachashvili N. *Phytocenoses of Reaumuria alternifolia in the Vashlovani State Reserve (East Georgia)*. Bull. Georg. Acad. Sci., **170**, 1, 122-124, 2004a.
 Lachashvili N. *Rare phytocenoses of Atraphaxis spinosa in the Vashlovani State Reserve (East Georgia)*. Bull. Georg. Acad. Sci., **170**, 2, 330-333, 2004b.
 Lachashvili N. *New data on Reaumuria alternifolia formation (Reaumurieta alternifoliae)*. Bull. Georg. Acad. Sci., **171**, 3, 503-505, 2005.
 Lachashvili N., Khachidze M. *Typology of the vegetation of Vashlovani State Reserve (East Georgia)*. Science and Technologies, **1-3**, 143-146, 2005 (in Georgian).
 Mikheeff A.A. *The vegetable corporation of Kabristan and the Bogaz plains, and the meliorative significance of this region*. Proc. of the society of survey and investigation of Azerbaijan. 7. Fascicle 11, Baku, p.103-150. 1929 (in Russian).

- Prilipko L.I. *Vegetation cover of Azerbaijan*. Baku, p. 169, 1970 (in Russian).
- Prilipko L.I. *Mountain xerophyte vegetation*. In: *Vegetation of the European part of USSR*. Leningrad, p. 277-280, 1980 (in Russian).
- Rabotnov T.A. *Phytocenology*. Moscow, p. 3-278, 1983, (in Russian).
- Sakhokia M.F. *Description of the vegetation of winter pastures of Sheki plateau*. Works on geobotanical investigation of winter pastures of Azerbaijan SSR. Series A. Winter pastures. Fascicle 9. Baku, p. 30, 1931 (in Russian).
- Sakhokia M.F. *Botanical description of the Tbilisi surroundings and along the itinerary: Tbilisi - Shiraki plateau*. In: *Botanical excursions through Georgia*. Tbilisi, p.7-30, 1958 (in Russian).
- Shennikov A.P. *Introduction to geobotany*. Leningrad, p. 9-412, 1954 (in Russian).
- Tsereteli D.V. *Iverian part of intermountain*. In: *Geomorphology of Georgia*. Tbilisi, p. 282-309, 1971 (in Russian).
- Vasilevich V. Ch. *Concerning methods of vegetation classification*. Bot. Journal, 70, 12, 1596-1604, 1985.

ივრის ზეგნის (აღმოსავლეთ საქართველო) თიხიანი ბედლენდების მცენარეულობა

ლანა შვილი ნ., ხაჩიძე მ.

საქართველოს მეცნიერებათა აკადემიის ნ. კეცხოველის სახელობის ბოტანიკის
ინსტიტუტი

(მიღებულია 10.06.2005)

რეზიუმე

შესწავლილია ივრის ზეგნის (აღმოსავლეთ საქართველო) თიხიანი ბედლენდების ეკოსისტემების მცენარეულობა. ძირითადად განვითარებულია ფრიგანისებრი და მთასწინების უდაბნოების მცენარეულობა. ფრიგანისებრი მცენარეულობა წარმოდგენილია რეუმურიანი (*Reumurieta alternifoliae*), ხორციფერიანი (*Atraphaxieta spinosae*) და უძრახელიანი (*Caraganeta grandiflorae*) ფორმაციების ფიტოცენოზებით, ხოლო მთისწინების უდაბნოს მცენარეულობა – ხურხუმოიანი (*Salsola nodulosae*), ავშნიანი (*Artemisieta lerchianae*), გამანთუსიანი (*Gamantheta pilosae*) და ატრიპლექსიანი (*Atriplexeta canae*) ფორმაციების ფიტოცენოზებით. ხშირია მათ შორის გარდამავალი ფიტოცენოზები. აღნიშნულის გარდა განვითარებულია *Stipa caspia*-ს ფიტოცენოზებიც (*Stipetum caspiae*), რომლებიც თავიანთი სტრუქტურით ზემოაღნიშნულ მცენარეულობას უახლოვდებიან. გამოყოფილია 25 ფიტოცენოზი. მოცემულია თითოეულის გავრცელების არეალი და მოკლე დიაგნოსტიკური დახასიათება.

STABILITY OF $11^{S(1)}$ *NOCARDIOPHISIS DESSONVILLEI* CELLS UNDER THE INFLUENCE OF DIFFERENT AGENTS

ATANELISHVILI I., JIMSHITASHVILI N., RATIANI K., TABATADZE M.,
GORDEZIANI M.

Department of Cellular and Molecular Biology, Iv. Javakhishvili Tbilisi state University.

(Received June 10, 2005)

Abstract

Effect of ionic and non-ionic detergents' various concentrations, increased doses of UV-radiation on quantitative changes of $11^{S(1)}$ *Nocardiohphis dessonvillei* cells has been investigated. Different nature between the influence of ionic and non-ionic detergents on cell viability have been demonstrated. $11^{S(1)}$ *Nocardiohphis dessonvillei* cells stimulation and/or inhibition seems to be the result of characteristic membrane structural changes caused by appropriated agents.

Keywords: Tween- θ , Triton-x100, SDS, $11^{S(1)}$ *Nocardiohphis dessonvillei*

Introduction

In nature and under standard conditions microorganisms can degrade various materials. To improve or turn down these processes new approaches have to be applied. Surfactants as well as UV-radiation contribute significantly to the biodegradation profile of various materials. Several studies conducted in various laboratories have revealed harmful effects of UV-radiation on growth, survival, development, nutrient uptake and various metabolic processes of microorganisms [Kumar et.al., 2004]. Besides, high concentrations of detergents significantly inhibit microbial growth. In addition, core role of biological membranes also have been emphasized [Dhouib et.al., 2003].

The goal of this study was investigation of effect of detergents' (ionic and non-ionic) as well as UV-radiation on the stability of $11^{S(1)}$ *Nocardiohphis dessonvillei* cells and also select detergents' concentration and UV-radiation doze, leading to considerable decrease of viable cells amount.

Materials and Methods

Microorganisms were isolated from soil of the river Aragvi gorge by the scientists of the N. Ketskhoveli Institute of Botany, Georgian Academy of Sciences and preserved in the Microbiological Departments collection. Experiments were carried out on $11^{S(1)}$ *Nocardiohphis dessonvillei* cells in stationary phase. Culture was grown on the medium described by Krassilnikov [Krassilnikov, 1950]. Ionic-Sodium Dodecylsulphate (SDS) and non-ionic detergents (Tween 80 and Triton X-100) were used in concentrations 10^{-7} M, 10^{-5} M, 10^{-2} M. Radiation source was BUV-ZOP lamp, dozes 200, 400, 600, 800, 1000 J/m² respectively. The quantity of viable cells was calculated according to number of colonies grown on solid medium. Quantity of viable cells is

given in percentage with respect to control (100%). Statistically refined experimental data are given in figures.

Results and discussion

Quantitative changes of *11^{S(I)}* y *Nocardiophasis dessonvillei* viable cells under the influence of various concentrations of detergents are illustrated in Fig. 1. The same characteristics in case of various dozes of UV-radiation are given in Fig. 2. Quantity of cells in control is regarded as 100%. Data represented in figures reveal concentration as well as radiation doze dependent changes of viable cell amount.

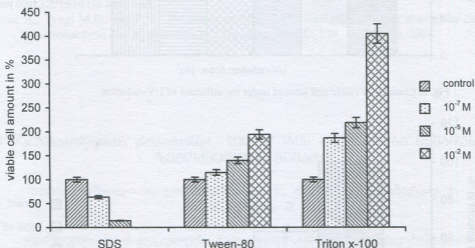


Fig.1. Changes of viable cell amount under the influence detergents' different concentrations

Hence, information available from Fig.1 shows different character of cell quantitative changes i.e. all selected concentrations of non-ionic detergents (Tween-8, Triton x-100) exhibit stimulatory effect upon viable cell amount. While the least concentration (10⁻⁷ M) of SDS resulted in considerable decrease of *11^{S(I)}* y *Nocardiophasis dessonvillei* cell amount (40%). It seems apparent that such discrepancy in the influence of ionic and non-ionic detergents is due to detergents differing nature and characteristic interactions with membrane lipid-protein complexes as well.

UV-radiation doze dependent changes of *11^{S(I)}* y *Nocardiophasis dessonvillei* viable cell amount is demonstrated in Fig.2, demonstrating decrease of viable cell amount in case of all selected dozes. Maximum of applied doze of UV-radiation resulted in sharp decrease of viable cells - 64%. Limited doze, LD₅₀ - 600-800j/m² has been established.

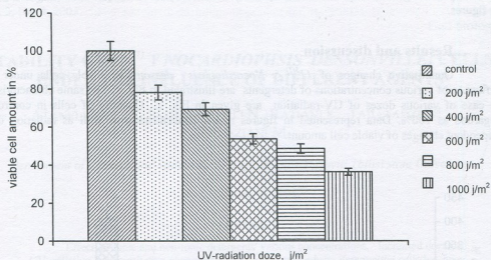


Fig. 2. Changes of viable cell amount under the influence of UV-radiation

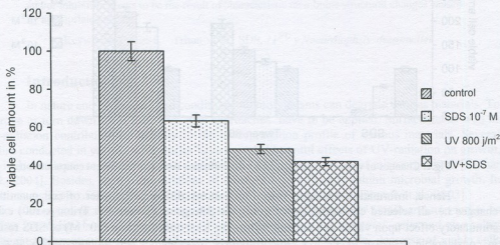


Fig. 3 Changes of viable cell amount under the influence of joint action of UV-radiation and SDS

On the basis of obtained experimental observations and existing literature data it has been proposed that microorganisms destructive activity can be regulated by quantitative changes of viable cells under the pretreatment of various factors. Considered experimental data have revealed decrease of *11^{5(1)}* y *Nocardiothis dessonvillei* cell amount in case of SDS and UV-radiation. According to previous experimental data [Gordeziani, 2004] UV-radiation increases membrane permeability not enhancing aggressive metabolites content in culture medium. Consequently, joint effect of SDS and UV-radiation for regulation of microorganisms' destructive activity seems far more beneficial. To substantiate this assumption series of experiments were carried out. *11^{5(1)}* y

Nocardiophasis dessonvillei cells were influenced by combined action of UV-radiation dose-800j/m² and SDS 10⁻⁷M. Data confirming our assumption are given in Fig. 3.

References:

- Dhouib A., Hamad N., Hassairi I., Sayadi S. *Degradation of anionic surfactants by Cyrtobacter braakii*. Process Biochemistry, **38**, 1245-1250, 2003.
- Gozeziani M., Tabatadze M., Atanelishvili I. and Bochorishvili N. *Effect of UV irradiation on several functional characteristics of 11⁵⁽¹⁾ y Nocardiophasis dessonvillei*. Journal of Biological Physics and Chemistry (JBPC), **4**, 4, 12/2004.
- Krassilnikov N.A. *Actinomycetes Antagonism and Antibiotic Compounds*. Moscow and Leningrad. 32, 1950 (in Russian).
- Kumar A., Tyagi M.B., Jha P.N. *Evidences showing UV-B-radiation-induced damage of DNA in cyanobacteria and its detection by PCR assay*. BBRC, **318**, 1025-1030, 2004.

11⁵⁽¹⁾ y *Nocardiophasis dessonvillei* უჯრედების მდგრადობა სხვადასხვა ზემოქმედების მიმართ

ათანელიშვილი ი., ჯიშშიტაშვილი ნ., რატიანი კ., ტაბატაძე მ.,
გორდეზიანი მ.

უჯრედული და მოლეკულური ბიოლოგიის კათედრა, ივ. ჯავახიშვილის
სახელობის თბილისის სახელმწიფო უნივერსიტეტი

(მიღებულია 10.06.2005)

რეზიუმე

შესწავლილია ულტრაიისფერი სხივების განსხვავებული დოზების, სხვადასხვა კონცენტრაციის იონური და არაიონური დეტერგენტების გავლენით 11⁵⁽¹⁾ y *Nocardiophasis dessonvillei*-ს ცოცხალი უჯრედების რაოდენობრივი ცვლილებები. ნაჩვენებია 11⁵⁽¹⁾ y *Nocardiophasis dessonvillei*-ს ცოცხალი უჯრედების რაოდენობრივი ცვლილებების განსხვავებული ხასიათი. უჯრედების ზრდის დათრგუნვა ან სტიმულაცია განპირობებული უნდა იყოს გამოყენებული აგენტების მიერ მემბრანის დამახასიათებელი სტრუქტურული ცვლილებებით.

THE MORPHOLOGICAL STUDY OF THE ACIDOPHILIC CELLS WITHIN THE DIGESTIVE GLAND TISSUE OF *HELIX LUCORUM*

CHILADZE N^{*}., MCHEDLIDZE T., MODEBADZE I., TUMANISHVILI T., DZIDZIGURI D.

Department of Cytology, Histology and Developmental Biology, The Laboratory of Developmental Biology, Iv. Javakishvili Tbilisi State University

(Received 23 June, 2005)

Abstract

It has been shown that the acidophilic cells in the digestive gland of *Helix lucorum* are mainly found within the central part of the calcium cells. Some dependence between the number of the acidophilic and calcium cells was revealed. The frequency of encounter of the acidophilic cells in the parenchyma of the hepatopancreas of *Helix lucorum* is varied with animal age.

Key words: gastropod snail, hepatopancreas, calcium cell, acidophilic cell.

Introduction

The multiple functions (production of digestive enzymes, absorption of nutrients, endocytosis of food substances, food storage and excretion) of the digestive gland of the gastropod snails are carried out by the following types of cells: digestive, calcium and excretory. Undifferentiated cells which give rise to the other cell types are also presented in the liver epithelium [Dimitriadis, 2001]. Moreover, the acidophilic cells, conditionally called "pink cells" were identified by us within the digestive gland tissue three years ago [Modebadze et al., 2002]. The goal of our work was the morphological study of the acidophilic cells in the digestive gland of the snail *Helix Lucorum*.

Materials and methods

The experiments were held on the gastropods – *Helix Lucorum* of different age (adults with shell diameter 35-40 mm and adolescents - with shell diameter 10-15 mm). The paraffin sections of about 5-6 μ m were stained by hematoxylin-eosin.

Results and discussion

It has been shown that the acidophilic cells within the digestive gland tissue of the snail *Helix lucorum* represent the cell population characteristic for this species in general. These cells have asymmetrically disposed nuclei and their cytoplasm contains some polysaccharides [Chiladze

* Corresponding Author: E-mail: n_chiladze@yahoo.com

et al., 2003]. The question consists in localization of the mentioned cells within the liver lobes. It turned out that they are always located inside the calcium cells. The idea about phagocytosis was suggested. Hence we decided to determine the localization area of the acidophilic cells within the calcium cell. The paraffin sections of the digestive gland tissue were investigated in the snails of different age for this purpose. It was established that the acidophilic cells in the snails of both ages are located mainly in the central part of the calcium cell (Table 1., Fig.1.), nevertheless sometimes they are observed within the other parts of the calcium cells, viz. basally and apically.

Table 1. The localization of the acidophilic cells within the digestive gland tissue

The snails	The localization of the acidophilic cells within the calcium cells		
	basal	central	apical
Adult	25±7,4%	60±5,8%	15±6,3%
Adolescent	22±10%	52±3,6%	26±4,8%

Table 2. The allocation of the cell types within the digestive gland lobes in the adult snails

Months	The cell types		
	Digestive	Calcium	Excretory
April	74±3%	14,6±3.8%	11.4±1.7%
June	85±4%	5.4±1.9%	9.6±1.8%

Table 3. The allocation of the cell types within the digestive gland lobes in the adolescent snails

Months	The cell types		
	Digestive	Calcium	Excretory
April	74±3%	14±1.1%	12±1.02%
September	73±3.3%	12±0.5%	15±0.9%

The phagocytosis in the digestive gland epithelium is not mentioned in the literature [Dimitriadis, 2001. The localization of three types described by us does not point to the phagocytosis. However the unambiguous negligence of phagocytosis requires further investigations.

It has been shown, that the number of acidophilic cells within the liver tissue decreases gradually with aging [Chiladze et al., 2003]. Furthermore, we established in these studies that the frequency of appearance of the acidophilic cells in the digestive gland is different in adult and adolescent individuals i.e. depends on the age of an animal. The acidophilic cells in adult individuals appear only in the certain time of the year. The first acidophilic cells appear in January, besides the number of these cells reaches their maximum in April, when the animal becomes functionally active (period after the hibernation). They nearly do not appear within the liver tissue during the rest of time of the year (Fig.2.).

The presence of the acidophilic cells within the digestive gland tissue in adolescent snails seems to carry the different character: these cells appear in every time of year, although their maximal number is reached in April similar to adults (Fig.3.).

The obtained results make us suggest that the higher level of appearance of the acidophilic cells in the adolescent snails is stipulated by the high metabolic activity of the digestive gland.

The nonstandard allocation of the acidophilic cells results in the great number of questions. Particularly, the additional observations were required to study the changes in the quantitative ratio of the main types of the digestive gland cells in the presence or absence of the acidophilic cells. The investigations on the liver tissue sections under the light microscope has shown that the number of calcium cells decreases three times in June, when there are no acidophilic cells in the adult animal liver tissue (Table 2.).

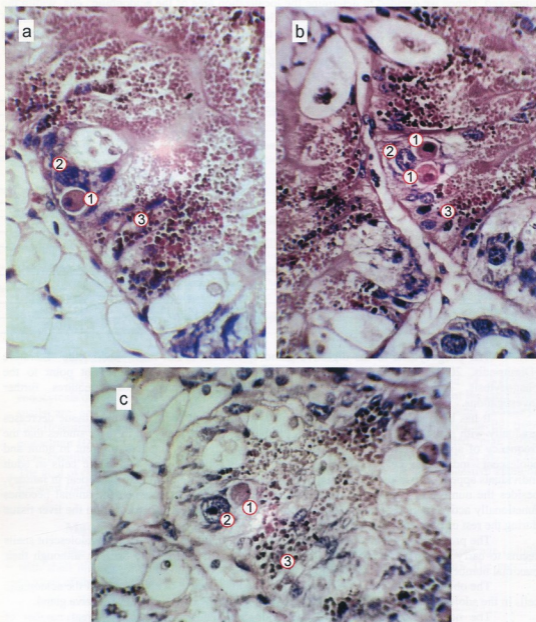


Fig. 1. The localization of the acidophilic cells within the digestive gland tissue; a – basal, b – central, c – apical; 1 – the acidophilic cell, 2 – the calcium cell nucleus, 3 – the digestive cell; *stained with Hematoxylin-Eosin (10X40).*

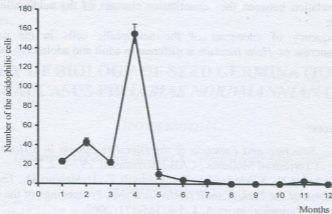


Fig. 2. The allocation of the acidophilic cells within the adult snail digestive gland tissue during the year.

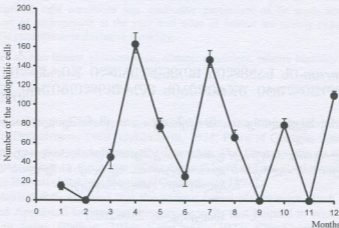


Fig. 3. The allocation of the acidophilic cells within the adolescent snail digestive gland tissue during the year.

The different picture is observed within the liver tissue of the adolescent snails: The number of calcium cells reduces lightly in September during the absence of the acidophilic cells (Table 3). This may be explained by the appearance of acidophilic cells within the digestive gland tissue of the adolescent snail again in October (Fig.3.), whereas they disappear completely in adults after June (Fig.2.) and the number of calcium cells reduces respectively.

On the basis of our investigations it may be concluded :

1. The acidophilic cells are located within the multifunctional calcium cell, basically in the central part;

2. The correlation between the quantitative changes of the acidophilic and calcium was revealed;
3. The frequency of encounter of the acidophilic cells in the parenchyma of the hepatopancreas of *Helix lucorum* is different in adult and adolescent individuals.

References:

- Dimitriadis V. K., *Structure and Function of the Digestive System in Stylommatophora*. In: The Biology of Terrestrial Molluscs., CAB-international, 237-252, 2001.
- Modebadze I., Chiladze N., Kakhidze I., Tumanishvili T., Dzidziguri D., *The study of the liver regeneration of the Turkish snail (Helix lucorum)*. Proceedings of the Georgian Academy of Sciences, Biological series A, 1, 5-6, 565-571, 2002.
- Chiladze N., Modebadze I., Tumanishvili T., Kvintradze T., Dzidziguri D., *The Peculiarities of the Digestive Gland Histoarchitectonics in the Snail Helix lucorum*. Proceedings of the Georgian Academy of Sciences, Biological Series B, 1, 1-2, 49-51. 2003.

Helix lucorum-ის საჭმლის მომნელებელი ჯირკვლის ძროვილში აციდოფილური უჯრედების მორფოლოგიური შესწავლა

ჭილაძე ნ., მჭედლიძე თ., მოდებაძე ი., თუმანიშვილი თ., ძიძიგური დ.

ციტოლოგიის, ჰისტოლოგიისა და განვითარების ბიოლოგიის კათედრა,
განვითარების ბიოლოგიის ლაბორატორია, ივ. ჯავახიშვილის სახ. თბილისის
სახელმწიფო უნივერსიტეტი

(მიღებულია 23.06.2005)

რეზიუმე

შესწავლილია მუცელფეხიანი მოლუსკის *Helix lucorum*-ის საჭმლის მომნელებელი ჯირკვლის ძროვილში აციდოფილური უჯრედების ("ვარდისფერი უჯრედების") ლოკალიზაცია და განაწილება. დადგინდა მათი ლოკალიზაცია მრავალფუნქციური კალციუმის უჯრედის შიგნით. ზრდასრულ ინდივიდებში გამოვლინდა კორელაცია აციდოფილური და კალციუმის უჯრედების რაოდენობრივ ცვლილებებს შორის; ნაჩვენებია იქნა, რომ წელიწადის სხვადასხვა დროს *Helix lucorum*-ის შეპატოპანკრეასის პარენქიმაში აციდოფილური უჯრედების შეხვედრის სიხშირე განსხვავებულია ზრდასრულ და მზარდ ინდივიდებში.

FOR STUDY OF BIOLOGY OF SEED GERMINATION OF THE CAUCASUS FIR (*ABIAS NORDMANNIANA*)

DZOTSENIDZE N.

Department of Botany, Kutaisi A. Tsereteli State University

(Received July 11, 2005)

Abstract

Peculiarities of seed germination and qualitative parameters of the fir seeds at the very east edge of the Caucasus fir distribution area was studied. Received data were compared with the same parameters of fir optimal climatic zone. At different light conditions characteristics of growth and development of fir seedling were studied. With complex morphological features the vital cenotypes of fir undergrowth were revealed. In spite of optimal light conditions low qualitative parameters of fir seeds and hindered development of undergrowth at the very east edge of habitat are mainly caused by low atmospheric precipitates and relative humidity.

Key words: habitat, phitocenotype, climatic conditions, relative humidity.

Introduction

According to literature data the east edge of the Caucasus fir is the gorges of the rivers Big and Small Liakhvi [Dolukhanov, 1989; Gulisashvili, 1974; Flora of Georgia, 2001]. Easter, at the mouth of the river Qsani this species gradually disappear. The Caucasus fir on the North edge of the Small Caucasus reaches to the gorge of the river Algeti and its single specimen have been described to village Bevreti (Mtskheta Region) [Gulisashvili, 1974].

Palaeobotanical data revealed that the ancient burial places were much more eastern, in Shiraqi steppes and Apsheroni tertiary sedimentary deposits that pointed to wider habitat of fir in historical past than today [Palibin, 1936; Kolakovski, 1973; Chochieva, 1975; Shatilova & Ramishvili, 1990]. Damp climatic conditions are optimal for fir residence in Talishi and Outer Kakheti, but this species does not occur there. The reason is that after glacial age damp climatic period was again changed by xerothermal climate and fir disappeared in this places. In some regions of East Caucasus (Inner Kakheti, Talishi) it was formed again damp climate, but fir has not distributed here.

Materials and Methods

We studied the Caucasus fir seed from the Algeti Reserve which is the utmost east edge of fir habitat. The qualitative parameters of this species were compared with the ones from the optimal climatic zone (Shovi). Dynamics of natural reproduction and development of vital forms of undergrowth at different light conditions were established.

The light by luxmeter, air temperature by thermograph, relative humidity by hygograph and temperature of soil surface by Saviniv's thermometer of the undergrowth of both species were studied. The lengths of undergrowths were 1.5 m.

Results and Discussions

The qualitative and quantitative parameters of fir seeds from the optimal climatic zone and utmost east edge of habitat are presented in the Table 1.

Table 1. Parameters of fir seeds from optimal climatic zone and utmost east edge of habitat

N	weight of 1000 seeds (kg)	potential of germination of 100 seeds (%)	amount of not germinated seeds (%)			total amount of not germinated seeds (%)	Yield of seeds from 1 cone (g)
			full, not germinated	Rotten	empty		
optimal climatic zone							
1	0.98±0.05	80±3	9.3±1.7	7.2±1.3	4.2±0.3	20±3	20.1±2.0
2	0.86±0.07	86±3	7.1±0.3	4.5±1.7	3.5±1.3	14±3	16.3±1.8
3	0.95±0.02	82±1	6.5±1.3	6.3±0.3	6.1±1.7	18±1	18.0±0.1
utmost east edge of habitat							
1	0.78±0.03	59±1	22±1	10.0±0.1	9.3±0.7	41±1	14.3±2.3
2	0.77±0.02	61±3	18±4	9.1±1.0	12.2±2.3	39±3	10.1±1.9
3	0.71±0.04	54±4	27±5	11.0±1.1	8.5±1.7	46±4	11.8±0.2

As is seen from the table qualitative parameters of fir seeds of optimal climatic zone are prevailed over corresponding parameters of the utmost east edge of habitat. Due to high relative humidity necessary for the fir as mesophyte specimen, in optimal climatic zone there are favourable conditions for growth and development of fir. At the utmost east edge, because of dry air and low atmospheric precipitates, fir development is confined and correspondingly seeds parameters are low.

To study the effect of light intensity on growth and development of fir seedling in natural conditions various light regime was settled: total light, middle light (50% of total light) and strong eclipse (20% of total light). Various light regime was conducted by covering of seedlings with gauze of different thickness; light intensity was controlled by luxmeter. At the end of vegetation period fir one-year sprouts were taken from the soil and their sizes and weights were determined.

At the middle light fir seedling was characterized with the best development and at the strong eclipse - with the worst. As for total light conditions mass death with root cervix burning was observed caused by direct sun radiation.

In Algeti Reserve vital forms of fir undergrowth were studied. On their development among with light conditions air relative humidity is considerably influenced. We revealed 4 types of undergrowth that were developed in different cenosis and at different light conditions. Normal developed undergrowths occur in middle size clearings where maximal light is 30 000-35 000 lux. Undergrowth which grow in middle frequency coniferous forest (0.5-0.6) are characterized with inhibited growth where maximal light is 20 000-22 000 lux. Lacking vitality undergrowth, annual growth of which is negligible, occur in high frequency coniferous forest (0.8-0.9) where maximal light is 7 000-8 000 lux. If light conditions of the last one should not be improved they will die.

Table 2. Fir undergrowth types according to their vital forms

vital forms of undergrowth	occurrence	age	height (m)	increase of height (cm)	shape of stem	shape of crown	colour of needles
normal developed	clearing	20-25	2.0	up to 12	straight	Conic	deep green
with inhibited growth	coniferous forest of 0.5-0.6 frequency	25-30	1.5	up to 5	straight, slight crooked	Conic	light green
lacking vitality	coniferous forest of 0.7-0.8 frequency	35-45	1.2	up to 1	often crooked	umbrella	light green, yellowish
dried up	coniferous forest of 0.9-1.0 frequency	45-50	1.0	-	crooked	Flag	yellow

As is seen from Table 2. type of fir undergrowth according to viability is estimated by complex of morphological characteristics (age, height, needles colour, crown form, increase of height). The most important characteristic of undergrowth viability is growth in height. According to literature data if during the last 5 years undergrowth's height increases more than 10 cm, it is considered as normal type [Japaridze, et al., 2001].

It is worth to mention that undergrowths height of which in vegetation period increased insignificantly, in forestry considered as "hopeless". This viewpoint is not correct, as fir being shade hardiness species during long period can exist in low light intensity conditions under the cover of high frequency coniferous forest. Improving the light conditions this category of undergrowth begin to grow intensely, as if it was not suppressed.

We can conclude that at the very east edge of habitat low quality, weak development of undergrowth, even in optimal light conditions, of Caucasus fir seeds is caused by low atmospheric precipitates and dry climatic conditions. So, it is quite justifiable to cultivate this subendemic species in humid regions of inner Kakheti where because of historical climatic changes this species doesn't exist. But now there are optimal conditions for fir growth and development. Such actions should artificially increase Caucasus fir habitat to the east.

References:

- Chochieva K. *About History of Dark Coniferous Forests of Georgia*. Proc. Acad. Sci. Georgian SSR, **8**, 2, 1975.
- Dolukhanov A. *Forest Flora of Georgia*. I, Tbilisi, "Metsniereba", 1989 (in Russ.).
- Flora of Georgia*. Tbilisi, "Metsniereba", 2001 (in Georgian).
- Gulisashvili V. *General Forestry*. Tbilisi, "Metsniereba", 1974 (in Georgian).
- Japaridze T., Kobakhidze N., Abulashvili T. *Spruce and Fir Sprouts Viable Forms in Dark Coniferous Biocenosis*. Proceedings of The Institute of Mining Forestry. **XIII**, Tbilisi, 'Metsniereba', 2001 (in Georgian).
- Kolakovski A. *Catalogue of Fossil Plants of Caucasus*. Tbilisi "Metsniereba", 1973 (in Russ.).
- Palibin I. *Steps of Development of Near Caspian Countries Flora of Cretaceous Period*. M.-L., "Nauka", 1936.
- Shatilova I., Ramishvili I. *Data of the History of Georgian Flora*. Tbilisi "Metsniereba", 1990 (in Georgian).

კავკასიური სოჭის (*Abies nordmanniana*) თესლმსხმოიარობა და აღმონაცენ - მოზარდის ფორმირება არეალის აღმოსავლეთ საზღვარზე

ძოწენიძე ნ.

ბოტანიკი კათედრა, ქუთაისის ა. წერეთლის სახ. სახელმწიფო უნივერსიტეტი

(მიღებულია 11.07.2005)

რეზიუმე

შესწავლილია კავკასიური სოჭის არეალის უკიდურეს აღმოსავლეთ საზღვარზე მისი თესლმსხმოიარობის თავისებურება და თესლების ხარისხობრივი მანვენებლები. მიღებული შედეგები შედარებულია ოპტიმალური სარტყლის სოჭის თესლების შესაბამის მანვენებლებთან. შესწავლილია სოჭის აღმონაცენის ზრდა-განვითარების თავისებურებები და მორფოლოგიური ნიშან-თვისებების ერთობლიობით გამოვლენილია სოჭის მოზარდის სახიცოცხლო ცენოტიპები. მიუხედავად სასინათლო ოპტიმუმისა, სოჭის თესლების დაბალი ხარისხობრივი მანვენებლები და მოზარდის შეფერხებული ზრდა-განვითარება არეალის უკიდურეს აღმოსავლეთ საზღვარზე ძირითადად გამოწვეულია ნალექების სიმცირით და ჰაერის დაბალი შეფარდებითი ტენიანობით.

FUNCTIONAL CHARACTERISTICS OF CHROMOSOMES IN PATIENTS WITH ALZHEIMER'S DISEASE AND VASCULAR DEMENTIA

TABATADZE N., DADUNASHVILI E., JOKHADZE T., ZOSIDZE N., LEZHAVA T.

Department of Genetics, Tbilisi State University, Tbilisi, Georgia.

(Received June 2, 2005)

Abstract

The level of chromosomal instability (structural and numerical), transcriptional activity of ribosomal genes and polymorphism of C-heterochromatin have been studied in patients affected with late-onset type of Alzheimer's disease (AD) and vascular dementia (VD). Cytogenetic studies were conducted on PHA-stimulated peripheral blood lymphocyte cultures derived from 26 demented individuals (11 with late-onset sporadic form of AD and 15 with VD) and 20 healthy donors (comprising equal numbers of elderly (70-80 yrs old) and young (20-30 yrs old)). According to the obtained results, no difference regarding the number of Ag-positive chromosomes per cell was observed in the patients affected with both forms of dementia and the indices were in the range of elderly control value. However, statistically significant increase in the frequencies of structural and numerical chromosome aberrations was revealed in case of VD but not in AD patients as compared with elderly controls. In addition, altered polymorphism of structural C-heterochromatin, in particular, increased level of large (d) variants was observed in AD group that might be helpful for distinguishing the two forms of dementia.

Key words: Alzheimer's disease, vascular dementia, aberration, heterochromatin, Ag-band, C-band, association of acrocentric chromosomes.

Introduction

Since the discovery of inherited dementias, their cytogenetic investigation has been intensively started [Hedera, Turner, 2003]. In spite of the numerous scientific data, it is not possible yet to distinguish one approach or direction from the other because the experimental results differ from each other within the vast range [Smith et al., 1983; Melargano et al., 1991; da Silva et al., 2000]. AD is an irreversible neurodegenerative disorder of the brain, robbing it of memory, and eventually, overall mental and physical functions, leading to death [Ritchie, Lovestone, 2002; Clark, Karlawish, 2003]. It is the most common cause of dementia among people over the age of 65 [Blanchard et al., 2004; Katzman, 2004]. Various susceptible genes (on chromosomes 1, 14, 19 and 21) are identified for the late-onset type of AD and genome screening experiments are still performed in order to reveal additional loci responsible for this harmful disorder [Panza, 2002]. The key neuropathological hallmarks consist of the extracellular senile plaques and intracellular neurofibrillary tangles [Rachakonda et al., 2004]. The exact diagnosis can be made only after detection of the mentioned abnormal structural signs in the post mortem brain sections [Clark, Karlawish, 2003; Blanchard et al., 2004; Katzman, 2004].

In vascular dementia, which is commonly caused by a stroke or a series of small strokes, brain cells are deprived of oxygen and die. It is sometimes difficult to determine whether people have AD or VD. It is very important to identify the type of dementia, because the treatment for AD and VD are quite different [Leach et al., 2004]. Various genetic analyses have been carried out in order to classify dementias and to understand the main genetic mechanisms involved in their pathogenesis. Therefore, the aims of our research were to assess the rate of chromosomal instability, to study the nucleolus organizer regions (NORs) and structural heterochromatin at these different forms of dementia.

Material and methods

Cytogenetic investigations were carried out on peripheral blood lymphocyte cultures of demented individuals (70-90 years of age): 11 patients with late-onset sporadic form of AD (diagnosed in the Institute of Neurology), 15 patients with VD and 20 healthy subjects (elderly control group - 10 individuals of 70-80 yrs. Old; the young control group - 10 individuals of 20-30 yrs. old).

Cultivation of lymphocytes, preparation and staining of chromosome slides were performed by standard methods. The level of spontaneous chromosomal aberrations (structural and numerical) was studied as described by Lezhava (1999). The frequencies of structural and numerical chromosomal disorders were assessed in 977 metaphases of 11 patients with AD, 1485 metaphases of 15 individuals with VD, in 1000 metaphases of young and 1000 metaphases of old control subjects.

The activity of ribosomal genes of acrocentric chromosomes was determined in 533 metaphases from 11 AD patients, 750 metaphases from 15 VD patients and in 1000 metaphases of both control groups. The experiments were carried out on the base of Ag-staining intensity and the frequency of acrocentric chromosome associations according to the Ag-banding methodology [Lezhava, 1999]. The probability of argyrophillic NORs and the frequency of satellite associations of acrocentric chromosomes were tested using the comparison of binomials.

The structural C-heterochromatin has been examined on the base of method described by Fernandez et al., (2002). 200 metaphases from demented individuals and 200 metaphases from healthy controls were analyzed. The types of C-segment variants were determined using Patil and Lubs' classification system: a<0,5x16p; b<0,5-1x16p; c>1,5x16p; d>1,5-2x16p; e>2x16p. Statistical analysis was performed by Zax formula:

$$\chi^2_{(k-1)} = (n+m) \frac{n}{m} \left\{ \sum_{i=1}^k \frac{\left(\frac{V_i}{n}\right)^2}{\frac{V_i + \mu}{n+m}} - 1 \right\}$$

Results and Discussion

Chromosomal aberrations. Noticeably, the variability towards the chromosome structural disorders was revealed among the affected (AD and VD) individuals. The frequency of aberrant chromosome-containing cells in AD group equaled $5,9 \pm 2,36\%$ that is approximately in the same range as the corresponding value in elderly controls ($3,1 \pm 0,55\%$), but is higher than in young controls ($1,6 \pm 0,4\%$). As for VD group, the statistically significant increase in chromosome

damages was revealed ($8,9 \pm 2,85\%$) in comparison with control subjects. The chromosome disorders mainly involved single and paired fragments, while chromatid and chromosome translocations were also registered. Chromosome breaks had a random distribution.

The increased frequency of numerical chromosomal disorders (both aneuploidy and polyploidy) was registered in the second (VD) group of demented individuals ($17,9 \pm 3,8\%$ and $0,18 \pm 0,09\%$ respectively) as compared with elderly and young control groups ($13,5 \pm 1,08\%$ and $0,04 \pm 0,02\%$ and $6,0 \pm 0,75\%$; $0,02 \pm 0,01\%$ respectively). Whereas, the total index of numerical disorders was slightly higher in AD group patients ($15,4 \pm 3,6\%$ and $0,07 \pm 0,03\%$ respectively) than in elderly controls. The obtained results are in agreement with the scientific data [Smith et al., 1983; Trippi et al., 2001].

Transcriptional activity of ribosomal genes. Human ribosomal genes are localized in the secondary constrictions (NORs) - in satellite stalks (heterochromatic regions) of acrocentric chromosomes. It was revealed that silver staining (Ag-banding) intensity corresponds to the level of NORs functioning in the previous interphase [Stitou et al., 2000]. The ability of acrocentric chromosomes to connect in order to form associations is determined by the presence of two chromatid satellite stalks [Lezhava, 1999, 2001]. The associative activity of the strands positively correlates with the intensity of Ag-banding. The absence of silver stained regions indicates inactivation of ribosomal genes [Trere, 2000; Kadotani et al., 2001].

The analysis of Ag-positive NORs in intact lymphocytes derived from AD and VD patients showed that both forms of dementia are characterized with the same level of transcriptional activity of ribosomal cistrons ($5,3 \pm 0,33\%$ AD group ($P < 0,01$); $5,2 \pm 0,32\%$ - VD group ($P < 0,01$). These indices correspond to the value of elderly controls ($5,23 \pm 0,32\%$) and are less than in young controls ($6,35 \pm 0,25\%$). The frequencies of acrocentric chromosome associations in both groups of affected individuals equaled to $42,1 \pm 4,9\%$ (AD) and $42,4 \pm 4,9\%$ (VD) that was in agreement with the corresponding value in old individuals ($42,2 \pm 2,4\%$) but not in young controls ($51,56 \pm 2,36\%$). It is notable that GG type of acrocentric chromosome associations was significantly increased in both cases of dementia ($14,5 \pm 3,5\%$ and $12,8 \pm 3,3\%$) in comparison with healthy donors ($6,0 \pm 2,37\%$ and $9,0 \pm 2,86\%$ respectively). Our data correspond to the scientific investigations [Lu et al., 1998].

Heteromorphism of structural heterochromatin. The polymorphism towards the variants of structural C-heterochromatic bands on chromosomes 1, 9 and 16 was revealed in both forms of dementia. Generally, the frequency of small a-segments was increased on chromosomes 1, 9 and 16 in case of whole VD group as compared with elderly controls while the frequency of large c-variants similar to the AD group was reduced on chromosome 16 in comparison with control subjects ($\chi^2_{16} = 11,62$ $P < 0,01$; $\chi^2_{9} = 13,47$ $P < 0,01$; $\chi^2_{1} = 11,69$ $P < 0,01$). The total index of large d-variants within the AD group patients was significantly increased on chromosome 1 and decreased on chromosomes 9 and 16 in comparison with VD patients and control subjects ($\chi^2_{16} = 8,05$ $P < 0,01$;

$\chi^2_{9} = 2,96$ $P > 0,05$; $\chi^2_{1} = 10,75$ $P < 0,01$). According to the obtained results we can conclude that the polymorphism of various C-structural heterochromatic bands revealed in the two groups of demented individuals.

Our results indicate that Alzheimer's disease and VD patients are characterized with different levels of chromosomal instability (statistically significant increase in the frequencies of structural and numerical chromosome disorders in case of VD but not in AD patients as compared with elderly controls). In addition to this, altered polymorphism of structural C-heterochromatin, in

particular, increased level of large (d) variants was observed in case of AD group that might be useful for the distinguishing of different forms of dementia. .

References:

- Blanchard B., Chen A., Rozeboom L., Stafford K., Weigle P., Ingram V. *Efficient reversal of Alzheimer's disease fibril formation and elimination of neurotoxicity by a small molecule*. Proc. Natl. Acad. Sci. USA, **101**, 4326-14332, 2004.
- Clark C., Karlawish J. H. *Alzheimer's disease: current concepts and emerging diagnostic and therapeutic strategies*. Ann. Intern. Med., **138**, 400-410, 2003.
- da Silva A., Payao S., Borsatto B., Bertolucci P., Smith M. *Quantitative evaluation of the rRNA in Alzheimer's disease*. Mech. Ageing Dev., **120**, 57-64, 2000.
- Fernandez R., Barragan M., Bullesos M., Marchal J., Diaz L., Sanchez A. *New C-band protocol by heat denaturation in the presence of formamide*. Hereditas, **137**, 145-148, 2002.
- Hedera P. *Turner R Inherited dementias*. Neurol. Clin., **20**, 779-808, 2003.
- Kadotani T., Watanabe Y., Saito T., Sawano K., Minatozaki K. *A chromosomal study on 100 cases of cerebral palsy*. Int. J. Hum. Genet., **1**, 109-112, 2001.
- Katzman R. *Neurologist's view of Alzheimer's disease and dementias*. Int. Psychogeriatr., **16**, 259-73, 2004.
- Leach N., Rehder C., Jensen K., Holt S., Jackson-Cook C. *Human chromosomes with shorter telomeres and large heterochromatin regions have a higher frequency of acquired somatic cell aneuploidy*. Mech. Ageing Dev., **125**, 563-573, 2004.
- Lezhava T. *Chromosomes in very senile age: 80 years and over*. M, Nauka, 1999.
- Lezhava T. *Chromosomes and aging: genetic conception of aging*. Biogerontology, **2**, 253-260, 2001.
- Lu W., Tang H., Fan M., Mi R., Wang L., Jia J. *Research on nucleolar organizer regions of hippocampal neurons in Alzheimer's disease*. Chin Med J., **26**, 282-184, 1998.
- Melargano M., Smith A., Kormann-Bortolotto M., Toniolo J. T. *Lymphocyte proliferation and sister chromatid exchange in Alzheimer's disease*. Gerontology, **37**, 293-298, 1991.
- Panza F., Solfrizzi V., D'Introno A., Capurso C., Colacicco A., Torres F., Altomare E. Capurso. *Genetics of late-onset Alzheimer's disease: vascular risk and beta- amyloid metabolism*. Recent. Prog.Med., **93**, 489-497, 2002.
- Patil S., Lubs M. *Classification of 9th regions in human chromosomes 1, 9 and 16 by C-banding*. Hum Genet., **38** 35-38, 1997.
- Rachakonda V., Hong Pan T., Dong Le W. *Biomarkers of neurodegenerative disorders: how good they are?* Cell. Research., **14**, 2004 349-360 2004.
- Ritchie K., Lovestone S. *The dementias*. Lancet, **360**, 1759-1766, 2002.
- Smith A., Broe G., Williamson M. *Chromosome aneuploidy in Alzheimer's disease*. Clin. Genet., **24**, 54-57, 1983.
- Stitou S., Diaz de la Guardia R., Jimenes R., Burgos M. *Inactive ribosomal cistrons are spread throughout the B chromosomes of Rattus (Rodentia, Muridae). Implications for their origin and evolution*. Chromosome Res., **8**, 305-311, 2000.
- Trere D. *Ag-NOR staining and quantification*. Micron, **31**, 127-131, 2000.
- Trippi E., Botto N., Scarpato R. *Spontaneous and induced chromosome damage in somatic cells of sporadic and familial AD patients*. Mutagenesis, 323-327, 2001.

**ქრომოსომათა ფუნქციური მახასიათებლები ალცჰეიმერის
დაავადებისა და მასკულარული დემენციის დროს**

ტაბატაძე ნ., დადუნაშვილი ე., ჯოხაძე თ., ზოსიძე ნ., ლეჟავა თ.

ივ. ჭავჭავაძის სახ. თბილისის სახელმწიფო უნივერსიტეტის გენეტიკის
კათედრა

(მიღებულია 02.06.2005)

რეზიუმე

შესწავლილ იქნა ქრომოსომათა დარღვევები, რიბოსომული გენების ტრანსკრიფციული აქტივობა და C-სტრუქტურული ჰეტეროქრომატინის პოლიმორფიზმი დემენციების სხვადასხვა ფორმების - ალცჰეიმერის დაავადებისა (აღ) და ვასკულარული დემენციის (ვღ) შემთხვევებში. ვღ დროს გამოვლენილ იქნა ქრომოსომათა სტრუქტურული-რაოდენობრივი დარღვევების სისშირის სტატისტიკურად მაღალსარწმუნო მატება. აღ შემთხვევაში აღნიშნული მანვენებლები ხანდაზმული ინდივიდებისათვის საკონტროლო დონეზე რჩებოდა. C-სტრუქტურული ჰეტეროქრომატინის პოლიმორფიზმის მაღალი დონე (დიდი ზომის d-ვარიანტები) დაფიქსირდა აღ შემთხვევაში. მიღებული მინაცემები დასაშვებია გამოყენებულ იქნას დემენციათა ფორმების დიფერენცირებისათვის.

EFFECT OF MERCURY CHLORIDE ON CONFORMATIONAL CHANGES OF CHROMATIN

TADUMADZE N., BABLISHVILI N., NIBLADZE N., SULABERIDZE S., BUADZE T.,
MONASELIDZE J.*

Department of Genetics, Iv. Javakishvili Tbilisi State University
** Institute of Physics, Georgian Academy of Sciences*

(Received June 2, 2005)

Abstract

The effect of mercury chloride on conformational parameters of chromatin of cultivated lymphocytes of peripheral blood was studied. In the conditions of influence of mutagenic concentrations of mercury chloride, conformational modifications as heterochromatinization was revealed. Capability of determination of mutagenesis and antimutagenesis mechanisms and management of these processes were discussed.

Key words: microcalorimeter, condensation, heterochromatinization.

Introduction

It is known that metal ions determine processes of stabilization-destabilization and different forms of equilibrium of secondary and tertiary structures of nucleic acids [Hakl, 2000; Jokhadze & Lezhava, 1994]. In scientific literature there are no data concerning the effect of mercury ions on conformational changes of chromatin. As it was established that conformational changes take decisive part in the processes of mutation [Jokhadze & Lezhava, 1994], it should be supposed that such influence may be connected with mutagenic activity of mercury ions

The goal of the work was to study effect of mercury ions on conformational parameters of chromatin.

Materials and Methods

As the object of research short time cultures of lymphocytes of peripheral blood of 20-30 years old clinically healthy donors were used. Cultivation and fixation of the cells were carried out by standard method.

Degree of chromatin condensation was studied by the method of differential scanning microcalorimetric method [Monaselidze & Bakradze, 1973]. This method is based on the fact that different fractions of chromatin revealed different ability of heat absorption. Data received by the effect of studied agent were compared with those of intact cells of the same individuals.

On the temperature scale condensed and decondensed chromatins are presented by pick and correspond to exactly defined positions. Intact cells melting take place in the temperature range: 40-120°C. Absorption curve has four maximums: 69±1°C, 83.5±1°C, 95±1°C and 106±1°C and sharply defined sides at - 46°C, 57°C, 61°C. It is known that the chromatin melting

corresponds to the picks - 95°C and 106°C, namely 95°C corresponds to 10 nm – fibril, and 106°C to 30 nm – solenoid.

Chromatin condensation graphically was expressed by thermal capacity curve. Parameter of thermal capacity of decondensed chromatin (euchromatin) reaches maximal value at 66°C which on temperature scale of heat absorption makes pick. This parameter for heterochromatin is about 80°C. Changes of maximal values of heat absorption, as well as displacement of the pick on the temperature scale are characteristics of changes of ratio of euchromatin and heterochromatin.

Results and Discussion

We considered that study of the influence of some substances in the viewpoint of modifier activity of chromatin as prerequisite of operation of activation-inactivation processes of chromosome domains and located in them genes should provide perspective guideline [Costa & Klein, 1999]. So we studied effect of mercury chloride on conformational state of chromatin of human cultivated lymphocytes.

On the base of mutagenic activities of different concentrations of $HgCl_2$ solutions effect of saline of maximal mutagenic concentration ($10^{-3}M$) on the degree of chromatin condensation was determined by differential scanning microcalorimetric method. Curves of thermal capacity of chromatin was studied on unstimulated lymphocytes of 5 donors of 20-30 years.

Mercury ions in unstimulated lymphocytes distinct to intact cells cause considerable changes of melting curve. Shift to the right of the curve of heat absorption takes place which indicates to the effect of studied agent on the chromatin condensation - heterochromatination (Fig.1).

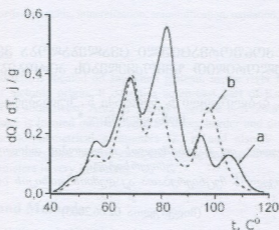


Fig. 1. Conformational changes of chromatin caused by effect of $HgCl_2$
a. Intact cells; b. Treated with $HgCl_2$ cells.

According to received data we should say that mechanism of mutagenic effect of mercury chloride on lymphocyte cultures should be connected with revealed by us ability of mercury ions to cause considerable conformational changes of chromatin and by this way to get DNA molecule less acceptable for reparational ferments, and as a result - to hinder reparation of DNA

Above mentioned is in accordance with the data which point to the role of weakening of DNA reparational system in mutagenesis caused by the effect of metal ions [Privezentsev et al., 1996; Guecheva et al., 2001; Meuron et al., 2001].

Our consideration is also supported by the data that chromosomal and gene mutations induced by heavy metals ions are specific for chromosomes' heterochromatic regions [Costa & Klein, 1999].

Thus, according to our studies conformational modification of chromatin caused by $HgCl_2$ should be considered as significant parameter for explanation of the mechanisms of the effect of mercury chloride.

References:

- Costa M., Klein C.B. *Nickel carcinogenesis mutation, epigenetic or selection*. EHP, 107-109, 1999.
- Guecheva T., Henriques J., Erdtman B. *Genotoxic effect of cooper sulphate in freshwater planarium in vivo studied with the single-cell gel test (comet assay)*. Genetic Toxicology and Environmental Mutagenesis, **497**, 1-2, 19-27, 2001.
- Hakl E. *Effect of metal ions on structural changes of DNA in alcoholic and urea solutions*. PhD abstract, 2000.
- Jokhadze T., Lezhava T. *Study on structural changes of chromosomes induced by heavy metal salts during in vivo and in vitro aging*. Genetics, **30**, 12, 1630-1632, 1994 (in Russian).
- Meuron S., Golichova C., Dulout F. *DNA damage by Cadmium and Arsenic salts assessed by the single cell gel electrophoresis assay*. Genetic Toxicology and environmental mutagenesis, **498**, 1-2, 47-55, 2001.
- Monaselidze J., Bakradze G. *Conformational changes of biopolymers in solution*. M., "Nauka", 300-304, 1973 (in Russian).
- Privezentsev K., Siroma N., Gaziev A. *Study of genetoxic effects of cadmium in vivo*. Cytology and Genetics, **30**, 3, 45-51, 1996 (in Russian).

ქრომატინის კონფორმაციული ცვალეზადობა ვერცხლისწყლის ქლორიდით ზემოქმედების პირობებში

თადუმაძე ნ., ბაბლიშვილი ნ. ნიბლაძე ნ., სულაბერიძე ს., ბუაძე თ., მონასელიძე ჯ.*

გენეტიკის კათედრა, ივ. ჯავახიშვილის სახელობის თბილისის სახელმწიფო უნივერსიტეტი

*საქართველოს მეცნიერებათა აკადემიის ფიზიკის ინსტიტუტი

(მიღებულია 02.06.2005)

რეზიუმე

შესწავლილია ადამიანის სისხლის ლიმფოციტებში ქრომატინის კონფორმაციული ცვალეზადობა ვერცხლისწყლის ქლორიდით ზემოქმედების პირობებში დიფერენციალური მიკროკალორიმეტრული მეთოდის საშუალებით. გამოვლენილია დენატურაციის მრუდის გადახრა და ქრომატინის კონფორმაციული ცვალეზადობა ჰეტეროქრომატინიზაციის სახით. ვერცხლისწყლის იონების მუტაგენური ზემოქმედების მექანიზმის ასახსნელად ქრომატინის სტრუქტურული მოდიფიკაცია მნიშვნელოვან პარამეტრად შეიძლება ჩაითვალოს.

CHEMICAL COMPOSITION OF THE WATER AND PLANKTON'S PRIMARY PRODUCTION OF THE LAKE KARTSAKHI (KHOZAPINI)

TSISKARISHVILI L., TSISKARISHVILI M.

Institute of Zoology, Georgian Academy of Sciences

(Received June 7, 2005)

Abstract

Basic abiotic parameters, the activity of photosynthesis and respiration, level of the primary production of the lake Kartsakhi (Khozapini) were studied. Common mineralization of the water varies from 727.32 to 915.17 mg/l; activity *A* viz. 6.04-12.41 mg/l at the water surface. In summer period ΣA viz. 3.21-4.50 g O₂/m² per day; ΣR viz. 2.26-3.84 g O₂/m² per day. The ratios $\Sigma A/\Sigma R$ and $\Sigma \Sigma A/\Sigma \Sigma R$ show that the primary production of plankton accumulates and inserts in the mud producing processes. The lake Kartsakhi belongs to the eutrophic reservoirs.

Key words: eutrophic, mineralization, ammonification, oxidation, biogenic.

Introduction

The lake Kartsakhi is located on Javakheti plateau, in the river Mtkvari basin, on 1799 m.a.s.l. The lake is of tectonic origin. It occupies south-east part of Kartsakhi synclinal basin spread from north-east to south-west. Area of the lake surface is $S=26.3$ km², $Z=1$ m, $z=0.73$ m, $V=19.3 \cdot 10^6$ m³. The lake is located on the Georgia-Turkey border and due to the absence of the bathymetrical map its morphometrical parameters (Z , z) and also V are roughly estimated [Aphazava, 1975]. So, to determine hydrological and biological resources of the lake, the precise definition of these parameters is very important.

Materials and Methods

Temperature, clarity and pH of the water were determined in two points located on 20 m and 200 m from the lakeside.

Photosynthesis activity (*A*) and respiration (*R*) of phytoplankton was measured in August 2001-2002 in two stations. In both of them the water was taken from the surface of lake; in one of the stations – at the depths of *0S*, *0.5S*, *1S*, *2S*, *3S*, *4S* (S – clarity of water by Sekky disc) and at the bottom. The bottles after being filled with water were exposed at corresponding depths *in situ* for 24 hours.

Annual gross primary production of plankton was estimated by increasing daily maximal values of gross primary production obtained 100 times during the vegetation period [Vinberg, 1960; Bulion, 1994].

Results and Discussion

In summer period temperature of water reaches to 23.4°C. The main reason of summer temperature differences is weather conditions of year, as well as time of measurement during the day (Table 1.)

pH of water is weak alkaline and don't exceed 8.0 which is caused by activity of photosynthesis of phytoplankton. This high photosynthetic activity also defines low means of clarity which don't exceed 30 cm. In the morning content of dissolved free O₂ in water of surface layer varies from 7.71 to 8.57 mg O₂/l, percentage of saturation by O₂ – 111.23-114.50%.

Table 1. Temperature, clarity, pH, dissolved free O₂ in water, percentage of water

Time of observation	30.08.01	08.08.02	14.06.04
Temperature (°C)	19.0	23.4	18.2
Clarity, S (cm)	20	30	25
pH	7.8	8.0	7.6
O ₂ mg/l	8.57	7.71	9.95
% of saturation by O ₂	114.45	111.23	114.50

Value of permanganate oxidation indicating content of organic compounds is high and varies from 24.65 to 28.95 mg O₂/l (Table 2).

Distinct to deep reservoirs for shallow reservoirs located both in low and hilly zones of Georgia similar and higher values of permanganate oxidation are characterized [Tsiskarishvili, 1989; 2000; 2002].

Comparative low content of NO₃⁻ and P is connected with their intensive consumption at synthesis of primary organic compounds by phytoplankton; with its aminization high content of NH₄⁺ - 0.15 mg/l is observed (Table 2).

Table 2. Values of permanganate oxidation of water and biogenic elements in the lake Kartsakhi.

Time of observation	30.08.01	08.08.02	14.06.04
Oxidation (mg O ₂ /l)	28.95	28.74	24.65
NH ₄ ⁺ (mg/l)	0.10	0.15	0.08
NO ₂ ⁻ (mg/l)	0.02	0.01	0.01
NO ₃ ⁻ (mg/l)	2.00	1.50	1.80
P _{dissolved mineral} (µg/l)	5	5	10

Common mineralization of water in summer varies from 727.32 to 915.17 mg/l. Due to melting of snow and spring rains common mineralization of water in the beginning of summer is considerably lower than at the end. Value of mineralization of the lake Kartsakhi 5 times and more is higher than in other reservoirs of this region [Apkhazava, 1975; Tsiskarishvili, 2000; 2002]. Distinct from region's other reservoirs water of this lake by classification of natural waters [Aleksin, 1970] belongs to hydrogen carbonate class of sodium group, while water of other reservoirs of this region is of calcium group. Water index of the lake Kartsakhi is C₁^{Na}. To establish the reason of this exclusion it is necessary to carry out corresponding complex studies.

Table 3. Values of the main ions of saline composition and common mineralization of the lake Kartsakhi water

Time of observation	Na ⁺ +K ⁺ (mg/l)	Ca ⁺² (mg/l)	Mg ⁺² (mg/l)	HCO ₃ ⁻ (mg/l)	SO ₄ ⁻² (mg/l)	Cl ⁻ (mg/l)	Σ _K +Σ _A (mg/l)
30.08.01	190.08	17.20	31.44	610.00	64.32	2.13	915.17
08.08.02	166.80	20.00	36.00	600.24	51.31	1.49	875.84
14.06.04	154.32	11.20	24.48	492.88	41.28	3.16	727.32

Data of the studies show that intensive development of phytoplankton determines high parameters of primary production at the surface, as well as at 1m² from the surface per volume unit for a day. Activity *A* at the surface varies from 6.04 to 12.41 mg O₂/l for a day. Pure photosynthesis (*A_p*) was recorded to the depths with values approximately equaling values of doubled *S* (clarity) of the water. With the increase of the depth activity - *A* was decreased and at the depth equaling tripled *S* it made up 4-10% of *A* at the surface of water (Fig.1). In the summer period gross primary production at 1m² (ΣA) varied from 3.21 to 4.50 g O₂/m² per day, and destruction (ΣR) viz. 2.26-3.84 g O₂/m² per day (Table 4).

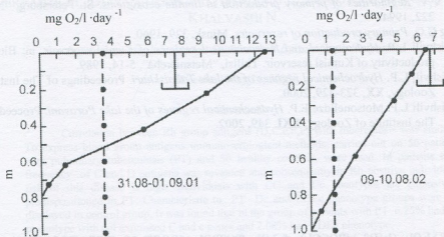


Fig. Dependence of the activity *A* (—) and destruction *R* (---) on the depth and clarity (Δ) of the water of the lake Kartsakhi, recorded in: a) August-September, 2001; b) August-September, 2002.

According to the method of Vinberg [Vinberg, 1960; Bulion, 1994] annual gross primary production of plankton of the lake Kartsakhi in 2001 was 1530 and in 2002 – 1091 g O₂/m² a year; destruction – 1306 and 768 g O₂/m² a year correspondingly (Table 4). Ratios $\Sigma A/\Sigma R$ and $\Sigma \Sigma A/\Sigma \Sigma R$ more than 1 indicate that bulk of organic compounds formed due to photosynthetic activity of phytoplankton accumulate in the lake and take part in the process of mud formation.

It was established that in the lake ecosystems fish catching makes about 0.11-0.13% of annual gross primary production of plankton [Bulion, 1994] and accordingly to preserve and defend natural reproduction of fish resources fish catching in the lake Kartsakhi should not exceed 12-18 kg/ha a year.

Table 4 Biotic balance of the lake Kartsakhi

daily	biotic	balance	annual	biotic	balance
Time of observation	2001	2002	Time of observation	2001	2002
ΣA g O ₂ /m ²	4.50	3.21	$\Sigma \Sigma A$ g O ₂ /m ²	450	321
ΣA kkal/m ²	15.30	10.91	$\Sigma \Sigma A$ kkal/m ²	1530	1091
ΣR g O ₂ /m ²	3.84	2.26	$\Sigma \Sigma R$ g O ₂ /m ²	1306	768
ΣR kkal/m ²	13.06	7.68	$\Sigma \Sigma R$ kkal/m ²	1306	768
$\Sigma A/\Sigma R$	1.17	1.42	$\Sigma \Sigma A/\Sigma \Sigma R$	1.17	1.42

By the value of gross primary production of photoplankton the lake Kartsakhi, as well as other shallow reservoirs of Javakheti plateau belongs to eutrophic reservoirs.

Acknowledgments. The work was supported by John D. & Kathryn T. Macarthur Grant N1-681-00.

References:

- Aleksin O.A. *Bases of Hydrochemistry*. Leningrad, "Nauka", 441, 1970.
Apkhazava I.S. *Lakes of Georgia*. Tbilisi, 180, 1975.
Bulion V.V. *Regularities of primary production in limnite ecosystems*. St. Petersburg, "Nauka", 222, 1994.
Vinberg G.G. *Primary production of reservoirs*. Minsk, 329, 1960.
Tsiskarishvili L.P. *Hydrological and hydrochemical regimes of Kumisi reservoir*. in: Biological productivity of Kumisi reservoir. Tbilisi, "Metsniereba", 5-14, 1989.
Tsiskarishvili L.P. *Hydrochemical regimes of the lake Tabatskhuri*. Proceedings of The Institute of Zoology, **XX**, 323-329, 2000.
Tsiskarishvili L.P. Motsonelidze E.P. *Hydrochemical regimes of the lake Paravani*. Proceedings of The Institute of Zoology, **XXI**, 349, 2002.

კარწახის (ხოზაფინის) ტბის წყლის ქიმიური შემადგენლობა და პლანქტონის პირველადი პროდუქცია

ცისკარიშვილი ლ. ცისკარიშვილი მ.

საქართველოს მეცნიერებათა აკადემიის ზოოლოგიის ინსტიტუტი

(მიღებულია 07.06.2005)

რეზიუმე

ნაჩვენებია, რომ ზაფხულის პერიოდში კარწახის ტბის წყლის საერთო მინერალიზაცია მერყეობს 727.32 მგ/ლ-დან 915.17 მგ/ლ-მდე. წყლის ზედაპირთან სინქარე A იცვლება 6.04 მგ/ლ-დან 12.41 მგ/ლ-მდე. ΣA და ΣR დღე-ღამეში ვარირებს შესაბამისად შემდეგ ინტერვალებში: 3.21-4.50 გ O₂/მ² და 2.26-3.84 გ O₂/მ² დღე-ღამეში. ფარდობები ΣA/ΣR და ΣΣA/ΣΣR გეიფენებს, რომ ტბაში პლანქტონის პირველადი პროდუქცია გროვდება და ირთვება ღამის წარმოქმნის პროცესში. თევზის რეწვა ტბაში არ უნდა აჭარბებდეს 12-18 კგ/ჰა წელიწადში. კარწახის ტბა ევტროფულ წყალსატევებს მიეკუთვნება.

CORRELATION BETWEEN BLOOD RH SYSTEMS GROUP ANTIGENS WITH PULMONARY TUBERCULOSIS

NAGERVADZE M., DIASAMIDZE A., AKHVLEDIANI L., GOGITIDZE T., DUMBADZE G.,
KHALVASHI N.

Immunogenetic laboratory, Sh Rustaveli Batumi State University

(Received May 16, 2005)

Abstract

Correlation between Rh group antigens (D,C,c,E,e) with tuberculosis was studied. To express blood group antigens immunoserological methods, carried out on 50 patients with pulmonary tuberculosis (PT) and 50 healthy controls, were used. In patients high frequency of C and D antigens was revealed and consequently in Rh positive individuals risk of this disease is high. Individuals with CC and Cc genotypes are exclusively predispositioned to PT. Characteristic to PT De and CDEc phenotype groups were not displayed in control group. It was found that in the group of patients with PT 6.25% had De phenotype with not expressed C and c genes and 2.08% had CDEc phenotype.

Key words: immunoserological method, blood group, antigen, genotype.

Introduction

Blood group systems immunogenetic polymorphism is widely used in ethnic anthropology, epidemiology, forensic medicine, transplantology, as well as in studies of human genetics and population biology [Systems of immunogenetical polymorphism, 2000].

Correlation between erythrocytic group antigens and infectious and noninfectious diseases such as: paratyphoid, measles, scarlet fever, coli-infection, diabetes, cancer, cirrhosis, pneumonia, bronchitis, tuberculosis was established [Platonova, 1999; Perea-Mejia, et al., 2000; Alberti et al., 1999; Helfand et al., 2005; Honore et al., 2001].

In scientific literature there are poor data about correlation of pulmonary tuberculosis with Rh system antigens [Vaskum , 1975; Havlir et al., 1991; Chiang et al., 1997;]. About 1/3 of humans is infected with *M.tuberculosis*. Among them only 10% is clinically revealed, and majority is bacteria carrier.

Marginal check and studies of the reasons of PT due to high frequency of this disease dissemination in Georgia is an urgent problem.

Materials and Methods

Studies have been carried out on blood erythrocytes of 50 individuals with PT and 50 healthy controls. Experiments were carried out on blood samples of the patients from Batumi Pneumonic-Pulmonary Clinic. To reveal of Rh systems antigens immunoserological method was used [Guide-book on blood transfusion and blood substitutes, 1982].

Anti - D, C, c, E, e monoclonal antibodies were used (Hemostandard, Russia).

Study of Rh system was carried out differentiated on D, C, c, E, e antigens. Rh phenotypes, homo- and heterozygote states of C and E genes were revealed.

Results and Discussion

Data of research showed correlation of tuberculosis with Rh systems antigens.

In diseased populations while studying separate antigens of Rh-Hr system high frequency of D and C antigens was noticed (Fig.1). Correspondingly in patients with tuberculosis frequency of Rh-positive factor carriers is higher than in control (Fig.2).

While studying homo- and heterozygote states of C and E genes it was shown that in patient group dominant homozygote CC and heterozygote Cc variants of C gene were presented in considerable high frequency, but the frequency of recessive homozygote cc is significantly lower compared to control group (Fig.3).

At the studies of Rh phenotypes two interesting phenomena occurred. Namely, in PT patients De - phenotype revealed in 6.25%, which in control group was not stated at all. As usually any genotype has C and c genes and in mentioned phenotype co-dominant alleles (Cc) of RHC locus are not revealed, we should considered that suppression of mentioned locus is caused by some reason which stimulate genetical predisposition towards this disease.

Besides, in diseased population CDEe phenotype was revealed in 2.08% (Fig.4) where simultaneously three dominant alleles C, D and E occur. These are crossed genes and they inherited together. But as a result of crossingover this regularity is violated and other phenotype groups appear. Absence of mentioned phenotype in control group should be explained by consideration that carriers of this phenotype are more submitted to elimination and are obliterated from population, i.e. it is characterized with negative selection.

Thus our studies showed correlation of D and C antigens of Rh systems with tuberculosis which have practical significance. Based on our research it is possible to stand out high risk group of disease and carry out preventive arrangements on the individuals of this group.

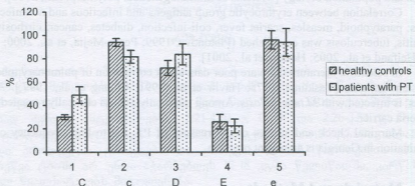


Fig. 1. Frequency of dissemination of Rh-Hh antigens

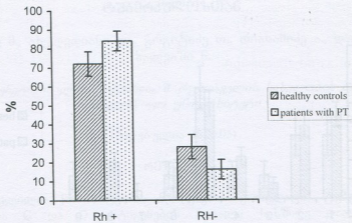


Fig. 2. Frequency of Rh-positive and Rh-negative factors carriers

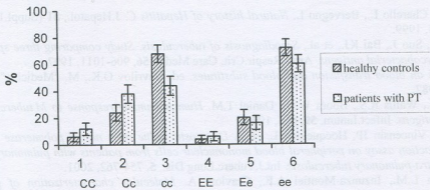


Fig. 3. Homo- and heterozygote states of C and E genes

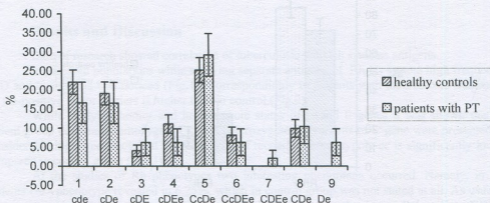


Fig. 4. Frequency of dissemination of Rh phenotype

References:

- Alberti A., Cherello L., Bervegnu L. *Natural history of Hepatitis C*. J.Hepatol., **31** (Suppl.1), 17-24, 1999.
- Chiang IH., Suo J., Bai KJ., et al., *Serodiagnosis of tuberculosis. Study comparing three specific mycobacterial antigens*. Am.J.Respir.Crit. Care Med., **156**, 906-1011, 1997.
- Guide-book on blood transfusion and blood substitutes*. ed. Gavrilov O.K., M. "Medicina", 304, 1982.
- Havliř D.V., Wallis R.S., Boom W.H., Daniel T.M. *Human immune response to M.tuberculosis antigens*. Infect.Innum. **59**, 665, 1991.
- Honore S., Vincensin JP, Hocqueloux L. et al. *Diagnostic value on a nested polymerase chain reaction assay on peripheral blood mononuclear cells from patients with pulmonary and extra-pulmonary tuberculosis*. Int.J.Tuberc.Lung Dis., **5**, 754-762, 2001.
- Perea-Mejia L.M., Inzunza-Montiel A.E., Cravioto A. *Molecular characterization of group A.Streptococcus strains isolated during a scarlet fever outbreak*. J.Clin. Microbiol., **40**, 11, 278-280, 2000.
- Platonova I.L. *Metabolic changes in blood in pulmonary tuberculosis patients from various blood groups*. Ukr. Biochim. Zh., **71**, 5, 94-96, 1999.
- Systems of immunogenetical polymorphism*. in: "Gene pool and gene geography of population". ed. Richkov Iu. Saint-Petersburg, "Nauka", **1**, 611, 2000.
- Vaskum K. *The ABO and rhesus blood groups in patients with pulmonary tuberculosis*. Tubercul., **56**, 4, 329-34, 1975.



სისხლის Rh სისტემის ჯგუფური ანტიგენების კორელაცია ტუბერკულოზთან

ნაგერვაძე მ., ახელედიანი ლ., გოგიტიძე თ., დიასამიძე ა., დუმბაძე გ., ხალვაში ნ.

იმუნოგენეტიკის ლაბორატორია, შ. რუსთაველის სახელობის ბათუმის სახელმწიფო უნივერსიტეტი

(მიღებულია 16.05.05)

რეზიუმე

შესწავლილია Rh სისტემის ანტიგენების კორელაცია ტუბერკულოზთან. გამოვლენილია C და D ანტიგენების მაღალი სიხშირე დაავადებულებში, შესაბამისად მაღალია დაავადების გამოვლენის რისკი Rh დადებით პირებში. დადგენილია, რომ CC და Cc გენოტიპების მატარებლები განსაკუთრებით ექვემდებარებიან ტუბერკულოზისადმი წინასწარგანწყობას. დაავადებულთათვის დამახასიათებელი De და CDEe ფენოტიპური ჯგუფები საერთოდ არ გამოვლინდა გამოკვლეულ ჯანსაღ პოპულაციაში.

STUDY OF AVIAN PATHOGENIC *ESCHERICHIA COLI*

ANTIA I., NATROSHVILI N., BALANCHIVADZE M., BATIASHVILI E.

Department of Microbiology and Virology, Zootechnical Veterinary University of Georgia

(Received May 12, 2005)

Abstract

Main properties of pathogenic *Escherichia coli* from poultry was studied. 13 strains from dead chicken were isolated. It was proved that all isolates are gram negative, 6 of them are motive. They have evident biochemical activity fermenting glucose, maltose, galactose, lactose and mannitol. Some of them digest sucrose. 7 of investigated cultures are hemolytic that proves their pathogenicity. All strains isolated from chicken were agglutinated by *E.coli* O-sera.

Introduction

Problems attributed to coliform infections are often caused by strains of the *Escherichia coli* organisms. There is marked variation in severity. Problems range from severe acute infections with sudden and high mortality to mild infections of a chronic nature with low morbidity and mortality. Infections may result in a respiratory disease from air sac infection, a septicemic (blood) disease from generalized infection, an enteritis from intestinal infection or a combination of any or all of these conditions. The disease may result from a coliform infection alone as in primary infection or in combination with other disease agents as a complicating or secondary infection. All age can be affected by colibacillus, but young growing birds are more vulnerable [Allan et al., 1993; Dho-Moulin & Fairbrother, 1999; Caprioli et al., 2005; Zhao et al., 2005].

There are many different strains or serological types within the group of *E.coli* bacteria. A marked variation exists between different strains by their degrees of pathogenicity. The main goals of our research are to secrete isolates of pathogenic *E.coli* from the poultry diseased with colibacillosis, morphological-cultural and biochemical studies and serological typing.

Materials and Methods

To secrete isolates of *E.coli* we studied internal - heart, liver, spleen, ovary and also intestine contents of 18 dead chickens suspected of colibacillosis. 13 strains of *E.coli* were isolated and studied. To investigate morphological-tinctorial features smears were prepared from internal and 18-20 hours cultures cultivated on nutrient media (meat-peptone broth and meat-peptone agar). They were dyed by Gram-stain method. To establish motility of cultures crushed drop and growth peculiarities in semisolid agar (0.3-0.4%) were used. At the same time on Endo and Lewin media growth peculiarities and relief of colonies were studied [Methods of General Bacteriology, 1983].

Results and Discussions

Studied isolates presented end-round gram-negative bacillus of 0.5-2 μm length and 0.3-0.6 μm width. 6 isolates prove to be motive (Table 1). *E.coli* isolates in Petri dish on agar surface form grayish bulging colonies of S type. Their diameter is 2-3 mm. Isolates uniformly stir up the broth with forming of slight precipitation. On Endo agar they give reddish metal shining colonies, and on Lewin nutrient medium – bluish colonies.

To determine biochemical activity of *E.coli* isolates carbohydrates fermentation, ability of production of ammonia, indole, hydrogen sulphide and hemolytic property were studied. It was shown that 18-20 hours incubated cultures of *E.coli* hydrolyze glucose, maltose, galactose, lactose and mannitol producing acids and gases. Only some isolates have capability of sucrose fermentation.

Cultures of *E.coli* produce ammonia, but not indole and hydrogen sulphide; they have evident catalytic activity. 7 isolates challenge erythrocytolysis that confirm their hemolytic property and pathogenicity (Table 2).

For serological typing isolates were studied in agaric (lamellate) agglutination reaction. All 13 isolates are O-sera positive.

Table 1. Morphological-cultural characteristics of *E.coli*

Isolated strains	dyeing by Gram method	motility
<i>E.coli-1</i>	-	-
<i>E.coli-2</i>	-	-
<i>E.coli-3</i>	-	+
<i>E.coli-4</i>	-	+
<i>E.coli-5</i>	-	-
<i>E.coli-6</i>	-	-
<i>E.coli-7</i>	-	-
<i>E.coli-8</i>	-	-
<i>E.coli-9</i>	-	+
<i>E.coli-10</i>	-	+
<i>E.coli-11</i>	-	+
<i>E.coli-12</i>	-	-
<i>E.coli-13</i>	-	+

Table 2. Biochemical characteristics of *E.coli* isolates

Isolated strains	H ₂ S	NH ₃	Indole	Catalytic activity	Hemolysis
<i>E.coli-1</i>	-	+	-	+	-
<i>E.coli-2</i>	-	+	-	+	-
<i>E.coli-3</i>	-	+	-	+	+
<i>E.coli-4</i>	-	+	-	+	+
<i>E.coli-5</i>	-	+	-	+	-
<i>E.coli-6</i>	-	+	-	+	-
<i>E.coli-7</i>	-	+	-	+	+
<i>E.coli-8</i>	-	+	-	+	+
<i>E.coli-9</i>	-	+	-	+	+
<i>E.coli-10</i>	-	+	-	+	+
<i>E.coli-11</i>	-	+	-	+	-
<i>E.coli-12</i>	-	+	-	+	-
<i>E.coli-13</i>	-	+	-	+	+

So, we can conclude that isolated from the poultry *E.coli* strains are pathogenic. Isolates reveal slight differences in morphological and cultural features. Biochemical activity of colibacillus isolates by parameter of sucrose fermentation and hemolytic properties differ. All strains isolated from chicken were agglutinated by *E.coli* O-sera.

References:

- Allan B.J., van den Hurk J.V., Potter A.A. *Characterization of Escherichia coli isolates from cases of avian colibacillosis*. Can. J. Vet. Res., **57**, 3, 146-51, 1993.
- Caprioli A., Morabito S., Brugereb H., Oswald E. *Enterohaemorrhagic Escherichia coli: emerging issues on virulence and modes of transmission*. Vet. Res., **36**, 3, 289-311, 2005.
- Dho-Moulin M., Fairbrother J.M. *Avian pathogenic Escherichia coli (APEC)*. Vet. Res., **30**, 2-3, 299-316.
- Methods of General Bacteriology*. ed. Ph.Gerhardt, M., "Mir", 1983.
- Zhao S., Maurer J., Hubert S. De Villena JF., et al. *Antimicrobial susceptibility and molecular characterization of avian pathogenic Escherichia coli isolates*. Vet. Microbiol., **107**, 3-4, 215-24, 2005.

ფრინველებიდან გამოყოფილი პათოგენური *E.coli*-ს შესწავლა

ანთია ი., ნატროშვილი ნ., ბალანჩივაძე მ., ბათიაშვილი ე., ნათიძე მ.

მიკრობიოლოგია-ვირუსოლოგიის კათედრა, საქართველოს ზოოტექნიკურ-სავეტერინარო უნივერსიტეტი

(მიღებულია 12.05.2005)

რეზიუმე

შესწავლილია ფრინველებიდან გამოყოფილი *E.coli*-ს იზოლატების ძირითადი თვისებები. კოლიბაქტერიოზით დაავადებული ქათმებიდან გამოყოფილია 13 იზოლატი. დადგენილია, რომ ყველა იზოლატი გრამუარყოფითია, მათგან 6 მოძრავია. ნაწლავის ჩხირის იზოლატებს გააჩნიათ მკვეთრად გამოხატული ბიოქიმიური აქტიობა. ისინი შლიან გლუკოზას, მალტოზას, გალაქტოზას, ლაქტოზას, და მანიტს, საქაროზას კი - მხოლოდ ზოგიერთი იზოლატი. შესწავლილი კულტურებიდან 7-ს გააჩნია ჰემოლიზური თვისება, რაც მიუთითებს მათ პათოგენურობაზე. ცამეტევე იზოლატმა *E.coli*-ს O შრატთან მოგვცა დადებითი რეაქცია.

ISOLATION AND IDENTIFICATION OF MICROSCOPIC FUNGI FROM SALTY SOILS

LASKHISHVILI M., KUTATELADZE L., ZAKHARIAHVILI N., GLONTI N.,
KHOKHASHVILI I., JOBAVA M.

S. Durmishidze Institute of Biochemistry and Biotechnology of the Georgian Academy of Sciences.
David Agmashenebeli Alley 10km, 0059 Tbilisi, Georgia

(Received June 14, 2005)

Abstract

52 different species of microscopic fungi have been isolated from salty soils of lower Kartli plane - surroundings of Kumisi and Krasnogorski salty lakes, for the purpose of searching halophytes. 7 genera, belonging to classes: Ascomycetes, Zygomycetes and Deiteromycetes were identified. The dominating genera of microscopic fungi, characteristic for surroundings of these two geographically distant salty lakes, and the frequency of their distribution were determined. The micro flora of Kumisi and Krasnogorski soils has been compared. Dominants for soils of both regions were genera: *Aspergillus* and *Penicillium*

Key words: pure culture, halophytes, *Aspergillus*, *Penicillium*

Introduction

Extremophilic microorganisms represent the special group of the earth inhabitants. They are found under ground in the depth of several hundred meters, in glaciers, in the depth of the world ocean, and in the waters of the nuclear reactors, etc. [Gavicchioli R. and Thomas T., 2003].

Mostly interesting among the extremophils are microorganisms growing at the areas with high concentrations of salt – halophils, which are known as “the inhabitants of the extreme bounds of life”.

Today halophils are considered as potential producers of biologically active substances [Vershinin A., 1999]. Halophils are rich of betoins and ectoins – substances which protect biological molecules and living cells from the extreme influences. Due to the strong oxidative system the preparations obtained from these microorganisms are successfully used in medicine and cosmetics [Vladimirov I., 1998]. The preparation “Baxin” – received from the liophilic biomass of halobacteria, is recommended as the nutrient addition. The halophilic collections of the world are presented mainly by the bacteria. Searching of halophils among the microorganisms and other groups, in particular, among the microscopic fungi, is of great interest. From this point of view, attention should be paid to the salty soils of Georgia, where all conditions for halophils’ developing are presented. At the same time the micro flora of these saline lands is practically not studied.

Materials and Methods

For searching halophils, among the 48 soil zones of Georgia the dry valley of lower Kartli plane – Soghanlugi valley was selected.

It is known that the salinity of rocks, constructing the Iaghluji Mountain, is responsible for salination of Soghanlugi valley soils. There are many salty lakes and hills in this valley [Sabashvili G., 1965]. Soil samples were picked up from geographically distant territories of Soghanlugi valley - Kumisi and Krasnogorski lakes, in particular, from lake waters, salty lakeside, from the rhizosphere of semi desert plants and lake algae, salty hills etc. It was supposed that among the microflora of such kind of saline lands the halophilic micromycetes may exist, as the extremophils of this group are able to grow at high concentrations of salt.

Micromycetes were removed at different nutrient media. Most abundant and diverse micro flora was characteristic for the universal (with agar) nutrient medium, where all isolated microscopic fungi developed well.

10 g of averaged soil samples were picked from two geographically distant territories of one of the salty valleys of lower Kartli plane - Soghanlugi valley [Fomin G., et al., 2001].

To obtain the homogenous suspension containing microorganisms as separately and freely moving cells, the samples were previously treated, using the method of soil aggregates dispersing [Zvyagintsev, 1980]. Treated material was sowed on a sterile Petri glasses by Waksman's method of soil dilution [Waksman, 1916] and direct sowing method [Warcup, 1950]. For this purpose suspensions of following dilution were prepared: 10^1 , 10^2 , 10^3 and 10^4 .

The microscopic fungi were isolated on the following nutrient media: (g/l) 1) The universal medium - 0.5l 7°B wort, 0.5l tap water, 20g agar, pH - 5.5-6.0. 2) Chapek's acidified medium (for inhibition of bacteria) - NaNO_3 -9.1, KH_2PO_4 -1.0, MgSO_4 -0.5, KCl -0.5, FeSO_4 -0.02, glucose-40.0, agar-20.0, pH-3.5 - 4.2. 3) Selective nutrient medium - NaNO_3 -3.2, KH_2PO_4 -2.0, MgSO_4 -0.5, yeast extract-10.0, microcrystal cellulose-1%, agar-20.0, pH-5.5 - 6.0. 4) Chapek's modified medium - NaNO_3 -9.1, KH_2PO_4 -1.0, MgSO_4 -0.5, KCl -0.5, FeSO_4 -0.02, starch-20.0, agar-20.0, pH-5.5 - 6.0. 5) Chapek-Dox's nutrient medium - NaNO_3 -2.0, KH_2PO_4 -1.0, $\text{MgSO}_4 \times 7\text{H}_2\text{O}$ -0.5, KCl -0.5, $\text{FeSO}_4 \times \text{H}_2\text{O}$ -0.02, sucrose-30.0, agar-20.0. Nutrient media were sterilized under 0.6 atm for 45 min, in autoclaves.

Incubation of the microscopic fungi, obtained from the soil samples was performed at 30°C in thermostat. Plate cultures were observed on the 3rd, 5th, 7th and 10th days. On the 10th day the separate colonies of Petri cultures were sowed once more and cultivated at 30°C for 10 days. If the pure culture developed after this procedure, the piece of mycelium or a small portion of spores from plate culture were placed in the test-tube containing sterilized universal medium. Test-tubes were placed into the thermostat at 30°C for 10 days.

Using the soil dilution method for purifying the microscopic fungi, the frequency of detection of particular species was determined by the ratio: $FD = \frac{IS}{TS}$ (FD - frequency of species detection, IS - amount of the investigated sample, TS - total amount of the sample).

While identification, using the light microscopy, morphological and cultural peculiarities of the cultures were taken into account. Also different guides were used [Pidoplichko, 1967; Bilayi, 1988; Malloch, 1981]. At first plate cultures were generally observed, at a small magnification, after - the preparations were made. In some cases the dry optical system microscopy was used.

Results and discussion

52 different species of microscopic fungi were released and identified till genus. Among them 32 species were from surroundings of Kumisi and 20 of Krasnogorski salty lakes. 7 genera were identified, belonging to classes: Ascomycetes, Zygomycetes, and Deiteromycetes (Table 1).

Table 1 Microscopic fungi distributed in Kumisi and Krasnogorski saline soils and lakes

Genera of Micromycetes	Frequency of distribution (%)	
	Kumisi	Krasnogorski
<i>Aspergillus</i>	25	45
<i>chaetomium</i>	18.75	-
<i>Penicillium</i>	31.25	20
<i>Fusarium</i>	12.50	-
<i>Trichoderma</i>	-	15
<i>Mucor</i>	6.25	10
<i>Rizopus</i>	6.25	10

Table 1 presents the genera of microscopic fungi and the frequency of their detection. It is clear that mainly three classes of micromycetes: Ascomycetes, Zygomycetes, and Deiteromycetes are disseminated all over Soghanlugi valley.

Soils of both regions possess the common dominants – genera *Aspergillus* and *Penicillium*. Genus *Trichoderma* is characteristic only for Krasnogorski saline lands, while genera *Chaetomium* and *Fusarium* were identified only in samples of Kumisi saline lands (Table 1).

Table 2. Microscopic fungi distributed in surrounding soils of Kumisi salty lake

Micromycete	Class	Frequency of distribution (%)					
		Place of sampling					
		Surface of salty soil	Salty lake-side	Semi desert's rhizosphere	Soil of Kumisi mountain	Water of Kumisi lake (uder the algae)	Saline hill
1. <i>Aspergillus</i>	Ascomy-cetes	53.0	50	16.6	-	100	-
2. <i>Chaetomium</i>		44.5	-	-	50.0	-	-
3. <i>Penicillium</i>		2.5	5	50.0	25.4	-	100
4. <i>Fusarium</i>	Deiteromy-cetes	-	-	33.4	-	-	-
5. <i>Mucor</i>	Zygomy-cetes	-	-	-	25	-	-
6. <i>Rizopus</i>		-	45	-	-	-	-

Differences were revealed not only between Kumisi and Krasniogirski microflora, but properly between the soil samples of particular habitat (Tables 2 and 3). In Kumisi soils dominating genera were *Aspergillus* and *Penicillium* (Table 2). But genus *Aspergillus* was not found in soils of Kumisi Mountain and saline hill, while genus *Penicillium* was absent only in Kumisi Lake.

Homogenous were Kumisi lake water, with absolutely dominating genus *Aspergillus*, and saline hill soils, only with *Penicillium* (Table 2). Zygomycetes class genera were found both in saline soils of lake surroundings (*Rizopus*), and Kumisi mountain soils (*Mucor*). While representatives of Deiteromycetes (genus *Fusarium*) were identified only for rhizosphere of semiarid plants (Table 2).

Table 3. Microscopic fungi distributed in surrounding soils of Krasnogorski salty lake

Micromycete	Class	Frequency of distribution (%)				
		Place of sampling				
		Silt of Krasnogorski lake (under the algae)	Salty lake-side	Soil under the salty lake	Soil between two salty lakes	Water of Krasnogorski lake (under the algae)
1. <i>Aspergillus</i> 2. <i>Penicillium</i>	Ascomy- cetes	40 20	25 25	100 -	- 25	75 25
3. <i>Trichoderma</i>	Deiteromy- cetes	-	-	-	75	-
5. <i>Mucor</i> 6. <i>Rizopus</i>	Zygomycetes	- 40	50 -	- -	- -	- -

In Table 3 Micromycetes genera from Krasnogorski salty lake and soils, and the frequency of their detection are presented. Genera *Aspergillus* and *Penicillium* are dominating here also. They present almost in all saline soils of Krasnogorski. In Krasnogorski Lake dominating was *Aspergillus*, but genus *Penicillium* is presented too (Table 3).

It must be mentioned that genus *Fusarium* (from Deiteromycetes class), which was characteristic for Kumisi soils, was absent in Krasnogorski soils. But this genus was replaced by another genus *Trichoderma* of the same class. This genus was distributed only in saline soils situated between the salty lakes. Species of genus *Rizopus* were found only in silt of Krasnogorski Lake, while genus *Mucor* - in salty lakeside. (Table3).

Thus, experimentally identified microscopic fungi belong to classes: Ascomycetes, Zygomycetes and Deiteromycetes. Obtained results coincide with the literature data that genera *Aspergillus*, *Penicillium*, *Mucor*, *Fusarium*, *Trichoderma* and *Rizopus* are mainly distributed in soil.

References:

- Bilay V. I., Koval E. Z. *Aspergillii*. Kiev, "Naukova dumka", 1988. (Russian)
- Fomin G.S., Fomin A.G. *The soil-control, quality and ecological security according to the international standards*. Moscow, 2001 (Russian).
- Gavicchioli R., and Thomas T. *Extremophiles In: The Encyclopedia of Microbiology*. Acad. Press, San Diego. 2003.
- Litvinov A. P. *Guide of the soil microscopic fungi*. Leningrad, 1967 (Russian).
- Malloch D., Moulds T. *Micromycetes, their isolation, cultivation and identification*. University of Toronto-press, Toronto-Buffalo-London, 1981.
- Pidoplichko N. M., Milko A. A. *Atlas of mucoral fungi*. Kiev, 1971 (Russian).
- Sabashvili G. *Soils of Georgia*. Tbilisi, "Metsniereba", 1965 (Georgian).
- Vershinin A. *Biological functions of carotenoids – diversity and evolution*. Biofactors. **10**, 2-3, 99-104, 1999.
- Vladimirov I. A. *Free radicals and antioxidants*. Proceedings of Russian Med. Acad., **7**, 43-51, 1998 (Russian).

სოღანღუღის ველის მარლიანი ნიადაგებიდან მიკროსკოპული
სოკოების გამოყოფა და იდენტიფიკაცია

ლახიშივილი მ., ქუთათელაძე ლ., ზაქარიაშვილი ნ., დლონტი ნ.,
ხოხაშვილი ი., ჯობავა მ.

საქ. მეცნიერებათა აკადემიის ს. ღურმიშვილის სახ. ბიოქიმიისა და
ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 14.06.2005)

რეზიუმე

პალოფილების ძიების მიზნით, ქვემო ქართლის ვაკის ზოგიერთი მარლიანი ნიადაგიდან – კუმისისა და კრასნოგორსკის მლაშე ტბების მიმდებარე ტერიტორიებიდან, გამოყოფილია 52 განსხვავებული სახეობის მიკროსკოპული სოკო. იდენტიფიცირებულია 7 გვარი, რომელიც მიეკუთვნება Ascomycetes, Zygomycetes-სა და eotermycetes კლასებს. დადგენილია სოღანღუღის ველის ამ ორი ერთმანეთისგან გეოგრაფიულად დაშორებული მარლიანი ტბის მიმდებარე ნიადაგებისთვის დამახასიათებელი მიკროსკოპული სოკოების დომინანტი გვარები და განსაზღვრულია მათი შეხვედრის სიხშირე. შედარებულია კუმისისა და კრასნოგორსკის ნიადაგების მიკროფლორა. ორივე რეგიონის ნიადაგების დომინანტებია *Aspergillus*-ის და *Penicillium*-ის გვარები.

PECULIARITIES OF *NOCARDIA DASSONVILEI* POLYSACCHARIDE METABOLISM

LOMTATIDZE Z., KOTIA N.

Department of Microbiology and Virology, Iv. Javakhishvili Tbilisi State University

(Received June 6, 2005)

Abstract

Peculiarities of polysaccharide metabolism of *Nocardia dassonvillei* according to the phases of culture growth have been studied. It has been established that the synthesis of polysaccharide begins in logarithmic phase of actinomycete growth, reaches its maximum in the stationary phase. In the phase of dying polysaccharide synthesis reduced. Monomeric content of general cellular polysaccharides and cell wall polysaccharides is identical and differs by quantitative correlation of monomers.

Key words: neutral polysaccharide, general cellular polysaccharide, cell wall polysaccharide, exopolysaccharide

Introduction

Polysaccharides appear to be active components of actinomycete cells playing an important role in vital activity of microorganisms. Topologically in the cells of microorganisms intracellular and extracellular polysaccharides are distinguished. Functionally intracellular polysaccharides appear to be modifiers, as well as reserve nutritive matter and structural glycoconjugates. Cell wall polysaccharides have structural and structural-metabolic functions. At the process of hyperproduction extracellular polysaccharides have a defensive function [Elinov, 1989]. Besides, polysaccharides appear to be stimulants of microorganism growth. In the population of microbes actinomycetes are one of the interesting group. On the ultrastructural and chemical basis, characteristic for procaryotes, they realize micellular organization in more perfect form which is characteristic for eucaryotes, fungi [Gauze et al., 1983; Gusev, Mineeva, 2003]. The interest to actinomycetes was arisen for their ability to synthesize a lot of biologically active substances.

The goal of our investigation was to study peculiarities of polysaccharide synthesis in the dynamics of culture growth.

Material and methods

Nocardia dassonvillei from the collection of microorganisms of Microbiology Chair of Institute of Botany, Georgian Academy of Sciences was used for the investigation.

Actinomycetes were grown on Krasilnikov synthetic medium: KNO_3 – 1 g, CaCO_3 – 1 g, MgSO_4 – 0,5 g, NaCl – 0,5 g, K_2HPO_4 – 0,5 g, FeSO_4 – a trace, agar – 20 g, water – 1 l.

Polysaccharides were obtained using the method of Elinov [Pract. work biochem, 2001]. Qualitative analysis of polysaccharides was done using thin-layer chromatography. The quantitative content of polysaccharide monosaccharides was determined using densitometry method [Gerhardt, 1984].

The results of our experiments have shown (Fig.1) that dynamics of *Nocardia dassonvillei* growth is determined by the following phases: logarithmic phase – 0-24 h, exponential phase – 24-96 h, stationary phase – 96-120 h, phase of dying – 120-168 h.

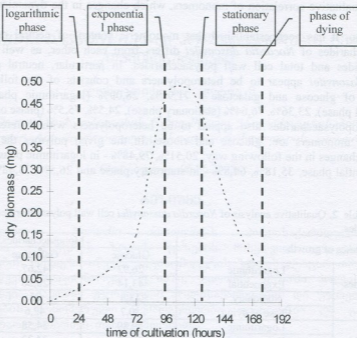


Fig. 1. Dynamics of *Nocardia dassonvillei* growth

Under these conditions of growth the intensity of polysaccharide synthesis has been studied. The analysis of experimental data has shown that polysaccharide synthesis takes place with different intensity during the whole cycle of actinomycete life.

The intensity of polysaccharides, particularly cell wall polysaccharides, neutral polysaccharides, total cell polysaccharide synthesis is less in logarithmic phase of culture growth, in exponential phase the intensity increases and reaches the maximum in stationary phase. A maximal decrease of synthesis of polysaccharide intensity is noted in the phase of dying (Table 1).

Table 1. Quantity of *Nocardia dassonvillei* polysaccharides according to growth phases

Phases of growth	Quantity of polysaccharides in 1 g of dry biomass (%)			
	wall polysaccharides	cell polysaccharides	neutral polysaccharides	exopolysaccharides
Logarithmic	4,7	14,5	5,1	5,5
Exponential	9,0	17,0	7,0	15,0
Stationary	17,9	20,0	11,0	20,0
Phase of dying	13,0	9,9	4,5	7,0

At the same time *Nocardia dasonvillei* is characterized by a high intensity of total cell polysaccharides and exopolysaccharides. In particular, total cell polysaccharides make 9-20% of dry biomass, exopolysaccharides - 5-20%. The synthesis of cell wall polysaccharides and neutral polysaccharides is relatively low and varies within the limits of 9-13% and 5-11%, correspondingly.

During the study of qualitative aspect of polysaccharides it has been established that monosaccharide content of cell wall polysaccharides and total cell polysaccharides is identical and differs by qualitative correlation of monomers, which changes in the dynamics of culture growth (Table 2).

Also, it has been established that monomeric content of neutral polysaccharides and exopolysaccharides of *Nocardia dasonvillei* differs from each other, as well as from cell wall polysaccharides and total cell wall polysaccharides. In particular, neutral polysaccharides of *Nocardia dasonvillei* appear to be heteropolymers and consists of the following quantitative correlation of glucose and galactose - 71,91%, 28,09% (logarithmic phase), 3,2%, 66,8% (exponential phase), 23,36%, 76,64% (stationary phase), 24,5%, 75,5% (phase of dying).

Exopolysaccharides also appear to be heteropolymers with different monosaccharide content. Its monomers are: glucose and ribose. In the given polymer the quantity of these monomers changes in the following way: 20,51%, 79,48% - in logarithmic phase, 75,32%, 29,43% - in exponential phase, 35,18%, 64,8% - in stationary phase and 26,5%, 73,48% - in the phase of dying.

Table 2. Qualitative analysis of *Nocardia dasonvillei* cell wall polysaccharides and total cell polysaccharides

Phases of growth		Monosaccharides, %		
		Glucose	Galactose	Ribose
wall polysaccharides	Logarithmic	26,92	42,67	30,41
	Exponential	43,14	20,26	36,6
	Stationary	43,3	25	31,7
	Phase of dying	30,7	42,6	26,6
cell polysaccharide	Logarithmic	39,89	34,58	25,53
	Exponential	43,64	34,32	22,03
	Stationary	37,01	41,82	21,15
	Phase of dying	47,05	26,05	26,89

So, the study of peculiarities of polysaccharide metabolism of *Nocardia dasonvillei* has shown that in spite of functional and topological difference of these biopolymers, they have a characteristic general scheme of the synthesis. The synthesis of polysaccharides in the process of actinomycete growth begins in logarithmic phase, reaches the maximum in stationary phase and is minimal in the phase of dying. At the same time, change in the quantitative correlation of monomers is observed in monosaccharide content of polysaccharides according to the phases of growth. However, under given conditions monosaccharide content of polysaccharides is not changed.

References:

- Gauze G.F., Preobrazhenskaia R.P., Sveshnikova M.A., Terekhova L.P., Maximova R.S. *Determinant of actinomycetes*. Moskva, Nauka, 1983 (in Russian).
 Gerkhardt F. *Methods of general bacteriology*. Moskva, Mir, 2, 1984 (in Russian).

Gusev M.V., Mineeva L.A. *Microbiology*. M., ACADEMA, p. 87, 2003 (in Russian).
Elinov N.P. *Chemical microbiology*. M., "Visshaia shkola", p. 38, 1989 (in Russian).
Practical work of biochemistry. Ed. N.P. Elinov, Publishing House of Moscow State University, 2001 (in Russian).

NOCARDIA DASSONVILEI-ის პოლისაქარიდული ცვლის თავისებურობა

ლომთათიძე ზ., კოტია ნ.

ივ. ჯავახიშვილის სახ. თბილისის სახელმწიფო უნივერსიტეტის ბიოლოგიის
ფაკულტეტის მიკრობიოლოგიისა და ვირუსოლოგიის კათედრა

(მიღებულია 06.06.2005)

რეზიუმე

შესწავლილია *Nocardia dassonvillei*-ს პოლისაქარიდული ცვლის
თავისებურობანი კულტურის ზრდის ფაზების მიხედვით. დადგენილია, რომ
პოლისაქარიდების სინთეზი იწყება აქტინომიცეტის ზრდის ლოგარითმულ ფაზაში,
მაქსიმუმს აღწევს სტაციონარულ ფაზაში, კვდომის ფაზაში პოლისაქარიდების
სინთეზი ნელდება. საერთო უჯრედული პოლისაქარიდების და უჯრედის კედლის
პოლისაქარიდების მონომერული შემადგენლობა იდენტურია და განსხვავდება
მონომერების რაოდენობრივი თანაფარდობით.

CLIMATIC AND HUMAN INFLUENCES ON VEGETATION DYNAMICS AROUND TBILISI OVER THE PAST 6000 YEARS

CONNOR S.E. ^{1*}, KVAVADZE E.V. ^{2*}

¹*Geography and Environmental Studies, University of Melbourne, VIC-3010, Australia*

²*L. Davitashvili Institute of Palaeobiology, Georgian Academy of Sciences*

(Received June 7, 2005)

Abstract

This paper presents new data on the vegetation history of the Tbilisi environs, based on analysis of pollen, charcoal and fungal remains preserved in a radiocarbon-dated sediment core. The results show a remarkable dynamism in vegetation structure, oscillating from scrub to forest to steppe and back again. Two statistical techniques (DCA and TWINSpan) are employed to classify the data into ecological groups and to differentiate climatic and human impacts on vegetation development. Although a climatic influence is apparent, human impact has been the dominant factor controlling vegetation change and forest cover on the hills around Tbilisi through the past 6000 years.

Key words: vegetation history, pollen analysis, charcoal, Holocene, Caucasus.

Introduction

Tbilisi is nowadays largely surrounded by cultural landscapes. Like most major cities in the Near East, Tbilisi's vegetation has been strongly modified by past human activities. Yet the Georgian capital has been more fortunate than most, retaining considerable tracts of forest. The city itself is located on the very border of forest and steppe, and thus its vegetation reflects not only human influences, but also climatic ones. Because changes in the position of the lower tree-line are traceable using pollen analysis [Davies and Fall, 2001; Connor et al., 2004], Tbilisi area provides an opportunity to examine these changes in the light of past climatic variations and a long, well-documented, socio-economic history.

It is widely believed that the city was surrounded entirely by forests up until the 19th century. Various authors have concluded that these original forests were of beech [Sosnovskii, 1915] or oak [Ketskhoveli, 1959], but available historical evidence is fragmentary and often ambiguous [Juansher, 1981; Tournefort, 1718; Vakhushti, 1983]. In this paper we adopt a palaeoecological approach to answer the following questions:

1. What was the past character of Tbilisi's vegetation?
2. How did it change through time?
3. Were these changes related to temperature, precipitation or human impact?

* Corresponding authors: 1. s.connor@pgrad.unimelb.edu.au; 2. eliso@paleobi.acnet.ge

Study area

The mountains to Tbilisi's west as far as the Mtkvari River are composed of Upper Eocene alkaline sedimentary rocks that have been warped and folded to create a rugged topography of sharp peaks and steep ravines. Preferential weathering of anticlines, the occurrence of landslips, and suffusion of gypseous strata within the rock units have led to the formation of a number of lake basins (Figure 1) within this much dissected landscape [Dzhanelidze, 1980].



Fig. 1. Map of the study area, showing lakes, forest areas and mountains above 1000 metres in elevation (Source: 1:200000 GUGK topographic map series 1984).

The climate of the area varies considerably depending on topography. The low lying areas to the southeast of Tbilisi are very dry and hot in summer, with an average annual temperature range of 2 - 23 °C (winter - summer) and annual precipitation less than 500 mm. In contrast, the mountains around the city have a temperature range of 0 to 20 °C and rainfall up to 800 mm. Two precipitation minima occur during the year: during winter and July-August [Svanidze and Papinashvili, 1992].

Complex topography, microclimates and land-use patterns enable a great variety of vegetation types to co-exist within a relatively small region. Tbilisi's vegetation is quite species-rich, with some 1643 species, being about 25% of the total number of species found in the Caucasus region as a whole. It also includes a number of local endemics (e.g. *Primula saguramica* Gavr. and *Anthemis saguramica* Sosn.) and can be classified into six major groups [Sakhokia, 1958]:

1. Semidesert – Composed largely of *Artemisia fragrans*-Chenopodiaceae (*Salsola*, *Suaeda*, *Petrosimonia* spp.) associations. This community is found only in the driest areas of the city, often in association with salt lakes.
2. Steppe – Dominated by *Bothriochloa ischaemum*, *Festuca valesiaca* and other grasses, this community is widespread on the left bank of the Mtkvari River and in some deforested areas.
3. Scrub – Composed of *Juniperus*, xerophile *Pyrus* species and deciduous maquis (forest species assuming shrubby form – *Quercus iberica*, *Carpinus orientalis*, *C. caucasica*,

Fraxinus excelsior etc.). This community is found on lower mountain slopes and in some cleared areas, especially near Mtskheta.

4. Forest – Most frequently an association of *Quercus iberica* and *Carpinus orientalis*, with a lesser role played by *Fagus orientalis* (on north slopes), *Carpinus caucasica* and *Pinus kochiana*. Found on most of the city's mountains.
5. Shibliak – A shrubby community whose indicator species is the Christ Thorn (*Paliurus spina-christi*). It often takes the place of forest where clearing has taken place and under the continual influence of livestock grazing. It may be found in association with steppe vegetation or with species typical of the forest understorey, including *Spiraea hypericifolia*, *Lonicera iberica*, *Rhamnus pallasii*, *Crataegus*, *Cotoneaster* and *Rosa* spp.
6. Phrygana – A community of widespread but fragmentary distribution, occurring amongst scrub and steppes. It is characterised by low, exceedingly prickly subshrubs (*Astragalus*, *Astracantha* and *Acantholimon* spp.) or perennials possessing pungently scented foliage (*Thymus tiflisiensis*, *T. transcausicus*, *Teucrium polium* etc.). Phrygana occurs particularly in places where livestock grazing is intensive.

It should be noted that much of the vegetation surrounding Tbilisi was artificially established during the Communist Period, especially pine plantations.

The study site, Tsavkisi wetland, is a reed-dominated mire located in the hills six kilometres west of Tbilisi, mid-way between the villages of Tsqneti and Tsavkisi (Figure 1). The elevation of the site is 1110 metres above sea level. Approximately 60% of the landscape within 1 km. of the site is forested, while the remainder consists of orchards, open steppe, patches of shibliak and fragments of phrygana vegetation.

Methods

Sediment cores were collected in 2002-2003 using a Russian D-section corer and a clay auger. Sedimentological changes were described in the field. Pollen was isolated from the sediment using standard techniques [Moore et al., 1991], mounted in glycerol and identified at $\times 400$ magnification. Between 400 and 1600 pollen grains were identified in each sample. Fauna and fungi considered useful for palaeoecological interpretation [van Geel, 1978] were identified, and charcoal particles quantified using the point-count method [Clark, 1982].

Pollen data were analysed statistically using Detrended Correspondence Analysis (DCA) [Hill and Gauch, 1980] and TWINSpan [Hill, 1979] in PC-ORD version 4.25 [McCune and Mefford, 1999]. DCA is a form of gradient analysis that is used to detect important trends in the data, while TWINSpan classifies samples into ecological groups and provides indicator species for each of these groups. Two radiocarbon dates were obtained, from the Australian Nuclear Science and Technology Organisation, on samples of concentrated pollen and charcoal.

Results

Sedimentology and Radiocarbon Dating

The lowermost section of the 7.8-metre Ts'avk'isi core consists of massive, dark grey, slightly calcareous silty sediment. At 6.65 m., these silts are interbedded with organic laminations, and above this depth the sediments are altogether more peaty, first dark brown, then yellow and unconsolidated. The sediment changes sharply to crumbly, dark brown peat above 5m, then changes again to dark grey silts that closely resemble those from the bottom of the core. Around 3 m. depth, the sediments yet again become peatier. Brown crumbly peat appears at 2.25 m. and the

upper part of the core is characterised by coffee-coloured reed peat with abundant gastropod remains. A schematic sedimentological log is provided on the pollen diagram (Figure 2).

Two accelerator mass-spectrometer (AMS) ^{14}C dates were obtained for the Tsavkisi material. The dates show that the record extends back some 6115 years (once differences between radiocarbon years and calendar years are accounted for). Sedimentation rates have decreased considerably in the past 2000 years because the sediments have accumulated to the same height as a small basin outflow, inhibiting further infilling.

Pollen Diagram

The sediments of Tsavkisi wetland are exceedingly rich in pollen. 95 stratigraphic levels were analysed and over 200 distinct pollen types were identified, as well as 15 types of fauna and fungi. The pollen diagram (Figure 2) is divided into 7 palynological zones based on statistical splitting [Birks and Gordon, 1985].

- Zone 1. The first zone has a high proportion of grass and herbaceous pollen (Asteraceae, *Galium*-type, *Potentilla*-type, *Filipendula*, *Polygonum aviculare*-type and *Plantago lanceolata*-type). In addition are many species characteristic of open vegetation – *Secale*-type, *Carduus*-type, Caryophyllaceae, *Colchicum*, *Echium*, *Gentianella campestris*-type, *Prunella*-type, *Saxifraga stellaris*-type and *Hornungia*-type. The arboreal component is dominated by *Quercus iberica* and *Carpinus caucasica*. Other important arboreal constituents include *Ephedra distachya*-type, *E. fragilis*-type, *Salix*, *Castanea*, *Pistacia*, *Tilia*, Rosaceae, *Paliurus*, *Vitis*, *Juniperus* and the juniper parasite *Arceuthobium oxycedri*. Charcoal particles are consistently represented throughout the zone, often in relatively high proportions. Of interest is the high proportion of *Sporormiella* dung fungal spores, indicative of livestock grazing [van Geel, 1978].
- Zone 2. The transition to the second zone is accompanied by a rapid increase in arboreal pollen, especially *Quercus*, *Carpinus caucasica* and *Fraxinus*. With the important exception of *Thalictrum*, grasses and herbs all decline. Charcoal concentrations and dung fungal spores also diminish.
- Zone 3. At the beginning of the third zone, *Fraxinus*, *Quercus* and *Corylus* decline. They are replaced by herbs such as *Hornungia*-type, *Potentilla*-type, *Filipendula*, *Polygonum aviculare*-type, *Gentiana*, Apiaceae, Asteraceae and rosaceous shrubs. In the middle part of the zone, *Juniperus* increases with *Plantago*, *Cirsium*, *Centaurea*, *Galium*-type, *Rumex*, *Urtica*, *Gentianella* and *Helianthemum*. Dung fungal spores again become important in this zone.
- Zone 4. In the fourth zone, Poaceae (grass pollen) increases dramatically, while arboreal pollen taxa all but disappear. Amongst herbaceous pollen otherwise dominated by *Plantago*, *Potentilla* and *Filipendula*, some relatively rare pollen types appear – *Scorzonera*, *Echinops*, *Cuscuta*, *Melampyrum*, *Polygala*, *Sideritis* and *Verbena*. Dung fungal spores are very well represented throughout the zone, and charcoal concentrations increase.
- Zone 5. The fifth zone includes the most remarkable change in the Ts'avk'isi pollen record. It begins with many fires (as indicated by charcoal peaks), a decrease in dung fungal spores and an increase in *Pinus* pollen. *Corylus*, *Alnus* and *Ulmus* increase initially, and the pollen of walnut (*Juglans regia*) and olive (*Olea europaea*) appears for the first time in the record. This is followed by an enormous increase in *Carpinus caucasica* pollen representation, up to 47% of dryland pollen. The poorly-represented pollen of *Carpinus orientalis* also peaks at this time. However, toward the end of the zone the role of *Carpinus* begins to diminish, and charcoal concentrations increase once again.

Zone 6. The sixth zone is most notable for its high proportion of fern spores (15%). These include spores of the Polypodiaceae, as well as *Polypodium vulgare*, *Adiantum* and *Pteridium*. Charcoal concentrations are consistently high, and the arboreal component is dominated by *Pinus* pollen.

Zone 7. In the last zone, fern spores decrease significantly, while grasses (Poaceae) and chenopods (Chenopodiaceae) increase. *Vitis* and *Cannabis* are important throughout. Arboreal pollen increases during the middle of the zone, particularly that of *Pinus*, *Quercus* and *Carpinus* spp. Charcoal concentrations are consistently, often exceedingly, high. Importantly, dung fungi (*Sporormiella* and *Podospora*) and grazing indicator pollen (*Potentilla*-type, *Ranunculus acris*-type, *Plantago major*-type and Fabaceae) are much reduced through zones 5-7 compared to zones 3-4.

Statistical Analyses

The pollen data were analysed using detrended correspondence analysis (DCA). This multivariate analysis has the great advantage of simplifying the many curves of the pollen diagram into just three major trends called axes. The first of these axes, and therefore the most significant one, shows a very strong relationship ($R^2: 0.73$) with the ratio between tree (arboreal) and non-arboreal pollen (Figure 2). *Carpinus orientalis*, *C. caucasica*, *Alnus*, *Fagus* and *Corylus* are all positively-correlated with this axis, while *Plantago coronopus*-type, *Sporormiella* (dung fungus), *Potentilla*-type, Poaceae and Fabaceae are negatively-correlated.

The second axis (Figure 3) is positively-correlated with Chenopodiaceae and *Pinus*, but negatively-correlated with *Quercus* and *Fraxinus* pollen, and microfossils of *Ceratophyllum*, an aquatic plant. The third axis (Figure 3) is positively-correlated with *Juniperus*, *Plantago lanceolata*-type, *Scleranthus*-type and *Centaurea nigra*-type pollen. Negative correlates include Poaceae and *Artemisia*.

The pollen data was then classified into ecologically-meaningful groups using TWINSpan. Indicators for each of these TWINSpan groups are given in Table I. The classification first splits the pollen record into two halves, at 185 cm. The first group (or 'stage'), from 780-185 cm., is indicated by *Juniperus* percentages of 2% or more. The other group, from 185-0 cm., is indicated by *Pinus* percentages of 5% or more and the presence of *Juglans* pollen. Each of these groups is divided into subgroups ('phases'). These results will be discussed at greater length below.

Table I. TWINSpan indicators for each of the classified groups.

Stage/phase	Indicator species (and cut levels)
Juniperus	Juniperus (2%)
Phase 1	<i>Quercus</i> (10%) and <i>Artemisia</i> (10%)
1a	<i>Gentianella campestris</i> -type (presence) and Rosaceae (presence)
1b	<i>Quercus</i> (20%), <i>Carpinus caucasica</i> (5%) and <i>Carpinus orientalis</i> (presence)
Phase 2	<i>Plantago coronopus</i> (2%), <i>Plantago lanceolata</i> (2%) and <i>Helianthemum</i> (presence)
2a	<i>Carpinus caucasica</i> (2%) and <i>Acer</i> (presence)
2b	Poaceae (20%)
Pinus-Juglans	Pinus (5%) and Juglans (presence)
Phase 3	<i>Plantago lanceolata</i> -type (2%) and <i>Polygonum persicaria</i> (presence)
3a	<i>Plantago lanceolata</i> -type (2%) and <i>Hordeum</i> -type (presence)
3b	Polypodiaceae (2%), <i>Cerastium</i> -type (presence) and <i>Polygonum amphibium</i> (presence)
Phase 4	<i>Carpinus caucasica</i> (5%), <i>Carpinus orientalis</i> (2%) and <i>Quercus</i> (10%)
4a	<i>Carpinus caucasica</i> (20%)
4b	Chenopodiaceae (10%)

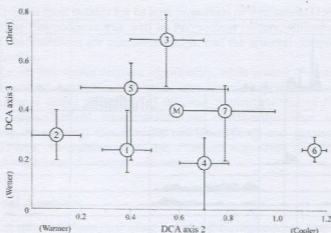


Fig. 3. Summary results of DCA axes 2 and 3, numbered according to pollen zones (see Fig. 2). The error-bars indicate the spread of data in each pollen zone in relation to temperature and precipitation gradients (see text). 'M' – modern sample.

Discussion

Vegetation History

The ecological groups provided by the statistical analysis provide a strong foundation for interpreting vegetation change on a landscape scale for the past 6115 years. In the following section, these groups are linked to vegetation communities and drawn into a timeline of vegetation development. The two major stages are, firstly, the *Juniperus* stage, lasting from 6115 years ago until the Hellenistic Period, and secondly, the *Pinus-Juglans* stage, from the Hellenistic Period to the present-day.

The *Juniperus* stage

Juniper is a shrub or tree characteristic of savanoid and scrub vegetation. It was clearly more prevalent at Ts'avk'isi prior to the Classical Period than it is today. Further evidence for this is the occurrence of its associated parasite, *Arceuthobium oxycedri*, in pollen spectra from that earlier time. Juniper is extremely sensitive to fire [Grove and Rackham, 2001], and this characteristic is exploited by pastoralists in the Alps to prevent juniper invading subalpine pastures. Although the charcoal record indicates that fires did occur during the *Juniperus* stage, their frequency and intensity appear to have been much less than in the post-Classical *Pinus-Juglans* stage. Grazing was obviously much more important during the *Juniperus* stage, as indicated by high proportions of the dung fungus *Sporormiella*. The *Juniperus* stage can be divided into two main phases:

Phase 1 (pollen zones 1-2; approx. 6000-4000 years ago): This phase characterised by relatively high proportions of *Quercus* and *Artemisia* pollen. *Quercus iberica* and *Artemisia* species are found together in open woodland and scrub communities in Georgia [Ketskhoveli, 1959], so the *Quercus-Artemisia* phase can be linked to an oak maquis community. This phase can further be divided into phases 1a and 1b, encompassing pollen zones 1 and 2, respectively. The indicators for each of these and their pollen spectra permit the conclusion that the earlier phase represents a heavily-grazed shibliak scrub and the later phase an oak-dominated woodland.

Phase 2 (pollen zones 3-4, approx. 4000-2300 years ago): Indicators for this phase are *Plantago* spp. and *Helianthemum*. In the Mediterranean region, *Helianthemum* is regarded as one of the most important indicator species of phrygana vegetation [Grove and Rackham, 2001]. *Helianthemum salicifolium* is commonly found in phrygana communities around Tbilisi along with grasses, *Onobrychis*, *Medicago*, *Teucrium*, *Thymus*, *Veronica*, *Polygala*, *Convovulus*, *Plantago*, *Filipendula*, *Potentilla*, *Galium*, *Falcaria*, *Trifolium*, *Eryngium* and *Astragalus* [Ketskhoveli, 1959: 246]. All of these taxa are well represented in the pollen spectra of pollen zones 3 and 4 of the Ts'avk'isi diagram. This *Plantago*-*Helianthemum* phase can be divided into phases 2a and 2b. The earlier phase (2a) is indicated by hornbeam and maple, the later phase (2b) by grasses. This suggests that the vegetation changed from a phrygana with elements of open woodland to one that was much more open and steppic.

The *Pinus*-*Juglans* stage

This stage begins in the Hellenistic Period or thereabouts. Its indicators are *Pinus* (representing *Pinus kochiana* and other species) and *Juglans* (representing the cultivated walnut, *Juglans regia*). Pine is usually found on poor, rocky soils where other more competitive deciduous trees will not grow, but it also competes with these trees in less marginal habitats by promoting fire through its resinous bark and flammable leaf litter [Rackham, 1990; Grove and Rackham, 2001].

The occurrence of *Juglans* pollen, along with *Olea europaea*, strongly suggest that these economically-important plants were introduced to the Tbilisi area by Greek or Roman colonists. While *Juglans* is cultivated near the Tsavkisi wetland today, the winters are too cold for olives, which are not grown in Georgia today. However, the 18th century Georgian geographer, Vakhushti, describes places on the lower Khrami River where olives, pomegranates and figs were grown in greater abundance than in other areas of eastern Georgia, because the climate there was warmer [Vakhushti, 1983: 428]. The low but consistent percentages of *Olea europaea* pollen suggest that olives grew there from the Hellenistic period until relatively recent times.

Phase 3 (pollen zones 4-7; approx. 2300-2000, 1300-1100, 400-50 years ago): This phase appears at three different periods during the pollen record. Its indicators are *Plantago lanceolata*-type and *Polygonum persicaria*. The plants that produce these pollen are found together on fallow land and along tracksides [Behre, 1986] and indicate open vegetation. Further division splits this phase into 3a and 3b. *Plantago lanceolata*-type remains an indicator of phase 3a, along with *Hordeum*-type pollen. Compared to phase 3b, whose indicators are all wet, ferny meadow taxa, phase 3a appears to indicate drier conditions. The Phase 3 community is always associated with high charcoal concentrations and elevated *Pinus* pollen percentages.

Phase 4 (pollen zones 5, 7; approx. 2000-1300, 1100-400 and 50-0 years ago): Like Phase 3, this phase appears three times during the site's vegetation history – once during the massive expansion of *Carpinus* pollen about 1900 years ago, again during the Middle Ages, and finally in the most recent times. Its indicators are *Carpinus caucasica*, *C. orientalis* and *Quercus*. These are, of course, the species most characteristic of the forests around Tbilisi today. The earlier occurrence of this forest community, during the Roman Period, is indicated by *Carpinus caucasica* pollen proportions greater than 20%. This suggests that *Carpinus caucasica* was dominant in the surrounding forests, as pollen percentages of the synonymous *C. betulus* indicate dominance when they exceed 10% [Huntley and Birks, 1983]. *Carpinus orientalis* was also of great importance in these forests, but produces small amounts of pollen. The second appearance of this phase (4b, during the Mediaeval Period and later) was obviously less wooded and more steppic, as the prevalence of

Chenopodiaceae attests. Pine was also of greater importance, and the vegetation was clearly moulded by fire.

Timeline

Using this information on plant communities and their appearance in at different times through the Ts'avk'isi record, the following stages of plant community development are proposed (dating is approximate, based on linear extrapolation):

- I. Open shibliak scrub or oak shrubland (6000-5000 years ago, phase 1a);
- II. Oak-dominated woodland (5000-4000 years ago, phase 1b);
- III. Phrygana in juniper-dominated open woodland (4000-3000 years ago, phase 2a);
- IV. Steppic-phrygana (3000-2300 years ago, phase 2b);
- V. Steppic fallow land with advancing forest (2300-2000 years ago, phase 3a);
- VI. Hornbeam forest (2000-1300 years ago, phase 4a);
- VII. Ferny meadow following forest destruction (1300-1100 years ago, phase 3b);
- VIII. Forest-steppe (1100-400 years ago, phase 4b);
- IX. Weedy steppe with forest elements (400-50 years ago, phase 3a);
- X. Forest-steppe (50 years ago to the present, phase 4b).

The conclusion to be drawn from this vegetation history is that the landscape near Tsavkisi wetland was, for much of the last 6000 years, much less forested than it is today. Around 2500-2000 years ago, conditions changed dramatically. Was this change related to climatic or anthropogenic factors? This is discussed in the following section.

Climate change during the past 6000 years

Perhaps the best evidence of climate change in the Caucasus region over the last 6000 years comes from an isotopic analysis of the laminated sediments of Lake Van, in eastern Anatolia [Lemke and Sturm, 1997]. This record has the advantage of coming from a very large lake (thus reflecting regional-scale changes), and being independent of vegetation (which is subject to many influences other than temperature and precipitation). It shows that the climate was relatively warm and moist from 6000-3500 years ago. Then the climate became briefly drier and somewhat cooler, and this cooler climate appears to have lasted until about 1500 years ago. Since that time, the climate varied considerably, but does not appear to have been significantly different to what we experience today.

In the Tsavkisi material, climatic gradients were extracted from the data using DCA. The dominant trend in the data (DCA axis 1) was not in any way related to the climatic trends seen in the Lake Van material. The second axis (Figure 3) appears to have some ecological relationship to temperature, since cold-tolerant *Pinus* is positively-correlated, whereas thermophiles such as *Quercus* and *Ceratophyllum* are negatively-correlated. This gradient exhibits an general trend similar to the Lake Van oxygen isotope curve, especially in the period 6000-2000 years ago. After this time the correlation between the two records is poor, and in some cases in opposition.

The same can be said of DCA axis 3. This is the least statistically important of the trends identified by DCA. Positive-correlates are xerophytes (*Juniperus*, *Plantago lanceolata*-type, *Scleranthus*-type and *Centaurea nigra*-type), while grasses and *Artemisia* are negatively-correlated. Grass and *Artemisia* pollen production is strongly dependant on rainfall [El-Moslimany, 1990], so this third axis (Figure 3) provides a rainfall gradient for the Tsavkisi record. The correlation with the Lake Van rainfall reconstruction is good from 6000-2000 years ago, clearly showing the rapid transition from moister to drier conditions about 3500 years ago (pollen zone 3) and the return to

moister conditions after 3000 years ago (pollen zone 4). However, the correlation is much poorer from 2500-2000 years ago to the present.

These trends suggest that the climate signal recorded by the Ts'avk'isi material reflects regional climatic changes until 2500-2000 years ago. The same trends are evident in the changing sedimentology of the wetland deposits. After this time, the Ts'avk'isi pollen-based climatic signal deviates considerably from the regional one, while the sedimentology does not change, so an alternative explanation must be sought.

Human Impacts on the Vegetation

The dominant statistical trend in the Tsavkisi pollen data is one of landscape openness. DCA axis 1 is positively-correlated with a suite of forest taxa and negatively-correlated with good indicators of open, grassy vegetation [Behre, 1986]. *Sporormiella* dung fungal spores are also strongly negatively-correlated ($R^2: 0.7$), indicating a strong influence from livestock grazing. This influence extended from at least 6000 years ago until the Hellenistic Period. After the massive expansion of hornbeam forests during the Roman Period, fire appears to have played a defining role in the ecology of forest and steppe communities.

On the Mediterranean island of Crete, a recent increase in pine has been attributed to a change from a grazing-dominated landscape to a fire-dominated one [Rackham, 1990]. If one compares the charcoal (fire indicator) and *Sporormiella* (grazing indicator) curves from Tsavkisi, they show that this same process probably occurred in the mountains around Tbilisi during the Classical Period. This indicates that the economy of the Tbilisi area changed considerably at the time that Greek and Roman cultural influences were first felt. Land-use of the mountains around the city changed from pastures to orchards.

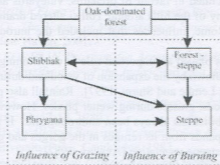


Fig. 4. A schematic model of plant community succession in the Tbilisi area under different and increasing human impacts. The ability of this process to operate in reverse (i.e. reafforestation) is determined largely by topography, since rugged terrain provides ample refugia for forest trees, while plains and plateaux, once deforested, are unlikely to recover quickly.

Forest composition changed as well – the importance of *Quercus*, *Juniperus* and *Fraxinus* declined considerably, and forests of *Carpinus caucasica* and *C. orientalis* expanded. A similar ‘explosive expansion’ of hornbeam forest, occurring during the Roman Period, has been observed in pollen diagrams in some parts of Europe, and the reasons for this have been debated for many years [Godwin, 1975; Huntley and Birks, 1983]. Climatic forces may have played a role, but there is such a poor correlation between the Lake Van isotope record and the Ts'avk'isi temperature gradient from the Roman Period that we must question this argument. The appearance of olive and walnut pollen, the decline of livestock grazing and an increasingly intensive fire regime led up to the expansion of these pure *Carpinus* forests.

The pollen record presented in this paper clearly shows that, in the vicinity of Tsavkiisi wetland, oak forest grew only during the early-middle Bronze Age, while since the Roman period, various forms of hornbeam secondary forest, forest-steppe and steppe prevailed. At many times during the past 6000 years, the vegetation was much more open and steppic than at present, a fact that may account for the region's high botanical diversity.

In order to conserve this biodiversity, it is important to understand how the various plant communities respond to different human impacts. Based on the Ts'avk'isi material, it seems that shibliak and phrygana are characteristic of heavy grazing, while secondary *Carpinus orientalis* forest-steppe and pure steppe are moulded to a greater degree by fire. Figure 4 provides a simple schematic representation of this theory.

Conclusion

In this paper we have presented a pollen-based reconstruction of vegetation changes near the city of Tbilisi over the past 6115 years. Specifically, the development of the vegetation can be traced from shibliak to oak woodland in the early-middle Bronze Age, through more steppic phrygana communities during the late Bronze and Iron ages, to cultural vegetation in the Hellenistic period, dense secondary forests in the Roman period, and finally to a stage where forests were destroyed by fire, recovered and were destroyed again, resulting in the present-day forest-steppe landscape.

Significantly, analysis of micro-charcoal and dung fungi preserved in the sediments has shown that land-use patterns changed considerably during the Hellenistic Period. From 6000 years ago until the Hellenistic Period, livestock grazing was of great importance in the area, leading to the expansion of shibliak and phrygana communities. But, around 2300 years ago, intensive grazing appears to have declined in favour of orchards, vineyards and other forms of cultivation. This means that the economy of the hills around Tbilisi altered dramatically as cultural influences arrived from Greece and Rome, influences that included the introduction and cultivation of the olive and walnut.

Climate has always played an important role in determining the character of the Tbilisi's surrounding vegetation, as shown by the expansion of woodland during a warmer phase 6000-3500 years ago [Kvavadze, 1999; Lemke and Sturm, 1997]. Rainfall also played a role, with xerophytes expanding during drier periods, grasses during wetter phases. However, the dominant force acting on the vegetation around Tbilisi has always been anthropogenic. If fire and grazing are removed, forest vegetation rapidly expands from its refuges in the gorges and gullies around Tbilisi to cover areas formerly occupied by steppe vegetation [Avakov, 1982].

The process of natural reforestation was witnessed by a Georgian chronicler during the Turkish occupation in 1080 AD: "In those times there was neither sowing nor harvest: the land was ruined and turned into forest" [Anon., 1982: 116]. Clearly since the most ancient of times, humankind has wittingly curtailed the expansion of forests onto agricultural land, and in doing so has created a cultural landscape with its own unique flora. We know that such a cultural landscape has existed near Tbilisi for at least 6000 years – the question that remains is: When was this landscape first created?

Acknowledgements

We wish to thank Dr. G. Avakov and Dr. O. Bendukidze (Institute of Palaeobiology) for their help in locating the Tsavkiisi site, Dr. Ian Thomas and Mr. Michael Fletcher (University of Melbourne) for fieldwork assistance. Thanks also to Prof. G. Mchelidze (Institute of Palaeobiology) Dr. G. Arabuli (State Museum of Georgia), Dr. Pim van der Knaap and Dr.

Jacqueline van Leeuwen (Swiss Institute of Plant Sciences) for their helpful comments, thanks to Caroline Connor for her great patience in preparing diagrams. Radiocarbon dates were provided through an AINSE PGRA.

References:

- Anonymous. *Tskhovreba mepet-mepisa Davitisi (Life of David, King of Kings)*. In: Z.Abesadze (ed.), *Kartuli P'roza III. Sabch'ota Sakartvelo*, Tbilisi, 1983 (in Old Georgian).
- Avakov, G.S. *Estestvennoe vosstanovlenie lesov v okrestnostiakh Tbilisi (Natural restoration of forest in the Tbilisi environs)*. *Priroda*, **11**, 43-49, 1982 (in Russian).
- Behre, K.-E. *The interpretation of anthropogenic indicators in pollen diagrams*. *Pollen Spores*, **23**, 225-245, 1986.
- Birks, H.J.B. and Gordon, A.D. *Numerical Methods in Quaternary Pollen Analysis*. Academic Press, London, 1985.
- Clark, R.L. *Point count estimation of charcoal in pollen preparations and thin sections of sediments*. *Pollen Spores*, **24**, 523-535, 1982.
- Connor, S.E., Thomas, I., Kvavadze, E.V., Arabuli, G.J., Avakov, G.S. and Sagona, A. *A survey of modern pollen and vegetation along an altitudinal transect in southern Georgia, Caucasus region*. *Rev. Palaeobot. Palynol.*, **129**, 229-250, 2004.
- Davies, C.P. and Fall, P.L. *Modern pollen precipitation from an elevational transect in central Jordan and its relationship to vegetation*. *J. Biogeogr.*, **28**, 1195-1210, 2001.
- Dzhanelidze, C. *Paleogeografia Gruzii v Golotsene (Holocene Palaeogeography of Georgia)*. Metsniereba, Tbilisi, 1980 (in Russian).
- El-Moslimany, A.P. *Ecological significance of common non-arboreal pollen: examples from drylands of the Middle East*. *Rev. Palaeobot. Palynol.*, **64**, 343-350, 1990.
- Godwin, H. *History of the British Flora*. Cambridge University Press, Cambridge, 1975.
- Grove, A.T. and Rackham, O. *The Nature of Mediterranean Europe: an Ecological History*. Yale University Press, New Haven, 2001.
- Hill, M.O. *TWINSpan: a FORTRAN Program for Arranging Multivariate Data on an Ordered Two-Way Table by Classification of the Individuals and Attributes*. Cornell University, Ithaca, NY, 1979.
- Hill, M.O. and Gauch, H.G. *Detrended correspondence analysis: an improved ordination technique*. *Vegetatio*, **42**, 47-58, 1980.
- Huntley, B. and Birks, H.J.B. *An Atlas of Past and Present Pollen Maps for Europe: 0-13000 years ago*. Cambridge University Press, Cambridge, 1983.
- Juansher. *Tskhovreba Vakhtang Gorgasali (Life of Vakhtang Gorgasali)*. In: L. Nanitashvili (ed.), *Kartuli Proza I. Sabchota Sakartvelo*, Tbilisi, 1981 (in Old Georgian).
- Ketskhoveli, N. *Sakartvelos Mtsenareuli Sapari (Vegetation Cover of Georgia)*. SSSR Metsnierebata Akademiis Gamomtsemloba, Tbilisi, 1959 (in Georgian).
- Kvavadze, E.V. *Golotsenovye kolebaniia urovniia ozera Lisi i izmeneniia polozhenii niznei granitsy lesa (Holocene fluctuations of the Lisi lake level and the shift of the lower forest line)*. *Paleobiologii Problemebi* **1**, 75-87, 1999 (in Russian).
- Lemke, G. and Sturm, M. *$\delta^{18}O$ and trace element measurements as proxy for the reconstruction of climate changes at Lake Van (Turkey): preliminary results*. In: H.N. Dalfes, G. Kukla and H. Weiss (eds.), *Third Millennium BC Climate Change and Old World Collapse*. NATO ASI Series, 1997.
- McCune, B. and Mefford, M.J. *PC-ORD: Multivariate Analysis of Ecological Data*. MjM Software, Gleneden Beach, Oregon, 1999.
- Moore, P.D., Webb, J.A. and Collinson, M.E. *Pollen Analysis*. Blackwell, London, 1991.

- Rackham, O. *The greening of Myrtos*. In: S. Bottema, G. Entjes-Nieborg and W. van Zeist (eds.), *Man's Role in the Shaping of the Eastern Mediterranean Landscape*. A.A. Balkema, Rotterdam, 1990.
- Sakhokia, M.F. *Botanicheskoe opisaniie okrestnostei goroda Tbilisi – plato Shiraki (Botanical description of the Tbilisi environs to the Shiraki Plateau)*. In: M.F. Sakhokia (ed.) *Botanicheskie ekskursii po Gruzii*. Tbilisi, 1958 (in Russian).
- Sosnovskii, D.I. *Protsessy ischeznovaniia lesov v okrestnostiakh Tiflisa (Process of forest disappearance near Tbilisi)*. Izv. K.O.R.G.O. **№ 1**, 1915.
- Svanidze, G.G. and Papinashvili, L.K. (eds.). *Klimat Tbilisi (Climate of Tbilisi)*. Gidrometeoizdat, St. Petersburg, 1992 (in Russian).
- Tournefort, J.P. *A Voyage into the Levant*. Browne et al., London, 1718.
- Vakhushti Bagrationi. *Aghts'era sameposa Sakartvelosa (Description of Georgia)*. In: N. Abesadze (ed.), *Kartuli P'roza V. Sabch'ota Sakartvelo*, Tbilisi, 1983 (in Old Georgian).
- van Geel, B. *A palaeoecological study of Holocene peat bog sections in Germany and the Netherlands*. Rev. Palaeobot. Palynol. **25**, 1-120, 1978.

ჰავისა და ადამიანის ზემავლენა თბილისის მიდამოების მცენარეულობაზე ბოლო 6000 წლის განმავლობაში

კონნორი ს.¹, ყვავაძე ე.²

¹ მელბურნის უნივერსიტეტის გეოგრაფიისა და გარემოს შესწავლის კათედრა, ავსტრალია

² საქართველოს მცენერებათა აკადემიის ლ. დავითაშვილის სახელობის პალეობოტანიკის ინსტიტუტის პალეობოტანიკის განყოფილება.

(მიღებულია 07.06.2005)

რეზიუმე

მოცემულია თბილისის მიდამოების მცენარეულობის განვითარების ისტორია. გამოყენებულია პალინოლოგიური მეთოდი, ნახშიროვანი ნაწილაკების და სოკოთა სპორების კვლევის შედეგები. შესრულებულია ტბური ნალექების რადიოკარბონული დათარიღება. მიღებული შედეგები აჩვენებს, რომ თბილისის მიდამოების მცენარეულობის განვითარების ისტორიაში შეიმჩნევა რამოდენიმე მნიშვნელოვანი ეტაპი. ბრინჯაოს ხანამდე, 6000 წლის წინ აქ გავრცელებული იყო შიბლიაკის ტიპის ცენოზები ნათელი ტყის ელემენტებით. ადრე და შუაბრინჯაოს დროს თბილისის მიდამოებში დომინირებდა მუხის ტყე. გვიანბრინჯაოსა და რკინის ხანაში კი გავრცელებულა უფრო ღია ლანდშაფტები, სადაც სტეპებსა და ფრიგანას მნიშვნელოვანი ფართობი ეკავათ. ელინისტურ დროს აქ კულტურულ მცენარეთა შორის ჩნდება ზეთისხილი. რომაულ პერიოდში გაბატონდა რცხილისა და ჯაგრციხლის ტყე. მოგვიანებით, ეს ტყე შეიცვალა ტყესტეპითა და სტეპით. სტატისტიკური მეთოდების საშუალებით (TWINSPAN და DCA) დადგენილია მცენარეთა ეკოლოგიური დაჯგუფებები და მათი ხასიათი, აგრეთვე განსხვავება ჰავისა და ადამიანის ზეგავლენას შორის; კერძოდ, გაირკვა, რომ ჰავასთან შედარებით, აქ ადამიანის ზეგავლენა უფრო მნიშვნელოვანია.

STUDY OF LECTIN ACTIVITY IN THE NUCLEAR PROTEIN FRACTION OF THE WHEAT (*T. AESTIVUM* V. *HOSTIANUM*) LEAVES

ALEKSIDZE G.* , TEVZADZE N.* , GABATASHVILI B., GIGOLASHVILI G.

Department of Plant Physiology, Iv. Javakhishvili Tbilisi State University

(Received May 16, 2005)

Abstract

The carioplasm, histone- and non-histone protein fractions were isolated from the cellular nuclei of the wheat (*T. aestivum* v. *hostianum*) leaves. Lectin activity in the histone protein fraction was revealed. By the chromatography it was shown that histone fraction consist of some protein fractions, and its basic properties were confirmed with isoelectric focusing. Ligand-receptor test showed that lectin activity of the histone proteins is inhibited with the non-histone protein fraction, which points at existence of lectin-bound glycoconjugates.

Key words: chromatin, chromatography, isoelectric focusing, histone protein fraction.

Introduction

The lectins for the first time were found in plants, later this group of proteins was also intensely studied in bacterial and animal organisms. Interaction between lectins and sugar-containing biopolymers plays a key role in the process of biological recognition [Lutsik, 1981]. In plants lectins are found in all parts of a plant, their content varies at different stages of development, and in different organs; mostly they are presented in actively growing and functioning tissues.

It is known that functioning of lectins is connected with the processes occurring on a cellular surface; however, there are some data certifying that lectin properties are characteristic of some membrane and soluble protein fractions, localized on the autonomous organelles – nuclei, chloroplasts, and mitochondria. This shows that functioning of lectin systems may occur in the intracellular processes as well [Aleksidze, 1980]. Subcellular localization of lectins, as concerned with their functions, is most perfectly studied in animals. In various animal cells the nuclear lectins have been found [Hubert, 1994]; presumably, they take part in transcription regulation, as well as in transport of a transcript [Laing, 1988]. Activity of the enzymes – ribonucleases and polymerases – in its turn may be dependent on their utilization of the carbohydrates [Kurl, 1988]. On the other hand, specific glycolisation may serve as a nucleic target signal or substantial recognition line in incorporation of some glycoproteins into the transcription process [Polet, 1988]. The data on biochemistry and physiological role of plant nuclear lectins are relatively sparse.

* Corresponding authors. giorgi_alexidze@yahoo.com tevza55@posta.ge

Considering above-mentioned, the goal of present study was fractionation of the cariolasm and chromatin proteins from the plant nuclei, and determining lectin activity in these fractions.

Material and Methods

The wheat plant (*T. aestivum* v. *hostianum*) was used as the study material; plant was grown in the laboratory conditions. For obtaining the nuclear fractions via centrifuging in saccharose density gradient and further purification of the nucleic fractions obtained with non-ionic detergents the two-week leaf shoots of wheat were used [Jokhadze, 1976]. All procedures of the nuclei fractionation were carried at 0-4°C. Integrity and purity of the nuclei was examined in the light microscope, after staining with methylen blue.

The nuclear sediment was dispersed in the lysis solution – phosphate buffer of 20 mM K (pH 6.0), 0.5 mM PMSF (phenylmethylsulphonilfluoride), in a ratio of 1:3. Then the suspension was quick-frozen and melted, in order to rupture the nuclei membranes. The melted suspension was centrifuged at 8000 g, for 10 min (K-24). The resulting supernatant was utilized as the soluble protein fraction of cariolasm, while the sediment contained membranes, chromatin, and nucleoli. The histone and non-histone protein fractions from the chromatin were obtained with the modified Bornkamm method [Bornkamm, 1972]. Extraction of the non-histone proteins from the sediment was performed with high ionic power solutions. Specifically, the sediment was added with extraction buffer – 0.35 mM NaCl, 0.5 mM PMSF, at a ratio of 1:3. Extraction was made at +4°C, for 30 min, and then it was centrifuged at 8000 g, for 10 min (K-24). The supernatant contained the non-histone protein fraction (the procedure was repeated twice). In order to extract remaining non-histone proteins, the sediment was treated with 0.5 M KCl (0.5 mM PMSF) solution, at a ratio of 1:3. Extraction was made at +4°C, for 30 min, and then it was centrifuged at 8000 g, for 10 min (K-24). Histones were extracted by acidic solutions. To remaining sediment 0.3 N HCl (0.5 mM PMSF) solution was added and extraction was centrifuged at 8000 g, for 10 min (K-24); the supernatant contained fraction of the histone proteins.

The protein fractions obtained were subjected to dialysis against the agglutination buffer (0.9% NaCl, phosphate buffer of 0.04 M K, pH 7.4 (PBS)). Lectin activity of the protein was assessed with hemagglutination test on the rabbit's trypsinized- and non-trypsinized erythrocytes, in immunological planchettes, according to Takachi micro-titration method. Lectin activity was evaluated by minimal concentration ($\mu\text{g/ml}$) of the protein, which still induces visible agglutination of the rabbit erythrocytes, or by specific activity (SA, mg/ml), which shows maximal dilution of the protein, which still induces agglutination ($\text{SA}=\text{T}/\text{c}$, where T – titer of hemagglutination or dilution of the lectin in the last cell, c – protein concentration (mg/ml) [Peumans, 1984]). Volume of the protein was assessed with the Bradford method [Bradford, 1976]. Revealing of lectin-binding capacity of the protein and assessment of specificity against carbohydrates were executed with hapten-inhibitory technique, based on inhibition of hemagglutination activity of a lectin [Lutsik, 1981].

Isoelectric focusing was performed in the 0.25 mm thick gels (pH 3-10). Size of the gel was 50/45 mm. Concentration of the protein under study was 1 mg/ml. Sample was applied in the volume of 1 μl , conditions of isoelectric focusing were as follows: 2000 V, 2.5 mA, 15°C, 500 Vh. Staining was made in the Coomassie Br. Bl. G-25 solution, dispersed in 3.5% solution of HClO_4 . Staining was performed at 50°C, for 30-45 min. Excess dye was washed out of the gel with 10% acetic acid. The following standard protein kits were used as markers (pH): Cytochrome C (horse, heart) (10.7); Ribonuclease A (bovine, pancreas) (9.5); Lectin (*Lens culinaris*) (8.3; 8.0; 7.8); Myoglobin (horse, muscle) (7.4; 6.9); Carbonic anhydrase (bovine, erythrocytes) (6.0); b-

Lactoglobulin (bovine, milk) (5.3; 5.2); Trypsin inhibitor (soybean) (4.5); Glucose oxidase (Aspergillus niger) (4.2); Amyloglucosidase (Aspergillus niger) (3.5); (Serva).

Gel-filtration of the histonic proteins was made on Toypearl HW-55 gel column (1.8 x 14 cm) and for elution PBS buffer (pH 6.8) was used. Elution of the proteins was made at 1 ml/min velocity, and in obtained fractions volume of proteins and lectin activity was assessed.

Results and Discussion

As pointed above, the first stage of our experiments implied obtaining the nuclei, from which, via various extraction solutions, the nuclear protein fractions were obtained – 1) fraction of the carioplasm soluble proteins; 2) non-histonic protein fraction; 3) histonic protein fraction.

In the rabbits' trypsinized and non-trypsinized erythrocytes, lectin activity of each fraction was evaluated with hemagglutination test. No hemagglutination activity was found in the carioplasm- and non-histonic fractions. Lectin activity against the native erythrocytes was revealed in the fraction of histonic proteins. Minimal concentration of the protein, which produced visually noticeable agglutination, was 0.6 $\mu\text{l/ml}$; the index of specific activity amounted 512 (Table 1).

Table 1. Lectin activity of the histonic proteins

Protein fraction	Protein, Mg	Titer	Lectin activity, $\mu\text{g/ml}$	Specific activity, ml/mg	Total activity, ml
Carioplasm proteins	0.5				
Non-histonic proteins	6.5				
Histonic proteins	2.5	512	0.6	512	1280

At the next stage of investigation, with an aim to reveal lectin-binding proteins in the overall nuclear proteins, the ligand-receptor inhibition method was used; this method implies shielding in the lectins of the carbohydrate-binding centers by glycoproteins and, as a result, suppression of the lectin-determined agglutination activity. The ligand-receptor test, performed in different nuclear proteins, has shown that lectin activity of histonic fraction obtained from the wheat leaves is inhibited by the non-histonic protein fraction, while the soluble protein fractions from carioplasm, in the same conditions, do not display the lectin-binding features. The results obtained demonstrate that in the non-histonic fraction there should be the lectin-binding glycoconjugates.

The results obtained in our experiments confirm also the literature data, which show that glycoproteins must participate in modulation of the nuclear fraction, specifically – in the transcription processes. Interaction between the histonic lectins and glycoproteins might be an additional mechanism for selective activation of chromatin. This hypothesis is supported by the data showing that in the nuclei there exist glycoproteins of highly mobile group [Putnam, 1994] and with a property of concentration in the chromatin loci, where activation and active transcription processes do occur.

In the next series of experiments a specificity of histonic proteins, possessing lectin activity, against carbohydrates was investigated; this property is considered as the main and most essential feature of lectins. The following carbohydrates were utilized as haptens – glucose, fructose, D-galactose, N-acetyl-D-glucosamine, manose, L-arabinose, D-xilose, D-lactose, D-raphinose (initial concentration was 400 mM). It was established that inhibition of the hemagglutination activity induces fructose, partly – xilose, and lactose (Table 2).

Table 2. Specificity of protein fractions, possessing lectin activity, against carbohydrates.

Carbohydrates (Initial concentration – 400mM)	Minimal inhibitory concentration of carbohydrate (mM)
Hexoses	
1. Glucose	–
2. Fructose	25
3. D-galactose	–
4. N-acetyl-D-glucosamine	–
5. Manose	–
Pentoses	
6. L-arabinose	–
7. D-xilose	100
Di- and trisaccharides	
8. D-lactose	200
9. D-raphinose	–

In order to purify histonic protein fraction, the chromatographic method was used. Studied protein divided into three fractions. Elution profile is presented on Fig. 1. In each protein fraction, we determined lectin activity. It was found that lectin activity was present in the proteins of first and second fractions; proteins of the second fraction were characterized with exceptionally high activity.

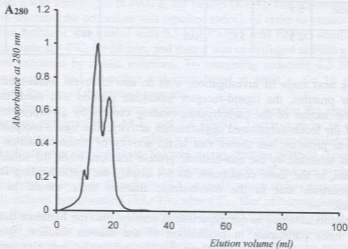


Fig Gel-filtration profile on the Toyopearl HW 55 column of the protein fraction possessing lectin activity. Elution buffer – 0.9% NaCl, phosphate buffer of 0.04M K (pH 6.8); elution velocity – 1 ml/min, A-280 nm, protein volume applied on the column – 1.5 mg.

To determine a chemical nature of the protein fractions extracted from chromatin, which have lectin activity, isoelectric focusing was performed via Phast System electrophoresis device (Pharmacia, Sweden), which clearly confirmed well-pronounced basic properties of these proteins. After staining of the gel two stripes of proteins were revealed with pH 10.7 and 10.2 respectively (Fig. 2).

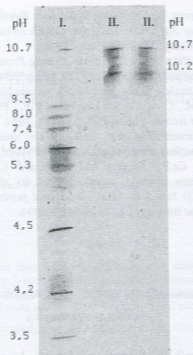


Fig. 3. Electrophoresis of histone protein fraction with isoelectric focusing I.—standart proteins: Cytochrome C (10.7); Ribonuclease A (9.5); Lectin (8.3; 8.0; 7.8); Myoglobin (7.4; 6.9); Carbonic anhydrase (6.0); b-Lactoglobulin (5.3; 5.2); Trypsin inhibitor (4.5); Glucose oxidase (4.2); Amyloglucosidase (3.5); (Serva). II— histone protein fraction with lectin activity.

Presented data certify existence of lectins and glycoconjugates in the protein fractions isolated from the cell nuclei of the wheat leaves. Their physiological role and molecular mechanisms of action in the plant cells are still unknown; therefore, further studies in this direction are in progress.

References:

- Aleksidze G.Y., Vyskrebtsseva E.I. *Subcellular localization of lectins in the roots of beet of different ages.* Fiziologia Rastenii, **37**, 2, 1980 (in Russian).
- Bornkamm G.W., Nobis P., Sonnenbichler J. *Concerning the specificity of histone and non-histone dissociation from calf thymus chromatin by salt.* Bioch. Bioph. Acta, **278**, 258-265, 1972.
- Bradford M.M. *A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding.* Anal. Biochemistry, **72**, 248-254, 1976.
- Hubert J., Seve A.P. *Nuclear Lectins.* Lectins: Biology, Biochemistry. Clinical Biochemistry, **10**, 220-226, 1994.
- Jokhadze D.I., Balashvili M.I. *Comparative investigation of RNA-polymerase activity in the cellular nuclei and chloroplasts.* Biokhimiia, **41**, 161-165, 1976 (in Russian).

- Kurl R. N., Holmes S. C., Verney E., Sidransky S. *Nuclear Envelope Glycoprotein with Poly (A) Polymerase Activity of Rat Liver: Isolation, Characterization and Immunohistochemical Localization*. *Biochemistry*, **27**, 8974-8980, 1988.
- Laing J. and Wang J. *Identification of Carbohydrate Binding Protein 35 in Heterogenous Nuclear Ribonucleoprotein Complex*. *Boichemistry*, **27**, 5329-5334, 1988.
- Lutsik M.D., Panasyuk E.N., Lutsik A.D. *Lectins*. M., "Vysshaya Shkola", 1981 (in Russian).
- Peumans W.J., Nsiba-lubeki M., Carlier A.R., Driessche V.E. *Proposal from a novel system of nomenclature of plant lectins*. *Planta*, **160**, 220-228, 1984.
- Polet H. and Molnar J. *Demonstration that some of the non-histone proteine, inducible to translocate into the nucleus are glycosylated*. *Cell Physiol.*, **135**, 47-54, 1988.
- Putnam Ch.D., Copenhaver Gr.P., Denton M.L., Pikaard Cr.S. *The RNA polymerase I transactivator upstream binding factor requires its dimerization domain and high-mobility-group (HMG) box 1 to bend, wrap, and positively supercoil enhancer DNA*. *Molecular and Cellular Biology*, **14**, 6476-6488, 1994.

ხორბლის (*T.aestivum v.hostianum*) ფოთლის უჯრედების ბირთვებში ლექტინურ
აქტივობის შესწავლა

ალექსიძე გ., თევზაძე ნ., გაბათაშვილი ბ., გიგოლაშვილი გ.

მცენარეთა ფიზიოლოგიის კათედრა, ივ. ჯავახიშვილის სახ. თბილისის
სახელმწიფო უნივერსიტეტი

(მიღებულია 16.05.2005)

რეზიუმე

ხორბლის (*T. aestivum v. hostianum*) ფოთლების უჯრედული ბირთვებიდან გამოყოფილია კარიოლაზმური, პისტონური და არაპისტონური ცილოვანი ფრაქციები. აღნიშნულ ცილებში შესწავლილია ლექტინური აქტივობა, რომელიც გამოვლინდა პისტონური ცილების ფრაქციაში. ქრომატოგრაფიის მეთოდით ნახევნებია, რომ პისტონური ფრაქცია შედგება რამოდენიმე ცილოვანი ფრაქციისგან, ხოლო მისი ფუძე თვისება დამტკიცდა იზოლექტროფოკუსირებით. ლიგანდ-რეცეპტორული ტესტით ნახევნებია, რომ პისტონური ცილების ლექტინური აქტივობა კავდება არაპისტონური ცილოვანი ფრაქციით, რაც მასში ლექტინ-დამაკავშირებელი გლიკოკონიუგატების არსებობაზე მიუთითებს.

THE SEASONAL CHANGES OF CHEMICAL CONTENTS OF BLACK SEA MUSSELS (*RAPANA THOMASIANA* CROSSE) BODIES

PALAVANDISHVILI N., IMNADZE V.

Department of Zoology and Water Biorecources, Sh Rustaveli Batumi State University

(Received March 14, 2005)

Abstract

Contents of some chemical composition and their changes depending on seasonal and ecological factors in bodies of Black Sea mussels - *Rapana thomasiana* were studied. It was shown that contents of proteins, lipids, ash and moisture depends on physiological state of mollusk, as well as on season, water temperature and nutrient medium.

Key words: gastropod, mussels, ecological factors, Black Sea.

Introduction

Rapana thomasiana (Crosse, 1861) (synonyms: *Rapana venosa* (Valenciennes, 1846), *Rapana pontica* (Nordsieck, 1969)) was introduced into the Black Sea in the 1940s and within a decade spread along the Caucasian and Crimean coasts and to the Azov Sea. Its range extended into the northwest Black Sea to the coastlines of Romania, Bulgaria and Turkey from 1959-1972. This species have been introduced and become established in the northern Adriatic and Aegean seas and the southeast coast of South America [Harding, 2003].

Rapana thomasiana is a gastropod with a characteristic deep orange aperture and columella. This species compact sandy bottoms where it can burrow deep into the substrate. Its native habitat is a region of wide annual temperature ranges. *Rapana thomasiana* has caused significant changes of the ecology of bottom-dwelling organisms and has the potential to damage native species [TheChesapeakeBay.com., 1999].

Rapana thomasiana is a prolific extremely versatile species, tolerating low salinities, water pollution and oxygen deficient waters [Mann & Harding, 2003]. It is an active predator of epifaunal bivalves, and its proliferation is a serious limitation to natural and cultivated populations of oysters and mussels [CEISM, 2000].

In Japan *Rapana thomasianahas* has been sold as seafood on Japanese markets and equivalent to other mollusks consumed in countries of Mediterranean culture. In Georgia from 1980s it has taken significant place in seafood industry.

Like other mollusks *Rapana thomasiana* is rich in biological active substances. To reveal nutritional value of mussels it is important to study chemical content of this species. The contents of proteins, lipids, free amino acids, ashes, microelements in bodies of the Black Sea mussels have been quantitatively estimated [Poliakov, 1999; Stepanyuk et al., 1990; Nekhoroshev et al., 1990].

The goal of our work was to study changes of chemical content of the mussels body from Ajara coastline depending on seasonal and ecological factors.

Materials and Methods

Studies were carried out 2000-2003 seasonally. Mussels were caught in Batumi coastline at 10-15 m depths.

The lengths of mussels were determined and material was divided into 3 classes: I class - mussels of 50 mm length, II - 60 mm and III - 70 mm.

Body was released from the shell of mussel and crushed. Biochemical studies were carried out by size classes. Protein, lipid ash and moisture contents were determined by the standard method [Lazarevski, 1955]. On every sample analyses was conducted three times and average value was calculated.

Results and Discussion

Data of chemical content of mussels by size classes and season are presented in the table 1. As it is seen from the table maximum amounts of protein occur in spring and winter periods. water temperatures were +18°C and +9°C correspondingly. In spring nutrient medium was favourable for mussels development.

Decrease of protein content in summer and autumn periods should be connected with the process of gametogenesis, violation of temperature regime, nutrient medium. In autumn the amount of mature oocytes and spermatozooids is decreased but of small ones is increased. At this time process of gametogenesis is intensified which endures to spring. In winter mussel gets over sandy bottoms and till spring it is in rest state. During the whole winter mussel is less active and accordingly its energy consumption is less compared to reproduction and growth periods. So in winter period intensive accumulation of proteins is noted.

Temperature is significantly influenced biological processes, such as quantitative and qualitative changes of metabolism, rate of growth, intensity of nutrition. At high temperature protein and carbohydrate metabolism prevail, at low temperature - metabolism of lipids. Received data show that in winter when temperature of water is 9°C content of lipids is lower. In winter mussels are in rest state and energy for their vital activity is nutrients supply. In summer lipids content increased which is energy supply for gonadotrophic tissue. It is seen evidently in III size-class (13.75%).

Table 1. Chemical composition of mussels body (*Rapana thomasiana*)

Season	size-classes	Quantity, %			
		proteins	lipids	moisture	ash
spring	I	23,83	0,89	70,61	1,57
	II	25,67	1,09	75,23	1,52
	III	27,31	0,85	79,47	2,01
summer	I	10,52	2,28	76,12	1,40
	II	11,2	2,24	76,69	1,77
	III	14,06	3,76	85,33	2,02
autumn	I	9,81	1,49	84,23	1,46
	II	13,36	0,64	81,85	1,84
	III	16,19	0,87	84,50	1,85
winter	I	37,06	0,28	55,57	2,0
	II	24,01	0,19	83,87	1,87
	III	28,76	2,34	83,13	2,25

Mussels chemical composition in autumn and summer is nearly similar. But decrease of lipid content in autumn compared to summer may be connected with temperature regime (in summer temperature +26°C, in autumn -21°C) and with pass of mussel into winter period.

As it is seen from the table moisture is gradually increased from spring and reaches maximum in all three size-classes in autumn (83.9%). As for mineral substances its content is higher in winter period (for three size-classes average amount is 2.07%) and in other seasons - 1.70%.

Compared to literature data [Poliakov, 1999; Stepanyuk et al., 1990; Nekhoroshev et al., 1990] chemical contents of mussels body of southeast coastline (Ajara) differ from the ones of other regions of the Black Sea. According to our data content of proteins, lipids, ash and moisture depends on physiological state and length of mollusk, as well as on season, water temperature and nutrient medium.

References:

- CIESM. *Rapana venosa*. CIESM atlas of exotic species in the Mediterranean Sea. vol.2, 2000.
 Harding J.M. *Predator by blue crabs, Callinectes sapidus, on rapa whelks, Rapana venosa possible natural controls for an invasive species?* J.Exp.Marine Biol. and Ecol., **297**, 161-177, 2003.
 Lazarevski A.A. *Technical-chemical control in fish industry*. M., "Pishchepromizdat", 1955 (in Russ).
 Mann R., Harding J.M. *Salinity tolerance of larval Rapana venosa: implications for disperse and establishment of an invading predatory gastropod on the North American Atlantic*. Biol.Bull., **204**, 161-177, 2003.
 Nekhoroshev M.V., Uss Yu.A., Shalyapin V.K. *Chemical composition of biodeposits and the rate of their extraction by cultivated mussels*. Ecology of the Sea, **36**, 37-41, 1990.
 Poliakov L.K. *Mussels - use for nutrition*. Crimean, "Vest", 1999 (in Russ).
 Stepanyuk I.A., Golovenko V.K., Poludina V.P. *Protein and amino acid pool of the Odessa Bay mussels*. Ecology of the Sea, **36**, 54-60, 1990.
 TheChesapeakeBay.com. *The Vined Rapa Whelk (Rapana venosa)*. 1999.

შავი ზღვის რაპანას (*Rapana thomasiana* Grosse) ზოობიოტი ქიმიური მანკმენებლების სეზონური ცვლილებები

ვალანდიშვილი ნ., იმნაძე ვ.

ბიოლოგიისა და წყლის ბიორესურსების კათედრა, შ. რუსთაველის
სახელობის ბათუმის სახელმწიფო უნივერსიტეტის

(მიღებულია 14.03.2005)

რეზიუმე

შესწავლილია რაპანის სხეულის ქიმიური შემადგენლობა. განსაზღვრულია ცილების, ცხიმების, ტენიანობის, მინერალური ნივთიერებების რაოდენობრივი შემცველობა. დადგენილია, რომ შავი ზღვის რაპანის ქიმიური შემადგენლობა იცვლება სეზონების მიხედვით, რაც დამოკიდებულია არა მხოლოდ მოლუსკის ბიოლოგიურ მდგომარეობაზე, არამედ წყლის ტემპერატურასა და კვებით ბაზაზე.

ინსტრუქცია ავტორთათვის

სამეცნიერო ნაშრომს ჟურნალი ბეჭდავს ინგლისურ ენაზე, მას უნდა დაერთოს რეზიუმე ინგლისურ და ქართულ ენაზე, სამეცნიერო მიმართულება, სათაური, ავტორთა გვარები და მათი სამუშაო დაწესებულების დასახელება, საკვანძო სიტყვათა მოკლე (4-6) სია.

სამეცნიერო წერილის მოცულობა არ უნდა იყოს 5 გვერდზე ნაკლები და 12 გვერდზე მეტი. წერილი უნდა გაფორმდეს შემდეგი რუბრიკაციით: შესავალი და მიზნები (introduction), მასალა და მეთოდები (materials and methods), შედეგები და მათი განხილვა (results and discussion), დამოწმებული ლიტერატურა. უკანასკნელი უნდა იყოს დალაგებული ანბანის მიხედვით, ხოლო ტექსტში წყაროების მითითება უნდა ხდებოდეს ფრწხილებში ჩასმული ავტორის გვართა და წლით [Lernmark, Hagglof 1981].

ლიტერატურა წარმოდგენილი უნდა იყოს შემდეგნაირად:

ჟურნალისთვის:

Carvalho C., Pereira H., Pina C. *Chromosomal G-dark bands determine the spatial organization of centromeric heterochromatin in nucleus*. Mol. Biol. Cell, 12, 5, 3563-3572, 2001.

წიგნისთვის:

Kuhn T.S. *The structure of scientific revolutions*. Chicago, IL, Chicago Press, 2000. an Gentner D., Brush S. *Flowing waters or teeming crowds*. In: Mental Models. D. Gentner (Ed.), Chicago IL., Chicago Press, 865-900, 2001.

ქართული ტექსტისთვის ოპტიმალური ფონტებია AcadNusx და AcadMtavr, ინგლისური ტექსტებისთვის - Times New Roman. შრიფტის ზომა - 12 პუნქტი, ინტერვალი - 1.5. ცხრილებში დასაშვებია უფრო მცირე ზომის შრიფტები. ჭერილი უნდა დაიბეჭდოს A4 ფორმატით, ზევით და ქვევით - 2,5 სმ., მარცხნივ - 3 სმ. და მარჯვნივ - 2სმ. დაშორებით. ცხრილები, გრაფიკები და დიაგრამები (მხოლოდ შავ-თეთრი) შესაძლებელია დამზადდეს როგორც Microsoft Word-ში, ისე Excel-ში, ფოტოსურათები მიიღება აგრეთვე ორიგინალების (არაელექტრონული) სახითაც.

მასალა რედაქციაში წარმოდგენილი უნდა იყოს როგორც ქაღალდზე ამობეჭდილი, ისე კომპიუტერულ დისკეტით ან CD-ზე. წერილი მთლიანად ერთი ფაილით უნდა იყოს შენახული, ხოლო ფაილის სახელწოდება წერილის პირველი ავტორის გვარს უნდა ატარებდეს. ცალკე ფაილად შეიძლება ილუსტრაციების წარმოდგენა.

ჟურნალის ბეჭდვა ავტორთა ხარჯებით ხორციელდება. თანხა რედაქციაში უნდა შემოვიდეს ნაშრომზე დადებითი რეცენზიის მიღებისთანავე. ნაშრომის რეცენზირება ანონიმურია და ავტორს აქვს უფლება მიიღოს ან არ მიიღოს რეცენზენტის შენიშვნები. უკანასკნელ შემთხვევაში ნაშრომი, დამატებით გაეგზავნება სარედაქციო საბჭოს ერთ-ერთ წევრს. მეორე უარყოფითი დასკვნის შემთხვევაში, ნაშრომი არ გამოქვეყნდება.

ნაშრომი ჩაბარება შეიძლება სამუშაო დღეებში, 12-დან 16 საათამდე, შემდეგ მისამართზე: თბილისი, რუსთაველის გამზირი 52, საქართველოს მეცნიერებათა აკადემია, ბიოლოგიის განყოფილება, IV სართული, 429 ოთახი, ტელ: 93-58-92, პასუხისმგებელი მდივანი - მაია გრიგოლავა.

ռ. 9/4.