ISSN 1512-2123

საქართველოს მეცნიერებათა აკადემიის

მაცნე

ბიოლოგიის სერია B
Biological series

2007
No. 1
Vol. 5

PROCEEDINGS

of the Georgian Academy of Sciences

ISSN 1512 – 2123

საქართველოს მეცნიერებათა აკადემიის

მაცნე

ბიოლოგიის სერია B
Biological series

2007
No. 1
Vol. 5

PROCEEDINGS

of the Georgian Academy of Sciences

Tbilisi. Georgia
2007

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

Published by NGO "Biomed", 14, L. Gotua st., Tbilisi 0160, Georgia

CONTENTS

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Biochemistry

THE IMPACT OF BIOACTIVE PREPARATIONS ON THE RESISTANCE OF RYEGRASS EXPOSED TO ORGANIC TOXICANTS – BENZENE, 3,4-BENZPYRENE AND TRINITROTOLUENE

*TSULUKIDZE N., BETSIASHVILI M., DZAMUKASHVILI N., SADUNISVILI T., KVESITADZE E.

Durmishidze Institute of Biochemistry and Biotechnology
**E-mail: ninuca_18@yahoo.com*

(Received October 16, 2006)

Abstract

Changes of the activities of glutathione S-transferase, phenoloxidase and peroxidase and of protein content in ryegrass, in response to the effect of organic pollutants – benzene, 3,4- benzpyrene (Bp), trinitrotoluene (TNT) and bioactive preparations, have been studied. It has been established that in most cases, during treatment of ryegrass seedlings with bioactive preparations – Fosnutren and Humiforte, enzyme activities and the protein amount have increased dramatically. Combined effects of benzene, Bp and TNT with bioactive preparations on enzymatic systems of ryegrass have been studied. It has been ascertained that treatment with bioactive preparations caused increase in the activities of glutathione S-transferase, phenoloxidase and peroxidase and protein amount in ryegrass that enhance the plant resistance to organic toxicants.

Key-words: Fosnutren, humiforte, glutathione S-transferase, ryegrass, organic pollutants

Introduction

The most effective remediation, perfect restoration and long-term preservation of chemically contaminated environment are possible by application of phytoremediation technologies. Phytoremediation involves clarification and restoration of chemically contaminated environment by means of plants and microorganisms, which are able to utilize and transform wide range of organic and inorganic toxicants. The plant (with its detoxification potential) capable to utilize the toxicants from – air, soil and water, all three elements of biosphere, is the most efficient mean for restoration of ecologically sound environment [Korte, 2000].

Detoxification process of toxic compounds in plant cell proceeds in three phases: reaction of activation, conjugation and compartmentation [Coleman, 1997]. In the first phase, hydrophilic group is formed in xenobiotic molecules at the expense of enzymatic transformation. As a result, the polarity and reactivity of toxicant molecule significantly increases. Various enzymes, including peroxidases and phenoloxidases, catalyze the activation reaction of xenobiotics [Kvesitadze et al., 2006].

Peroxidase (EC 1.11.1.7) is a widely spread enzyme, found in all green plants, fungi and aerobic bacteria. In a plant cell these enzymes display diverse functions, participate in a number of physiological and detoxification processes: hormonal regulation, lignification, and response to stress conditions, protection of a cell from infections and hydroperoxides. Free radical containing products formed as a result of reactions catalyzed by peroxidase are able to oxidize other compounds, including xenobiotics. Peroxidases of different plants are able to oxidize dimethylaniline, benzpyrene, phenol, aminofluorine and hydroxianysoles [Siegel, 1993].

Phenoloxidase (EC 1.14.18.1) is a copper containing metaloenzyme widely spread in microorganisms, plants, insects and animals. Phenoloxidase actively participate in oxidative degradation of organic toxicants. If the xenobiotic is of phenolic nature, then it is a substrate of phenoloxidase and oxidized in monophenolase and diphenolase reactions. In other cases, xenobiotic oxidation is carried out by co-oxidative mechanism by endogenous phenols [Papunidze et al., 2005].

In the second, conjugation phase xenobiotic or metabolite, activated in the first phase, connect with endogenous hydrophilic molecule. The obtained conjugate is polar and less toxic. In the third phase, compartmentation of inactive water-soluble conjugates takes place in vacuoles or cell wall.

Glutathione S-transferase (GST, EC. 2.5.1.18) is a representative of cytosolic enzymes. This dimmeric enzyme catalyzes bonding of tripeptide glutathione to electrophylic sites of various organic and inorganic molecules. Detoxification of various endo and xenobiotic compounds through this enzyme occurs as a result of covalent bonding between hydrophobic substrate and SH-group of cysteine residue in glutathione [Armstrong, 1997]. The obtained conjugate is less reactive and polar that simplifies its further compartmentation [Coskun, 2002]. In analogue with other detoxification enzymes, some isomers of this enzyme are inducible. Their intracellular level can increase as a result of effect of plant hormones, pathogens and xenobiotics (e.g. herbicides) [Lamoureux, 1989; Mars, 1996].

Frequently, under the effect of toxic compounds enhancement of protein biosynthesis processes in plant cells is observed. Increase of protein amount on one hand promotes balance of protein deficiency, found at conjugation with toxic compounds as a result of protein expenditure, and on the other hand is connected with induction of enzymes participating in detoxification processes [Kvesitadze et al., 2005].

At elaboration of new technologies, great importance is attached to the approaches that enable to regulate ecophysiological characteristics of plants without interfering their genome. In this point of view, the preparations of Spanish firm INAGROSA, particularly Fosnutren and Humiforte are of interest. These preparations are complex of synthesized free amino acids and microelements, applying of which significantly enhance plant productivity, and at the same time, their resistance to different toxicants effects. It should be mentioned that their absorption does not consume plant energy and depend on chlorophyll activity.

Ryegrass is widely applied for planting lawns in cities and along motorways. Consequently, evaluation of the plant resistance to different toxicants, prevalent in the environment and its detoxification capability is of importance.

Some compounds, widely spread in the environment and characterized by high toxicity have been chosen for the experiments. At present, motor transport takes the main place in environmental pollution in developed countries. Exhaust, together with different toxic compounds contains benzene and 3,4-benzpyrene. Compounds containing aromatic rings are extremely toxic and carcinogenic [Samoillloff, 1998; Curfs, 2003]. Among explosives, trinitrotoluene is the most toxic. Hundreds of hectares of contaminated soil remain after hostilities and military exercises [Robidoux, 1999].

The goal of our investigation was to estimate combined impact of bioactive preparations and organic pollutants on ryegrass seedlings in order to increase its phytoremediation capability.

Materials and Methods

The object of the study was ryegrass, seeds of which were swollen and after grown in tap water during 10 days. To induce enzymes the seedlings were placed in solutions containing benzene (0,1mM), 3,4-benzpyrene (0,1mM) and trinitrotoluene (0,1mM), during 5 days. Fosnutren and Humiforte, bioactive preparations were added together with toxicants in concentrations of 5 ml/l. After exposure, roots and leaves were cut and homogenized in a mortar in 0.05M phosphate buffer pH 7.5, in the ratio 1:3. After centrifugation at 12000g, 30 min, the obtained supernatant was studied for enzymes activity.

Glutathione S-transferase activity was determined spectrophotometrically at 340 nm, by measuring the rate of 1-chloro 2,4-dinitrobenzene (CDNB) conjugation with reduced glutathione [Habig, 1974]. Reaction mixture contained 1mM glutathione and 0.1ml enzyme preparation in 0.2M phosphate buffer, pH 6.5, final volume 3ml. Reaction started by addition of 1mM CDNB. Analogous mixture of the same content without enzyme preparations was used as a control.

As a unit of glutathione S-transferase activity the enzyme amount, which catalyzes conjugation of 1mM CDNB with glutathione in 1 min at 25°C is taken.

Peroxidase activity was determined spectrophotometrically at 470 nm according to the rate of guaiacol oxidation [Gregori 1972]. Reaction mixture contained 10mM guaiacol, 0.5 ml H_2O_2 solution (0.3%) and 0.01ml enzyme preparation in 0.05M phosphate buffer, pH 5.4, final volume 3ml.

As a unit of peroxidase activity the enzyme amount catalyzing oxidation of 1mM guaiacol in1 min at 25°C is taken.

Phenoloxidase activity was determined spectrophotometrically at 430 nm according to the rate of pyrocatechine oxidation [Lanzarini, 1972]. Reaction mixture contained 2mM pyrocatechine and 0.1ml enzyme preparation in citrate buffer, pH 4.7, final volume 3ml. Enzyme activity is expressed in ΔE/min.

The enzyme activities were calculated per g of fresh weight and expressed in percents in Tables. Protein was determined by Bradford's method [Bradford, 1974]. As a standard - bovine serum albumin was used.

Results and Discussion

Changes in the activities of glutathione S-transferase, peroxidase and phenoloxidase and protein content have been studied in roots and leaves of ryegrass, treated by biopreparations (Fig. 1). According to the obtained results, glutathione S-transferase and peroxidase activities were enhanced in roots and leaves of plants treated with biopreparations. Increase of peroxidase activity was observed only in plant roots, where the enzyme activity, in comparison with leaves, is higher, according to the literature data as well [Siegel, 1993]. Protein content in treated plants increases significantly both in roots and in leaves. Especially significant increase of protein amount, by 100% was observed in plants treated with Humiforte.

Changes in activities of glutathione S-transferase, peroxidase and phenoloxidase and in protein content were studied at exposure to different toxicants together with bioactive preparations in seedlings of ryegrass. At exposure of seedlings to benzene, significant increase in activities of enzymes and protein content was observed in experiments, when the plant was treated with toxicants together with bioactive preparation (Fig. 2). Different pictures were observed in roots and

leaves. Increase of activities in enzymes participating in detoxification was clearer; presumably, it is connected with treatment method of plants as roots are in direct contact with xenobiotics and with the ability of biopreparations to resist and limit transportation of toxicants to aboveground organs [Kvesitadze, 2005]. On such ability of Fosnutren and Humiforte indicates also significant increase of protein amount in roots, and decrease of protein content in leaves. In that case, obvious difference between stimulating effects of Fosnutren and Humiforte was not observed.

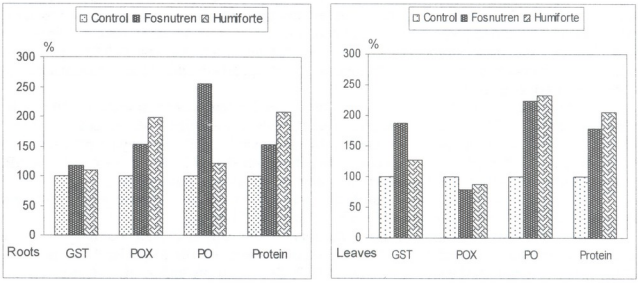


Fig. 1. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to Fosnutren and Humiforte. Exposure time – 5 days. Concentrations of Biopreparations – 5ml/l.

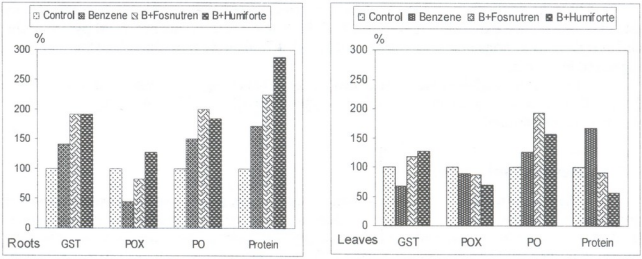


Fig. 2. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to benzene, Fosnutren and Humiforte. Exposure time – 5 days. Concentration of benzene – 0.1mM/l. Concentrations of Biopreparations – 5ml/l.

Changes in enzymes activities in response to exposure to 3,4-benzpyrene (Bp), organic toxicant, have been studied (Fig. 3). As a result of the experiments, it has been established that mainly, glutathione S-tranferase and oxidation enzymes activities increase in roots and leaves of ryegrass, in response to treatment with xenobiotic. Decrease of protein amount in seedlings, treated with toxicants only, indicates on higher toxicity of benzpyrene in comparison with that of benzene.

However, under the influence of bioactive preparations this toxic effect is significantly decreased, while protein content and enzyme activity increased.

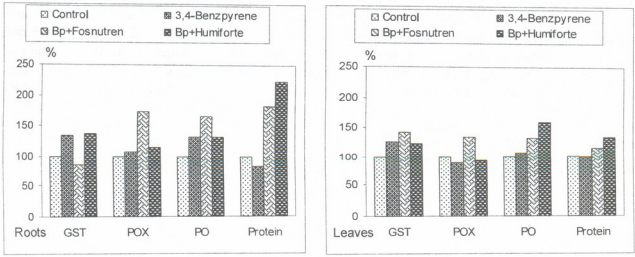


Fig. 3. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to 3,4-benzpyrene (Bp), Fosnutren and Humiforte. Exposure time – 5 days. Concentration of 3,4-benzpyrene – 01,mM/l. Concentrations of Biopreparations – 5ml/l.

The influence of trinitrotoluene (TNT) on seedlings of ryegrass has also been studied (Fig. 4). According to the obtained results, decrease in activity of oxidation enzymes is mainly observed in most cases, connected with another pathway of xenobiotic transformation in the studied plant. Increase of glutathione S-tranferase activity is observed in roots. Presumably, the enzyme renders safe active metabolites formed in the cell under toxicants exposure. Increase of protein content is found both in roots and leaves; it is especially sharp in the presence of bioactive preparations.

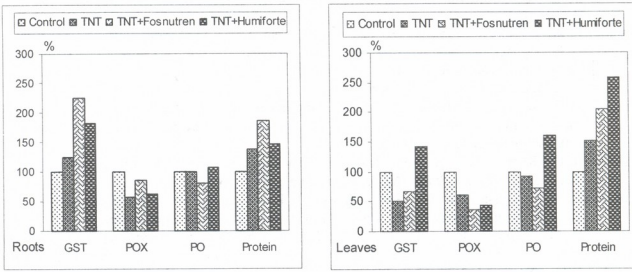


Fig. 4. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to TNT, Fosnutren and Humiforte. Exposure time – 5 days. Concentration of TNT – 0.1 mM/l. Concentrations of biopreparations – 5ml/l.

According to the obtained results, it might be concluded that at treatment with Fosnutren and Humiforte, in most of cases, activities of glutathione S-transferase, phenoloxidase, peroxidase, and protein content enhance dramatically in ryegrass. Application of bioactive preparations together with toxicants increases the plant resistance to xenobiotics as free amino acids and microelements are essential substrate for synthesis of enzymes and/or their substrates (glutathione in case of glutathione S-transferase). Ryegrass, as evergreen plant is widely used in decorating of cities and along motorways. On the base of our experiments, it might be concluded that application of the plant is desirable in combination with bioactive preparations, which significantly improve its phytoremediation capability. It should be also mentioned that the plant genome would not be affected; besides, it is safe for the environment. Phytoremediation itself is another method of nature protection from the dangerous impact of humans.

Acknowledgement: Biologically active preparations – Humiforte and Fosnutren were kindly provided by INAGROSA, Industrias Agrobiologicas, S.A.

References:

Armstrong R. *Structure, catalytic mechanism, and evolution of the glutathione transferases*. Chem. Res. Toxicol. **10**, 2-18, 1997.

Bradford M. *A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the triple of protein dye binding.* Anal. Biochem., **59**, 277-282, 1974.

Coleman J., Mechteld M., Emyr Davis T. *Detoxification of xenobiotics by plants: chemical modification and vacuolar compartmentation.* Trends in Plant Science, **2**, 141-151, 1997.

Coskun G., Zihnioglu F. *Effect of some biocides on glutathione-s-transferase in barley, wheat, lentil and chickpea plants.* Turk J. Biol., **26**, 89-94, 2002.

Cufts D., Beckers L., Godschalk R., Gijbels M., Van Schooten F. Environ. Mol. Mutagen. **42**, 243-249, 2003.

Gregory R. *A rapid assay for peroxidase activity*. Biochem. J., **101**, 582-583, 1966.

Habig W., Jakoby M. *Glutathione S-transferases – the first enzymatic step in mercapturic acid formation*, J. Biol. Chem., **22**, 7130-7139, 1974.

INAGROSA, Industrias Agrobiologicas, S.A.

Korte F., Kvesitadze G., Ugrekhelidze D., Gordeziani M., Khatisashvili G., Buadze O., Zaalishvili G., Coulton F. *Review: Organic toxicants and plants*. Ecotoxicology end Environmental Safety. **47**, 1-26, 2000.

Kvesitadze G., Khatisashvili G., Sadunishvili T., Evstigneeva Z. *Metabolism of Anthropogenic Toxicants in Higher Plants*. Moscow, "Nauka", 2005 (In Russian).

Kvesitadze G., Khatisashvili G., Sadunishvili T., Ramsden J. *Biochemical mechanisms of detoxification in higher plants. Basis of phytoremediation.* Springer, 2006.

Lamoureux G., Rusness D. *The role of glutathione and glutathione-S-transferases in pesticide metabolism, selectivity, and mode of action in plants and insects*. Wiley Interscience, 153-196, 1989.

Lanzarini G., Pifferi P., Samorani A. *Specificty of an o-diphenol oxidase from Punus avium fruits*. Phytochemistry, **11**, 89-94, 1972.

Marrs K.A. *The functions and regulation of glutathione S-transferases in plants.* Plant Mol. Biol. **47**, 127-158, 1996.

Papunidze V., Khatisashvili, G., Sadunishvili T. *Plant for a Healthy Environment*. 2005 (In Georgian).

Robidoux P., Hawari J., Thiboutot S., Ampleman G., Sunahara G. Ecotoxicol. Environ. Saf., **44**, 311-321, 1999.

Samoiloff M. *Benezene toxicity. Benzene and derivatives*. Organic chemistry **3**, 1998.

Siegel B. *Plant peroxidases - an organismic perspective*. J Plant Growth Regul., **12**, 303-312, 1993.

ბიოსასუქების გავლენა კოინდარის მდგრადობაზე ორგანული ტოქსიკანტების – ბენზოლის, 3,4-ბენზპირენისა და ტრინიტროტოლუოლის ზემოქმედებისას

წულუკიძე ნ., ბეციაშვილი მ., ძამუკაშვილი ნ., სადუნიშვილი თ., კვესიტაძე ე.

დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 16.10.2006)

შესწავლილია ფერმენტების – გლუტათიონ S-ტრანსფერაზას, ფენოლოქსიდაზას, პეროქსიდაზას აქტივობებისა და ცილის შემცველობის ცვლილება კოინდარში ორგანული ტოქსიკანტების – ბენზოლის, 3,4-ბენზპირენის, ტრინიტროტოლუოლისა (TNT) და ბიოსასუქების ზემოქმედების საპასუხოდ. დადგენილია, რომ კოინდარის ნაზარდების ბიოაქტიური პრეპარატებით – ფოსფუტრენითა და ჰუმიფორტეთი დამუშავებისას უმრავლეს შემთხვევაში მკვეთრად იმატებს ფერმენტების აქტივობა და ცილის რაოდენობა. შესწავლილია ბენზოლის, 3,4-ბენზპირენის, TNT-ს და ბიოსასუქების ერთობლივი გამოყენების გავლენა მცენარის ფერმენტულ სისტემებზე. დადგენილია, რომ ბიოაქტიური პრეპარატებით დამუშავება იწვევს კოინდარში გლუტათიონ S-ტრანსფერაზას, ფენოლოქსიდაზას, პეროქსიდაზას აქტივობისა და ცილის რაოდენობის მატებას, რაც ზრდის ამ მცენარის გამძლეობას ორგანული ტოქსიკანტების მიმართ.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Biotechnology

EFFECTIVE CONTROLLING OF BACTERIAL SPOT IN TOMATO WITH BACTERIOPHAGE

GHUDUMIDZE N.[1], ALAVIDZE Z.[2], CHKONIA I.[2], ELIASHVILI P.[3], GIORGOBIANI N.[3], SHAPOVALOVA N.[1], MEIPARIANI A.[2], SADUNISHVILI T.[1]

[1]*S. Durmishidze Institute of Biochemistry and Biotechnology*
[2]*G. Eliava Institute of Bacteriophages, Microbiology and Virology*
[3]*L. Kanchaveli Institute of plant protection*

(Received October 16, 2006)

Abstract

Tomato foliages were artificially contaminated with the culture washed out of 4 pathogenic strains of *Xanthomonas vesicatoria*. The disease developed on lower side as well as on upper side of foliages. The disease was caused by infection with pathogenic strains isolated from both damaged foliages and fruits. Spraying foliages with $7x10^6$ p.f.u. of phage just at the moment of infection or 24h later hindered the disease onset, but a week after affection suppressed the disease development. Unlike chemical substances phage did not require to be sprayed twice or several times. Phage does not damage the plant foliages and does not provoke the disease outbreak. Phage application is safe for a human as well as for a plant and it can be used as a salutary agent in dealing with damages caused by Bacterial Spot of sowing areas.

Keywords: Bacterial Spot, tomato foliages, chemical substances, bacteriophage.

Introduction

Bacterial Spot in tomato leaves is caused by phytopathogenic bacteria-*Xanthomonas vesicatoria*. Damage from these diseases may range from a light spotting of the foliage to almost complete defoliation of the plant, with corresponding impacts on the ability of photosynthesis and production potential [Leboeuf et al., 2005], and therefore decreases the yield of tomato crops. When the conditions are optimal for bacterial multiplication (high humidity, 28-32°C.) loss in tomato crops marketable yield can be great.

In order to control the Bacterial Spot the chemical substances are used. Treatment with acid or chlorine may be comparatively effective, if properly manage. But chlorine is inactivated by organic matter and its activity is affected by the pH of solution. Clearly, maintaining the accurate pH is a critical moment for successful disinfection with acid [Leboeuf et al., 2005].

Today the most effective bacteriocide against tomato Bacterial Spot in Georgia still has been copper sulfate spray. But copper sprays are less effective, when spray intervals are extended (7 days or less interval is required). Bacterial populations may show wide resistance to the given solution. Application of high concentrations of copper ions can damage plant tissue leading to a rapid bacteria reproduction. Copper spray may suppress the bacteria on the foliage surfaces, but bacteria located deeply may survive, multiply and cause an outbreak [Leboeuf et al., 2005].

Application of Cixomi and other copper-containing preparations including Kupxodat, Kuprophlo also are used for controlling the tomato Bacterial Spot.

All these compounds in certain amounts may penetrate into a human organism. The prevalence of the toxic substances can hinder such biological processes as growth, development, propagation and in some cases even stop them. Pesticides play a key role in xenobiotics, although, humans have to use pesticides, which finally get into biosphere and humans become a target of their action [Jurin, 2002].

At present the researchers try to study other desinfectants and alternative methods including treatment by microware, sonication and hydrostatic pressure.

One of the encouraging alternative ways in combating against Bacterial Spot in tomato is an application of bacteriophages for treatment purposes, as phages are a specific kind of viruses that attack suitable bacteria to kill pathogenic microorganisms.

The goal of our paper was to show the possibilities of phage application as an alternative tool to chemical substances and a natural biocontrolling agent against Bacterial Spot caused artificially on the tomato foliages.

Materials and Methods

In our experiment was used: *Lycopersicum esculentum* 45 day seedlings; 24h. the culture washed out of 4 various pathogenic strains of *Xanthomonas vesicatoria,* among them 3 strains isolated from the damaged tomato foliages and 1 - from damaged fruit; $7x10^6$ p.f.u. phage mixture of pure lines of mixture of phages isolated from sewages and damaged tomato materials.

The leaves selected for controlling were mechanically damaged by scratching the leaves with a needle. Infection was performed by dropping the bacterial culture onto the mechanically damaged plant leaves [Baltyukova et al., 1968].

The culture was dropped by micropipette. One strain infected some foliages located on various branches of one plant, which were mechanically damaged and added with drops of culture on both, lower and upper sides of foliages; the phage was sprayed by means of special sprayer.

Experiment procession. Tomato seedlings were planted separately into the pots and placed in the greenhouse. For trial 25 days later healthy plants were selected and divided into 5 groups. Control plants were included into I group - a total, 2 plants. The plant foliages of II group were infected with *Xanthomonas vesicatoria* culture - a total, 4 plants. This group was designed for bacterial controlling. The plants, foliages of which were sprayed with phage just at the moment of affection, were included into III group - a total, 4 plants. The plant foliages of IV group were sprayed with phage 24h after affection - a total, 4 plants. The plant foliages of V group were sprayed with phage after a week - a total, 4 plants.

The plants were placed in the thermostat room at constant temperature 28°C. The aeration was performed by airing the room. By day the light was switched on. High humidity was provided by frequent watering and natural evaporation of water from watery vessels.

Results and Discussion.

Observation was carried out within 4 week. It is remarkable, that the signs of bacterial spotting among the plants infected with bacteria were detected only 4 days after affection (II and IV groups) and only on the 5th day the disease developed in a shape of brownish spots on the lower and upper sides of the foliages, infected with pathogenic strains isolated from both damaged foliages and fruits.

During the whole experiment no signs of disease development were observed on the foliages of I group plants (Fig.1). As a result of the disease development on the plant foliages in II group, damaged leaves appear yellow (10 days) (Fig.2). Subsequently all leaves of the branches entirely yellowed. No signs of disease were observed on the plant foliages in III and IV groups within the whole period. After production of Bacterial Spot on the plant leaves in V group the disease was eliminated by phage spray - light spots remained on the infected sites (Fig.4), but the disease did not develop. Evidence of disease was not observed within 3 weeks. Thus, the results of the experiment demonstrated that bacteriophage application succeeded in dealing with bacterial spotting developed on the tomato foliages being infected with *Xanthomonas vesicatoria*. Phage hindered the disease onset, when it was sprayed just at the moment of bacteria affection (Fig.3); after 24h and even after 7 days. It is remarkable, that during the experiment in the case of each variation phage was used only once.

Fig.1. The control plant. (10th day of observation).

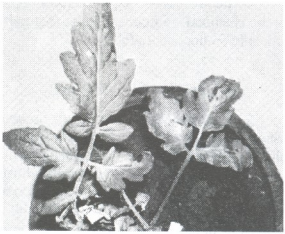


Fig.2. The bacterial-control plant (10th day of observation).

Fig.3. Phage was sprayed at the moment of infection (the 10-th day of observation).

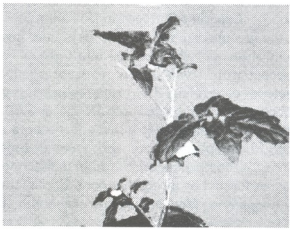


Fig.4. Phage was sprayed after a week of Infection (the 10-th day of observation).

Thus, phages can be used as an effective treatment remedy against Bacterial Spot produced on the tomato foliages. In comparison with chemical substances phages have some advantages: phage preparation is cheaper; when penetrating into any infected site the phage remains there until suitable bacterium exists; phage application does not damage plant organisms even in the case of a high concentration. When getting into a human organism through tomato the phages are safe unlike the chemical substances.

Acknowledgement: This work was carried out with financial assistance of ISTC within the framework of the project NG-1129.

References:

Baltjukova K., Matishevskaja M., Kulikovskaja M., Sudorenko C. *Methods for studying the causative against of plant bacterial disease*. Kiev. Nauka Dumkova. 1968.

Jurin V. *Basis of xenobiology*. Minsk. Novaja Znanie. 2002.

Leboeuf J., Cuppels D., Dick J., Pitblado R., Poewen St., Celetti M. *Bacterial diseases of tomato*; *Bacterial Spot*, *Bacterial Speck*, *Bacterial Canker*. Queen's Printer for Ontario. Factsheet ISSN 1198-712X, 363-365, 2005.

პამიდვრის ბაქტერიული სილაქავის ეფექტური მკურნალობა ბაქტერიოფაგით

დუდუმიძე ნ.[1], ალავიძე ზ.[2], ჭყონია ი.[2], ელიაშვილი ფ.[3], გიორგობიანი ნ.[3], შაპოვალოვა ნ.[1], მეიფარიანი ა.[2], სადუნიშვილი თ.[1]

[1]ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი
[2]გ. ელიავას ბაქტერიოფაგის, მიკრობიოლოგიისა და ვირუსოლოგიის ინსტიტუტი
[3]ლ. ყანჩაველის მცენარეთა დაცვის ინსტიტუტი

(მიღებულია 16.10.2006)

რეზიუმე

Xanthomonas vesicatoria-ს 4 პათოგენური შტამის ჩამონარეცხი კულტურით მოხდა პამიდვრის ფოთლების ხელოვნური ინფიცირება. დაავადება განვითარდა ფოთლების როგორც ქვედა, ასევე ზედა მხრიდან ინფიცირების შემთხვევაში. ფოთლებში დაავადება გამოიწვია როგორც დაავადებული ფოთლებიდან, ასევე ნაყოფიდან გამოყოფილი პათოგენური შტამებით დახნებოვნებამ. $7x10^6$ ტიტრის მქონე ფაგის ფოთლებზე შესხურებამ ინფიცირებისთანავე, ან 24 საათის შემდეგ ხელი შეუშალა დაავადების დაწყებას, ხოლო ინფიცირებიდან 1 კვირის შემდგომმა შესხურებამ შეაჩერა დაავადების განვითარება. ქიმიური საშუალებებისგან განსხვავებით, ფაგის მეორე ან მრავალჯერადი შესხურება არ იყო აუცილებელი, ფაგი არ აზიანებს მცენარის ქსოვილებს და ამით არ ახდენს დაავადების აფეთქების პროვოცირებას. ფაგის გამოყენება უსაფრთხოა როგორც თვით მცენარისთვის, ასევე ადამიანისთვის და შესაძლებელია გამოყენებული იყოს ნათესი ფართობების ბაქტერიული სილაქავისგან გამოწვეული დაზიანების საწინააღმდეგო სამკურნალო საშუალებად.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Botany

INTRASPECIFIC CHEMICAL DIFFERENTIATION OF *URTICA DIOICA* L. GROWING IN GEORGIA

KAVTARADZE N., GVINIASHVILI TS., ALANIA M., KUCHUKHIDZE J.

Kutateladze Institute of Pharmacochemistry

(Received May 15, 2006)

Abstract

Distinctive chemical and morphological features of *Urtica dioica* L. growing in Georgia have been revealed. *Utrica dioica* with red coloration was defined as a new variety - *Utrica dioica* L. var. rubescens Gviniaschvili et Kavtaradze – planta cum anthocyanea.

Key words: *Urtica dioica* L., flavonols, anthocyans.

Introduction

80 species of the genus Urtica L. are spread in temperate and tropical zones of both hemispheres [Mabberley, 1998]. Two species *Urtica dioica* L. and *Urtica urens* L. grow in Georgia [Shkhian, 1975]. They are cosmopolites, follow humans and are met in the conditions of violated natural plant cover. *Urtica dioica* L. is distributed everywhere. It grows in ruderal places, near housings, drafts and banks.

In folk and scientific medicine tincture and decoction of *Urtica dioica* L. is used as hemostatic agent [Budantsev et al., 1985]. Roots and rhizomes of this plant present a raw material for medicinal preparation used at prostate adenoma [Gide-book Vidal, 2002]. Data about phenolic composition of *Urtica dioica* occur in scientific literature. Flavonoids [Chaurasia, 1987], lignins [Kraus et al., 1990], coumarins [Wichtl, 1996] were isolated from *U. dioica*.

Chemical characteristics of *Urtica dioica* growing in Georgia were not studied yet. Phytochemical analysis revealed that qualitative chemical compositions of above-ground parts of *Urtica dioica* growing in various regions of Georgia are different.

The goal of our research was to find out differences of chemical compounds in correlation with morphological characteristics of the plant.

Materials and Methods

U. dioica was studied during 2000-2003. Herbarium material was collected in East Georgia (Kartli, Gori district) and West Georgia (Khobi district, village Alioni) during mass florescence of plant (July-August).

Traditional methods of chemical analysis were used [Alania et al., 2002]. Macromorphological studies of plant were also carried out (Table 2).

Results and Discussion

Our field investigations showed that plants of common *U. dioica* growing throughout the country develop green above-ground parts, but the specimens collected in West Georgia – red ones. Flavonoid glycosides were isolated from those plants and characterized (Table 1).

As is seen from the data given in the table *U. dioica* green is sharply distinct by qualitative flavonoid composition from that of *U. dioica* red. Standard green specimens synthesized only flavonol derivatives - kaempferol and quercetin, but red specimens – flavonols and in addition anthocyans, derivatives of pelargonidin [Kavtaradze et al., 2001; Alania et al., 2002; Kavtaradze et al., 2003].

In order to establish the relation of chemical heterogeneity of material with its morphology we have carried out comparative study of macromorphological characteristics of researched plants (Table 2).

As is seen from the data given in the table specimens are distinguished by the level of pubescence, by the size and level of lignification of stem, by coloration of rhizome, stem, footstalk, rib and lamina.

While field investigations it was noted that red coloration of edges and nodes of rhizomes, from which shoots of stems, footstalks, ribs and lamina are grown, is persisted during the whole vegetation and generational period, which is the very feature that distinguishing it. There are some data in scientific literature that among *U. dioica* occur some specimens, which stems may have coloration caused by presence of anthocyanin pigments changing into brown coloration at the moment of florescence [Medvedev, 1934].

It was revealed that in West Georgia red and green specimens of *U. dioica* could grow together maintaining their characteristic morphology and qualitative chemical composition (Tables 1, 2).

Table 1. Flavonoids isolated from above-ground parts of *Urtica dioica*.

Specimens	Isolated compounds	Empiric formula	Melting temperature, °C	Literature
Green specimens	Flavonols Quercetin (3,5,7,3',4'-pentahydroxiflavon)	$C_{15}H_{10}O_7$	303-306	Kavtaradze et al., 2001
	Isoquercitrin (quercetin-3-O-β-D-glucopyranside)	$C_{21}H_{20}O_{12}$	221-224	" "
	Hyperin (quercetin-3-O-β-D-galactopyranoside)	$C_{21}H_{20}O_{12}$	232-235	" "
	Rutin (quercetin-3-O-β-D-rutinoside)	$C_{27}H_{30}O_{16}$	187-189	" "
	Kaempferol-3-O-tri-galactoside	$C_{33}H_{40}O_{20}$	-	Alania et al., 2004
Red specimens	Anthocyans Pelargonidin-3-xyloside	$C_{20}H_{20}O_9$	260 (with decomposition)	Kavtaradze, Alania, 2003
	Pelargonidin-3-xylobioside	$C_{25}H_{28}O_{14}$	170 (with decomposition)	" "
	Pelargonidin-3-gluco-galactoside	-	-	" "
	Flavonols Nicotiflorin (kaempferol-3-0-β-D-rutinoside)	$C_{27}H_{28}O_{16}$	178-180	Alania et al., 2002
	Rutin (quercetin-3-O-β-D-rutinoside)	$C_{27}H_{30}O_{16}$	187-189	" "

Table 2. Comparative morphological characteristics of *Urtica dioica* L. specimens

Organs and the main morphological features		Characterization	
		Green specimens [Medvedev, 1934]	Red specimens
Rhizome	Form	Tetraquetrous, nodular	Tetraquetrous, nodular
	Coloration	Yellow (sapling) Brownish (mature)	Orange-yellow Edges and nodes of rhizome are red (both, saplings and mature ones)
	Pubescence	Not pubescent	Not pubescent
Stem	Form	Tetraquetrous, rather drooping, thin;	Tetraquetrous, rather drooping, thick
	Height	0.5-1.0 m, in average	1.2-1.8 m, in average
	Butt diameter	5-10 mm, in average	10-17 mm, in average
	Coloration	Green	Red
	Pubescence per 1 cm of the length Number of stinging hairs	10-15	25-35
	Number of usual hairs	300-340	430-460
Leaves	Form and average sizes	Bottom – cordiform-elongated, sharpened; Middle – cordiform-lancet. Upper – nearly lancet (male plants), spear-shaped (mother plants)	Bottom – cordiform-elongated, sharpened; Middle – cordiform-lancet. Upper – nearly lancet (male plants), spear-shaped (mother plants)
	Leaf edges of the bottom layers	Crenate, coarse-toothed	Crenate, coarse-toothed
	Leaf edges of the upper layers	Serrate	Serrate
	Leaf edges of the lateral shoots	Serrate	Serrate
	Coloration of the upper side	Succulent green or dark green	Succulent green with dark reddish coloration
	Coloration of the bottom side	Lighter compared to upper side of lamina	Saplings –succulent red Mature leaves - lighter
	Number of the main ribs	3	3
	Coloration of the ribs	Green	Red
	Pubescence per 1 cm Number of stinging hairs	15-22	38-46
	Number of usual hairs	370-395	525-540
Footstalk	Form	Round, from above - with deep sulcus lengthwise	Round, from above - with deep sulcus lengthwise
	Coloration	Green	Red during the whole vegetative and generational period
	Pubescence per 1 cm Number of stinging hairs	20-55	35-70
	Number of usual hairs	260-380	450-690

Thus, results of chemical analysis and comparative morphological study show morphological and chemical intraspecies heterogeneity of studied specimens of *U. dioica*. Hence,

the plant with red coloration is distinguished as a variety – *Urtica dioica* L., var. rubescens Gviniaschvili et Kavtaradze – planta cum anthocyanea (Georgia, Samegrelo, Khobi district, vil. Alioni, 07.08.2002, near private farm).

References:

Alania M.D., Gvazava L.N., et al. *Phenolic compounds of some plant species growing in Georgia.* Abstracts of the VI symposium on phenolic compounds. Moscow, p. 95, 2004 (in Russian).

Alania M.D., Kemertelidze E.P., Komissarenko H.F. *Flavonoids of some species of Astragalus L. of Georgian flora.* Tbilisi, 2002 (in Russian).

Alania M., Gvazava L., et al. *Polyphenol and cycloartane compounds from some genera of plants of Georgian flora.* Abstracts of the 1st International Pharmaceutical Congress. Tbilisi, p.4, 2002.

Budantsev A.L., Fokina G.A. *Urticaceae Family.* In: Plant resources of USSR: flowering plants, their chemical composition and usage. Leningrad, p. 137-140, 1985 (in Russian).

Chaurasia N., Wichtl M. *Flavonoglycoside aus U. dioica.* Planta Med., **53**, 432-434, 1987.

Gide-Book Vidal. Medicinal Preparations in Russia. Loviisa, "Prostavern Urtica Likvid", p. 710, 2002.

Kavtaradze N.Sh., Alania M.D., Aneli J.N. *Chemical components of U. dioica growing in Georgia.* Chemistry of Natural Compounds, **3**, p.244, 2001 (in Russian).

Kavtaradze N.Sh., Alania M.D. *Anthocyan glycosides of U. dioica.* Chemistry of Natural compounds, **3**, p. 249, 2003 (in Russian).

Kraus R., Spitteler G. *Phenolic compounds from roots of U. dioica.* Phytochem., **29**, 5, 1653-1659, 1990.

Marrberley D.J. *The Plant-Book.* Cambridge University Press, p.55, 1998.

Medvedev P.F. *Utrica of USSR.* Leningrad, 1934 (in Russian).

Shkhian A.S. *Flora of Georgia.* Tbilisi, p.84-85, 1975 (in Georgian).

Wichtl M. *Brennesselblatter. DAB-Kommentar. Wesenschaftliche Erlauterungen zum Deutschen Arzneibuch. Monographin A-C.* Stuttgart-Frankfurt a.m./Eschborn, Bd.II/1, 1996.

საქართველოში გავრცელებული *Urtica dioica* L.-ს შიდასახეობრივი ქიმიური დიფერენციაცია

ქავთარაძე ნ., ღვინიაშვილი ც., ალანია მ., კუჭუხიძე ჯ.

ი. ჭუთათელაძის ფარმაკოქიმიის ინსტიტუტი

(მიღებულია 15.05.2006)

რეზიუმე

შესწავლილია საქართველოს სხვადასხვა რაიონში გავრცელებული *U. dioica*–ის ქიმიური და მორფოლოგიური თავისებურებები. გამოვლენილია *U. dioica* – ის ახალი სახესხვაობა წითელი შეფერილობით - *Urtica dioica* L. var. rubescens Gviniaschvili et Kavtaradze.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Botany

ENDEMIC MEDICINAL PLANTS OF KHEVI (KAZBEGI REGION, GEORGIA)

KHAIKASHVILI KH.

Tbilisi Botanic Garden

(Received August 29, 2006)

Abstract

The paper deals with results of inventory of 18 endemic medicinal plant species, concerning their number, abundance, frequency, and life forms. Degree of threats with extinction of such rare species as *Thymus collinus, Valeriana cardanines* and *V. tilifolia* is in relation to their collection by local population.

Key words: endemic medicinal plants, endangered species, Khevi region, Georgia.

Introduction

Kazbegi region is located on the highest part of the Great Caucasus. Due to contrasting orographic, climate and edaphic conditions the region is characterized by its florocoenotic diversity and richness in endemic species. Thus, 26% of angiosperms of the Khevi flora are endemic, including 6 out of 11 Caucasian endemic genera.

Total number of endemic plants in Khevi flora accounts for 247 species, i.e. 51,1% of high-mountain endemic flora of the Greater Caucasus [Shetekauri, Gagnidze, 2000; Nakhutsrishvili et al., 2005].

It should be noted that among endemic plants of Khevi about 20 species are considered to be used in traditional and officinal medicine from which two species, *Betula raddeana* and *Senecio rhombifolius*, are included in Red Data Book of Georgia (1982). *B. raddeana* is also included in RDB of the former USSR (1984), and IUCN Red List (1997, 2000).

Materials and Methods

Inventory of the abundance of species was done according to Drude's 6-point scale [Drude, 1890]. Frequency has been evaluated after Braun-Blanquet (1951). Spectra of life forms were identified according to Raunkier (1934). Plot size for herbaceous plant was $10m^2$, for woody species - $20m^2$. Experimental plots were chosen randomly within the area of population investigated.

Results and Discussion

6 species (*Betula raddeana* Trautv., *Sorbus caucasigena* Kom. ex Gatsch., *Rosa buschiana* Chrshan., *R. didoensis* Boiss., *R. galuschkoi* Demurova, *R. oplisthes* Boiss.), out of

investigated 18 endemic medicinal plants, belong to woody plants. One species (*Thymus collinus* Bieb.) is semishrub. Herbaceous plants are represented by one bienial species, *Heracleum asperum* (Hoffm.) Bieb. and 10 perennial ones: *Cephalaria gigantea* (Ledeb.) Born., *Galanthus platyphyllus* Traub & Moldenke, *Galega orientalis* Lam., *Potentilla agrimonioides* Bieb., *P. caucasica* Juz., *Senecio rhombifolius* (Adams) Sch. Bip., *Thalictrum buschianum* Kem.-Nath., *Trifolium fontanum* Bobr., *Valeriana cardamines* Bieb., *V. tiliifolia* Troitzk.

The data on species inventory for plants under consideration are given in the following table.

Table. Data on medicinal plant species inventory for 10 m^2 and 20m^2 plots

Plants species	Elev.	Ex.	Steep.	Num.	Abun.	Freq.	Life forms
Betula raddeana	2000	N	45	4	Cop_2	2	Mezophanerophyte
Cephalaria gigantea	1700	-	15	8	Cop_2	1	Hemicryptophyte
Galanthus platyphyllus *	2350	E	20	41	Sp	4	Cryptophyte
Galega orientalis	2000	N	45	9	Cop_1	1	Hemicryptophyte
Heracleum asperum	1600	-	-	11	Cop_2	2	Hemicryptophyte
Potentilla agrimonioides	1800	S	40	-	Cop_2	2	Hemicryptophyte
P. caucasica	1700	-	-	16	Cop_2	2	Hemicryptophyte
Rosa buschiana	2000	N	40	3	Cop_1	2	Nanophanerophyte
R. didoensis	1700	W	20	-	Cop_1	2	Nanophanerophyte
R. galushkoi	1600	-	-	-	Cop_2	2	Nanophanerophyte
R. oplisthes	1900	N	45	-	Cop_2	2	Microphanerophyte
Senecio rhombifolius	1900	N	30	5	Sp	1	Hemicryptophyte
Sorbus caucasigena	2000	E	45	4	Cop_2	2	Microphanerophyte
Thalictrum buschianum	1900	E	40	12	Cop_2	2	Hemicryptophyte
Thymus collinus	1700	E	40	-	Cop_2	3	Chamaephytes
Trifolium fontanum	1700	-	-	-	Cop_2	3	Hemicryptophyte
Valeriana cardamines	2000	N	45	-	Cop_2	1	Hemicryptophyte
V. tiliifolia	2000	N	40	7	Cop_2	1	Hemicryptophyte

Abbreviations and sign: Elev. - Elevation m a.s.l., Ex. - Exposition, Steep. - Steepness in ^{0}, Num. - Number, Abun. - Abundance after Drude (1890), Freq. - Frequency after Braun-Blanquet (1951), Life forms after Raunkier (1934), *Endemic species of Georgia

As is shown in the table *Galanthus platyphyllus* growing on alpine meadows is very rare species. In spite of its rarity in Khevi region it is not utilized as a medicinal plants. Consequently this species is not under threat. According to Miller and others [Miller et al., 2006] *G. platyphyllus* is considered as IUCN Vulnerable category (VU) species.

Next rare endemic medicinal plant in the Khevi region is *Senecio rhombifolius* growing in subalpine tall herbaceous vegetation. Unlike other regions of Georgia in Khevi this wellknown and utilized medicinal plant is not collected for medicinal purposes. Consequently, in spite of scarce resources of this species it is not under threat in this region.

Comparatively abundant species is *Galega orientalis.* In Khevi it occurs in subalpine forests and forest margins, and not utilized by local population for medicinal purposes.

Rosa buschiana occupy dry rock and scree habitats in restricted area in subalpine and alpine belts. Next species, *R. didoensis* occurs in the forest margins and shrublands of upper mountain forests and subalpine belts. It must be noted that only fruits are used for medicinal purpose.

Valeriana cardamines and *V. tiliifolia* are more widely distributed and sufficiently abundant species. Their roots are intensively collected by local population for medicinal purpose. Consequently these species are under serious threat.

Thymus collinus is utilized by local population as medicinal and spice means. Consequently, its resources are gradually diminished.

As a result of cutting number of *Betula raddeana* as well as other components of subalpine forests, viz., *B.litwinowii, B. pendula, Acer trautvetteri* are also little by little diminished.

It is concluded that to maintain endangered medicinal and other species special protective measures need to be taken including strengthening the regime of Kazbegi State Reserve on that part of territory where medicinal plants are represented.

References:

Braun-Blanquet, J. *Planzensoziologie*, 2-e aufl. Wien, 1951, (After Jaroshenko P.A. Geobotany, Moscow, Leningrad, 1961 (in Russ.).

Drude, O. *Handbuch der Pflanzengeographie*, Stuttgart, 1890, (After Jaroshenko P.A. Geobotany, Moscow, Leningrad, 1961 (in Russ.).

IUCN Red List of Threatened Plants. Eddited by K. S. Walter, H.J. Cillet, World Conservation Monitoring Centre, 1997.

IUCN Red List of Threatened Species. Compiled by C. Hilton-Taylor, IUCN, Gland, Switzerland and Cambridge, UK, 2000.

Miller J.S., McCue K., Consiglio T., Stone J., Eristavi M., Sikharulidze Sh., Mikatadze-Pantsulaia Ts., Khutsishvili M. *Endemic medicinal plants of Georgia (Caucasus)*. Tbilisi, 2006.

Nakhutsrishvili G., Abdaladze O., Kikodze A. *Khevi, Kazbegi Region.* Tbilisi, 2005.

Raunkier C. *Life forms of plants and statistical plant geography.* N.Y., 1934 (After Jaroshenko P.A. Geobotany, Moscow, Leningrad, 1961 (in Russ.).

Red Data Book of Georgia. Tbilisi, 1982 (in Georg.).

Red Data Book of USSR, Moscow, 1984 (in Russ.).

Shetekauri Sh., Gagnidze R. *Diversity of high-mountain endemic flora of the Greater Caucasus.* In: Biological and landscape diversity of Georgia. Tbilisi, 151-158, 2000 (in Georg.).

ხეშო (ყაზბეგის რაიონი) გავრცელებული ენდემური სამკურნალო მცენარეები

ხაიკაშვილი ხ.

თბილისის ბოტანიკური ბაღი

(მიღებულია 29.08.2006)

რეზიუმე

სტატია ეხება ხეშის ფლორის 18 სამკურნალო ენდემურ სახეობას. მოცემულია მათი რაოდენობა, სიხშირე, შეხვედრილობა, სახიცოცხლო ფორმების სპექტრი. გამოვლენილია გადაშენების საფრთხის ქვეშ მყოფი ენდემური სახეობები.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Botany

ANALYSIS OF BRYOPSIDA SPECIES ACCORDING TO THE OCCURRENCE FREQUENCY IN THE FOREST BELT OF LAGODEKHI RESERVE

TIGISHVILI K.

N. Ketskhoveli Institute of Botany

(Received May 8, 2006)

Abstract

125 species of Bryopsida are divided into 5 groups according to the occurrence frequency. The highest mark, 5 points was conferred to the 6% of total number of mosses, 4 points - to the 19%, 3 points - to the 28%, 2 points- to the 30%, 1 point - to the 17 %. The obtained results show, that the species with medium and lower occurrence frequency comprise more than half of bryoflora of the lower forest belt. Widely spread species are characterized by the most limited occurrence frequency. Together with this, the comparison of systematical structure of bryoflora of the mentioned belt with the bryoflora of several regions of the East Trans-Caucasus is made on the example of 10 leading families.

Key words: East Trans-Caucasus, bryoflora, leading families.

Introduction

Mosses comprise a significant part of natural plant cover. Natural reserves are the objects for the protection of information resources and the investigation of untouched ecosystems in them is particularly important [Reimers, 1978].

The main objective of our investigation was thorough study of bryoflora of Lagodekhi Reserve. Revelation of the occurrence frequency of bryoflora is connected with many difficulties [Maslovski, 1991]. Despite this we made an attempt to solve the problem on the example of bryoflora of forest belt. The obtained results are of preliminary character.

Materials and Methods

Bryoflora of main ecosystems of forest belt of Lagodekhi Reserve has been studied using itinerary and semistationary methods. Material was taken and treated on sample plots according to the geobotanical method [Neshataev, 1987].

Results and Discussion

On the basis of rich bryological material, obtained in the forest belt of Lagodekhi Reserve (1200 samples) we made an attempt to establish the occurrence frequency of Bryopsida species. According to this feature Bryopsida species of the mentioned region have been divided into 5 main

groups: 1. species, spread in the most of coenoses and ecotopes, characterized with constant occurrence - 5 points; 2. species, which are not spread in the majority of coenoses and ecotopes, but are distinguished with rather high occurrence frequency in the main formations and the most significant ecotopes - 4 points; 3. species, which do not have wide distribution, but are distinguished by the certain occurrence frequency in some ecotopes - 3 points; 4. species, characterized by sporadic distribution and low frequency of occurrence - 2 points; 5. extremely rare species - 1.

Analysis, performed in order to reveal the frequency of occurrence of moss species has shown, that the highest evaluation was conferred to 6% of the total number of bryoflora species or to only 9 species. The following taxons: *Hypnum cupressiforme*, *Leucodon* spec., *Brachythecium rutabulum*, *Brachythecium populeum*, *Neckera besseri*, *Anomodon attenuatus* are characterized by wide distribution and constant frequency of occurrence. The 17 % of the total number of bryoflora species were evaluated by 1 point. Despite the fact, that the material was taken several times, some of these taxons ware found only once. The species *Pottia truncata*, *Phasum cuspidatum*, *Pseudoscleropodium purum*, *Breidleria arcuata*, *Pleurochaete squarrosa* belong to rare species.

The rest species, comprising the bryoflora of forest belt, are positioned between these two extreme groups. The taxons, evaluated by 4 points are not everytypes of the lower forest belt, but are widely spread in favourable econiches. Their number makes 19% of the total number of species. Sometimes, within the limits of synusia, they are distinguished by significant development of the biomass. The epilyths, spread in humid forests of river ravines: *Thamnium alopecurum*, *Ctenidium molluscum*, *Mnium undulatum,* the species, characteristic to Fagetum nudum: *Isothecium myurum*, *Pterigynandrum filiforme;* and *Thuidium philibertii*, characteristic to hornbeam forests and some others belong to such species.

The species, evaluated by 3 and 4 points comprise 30% and 28% of the main list. Taken separately they exceed the number of taxons of other groups and together they make more than half of the bryoflora species. Species, evaluated by 3 points, found only in some coenoses and ecotopes, are represented by the following taxons: *Eurhynchium striatum*, *Euryinchium zetterstedtii*, *Rhynchostegium riparioides*, *Mnium stellare*, *Orthorhichum diaphanum* and others. Distribution of 2-point species is rather limited, though their total number reaches 42. *Weisia controversa*, *Mnium selligeri*, *Bryum bicolor* and others can be listed among such species.

Based on the mentioned data it can be supposed, that bryoflora of the lower forest belt of Lagodekhi Reserve is mainly presented by the species of medium and low frequency of occurrence. The quantity of species of high frequency of occurrence is the most limited. Despite the wide ecological amplitude of mosses, narrow ecological conditions of the environment are the main factors, determining their distribution. Our data are in agreement with those presented in the scientific literature [Maslovski, 1991].

Taxonomic structure of mosses allows to establish the pattern of bryoflora for the mentioned region. Acoording to Tolmachev [Tolmachev, 1974] the pattern of bryoflora is reflected in the best way by the specific composition of ten leading families, which hold the dominant position in bryoflora by the number of species. Composition of bryoflora of the studied region, presented by the leading 10 families is as follows: **Brachytheciaceae** - 22 species, **Mniaceae** - 14 species, **Amblystegiaceae** - 15 species, **Bryaceae** - 13 species, **Pottiaceae** - 11 species, **Dicranaceae** - 9 species, **Grimmiaceae** - 8 species, **Thuidiaceae** - 5 species, **Hypnaceae** - 6 species, **Trichostomaceae** - 4 species. The 104 species or more than half of total bryoflora are united in 10 leading families. The zonal-floristic peculiarities of the region are well reflected by systematical composition of bryoflora. The presence of **Brachytheciaceae**, **Mniaceae** and **Amblystegiaceae** among leading families points to the prevalence of forest landscapes in the mentioned region and its mesophilic character. The families **Grimmiaceae** and **Dicranaceae** serve as the evidence for its mountainous relief. Despite the physicogeographical and vegetative

contrasts, characteristic to the Caucasus, we made an attempt to compare bryoflora of forest belt of Lagodekhi Reserve with that of some regions of East Trans -Caucasus [Chikovani, 1965; Lubarskaya, 1974; Manakian, 1989] (Table 1). It turned out, that 8 families are common for 10 leading families of all regions. This points to the common botanical-geographical character of the mentioned regions.

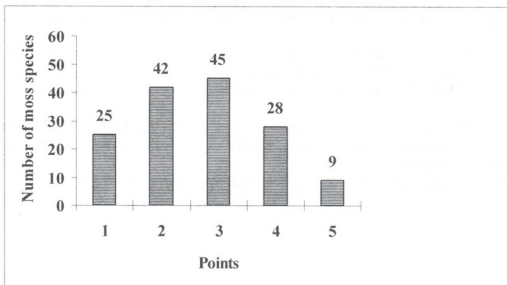


Fig. 1. Groups of Bryopsida species according to the frequency of occurrence.
1 point - 25 species (17% of total); 2 points - 42 species (28% of total); 3 points 45 species (45% of total); 4 points - 28 species (19% of total); 5 points 9 species (6% of total)

Table 1. Comparison of Lagodekhi Reserve forest belt bryoflora with that of some regions of East Trans-Caucasus.

Moss	Lagodekhi reserve		Nukha-Zaqatala		Gombori		NE Armenia		SE Armenia	
Family	Species number	Posi-tion	Species number	Position	Species number	Position	Species number	Position	Species number	Position
Brachytheciaceae	22	1	20	1	13	2	4	7	19	1
Mniaceae	14	3	6	8	6	7	4	8	5	8
Bryaceae	13	4	8	5	7	6	10	2	10	3
Amblystegiaceae	15	2	10	2	9	5	4	9	8	7
Dicranaceae	9	6	5	9	4	10	3	10	-	-
Grimmiaceae	8	7	-	-	5	8	5	5	9	6
Orthotrichaceae	7	-	-	-	10	4	6	4	10	4
Pottiaceae	11	5	9	4	14	-	7	3	9	5
Hypnaceae	6	8	10	3	-	-	-	-	4	9
Thuidiaceae	5	9	7	6	4	9	5	6	3	10
Trichostomaceae	4	10	6	7	13	3	11	1	11	2
Neckeraceae	-	-	5	10	-	-	-	-	-	-

References:

Chikovani N. *Checklist of bryophyte species of Georgia.* Braun-Blanquetia, vol. **34**, Camerino, 2004.

Lubarskaya L. B. *Bryopsida mosses of South-East Part of the Main Caucasus (Azerbaijan).* pp. 180-200, Baku, 1989 (in Russian).

Maslovski O. M. *Analysis of bryoflorotopological structure of Bryoflora complexes of Byelorussian Depression.* In: Advances and Perspectives in Bryology, L'vov, pp. 147-148, 1991 (in Russian).

Manakyan V. A. *Bryopsida of the Main Forest Massifs of Armenian Forest.* Collection of Works of the Armenian Academy of Sciences, **11**, Erevan, 1988.

Neshataev U. N. *Methods of Analysis of Geobotanical Material.* Leningrad, pp.53-98, 1984 (in Russian).

Reimers N. F. *Particularly Protected Natural Territories.* pp. 25-30, Moscow, 1978 (in Russian).

Tigishvili K. T. *Bryoflora of Lagodekhi State Reserve.* Report of the Project, Manuscript, Tbilisi, 1988 (in Georgian).

Tolmachev L. I. *Introduction to the Geography of Plants.* Leningrad, 243 p., 1974.

ღეროფოთლოვანი ხავსების ანალიზი შეხვედრილობის სიხშირის მიხედვით ლაგოდეხის ნაკრძლის ტყის სარტყელში

ტ. ციციშვილი

ნ. კეცხოველის ბოტანიკის ინსტიტუტი

(მიღებულია 08.05.2006)

რეზიუმე

ლაგოდეხის ნაკრძლის ტყის სარტყლის 125 სახეობის ღეროფოთლოვანი ხავსი, შეხვედრილობის სიხშირის მიხედვით, დაყოფილია 5 ჯგუფად. უმაღლესი შეფასება, 5 ბალი, მიიღო ხავსების საერთო რაოდენობის 6%, 4 ბალი - 19%, 3 ბალი - 28%, 2 ბალი - 30%, 1 ბალი -17%. მიღებული მონაცემებიდან ჩანს, რომ ტყის სარტყლის ბრიოფლორის ნახევარზე მეტი წარმოდგენილია საშუალო და უფრო დაბალი შეხვედრილობის სიხშირის მქონე სახეობებით. ფართოდ გავრცელებული სახეობების შეხვედრილობის სიხშირე კი ყველაზე მეტად არის შეზღუდული. აღნიშნული სარტყლის ბრიოფლორის სისტემატიკური სტრუქტურა, 10 წამყვანი ოჯახის მაგალითზე, შედარებულია აღმოსავლეთ ამერიკავკასიის ზოგიერთი რეგიონის ბრიოფლორასთან.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Ecology

THE ORIBATID MITES (ACARI, ORIBATIDA) OF GOMBORI RIDGE BEECH FOREST

ARABULI T.

Institute of Zoology, Chavchavadze ave. 31, 0179, Tbilisi, Georgia

(Received September 20, 2006)

Abstract

119 species of oribatid mites were registered in Gombori Ridge beech forests. Among them 3 species: *Phthiracarus balogi* (Feider, Suciu, 1957), *Tricheremaeus pilosus* Michael, 1988, and *Suctobelba granulate* Hammer, 1952, were recorded for the first time for Georgian fauna. According to faunal likeness distinct groupings of oribatid mites were formed, which is stipulated by distribution of beech forests at various heights. Fauna of Oribatid mites of soil of beech forests is richer than fauna of moss. Changes of number dynamics of oribatids are mutually opposed in those bitopes: while the number is high in moss, it decreases in soil, and visa versa, which is caused by temperature and moisture changes in ecosystem and migration ability of oribatids.

Introduction

Plant cover of Gombori Ridge is characterized with wide diversity and complicated floristic composition, which is caused by its ecological past. 9 zonal types are distinguished across the ridge [Sakhokia, 1960]. The basic component of Fagus forest is beech; beech forests represented by deadcovering and not evergreen subforests prevail. They occur on north exposition, forest edge at the west part of forest is relatively descended and begins from 800-900 m, and at the south exposition, where influence of dryness is higher it begins from 1000 m. On north exposition of south slopes, in humid places the beech comes to 900 m.

The number dynamics of oribatid mites in Georgia has been studied [Darejanashvili, 1965; Murvanidze, 1999; Arabuli, 2003], but number dynamics of oribatid mites inhabited simultaneously in two different stations was not studied yet. Hence, the goal of our work was to study number dynamics of oribatid mites of soil and moss of deadcovering beech forest.

Material and Methods

The material was collected during September 2001 – November 2003. Beech forests of both, south-west and north-east slopes were investigated, particularly: 1. deadcovering beech forest on Akhmeta-Tianeti pass (the utmost north-east of the ridge, near mount Shakhvetila), 2. deadcovering beech forest in Nagubrebi (on north exposition, village Tetrtsklebi), 3. beech forest with azalea subforest in central part of Gombori Ridge (Mount Tsivi, north exposition), 4. deadcovering beech forest (on north exposition of mount Tsivi), and 5. beech forest with deadcovering in Mariamjvari reserve (Table 1.).

Material was collected and worked out according to received methods of soil zoology [Krivolutski, 1973]. At each site 3 samples of soil and moss of 10 cm^3 area were taken. Extracted mites were fixed and temporary preparations were made [Balogh & Mahunka, 1983]. Mite number was counted on 1 m^2. Identification of mites was carried out by special guide-books.

The coefficient of faunal likeness was calculated by Jaccard's formula and the cluster was constructed according to the known method [Krebs, 1989] (Fig.1).

Results and Discussion

119 species united in 65 genera, 44 families and 24 upper families were registered in Gombori Ridge beech forests. Among them 3 species: *Phthiracarus balogi* (Feider, Suciu, 1957), *Tricheremaeus pilosus* Michael, 1988, and *Suctobelba granulate* Hammer, 1952, were recorded for the first time for Georgian fauna.

Our studies have shown that by species diversity of oribatid mites the soil is richer than moss. Among 119 species revealed in beech forests 100 ones were registered in soil, and 54 species – in moss. Characteristic species for every biotope were also studied; it was registered: 65 characteristic species in soil, and 10 – in moss, 44 - common for soil and moss (Table 1).

Table 1. Oribatid mites of Gombori Ridge beech forests

N	species	moss	soil					
			station	single samples				
				1	2	3	4	5
1	*Liochthonius lapponicus* (Tragardh, 1910,)		+					
2	*Hypochthonius rufulus* C.L. Koch, 1835		+					
3	*Hypochthoniella minutissima* (Berlese, 1904)		+	+				
4	*Mesoplophora pulchra* Sellnick, 1928		+		+			
5	*Epilohmannia gigantea* Berlese, 1917		+					
6	*Hoplophthiracarus vanderhammeni* Nied, 1991	+	+	+		+	+	+
7	*Phthiracarus ferrugineus* (C. L. Koch, 1841)	+	+	+		+	+	
8	*Phth. globosus* (C. L. Koch, 1841)		+				+	+
9	*Steganacarus csiszae* Balogh & Mahunka, 1979		+					
10	*St. striculus* (C. L. Koch, 1836)		+				+	
11	*St.serratus* (Feider & Suciu, 1957)		+					
12	*St. spinosus* (Sellnick, 1920)		+		+			
13	*St. (T) carinatus* (C. L. Koch, 1841)	+	+		+	+	+	+
14	*St. (T) phyllophorus* (Berlese, 1904)		+				+	
15	*St. balearicus* Perez-Inigo, 1969	+	+					
16	*St. bicarinatus* Jeleva,1970		+					
17	*Phthiracarus baloghi* (Feider, Suciu, 1957)		+					
18	*Archiphthiracarus. murphyi* (Harding, 1976)		+					
19	*A. lanatus* (Feider, Suciu, 1957)		+					
20	*A. ligneus* (Willmann, 1931)		+				+	+
21	*A.. clemens* (Aoki, 1963)		+					
22	*Rhysotritia ardua* (C.L. Koch, 1841)	+	+	+		+		+
23	*Oribotritia serrata* Feider et Suciu, 1958		+					
24	*Nothrus silvestris* Nicolet, 1855		+			+		
25	*Platynothrus grandjeani* Sitnikova, 1975		+					
26	*Camisia horrida* (Hermann, 1804)	+						
27	*Nanhermannia nana* (Nicolet, 1855)					+		
28	*Hermanniella granulata* (Nicolet, 1855)	+	+	+				

29	*H. punctulata* Berlese, 1908		+	+				
30	*Liodes theleproctus* (Hermann, 1804)	+						
31	*Arthrodamaeus femoratus* (C. L. Koch, 1840)	+	+	+				
32	*Metabelba filippova* Bul.-Zachvatkina, 1965					+		
33	*M. flagelliseta* Bulanova-Zachvatkina, 1965		+	+			+	
34	*M. pulverulenta* (C. L. Koch, 1839)	+	+			+		
35	*Metabelbella macerochaeta* Bul-Zach, 1967					+		
36	*Eupterotegeus ornatissimus* (Berlese, 1908)		+				+	
37	*Amerus troisii* (Berlese, 1883)		+					
38	*Amerobelba decedens* Berlese, 1908		+					
39	*Damaeolus ornatissimus* Csiszar, 1962	+	+				+	
40	*Eremobelba geographica* Berlese, 1908			+		+		
41	*Eremaeus hepaticus* C. L. Koch, 1836	+	+	+		+	+	
42	*E. oblongus* C. L. Koch, 1836	+	+					
43	*E. triglavensis* Tarman, 1958	+						
44	*Tricheremaeus pilosus* Michael, 1888		+					
45	*Zetorchestes microtychus* (Berlese, 1883)		+					
46	*Cultoribula bicultrata* Berlese, 1908		+					
47	*Gustavia microcephala* (Nicolet, 1855)		+					
48	*Adoristes ovatus* (C.L. Koch, 1840)		+					
49	*Liacarus brevilamellatus* Mihelcic, 1955						+	
50	*L. coracinus* (C. L. Koch, 1840)	+	+					
51	*L. tubifer* Djaparidze & Melamud, 1990		+	+		+		
52	*L. lencoranicus* Krivolutsky, 1967		+					
53	*Ceratoppia bipilis* (Hermann, 1804)	+		+				
54	*C. quadridentata* (Haller, 1882)	+						
55	*Carabodes femoralis* (Nicolet, 1855)	+	+					
56	*C. rugosior* Berlese, 1916		+			+	+	+
57	*C. procerus* Weigmann & Murvanidze 2003					+		
58	*Tectocepheus punctulatus* Djaparidze, 1985	+	+		+			
59	*T. sarekensis* (Tragardh, 1910)	+	+					
60	*T. velatus* (Michael, 1880)	+	+	+	+	+		+
61	*Berniniella bicarinata* Paoli, 1908						+	
62	*B. conjucta* (Strenzke, 1951)		+					
63	*B. exempta* (Mihelcic, 1959)		+	+				
64	*B. sigma* (Strenzke, 1951)	+	+	+				
65	*Microppia minus* (Paoli, 1908)		+					
66	*Oppiella maritima* (Willmann, 1928)		+					
67	*O. nasuta* (Moritz, 1965)		+			+		
68	*O. nova* (Oudemans, 1902)	+	+	+	+	+	+	+
69	*O. (R) hygrophila* (Mahunka, 1987)		+					
70	*O. obsoleta* (Paoli, 1908)		+					
71	*O. (R) simifallax* (Subias & Minguez, 1986)						+	
72	*O. (R) subpectinata* (Oudemans, 1900)	+	+	+	+	+	+	+
73	*O. unicarinata* (Paoli, 1908)	+						
74	*Oxyoppioides decipiens* (Paoli, 1908)	+	+					
75	*Ramusella insculpta* (Paoli, 1908)	+	+		+		+	
76	*R. mihelcici* (Perez-Inigo, 1964)			+				
77	*Quadroppia michaeli*, Mahunka, 1977	+	+		+	+		
78	*Q. quadricarinata* (Michael, 1885)	+	+					
79	*Suctobelba granulata* Hammer,1952		+		+	+		

80	*S. trigona* (Michael, 1888)	+	+					
81	*Suctobelbella acutidens* (Forsslund, 1941)	+	+					
82	*S. duplex* (Strenzke, 1950)	+	+					
83	*S. subcornigera* (Forsslund, 1941)		+				+	
84	*Banksinoma lanceolata* (Michael, 1888)					+		
85	*Cymbaeremaeus cymba* (Nicolet, 1885)	+	+			+		
86	*Eupelops acromios* (Hermann, 1804)		+					
87	*E. plicatus* (C. L. Koch, 1836)		+		+			
88	*E. torulosus* (C. L. Koch, 1840)	+		+		+		
89	*Achipteria coleoptrata* (Linne, 1746)		+					
90	*A. nitens* (Nicolet, 1855)	+						
91	*Parachipteria georgica* Murv., Weigm., 2003	+	+	+	+	+		
92	*P. punctata* (Nicolet, 1855)	+	+				+	
93	*P. nicoleti* (Berlese, 1883)						+	+
94	*Umbellozete fuscus* Krivolutsky, 1969	+	+	+		+		+
95	*Acrogalumna longipluma* (Berlese, 1904)		+					
96	*Pilogalumna tenuiclava* (Berlese, 1908)	+	+				+	+
97	*Ceratozetella sellnicki* (Rajski, 1958)	+	+					
98	*Ceratozetes gracilis* (Michael, 1884)	+	+	+	+	+	+	+
99	*C. laticuspidatus* Menke, 1964		+					
100	*C. longicuspidatus* Kulijev, 1962		+					
101	*C. mediocris* Berlese, 1908		+					
102	*Sphaerozetes piriformis* (Nicolet, 1855)	+	+					
103	*Trichoribates trimaculatus* (C.L.Koch, 1836)	+						
104	*T. caucasicus* Shaldybina, 1971	+						
105	*Chamobates caucasicus* Shaldybina, 1969			+				
106	*Ch. interpositus* Pschorn-Walcher, 1953	+						
107	*Ch. voigtsi* (Oudemans, 1902)		+					
108	*Euzetes globosus* (Nicolet, 1855)					+		
109	*Minunthozetes pseudofusiger* (Schwc, 1922)	+	+	+	+	+	+	
110	*Mycobates parmeliae* (Michael, 1884)	+						
111	*M. tridactylus* Willmann, 1929	+	+					
112	*Punctoribates punctum* (C. L. Koch, 1893)	+	+					
113	*Protoribates capucinus* (Berlese, 1908)	+	+					
114	*P. pannonicus* Willmann, 1951					+		
115	*Scheloribates laevigatus* (C. L. Koch, 1836)	+	+	+				
116	*Sch. latipes* (C. L. Koch, 1840)		+					
117	*Oribatula tibialis* (Nicolet, 1855)	+	+	+				+
118	*Phaulopi saakadzei* Djaparidze, 1985	+	+					
119	*Zygoribatula exilis* (Nicolet, 1855)	+	+		+			
	number of species	54	93	26	15	29	24	14

Beech forest with azalea subforest was distinguished by oribatid mite species diversity, where 29 species were recorded, beech forest with deadcovering (Akhmeta-Tianeti pass) – 26 species, and beech forest with deadcovering on north exposition of mount Tsivi – 24 species. It should be mentioned that beech forest with azalea subforest is rich in by species number, as well as by characteristic species (7 characteristic species were registered), which should be caused by diversity of plant cover.

In spite of studying of more or less similar ecosystems, while calculating the faunal likeness of Gombori Ridge oribatid mites, three distinct groupings have been formed (Fig. 1): the highest coefficient of likeness (35%) was noted between deadcovering beech forest located on

north exposition of mount Tsivi and Mariamjvari deadcovering beech forest, which is caused by location of those forests at the same heights. In spite of territorial distance, Akhmeta-Tianeti deadcovering beech forest and beech forest with azalea subforest of mount Tsivi located at higher regions of ridge were grouped together (34%), and beech forest of Nagubrebi placed at lower part of the ridge was absolutely isolated from them. Formation of distinct groupings should be stipulated by special sensitivity of oribatid mites to environmental conditions. As it was mentioned above beech forests on Gombori Ridge begin at various heights due to humidity, which affects the faunal composition of oribatid mites.

Number dynamics was studied in soil and moss simultaneously during 26 months. In September 2001 number of mites in moss consist of 22 500 specimens/m^2, while in soil 5159spec/m^2 were registered. In October insignificant increase of mite number was noted both, in soil and moss, but in November with temperature decrease number of specimens in moss decreased significantly, and at the expense of this their number increased up to 13 657 spec/m^2 in soil, which is caused by migration of mites from the moss to the soil at the beginning of adverse weather conditions. In December mite number was decreased in both biotopes and winter minimums were recorded.

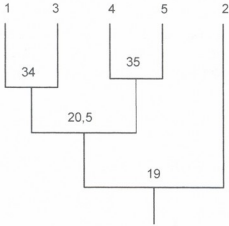


Fig.1. Cluster of faunal likeness of Oribatid mites in Fagus forest.

In January 2002 number of oribatid mites increased both, in soil and moss, and in February winter minimum (4 500 spec/m^2) was fixed in moss. In March and April together with the increase of humidity the minimal number was registered in soil. Just at the beginning of spring mite number increased in moss; increase was continued in April too, and in May maximum number was fixed – 37 000 spec/m^2. As for oribatid mites inhabited in soil, maximum number was revealed in June – 22 324 spec/m^2. In June and July, due to high temperature and dryness, mite number was decreased in moss, but in August it reached maximum – 41 500 spec/m^2.

In October 2002 the picture in soil and moss was contrary: in moss mite number minimum was recorded (4 500 spec/m^2), in soil – maximum (60 990 spec/m^2). In November situation appeared opposite: number increased up to 78 500 spec/m^2 in moss, but in soil – decreased to 16 999 spec/m^2.

In December 2002, as in 2001, number of oribatid mites decreased both in soil and moss, though mite number recorded in 2002 in soil was twice as much than that of moss.

In January 2003 the number of oribatid mites inhabited in soil increased, spring maximum (132 157 spec/m^2) was registered in March, but in moss - later, in May.

In spring 2003 soil mite number revealed considerably high, especially in July – 63 156 spec/m^2. In August and September and especially in October decrease of number was noted. As for oribatids inhabited in moss in June their number was grossly low. In the following months

intensive growth of their number was recorded and in September it reaches maximum – 337 000 spec/m^2.

Thus, as a result of our investigations it was found out that changes of number dynamics of oribatids are mutually opposed in soil and moss: while the number is high in moss, it decreases in soil, and visa versa, indicating the fact that oribatids respond rapidly to temperature and moisture changes and migrate actively towards optimal conditions.

References:

Arabuli T. *The quantiteitive dynamics of Oribatid Mites (Acari, Oribatei) in natural ecosystems (Hornbeam, (Carpinus caucasica Grossh) Forest).* Bulletin of the Georgian Academy of Sciences, **169**, 1, 169-171, 2004.

Balogh J. Mahunka S. *Primitiv Oribatids of the Palearctic region.* Akademiai Kiado. Budapest. p.372, 1983.

Darejanashvili Sh.D. *Seasonal and stational distribution of Acariformes, Oribatei in Tbilisi surroundings.* Bull.Acad.Sci.of Georgian SSR, **XXXVII**, 1, 175-179, 1965 (in Russian).

Krivolutski D.A. *Methods of complex investigation of soils on population density of Oribatid Mites.* In: Methods of Soil-Zoological Studies. Moscow, "Nauka", p. 44-48, 1973 (in Russian).

Krebs CH.J. *Ecological Methodology.* New York, Harper & Row Publisherrs, p.654, 1989.

Murvanidze M. *To the study of quantiteitive dynamics of Oribatid Mites (Acari, Oribatei) in urban conditions.* Bulletin of the Georgian Academy of Sciences, **160**, 2, 377-379, 1999.

გომბორის ქედის წიფლნარების ჯავშნიანი ტკიპები (Acari, Oribatida)

თ. არაბული

ზოოლოგიის ინსტიტუტი

(მიღებულია 20.09.2006)

რეზიუმე

გომბორის ქედზე წიფლნარების გამოკვლევის შედეგებად აღირიცხა ჯავშნიანი ტკიპების 119 სახეობა, რომელთაგან სამი: *Archiphthiracarus balogi, Tricheremaeus pilosus* და *Suctobelba granulata* პირველად იქნა რეგისტრირებული საქართველოს ფაუნისათვის. ჯავშნიანიან ტკიპებს შორის ფაუნისტური მსგავსების მიხედვით წარმოიქმნა განსხვავებული დაჯგუფებები, რაც განპირობებულია ქედზე წიფლნარების სხვადასხვა სიმაღლეზე გავრცელებით. დადგენილია, რომ წიფლნარების ნიადაგის ჯავშნიანი ტკიპების ფაუნა უფრო მდიდარია, ვიდრე ხავსის. ამ ორ ბიოტოპში ტკიპების რიცხოვნობის დინამიკა ურთიერთსაწინააღმდეგოდ იცვლება: როცა ტკიპების რიცხოვნობა მაღალია ხავსში, მაშინ მათი რაოდენობა დაბალია ნიადაგში და პირიქით, რაც ეკოსისტემაში ტენიანობის და ტემპერატურის ცვალებადობით და ორიბატიდების მიგრაციის უნარით არის განპირობებული.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Ecology

DYNAMICS OF CONDITION FACTOR OF VENDACE (*COREGONUS ALBULA* L.) IN THE LAKE PARAVANI

*JAPOSHVILI B.[1], KOKOSADZE T.[2], JAPOSHVILI O.[1]

[1] *Chavchavadze Tbilisi State Univeristy*
[2] *Institute of Zoology*
*Tel: (+ 995 99) 971827, *E-mail: bellaj99@yahoo.com*

(Received September 25, 2006)

Abstract

The dynamics of condition factor among both sexual groups of vendace has been studied for the first time. Condition factor of males and females were compared. The reason of differences is discussed. It was shown, that decrease of condition factor during last years is related with climate changes and global warming.

Key words: *Coregonus albula*, condition factor, Lake Paravani.

Introduction

Lake Paravani is the largest by its surface area among lakes of Georgia (37.5 km^2). It is situated in the Southern Part of Georgia on Javakheti upland on the 2080 m a.s.l. Volume of the lake is 90,8 mln m^3. Lake usually freezes in the second half of the December, while ice layer reaches its maximal thickness in March, very seldom it can be observed in the second half of February. In various years ice layer equaled to 47-73 cm, 80-90 cm, in very cold winter season it was even 1-1.2 m. Melting starts in the third decade of April. At the end of April or in the early May lake tends to be totally free from the ice cover [Barach, 1964, Apkhazava, 1975].

In 30s of 20th century vendace (*Coregonus albula* L.) was introduced in Paravani Lake from the Lagoda Lake (Volkhov hatchery). It was easily adapted to new environment and soon became object for commercial fishing [Demetrashvili, 1960, Japoshvili, 2002]. Data for condition factor for *Coregonus albula* is very poor and insufficient [Demetrashvili, 1960, Japoshvili, 2004, Kokhia, 1961, Peskova, 1960].

Materials and Methods

We have studied dynamics of condition factor for male and female *Coregonus albula* of Paravani Lake during 1999-2005. To calculate condition factor we have used Fulton's equation:

$K=(W/L^3)x100$

K is condition factor, W is the weight of the whole fish weight, L is total length of fish [Nikolskii, 1974, Murphy, Willis, 1996]

Age determinations were based on scales [Pravdin, 1966].

Results and Discussion

During the study males and females of vendace in Paravani Lake were represented by 4 age groups. Studies and calculations have shown that condition factor for females under the age group of 1+ reaches its peak in September. This indicator is increasing between May and September (from 0.65% to 0.81%), later on the indicator reduces and reaches the index observed in May (Fig. 1).

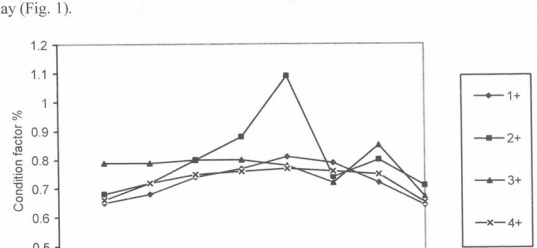


Fig. 1. Dynamics of condition factor of vendace females in Lake Paravani over the months.

In females under the age group of 2+, condition factor rises intensively and reaches its maximum in September -1.09%, while in October the curve is reduced and equals to 0.74%, however before the hatching it is risen a little bit achieving 0.80%, and falling again in December to 0.71%.

Female age group 3+ was observed in the period between May and September. They have shown condition factor of the same value approximately. It is well reflected on the curve that is almost linear. At the beginning of October coefficient equals to 0.72%, in November it reaches 0.85%, while in December it decreases to 0.67%.

In females of the age group 4+ condition factor tends to be 0.66% in May. In summer it rises to 0.77% and in December, following the hatching period the index decreases and equals to 0.65%.

Alteration of condition factor has been studied in males likewise (Fig. 2). Studies have shown that condition factor of males of age group 1+ reaches 0.58% in May, in July it rises to 0.78%, later on the value gradually falls down to 0.70% in September. In November the index increases for a while, equalling to 0.75% and decreases in December to 0.62%. In age group of 2+ coefficient is 0.60% in May. In summer period it increases dramatically and achieves to 0.84% in September. In October condition factor falls to 0.70%. At this period of time the curves are intercrossed for the age groups 1+ and 2+ in males. At the beginning of November it rises and reaches 0.78%, while in December it falls again and equals to 0.64%.

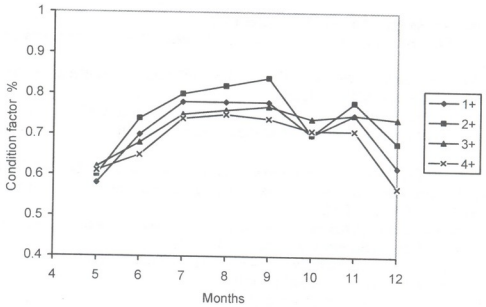


Fig. 2. Dynamics of condition factor of vendace males over the months in Lake Paravani.

Different results were found in males of the age group of 3+ and 4+. In both groups condition factor is rising from May to August. In the following months coefficient falls for the age group 4+, while 3+ age group preserves uniformity and the curve is almost linear.

Conclusions

Studies of females revealed that between May and August-September females of the age group of 1+ and 2+ are characterized with intensified diet. In addition in the age group 2+, before hatching, condition factor is decreased. It should be caused by falling of feeding rate due to the preparatory stage for hatching. This phenomenon for *Coregonus albula* is observed in other European lakes as well. In October-December species under the age group of 2+ and 3+ reflect similarity in nutrition curves caused by active involvement of those age groups in the hatching. Our studies have shown that in comparison with female species condition factor in males is less altered monthly. We suppose that condition factor is more exposed to seasonal changes due to generative synthesis in females.

We have recorded pretty low indices of the condition factor, which can be caused by several reasons, including: illegal catches intensified in the last period. As for discrepancy of our figures with previous data we suggest that differences are caused by altered terms of hatching, ice-cover formation, and warming of the lake as a result of global warming.

References:

Apkhazava I. *Lakes of Georgia.* Tbilisi, "Metsniereba", 1975.

Barach G. *Lakes of Georgia and their importance for fisheries.* Tbilisi, "Sabchota Sakartvelo", 1964.

Demetrashvili M. *For study of biology of lake Paravani vendace.* Bulletin of the Georgian Academy of sciences, **XXV**, N.1, 65-70, 1960.

Japoshvili B. *Results of visual observation on gonadogenesis of Vendace (Coregonus albula L.) in conditions of Paravani Lake.* Bull. of the Georgian Academy of sciences, **166**, 3, 591-594, 2002.

Japoshvili B. *Some Biological and morphometric characteristics of a vendace (Coregonus albul aL.) of the Lake Paravani.* Proceedings of the Georgian academy of Sciences, Biological series B, **2**, 1-2, 97-100, 2004.

Kokhia A. *Materials of diet of fishes of Paravani Lake.* Proceedings of the institute of zoology, Georgian Academy of Sciences, **XVIII,** 85-94, 1961.

Murphy B.R., Willis D. W. *Fisheries Techniques*. Second edition, American Fisheries society, Bethesda, Maryland, USA, XX, pp732, 1996.

Nikolski G. *Ecology of Fishes*. Moscow, "Visshaya Shkola", 1974.

Peskova O. Biology of vendace from Paravani and Sagamo lakes. Proceedings of scientific-research fishery station of Georgia, vol. **V**, 3-37, 1960.

Pravdin I. *Leading on fish study.* M., "Pishchevaya promishlennost", 373, 1966.

ევროპული ჭაფალას (*Coregonus albula* L.) ნაკვეგობის კოეფიციენტის დინამიკა ფარავნის ტბაში

ჯაფოშვილი ბ.[1], ქოქოსაძე თ.[2], ჯაფოშვილი, ო.[1]

[1] ი. ჭავჭავაძის თბილისის სახელმწიფო უნივერსიტეტი
[2] ზოოლოგიის ინსტიტუტი

(მიღებულია 25.09.2006)

რეზიუმე

ევროპული ჭაფალას ორივე სქესის წარმომადგენლებში პირველადაა შესწავლილი ნაკვეობის კოეფიციენტის დინამიკა. შედარებულია მდედრებისა და მამრების ნაკვეობის კოეფიციენტი. განხილულია განსხვავებების მიზეზები. ნაჩვენებია, რომ ბოლო წლებში ნაკვეობის კოეფიციენტის დაცემა კლიმატურ ცვლილებებთან და გლობალურ დათბობასთანაა დაკავშირებული.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Entomology

NEW SPECIES OF MERMITHID *HEXAMERMIS DECEMLINEATAE* SP.N. (NEMATODA, MERMITHIDAE) FROM COLORADO BEETLE

GORGADZE O., LORTKIPANIDZE M., KOKHIA M., MELASHVILI N., KUCHAVA M.

Institute of Zoology

(Received October 9, 2006)

Abstract

The paper deals with the description of the new species of mermithid *H. decemlineatae* sp.n. The measurements of adult female, male and post parasitic larvae are given. Host: Colorado beetle (*Leptinotarsa decemlineata* Say). Parasitic and postparasitic mermithid larvae are revealed in the body of beetle, but adult females and males were recorded in the soil. The minimal number of mermithid larvae in each beetle was 1 specimen, and maximal - 30 specimens. In the host the number of parasitic larvae amounts to 1-3 specimens. In natural environment 47.5% of the beetles and 64.5% of larvae were infected with mermithid larvae. Nematode is localized in adipose tissue of the beetle and larvae.

Key words: parasitic and postparasitic nematodes, anatomical and morphological studies, *Hexamermis stepposis, Hexamermis angusta.*

Introduction

Colorado beetle *Leptinotarsa decemlineata Say* (Coleoptera) belongs to the Chrysomelidae family. It is harmful pest of potato. This pest was introduced from North America and was spread in most territories of Europe and Asia [Briantsev, 1966].

According to the studies carried out in biocenosis of potato sowings it was found out that those organisms (nematodes, bugs, carabus, ladybirds), which significantly reduce number of Colorado beetle, were consequently adapted on them. In this way entomopathogenic nematodes of the Mermithidae family are especially significant. Those nematodes in humid conditions can infest beetle, as well as larvae of Colorado beetle, and stimulate their death with 80-95% rate [Mishachkov, 1980]. Due to this fact mermithids appear to be perspective control agents against pests [Ipatieva, Pimenova, 1985; Rubtsov, 1978].

The goal of our work was to study nematodes of Colorado beetles distributed in some regions of East Georgia.

Materials and Methods

To study parasitic nematodes of Colorado beetle and its larvae potato sowings of private farms of mountain regions of East Georgia were researched. Places of collection were: villages Thesami, Ghulelebi, Trani, (Mtskheta-Mtianeti region).

665 specimens of the beetle and 1225 specimens of its larvae were dissected using Pavlovski method [Pavlovski, 1957]. 511 specimens of parasitic and postparasitic nematodes of one species were revealed in beetles and larvae, but the adult forms of the same species - in the soil of potato sowings. For anatomical and morphological study of collected nematodes temporary and long-term preparations were prepared [Poinar, 1975]. For nematode identification international index formulae of nematodology were used [De Man, 1884; Micoletzky, 1914]. It was established that recorded nematode belongs to the genus Hexamermis and family Mermithidae.

Genus diagnosis - Hexamermis Steiner, 1924 [Steiner, 1924].

Nematodes of this genus are of middle or big sizes (50-80 mm). Frontal part of the head of female, unlike male, is of mainly conic form. Tail end is rounded. Cuticle of the end parts of head and tail of parasitic and postparasitic larvae is thicker, than of the body middle part. Mouth opening in the frontal part of the head is placed symmetrically. Has 6 cephalic papillae; has no labial papillae; Amphids of small size. Vulva is straight and has significantly thickened stoma. Vagina is of pear-form; spicule - pair, straight and short. Has thick sexual papillae. Tail ends of parasitic and postparasitic larvae are rounded.

Typical species: *Hexamermis angusta* Rubzov, 1971 [Rubtsov, 1971].

Results and Discussion

Host: Colorado beetle (*Leptinotarsa decemlineata* Say).

Localization: in adipose tissue of the beetle and larvae.

The apical part of head of female nermithid is speculated, but of male – rounded. (Fig. 1 A, C). Neck gland is seen under cuticle. There are not protrudent tubers on the head. Amphids are small (4-6 μm) and oval. Their ducts are opened a bit lower of cephalic papillae. Cuticle oesophagus is not spread up to the mouth opening. Width of mouths opening walls is of 3-5 μm.

Female: n=7; L=42.3 (32.0-62.5) mm; a = 172.1 (155.3-203.6); V (%) = 55 (54-57);

Body width: near cephalic papillae consists of 70 (53-115) μm, near nerve ring – 162 (92-222) μm, near vulva – 250 (157-380) μm and near the end of trophosome – 158 (120-277) μm. Distance from the frontal part of the head to nerve ring is 269 (179-335) μm; up to vulva – 25.5 (17.3-30.8) mm; from the end of trophosome to the tail end – 168 (75-280) μm. Structure of vagina is not distinguished from that of species described by Rubtsov [Rubtsov, 1971]. Width of cuticle near mouth opening is 21 (19-32) μm, near vulva – 14 (9-18) μm, at the tail end – 25 (24-33) μm.

Male: n=23.5 (19.3-38.6) mm; a=135.1 (105-183.7); c=130.6 (83.8-159.4).

Body width: near cephalic papillae consists of 70 (56-93) μm; near nerve ring 115 (80-193) μm; at anus –155 (120-240) μm; the widest part of body - 198(153-322) μm. Distance from the frontal part of head to nerve ring is 305 (240-396) μm. Male has weakly bent pair spicule (Fig. 1. D), which length is 161 (103-250) μm, diameter – 20 (15-36) μm, but its end is acute. Cuticle width at head opening in the front part of the body is 18 (14-23) μm, in the middle part – 13 (10-16) μm and near tail - 13 (6-30) μm. Tail length is 200 (150-304) μm.

Postparasitic larvae

Female: n=1; L=25.3 mm.

Body width: at cephalic papillae – 98 μm, at nerve ring – 170 μm, at vagina – 335 μm, at the end of trophosome – 225 μm. Distance from the apical part of the head to nerve ring consists of 350 μm, from the end of trophosome to the end of tail – 345 μm. Distance from front of the head to vagina equals to 20.2 mm. Cuticle width is: near head opening in the front of the head – 41 μm, at vagina in the middle part of the body – 13 μm, and at the tail – 102 μm.

Differential diagnosis

By anatomical and morphological characteristics species described above resembles species *Hexamermis stepposis* Artyukhovsky et Khartschenko, (1965) [Artyukhovsky, Khartschenko, 1965], but is more similar to the species *Hexamermis angusta* Rubzov [Rubtsov, 1971], from which it is distinguished by the form of vulva lips, by form and size of amphids.

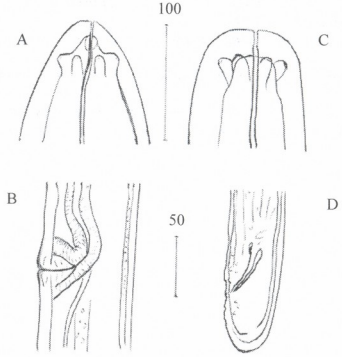


Fig. 1. Hexamermis decemlineatae sp. n
Female: A – frontal end of the body; B – vulva segment
Male: C – frontal end of the body; D – tail segment with spicule.

According to anatomical-morphological features *Hexamermis decemlineatae* sp.n. is considered as a new species for Georgia.

References:

Anderson R.S. *Methode pour I examen des nematodes en vue apicale.* Ann. Parasitol. Hum. Comp., **33**, 171-172, 1958.

Artiukhovski A.K., Kharchenko N.A. *Study of mermithids of Streletskaya Steppe.* Proceedings of Central Black Sea Reservoir, M., p.9, 1965 (in Russian).

Briantsev B.A. *Agricultural entomology.* L., "Kolos", pp.342, 1966 (in Rusian).

De Man J. G.. *Die frei in d. reinen Erde u. in sussen Wasser lebenden nematoden d. niederl. fauna Leiden.* p. 313-321, 1884.

Ipatieva G.V., Pimenova I.N. *Mermithids of the Altai Mountains.* Publ. Saratov University, p.1-149, 1985 (in Russian).

Micoletzky H. *Freilebende Subwasser-Nematoden der Ost- Alpen mit besonderer Beruckichtigung des Lunzer Seengebietes.* Zool. Jahrb. Abt., **1**, 36, 331-546, 1914.
Mishachkov G.A. *Biology and ecology of mermithid Hexamermis albicans - parasite of Colorado beetle.* In: Helminths of insect. M., "Nauka", 85-89, 1980 (in Russian).
Pavlovski E.N. *Methods of hand-anatomy of insect.* M., "Nauka", p.1-45, 1957 (in Russian).
Poinar G.O.jr. *Entomogenous nematodes. A manual and host list of insect – nematode associations.* Leiden E. J. Brill., p. 1-317, 1975.
Rubtsov I.A. *New species of Mermithidae from southern seaside.* Zool. J., **50**, 8, 1143-1152, 1971 (in Russian).
Rubtsov I.A. *Mermithids. Classification, significance, usage.* L., "Nauka", p.207, 1978 (in Russian).
Steiner G. *Beitrage zue Kenntnis der Mermithiden, 2. Mermithiden aus Paraguay in der Sammlung des Zoologischen Museum zu Berlin.* Zbl. Bacteriol., Parasitenkunde. **62**, 1-6.1924.

ახალი სახეობის ნემატოდა *Hexamermis decemlineatae* sp. n. (Nematoda, Mermithidae) კოლორადოს ხოჭოდან

გორგაძე ო., ლორთქიფანიძე მ., კოხია მ., მელაშვილი ნ., კუჭავა მ.

ზოოლოგიის ინსტიტუტი

(მიღებულია 09.10.2006)

რეზიუმე

აღწერილია მერმიტიდას *Hexamermis decemlineatae* sp.n. ახალი სახეობა. მოცემულია ზრდასრული ინდივიდების და პოსტპარაზიტული ლარვების განაზომები. მასპინძელი: კოლორადოს ხოჭო (*L. decemlineata* Say). ლოკალიზაცია: ხოჭოსა და ლარვების ცხიმოვანი ქსოვილი. ხოჭოს სხეულში გამოვლენილია მერმიტიდას პარაზიტული და პოსტპარაზიტული ლარვული ფორმები, ხოლო ზრდასრული ეგზემპლარები გამოვლენილია ნიადაგში. მერმიტიდების ლარვების მინიმალური რაოდენობა თითო ხოჭოში შეადგენდა 1 ეგზემპლარს, ხოლო მაქსიმალური - 30. მასპინძელში პარაზიტული ლარვების რაოდენობა აღწევდა 1-3 ეგზემპლარს. ბუნებრივ პირობებში მერმიტიდების ლარვების მიერ დაინვაზირებულია ხოჭოების 47,5%, ხოლო ლარვების 64,5%.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Genetics

GENE DRIFT IN THE MIRZAANI POPULATION OF WINE YEAST

SHATIRISHVILI A.[1], MENABDE M.[2], SADAGISHVILI T.[2], ZARNADZE T.[3]

[1]*I. Chavchavadze Tbilisi State University*
[2]*Iv. Javakhishvili Tbilisi State University*
[3]*A. Tsereteli Kutaisi State University*

(Received March 1, 2006)

Abstract

During 1996 – 2004 the antagonistic activity was studied in the Mirzaani (Kakheti) population of wine yeast. The population was found to be polymorphic by the feature. Three phenotypic classes were identified in the population: killer (K), neutral (N) and sensitive (S) classes. Gene drift resulting in periodic fluctuation of the phenotypic classes was revealed.

Key words: yeast population, antagonistic activity, killer-system.

Introduction

Among other factors affecting the gene pool of population the gene drift is of particular importance. As a result of its activity allele concentrations change in the gene pool. This process is especially intensified in the populations with great decrease in the number of members [Hedrick, 1999; Shatirishvili, 2002]. Such periodic (cyclic) number fluctuation in the wine yeast is due to abiotic, biotic or anthropogenic factors. In the regions of well-developed domestic wine production of Georgia fermentation of wine occurs spontaneously. The process is first prompted by a small group of yeasts, i.e. "the founder's principle" acts [Shatirishvili, 2002; Mayr, 1970]. Small numbers of yeasts give rise to tens of millions of new ones. The yeast gets into the grape juice from the philosphere, ripened grape berries and wine cellar implements, while the stream depends on the Drosophila [Ribero-Gasion et al., 1980].

Materials and Methods

In Saccharomyces, and particularly in the wine yeast, no traits identifying the population have been worked out yet. Its reproductive area depends on the Drosophila. Therefore, it occupies about 400-500 meters [Sadagishvili et al., 2001; Menabde et al., 2004]. Proceeding from that we considered the forms of wine yeast spread over village Mirzaani and nearby regions to be a population. The material (sediment) was taken from 10 different remote districts by the method described before [Menabde et al., 2004].

The antagonistic activity of strains was detected by means of the testing strains: K7 {KIL – K1}; S14 (sensitive to the system K1); Oxford genetic lines; the line M437 {KIL-K2} created at the Institute of Genetics of Russian Academy of Sciences; the line RA p192 (sensitive to the system K2), a Petergof genetic line. Special culture media were applied for studying the

antagonistic activity of the strains. The specificity of discovery of the killer systems was described before [Shatirishvili et al., 2001].

Results and Discussion

The alcoholic fermentation of grape juice represents a complex multi-stage process. Some microorganisms are involved in it and the yeast fungi join the process at the final stage. During the fermentation inter- and intra-specific competitions take place [Shatirishvili et al., 2001]. The inter-strain antagonism occurs in the wine yeast, that becomes apparent when the cells of sensitive strains are eliminated under the effect of a toxin released by another strain [Menabde et al., 2004].

In order to study the variability of gene frequencies in natural populations of wine yeast, in 1996-2004 we investigated the Mirzaani (Kakheti) population. With the interval of 2-3 years the strains were repeatedly isolated from 10 micropopulations (500 cultures in total) and analyzed genetically. The antagonistic activity was defined by introducing strokes of the strains under study to the nutrient media with a special loop. If the strain contains a killer system, the latter causes a lysis of sensitive test-cultures forming a sterile ring around the developed culture. If a strain is sensitive to the test culture the area of eliminated cells gets stained in dark blue color by Methylen – blue stain [Shatirishvili et al., 2001]. The strain that has a killer-system produces and releases the protein – a toxin that induces elimination of sensitive strain cells. The strains with neutral phenotype are sensitive to the toxin. The results obtained for the strains in 1996 are given as patterns in Tables 1 and 2. The other results have already been published [Sadagishvili et al., 2001; Menabde et al., 2004; Shatirishvili et al., 2001].

The strains constituting the population are arranged in three phenotype classes: killer (K), neutral (N) and sensitive (S) strains. The whole population as well as each micro population appeared to be polymorphic. When compared the structures of different populations studied in different years, we found that in 1998 and 2000 the rates of killer strains reached maximum (17,3% and 15,8 % respectively), while in 2004 the content of killer strains was minimal – 1,4% (see Fig. 1).

Table 1. Determination of antagonistic activity of Mirzaani "Rkatsiteli" population

Phenotype	Number of classes	Test-strains			
		M437	7A – P192	K7	S14
I	1	K	K	K	N
II	7	K	K	N	N
III	4	K	N	K	K
IV	2	K	N	K	N
V	9	N	K	K	N
VI	11	N	N	K	K
VII	3	N	N	K	N
VII	6	N	N	K	K
IX	451	N	N	N	N
X	2	N	N	N	S
XI	2	N	N	S	N
XII	1	S	N	N	S
XII	1	N	S	S	S

Changes in environmental conditions sharply affect the ratio of the strains with K, N and S phenotypes and cause variations in natural populations of the yeast. The reason for that is number fluctuation. Under the severe climate conditions in winter and spring the quantity of the yeast

dramatically decreases causing so called "bottleneck" effect. Thus, it comes the period when the number of strains in population and the density of the population become minimal [Shatirishvili, 2002].

In autumn (in vintage) in the sites of domestic wine production, where the wine fermentation is of spontaneous character, the number and density of the yeast within the population sharply rise. Thus, after passing the "bottleneck" the quantity of population members in the natural population reaches maximum. The small groups with occasionally survived yeasts give rise to the yeast population that means that "the founder's principle" works.

Table 2. Determination of the frequencies of phenotypes K, N and S in Mirzaani micropopulations

Micro-population	Number of analyzed strains	Killer		Neutral		Sensitive	
		Number	%	Number	%	Number	%
I	50	3	6	47	94	-	-
II	50	2	4	48	96	-	-
III	50	5	10	43	86	2	4
IV	50	-	-	50	100	-	-
V	50	1	2	48	96	1	2
VI	50	4	8	46	92	-	-
VII	50	5	10	43	86	2	4
VII	50	6	12	43	86	1	2
IX	50	12	24	38	76	-	-
X	50	5	10	45	90	-	-
Total	500	43	8.6	451	90.2	6	12

References:

Hedrick P. W. *Genetics of population*. Sones & Barlett Publishers, 1999.

Mayr E. *Populations, species and evolution.* Harvard university press, 1970.

Menabde M., Sadagishvili T., Kacharava M., Shatirishvili A. *Antagonistic activity in endemic population of wine yeast (Saccharomyces vini)*. Proc. Georgian Acad. Sci., Biol. Ser. B, **2**, 5-6, 57-60, 2004.

Ribero-Gasion J., Reino E., Ribero-Gasion P. *Theory and practice of wine-making*. v.3, M., 1980

Sadagishvili T., Zarnadze T., Menabde M., Shatirishvili A. *Killer systems of wine yeasts*. Proc.Georgian Acad.Sci., Biol.Ser. **27**, 1-3, 225-229, 2001.

Shatirishvili A., Sadagishvili T., Zarnadze T., Menabde M. *Antaginistic activity of natural saccharomyces*. Georgian Med. News, **70**, 1, 48-51, 2001.

Shatirishvili A. *Genetic mechanisms of evolution in natural population of wine yeasts.* In: Biology and contemporaneity, Tbilisi, p. 254-260, 2002.

ბენთა დრეიფის მოქმედება ღვინის საფუარის მირზაანის პოპულაციაში

შათირიშვილი ა.[1], მენაბდე მ.[2], სადაგიშვილი თ.[2], ზარნაძე თ.[3]

[1]ი. ჭავჭავაძის თბილისის სახელმწიფო უნივერსიტეტი
[2]ივ. ჯავახიშვილის თბილისის სახელმწიფო უნივერსიტეტი
[3]ა. წერეთლის ქუთაისის სახელმწიფო უნივერსიტეტი

(მიღებულია 01.03.2006)

რეზიუმე

შესწავლილია ანტაგონისტური აქტივობა ღვინის საფუარის მირზაანის პოპულაციაში. პოპულაციის სტრუქტურა პოლიმორფული აღმოჩნდა. იგი სამი ფენოტიპური კლასითაა წარმოდგენილი: კილერი (K), ნეიტრალური (N), მგრძნობიარე (S). მიკროევოლუციის მამოძრავებელი ფაქტორის ხემოქმედების შედეგად ფენოტიპური კლასები პერიოდულად ფლუქტუირებს.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Immunology

DISTRIBUTION FEATURES OF SOME ERYTHROCYTIC GROUP-SPECIFIC ANTIGENS SIGNIFICANT FOR CLINICAL MEDICINE IN ADJARA REGION

*NAGERVADZE M., DIASAMIDZE A., AKHVLEDIANI L., DUMBADZE G., KHUKHUNAISHVILI R., KORIDZE M.

Sh. Rustaveli Batumi State University
**E-mail: mari-na@nm.ru*

(Received September 20, 2006)

Abstract

Erythrocytic group antigens interesting in the viewpoint of transfusion, such as *c, K, D^u*, were studied in Adjara population. Those antigens are characterized with high immunogenicity but their screening is not conducted. It was shown that distribution frequency of *c* antigen in Adjara population is 90.2±3.03%. 4.2% of population is carrier of *K* factor. Distribution frequency of D^u antigen is 0.625±0.12%. Number of *CC* genotype carriers within the population appeared to be 8.0±2.7%.

Key words: immunoserological methods, *ABO* system, *Rh* system, *Kell* system.

Introduction

Erythrocytic group antigens stipulating for blood compatibility and appearing as the main reason of posttransfusion complication are significant for clinical medicine [Anstee et al., 1999]. Significance of those group antigens is associated with immune characteristics of the living organism. They take important role in transfusiology [Schonewille et al., 2006], epidemiology [Vojvodic, 2000] and transplantology [Bucin, 2006], in human genetics [Shubin, 1997], and especially in the studies of population genetics [Kucher, 2000]. Study of erythrocytic group systems is also important in ethnic anthropology [Shneider et al., 2002]. Heredity of those systems is so stable that their study for establishment of some ethnic group origin gives accurate data [Schmidt et al., 2003].

Today in Adjara two antigens (*A, B*) of *ABO* system and *D* antigen of *Rh* system are taken into account during blood transfusion. For the individuals where those antigens do not occur the theoretical risk of alloimmunosensitization is high [Judd et al., 1992]. In the viewpoint of transfusion *c* antigen, among rhesus system antigens, is also significant. Numerous data about alloimmunosensitization caused by this antigen are presented in scientific literature [Regan et al., 1997]. Distribution frequency of *c* antigen within world population is 80-82%. 18-20% of humans do not have this antigen and are revealed in *CC* state. Individuals with just this genotype belong to high-risk group of alloimmunosensitization.

Immune activity of *K* antigen is slightly minor than rhesus antigen (*RhD*) activity. Immunosensitization caused by *K* antigen is a frequent case [Donskov, 1996], which is certified by the numerous data of posttransfusion complications described in literature.

Today it is necessary to carry out screening of donors by *K* antigen. At present *Kell*-positive blood is not used in transfusion in many countries.

The majority of mistakes during rhesus system determination are related with weak variation of *D* antigen – D^u. Unlike *D* antigen, D^u antigen has latent antigen determinants. For their discovery, first, it is necessary to fix those determinants on the surface of erythrocytes, and further, to reveal them. Such study is carried out in all those cases when *Cde, cdE* phenotypes are revealed during the primary phenotyping of erythrocytes. Complex methods enable to delete individuals having CD^ue and cD^uE phenotypes from rhesus-negative donors.

Unfortunately, proceeding from high transfusion significance of abovementioned antigens, their screening is not conducted today.

The aim of our work was to study regularities of distribution of erythrocytic group antigens in Adjara region and to forecast theoretically expected alloimmunosensitization.

Materials and Methods

The study was carried out by immunoserological methods. Test-systems having anti -*c*, -*K* specificities, anti –*D* incomplete antibody, antiglobulin serum, standard group erythrocytes and standard serums, were used.

512 individuals of Adjara population were studied.

The obtained data-processing was carried out using statistical methods.

Results and Discussion

In the majority of studied individuals *c* antigen was registered (90.2±3.03%). Distribution frequency of *C* antigen was 53.0±5.3% [Nagervadze et al., 2006] (Fig.1). According to the research of allele concentrations it was revealed that concentration of *c* allele is high and equals to 0.74 (Fig. 2).

Forecasting of theoretically expected immunosensitization caused by *c* antigen was carried out. Carriers of *CC* genotype were separated out. Distribution frequency of *CC* genotype in Adjara population is equaled to 8.0±2.7% (*Cc* – 54.0±4.9 and *cc* – 38.0±4.8), implying that carriers of this genotype do not consist in *c* antigen and during transfusion in 92% cases incompatibility should be revealed (Fig. 3).

With low frequency, but nevertheless, D^u antigen was recorded in Adjara population. Distribution frequency of D^u antigen is 0.625±0.12% (Fig. 4).

Donors having D^u antigen should be belonged to rhesus-positive, but recipients – to rhesus-negative group and rhesus-negative blood should be transfused to them, because normal *D* antigen may cause immune response therein.

As a result of our studies it was revealed that 4.2% of Adjara population is carrier of *K* factor (Fig. 5).

Thus, it was established that distribution frequency of *c* antigen is high in Adjara population, and respectively theoretical risk of immunosensitization caused by this antigen is high. It is necessary to provide medical laboratories with information that erythrocyte screening of donor-recipients upon such antigens, as *c*, C^w, D^u, *K*, must be carried out. Such approach should

decrease cases of posttransfusion complications and the risk of immunosensitization should be brought to minimum.

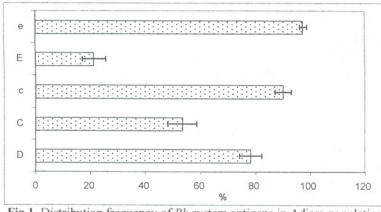


Fig.1. Distribution frequency of *Rh* system antigens in Adjara population

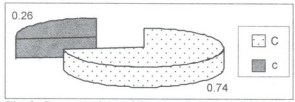


Fig. 2. Concentrations of *C* and *c* alleles in Adjara population

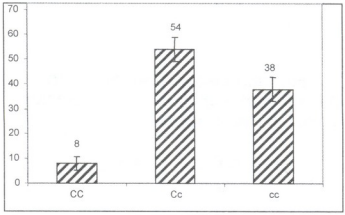


Fig. 3. Analysis of *Cc, CC* and *cc* genotypes in Adjara population

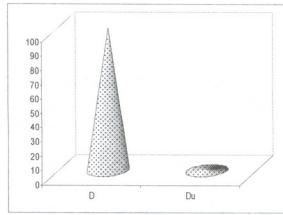


Fig. 4. Distribution frequency of *D* and *D*^{u} antigens in Adjara population

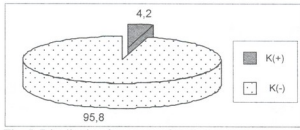


Fig. 5. Distribution frequency of *Kell* system phenotypes in Adjara population.

References:

Anstee D.J., Cartron J.P. *Toward an understanding of the red cell surface//Red cell immunohaemato logy towards its second century.* ESTM. TelAviv, p.12-44, 1999.

Bucin D., Johansson S., Malm T., Jogi P., Johansson J., Westrin P., et al. *Heart transplantation across the antibodies against HLA and ABO.* Transpl Int., **19**, 3, 239-44, 2006.

Kucher AN., Puzyrev VP., Chernetsov DB., Erdynieva LS., Sanchat NO. *Polymorphism of immunological and biochemical marker genes in rural populations of the Tuva Republic.* Genetika, **36**, 4, 562-9, 2000.

Nagervadze M., Akhvlediani L., Dumbadze G., Bagrationi K., Diasamidze A. *Distribution of Rh-Hr antigens in Adjara population.* Proc. Georgian Acad. Sci., Biol.Ser. B, **4**, 1, 48-51, 2006.

Schmidt HD, Scheil HG. *Blood group frequencies in Romania: microregional and ethnic differences.* Anthropol Anz., **61**, 4, 381-93, 2003.

Shneider Iu.V., Shilnikova I.N., Zhukova O.V. *Materials for studying the gene pools of Russia and neighboring countries: the Russian population of the Pskov region.* Genetika, **38**, 11, 1561-5, 2002.

Shubin PN., Morokov VA., Efimtseva EA., Chelpanova TI. *Ethnogenetics of the Komi-Zyrians according to data on the distribution of gene frequencies of erythrocyte and serum systems in the blood.* Genetika, **33**, 2, 235-42, 1997.

Vojvodic S. *Inhibitory activity of blood group antigens M and N in inhibition of virus hemagglutination reactions of influenza viruses.* Med Pregl., **53,** 1-2, 7-14, 2000.

კლინიკური მედიცინისათვის მნიშვნელოვანი ზოგიერთი ერითროციტური ჯგუფსპეციფიკური ანტიგენების გავრცელების თავისებურებანი აჭარის რეგიონში

ნაგერვაძე მ., დიასამიძე ა., ახვლედიანი ლ., დუმბაძე გ., ხუხუნაიშვილი რ., ქორიძე მ.

ჭ. რუსთაველის ბათუმის სახელმწიფო უნივერსიტეტი

(მიღებულია 20.09.2006)

რეზიუმე

აჭარის მკვიდრ მოსახლეობაში შესწავლილია ტრანსფუზიური თვალსაზრისით საინტერესო ერითროციტური ჯგუფური ანტიგენები, როგორიცაა *c, K, D^u*. აღნიშნული ანტიგენები საკმაოდ მაღალი იმუნოგენურობით ხასიათდებიან, მაგრამ არ ხდება მათი სკრინინგი. ნანახებია, რომ აჭარის მოსახლეობაში *c* ანტიგენის გავრცელების სიხშირე 90.20±3.03%-ია. აჭარის მოსახლეობის 4.2% ფაქტორის მტარებელია. *D^u* ანტიგენის გავრცელების სიხშირე – 0.625±0.12%-ია. გამოყოფილი იქნა *CC* გენოტიპების მტარებელი პირები; მათი გავრცელების სიხშირე 8.0 2.7%-ია.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

DETERMINATION OF ANTIMICROBIAL ACTIVITY OF NITROGEN FIXING BACTERIA

BAGALISHVILI M., BASILASHVILI L., KIKVIDZE M., GURIELIDZE M., NUTSUBIDZE N.

S. Durmishidze Institute of Biochemistry and Biotechnology

(Received October 6, 2006)

Abstract

Antagonistic features of rhizosphere of nitrogen-fixing microorganisms, which they reveal towards test-organisms, including phytopatogenic fungi and actinomyces have been studied. Most of investigated nitrogenfixers (28 cultures out of 30) display antimicrobial activity in different degree. Antimicrobial activity of rhizospere nitrogenfixing microorganisms together with other positive characteristics (fixation of molecular nitrogen, production of growth stimulators) enable to use these microorganisms in agriculture on the purpose of soil remediation and enhancing of productivity.

Key words: phytopatogenic fungi, actinomyces, nitrogen-fixing microorganisms

Introduction

Microorganisms are never found in isolated state in natural conditions. They can be obtained as pure cultures only artificially, but in nature, they represented in association, where different interactions are established between microbes. One of the most wide spread forms of interaction is antagonism - when one strain of organisms suppresses development of other organisms. Antagonistic properties of microorganisms are mostly manifested by formation of antimicrobial substances [Crawford, 1988].

28 pure cultures of nitrogen fixers were isolated from rhizosphere of cereals. These rhizospherous microorganisms use the energy of photoassimilates isolated by plant roots for the energy – consuming nitrogen fixation processes. It is supposed that these organisms obtain other advantages that make them competitive among various microbes around the plant roots [Shah et al., 1992].

That is the reason for study whether antimicrobial substances are isolated from rhizosphere of nitrogen fixing microorganisms and what the model is of action of these substances on other microbes.

Materials and Methods

30 nitrogen-fixing microorganisms were studied in all. As test-organisms were used actinomycetes: *Streptomices* 82; *Streptomices fradiae* 110, Phytopathogenic fungi: *Fusarium solani*, *Rhizoctonia* sp; yeasts: *Saccharomyces fragilis*, *Candida utilis*; gram-positive bacteria: *E.coli*, *Ps. fluorescens* (Table 1).

Antimicrobial activity of isolated by us and collectional nitrogen fixing microorganisms was studied by the method of agar blocks [Egorov, 1965] as follows: on Petri dishes with corresponding agar nutrient medium, the studied microorganisms were platted in a form of a lawn. After good development of a cultures and formation of antimicrobial substance, which is diffused in agar, we cut 10 mm diameter agar blocks with a special sterile lancet and replace them on other Petri dishes with previously platted test-organisms on a nutrient medium. After 18-20 h of incubation at the optimal for test-organisms temperature light zones were formed around agar blocks, indicating suppression of development of test-organisms. The results were registered 36- 48 h after incubation. According to sterile zones diameter around agar blocks we could judge about antimicrobial activity of microorganisms.

Results and Discussion

28 nitrogen fixing microorganisms, among studied by us 30 ones, reveal antimicrobial activity against 9 out of 11 test-organisms. The widest spectrum was characteristic for *A. brasilense* Г3 and *A. brasilense* G3 (7 test-organisms). Many microorganisms were found to have antimicrobial activity to *Fusarium solani* (19 microorganisms) and *Streptomyces fradiae* (17 microorganisms). The highest antimicrobial activity of microorganisms was manifested to *Fusarium solani*: G-41 - 19 mm, G111 - 19 mm and G71 - 18 mm (Table 1).

Table 1. Antimicrobial activity of nitrogen fixing microorganisms

Nitrogen fixing microorganisms	test-organisms (size of zones, mm)										
	Strept. fradiae 82	*Stept. fradiae 110*	*Fusarium solani*	*Rhizoctonia sp.*	*Sacch. fragilis*	*Candida utilis*	*Myc. phlei*	*Rhodococcus sp.*	*Staph. aureus*	*E. coli*	*Ps.fluorescens*
A.bras. ATCC 9825	11	12	-	14	-	-	13	11	14	-	-
A.brasilense Г3	11	14	13	14	-	15	12	16	-	-	-
A.brasilense G1	11	12	-	-	-	-	11	-	14	-	-
A.brasilense G2	12	13	-	11	-	13	12	-	-	-	-
A.brasilense G3	13	12	12	15	-	-	12	13	14	-	-
A.brasilense G4	12	-	11	15	-	11	13	-	-	-	-
A.brasilense G5	13	12	13	-	12	-	12	-	-	-	-
A.brasilense G12	11	-	11	-	-	-	-	-	-	-	-
A.brasilense G16	-	-	-	-	-	-	-	13	-	-	-
A.brasilense G20	-	-	12	-	-	11	-	-	-	-	-
G22	-	-	-	-	-	-	-	-	-	-	-
G23	-	12	-	-	-	-	13	11	-	-	-
G24	-	13	-	-	-	-	-	-	11	-	-
G26	11	16	17	-	11	-	-	-	-	-	-
G41	-	14	19	-	-	-	13	14	12	-	-
G43	-	15	14	-	-	-	-	-	-	-	-
G44	-	14	-	-	-	-	-	-	-	-	-

G45	-	13	11	11	-	-	-	-	-	-	-
G61	-	-	13	15	13	13	-	15	17	-	-
G62	-	-	12	-	12	-	-	-	11	-	-
G64	-	-	12	-	-	-	-	-	-	-	-
G66	-	12	13	-	-	-	-	14	-	-	-
G68	-	12	-	-	11	-	11	11	13	-	-
G70	-	-	14	-	11	11	-	-	-	-	-
G71	-	-	18	12	-	11	-	-	-	-	-
G72	-	-	12	-	11	-	-	-	-	-	-
G110	-	-	-	-	-	-	-	-	-	-	-
G111	-	11	19	-	-	-	-	-	-	-	-
G120	-	11	18	-	11	-	-	-	-	-	-
G121	-	-	17	-	11	-	-	-	11	-	-

Fusarium solani – is phytopathogenic fungi, it is characterized by high capability to infection and preserves in soil for a long time, as it suppresses development of useful microorganisms and simplifies the process of penetration into a plant tissue [Meyer et al 1998]. Issuing from the said above the fact that nitrogen fixing microorganisms and among them *Azospirillum* excrete antimicrobial substances towards such strong phytopathogenic organisms is very interesting (Fig.1, 2).

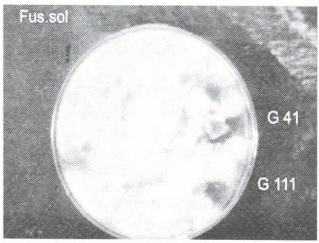


Fig. 1. Antagonism of nitrogen-fixing strains G41 and G111 towards *Fusarium solani*

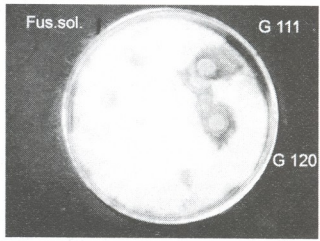


Fig. 2. Antagonism of nitrogen-fixing strains G111 and G120 towards *Fusarium solani*

Thus, most of the studied nitrogen fixers (28 cultures from 30) obtain antimicrobial activity to 9 from 11 test-organisms that gives additional advantages to rhizosphere to develop in microflora. Issuing from the said above introduction of active strains of nitrogen fixers into rhizosphere of agricultural cultures in our opinion will have positive effect on their growth and development.

References:

Egorov N. S. *Microbes – antagonists and biological methods of determination of antibiotic activity.* M., "Visshaya shkola", 1965.

Crawford D.L. *Development of recombinant Streptomyces for biotechnological and environmental uses.* Adv. Biotechnol., **6**, 183-206, 1988.

Meyer L., Weher F.C., Nel & Carling D.F. Characterization of the crater disease strain of *Rhizoctonia solani.* J. Phytopathology, **88**, 4, 366-371, 1998.

Shah S., Karkhanis V., Desai A. *Isolation and characterization of siderophore, with antimicrobial activity, from Azospirillum lipoferum M.* Curr. Microbiol., **25**, 34-35, 1992.

აზოტმაფიქსირებელი ბაქტერიების ანტიმიკრობული აქტივობის განსაზღვრა

ბაგალიშვილი მ., ბასილაშვილი ლ., კიკვიძე მ., გურიელიძე მ., ნუცუბიძე ნ.

ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 06.10.2006)

რეზიუმე

შესწავლილია რიზოსფერული აზოტმაფიქსირებელი მიკროორგანიზმების ანტაგონისტური თვისებები, რომლებსაც ისინი ავლენენ სხვადასხვა ტესტ-ორგანიზმების, მათ შორის ფიტოპათოგენური სოკოებისა და აქტინომიცეტების მიმართ. გამოკვლეულ აზოტფიქსატორთა უმრავლესობამ (30-დან 28 კულტურამ) გამოავლინა ანტიმიკრობული აქტივობა მეტ-ნაკლები ხარისხით. რიზოსფერული აზოტმაფიქსირებელი მიკროორგანიზმების აქტივობა სხვა დადებით მახასიათებლებთან ერთად (მოლეკულური აზოტის ფიქსაცია, ზრდის სტიმულატორების პროდუქციის უნარი) საშუალებას იძლევა ეს მიკროორგანიზმები გამოყენებულ იქნას სოფლის მეურნეობაში ნიადაგების გაჯანსაღებისა და პროდუქტიულობის გაზრდის მიზნით.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

THERMOPHILIC MICROSCOPIC FUNGI OF SOILS OF EAST GEORGIA

*DAUSHVILI L., BURDULI T., KUTATELADZE L., JOBAVA M., DZALAMIDZE I.

S. Durmishidze Institute of Biochemistry and Biotechnology. David Agmashenebeli Alley 10km, 0059 Tbilisi, Georgia
**E-mail: lela_d@mail.ru*

(Received August 14, 2006)

Abstract

70 cultures of microscopic fungi, isolated from dry subtropical-steppe zone of Sighnaghi region (east Georgia) have been investigated. 5 thermophilic, 5 thermotolerant and 4 psychrophilic (facultative) strains of micromycetes were revealed. The optimal ranges of growth and spore-formation of thermophilic micromycetes have been established.

Key words: thermophilic, thermotolerant, psychrophilic, *Aspergillus*.

Introduction

Thermophilic microorganisms, in particular thermophilic fungi are the subjects of intensive investigation. This interest is determined by preferred application of thermophilic fungi and their enzymes in different fields of industry, agriculture and medicine, compared with mesophilic microorganisms and their enzymes [Bilay, 1979].

Thermophilic microscopic fungi are potential sources of various industrially important thermostable enzymes, such as lipases, xylanases, proteases, amylases and pectinases. These enzymes have numerous applications in the detergent, starch, paper, food and pharmaceutical industries [Phutela et al., 2005]. Due to their increased thermostability, enzymes of thermophilic micromycetes are potentially useful in the starch industry for production of maltose and glucose. Thermostable amylase are generally preferred as their application minimizes contamination risk and reduces reaction time, thus providing considerable energy saving. Hydrolysis carried out at higher temperatures also minimizes polymerization of D-glucose to iso-maltose [Kunamneni et al., 2005]. Thermophilic fungi have a strong capacity to degrade polysaccharide constituents in plants, therefore having potential for biotechnological applications such as bioconversion of plant biomass into animal feed, plant fertilizers and chemicals for the food industry [De Faria et al., 2004].

The goal of the study was to reveal the extremophilic by temperature (thermopiles, thermotolerants and psychrophiles) strains of microscopic fungi among the collection of cultures, isolated from different type soils of dry subtropical-step zone of Sighnaghi (east Georgia); also to establish the extreme and optimal ranges of temperature for growth and spore-formation of micromycetes.

Materials and Methods

Cultures of microscopic fungi from the collection of the laboratory of biotechnology of S. Durmishidze Institute of biochemistry and biotechnology served as investigation objects.

The microscopic fungi were grown on the universal nutrient medium: wort (content of sugar 7.0%)-1.0l, agar-20.0.

Surface cultivation of cultures was performed on Petri dishes at temperature range -0°C - 60°C with 5°C intervals.

The growth of microscopic fungi was determined by means of measuring two parameters – the diameter of the colony in two perpendicular directions after 3-fold cultivation, 3, 5 and 7 days later. On the other hand the density of hyphae of the developing colonies in different parts was measured. The final sum of both parameters was appreciated via 3-point system.

Results and Discussion

Experiments were done on the cultures of microscopic fungi isolated from brown carbonate, chestnut and meadow-chernozem soils of Sighnaghi region. The collection consisted of 70 cultures representing 10 different genera of the 3 main classes of microscopic fungi, in particular: Zygomycetes (*Mortierella, Mucor, Rhizopus*), Ascomycetes (*Aspergillus, Penicillium, Chaetomium*) and Deuteromycetes (*Botrytis, Cladosporium, Trichoderma*) [Daushvili et al., 2004].

Microscopic fungi were grouped as thermophiles (obligative thermophiles), thermotolerants (facultative thermophiles) and psychrophiles (facultative psychrophiles) following Cooney and Emerson.

Microscopic fungi with not less than 20°C minimal growth temperature and with maximum at 50°C, were regarded as obligative thermophiles. The facultative thermophiles grew at lower than 20°C and at higher than 50°C. Facultative psychrophyles grew at 0°C and lower temperature and at the same time at 20-25°C too.

According to experimental results 20.0% of the studied fungi were extremophiles by temperature (Fig. 1). Since the tropical and subtropical climatic zones represent a favorable habitat for thermophilic fungi, 14.2% of obligate and facultative thermophiles is a natural phenomenon for Sighnaghi region soils [Bilay, 1985]. Presence of psychrotrophic micromycetes was less expectable here, while existence of facultative psychrophiles may be explained by their tolerance to moderate temperatures (20°-25°C).

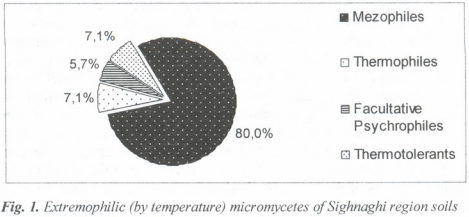


Fig. 1. *Extremophilic (by temperature) micromycetes of Sighnaghi region soils*

Majority of thermophilic and psychrophilic microscopic fungi were isolated from meadow-chernozem soils. This may be explained by the fact that this type of soil was distinguished with abundance and diversity of genera of microscopic fungi.

Among 10 studied genera of fungi thermophilic features were revealed only in genera: *Aspergillus, Chaetomium* and *Penicillium*. Psychrophiles were found among the genera: *Mortierella, Mucor* and *Cladosporium*. Majority of obligate and facultative thermophiles belonged to *Aspergillus* genus (Fig. 2).

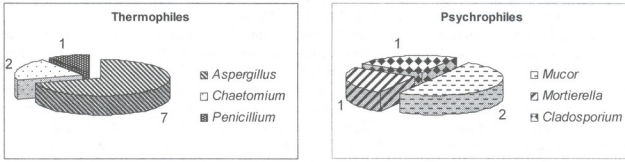


Fig. 2. *Genera of extremophilic fungi of Sighnaghi region soils*

The temperature amplitudes for optimal growth of thermophilic and thermotolerant microscopic fungi were established (Table 1). The temperature amplitude of optimal growth distinguished the thermotolerant representatives of *Aspergillus* genus. More over, the morphological changes, caused by the approaching the extreme temperature limit were less evident, while in genera *Chaetomium* and *Penicillium* the changes were clearly revealed. These morphological changes were mainly expressed in significant decline of spore-formation and transformation of colony color. In some cases changes of hypae length and surface consistence were observed.

Table 1. The optimal temperature ranges for growth of thermophilic fungi.
1 - *Chaetomium sp. S77,* 2 - *Aspergillus sp. S73,* 3 - *Aspergillus niger S60,* 4 - *Aspergillus niger S64,* 5 - *Aspergillus niger S65,* 6 – *Penicillium sp. S57,* 7 – *Chaetomium sp. S67,* 8 - *Aspergillus sp. S51,* 9 - *Aspergillus sp. S52,* 10 - *Aspergillus sp. S58.*

Minimum	Optimal temperature for growth							Maximum
22, 23				1				53, 55
23, 24				2				53, 55
17, 19				3				54, 55
17, 18				4				53, 54
23, 24				5				55
15, 16				6				54
15, 17				7				53
17, 19				8				55
22, 23				9				53, 54
23				10				51, 52
	17-19	28-30	38-40	41-42	44-45	47-48	51-52	

Temperature °C

References:

Bilay T. *Thermophilic fungi*. Kiev, 1985 (in Russian).

Bilai T. *Thermostable enzymes of fungi*. Kiev, 1979 (in Russian).

Daushvili L., Kutateladze L., Burduli T., Jobava M., Dzalamidze I., Aleksidze T., Tinikashvili L. *Isolation of microscopic fungi from soils of different regions of the Caucasus*. Proc. Georgian Acad. Sci., Biol. Ser. B. **2**, 5-6, 68-73, 2004.

De Faria F., Te'o V., Begquist P., Azevedo M., Nevalainen K. *Expression and processing of a major xylanase (XYN2) from the thermophilic fungus Humicola grisea var. thermoidea in Trichoderma reesei*. Letters in Appl. Microbiology, **34**, 119-123, 2002.

Kunamneni A., Kumar K., Singh S. *Response surface methodological approach to optimize the nutritional parameters for enhanced production of α-amylase in solid state fermentation by Thermomyces lanuginosus*. African Journal of Biotechnology, **4,** 7, 708-716, 2005.

Phutela U., Dhuna V., Sandhu Sh., Chadha B.S. *Pectinase and polygalacturonase production by a thermophilic Aspergillus fumigatus isolated from decomposting orange peels*. Brzailian Journal of Microbiology, **36**, 63-69, 2005.

სიღნაღის რეგიონის ნიადაგებში გავრცელებული თერმოფილური მიკროსკოპული სოკოები

დაუშვილი ლ., ბურდული თ., ქუთათელაძე ლ., ჯობავა მ., ძალამიძე ი.

ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 14.08.2006)

რეზიუმე

შესწავლილია სიღნაღის რეგიონის მშრალი სუბტროპიკული სტეპის ზონის ნიადაგებიდან გამოყოფილი მიკროსკოპული სოკოების 70 კულტურა. გამოვლენილია 5 თერმოფილური, 5 თერმოტოლერანტული და 4 ფსიქროფილური (ფაკულტატური) მიკრომიცეტი. დადგენილია თერმოფილური მიკრომიცეტების ზრდისა და სპორაწარმოქმნის ოპტიმალური ტემპერატურული ამპლიტუდები.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

STUDY OF ANTIBIOTIC AND PHAGE SENSITIVITY OF SOME AEROBIC PYOGENIC BACTERIA

GIORGADZE M.[1], NATIDZE T.[2]

[1]*Ltd "Doagnosis-90"*
[2]*Ltd "Immunogen"*

(Received May 15, 2006)

Abstract

It was shown that isolates of streptococcus, staphylococcus and Escherichia coli isolated from suppurative inflammation areas and from the blood of the dog diseased with sepsis are more sensitive to enco- and intestibacteriophages, as compared to antibiotics. At the same time lysis degree is mostly equaled to 4+ and 3+. The obtained results revealed outlook for usage of bacteriophages for treatment of infectious diseases of bacterial etiology in dogs, as preparation without side effects.

Key words: suppurative inflammation of skin, enco- and intestibacteriophages

Introduction

Forming of microbial populations resistant to antibiotics is the actual problem of medicine and veterinary. This phenomenon, which is mainly caused by specific plasmids occurring in bacteria, is connected with purposeless and sometimes uncontrolled usage of antibiotics. Usage of some antibiotics, due to resistance against them, is noneffective and improper.

Medical and veterinary practice demands to look for new antibiotics and to work out alternative preparations. Such preparation is a bacteriophage, which is successfully used in medical practice for treatment of infectious diseases, and among them for suppurative inflammation of skin. Priority of bacteriophages compared to antibiotics is impossibility of forming of resistant populations, absence of allergy, reproduction in the nidus of infection, rapidity of preparation, etc. [Barrow, Soothill, 1997; Carlton, 1999; Smith, Huggins, 1982; Marks, Sharp, 2000]

The aim of our work was comparative study of antibiotic and phage sensitivity of some aerobic pyogenic bacteria isolated from dogs sick with skin diseases (dermatitis, pyoderma), and from the dogs sick with sepsis.

Materials and Methods

In our experiments we used ten isolates of staphylococcus (*St. aurous* - 6, *St. epidermidis* – 4), six isolates of streptococcus (*Str. pyogenes* – 4, *Str.viridans* – 2), and seven isolates of Escherichia coli (*E.cili hemolytic* – 3, *E.coli nonhemolytic* – 4).

Antibiotic sensitivity was studied by disk-diffusion method, phage sensitivity – by Fisk modified method [Overturf et al., 1991].

In our study we used the following antibiotics: amoxacillin, ampicillin, ampicide, gentamicin, doxicicline, erythromycin, kanamycin, chloramphenicol, penicillin, streptomycin, tetracycline, triaxon, cipro-bai and ciprofloxacin.

Among bacteriophages were used the following ones: encophage, intestibacteriophage, pyophage, Sisphage, Fersisphage.

Results and Discussion

As a result of our studies it was established that the effect of antibiotics and phages on staphylococcus, streptococcus and Escherichia coli is different (Table 1). Some staphylococcus are polyresistant against antibiotics. For example, the majority among them revealed resistance against erythromycin, kanamycin, streptomycin, tetracycline, and partially against gentamicin and penicillin. Amoxicillin, ampicillin, ampicide, triaxon have high influence on staphylococcus. To those antibiotics *St.epidermis*-2 appeared to be resistant.

Table 1. Antibiotic and phage sensitivity of microbes

№	Microbes	Antibiotic													Bacteriophage					
		Amoxacillin	Ampicillin	Ampicide	Gentamicin	Doxicicline	Erythromycin	Kanamicin	Chloramphenicol	Penicillin	Tetracycline	Triaxon	Streptomycin	Cipro-bai	Ciprofloxacin	Encophage	Intestiphage	Pyophage	Sisphage	Fersisphage
1	*St. aureus - 1*	4+	4+	4+	4+	R	R	2+	4+	4+	R	R	3+	R	2+	4+	4+	4+	2+	2+
2	*St. aureus - 2*	4+	2+	2+	R	R	R	R	R	R	R	2+	2+	R	2+	3+	3+	2+	2+	2+
3	*St. aureus - 3*	4+	3+	4+	2+	3+	R	R	R	R	3+	4+	R	R	3+	2+	3+	2+	2+	R
4	*St. aureus - 4*	4+	4+	4+	3+	2+	R	2+	2+	4+	4+	4+	R	3+	3+	R	2+	2+	2+	2+
5	*St. aureus - 5*	4+	4+	4+	3+	4+	R	2+	4+	2+	R	3+	R	3+	4+	4+	4+	3+	R	3+
6	*St. aureus - 6*	4+	4+	4+	2+	2+	R	R	R	R	R	3+	R	3+	2+	2+	R	R	2+	2+
7	*St. epidermidis - 1*	3+	3+	2+	R	R	3+	3+	4+	2+	R	2+	2+	R	R	R	3+	2+	R	R
8	*St. epidermidis - 2*	R	R	R	R	R	R	R	3+	R	R	3+	R	R	2+	2+	2+	2+	2+	R
9	*St. epidermidis - 3*	4+	3+	-	3+	R	R	R	4+	4+	2+	4+	R	2+	4+	3+	2+	2+	2+	2+
10	*St. epidermidis - 4*	4+	4+	2+	2+	4+	R	R	R	3+	R	3+	R	3+	4+	2+	3+	3+	2+	2+
11	*St. pyogenes - 1*	3+	R	3+	4+	4+	-	2+	4+	2+	2+	4+	2+	4+	4+	3+	3+	2+	2+	2+
12	*St. pyogenes - 2*	4+	2+	4+	4+	4+	-	3+	4+	3+	3+	4+	3+	3+	4+	2+	3+	2+	R	2+
13	*St. pyogenes - 3*	4+	3+	4+	4+	4+	-	3+	4+	3+	3+	4+	3+	3+	4+	2+	2+	2+	R	R
14	*St. pyogenes - 4*	4+	3+	2+	3+	3+	2+	3+	4+	2+	3+	2+	2+	4+	3+	4+	4+	3+	2+	2+
15	*St. viridans - 1*	3+	3+	3+	3+	4+	2+	3+	4+	2+	4+	3+	3+	3+	3+	3+	4+	3+	2+	R
16	*St. viridans - 2*	3+	R	4+	3+	3+	2+	2+	3+	2+	4+	4+	2+	4+	4+	3+	3+	3+	2+	2+
17	*E. coli (H) - 1*	4+	4+	4+	3+	4+	R	2+	4+	R	4+	4+	2+	4+	4+	2+	3+	3+	2+	2+
18	*E. coli (H) - 2*	4+	4+	4+	3+	4+	R	4+	4+	R	4+	3+	3+	4+	3+	3+	3+	3+	3+	2+
19	*E. coli (H) - 3*	3+	3+	2+	4+	-	R	2+	4+	2+	3+	2+	3+	4+	2+	3+	2+	2+	2+	3+
20	*E. coli (NH) - 4*	4+	3+	2+	3+	-	R	3+	4+	3+	4+	3+	3+	3+	2+	2+	R	R	R	2+
21	*E. coli (NH) - 5*	4+	4+	2+	3+	-	2+	4+	3+	2+	4+	2+	3+	4+	3+	2+	3+	3+	2+	R
22	*E. coli (NH) - 6*	3+	2+	2+	4+	3+	3+	R	3+	3+	4+	3+	2+	4+	3+	2+	3+	3+	3+	2+
23	*E. coli (NH) - 7*	4+	4+	2+	3+	4+	4+	3+	3+	R	3+	3+	2+	3+	3+	3+	3+	2+	3+	2+

Note: R – resistant; "-" – not studied; H – hemolytic; NH – nonhemolytic.

Staphylococcus, as compared to antibiotics, lyses intensively encophage and intestibacteriophage. It should be mentioned that poly-antibiotic resistant staphylococcus (*St.aureus*-2, *St.aureus*-3, *St.epidermidis*-1, *St. epidermidis*-3, *St.epidermidis*-4) in most cases are lysed by bacteriophages in various degrees. Enco- and intestibacteriophage are especially characterized by this feature.

Unlike staphylococcus, streptococcus are more sensitive to the influence of antibiotics and bacteriphages. Among antibiotics with especially intensive effect are distinguished: amoxacillin, ampicillin, ciprofloxacin, triaxon and cipro-bai, (4+, 3+), and among bacteriophages – enco- and intestibacteriophages (4+, 3+, 2+).

Antibiotic- and phage-sensitivities of Escherichia coli are nearly similar. For example, *E.coli*-4, which is resistant against erythromycin, is sensitive against enco- and Fersisphage, and *E.coli-7*, which is resistant against penicillin, turned out to be sensitive against all bacteriophages. In other cases E.coli strains are sensitive to antibiotics (amoxacillin, ampicillin, ciprofloxacin, cipro-bai) and to bacteriophages, especially to enco- and intestiphages (3+, 2+).

Thus, Sensitivity of staphylococcus, streptococcus and Escherichia coli to antibiotics and phages is different. Staphylococcus shows distinct resistance to antibiotics. Strains are sensitive to enco- and intestibacteriophages that enables to use them as alternative agent.

References:

Barrow PA., Soothill JS. *Bacteriophage therapy and prophylaxis: rediscovery and renewed assessment of potential.* Trends Microbiol., **5**, 268-271, 1997.

Carlton RM. *Phage therapy: past history and future prospects.* Arch. Immunol. Ther. Exp., **47**, 267-274, 1999.

Marks T., Sharp R. *Bacteriophages and biotechnology.* J.Chem.Technolgy and Biotechnology, **75**, 1, 6-17, 2000.

Overtruf G.D., Talan D.A., Singer K. et al. *Phage Typing of Staphylococcus.* J.Clin.Microbiol., 29, 2, 373-375, 1991.

Smith HW., Huggins MB. *Successful treatment of experimental Escherichia coli infections in mice using phage: its general superiority over antibiotics.* J. Gen. Microbiol., **128**, 307-318, 1982.

ზოგიერთი აერობული ჩირქმბადი ბაქტერიის ანტიბიოტიკო- და ფაგომგრძნობელობის შესწავლა

გიორგაძე მ.[1], ნათიძე თ.[2]

[1]შპს „დიაგნოზი-90“;
[2]შპს „იმუნოვენი“

(მიღებულია 15.05.2006)

რეზიუმე

დადგენილია, რომ კანის ჩირქოვანი უბნებიდან და სეფსისით დაავადებული ძაღლის სისხლიდან გამოყოფილი სტრეპტოკოკების, სტაფილოკოკების და ეშერიხიების ი'ზოლატები ანტიბიოტიკებთან შედარებით გაცილებით მგრძნობიარეა ენკო- და ინტესტიბაქტერიოფაგების მიმართ. ამასთან, ლიზისის ხარისხი უმეტესად ტოლია 4+ და 3+. მიღებული შედეგები იძლევა ძაღლებში ბაქტერიოფაგის გამოყენების პერსპექტივას.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

DETERMINATION OF EFFECTIVE SCHEME OF ANTI-BRUCELLOSIS IMMUNIZATION OF HORNED CATTLE

GVINJILIA G., GVINJILIA M.

Laboratory of Ministry of Agriculture of Georgia

(Received August 10, 2006)

Abstract

Stability of immune response at vaccination of animals with strain-19 of anti-brucellosis vaccine was studied. Vaccination scheme of adult animals in stationary conditions was worked out. This regimen enables us to conduct animal vaccination by small dose once a year.

Key words: Rose-Bengal Reaction, Agglutination reaction, serological methods

Introduction

It is well known that Brucella is the microorganism causing worldwide spread disease – brucellosis. Brucella mainly infects horned cattle, sheep, goat, pig. Brucella infection can be also seen in wild species of animals [Callahan, 2006 a].

Brucellosis is a persistent disease conducted with elimination of microbe from reproductive system and mammary gland [Sigafoose, 2006]. On account of an economic impact connected with animal health and infection risk of humans the most countries have program of brucellosis control, which involves vaccination of young and adult animals by strain-19. Today, in epizootic and epidemiologic viewpoint the situation is alarm in Georgia [Callahan, 2006 b].

Thus, the goal of our work is to improve diagnostic methods, and special prophylactic agents, and schemes of their usage [Payeur, 2006]. We aimed to determine effectiveness of strain-19 and immune status of animal at various dosing regimen. One link of chain – recipient animal – must become as no recipient. It should be realized by usage of specific immunization; i.e. for extermination of brucellosis immune barrier should be used. Vaccinal prevention may directly set up the precondition of further irreversible liquidation of the disease.

Materials and Methods

88 calves of 4-6 months old and 2000 adult cows from safe on brucellosis farms were used in experiments. Their immunization with strain-19 was carried out in the following dosing regimes: 0.5 billions, 3 billions, 9 billions, and standard 80 billions of microbial cells.

The following standard serological methods were used:

1. Rose-Bengal Reaction (RBR),
2. Agglutination (Raite) reaction (AR)

Results and Discussion

The obtained data show that with the decrease of dose antigenic effect of preparation decreases, postvaccinal reactions disappear earlier than usual (Table 1.).

The obtained results reveal that calves, which were immunized with small dose of vaccine, maintain postvaccinal reactions during 4 months. The calves vaccinated with 9 and 80 billions of microbial cells maintain stable immune status during one year.

Among 22 calves immunized with 9 billion microbial cells 21 turned out to be positive. Animals immunized with standard dose show the same result.

On the second stage of study experiments were carried out on cows in order to determine postvaccinal state in adult cows. Experimental animals (2000 ones) were divided into two groups, vaccinated with 9 and 80 billion microbial cells, respectively.

As a result of serological investigations conducted after 4 months, as well as after 1 year it was revealed that postvaccinal reaction is positive during one year almost in all animals. Percentage of animals having positive reaction, which were vaccinated with 9 billion microbes consists of 92.7% at the end of year, but of animals vaccinated with 80 billion microbes – 93.3%.

Thus, we consider that high dosing regime is not needed, as post-immunization reactions are similar. The common scheme of anti-brucellosis vaccination of adult animals should be the following: cows being in stationary conditions must be vaccinated with the dose of 9 billion microbes once in a year. In that way we should prevent the problem of postvaccinal reaction without reduction of immunogenic characteristics of preparation.

Table 1. Results of serological studies carried out on 4-6 months old calves

Animal groups	Number of animals	Dose of immunization (billion microbes)	After 4 months		After year	
			positive	negative	positive	negative
1	22	0.5	12	19	-	22
2	22	3	15	7	2	20
3	22	9	21	1	21	1
4	22	80	22	-	21	21

Table 2. Results of serological studies carried out on adult caws

Number of animals	Dose of immunization (billion microbes)	After 4 month		After year	
		positive	negative	positive	negative
1000	9	990	10	927	73
1000	80	994	6	933	67

References:

Callahan J. *Brucellosis.* In: Collection of works in microbiology and virology. N24914-101, 2006a.

Callahan J. *Epidemiology of brucellosis.* In: Collection of works in microbiology and virology. N24914-102, 2006b.

Sigafoose T.M. *Brucellosis.* In: Collection of works in microbiology and virology. N24914-100, 2006.

Payeur J.B. *Laboratorial methods of brucellosis investigation.* In: Collection of works in microbiology and virology. N24915, 2006.

მსხვილფეხა რქოსანი პირუტყვის ბრუცელოზის საწინააღმდეგო იმუნიზაციის სქემის განსაზღვრა

ღვინჯილია გ., ღვინჯილია მ.

სხია სოფლის მეურნეობის სამინისტროს ლაბორატორია

(მიღებილია 10.08.2006)

რეზიუმე

შესწავლილია იმუნური პასუხის სტაბილურობა ცხოველის ორგანიზმში ბრუცელოზის საწინააღმდეგო ვაქცინის შტამი-19 მცირე დოზის შეყვანისას. შემუშავებულია ზრდასრული ცხოველების ვაქცინაციის სქემა სტაციონარულ პირობებში, რომელიც საშუალებას იძლევა განვახორციელოთ ცხოველთა ვაქცინაცია მცირე დოზით წელიწადში ერთხელ.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

STUDY OF THE EXTENT OF EXTREMOPHILICY OF HALOPHILIC MICROSCOPIC FUNGI FROM SALINE SOILS OF KAKHETI PLAIN

ZAKARIASHVILI N., *KUTATELADZE L., JOBAVA M., KHOKHASHVILI I., DZALAMIDZE I., KVESITADZE E., ALEKSIDZE T.

S. Durmishidze Institute of Biochemistry and Biotechnology
D. Agmashenebeli Alley 10km, 0059 Tbilisi, Georgia
**E-mail: kutateladze@geomail.ge*

(Received May 15, 2006)

Abstract

The extent of extremophilicy of halophilic microscopic fungi, isolated from saline soils of Kakheti plain has been investigated. The pH and temperature ranges of 44 cultures of fungi were established. The optimal pH and temperature of cultivation for each halophile has been selected. 23 extremophiles were revealed: 8 of them by temperature and 15 – by pH. 3 of them were thermophiles, 2 – psycrotolerants, 3 – thermotolerants, 10 – alkaliphiles, and 5 – pH-tolerants. 5 microscopic fungi – *Aspergillus sp.* A26, *Chaetomium sp.* A36, *Trichoderma sp.* A40, *Trichoderma sp.* A41 and *Fusarium sp.* A10 were regarded as extremophiles. They revealed extremophilic properties by three parameters simultaneously – temperature, pH and resistance to high concentrations of NaCl.

Key words: extremophiles, halophiles, thermophiles, pH-tolerant, microscopic fungi

Introduction

Determination of the limits of existence of living organisms is one of the central problems of present-days biology. Due to their high genetic and physiological adaptivity, microorganisms belong to the unique forms of life, managing to exist under the extreme environmental conditions (high and low temperatures, acid and alkali surrounding, high concentrations of salt, etc.) [Aguliar, 1996; Dix, 1995; Mouchacca, 1997; Stetter, 1999]. These types of microorganisms are named as extremophiles. They comprise 3 main groups: 1. thermophiles and psycrophiles 2. alkali- and acidophiles, 3. halophiles. Halophiles deserve a special interest among the extremophiles. Resistance to unfavorable environmental conditions, unique chemical structure and nonpathogenous properties of this group of microorganisms are responsible for involving them into the biotechnological processes. The halophilic cell is a "live laboratory", which makes possible to create several commercial products simultaneously (enzymes, nucleic acids, betonies, ectoines, carotinoids, etc.). The inhabitants of hyper-alkali soils are regarded as "rectifiers" of the environment polluted with organic toxins. In spite of the mentioned biopotential, the abilities of halophils are less used in large technological processes. From the practical experience it is clear that the stable enzymes, acting in extreme regimen (high temperature, alkali or acid surrounding)

possess great advantages in enzyme technology and biotechnology. Extremophiles are producers of this type of enzymes. According to this fact, selection of a high quality extremophiles among the halophilic microscopic fungi, isolated in our early experiments from saline soils of Kakheti plain [Laskhishvili et al., 2005], was of great interest.

Materials and Methods

44 microscopic fungi, isolated from saline soils of Kakheti plane served as objects for experiment. To select extremophiles, microscopic fungi were cultivated under the extreme conditions, on an universal, agar nutrient medium, containing the optimal concentration of NaCl. According to early experiments, the optimal concentrations of NaCl were established for particular halophile [Stetter, 1999].

Microscopic fungi were cultivated under the wide range of temperature (0-55°C, with intervals of 5°C), to reveal thermo- and psycrophiles. The wide amplitude of pH was tested to select acid and alkaliphiles (pH 2.0-10.0, with 0.5 intervals). Cultivation prolonged for 10 days. Values of temperature or pH, resulting in maximal increase, were regarded as optimal. Intensity of growth was evaluated using 3-mark system, taking into account the diameter and growth velocity of the colony of micromycetes.

While the thermophilicy was detected, the determinations offered by Quean and Emerson were used. In particular, the cultures of micromycetes with existence ranges from 20°C to 55°C were regarded as thermophiles. The cultures with maximal growth at ~50°C and able to develop at lower than 20°C were distinguished as thermotolerants.

Microscopic fungi growing at low temperature, but able to develop at 40°C too, were grouped as psycrotolerants.

Micromycetes well developing equally at pH ranging from 5 to 10 were regarded as alkaliphiles, while others, growing at a wide range of pH (2.0 - 10.0) were grouped as pH-tolerant.

Results and Discussion

While determining the extent of extremophilicy of halophilic microscopic fungi isolated from saline soils of Kakheti plane the existing ranges and optimal temperatures were established first of all. For this purpose halophiles were grown at universal agar nutrient medium with optimal concentrations of NaCl, at a wide range of temperature – from 0°C to 55°C, with 5°C intervals.

In table 1 the characteristics of microscopic fungi from saline soils of Kakheti plane are given following the temperature of cultivation. From the table it is clear that the majority of microflora consisted of mesophiles with optimal growing temperature 28-30°C. These meanings are in accordance with the literature data about the prevailing of mesophiles among the microorganisms in nature.

Some representatives of investigated strains sharply changed its morphological and cultural features while approaching the critical temperature. Representatives of different genera of fungi diversely reacted on a temperature fluctuations, e.g. degeneration of spores was mentioned in some species of *Fusarium* genus. The fluffy mycelium of several strains of the genus Mucor turned into skinny one. In some cultures of *Aspergillus* changes in colour or difficulties in spore merging was mentioned.

8 extremophiles by temperature has been selected among the microscopic fungi, isolated from saline soils of Kakheti plain. Among them 3 cultures – *Aspergillus sp.* A2, *A. sp.* A26, and *Chaetomium sp.* A36 were thermophiles, 2 – *Fusarium sp.* A10 and *F. sp.* A43 were psicrotolerants, and 3 – *Trichoderma sp.* A40, *T. sp.* A41 and *T. sp.* A42 were thermotolerants.

After arranging the extremophiles by temperature we aimed to reveal microscopic fungi, growing at extremal pH. For this purpose the cultures from the collection were grown on universal agar nutrient medium, with optimal concentration of NaCl and optimal temperature, changing the pH of the medium within wide range (from 2.0 to 11.0).

In Table 2 the halophilic microscopic fungi spread in saline soils of Kakheti plane are presented according to pH meanings. Among 44 strains of the collection, 15 turned to be halophiles. Between them 10 were alkaliphiles and 5 – pH-tolerant.

Table 1. The extent of extremophilicy of microscopic fungi from saline soils of Kakheti plane

Microscopic fungi	Temperature ranges of culture growth	Optimal temperature of growth, °C	Group of micromycetes by temperature	Relation of culture to NaCl concentrations
1. *Aspergillus sp. A-1*	15°C-40°C	30°C	mesophile	weak
2. *Aspergillus sp. A-2*	20°C-55°C	40°C-45°C	thermophile	weak
3. *Aspergillus sp. A-3*	15°C-45°C	28°C-32°C	mesophile	weak
4. *Aspergillus sp. A-4*	15°C-45°C	30°C	mesophile	weak
5. *Aspergillus sp. A-13*	15°C-40°C	28°C	mesophile	moderate
6. *Aspergillus sp. A-14*	15°C-45°C	25°C	mesophile	moderate
7. *Aspergillus sp. A-25*	15°C-40°C	30°C	mesophile	extreme
8. *Aspergillus sp. A-26*	20°C-55°C	40°C-45°C	thermophile	extreme
9. *Aspergillus sp. A-27*	15°C-40°C	30°C	mesophile	halotolerant
10. *Aspergillus sp. A-28*	15°C-45°C	30°C	mesophile	moderate
11. *Aspergillus sp. A-29*	15°C-45°C	30°C	mesophile	extreme
12. *Aspergillus sp. A-30*	15°C-45°C	30°C	mesophile	moderate
13. *Aspergillus sp. A-31*	15°C-45°C	25 °C -30°C	mesophile	halotolerant
14. *Penicillium sp.A-5*	15°C-45°C	25 °C -30°C	mesophile	weak
15. *Penicillium sp.A-6*	15°C-40°C	25 °C -30°C	mesophile	moderate
16. *Penicillium sp.A-15*	15°C-40°C	25 °C -30°C	mesophile	moderate
17. *Penicillium sp.A-16*	15°C-40°C	25 °C -30°C	mesophile	moderate
18. *Penicillium sp.A-17*	15°C-40°C	25 °C -30°C	mesophile	moderate
19. *Penicillium sp.A-18*	15°C-40°C	25 °C -30°C	mesophile	moderate
20. *Penicillium sp.A-19*	15°C-40°C	25 °C -30°C	mesophile	moderate
21. *Penicillium sp.A-20*	15°C-40°C	30°C	mesophile	halotolerant
22. *Penicillium sp.A-32*	15°C-45°C	30°C	mesophile	moderate
23. *Penicillium sp.A-33*	15°C-45°C	25 °C -30°C	mesophile	halotolerant
24. *Penicillium sp.A-34*	15°C-40°C	30°C	mesophile	halotolerant
25. *Penicillium sp.A-35*	15°C-40°C	30°C	mesophile	halotolerant
26. *Chaetomium sp. A-36*	20°C-55°C	40°C-45 °C	thermophile	halotolerant
27. *Chaetomium sp. A-37*	15°C-40°C	30°C	mesophile	halotolerant
28. *Chaetomium sp. A-38*	15°C-40°C	30°C	mesophile	moderate
29. *Chaetomium sp. A-39*	15°C-40°C	25 °C -30°C	mesophile	extreme
30. *Allescheria sp. A-11*	15°C-40°C	30°C	mesophile	weak
31. *Allescheria sp. A-12*	15°C-45°C	25 °C -30°C	mesophile	weak
32. *Cladosporium sp. A-21*	15°C-40°C	30°C	mesophile	moderate
33. *Cladosporium sp. A-22*	15°C-40°C	30°C	mesophile	moderate
34. *Fusarium sp. A-7*	15°C-40°C	25 °C -30°C	mesophile	weak
35. *Fusarium sp. A-8*	15°C-40°C	30°C	mesophile	weak
36. *Fusarium sp. A-9*	5°C-40°C	10°C-20 °C	mesophile	weak
37. *Fusarium sp. A-10*	5°C-40°C	10°C-20 °C	psicrotrophe (psicrotolerant)	halotolerant
38. *Fusarium sp. A-43*	15°C-45°C	30°C	mesophile	halotolerant
39. *Fusarium sp. A-44*	5°C-55°C	20°C-35 °C	thermotolerante	halotolerant
40. *Trichoderma sp. A-40*	5°C-55°C	20°C-35 °C	thermotolerante	halotolerant
41. *Trichoderma sp. A-41*	5°C-55°C	20°C-35 °C	thermotolerante	moderate
42. *Trichoderma sp. A-42*	15°C-45°C	20 °C -30°C	thermotolerant	halotolerant
43. *Mucor sp. A-23*	15°C-45°C	20°C -30°	mesophile	moderate
44. *Mucor sp. A-24*	15°C-45°C	20°C -30°	mesophile	moderate

Table 2. The extent of extremophilicy of microscopic fungi by pH

Microscopic fungi	Place of sampling	The range of living pH of the culture	Optimal pH	Characterization of culture
1. *A.sp.A25*	Soils with high salinity	5.0-10.0	9	alkaliphile
2. *A.sp.A26*		5.0-10.0	9	alkaliphile
3. *A.sp.A27*		4.5-8.5	5.0	-
4. *A.sp.A28*		4.5-7.5	5.0	-
5. *A.sp.A29*		2.5-10.0	4-8.0	pH-tolerant
6. *A.sp.A30*		4.0-7.5	6.0	-
7. *A.sp.A31*		2.5-10.0	4-8.0	pH-tolerant
8. *P.sp.A32*		4.5-7.5	6.0	-
9. *P.sp.A33*		2.5-10.0	3.5-7.5	pH-tolerant
10. *P.sp.A34*		4.5-10.0	9	alkaliphile
11. *P.sp.A35*		4.5-7.5	5.0	-
12. *Ch.sp.A37*		4.5-7.5	5.0	-
13. *Ch.sp.A36*		4.5-10.0	9	alkaliphile
14. *Ch.sp.A38*		5.0-8.0	6	-
15. *Ch.sp.A39*		4.5-7.5	6.0	-
16. *F.sp.A43*		5.0-7.5	6.0	-
17. *F. sp.A44*		4.5-7.5	5.5	-
18. *T. sp.A40*		4.5-10.0	9.0	alkaliphile
19. *T. sp.A41*		5.0-10.0	8.5	alkaliphile
20. *T. sp.A42*		4.5-8.0	6.0	-
21. *A. spA.13*	Soils with moderate salinity	5.0-8.0	6.0	-
22. *A. spA.14*		4.5-7.5	5.5	-
23. *P.sp.A15*		4.5-7.5	5.0	-
24. *P.sp.A16*		4.5-8.0	5.5	-
25. *P.sp.A17*		4.5-7.5	5.5	-
26. *P.sp.A18*		4.5-7.5	5.5	-
27. *P.sp.A19*		4.5-8.0	6.0	-
28. *P.sp.A20*		4.5-7.5	5.5	-
29. *Cl. sp.A21*		4.5-8.0	6.0	
30. *Cl. sp.A22*		4.5-7.5	6.0	-
31. *M. sp.A23*		4.5- 10.0	9.0	alkaliphile
32. *M. sp.A24*		4.5-10.0	9.0	alkaliphile
33. *A. sp.A1*	Soils with weak salinity	4.5-7.5	5.5	-
34. *A. sp.A2*		4.5-10.0	8.5	alkaliphile
35. *A. sp.A3*		4.5-7.5	5.5	-
36. *A. sp.A4*		4.5-7.5	6.0	-
37. *P.sp.A5*		4.5-7.5	5.5	-
38. *P.sp.A6*		4.5-8.0	6.0	-
39. *A. sp.A11*		5.0-8.0	6.0	-
40. *A.sp.A12*		4.5-7.5	6	-
41. *F. sp.A7*		2.5-10.0	4.5-8.0	pH-tolerant
42. *F. sp.A8*		2.5-10.0	4.5-8.0	pH-tolerant
43. *F. sp.A9*		4.5-7.5	6.0	-
44. *F. sp.A10*		4.5-10.0	8.5	alkaliphile

From the literature it is known that some representatives of *Aspergillus* and *Penicillium* genera are able to grow at a wide range of pH – from 2.0 to 10.0. The pH-tolerant microscopic fungi revealed in our experiments, belong also to these genera. Alkaliphils were found almost among all genera of fungi, except *Cladosporium* and *Allescheria* (Table 2). Extremophilic cultures by simultaneously three parameters (pH, temperature and NaCl concentration) were revealed on the base of selecting the extremophiles by pH and temperature. In particular, as high-quality extremophiles were evaluated: 1. thermophilic *Aspergillus sp.* A26, which is the extreme halophile and alkaliphile, at the same time. 2. *Chaetomium sp.* A36 – as halotolerant, alkaliphile and

thermotolerant. 3, 4. *Trichoderma sp.* A40 and *Trichoderma sp.* A41– as moderate halophile, alkaliphile and thermotolerant, and 5. *Fusarium sp.* A10 – as halotolerant, alkaliphile and psicrotolerant.

Selection of cultures, resistant to extreme conditions of temperature, pH and NaCl concentrations, and revealing the active producers of enzymes among them, is the perspective base for creating of new technologies.

References:

Aguliar A. *Extremophile research in the Europian Union: from fundamental aspects to industrial expectation.* FEMS Microbiol. Rev. 18, 89-92, 1996.

Dix N. I., Webster I. F*ungal ecology.* Chapman & Hall, London, U. K., 1995.

Laskhishvili M., Kutateladze L., Zakariashvili N., et al. *Izolation and identification of microscopic fungi from salty soils of Kakheti.* Proc. Georg. Acad. Sci. Biol. Ser. B, 3, 4, 60-63, 2005.

Mauchacca I. *Thermophilic fungi: biodiversity and taxonomic status.* Cryptogam. Mycol., 18, 19-69, 1997.

Stetter K. O. *Extremophiles and their adaptation to hot environments.* FEBS Lett., 452, 22-25, 1999.

კახეთის ვაკის მარილიან ნიადაგებში გავრცელებული ჰალოფილური მიკროსკოპული სოკოების ექსტრემოფილობის ხარისხის დადგენა

ზაქარიაშვილი ნ., ქუთათელაძე ლ., ჯობავა მ., ხოხაშვილი ი., ძალამიძე ი., კვესიტაძე ე. ალექსიძე თ.

ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 15.05.2006)

რეზიუმე

განსაზღვრულია კახეთის ვაკის მარილიანი ნიადაგებიდან გამოყოფილი ჰალოფილური მიკროსკოპული სოკოების ექსტრემოფილობის ხარისხი. დადგენილია კოლექციის 44 კულტურის სახიცოცხლო pH-ისა და ტემპერატურული დიაპაზონი. თითეული ჰალოფილისთვის შერჩეულია კულტივირების ოპტიმალური პირობები – pH და ტემპერატურა. გამოვლენილია 23 ექსტრემოფილი (მათ შორის 8 ტემპერატურის მიხედვით, 15 – pH-ის მიხედვით): 3 თერმოფილი, 2 – ფსიქროტოლერანტი, 10 – ალკალიფილი და 5 – pH-ტოლერანტი. მაღალი ხარისხის ექსტრემოფილებად შეფასებულია 5 მიკროსკოპული სოკო, რომელიც ერთდროულად სამი პარამეტრით (ტემპერატურა, pH და NaCl-ის მაღალი კონცენტრაციებისადმი მდგრადობა) აშკარავნებდა ექსტრემოფილობას: *Aspergillus sp.* A26, *Chaetomium sp.* A36, *Trichoderma sp. A*40, *Trichoderma sp.* A41 და *Fusarium sp.* A10

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Paleobiology

THE MAIN STAGES OF DEVELOPMENT OF *HAMAMELIDACEAE* ON THE TERRITORY OF EURASIA

SHATILOVA I., RUKHADZE L., MCHEDLISHVILI N.

Georgian National Museum, Davitashvili Institute of Paleobiology

(Received November 6, 2006)

Abstract

About the fossil *Hamamelidaceae* a big factual material is accumulated. In Europe and non-tropical Asia the findings of these plants are more often connected with the deposits of Oligocene, Early and Middle Miocene. In Georgia the greatest number of *Hamamelidaceae* were found in the Upper Miocene deposits. This material is of great interest, as it reflects the latest stage of florescence of the family in such region of Eurasia, where at present its representatives are fully absent.

Key words: *Hamamelidaceae*, development history, Western Georgia, Europe, non-tropical Asia.

Introduction

During the process of study of the palynological complexes of Sarmatian and Meotian deposits of Western Georgia our attention was attracted by the abundance of pollen grains of family *Hamamelidaceae.* That stimulated us to lead the monographycal investigation of this taxon and to trace its history on the territory of Eurasia. The big material was analyzed as nearly all Paleogene and Neogene floras described in literature contain one or another genus of *Hamamelidaceae*, except *Liquidambar*, which is the constant component of Cenozoic floras [Shatilova, Stuchlik, 2001].

Materials and Methods

In Georgia the earliest discoveries of *Hamamelidaceae* are dated by Paleogene. In the composition of Eocene, Oligocene, Early and Middle Miocene floras four genera (*Hamamelis, Corylopsis, Sycopsis, Liquidambar*) are known [Panova et al., 1984; Ramishvili, 1982].

In Late Miocene (Sarmatian, Meotian) the part of *Hamamelidaceae* in composition of flora increased and the family was represented by the following genera: *Hamamelis, Corylopsis, Eustigma, Fortunearia, Fothergilla, Parrotiopsis, Parrotia, Sycopsis, Distyliopsis, Distylium, Disanthus, Chunia, Liquidambar, Altingia.* The list is given by system of Endress [Endress, 1989].

All representatives of family *Hamamelidaceae* were probably the components of subtropical forests of plains and lower mountain belt (Fig.1.). In their composition both, evergreen and deciduous plants occur: *Carya, Lauraceae, Myrica, Quercus, Castanopsis, Araliaceae,* etc. [Kolakovsky, Shakryl, 1976; Shatilova et al., 1999].

After the Sarmatian the great number of subtropical forms died out. But this process didn't touch the family *Hamamelidaceae*, which in Meotian time continued to preserve the rich systematical composition. Between Sarmatian and Meotian some differences revealed in generic composition of family, but the number of genera was the same.

The main way of development of Pontian and Kimmerian vegetation was the widening of the area of deciduous cenosis and reduction of subtropical forests, which at the end of Middle Pliocene (Kimmerian) finished to exist as a separate formation. Subtropical plants preserved on the territory of Western Georgia after the Kimmerian became the components of warm-temperate forests. The single representatives of family *Hamamelidaceae (Liquidambar, Altingia, Corylopsis, Fortunearia, Parrotia)* were referred to such plants.

Results and Discussion

In the history of development of *Hamamelidaceae* on the territory of Georgia three stages can be distinguished. The initial one began after the first appearance of the taxon in geological chronology and continued till the time of its florescence. In Georgia it was the time of Paleogene, Lower and Middle Miocene. The second stage corresponded to the period of florescence of family. It was comparative short and embraced the Late Miocene (Sarmatian, Meotian). The third stage (time of decline of *Hamamelidaceae* and their extinction) was longer and corresponded to the whole Pliocene, Early and Middle Pleistocene.

The analysis of rich scientific literature shows that the first stage, during which the process of increasing of the systematical composition of *Hamamelidaceae* was going, finished in the Mesozoic in the non-tropical Asia. In Europe it was continued till the end of Eocene. During this time the representatives of *Hamamelidaceae* did not play significant role in plant communities, main components of which were palms and different *Lauraceae* [Sinitsin, 1980].

The second stage corresponded to the time of florescence of *Hamamelidaceae*, occurred at the end of Palaeogene and at the beginning of the Miocene in non-tropical Asia. In the Eocene, as a result of temperature fall, the flora of Early Palaeogene disappeared giving way to Turgaian flora. The plants forming the nucleus of this flora were concentrated in the southern mountainous regions on the border of Tethysian district. They revealed great tolerance spreading north and south along the mountain ranges and turned out to be capable to survive the drop of the temperature, which occurred at the boundary of Eocene and Oligocene [Meyen, 1987]. Representatives of *Hamamelidaceae* were among such plants. The great majority of them were shrubs ensuring their survival.

According to V. Sinitzin [1980], at the end of Late Oligocene all tropical forms (*Palmae, Proteaceae, Lauraceae, Myrtaceae*) were extinct in Asia, but warm-temperate trees as *Liquidambar, Liriodendron, Nyssa, Rhus, Magnolia*, characteristic for forests of Central-Chinese floristic province at present, continued to exist in the mesophylous forests of Siberia and in North-Eastern part of Asia.

By the end of Palaeogene and during the Miocene the Turgaian flora spread to the south and south-western (in Europe), replacing the retreating subtropical vegetation [Sinitsin, 1980]. After this time the florescence of *Hamamelidaceae* began on the territory of Europe. Here the typical polydominant forests, preserved after the Oligocene, were distributed. Judging from the localities of fossil remains of *Hamamelidaceae*, they were mainly connected with swamp forests. This formation was wide distributed in Central and South-Eastern Europe.

The third stage of development of *Hamamelidaceae*, which corresponds to the period of extinction, began in Early Miocene and proceeded rather rapidly in Asia. In Europe this process began in Late Miocene and proceeded gradually.

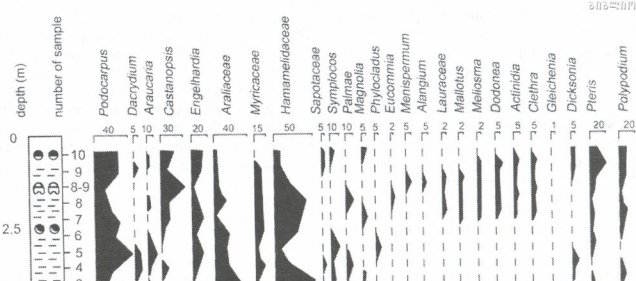


Fig.1. The percentage composition of subtropical plants of the components of lower mountain belt forests on the territory of Western Georgia in Late Miocene.

Conclusion

So, in the history of family *Hamamelidaceae* on the territory of Georgia, Europe and non-tropical Asia three main stages can be distinguished. In the several regions of Eurasia some phases of these stages were nonsynchronous and occupied different stretches of geological time. In Asia and Europe the evolution of *Hamamelidaceae* was closely related with the history of Turgaian flora. In Georgia the development of this family proceeded against the background of evolution of subtropical vegetation, determined the landscape of plain and lower mountain belt. Due to isolate position of Western Georgia, this formation preserved longer, than in other regions of Eurasia and should be traced till the end of Middle Pliocene.

References:

Endress P.K.. *A suprageneric taxonomic classification of the Hamamelidaceae.* Taxon, **38**, 3, 271-376, 1989.

Kolakovskiy A.A., Shakryl A.K. *Sarmatian floras of Abkhazeti.* Proceedings of Sokhumi Botanic Garden, **XXII**, 98-148, 1976 (in Russian).

Meyen S.V. *Basis of Paleobiology.* Moscow, 1987 (in Russian).

Panova L.A., Maligonova E.Yu., Tabachnukova I.P. *Miospore and nannoplankton of Eocene-Oligocene deposits of the north side of Akhaltsikhe depression. Spores and pollen in the deposits of phanerozoic.* Proceedings of VSEGEI, Nov. Ser., **327**, 74-93, 1984 (in Russian).

Ramishvili I.Sh. *Middle Miocene floras of Georgia according to the data of palynological analysis.* Tbilisi, 1982 (in Russian).

Shatilova I., Stuchlik L. *On the history of the development of family Hamamelidaceae.* Palaeontographica, Abt. **B**, Bd. **259**, 235-244, 2001.

Shatilova I.I., Rukhadze L.P., Mchedlishvili N.Sh., Makharadze N.V. *The main stages of development of vegetation and climate of Western Georgia.* Problems of Paleobiology, Tbilisi, **I**, 27-44, 1999 (in Russian).

Sinitsin V.L. *Natural conditions and climate of the territory of USSR in early and middle cenozoic.* Leningrad, 1980 (in Russian).

ოჯახ *Hamamelidaceae*-ს წარმომადგენლები დასავლეთ საქართველოს ნეოგენურში

შატილოვა ი., რუხაძე ლ., მჭედლიშვილი ნ.

საქართველოს ეროვნული მუზეუმი, ლ. დავითაშვილის პალეობიოლოგიის ინსტიტუტი

(მიღებულია 06.11.2006)

რეზიუმე

დღეისათვის დაგროვილია მდიდარი ფაქტიური მასალა ოჯახ *Hamamelidaceae*-ს შესახებ. ამ მცენარეთა ნამარხ ნაშთებს ევროპასა და არატროპიკულ აზიაში ყველაზე ხშირად პოულობენ ოლიგოცენურ, აღრეულ და შუა მიოცენურ ნალექებში. საქართველოში, ძირითადად, მას დასავლეთ ნაწილში, ე.წ. კოლხეთის რეფუგიუმში ამ ოჯახის სხვადასხვა გვარების წარმომადგენლები აღმოჩენილია გვიანმიოცენური (სარმატული, მეოტური) ფლორის შემადგენლობაში. აღნიშნული მასალა ძალზე მნიშვნელოვანია, რადგან იგი ახახავს ოჯახ *Hamamelidaceae*-ს ბატონობის ბოლო ეტაპს ევრაზიის იმ რეგიონში, სადაც დღეს ისინი სრულიად აღარ გვხვდება.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Plant Physiology

THE EFFECT OF BORON, ZINC AND MANGANESE ON THE ACTIVITY OF AMYLASES IN THE SEEDS OF *RAPHANUS SATIVUS, SPINACIA OLERACEA AND CORIANDRUM SATIVUM*

MANGALADZE N., TUTBERIDZE R., KILADZE N.

A. Tsereteli Kutaisi State University

(Received September 18, 2006)

Abstract

Seeds of *Raphanus sativus, Spinacia oleracea and Coriandrum sativum* were processed for 24 hours by 0.02% solutions of $ZnSO_4$, $KMnO_4$ and H_3BO_3. Control seeds were kept in distillate for the same time. To determine amylase activity on the second day of seed germination photocalorimetric method was used. At the seed processing with $ZnSO_4$, $KMnO_4$ and H_3BO_3 it was shown that the activity of α- and αβ-amylases were increased. Boron is suggested to have common positive effect on the amylase activity in seeds of all experimental species.

Key wards: microelements, amylase activity, photocalorimetric method

Introduction

It is well established that microelement deficiency may lead to alimentary diseases of plants [Braun et al., 1962; Katalimov, 1956; Marschner, 1995; Snowball et al., 1980]. Modern authors confirm that microelements are necessary for seed germination, development of vegetative organs, florescence and fruitage. There are some data indicating that microelements take significant role in the formation of anatomical structure of plant organs during the metabolic processes [Szpunar, 2004; Wang et al., 2003; Williams, 2001].

The present work was aimed to examine the role of microelements in the activity of enzymes in the plant cell. Particularly, the effect of boron, zinc and manganese on the activity of amylases in the seeds of *Raphanus sativus, Spinacia oleracea and Coriandrum sativum* was studied.

Materials and Methods

Changes of activities of amylases in the seeds of radish, spinach and coriander treated with solutions containing boron, zinc and manganese were examined. Seeds (300 of each plant) were placed on filtration paper treated with 0.02% solution of microelements for 24 hours. Particularly, seeds (100 ones from every plant) were processed in the $ZnSO_4$, $KMnO_4$ and H_3BO_3 solutions. Control seeds (100 ones from every plant) were set on filtration paper impregnated by distilled water for 24 hours. After processing seeds were placed on Petri dishes. On the second day after appearance of the sprout tips the activities of amylases were determined.

The enzymes were isolated in NaCl solution, incubated in standard starch solution; amount of starch unhydrolyzed by amylases was determined calorimetrically. As a result of hydrolysis and phosphorolysis starch is degraded to monosaccharides during seed germination. α and αβ-amylases, glycoamylase and aminopectin-1-6-glycosidase take part in hydrolysis. During swelling and germination of dry seeds hydrolysis activity of enzymes increases and as a result, starch content decreases and sugar content increases. Activity of amylases is evaluated by hydrolyzed starch in milligrams.

For isolation of amylases 4 g of germinated seeds were put in the mortar, 15 ml of 1% NaCl solution was added and crushed up to homogeneous material. Homogenate was put in the tubes cooled in fridge and centrifuged for 15 min. Substrate was prepared as follows: two tubes (each of 10 ml) were filled with 3 ml of acetate buffer (pH - 5.5) and 3 ml of 2% starch solution, mixed and heated up to 40°C. Further 0.5 ml enzyme preparation was added to one tube, and 5 ml of distilled water - to another one. Content of tubes was heated for 30 min. Then 2 ml of 0.1% of NaCl solution was added to the tubes, mixed and 0.5 ml of solution was taken from each one. Samples were brought into the flasks filled with 25 ml water. 1 ml of 0.1% of NaCl solution, 5 drops of 0.3% of iodine solution were added to the flasks and filled with water up to 50 ml and mixed. Solutions were examined on dyeing in the calorimeter. Red colour filter was used. Amylase activity was calculated by the formula: $A=E_c-E_0 / E_c \times 2.2/60$, where A enzyme activity, E_c and E_0 - optical density of control and experimental solutions, 2.2 coefficient per 1 ml of enzyme solution, 60 - coefficient per 1 mg starch (3 ml of 2% solution corresponds to 60 ml). The obtained data were processed by Student statistical method.

Results and Discussion

As is seen from Fig. 1 activities of α-amylase in the seeds of radish processed with boron and manganese were increased. Difference between activities of the enzyme of the seeds processed by zinc solution and the control is not statistically true. At the same tame processing by boron had stronger influence on the activity of α-amylase compared to manganese. Treatment of the radish seeds with all three experimental solutions increased activity of α-amylase as compared to the control. Processing with the boron caused the strongest effect.

Processing of spinach seeds with all three experimental solutions raised α-amylase activity, though difference with the control is reliable only in the cases of zinc and manganese. At the same time difference in the effects of zinc and manganese is not statistically true.

Processing of coriander seeds with experimental solutions caused statistically true increase in activity of α-amylase in cases of boron and zinc.

As a whole, treatment of radish seeds with H_3BO_3 showed the highest influence on the activity of α-amylase. The effect of $ZnSO_4$ was the biggest in case of spinach seeds, and $KMnO_4$ - in the seeds of radish and spinach.

While processing of radish seeds with experimental solutions activity of αβ-amylase in all three cases compared to control was increased. Differences between effects of boron, zinc and manganese solutions are unreliable. At the same time, activity of αβ-amylase increased much more than activity of α-amylase.

Activity of αβ-amylase in spinach seeds was increased in all three cases, though the strongest, statistically reliable effect was revealed in case of $KMnO_4$. Distinction between effects of H_3BO_3 and $ZnSO_4$ is not significant.

Activity of αβ-amylase in coriander seeds was increased in the cases of H_3BO_3 and $ZnSO_4$. Effect of $KMnO_4$ did not differ reliably from control. The highest influence had $ZnSO_4$.

Activities of amylases play significant role in the process of seed germination, as these enzymes promote degradation of starch and consumption of sugars by plant cells. Obtained results enable to consider that microelements, boron zinc and manganese make favour for increasing amylase activities. At the same time activity of αβ-amylase compared to α-amylase appeared to be more dependant from the presence of boron, zinc and manganese in the cells. Microelements turned to have species-specific character for amylase activities. α-amylase activities in radish and coriander are increased greatly at the effect of boron, but in spinach - at the effect of zinc. Activity of αβ-amylase in radish is equally dependant on boron, zinc and manganese, while in spinach the highest effect revealed - manganese, and in coriander - boron. Effect of boron on amylase activities is nearly similar to the effects of zinc and manganese. In a whole, for germination of seeds of radish, spinach and coriander presawing processing of seeds with the H_3BO_3 solutions should be considered as the most desirable.

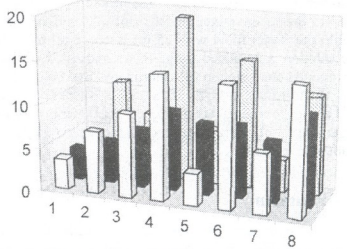


Fig. 1. Activities of amylases in the seeds of *Raphanus sativus, Spinacia oleracea* and *Coriandrum sativum.* 1st row - radish, 2nd row - spinach, 3rd row -coriander; 1 and 2 - control, 3 and 4 - with boron, 5 and 6 with zinc, 7 and 8 - with manganese; in all three cases the first column corresponds to α-amylase and the second column - to αβ-amylase.

References:

Braun T., Zister K., Tanner G. *Physiological consequences of manganese deficiency.* In: Microelemets. M.-L., "Nauka", 1962 (in Russian).

Katalimov M.V. *Microelement content in plants according to species characteristics and features.* In: Microelements in agriculture and medicine. Riga, 1956 (in Russian).

Marschner H. *Mineral nutrition of higher plants.* ed. 2nd, Academic Press, London, 1995.

Snowball L., Robson A.D. Lorenagan. *The effect of cooper on nitrogen fixation in subterranean clover (Trifolium subterraneum).* New Phytol., **85**, 63-72, 1980.

Szpunar J. *Metallomics: a new frontier in analytical chemistry.* Anal. Bioanal. Chem., **204**, 54-56, 2004.

Wang R., Okamoto M., Xing X., Crawford N.M. *Microarray analysis of the nitrate response in Arabidopsis roots and shoots reveal over 1000 rapidly responding genes and new*

linkages to glucose, trehalose-6-phosphate, iron, and sulfate metabolism. Plant Physiol., **132**, 556-567, 2003.

Williams R.J.P. *Chemical selection of elements by cells.* Coordin.Chem.Rev., **216-217**, 583-595, 2001.

ბორის, ცინკის და მანგანუმის ზეგავლენა ამილაზას აქტივობაზე *Raphanus sativus, Spinacia oleracea and Coriandrum sativum* თესლებში

მანგალაძე ნ., თუთბერიძე რ., კილაძე ნ.

ა. წერეთლის ქუთაისის სახელმწიფო უნივერსიტეტი

(მიღებულია 18.09.2006)

რეზიუმე

Raphanus sativus, Spinacia oleracea and Coriandrum sativum-ის თესლები დამუშავებულ იქნა 24 საათის განმავლობაში $ZnSO_4$, $KMnO_4$ და H_3BO_3 0.02% ხსნარებით. საკონტროლო თესლები იგივე დროით თავსდებოდა დისტილირებულ წყალში. განსაზღვრულია ამილაზას აქტივობა თესლის გაღივებიდან მეორე დღეს ფოტოკალორიმეტრული მეთოდით. ნაჩვენებია, რომ $ZnSO_4$, $KMnO_4$ და H_3BO_3 -ით დამუშავებული თესლების α- და αβ- ამილაზას აქტივობები იზრდება. გამოვლენილია, რომ ბორი დადებითად მოქმედებს ამილაზას აქტივობაზე სამივე სახეობის თესლში.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Plant Physiology

INFLUENCE OF SIMULATED ACID RAINS ON PHYSIOLOGICAL INDICES OF WHITE AND RED FORMS OF CABBAGE *(BRASSICA CAPITATA* L.)

RAPAVA L., CHANISHVILI SH., BADRIDZE G., BARBLISHVILI T., ABRAMIDZE S.

N. Ketskhoveli Institute of Botany

(Received October 10, 2006)

Abstract

Effect of pH2.5 water solution of sulphuric acid on seeds and leaves of red and white forms of cabbage (*Brassica capitata* L.) has been studied. Investigated physiological indices – photosynthetic activity of leaves, total activity of growth regulators, dry matter accumulation and water content have revealed higher sensitivity of white form of cabbage to simulated acid precipitations, while red form turned out to be more resistant to acid treatment. This may be due to the high content of anthocyanins in leaves of red cabbage.

Key words: acid rains, photosynthesis, dry matter accumulation, water content, cabbage

Introduction

Increasing of environmental acidity remains one of the most important ecological problems. The influence of acid rains has been mainly studied on woody plants for years [Eds. Muller C. et al., 1999]. During the last period scientists' attention has been focused on investigation of growth and productivity of cultivated plants under the influence of polluted environment [Evans et al., 1986; Hippeli, Elster, 1996].

Phytotoxic effect of polluted environment manifests itself through the changes in plant appearance and dry matter accumulation [Olson et al., 1987]. Therefore, it has been established that plants possess the ability of partial compensation of the primary effect of acid precipitation in the course of development, thus avoiding productivity decrease [Jay et al., 1987]. The opinion exists that plant organism is more resistant to acid pollution at early stages of development [Adams, Hutchinson, 1987].

Proceeding from the above mentioned the objective of our investigation was to study the influence of simulated acid precipitations on seeds and leaves of different varieties of the same species of cultivated plants.

Material and Methods

White and red forms of cabbage (*Brassica capitata* L.) were selected as test objects.

Anthocyanins are known to be one of the principal antioxidant substances, protecting plant organism against different environmental stresses like radiation, O_3, acid rains etc. [Filippovich, et al., 1975]. As the red form of cabbage is rich in substances of anthocyanic nature, presumably it

might be resistant to acid pollution. The comparative study of the effect of acid rains on two forms of cabbage served as a reason of their selection as test objects.

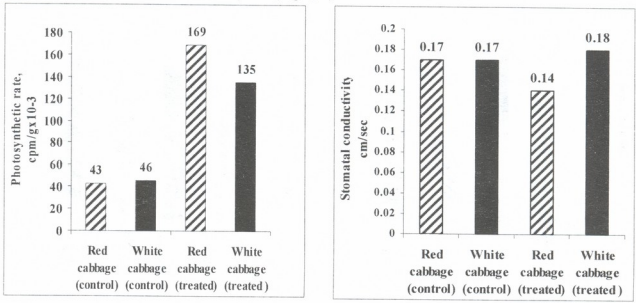
Water solution of sulphuric acid with pH2.5 was used for treating experimental seeds and plants. Cabbage seeds were soaked in acid solution for 24h. In plants, emerged from acid-soaked seeds photosynthetic activity [Voznesensky et al., 1965], stomatal conductivity and total activity of growth regulators (Kefeli, Turetskaya, 1966] were measured.

In other series of experiments leaves of red and white varieties of cabbage plants of the first year of vegetation were sprayed with acid three times with five days interval. Control plants were treated with the same amount of tap water. Material for analysis was taken 10 days after the last spraying.

In addition to the abovementioned indices (photosynthetic activity, stomatal conductivity, total activity of growth regulators) in cabbage plants sprayed with simulated acid rain some indices of water regime and dry matter accumulation were also determined.

Results and Discussion

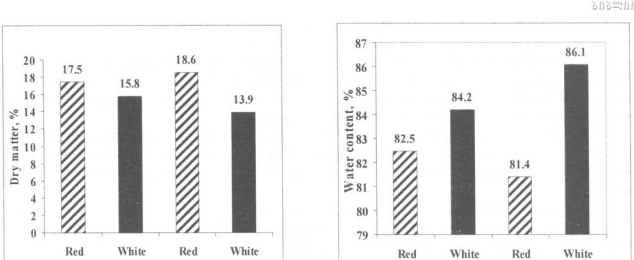
The obtained results show that significant intensification of photosynthetic rate took place in leaves of cabbage plants, emerged from acid-soaked seeds (Fig.1, a). The effect was more pronounced in red form of cabbage. No essential differences were found in stomatal conductivity of control and experimental variants of plant (Fig 1, b).



a b

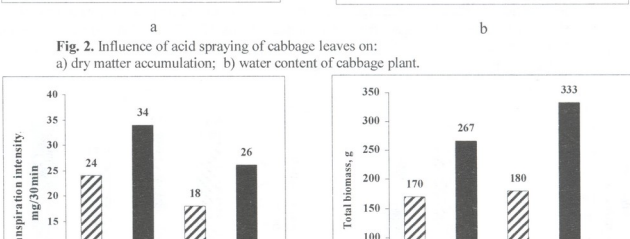
Fig. 1. Influence of acid treatment of cabbage seeds on:
a) photosynthetic activity of leaves; b) leaf stomatal conductivity.

Measurements of dry matter accumulation and water content have revealed that the red form of cabbage was distinguished with higher dry matter and less content of water compared with white form (Fig. 2, a, b). Spraying leaves with acid solution increased dry matter accumulation in red cabbage, while in white form the opposite results were mentioned (Fig 2, a, b). Transpiration index was higher in white cabbage. Spraying with acid solution diminished the index in both forms of cabbage but the effect was more apparent in white cabbage (Fig. 3, a). At the same time the total weight of plants increased (Fig. 3, b).



a b

Fig. 2. Influence of acid spraying of cabbage leaves on:
a) dry matter accumulation; b) water content of cabbage plant.

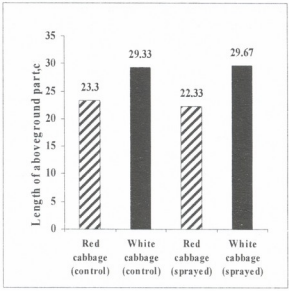


a b

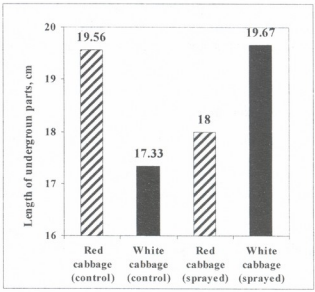
Fig. 3. Influence of acid spraying of cabbage leaves on:
a) transpiration intensity of leaves; b) plant total biomass.

Examination of above and under ground parts of tested plants has shown that as a result of leaf spraying in white cabbage the length of under ground parts increased, while no effect was mentioned on above ground parts (Fig. 4, a, b). In red cabbage spraying with acid caused diminishing of length of both above and under ground parts.

Testing of the total activity of growth regulators manifested essential reduction of the index of growth stimulators in leaves of plants emerged from acid-treated seed (Fig. 5 b; Fig. 6 b). Especially clear effect was mentioned in white cabbage. Opposite effect was revealed in case of plant spraying with acid solution: here significant activation of growth stimulators was detected, which was reflected on changes in biomass accumulation (Fig. 5 c, Fig. 6 c, Fig. 3 b).



a



b

Fig. 4. Influence of acid spraying of cabbage leaves on length of:
a) above- and b) underground parts of a plant.

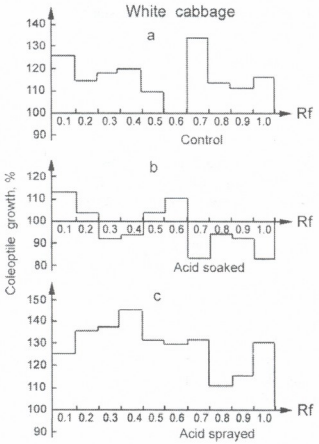


Fig. 5. Total activity of growth regulators in white cabbage leaves

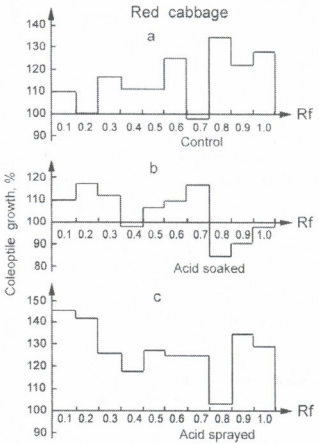


Fig. 6. Total activity of growth regulators in red cabbage leaves

According to the analysis of the obtained data white form of cabbage seems to be more sensitive to simulated acid rains, while red one is more resistant. This fact may be explained by high content of protective substances - anthocyanins in leaves of red cabbage.

References:

Adams C. M., Hutchinson T. C. *New Phytologist,* **106,** 3, 437-456, 1987.

Evans L. S., Sarrantonio M. J., Owen E. M. *New Phytologist,* **103**, 689-693, 1986.

Filippovich J. B., Egorova T. A., Sevast'yanova G. A. *Practical work in general biochemistry.*, M., "Prosveshchenye", 317p., 1975 (in Russian).

Hippeli S., Elster E. F. J. *Plant Physiology*, **148**, 249-257, 1996.

Jay J. S., Osmeloski J., Vamada K., Hellor L. *New Phytologist,* **105**, 1, 139-147, 1987.

Kefeli V.N., Turetskaya R.Kh. *Methods of determination of growth regulators and inhibitors.*, M., 1966 (in Russian).

Ten Years of Monitoring Forest Condition in Europe. Studies of Temporal Development, Spatial Distribution and Impact of Natural and Anthropogenic Stress Factors. Eds. **Muller C.** et al., Brussels: UN/ECE and EC. p.205-275, 1999.

Olson R. L., Winner W. E., Moore L. D. *Environment and Exp. Botany*, **27**, 2, 239-244, 1987.

Voznesensky V.L., Zalensky O.V. and Semikhatova O.A. *Methods of investigation of photosynthesis and respiration of plants.* Moscow, "Nauka", 1965(in Russian).

ხელოვნური მჟავე ნალექებით თესლისა და ფოთლების დამუშავების ბავლენა კომბოსტოს (*Brassica capitata* L.) წითელი და თეთრი ნაირსახეობების ფიზიოლოგიურ მაჩვენებლებზე

რაფავა ლ., ჭანიშვილი შ., ბადრიძე გ., ბარბალიშვილი თ., აბრამიძე ს.

(მიღებულია 10.10.2006)

რეზიუმე

შესწავლილია გოგირდმჟავას pH2.5 წყალხსნარით კომბოსტოს (Brassica capitata L.) წითელი და თეთრი ნაირსახეობების თესლისა და ფოთლების დამუშავების ეფექტი ფოთლების ფოტოსინთეზურ აქტივობაზე, ზრდის რეგულატორების აქტივობაზე, მშრალი ნივთიერების აკუმულაციასა და წყლის შემცველობაზე. კომბოსტოს თეთრი ნაირსახეობის როგორც თესლი, ისე ფოთლები უფრო მგრძნობიარე აღმოჩნდა მჟავე ნალექებით დამუშავებისადმი. კომბოსტოს წითელი ნაირსახეობის შედარებით მაღალი მდგრადობა მჟავათი დამუშავებისადმი საკარაუდოდ ამ ნაირსახეობის ფოთლებში დამცველობითი ფუნქციის მქონე ნივთიერების - ანთოციანების მაღალი შემცველობით უნდა იყოს განპირობებული.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Zoology

EFFICIENCY OF NEMATODE *STEINERNEMA CARPOCAPSAE* SAY AGAINST FALL-PLANTING CUTWORM (*AGRIOTIS SEGETUM* SCHIFF)

KAKULIA I.[1], KOKHIA M.[1], SHEKILADZE K.[2]

[1]*Institute of Zoology*
[2]*Agricultural State University*

(Received May 20, 2006)

Abstract

Bioecology of fall-planting cutworm - *Agriotis segetum* Schiff and the effect of the nematode *Steinernema carpocapsae* Say on the fall-planting cutworm was studied in Sachkhere-Chiatura district, West Georgia, for further usage of obtained results in pest biocontrol. In the field environment pest death rate consists of 68 % in the suspension of the concentration - 150 nematodes/ml, 66.5% - in case of 200 nem/ml, and 67.2% - in case of 250 nem/ml. Effect of entomopathogenic nematodes appeared the highest on the I plot, which was processed with suspension of 150 nem/ml concentration (68%) indicating that efficiency of the nematode *S. carpocapsae* against pests is high even at low concentration of suspension.

Key words: bioecological control, entomopathogenic nematode, nematode suspension.

Introduction

Damage to agriculture caused by pest is great. They obliterate significant part of the yield (nearly 25%) and decrease quality of product. Fall-planting cutworm *Agriotis segetum* Schiff is considered as the most dangerous pest of crops, and particularly of maize. As the maize is one of the main crops in Georgia to study the effect of entomopothogenic nematodes on fall-planting cutworm is the urgent problem.

Fall-planting cutworm is widespread pest in Georgia [Kanchaveli, Supatashvili, 1968]. Its larvae cause great damage to maize, horticultural crops; they injure not only seed germs, but root system too. Larvae cut aslant the root collar of saplings causing their death. Pest larvae of different ages hibernate in the soil. At the beginning of frosts young larvae die, but mature ones make soil bed in early spring and pupate there. Two weeks later nymph flies out of pupae, which generally occur in soil at daytime. Nymph blows 2500 eggs both on weed and cultural plants. Larvae are characterized with negative phototaxis and hence they are hidden in soil. Larvae stage lasts 28-38 days. In Georgia this pest gives 3 generations, and so, their number and respectively damage is great. Damage level caused by fall-planting cutworm belongs to high harmfulness categories.

Materials and Methods

Studies were carried out in the villages Kvatsikhe and Biga of Chiatura region. Plots of maize sowings were located on 10 m from river bank. For experiment 4 plots (3 - experimental and

1 - control) were chosen, each of them of $5m^2$ area. Experiments were carried out in autumn, 2005 (September) and spring, 2006 (May).

Experimental plots were treated with nematode (*Steinernema carpocapsae*) suspension of various concentrations. Suspensions were kept in thermos. Plots were treated early in the morning, at quiet whether conditions (air temperature – 16-20°C). Prepared nematode suspension was sprayed into plants by automax. 3, 8 and 10 days after live and dead larvae were counted both on experimental and control plots according to Franz method [Franz, 1968]. The obtained data are presented in the Table.

Results and Discussion

It was found that pest death rate in I plot was 68%, in II plot – 66.5%, and in III plot – 67.2% (see the table).

Table. Efficiency of nematoda *S. carpocapsae* against fall-planting cutworm (*Agriotis segetum*)

#	Nematode species	Concentration of nematode suspension nem/1ml	Number of larvae	Death rate of larvae			Total number of dead larvae	%
				3 days	8 days	10 days		
1.	*S.carpocapsae*	150 (I plot)	50	10	16	8	34	68
2.	"----------"	200 (II plot)	122	34	32	18	74	66,5
3.	"----------"	250.(III plot)	110	30	32	12	74	67,2
4.	control	clean water	100	-	-	1	1	1

As is seen from the table effect of entomopathogenic nematodes is a bit higher on I plot, which was processed with nematode suspension of 150nem/ml concentration (68%). Results of experiments carried out earlier in laboratory environment showed that while using nematode suspension of 200nem/ml concentration the death rate of larvae was 97%. We consider that the obtained results are caused by the closeness of I plot with the river, and its location in shadow.

Thus, efficiency of nematode *S. carpocapsae* against pests is rather high in spite of low concentration of suspension, and efficiency often depends not on the concentration of suspension but on the experimental conditions.

Obtained data confirm the literature data [Lortkipanidze et al., 2004; Hominick, Reid, 1990] according to which *S. capocapsae* is high-efficient agent for the control of fall-planting cutworm.

References:

Franz J.M. *Zur Berechung des Wirkunsrates einer Microbiologchen Bekamfung von Schädlinskten.* Planzensch., **80**, p. 65-71, 1968.

Hominick W.M., Reid A.P. *Perspectives on entomopathogenic nematology.* In: Entomopathogenic Nematodes in Biological Control (Gaugler R. and Kaya H.K. eds) CRC. Boca Raton FL. p.327-345, 1990.

Kanchaveli G., Supatashvili M. *Forest entomology.* Tbilisi. p. 1-375, 1968 (in Georgian).

Lortkipanidze M., Devdariani Ts., Maglakelidze L., Kokhia M. *Effect of nematodobacterial complex on some pests.* Proceeding of The Institute of Zoology, **XXII**, 45-48, 2004 (in Georgian).

ნემატოდა *STEINERNEMA CARPOCAPSAE* Say -ს ეფექტურობა შემოდგომის ნათესების ხვატარის – *AGRIOTES SEGETUM* Schiff მიმართ

კაკულია ი.[1], კოხია მ.[1], შეყილაძე ქ.[2]

[1]*ზოოლოგიის ინსტიტუტი*
[2]*საქართველოს სასოფლო-სამეურნეო უნივერსიტეტი*

(მიღებულია 20.05.2006)

რეზიუმე

შესწავლილია შემოდგომის ნათესების ხვატარის – *Agriotis segetum* Schiff ბიოეკოლოგია და ნემატოდა *Steinernema capocapsae* Say-ს მოქმედების ეფექტი შემოდგომის ნათესების ხვატარის მიმართ სახნერ-ჰიათურის რეგიონში, შემდგომ მავნე მწერების ბიოკონტროლში გამოსაყენებლად. საველე პირობებში მწერების სიკვდილიანობის პროცენტული მაჩვენებელი 150 ნემატოდა/მლ კონცენტრაციის ნემატოდურ სუსპენზიაში შეადგენდა 68%, 200 ნემ/მლ შემთხვევაში – 66.5%, ხოლო 250 ნემ/მლ-ში კი 67.2%-ია. ენტომოპათოგენური ნემატოდების მოქმედების ეფექტი ყველაზე მაღალი იყო I ნაკვეთზე, რომელიც დამუშავდა 150 ნემ/მლ კონცენტრაციის მქონე სუსპენზიით (68%), რაც იმის მაჩვენებელია, რომ ნემატოდა *S. carpocapsae*-ს ეფექტურობა მავნე მწერების წინააღმდეგ მაღალია მიუხედავად სუსპენზიის დაბალი კონცენტრაციისა.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Zoology

THE ORIBATID MITES (ACARI, ORIBATIDA) OF FLOOD PLAIN ALDER FORESTS OF CENTRAL COLCHIC LOWLAND

*MURVANIDZE M., KVAVADZE E.

Institute of Zoology
**E-mail: makam94@hotmail.com maka.murvanidze@gmail.com*

(Received October 16, 2006)

Abstract

The researches are provided in flooded ecosystems of Central Colchic Lowland on the territories of Colchic National Park, Kobuleti Reserve. These territories are absolutely unique and have great international importance. Oribatid mite fauna of flood plain alder forests in Central Colchic Lowland is studied. 79 species are registered. The ecological analysis shows that in biotops with frequent inundation similar oribatid communities are formed. The diversity of species and high density indicates on a leading role of oribatid mites in processes of decomposition in studied ecosystems.

Key words: Colchic Lowland, *Oribatida*, Ramsar Site, bioindication.

Introduction

The flood plain forests are widely distributed on Colchic Lowland. The main composer of these forests is alder (*Alnus barbata*). Here are also *Pterocarya pterocarpa, Populus canescens, Salix micans* and *S. alba* found. In understorey grow *Rhododendron luteum, Viburnum opulus, Clemantis vitalba, Crataegus pentagina* etc; from liana's group are found - *Smilax excelsa, Hedera colchica, Pericploca graeca* etc [Ketskhoveli, 1959]. The flood plain alder forests are important to maintain the biodiversity of not only Colchic Lowland, but of the whole Black Sea biogeographical region.

In XX century the major part of flood plain forests of Colchic Lowland were drought up and on meliorated soils citrus plantations were mainly cultivated. Currently the floodplain forests and bogs of Colchic Lowland have great international importance, as they make habitats for migrating, hibernating and nesting birds. The flooded habitats have great importance in maintaining the biodiversity of species on the concrete territories [Weigmann, 1997 b]. Currently part of the flooded ecosystems of Colchis are on the territories of Colchic National Park, Kobuleti Reserve and Ramsar Site and are protected by law.

It is known that oribatid mites are one of the main decomposers of organic matters [Ghilarov, Krivolutsky, 1975; Haq, 1987]. They are known as one of the best bioindicators of environment conditions as well [Klausnitzer, 1990, Weigmann, 1991, 1997 b]. Their diversity and density on the studied territory may indicate on condition of these ecosystems.

Information on oribatid mites of Colchic Lowland alder forests is very poor. Lagidze (1981) has registered 29 species of oribatids in bogs with alder forests. Three of them (*Nothrus*

palustris, Minunthozetes pseudofusiger and *Phthiracarus* sp.) were typical for swamped soils and were frequently found. 95 % of registered mites were found in 0-5 cm depth.

Within the animal researches regarding typical biocenoses of Colchic Lowland [Kurashvili, (edt.) 1984], 12 species of oribatid mites were registered on territory of Kulevi in the alder forest. Most of them were found in upper, 0-10 cm layer of soil.

Species that are registered in literature mainly coincide with our data, but this coincidence belongs to such wide distributed species as *Tectocepheus velatus, Platynothrus peltifer, Quadroppia quadricarinata, Scheloribates latipes* and *Minunthozetes pseudofusiger*.

Materials and Methods

Material was taken in June and July 2005. At each site three soil samples (10 cm^3, 0-10 cm depth from surface) were taken and animals were extracted by Tullgren–apparatus. On the studied territory 8 plots were investigated:

1. Anaklia. The left bank of riv. Tikori. Bog with alder forest. N = 42°2,793' E = 41° 37,017'; H = 5m;
2. Anaklia. The left bank of riv. "Didi Gali". Alder forest with *Buxus* understorey.
3. Parpala. The right bank of riv. Churia. Wet alder forest;
4. Imnati. Polydominant wooded bog. N = 42° 08, 283' E = 41° 57,299'; H= 5-7m;
5. Imnati. Kalamona forest. Bog with ash - alder forest. N = 42° 08,190'; E = 41° 96, 980';
6. Kulevi. Alder forest;
7. 6 km from Kulevi. Wet alder forest;
8. Partotskali Lake coast. Bog with alder forest.

As coefficient of faunal likeness between different plots, indicating species identity, Jaccard's coefficient was calculated [Chernov, 1975]. The calculation of community likeness was based on Renkonen's coefficient [Krebs, 1989]. The dominance identities and faunal likeness have been clustered to a dendrogram.

In this investigation only the adult mites were identified and counted.

For the identification of the oribatid mites mainly Ghilarov and Krivolutsky (1975), Balogh and Mahunka (1983), Niedbala (1983) Weigmann (2006) articles were used. For determination of the biogeographycal belonging of oribatid mites work of Subias (2004) was used.

Results and Discussion

79 species of oribatid mites united in 42 families and 50 genera were registered on studied territory (Tab. 1).

Great number of mesophyllic species is presented in the fauna and mainly found in humid ecosystems of Colchic Lowland. Such species are *Mesoplophora pulchra, Microtritia minima, Dissorhina ornata, Eupelops hygrophilus, Achipteria longisetosa, Pergalumna minor* and *Punctoribates mansanoensis*.

No species was registered in every plot. *Steganacarus personatus* was found everywhere except Kulevi (plot 6), and *Protoribates capucinus* was found everywhere except the bank of riv. Churia (plot 7). 34 species were found only in one plot (Tab. 1).

In fauna of oribatid mites predominate widely distributed mites: Palaearctic – 26 species (32 %), Holarctic – 20 species (25 %), Cosmopolits – 16 species (20 %) and European – 8 species (10 %). With less quantity are presented Mediterranean (5 species – 6 %), Caucasian, Endemic (2-2 species – 2-2 %) and Euro-Atlantic (1 species – 1 %) species (Tab. 1).

Table 1. The list of oribatid mites of floodplain alder forests in Central Colchic Lowland and their biogeographycal distribution (+ dominance %; dom. +: % < 1)

#	species	1	2	3	4	5	6	7	8	Distr.
1	*Hypochthonius rufulus* C. L. Koch, 1836	+	+						3	Cosm
2	*Mesoplophora pectinata* Mahunka, 1979				4					Pal
3	*M. pulchra* Sellnick, 1928	+							1	Pal
4	*Phthiracarus assimilis* Niedbala, 1983	+	+							Cauc
5	*Ph. crassus* Niedbala, 1983				+					Md
6	*Ph. ferrugineus* (C. L. Koch, 1841)	7	1	6			3		8	Pal
7	*Ph. globosus* (C. L. Koch, 1841)		11						+	Ho
8	*Ph. incertus* Niedbala, 1983								1	Hol
9	*Ph. lanatus* (Feider & Suciu, 1957)	+			+			9		Eu
10	*Ph. lentulus* (C. L. Koch, 1841)		3		4	16		36		Hol
11	*Ph. ligneus* Willmann, 1931		1				8		14	Hol
12	*Ph. nitens* (Nicolet, 1855)	1	1	3						Pal
13	*Hoplophthiracarus vanderhammeni* Niedbala, 1991	+	1				2	3		Cosm
14	*Steganacarus carinatus* (C. L. Koch, 1841)	+	+			17	2	3		Pal
15	*St. conjuctus* Niedbala, 1983								+	Md
16	St. *personatus* Niedbala, 1983	5	5	70	46	35		3	2	Eu
17	*St. striculus* (C. L. Koch, 1836)		+				4		2	Hol
18	*Microtritia minima* (Berlese, 1904)				+					Cosm
19	*Rhyzotritia ardua* (C. L. Koch, 1841)						3		1	Cosm
20	*Camisia horrida* (Hermann, 1804)								+	Hol
21	*Platinothrus peltifer* (C. L. Koch, 1839)	+	4		+		13	3		Cosm
22	*Nanhermannia nana* (Nicolet, 1855)	+	1			2	8	39	8	Cosm
23	*Belba sculpta* Mihelcic, 1957	+					1			Md
24	*Metabelba pulverulenta* (C. L. Koch, 1840)	+			+					Hol
25	*Arthrodamaeus femoratus* (C. L. Koch, 1840)								+	Pal
26	*Hypocepheus mirabilis* Krivolutski, 1971	1								Eu
27	*Amerobelba decedens*Berlese, 1908	1	2						+	Md
28	*Eremobelba geographica* Berlese, 1908	+	4						2	Eu
29	*Damaeolus ornatissimus* Csiszar, 1962		2							Pal
30	*Gustavia microcephala* (Nicolet, 1855)	2	+	3						Pal
31	*Liacarus brevilamellatus* Mihelcic, 1955	+								Md
32	*L. coracinus* (C. L. Koch, 1841)						1			Pal
33	*Xenillus tegeocranus* (Hermann, 1804)	+	+		+					Pal
34	*Ceratoppia quadridentata* (Haller, 1882)				+	5	1		1	Hol
35	*Carabodes femoralis* (Nicolet, 1855)				1					Pal
36	*C. rugosior* Berlese, 1916	+			3					Hol
37	*Tectocepheus velatus* (Michael, 1880)		+							Cosm
38	*Dissorhina ornata* (Oudemans, 1900)	4	1						1	Hol
39	*Oppia nitens* C. L Koch, 1836	28							7	Hol
40	*Oppiella (Rhinoppia) fallax* (Paoli, 1908)	2	6		+	2			2	Cosm
41	*O. obsoleta* (Paoli, 1908)	+	1							Pal
42	*O.neerlandica* (Oudemans, 1900)	+			+					Hol
43	*O. nova* (Oudemans, 1902)	+			+		16		6	Cosm
44	*O. tuberculata* (Bulanova-Zachvatkina, 1964)		15							Eu
45	*O. unicarnata* (Paoli, 1908)		+						+	Hol
46	*Ramusella insculpta* (Paoli, 1908)		2							Pal
47	*R. mihelcici* (Perez-Inigo, 1965)				1					Pal
48	*Quadroppia michaeli* Mahunka, 1977		+							Pal

49	*Q. quadricarinata* (Michael, 1885)	+								Hol
50	*Suctobelba trigona* (Michael, 1888)	+								Pal
51	*Suctobelbella duplex* (Strenzke, 1950)	+								Pal
52	*S. subcornigera* (Forsslund, 1941)		+							Cosm
53	*Autogneta tragardhi* (Forsslund, 1941)	+							1	Hol
54	*Eupelops acromios* (Hermann, 1804)				+					Pal
55	*E. hygrophilus* (Knuelle, 1954)									Eu
56	*Achipteria longisetosa* Murvanidze & Weigmann, 2003								1	End
57	*Parachipteria georgica* Murvanidze & Weigmann, 2003	13	+						42	End
58	*P. punctata* (Nicolet, 1855)				1					Hol
59	*P. nicoleti* (Berlese, 1883)						14			Hol
60	*Oribatella nigra* Kulijev, 1967		+							Cauc
61	*Galumna obvia* Berlese, 1914						2			Cosm
62	*Pergalumna minor* (Willmann, 1928)		28							Hol
63	*Ceratozetes gracilis* (Michael, 1884)		17	3						Cosm
64	*Edwardzetes edwardsi* (Nicolet, 1855)		+							Eu-Atl
65	*Latilamellobates incisellus* (Kramer, 1897					5				Pal
66	*Sphaerozetes piriformis* (Nicolet, 1885)					7				Pal
67	*S. tricuspidatus* Willmann, 1923						2			Pal
68	*Chamobates kieviensis* Shaldybina, 1980		1						1	Eu
69	*Ch. voigtsi* (Oudemans, 1902)	53	1		2					Pal
70	*Minunthozetes pseudofusiger* (Schweizer, 1956)	+	1		3	2			1	Pal
71	*Punctoribates mansanoensis* (Hammer, 1958)						1			Eu
72	*P. punctum* (C. L. Koch, 1908)		+		17	2				Cosm
73	*Protoribates capucinus* Berlese, 1908	+	2		1	5	20	3	2	Cosm
74	*Scheloribates laevigatus* (C. L. Koch, 1835)				+					Cosm
75	*Sch. latipes* (C. L. Koch, 1840)	+								Pal
76	*Dometorina plantivaga* (Berlese, 1895)				+					Cosm
77	*Oribatula tibialis* (Nicolet, 1855)	1	+		5				2	Hol
78	*Zygoribatula exilis* (Nicolet, 1855)				3	2				Hol
79	*Z. microporosa* (Bulanova-Zachvatkina, 1967)								+	Pal
	total	37	37	6	26	12	17	8	29	
	density 1000 ind/m^2	71	32	17	51	13	50	8	30	

The maximum density of oribatid mites was registered in Anaklia, on the bank of riv. Tikori (71 730 ind/m^2). The density was also high in polydominant wooded bog in Imnati (51 750 ind/m^2) and in alder forest in Kulevi (50 500 ind/m^2). The lower indexes were marked in wet alder forest 6 km from Kulevi (8 250 ind/m^2) (Tab. 1).

High dominance showed *Steganacarus personatus* (70 %, 46 % and 35 % in different sites), *Phthiracarus lentulus* (36%), *Nanhermannia nana* (39%), *Oppia nitens* (28 %), *Parachipteria georgica* (42%), *Pergalumna minor* (28%), *Chamobates voigtsi* (53%) and *Protoribates capucinus* (20%). Dominance value of other species is much less (Tab. 1).

Based on the calculations of faunal likeness and dominance analyses the ecological clusters were build. Three groups are distinguished in cluster of faunal likeness. In the first group there are united mites from Anaklia and Partotskali Lake (plots 1, 2, 8), what is caused by extreme wetness of these plots. Plots 1 and 2 are located on the banks of the river and inundate several times in the year. In the second group oribatid mites of Imnati and 6 km from Kulevi are united (plots 4, 5, 7). Those sites are distinguished by less humidity. The third plot (bank of riv. Churia) is located

a bit higher compared with other plots and inundates rarely. In this plot less number of oribatid mites was registered, what is also a reason for its low likeness with mites of other plots (Fig. 1).

Three groups were divided in cluster of dominance identities as well. The first group includes the dominant species of oribatid mites of riv. Churia and Imnati (plots 3, 4, 5). These plots are territorially close and ecologically similar. In the 3rd plot number of species was low, but their density was high (16 500 ind/m^2) and dominance identities were also high. The second group unites dominant species of Anaklia and Partotskali Lake (plots 1, 2, 8). As it was already mentioned, these plots are ecologically similar. Plots 6 and 7 are grouped together because they are close both, territorially and ecologically (Fig. 2).

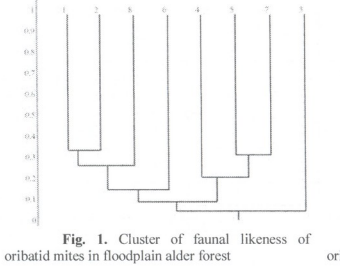


Fig. 1. Cluster of faunal likeness of oribatid mites in floodplain alder forest

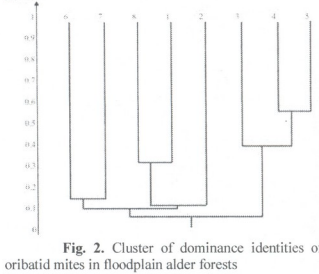


Fig. 2. Cluster of dominance identities of oribatid mites in floodplain alder forests

High diversity and density of oribatid mites are rather unexpected. It is known that mites prefer humid environment, but can not resist high humidity for long period and die because of invasion of microorganisms [Smrz, 1996]. The received results can be explained with low concurrence among the groups which is provoked by extreme conditions and only oribatid mites provide humification and decomposition processes. Low concurrence increases diversity of species of concrete group [Heaney, 2001].

Researches provided in flooded biotops of riv. Oder valley (Germany) showed that oribatid mites adapted to inundation during the winter period and hibernated in early stages; when inundation happened in summer, most of imagoes died [Weigmann, 2004]. In our case the studied territory inundates in spring, summer and autumn. The level of the water decreases in winter, but humidity remains high. We suppose that the main species of oribatid mites of Colchic Lowland alder forests are adapted to the several inundations and fluctuation of their quantity is less discernable during the year.

The species diversity and high density of oribatid mites on the studied territory indicates the unique flora of these ecosystems, active processes of decomposition and soil formation and proves necessity of its international protection.

References:

Balogh J., Mahunka. S. *Primitive Oribatis of Palaearctic Region.* Akademiai Kiado, Budapest,1983.

Chernov Yu. I. *The General Synecological Characteristic of Soil Invertebrates and Methods of Their Analysis.* In: Methods of soil-zoological investigations. Moscow, 160-216, 2002 (in Russian).

Ghilarov M. S., Krivolutsky D. A. *The Identification Keys of Soil Inhabiting Mites. Sarcoptiformes.* "Nauka", Moscow, 1975 (in Russian).

Heaney L. R. *Small Mammal Diversity Along Elevation Gradients in the Phillipines: an Assasment of Patterns and Hypotheses.* Global Ecology and Biogeography. **10,** 15-39, 2001.

Ketskhoveli N. *Vegetation Cover of Georgia.* Metsnierebata akademiis gamomcemloba, Tbilisi, 1959 (in Georgian).

Klausnitzer B. *Ecology of the City Fauna.* "Mir", Moscow, 1990 (in Russian).

Krebs Ch. J. *Ecological Methodology.* Harper & Row Publishers, New York, 1989.

Kurashvili B. (edt). In: *Animal Composition of Typical Biocenoses of Colchic Lowland.* "Mecniereba", pp. 38-43, 1985 (in Georgian).

Lagidze O. I. *Fauna of Podsol-Clay Soils of Colchic Lowland and Their Changes Within Drainage and Cultivating.* PhD abstract, 1981 (in Russian).

Niedbala W. *Les Noueaux Phthiracaridae (Acari, Oribatida) du Caucase.* Ann. Zool. **37,** 62, 1983.

Smrz J. *Some Aspects of the Strategy of Oribatid Mites* (*Oribatida*). Acarology. **IX**. Proceedings, 553-555, 1993.

Subias L. S. *Listado Sistematico, Sinonimico y Biogeografico de los Acaros Oribatidos (Acariformes, Oribatida) del Mundo.* (1758-2002). Graellsia, 60, 2004.

Weigmann G. *Oribatid Communities in Transtcts from Bogs to Forests in Berlin Indicating the Biotope Qualities.* In: F. Dusbabek and V. Bukva (Eds): Modern Acarology, vol. **1**. Academia Prague and SPB Academic Publishing bv. The Hague, 359-364, 1991.

Weigmann G. *Bioindication by means of Isovalent species Groups.* Abh. Ber. Naturkundemus. Goerlitz. **69,** 2, 59-65, 1997a.

Weigmann G. *Die Hornmilben-Fauna (Acari, Oribatida) in Auenböden des Unteren Odertals.* Faun. - Ökol. Mitt. **7,** 319-333, 1997b.

Weigmann G. *Recovery of the Oribatid Mite Community in a Flood Plain After Decline due to Long Time Inundation.* Phytophaga. **XIV,** 201-207, 2004.

Weigmann G. *Hornmilben (Oribatida). Die Tierwelt Deutschlands.* 76 Teil. Goecke, Evers, Kelter Publ. 2006.

კოლხეთის ცენტრალური დაბლობის ჭარბტენიანი მურქნარების ჯავშნიანი ტკიპები (ACARI, ORIBATIDA)

მურვანიძე მ., ყავაძე ე.

ზოოლოგიის ინსტიტუტი

(მიღებულია 16.10.2006)

რეზიუმე

გამოკვლევები ჩატარებულია კოლხეთის ცენტრალური დაბლობის ჭარბტენიან ეკოსისტემებში კოლხეთის ეროვნული პარკის, ქობულეთის ნაკრძალისა და რამსარის საიტის ტერიტორიებზე. ეს ტერიტორიები სრულიად უნიკალურია და აქვთ დიდი საერთაშორისო მნიშვნელობა. შესწავლილია ჯავშნიანი ტკიპების ფაუნა კოლხეთის ცენტრალური დაბლობის ჭარბტენიან მურქნარებში. რეგისტრირებულია 80 სახეობა. ეკოლოგიური ანალიზი გვიჩვენებს, რომ ბიოტოპებში, რომლებიც ხშირად იფარება წყლით, მსგავსი ორიბატიდული ფაუნა ყალიბდება. სახეობათა მრავალფეროვნება და მაღალი სიმჭიდროვე მიუთითებს გამოკვლეულ ეკოსისტემებში დეკომპოზიციის პროცესებში ჯავშნიანი ტკიპების წამყვან როლზე.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Letter to editor

NEW DATA ON FUNGAL DISEASES OF BULB ONION (*ALLIUM CEPA* L.)

*METREVELI I., KUPRASHVILI T.

L. Kanchaveli Institute of Plant Protection
**E-mail: metreveli_i@hotmail.com*

(Received November 6, 2006)

Abstract

During 2004-2006 the samples of bulb onion (*Allium cepa* L.), harvested in different regions of Georgia or imported from abroad have been examined on the presence of fungi. The list of the revealed fungi supplemented with short diagnoses, the information on sites and time of collection are presented. 5 species are registered for the first time on bulb onion in Georgia. One of them - *Epicoccum* sp. is not identified up to species. The descriptions of 4 new species of fungi are given. Among them 2 species was revealed on the local bulb onion cultivars, and 2 - on imported cultivars.

Bulb onion (*Allium cepa* L.), one of the significant vegetable crops of Liliaceae family, is widely cultivated in Georgia. Onion bulbs are used as food and in medicinal purposes. Onion skin is widely exploited as natural dye-staff. Obtaining big yield of high quality bulb onion is of great importance for the country agriculture. The yield and quality of bulb onion is significantly affected by fungal diseases, both in the open ground and during the storage.

The following most common and harmful fungal diseases have been registered on bulb onion – onion mildew (*Peronospora schleideni* Ung.), smut (*Urocystis cepulae* Frost.), onion rust (*P.mixta Fuckel porii* Wint.), grey rot (*Botrytis alli* Munn.) [Khazaradze, 1952; Shoshiashvili, Kirimelashvili, 1952; Zhvania, 1985; Nebulishvili, 1988]. The mentioned pathogens heavily reduce the yield of bulb onion and worsen its quality.

Active trade relations with foreign countries, uncontrolled situation at customs offices have had effect on agrocoenoses of cultivated crops. Reaction of microorganisms and saprophytic fungi, associated with the material introduced from abroad, undergoes changes in the process of competition in a new environment and the organisms often become pathogenic. This may cause wide expansion of the diseases in a new environment. This very phenomenon prompted us to investigate species composition of fungi associated with both local and imported onion bulbs.

The list of new species of fungi registered by us on the material obtained as a result of observations carried out during 2004-2006 in Georgia, is supplemented with short diagnoses and designations of collecting site and time. Information on collecting sites is appended to the species registered in Georgia and corresponding determination keys are cited. Species of fungi are arranged according to E. Muller and V. Lefler [Muller, Leffler, 1995].

The fungi which are registered for the first time in Georgia are indicated by *.

1. *Peronospora destructor* Berk. Casp [7:147] onion mildew.

Tbilisi, Central Market, green onion leaves brought from village Dzalisi Mtskheta District, 11.05.2004; Agrarian market, Marneuli, onion bulbs, 24.07.2005.

2. *Mortierella jenkini* (Smith) Naumov [9:14]

Tbilisi, Didi Dighomi, private commercial greengrocery. Onion bulbs, introduced from Akhalkalaki district, 17.07.2005.

3. *Choanephora conjuncta Couch. (9:88)

Hyphae of the colony are of filiform, of yellowish-grey color. Conidiophores 0.8 cm high and 10-35μm in diameter. Conidia oblong, ovate, pyriform, globose or elliptic, 8-20(24) x 6-10(12) μm. Brown sporangiospores are elliptic or spindle-shaped, 14-26 x 8-15 μm. Stretched mycelial mat of yellowish light grey color develops on onion skin.

Tbilisi, Saburtalo District, Vazha-Pshavela ave., private commercial greengrocery, 04.05.2004.

4. *Urocystis cepulae* Frost (6:517; 7:211; 10:319)

Gori District, village Kheltubani, 11.05.2004; Kareli district, village Khviti, 13.05.2004.

Tbilisi, Didi Dighomi, private vegetabl stall, 15.09.2006.

5. *Puccinia porii* G. Winter [11:161]

Agrarian market, gori, 13.09.2005; Telavi District, village Gulgula, 21.08.2004.

6. *Aspergillus niger v.* Tiegh. [12:547]

Kaspi District, village Kavtiskhevi, private plot, 16.09. 2005.

7. **Glioklad ium vermoeseni (*Biourge)Thom [13:39]

Aerial mycelium of white color later on becomes granular and turns white-pink. Thick twisted mycelial hyphae of 3-6 μm diameter with numerous vacuoles. Conidiophores 100-200 x 4-5 μm, sterigmas usually 8-12 μm, colorless ones usually 4-6 x 3-4 μm. Conidia elliptic in 1-2 mm long chains.

Tbilisi, Agrarian market, private commercial greengrocery, 18.07.2006.

8. *Verticillium lateritium* Berk. [13:79]

Lagodekhi District, village Ninigori., private greengrocery, 13.06.2005.

9. *Botrytis alli* Munn.. [12:179; 13:67; 10:485]

Telavi District, village Vardisubani, 16.08.2004; Tbilisi, Saburtalo District, private greengrocery, 23.10.2005.

10. *Cladosporium herbarum* (Pers.) Link. [12:313]

Kvareli District, village Shilda, Private plot 17.08.2005; Tbilisi, Didube District, Agrarian market, 23.10.2005.

11. *Periconia atra Corda* [12:349]

Samtredia District, Village Ivandidi, 11.07.2005; Tbilisi, agrarian market 01.05.2006.

12. *Alternaria porri* (Ellis) Cif. Ellis [13:177; 10:512]

Kutaisi, agrarian market, 17.03.2004.

13. *Macrosporium parasiticum* Thum. [11:161]

Samtredia, agrarian market, 17.08.2004.

14. *Stemphyllium allii* Oudem. [13:184; 10:537]

Samtredia, agrarian market, 16.08.2006.

15. *Cercospora duddiae* Welles [12:278; 13:112; 11:161; 10:517]

Tbilisi, Didi Dighomi, private plot 17.08.2005.

16. **Heterosporium alli-cepae* Ran. [13:144]

Conidiphores yellowish-brown, 200x7.5-20 μm wide. Conidia yellowish-grey, unicellular, conidia echinulate; pyriform with 1-2 septa. 31-78 x 8.5-18 μm ([13] 32-76 x 9.5-20 μm).

Lagodekhi District, village Ninigori, private plot, 09.2006; Rustavi, private greengrocery, 25.11.2005.

17. *Fusarium oxysporum* Schlecht [13:261]

Lagodekhi District, private plot, 22.12.2005.

18. **Fusarium avenaceum var. anguioides* (Sherb) Bilai [8:182].

Infected bulbs are darkened, in tissue constructing cells and intercellular spaces mycelial hyphae are developed. Mycelial scab of white color occurs between the bulb scales. Macroconida 20-38 x 3.9-5.3 µm. The fungus occurs in the storage conditions.

Gori, agrarian market, 18.06.2004.

19. **Epicoccum sp.*

Lagodekhi District, village Vardisubani, private plot, 13.06.2005.

20. *Colletotrichum circinans* (Berk.) Voglino. (13:196)

Tbilisi, Saburtalo District, agrarian market "Soplis nobati", 3.03.2004.

References:

Bilai V. I. *Fuzarii*, Kiev, 442 p., 1985 (in Russian).

Ellis M. B. *Dematiaceus Hyphomycetes.* Commonwealth Mycological Institute, Kew, Surrey, England, p.608, 1971.

Flora of spore plants of Georgia (Synopsis). Tbilisi, 1986 (in Russian).

Khazaradze E. Proceedings of Inst.Plant Prot. of Georg.SSR, **VIII**, 97-109, 1952 (Georgian).

Khokhryakov M. K., PotlaichukV. I., SemenovA. Ya, ElbakyanM. A. *Guide-book of diseases of agricultures.* Leningrad, p.303, 1984 (in Russian).

Muller E., Leffler V. *Mycology.* Moscow, p.344, 1995 (in Russian).

Nebulishvili E. Cand. (PhD) Thesis, Tbilisi, p.23, 1988 (in Russian).

Pidoplichko N. M., Milko A. A. *Atlas of mucoraceous molds.* Kiev, p.114, 1976 (in Russian).

Pidoplichko N.M. *Parasitic fungi of cultural plants. Imperfect fungi.* Guide-book, vol. **2**., Kiev, p.298, 1977 (in Russian).

Saccardo P. A. *Syllore Fungorum*, vol. **XIII**, Berlin, 1888.

Shoshiashvili I., Kirimelashvili N. Proceedings of Inst.Plant Prot. of Georg.SSR, **VIII**, 109-118, 1952 (in Georgian).

Yatchevskii A. A. *Fungi identification, imperfect fungi.* Vol. **2**, Petrograd, 580 p.1917, (Russian).

Zhvania N. Cand. (PhD) Thesis, Tbilisi, p.23, 1985 (in Russian).

ახალი მონაცემები ხახვის (*Allium cepa* L.) სოკოვანი დაავადებების შესახებ

მეტრეველი ი., კუპრაშვილი თ.

ლ. ყანჩაველის მცენარეთა დაცვის ინსტიტუტი

(მიღებულია 06.11.2006)

რეზიუმე

ნაშრომში წარმოდგენილია 2004-2006 წლებში საქართველოს სხვადასხვა რაიონებში კერძო პირთა და ფერმერთა ნაკვეთებზე მოყვანილი და საზღვარგარეთიდან იმპორტირებული სარეალიზაციო ხახვის ბოლქვებზე გამოვლენილი სოკოების სია მოკლე დიაგნოზით, მოპოვების ადგილისა და დროის ჩვენებით. გამოვლენილია 5 სახეობის სოკო, რომლებიც დღემდე არ იყო რეგისტრირებული ხახვზე საქართველოში. ერთერთი მათგანი - *Epicoccum sp.* არ არის იდენტიფიცირებული სახეობამდე. აღწერილია სოკოს 4 სახეობა, ამათგან 2 სახეობა გამოვლენილია ადგილობრივ, და 2 - სხვა ქვეყნებიდან შემოტანილი ხახვის მასალაზე.

ინსტრუქცია ავტორთათვის

სამეცნიერო ნაშრომი გამოიცემა 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.

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 ფრჩხილებში ჩასმული ავტორის გვარითა და წლით [Lernmark, Hagglof 1981].

მითითებული ლიტერატურა წარმოდგენილი უნდა იყოს შემდეგნაირად:

ჟურნალის შემთხვევაში

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.

წიგნის შემთხვევაში

Kuhn T.S. *The structure of scientific revolutions.* Chicago, IL, Chicago Press, 2000.

Brush S. *Flowing waters or teeming crowds.* In: Mental Models. D. Gentner (Ed.), Chicago IL., Chicago Press, 865-900, 2001.

მასალა რედაქციაში წარმოდგენილი უნდა იყოს ქაღალდზე ამობეჭდილი და დისკეტით (ან CD-ით). წერილი ერთი ფაილით უნდა იყოს შენახული (ცალკე ფაილად შეიძლება ილუსტრაციების წარმოდგენა), ხოლო ფაილის სახელწოდება წერილის პირველი ავტორის გვარს უნდა ატარებდეს.

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. წერილი 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.

ჟურნალის გამოცემა 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.

ნაშრომის 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.

ISSN 1512-2123

საქართველოს მეცნიერებათა აკადემიის

მაცნე

ბიოლოგიის სერია B
Biological series

2007
No. 1
Vol. 5

PROCEEDINGS

of the Georgian Academy of Sciences

ISSN 1512 – 2123

საქართველოს მეცნიერებათა აკადემიის

მაცნე

ბიოლოგიის სერია B
Biological series

2007
No. 1
Vol. 5

PROCEEDINGS

of the Georgian Academy of Sciences

Tbilisi. Georgia
2007

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

Published by NGO "Biomed", 14, L. Gotua st., Tbilisi 0160, Georgia

CONTENTS

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Biochemistry

THE IMPACT OF BIOACTIVE PREPARATIONS ON THE RESISTANCE OF RYEGRASS EXPOSED TO ORGANIC TOXICANTS – BENZENE, 3,4-BENZPYRENE AND TRINITROTOLUENE

*TSULUKIDZE N., BETSIASHVILI M., DZAMUKASHVILI N., SADUNISVILI T., KVESITADZE E.

Durmishidze Institute of Biochemistry and Biotechnology
**E-mail: ninuca_18@yahoo.com*

(Received October 16, 2006)

Abstract

Changes of the activities of glutathione S-transferase, phenoloxidase and peroxidase and of protein content in ryegrass, in response to the effect of organic pollutants – benzene, 3,4- benzpyrene (Bp), trinitrotoluene (TNT) and bioactive preparations, have been studied. It has been established that in most cases, during treatment of ryegrass seedlings with bioactive preparations – Fosnutren and Humiforte, enzyme activities and the protein amount have increased dramatically. Combined effects of benzene, Bp and TNT with bioactive preparations on enzymatic systems of ryegrass have been studied. It has been ascertained that treatment with bioactive preparations caused increase in the activities of glutathione S-transferase, phenoloxidase and peroxidase and protein amount in ryegrass that enhance the plant resistance to organic toxicants.

Key-words: Fosnutren, humiforte, glutathione S-transferase, ryegrass, organic pollutants

Introduction

The most effective remediation, perfect restoration and long-term preservation of chemically contaminated environment are possible by application of phytoremediation technologies. Phytoremediation involves clarification and restoration of chemically contaminated environment by means of plants and microorganisms, which are able to utilize and transform wide range of organic and inorganic toxicants. The plant (with its detoxification potential) capable to utilize the toxicants from – air, soil and water, all three elements of biosphere, is the most efficient mean for restoration of ecologically sound environment [Korte, 2000].

Detoxification process of toxic compounds in plant cell proceeds in three phases: reaction of activation, conjugation and compartmentation [Coleman, 1997]. In the first phase, hydrophilic group is formed in xenobiotic molecules at the expense of enzymatic transformation. As a result, the polarity and reactivity of toxicant molecule significantly increases. Various enzymes, including peroxidases and phenoloxidases, catalyze the activation reaction of xenobiotics [Kvesitadze et al., 2006].

Peroxidase (EC 1.11.1.7) is a widely spread enzyme, found in all green plants, fungi and aerobic bacteria. In a plant cell these enzymes display diverse functions, participate in a number of physiological and detoxification processes: hormonal regulation, lignification, and response to stress conditions, protection of a cell from infections and hydroperoxides. Free radical containing products formed as a result of reactions catalyzed by peroxidase are able to oxidize other compounds, including xenobiotics. Peroxidases of different plants are able to oxidize dimethylaniline, benzpyrene, phenol, aminofluorine and hydroxianysoles [Siegel, 1993].

Phenoloxidase (EC 1.14.18.1) is a copper containing metaloenzyme widely spread in microorganisms, plants, insects and animals. Phenoloxidase actively participate in oxidative degradation of organic toxicants. If the xenobiotic is of phenolic nature, then it is a substrate of phenoloxidase and oxidized in monophenolase and diphenolase reactions. In other cases, xenobiotic oxidation is carried out by co-oxidative mechanism by endogenous phenols [Papunidze et al., 2005].

In the second, conjugation phase xenobiotic or metabolite, activated in the first phase, connect with endogenous hydrophilic molecule. The obtained conjugate is polar and less toxic. In the third phase, compartmentation of inactive water-soluble conjugates takes place in vacuoles or cell wall.

Glutathione S-transferase (GST, EC. 2.5.1.18) is a representative of cytosolic enzymes. This dimmeric enzyme catalyzes bonding of tripeptide glutathione to electrophylic sites of various organic and inorganic molecules. Detoxification of various endo and xenobiotic compounds through this enzyme occurs as a result of covalent bonding between hydrophobic substrate and SH-group of cysteine residue in glutathione [Armstrong, 1997]. The obtained conjugate is less reactive and polar that simplifies its further compartmentation [Coskun, 2002]. In analogue with other detoxification enzymes, some isomers of this enzyme are inducible. Their intracellular level can increase as a result of effect of plant hormones, pathogens and xenobiotics (e.g. herbicides) [Lamoureux, 1989; Mars, 1996].

Frequently, under the effect of toxic compounds enhancement of protein biosynthesis processes in plant cells is observed. Increase of protein amount on one hand promotes balance of protein deficiency, found at conjugation with toxic compounds as a result of protein expenditure, and on the other hand is connected with induction of enzymes participating in detoxification processes [Kvesitadze et al., 2005].

At elaboration of new technologies, great importance is attached to the approaches that enable to regulate ecophysiological characteristics of plants without interfering their genome. In this point of view, the preparations of Spanish firm INAGROSA, particularly Fosnutren and Humiforte are of interest. These preparations are complex of synthesized free amino acids and microelements, applying of which significantly enhance plant productivity, and at the same time, their resistance to different toxicants effects. It should be mentioned that their absorption does not consume plant energy and depend on chlorophyll activity.

Ryegrass is widely applied for planting lawns in cities and along motorways. Consequently, evaluation of the plant resistance to different toxicants, prevalent in the environment and its detoxification capability is of importance.

Some compounds, widely spread in the environment and characterized by high toxicity have been chosen for the experiments. At present, motor transport takes the main place in environmental pollution in developed countries. Exhaust, together with different toxic compounds contains benzene and 3,4-benzpyrene. Compounds containing aromatic rings are extremely toxic and carcinogenic [Samoillloff, 1998; Curfs, 2003]. Among explosives, trinitrotoluene is the most toxic. Hundreds of hectares of contaminated soil remain after hostilities and military exercises [Robidoux, 1999].

The goal of our investigation was to estimate combined impact of bioactive preparations and organic pollutants on ryegrass seedlings in order to increase its phytoremediation capability.

Materials and Methods

The object of the study was ryegrass, seeds of which were swollen and after grown in tap water during 10 days. To induce enzymes the seedlings were placed in solutions containing benzene (0,1mM), 3,4-benzpyrene (0,1mM) and trinitrotoluene (0,1mM), during 5 days. Fosnutren and Humiforte, bioactive preparations were added together with toxicants in concentrations of 5 ml/l. After exposure, roots and leaves were cut and homogenized in a mortar in 0.05M phosphate buffer pH 7.5, in the ratio 1:3. After centrifugation at 12000g, 30 min, the obtained supernatant was studied for enzymes activity.

Glutathione S-transferase activity was determined spectrophotometrically at 340 nm, by measuring the rate of 1-chloro 2,4-dinitrobenzene (CDNB) conjugation with reduced glutathione [Habig, 1974]. Reaction mixture contained 1mM glutathione and 0.1ml enzyme preparation in 0.2M phosphate buffer, pH 6.5, final volume 3ml. Reaction started by addition of 1mM CDNB. Analogous mixture of the same content without enzyme preparations was used as a control.

As a unit of glutathione S-transferase activity the enzyme amount, which catalyzes conjugation of 1mM CDNB with glutathione in 1 min at 25°C is taken.

Peroxidase activity was determined spectrophotometrically at 470 nm according to the rate of guaiacol oxidation [Gregori 1972]. Reaction mixture contained 10mM guaiacol, 0.5 ml H_2O_2 solution (0.3%) and 0.01ml enzyme preparation in 0.05M phosphate buffer, pH 5.4, final volume 3ml.

As a unit of peroxidase activity the enzyme amount catalyzing oxidation of 1mM guaiacol in1 min at 25°C is taken.

Phenoloxidase activity was determined spectrophotometrically at 430 nm according to the rate of pyrocatechine oxidation [Lanzarini, 1972]. Reaction mixture contained 2mM pyrocatechine and 0.1ml enzyme preparation in citrate buffer, pH 4.7, final volume 3ml. Enzyme activity is expressed in ΔE/min.

The enzyme activities were calculated per g of fresh weight and expressed in percents in Tables. Protein was determined by Bradford's method [Bradford, 1974]. As a standard - bovine serum albumin was used.

Results and Discussion

Changes in the activities of glutathione S-transferase, peroxidase and phenoloxidase and protein content have been studied in roots and leaves of ryegrass, treated by biopreparations (Fig. 1). According to the obtained results, glutathione S-transferase and peroxidase activities were enhanced in roots and leaves of plants treated with biopreparations. Increase of peroxidase activity was observed only in plant roots, where the enzyme activity, in comparison with leaves, is higher, according to the literature data as well [Siegel, 1993]. Protein content in treated plants increases significantly both in roots and in leaves. Especially significant increase of protein amount, by 100% was observed in plants treated with Humiforte.

Changes in activities of glutathione S-transferase, peroxidase and phenoloxidase and in protein content were studied at exposure to different toxicants together with bioactive preparations in seedlings of ryegrass. At exposure of seedlings to benzene, significant increase in activities of enzymes and protein content was observed in experiments, when the plant was treated with toxicants together with bioactive preparation (Fig. 2). Different pictures were observed in roots and

leaves. Increase of activities in enzymes participating in detoxification was clearer; presumably, it is connected with treatment method of plants as roots are in direct contact with xenobiotics and with the ability of biopreparations to resist and limit transportation of toxicants to aboveground organs [Kvesitadze, 2005]. On such ability of Fosnutren and Humiforte indicates also significant increase of protein amount in roots, and decrease of protein content in leaves. In that case, obvious difference between stimulating effects of Fosnutren and Humiforte was not observed.

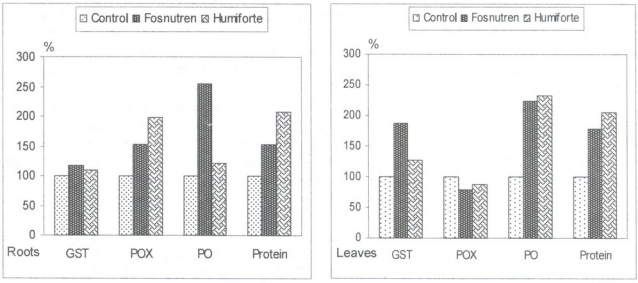


Fig. 1. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to Fosnutren and Humiforte. Exposure time – 5 days. Concentrations of Biopreparations – 5ml/l.

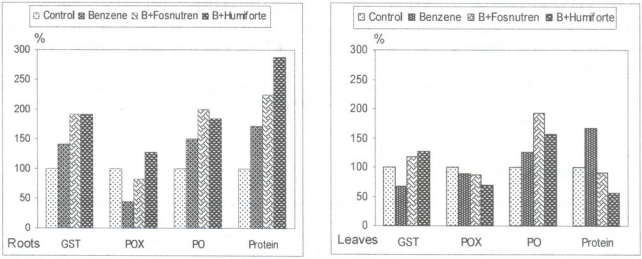


Fig. 2. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to benzene, Fosnutren and Humiforte. Exposure time – 5 days. Concentration of benzene – 0.1mM/l. Concentrations of Biopreparations – 5ml/l.

Changes in enzymes activities in response to exposure to 3,4-benzpyrene (Bp), organic toxicant, have been studied (Fig. 3). As a result of the experiments, it has been established that mainly, glutathione S-tranferase and oxidation enzymes activities increase in roots and leaves of ryegrass, in response to treatment with xenobiotic. Decrease of protein amount in seedlings, treated with toxicants only, indicates on higher toxicity of benzpyrene in comparison with that of benzene.

However, under the influence of bioactive preparations this toxic effect is significantly decreased, while protein content and enzyme activity increased.

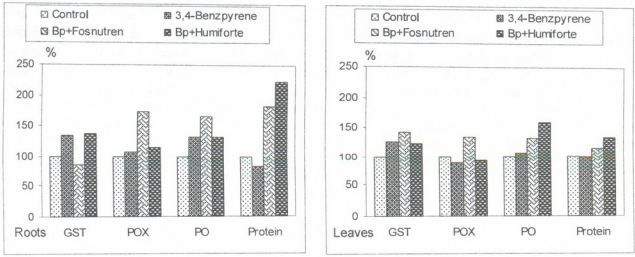


Fig. 3. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to 3,4-benzpyrene (Bp), Fosnutren and Humiforte. Exposure time – 5 days. Concentration of 3,4-benzpyrene – 01,mM/l. Concentrations of Biopreparations – 5ml/l.

The influence of trinitrotoluene (TNT) on seedlings of ryegrass has also been studied (Fig. 4). According to the obtained results, decrease in activity of oxidation enzymes is mainly observed in most cases, connected with another pathway of xenobiotic transformation in the studied plant. Increase of glutathione S-tranferase activity is observed in roots. Presumably, the enzyme renders safe active metabolites formed in the cell under toxicants exposure. Increase of protein content is found both in roots and leaves; it is especially sharp in the presence of bioactive preparations.

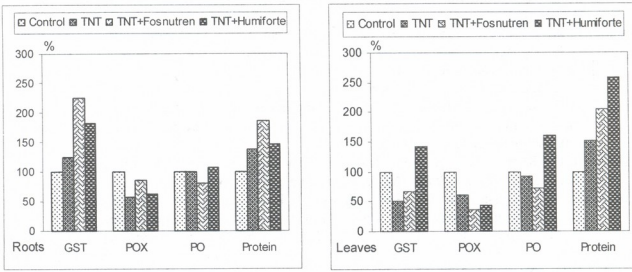


Fig. 4. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to TNT, Fosnutren and Humiforte. Exposure time – 5 days. Concentration of TNT – 0.1 mM/l. Concentrations of biopreparations – 5ml/l.

According to the obtained results, it might be concluded that at treatment with Fosnutren and Humiforte, in most of cases, activities of glutathione S-transferase, phenoloxidase, peroxidase, and protein content enhance dramatically in ryegrass. Application of bioactive preparations together with toxicants increases the plant resistance to xenobiotics as free amino acids and microelements are essential substrate for synthesis of enzymes and/or their substrates (glutathione in case of glutathione S-transferase). Ryegrass, as evergreen plant is widely used in decorating of cities and along motorways. On the base of our experiments, it might be concluded that application of the plant is desirable in combination with bioactive preparations, which significantly improve its phytoremediation capability. It should be also mentioned that the plant genome would not be affected; besides, it is safe for the environment. Phytoremediation itself is another method of nature protection from the dangerous impact of humans.

Acknowledgement: Biologically active preparations – Humiforte and Fosnutren were kindly provided by INAGROSA, Industrias Agrobiologicas, S.A.

References:

Armstrong R. *Structure, catalytic mechanism, and evolution of the glutathione transferases*. Chem. Res. Toxicol. **10**, 2-18, 1997.

Bradford M. *A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the triple of protein dye binding.* Anal. Biochem., **59**, 277-282, 1974.

Coleman J., Mechteld M., Emyr Davis T. *Detoxification of xenobiotics by plants: chemical modification and vacuolar compartmentation.* Trends in Plant Science, **2**, 141-151, 1997.

Coskun G., Zihnioglu F. *Effect of some biocides on glutathione-s-transferase in barley, wheat, lentil and chickpea plants.* Turk J. Biol., **26**, 89-94, 2002.

Cufts D., Beckers L., Godschalk R., Gijbels M., Van Schooten F. Environ. Mol. Mutagen. **42**, 243-249, 2003.

Gregory R. *A rapid assay for peroxidase activity*. Biochem. J., **101**, 582-583, 1966.

Habig W., Jakoby M. *Glutathione S-transferases – the first enzymatic step in mercapturic acid formation*, J. Biol. Chem., **22**, 7130-7139, 1974.

INAGROSA, Industrias Agrobiologicas, S.A.

Korte F., Kvesitadze G., Ugrekhelidze D., Gordeziani M., Khatisashvili G., Buadze O., Zaalishvili G., Coulton F. *Review: Organic toxicants and plants*. Ecotoxicology end Environmental Safety. **47**, 1-26, 2000.

Kvesitadze G., Khatisashvili G., Sadunishvili T., Evstigneeva Z. *Metabolism of Anthropogenic Toxicants in Higher Plants*. Moscow, "Nauka", 2005 (In Russian).

Kvesitadze G., Khatisashvili G., Sadunishvili T., Ramsden J. *Biochemical mechanisms of detoxification in higher plants. Basis of phytoremediation.* Springer, 2006.

Lamoureux G., Rusness D. *The role of glutathione and glutathione-S-transferases in pesticide metabolism, selectivity, and mode of action in plants and insects*. Wiley Interscience, 153-196, 1989.

Lanzarini G., Pifferi P., Samorani A. *Specificty of an o-diphenol oxidase from Punus avium fruits*. Phytochemistry, **11**, 89-94, 1972.

Marrs K.A. *The functions and regulation of glutathione S-transferases in plants.* Plant Mol. Biol. **47**, 127-158, 1996.

Papunidze V., Khatisashvili, G., Sadunishvili T. *Plant for a Healthy Environment*. 2005 (In Georgian).

Robidoux P., Hawari J., Thiboutot S., Ampleman G., Sunahara G. Ecotoxicol. Environ. Saf., **44**, 311-321, 1999.

Samoiloff M. *Benezene toxicity. Benzene and derivatives*. Organic chemistry **3**, 1998.

Siegel B. *Plant peroxidases - an organismic perspective*. J Plant Growth Regul., **12**, 303-312, 1993.

ბიოსასუქების გავლენა კოინდარის მდგრადობაზე ორგანული ტოქსიკანტების – ბენზოლის, 3,4-ბენზპირენისა და ტრინიტროტოლუოლის ზემოქმედებისას

წულუკიძე ნ., ბეციაშვილი მ., ძამუკაშვილი ნ., სადუნიშვილი თ., კვესიტაძე ე.

დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 16.10.2006)

შესწავლილია ფერმენტების – გლუტათიონ S-ტრანსფერაზას, ფენოლოქსიდაზას, პეროქსიდაზას აქტივობებისა და ცილის შემცველობის ცვლილება კოინდარში ორგანული ტოქსიკანტების – ბენზოლის, 3,4-ბენზპირენის, ტრინიტროტოლუოლისა (TNT) და ბიოსასუქების ზემოქმედების საპასუხოდ. დადგენილია, რომ კოინდარის ნაზარდების ბიოაქტიური პრეპარატებით – ფოსფუტრენითა და ჰუმიფორტეთი დამუშავებისას უმრავლეს შემთხვევაში მკვეთრად იმატებს ფერმენტების აქტივობა და ცილის რაოდენობა. შესწავლილია ბენზოლის, 3,4-ბენზპირენის, TNT-ს და ბიოსასუქების ერთობლივი გამოყენების გავლენა მცენარის ფერმენტულ სისტემებზე. დადგენილია, რომ ბიოაქტიური პრეპარატებით დამუშავება იწვევს კოინდარში გლუტათიონ S-ტრანსფერაზას, ფენოლოქსიდაზას, პეროქსიდაზას აქტივობისა და ცილის რაოდენობის მატებას, რაც ზრდის ამ მცენარის გამძლეობას ორგანული ტოქსიკანტების მიმართ.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Biotechnology

EFFECTIVE CONTROLLING OF BACTERIAL SPOT IN TOMATO WITH BACTERIOPHAGE

GHUDUMIDZE N.[1], ALAVIDZE Z.[2], CHKONIA I.[2], ELIASHVILI P.[3], GIORGOBIANI N.[3], SHAPOVALOVA N.[1], MEIPARIANI A.[2], SADUNISHVILI T.[1]

[1]*S. Durmishidze Institute of Biochemistry and Biotechnology*
[2]*G. Eliava Institute of Bacteriophages, Microbiology and Virology*
[3]*L. Kanchaveli Institute of plant protection*

(Received October 16, 2006)

Abstract

Tomato foliages were artificially contaminated with the culture washed out of 4 pathogenic strains of *Xanthomonas vesicatoria*. The disease developed on lower side as well as on upper side of foliages. The disease was caused by infection with pathogenic strains isolated from both damaged foliages and fruits. Spraying foliages with $7x10^6$ p.f.u. of phage just at the moment of infection or 24h later hindered the disease onset, but a week after affection suppressed the disease development. Unlike chemical substances phage did not require to be sprayed twice or several times. Phage does not damage the plant foliages and does not provoke the disease outbreak. Phage application is safe for a human as well as for a plant and it can be used as a salutary agent in dealing with damages caused by Bacterial Spot of sowing areas.

Keywords: Bacterial Spot, tomato foliages, chemical substances, bacteriophage.

Introduction

Bacterial Spot in tomato leaves is caused by phytopathogenic bacteria-*Xanthomonas vesicatoria*. Damage from these diseases may range from a light spotting of the foliage to almost complete defoliation of the plant, with corresponding impacts on the ability of photosynthesis and production potential [Leboeuf et al., 2005], and therefore decreases the yield of tomato crops. When the conditions are optimal for bacterial multiplication (high humidity, 28-32°C.) loss in tomato crops marketable yield can be great.

In order to control the Bacterial Spot the chemical substances are used. Treatment with acid or chlorine may be comparatively effective, if properly manage. But chlorine is inactivated by organic matter and its activity is affected by the pH of solution. Clearly, maintaining the accurate pH is a critical moment for successful disinfection with acid [Leboeuf et al., 2005].

Today the most effective bacteriocide against tomato Bacterial Spot in Georgia still has been copper sulfate spray. But copper sprays are less effective, when spray intervals are extended (7 days or less interval is required). Bacterial populations may show wide resistance to the given solution. Application of high concentrations of copper ions can damage plant tissue leading to a rapid bacteria reproduction. Copper spray may suppress the bacteria on the foliage surfaces, but bacteria located deeply may survive, multiply and cause an outbreak [Leboeuf et al., 2005].

Application of Cixomi and other copper-containing preparations including Kupxodat, Kuprophlo also are used for controlling the tomato Bacterial Spot.

All these compounds in certain amounts may penetrate into a human organism. The prevalence of the toxic substances can hinder such biological processes as growth, development, propagation and in some cases even stop them. Pesticides play a key role in xenobiotics, although, humans have to use pesticides, which finally get into biosphere and humans become a target of their action [Jurin, 2002].

At present the researchers try to study other desinfectants and alternative methods including treatment by microware, sonication and hydrostatic pressure.

One of the encouraging alternative ways in combating against Bacterial Spot in tomato is an application of bacteriophages for treatment purposes, as phages are a specific kind of viruses that attack suitable bacteria to kill pathogenic microorganisms.

The goal of our paper was to show the possibilities of phage application as an alternative tool to chemical substances and a natural biocontrolling agent against Bacterial Spot caused artificially on the tomato foliages.

Materials and Methods

In our experiment was used: *Lycopersicum esculentum* 45 day seedlings; 24h. the culture washed out of 4 various pathogenic strains of *Xanthomonas vesicatoria,* among them 3 strains isolated from the damaged tomato foliages and 1 - from damaged fruit; $7x10^6$ p.f.u. phage mixture of pure lines of mixture of phages isolated from sewages and damaged tomato materials.

The leaves selected for controlling were mechanically damaged by scratching the leaves with a needle. Infection was performed by dropping the bacterial culture onto the mechanically damaged plant leaves [Baltyukova et al., 1968].

The culture was dropped by micropipette. One strain infected some foliages located on various branches of one plant, which were mechanically damaged and added with drops of culture on both, lower and upper sides of foliages; the phage was sprayed by means of special sprayer.

Experiment procession. Tomato seedlings were planted separately into the pots and placed in the greenhouse. For trial 25 days later healthy plants were selected and divided into 5 groups. Control plants were included into I group - a total, 2 plants. The plant foliages of II group were infected with *Xanthomonas vesicatoria* culture - a total, 4 plants. This group was designed for bacterial controlling. The plants, foliages of which were sprayed with phage just at the moment of affection, were included into III group - a total, 4 plants. The plant foliages of IV group were sprayed with phage 24h after affection - a total, 4 plants. The plant foliages of V group were sprayed with phage after a week - a total, 4 plants.

The plants were placed in the thermostat room at constant temperature 28°C. The aeration was performed by airing the room. By day the light was switched on. High humidity was provided by frequent watering and natural evaporation of water from watery vessels.

Results and Discussion.

Observation was carried out within 4 week. It is remarkable, that the signs of bacterial spotting among the plants infected with bacteria were detected only 4 days after affection (II and IV groups) and only on the 5th day the disease developed in a shape of brownish spots on the lower and upper sides of the foliages, infected with pathogenic strains isolated from both damaged foliages and fruits.

During the whole experiment no signs of disease development were observed on the foliages of I group plants (Fig.1). As a result of the disease development on the plant foliages in II group, damaged leaves appear yellow (10 days) (Fig.2). Subsequently all leaves of the branches entirely yellowed. No signs of disease were observed on the plant foliages in III and IV groups within the whole period. After production of Bacterial Spot on the plant leaves in V group the disease was eliminated by phage spray - light spots remained on the infected sites (Fig.4), but the disease did not develop. Evidence of disease was not observed within 3 weeks. Thus, the results of the experiment demonstrated that bacteriophage application succeeded in dealing with bacterial spotting developed on the tomato foliages being infected with *Xanthomonas vesicatoria*. Phage hindered the disease onset, when it was sprayed just at the moment of bacteria affection (Fig.3); after 24h and even after 7 days. It is remarkable, that during the experiment in the case of each variation phage was used only once.

Fig.1. The control plant. (10th day of observation).

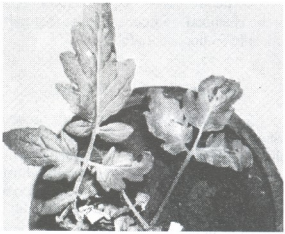


Fig.2. The bacterial-control plant (10th day of observation).

Fig.3. Phage was sprayed at the moment of infection (the 10-th day of observation).

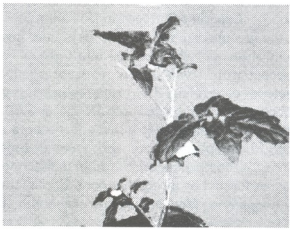


Fig.4. Phage was sprayed after a week of Infection (the 10-th day of observation).

Thus, phages can be used as an effective treatment remedy against Bacterial Spot produced on the tomato foliages. In comparison with chemical substances phages have some advantages: phage preparation is cheaper; when penetrating into any infected site the phage remains there until suitable bacterium exists; phage application does not damage plant organisms even in the case of a high concentration. When getting into a human organism through tomato the phages are safe unlike the chemical substances.

Acknowledgement: This work was carried out with financial assistance of ISTC within the framework of the project NG-1129.

References:

Baltjukova K., Matishevskaja M., Kulikovskaja M., Sudorenko C. *Methods for studying the causative against of plant bacterial disease*. Kiev. Nauka Dumkova. 1968.

Jurin V. *Basis of xenobiology*. Minsk. Novaja Znanie. 2002.

Leboeuf J., Cuppels D., Dick J., Pitblado R., Poewen St., Celetti M. *Bacterial diseases of tomato*; *Bacterial Spot*, *Bacterial Speck*, *Bacterial Canker*. Queen's Printer for Ontario. Factsheet ISSN 1198-712X, 363-365, 2005.

პამიდვრის ბაქტერიული სილაქავის ეფექტური მკურნალობა ბაქტერიოფაგით

დუდუმიძე ნ.[1], ალავიძე ზ.[2], ჭყონია ი.[2], ელიაშვილი ფ.[3], გიორგობიანი ნ.[3], შაპოვალოვა ნ.[1], მეიფარიანი ა.[2], სადუნიშვილი თ.[1]

[1]ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი
[2]გ. ელიავას ბაქტერიოფაგის, მიკრობიოლოგიისა და ვირუსოლოგიის ინსტიტუტი
[3]ლ. ყანჩაველის მცენარეთა დაცვის ინსტიტუტი

(მიღებულია 16.10.2006)

რეზიუმე

Xanthomonas vesicatoria-ს 4 პათოგენური შტამის ჩამონარეცხი კულტურით მოხდა პამიდვრის ფოთლების ხელოვნური ინფიცირება. დაავადება განვითარდა ფოთლების როგორც ქვედა, ასევე ზედა მხრიდან ინფიცირების შემთხვევაში. ფოთლებში დაავადება გამოიწვია როგორც დაავადებული ფოთლებიდან, ასევე ნაყოფიდან გამოყოფილი პათოგენური შტამებით დახნებოვნებამ. $7x10^6$ ტიტრის მქონე ფაგის ფოთლებზე შესხურებამ ინფიცირებისთანავე, ან 24 საათის შემდეგ ხელი შეუშალა დაავადების დაწყებას, ხოლო ინფიცირებიდან 1 კვირის შემდგომმა შესხურებამ შეაჩერა დაავადების განვითარება. ქიმიური საშუალებებისგან განსხვავებით, ფაგის მეორე ან მრავალჯერადი შესხურება არ იყო აუცილებელი, ფაგი არ აზიანებს მცენარის ქსოვილებს და ამით არ ახდენს დაავადების აფეთქების პროვოცირებას. ფაგის გამოყენება უსაფრთხოა როგორც თვით მცენარისთვის, ასევე ადამიანისთვის და შესაძლებელია გამოყენებული იყოს ნათესი ფართობების ბაქტერიული სილაქავისგან გამოწვეული დაზიანების საწინააღმდეგო სამკურნალო საშუალებად.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Botany

INTRASPECIFIC CHEMICAL DIFFERENTIATION OF *URTICA DIOICA* L. GROWING IN GEORGIA

KAVTARADZE N., GVINIASHVILI TS., ALANIA M., KUCHUKHIDZE J.

Kutateladze Institute of Pharmacochemistry

(Received May 15, 2006)

Abstract

Distinctive chemical and morphological features of *Urtica dioica* L. growing in Georgia have been revealed. *Utrica dioica* with red coloration was defined as a new variety - *Utrica dioica* L. var. rubescens Gviniaschvili et Kavtaradze – planta cum anthocyanea.

Key words: *Urtica dioica* L., flavonols, anthocyans.

Introduction

80 species of the genus Urtica L. are spread in temperate and tropical zones of both hemispheres [Mabberley, 1998]. Two species *Urtica dioica* L. and *Urtica urens* L. grow in Georgia [Shkhian, 1975]. They are cosmopolites, follow humans and are met in the conditions of violated natural plant cover. *Urtica dioica* L. is distributed everywhere. It grows in ruderal places, near housings, drafts and banks.

In folk and scientific medicine tincture and decoction of *Urtica dioica* L. is used as hemostatic agent [Budantsev et al., 1985]. Roots and rhizomes of this plant present a raw material for medicinal preparation used at prostate adenoma [Gide-book Vidal, 2002]. Data about phenolic composition of *Urtica dioica* occur in scientific literature. Flavonoids [Chaurasia, 1987], lignins [Kraus et al., 1990], coumarins [Wichtl, 1996] were isolated from *U. dioica*.

Chemical characteristics of *Urtica dioica* growing in Georgia were not studied yet. Phytochemical analysis revealed that qualitative chemical compositions of above-ground parts of *Urtica dioica* growing in various regions of Georgia are different.

The goal of our research was to find out differences of chemical compounds in correlation with morphological characteristics of the plant.

Materials and Methods

U. dioica was studied during 2000-2003. Herbarium material was collected in East Georgia (Kartli, Gori district) and West Georgia (Khobi district, village Alioni) during mass florescence of plant (July-August).

Traditional methods of chemical analysis were used [Alania et al., 2002]. Macromorphological studies of plant were also carried out (Table 2).

Results and Discussion

Our field investigations showed that plants of common *U. dioica* growing throughout the country develop green above-ground parts, but the specimens collected in West Georgia – red ones. Flavonoid glycosides were isolated from those plants and characterized (Table 1).

As is seen from the data given in the table *U. dioica* green is sharply distinct by qualitative flavonoid composition from that of *U. dioica* red. Standard green specimens synthesized only flavonol derivatives - kaempferol and quercetin, but red specimens – flavonols and in addition anthocyans, derivatives of pelargonidin [Kavtaradze et al., 2001; Alania et al., 2002; Kavtaradze et al., 2003].

In order to establish the relation of chemical heterogeneity of material with its morphology we have carried out comparative study of macromorphological characteristics of researched plants (Table 2).

As is seen from the data given in the table specimens are distinguished by the level of pubescence, by the size and level of lignification of stem, by coloration of rhizome, stem, footstalk, rib and lamina.

While field investigations it was noted that red coloration of edges and nodes of rhizomes, from which shoots of stems, footstalks, ribs and lamina are grown, is persisted during the whole vegetation and generational period, which is the very feature that distinguishing it. There are some data in scientific literature that among *U. dioica* occur some specimens, which stems may have coloration caused by presence of anthocyanin pigments changing into brown coloration at the moment of florescence [Medvedev, 1934].

It was revealed that in West Georgia red and green specimens of *U. dioica* could grow together maintaining their characteristic morphology and qualitative chemical composition (Tables 1, 2).

Table 1. Flavonoids isolated from above-ground parts of *Urtica dioica*.

Specimens	Isolated compounds	Empiric formula	Melting temperature, °C	Literature
Green specimens	Flavonols Quercetin (3,5,7,3',4'-pentahydroxiflavon)	$C_{15}H_{10}O_7$	303-306	Kavtaradze et al., 2001
	Isoquercitrin (quercetin-3-O-β-D-glucopyranside)	$C_{21}H_{20}O_{12}$	221-224	" "
	Hyperin (quercetin-3-O-β-D-galactopyranoside)	$C_{21}H_{20}O_{12}$	232-235	" "
	Rutin (quercetin-3-O-β-D-rutinoside)	$C_{27}H_{30}O_{16}$	187-189	" "
	Kaempferol-3-O-tri-galactoside	$C_{33}H_{40}O_{20}$	-	Alania et al., 2004
Red specimens	Anthocyans Pelargonidin-3-xyloside	$C_{20}H_{20}O_9$	260 (with decomposition)	Kavtaradze, Alania, 2003
	Pelargonidin-3-xylobioside	$C_{25}H_{28}O_{14}$	170 (with decomposition)	" "
	Pelargonidin-3-gluco-galactoside	-	-	" "
	Flavonols Nicotiflorin (kaempferol-3-0-β-D-rutinoside)	$C_{27}H_{28}O_{16}$	178-180	Alania et al., 2002
	Rutin (quercetin-3-O-β-D-rutinoside)	$C_{27}H_{30}O_{16}$	187-189	" "

Table 2. Comparative morphological characteristics of *Urtica dioica* L. specimens

Organs and the main morphological features		Characterization	
		Green specimens [Medvedev, 1934]	Red specimens
Rhizome	Form	Tetraquetrous, nodular	Tetraquetrous, nodular
	Coloration	Yellow (sapling) Brownish (mature)	Orange-yellow Edges and nodes of rhizome are red (both, saplings and mature ones)
	Pubescence	Not pubescent	Not pubescent
Stem	Form	Tetraquetrous, rather drooping, thin;	Tetraquetrous, rather drooping, thick
	Height	0.5-1.0 m, in average	1.2-1.8 m, in average
	Butt diameter	5-10 mm, in average	10-17 mm, in average
	Coloration	Green	Red
	Pubescence per 1 cm of the length Number of stinging hairs	10-15	25-35
	Number of usual hairs	300-340	430-460
Leaves	Form and average sizes	Bottom – cordiform-elongated, sharpened; Middle – cordiform-lancet. Upper – nearly lancet (male plants), spear-shaped (mother plants)	Bottom – cordiform-elongated, sharpened; Middle – cordiform-lancet. Upper – nearly lancet (male plants), spear-shaped (mother plants)
	Leaf edges of the bottom layers	Crenate, coarse-toothed	Crenate, coarse-toothed
	Leaf edges of the upper layers	Serrate	Serrate
	Leaf edges of the lateral shoots	Serrate	Serrate
	Coloration of the upper side	Succulent green or dark green	Succulent green with dark reddish coloration
	Coloration of the bottom side	Lighter compared to upper side of lamina	Saplings –succulent red Mature leaves - lighter
	Number of the main ribs	3	3
	Coloration of the ribs	Green	Red
	Pubescence per 1 cm Number of stinging hairs	15-22	38-46
	Number of usual hairs	370-395	525-540
Footstalk	Form	Round, from above - with deep sulcus lengthwise	Round, from above - with deep sulcus lengthwise
	Coloration	Green	Red during the whole vegetative and generational period
	Pubescence per 1 cm Number of stinging hairs	20-55	35-70
	Number of usual hairs	260-380	450-690

Thus, results of chemical analysis and comparative morphological study show morphological and chemical intraspecies heterogeneity of studied specimens of *U. dioica*. Hence,

the plant with red coloration is distinguished as a variety – *Urtica dioica* L., var. rubescens Gviniaschvili et Kavtaradze – planta cum anthocyanea (Georgia, Samegrelo, Khobi district, vil. Alioni, 07.08.2002, near private farm).

References:

Alania M.D., Gvazava L.N., et al. *Phenolic compounds of some plant species growing in Georgia.* Abstracts of the VI symposium on phenolic compounds. Moscow, p. 95, 2004 (in Russian).

Alania M.D., Kemertelidze E.P., Komissarenko H.F. *Flavonoids of some species of Astragalus L. of Georgian flora.* Tbilisi, 2002 (in Russian).

Alania M., Gvazava L., et al. *Polyphenol and cycloartane compounds from some genera of plants of Georgian flora.* Abstracts of the 1st International Pharmaceutical Congress. Tbilisi, p.4, 2002.

Budantsev A.L., Fokina G.A. *Urticaceae Family.* In: Plant resources of USSR: flowering plants, their chemical composition and usage. Leningrad, p. 137-140, 1985 (in Russian).

Chaurasia N., Wichtl M. *Flavonoglycoside aus U. dioica.* Planta Med., **53**, 432-434, 1987.

Gide-Book Vidal. Medicinal Preparations in Russia. Loviisa, "Prostavern Urtica Likvid", p. 710, 2002.

Kavtaradze N.Sh., Alania M.D., Aneli J.N. *Chemical components of U. dioica growing in Georgia.* Chemistry of Natural Compounds, **3**, p.244, 2001 (in Russian).

Kavtaradze N.Sh., Alania M.D. *Anthocyan glycosides of U. dioica.* Chemistry of Natural compounds, **3**, p. 249, 2003 (in Russian).

Kraus R., Spitteler G. *Phenolic compounds from roots of U. dioica.* Phytochem., **29**, 5, 1653-1659, 1990.

Marrberley D.J. *The Plant-Book.* Cambridge University Press, p.55, 1998.

Medvedev P.F. *Utrica of USSR.* Leningrad, 1934 (in Russian).

Shkhian A.S. *Flora of Georgia.* Tbilisi, p.84-85, 1975 (in Georgian).

Wichtl M. *Brennesselblatter. DAB-Kommentar. Wesenschaftliche Erlauterungen zum Deutschen Arzneibuch. Monographin A-C.* Stuttgart-Frankfurt a.m./Eschborn, Bd.II/1, 1996.

საქართველოში გავრცელებული *Urtica dioica* L.-ს შიდასახეობრივი ქიმიური დიფერენციაცია

ქავთარაძე ნ., ღვინიაშვილი ც., ალანია მ., კუჭუხიძე ჯ.

ი. ჭუთათელაძის ფარმაკოქიმიის ინსტიტუტი

(მიღებულია 15.05.2006)

რეზიუმე

შესწავლილია საქართველოს სხვადასხვა რაიონში გავრცელებული *U. dioica*–ის ქიმიური და მორფოლოგიური თავისებურებები. გამოვლენილია *U. dioica* – ის ახალი სახესხვაობა წითელი შეფერილობით - *Urtica dioica* L. var. rubescens Gviniaschvili et Kavtaradze.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Botany

ENDEMIC MEDICINAL PLANTS OF KHEVI (KAZBEGI REGION, GEORGIA)

KHAIKASHVILI KH.

Tbilisi Botanic Garden

(Received August 29, 2006)

Abstract

The paper deals with results of inventory of 18 endemic medicinal plant species, concerning their number, abundance, frequency, and life forms. Degree of threats with extinction of such rare species as *Thymus collinus, Valeriana cardanines* and *V. tilifolia* is in relation to their collection by local population.

Key words: endemic medicinal plants, endangered species, Khevi region, Georgia.

Introduction

Kazbegi region is located on the highest part of the Great Caucasus. Due to contrasting orographic, climate and edaphic conditions the region is characterized by its florocoenotic diversity and richness in endemic species. Thus, 26% of angiosperms of the Khevi flora are endemic, including 6 out of 11 Caucasian endemic genera.

Total number of endemic plants in Khevi flora accounts for 247 species, i.e. 51,1% of high-mountain endemic flora of the Greater Caucasus [Shetekauri, Gagnidze, 2000; Nakhutsrishvili et al., 2005].

It should be noted that among endemic plants of Khevi about 20 species are considered to be used in traditional and officinal medicine from which two species, *Betula raddeana* and *Senecio rhombifolius*, are included in Red Data Book of Georgia (1982). *B. raddeana* is also included in RDB of the former USSR (1984), and IUCN Red List (1997, 2000).

Materials and Methods

Inventory of the abundance of species was done according to Drude's 6-point scale [Drude, 1890]. Frequency has been evaluated after Braun-Blanquet (1951). Spectra of life forms were identified according to Raunkier (1934). Plot size for herbaceous plant was $10m^2$, for woody species - $20m^2$. Experimental plots were chosen randomly within the area of population investigated.

Results and Discussion

6 species (*Betula raddeana* Trautv., *Sorbus caucasigena* Kom. ex Gatsch., *Rosa buschiana* Chrshan., *R. didoensis* Boiss., *R. galuschkoi* Demurova, *R. oplisthes* Boiss.), out of

investigated 18 endemic medicinal plants, belong to woody plants. One species (*Thymus collinus* Bieb.) is semishrub. Herbaceous plants are represented by one bienial species, *Heracleum asperum* (Hoffm.) Bieb. and 10 perennial ones: *Cephalaria gigantea* (Ledeb.) Born., *Galanthus platyphyllus* Traub & Moldenke, *Galega orientalis* Lam., *Potentilla agrimonioides* Bieb., *P. caucasica* Juz., *Senecio rhombifolius* (Adams) Sch. Bip., *Thalictrum buschianum* Kem.-Nath., *Trifolium fontanum* Bobr., *Valeriana cardamines* Bieb., *V. tiliifolia* Troitzk.

The data on species inventory for plants under consideration are given in the following table.

Table. Data on medicinal plant species inventory for 10 m^2 and 20m^2 plots

Plants species	Elev.	Ex.	Steep.	Num.	Abun.	Freq.	Life forms
Betula raddeana	2000	N	45	4	Cop_2	2	Mezophanerophyte
Cephalaria gigantea	1700	-	15	8	Cop_2	1	Hemicryptophyte
Galanthus platyphyllus *	2350	E	20	41	Sp	4	Cryptophyte
Galega orientalis	2000	N	45	9	Cop_1	1	Hemicryptophyte
Heracleum asperum	1600	-	-	11	Cop_2	2	Hemicryptophyte
Potentilla agrimonioides	1800	S	40	-	Cop_2	2	Hemicryptophyte
P. caucasica	1700	-	-	16	Cop_2	2	Hemicryptophyte
Rosa buschiana	2000	N	40	3	Cop_1	2	Nanophanerophyte
R. didoensis	1700	W	20	-	Cop_1	2	Nanophanerophyte
R. galushkoi	1600	-	-	-	Cop_2	2	Nanophanerophyte
R. oplisthes	1900	N	45	-	Cop_2	2	Microphanerophyte
Senecio rhombifolius	1900	N	30	5	Sp	1	Hemicryptophyte
Sorbus caucasigena	2000	E	45	4	Cop_2	2	Microphanerophyte
Thalictrum buschianum	1900	E	40	12	Cop_2	2	Hemicryptophyte
Thymus collinus	1700	E	40	-	Cop_2	3	Chamaephytes
Trifolium fontanum	1700	-	-	-	Cop_2	3	Hemicryptophyte
Valeriana cardamines	2000	N	45	-	Cop_2	1	Hemicryptophyte
V. tiliifolia	2000	N	40	7	Cop_2	1	Hemicryptophyte

Abbreviations and sign: Elev. - Elevation m a.s.l., Ex. - Exposition, Steep. - Steepness in ^{0}, Num. - Number, Abun. - Abundance after Drude (1890), Freq. - Frequency after Braun-Blanquet (1951), Life forms after Raunkier (1934), *Endemic species of Georgia

As is shown in the table *Galanthus platyphyllus* growing on alpine meadows is very rare species. In spite of its rarity in Khevi region it is not utilized as a medicinal plants. Consequently this species is not under threat. According to Miller and others [Miller et al., 2006] *G. platyphyllus* is considered as IUCN Vulnerable category (VU) species.

Next rare endemic medicinal plant in the Khevi region is *Senecio rhombifolius* growing in subalpine tall herbaceous vegetation. Unlike other regions of Georgia in Khevi this wellknown and utilized medicinal plant is not collected for medicinal purposes. Consequently, in spite of scarce resources of this species it is not under threat in this region.

Comparatively abundant species is *Galega orientalis.* In Khevi it occurs in subalpine forests and forest margins, and not utilized by local population for medicinal purposes.

Rosa buschiana occupy dry rock and scree habitats in restricted area in subalpine and alpine belts. Next species, *R. didoensis* occurs in the forest margins and shrublands of upper mountain forests and subalpine belts. It must be noted that only fruits are used for medicinal purpose.

Valeriana cardamines and *V. tiliifolia* are more widely distributed and sufficiently abundant species. Their roots are intensively collected by local population for medicinal purpose. Consequently these species are under serious threat.

Thymus collinus is utilized by local population as medicinal and spice means. Consequently, its resources are gradually diminished.

As a result of cutting number of *Betula raddeana* as well as other components of subalpine forests, viz., *B.litwinowii, B. pendula, Acer trautvetteri* are also little by little diminished.

It is concluded that to maintain endangered medicinal and other species special protective measures need to be taken including strengthening the regime of Kazbegi State Reserve on that part of territory where medicinal plants are represented.

References:

Braun-Blanquet, J. *Planzensoziologie*, 2-e aufl. Wien, 1951, (After Jaroshenko P.A. Geobotany, Moscow, Leningrad, 1961 (in Russ.).

Drude, O. *Handbuch der Pflanzengeographie*, Stuttgart, 1890, (After Jaroshenko P.A. Geobotany, Moscow, Leningrad, 1961 (in Russ.).

IUCN Red List of Threatened Plants. Eddited by K. S. Walter, H.J. Cillet, World Conservation Monitoring Centre, 1997.

IUCN Red List of Threatened Species. Compiled by C. Hilton-Taylor, IUCN, Gland, Switzerland and Cambridge, UK, 2000.

Miller J.S., McCue K., Consiglio T., Stone J., Eristavi M., Sikharulidze Sh., Mikatadze-Pantsulaia Ts., Khutsishvili M. *Endemic medicinal plants of Georgia (Caucasus)*. Tbilisi, 2006.

Nakhutsrishvili G., Abdaladze O., Kikodze A. *Khevi, Kazbegi Region.* Tbilisi, 2005.

Raunkier C. *Life forms of plants and statistical plant geography.* N.Y., 1934 (After Jaroshenko P.A. Geobotany, Moscow, Leningrad, 1961 (in Russ.).

Red Data Book of Georgia. Tbilisi, 1982 (in Georg.).

Red Data Book of USSR, Moscow, 1984 (in Russ.).

Shetekauri Sh., Gagnidze R. *Diversity of high-mountain endemic flora of the Greater Caucasus.* In: Biological and landscape diversity of Georgia. Tbilisi, 151-158, 2000 (in Georg.).

ხეშო (ყაზბეგის რაიონი) გავრცელებული ენდემური სამკურნალო მცენარეები

ხაიკაშვილი ხ.

თბილისის ბოტანიკური ბაღი

(მიღებულია 29.08.2006)

რეზიუმე

სტატია ეხება ხეშის ფლორის 18 სამკურნალო ენდემურ სახეობას. მოცემულია მათი რაოდენობა, სიხშირე, შეხვედრილობა, სახიცოცხლო ფორმების სპექტრი. გამოვლენილია გადაშენების საფრთხის ქვეშ მყოფი ენდემური სახეობები.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Botany

ANALYSIS OF BRYOPSIDA SPECIES ACCORDING TO THE OCCURRENCE FREQUENCY IN THE FOREST BELT OF LAGODEKHI RESERVE

TIGISHVILI K.

N. Ketskhoveli Institute of Botany

(Received May 8, 2006)

Abstract

125 species of Bryopsida are divided into 5 groups according to the occurrence frequency. The highest mark, 5 points was conferred to the 6% of total number of mosses, 4 points - to the 19%, 3 points - to the 28%, 2 points- to the 30%, 1 point - to the 17 %. The obtained results show, that the species with medium and lower occurrence frequency comprise more than half of bryoflora of the lower forest belt. Widely spread species are characterized by the most limited occurrence frequency. Together with this, the comparison of systematical structure of bryoflora of the mentioned belt with the bryoflora of several regions of the East Trans-Caucasus is made on the example of 10 leading families.

Key words: East Trans-Caucasus, bryoflora, leading families.

Introduction

Mosses comprise a significant part of natural plant cover. Natural reserves are the objects for the protection of information resources and the investigation of untouched ecosystems in them is particularly important [Reimers, 1978].

The main objective of our investigation was thorough study of bryoflora of Lagodekhi Reserve. Revelation of the occurrence frequency of bryoflora is connected with many difficulties [Maslovski, 1991]. Despite this we made an attempt to solve the problem on the example of bryoflora of forest belt. The obtained results are of preliminary character.

Materials and Methods

Bryoflora of main ecosystems of forest belt of Lagodekhi Reserve has been studied using itinerary and semistationary methods. Material was taken and treated on sample plots according to the geobotanical method [Neshataev, 1987].

Results and Discussion

On the basis of rich bryological material, obtained in the forest belt of Lagodekhi Reserve (1200 samples) we made an attempt to establish the occurrence frequency of Bryopsida species. According to this feature Bryopsida species of the mentioned region have been divided into 5 main

groups: 1. species, spread in the most of coenoses and ecotopes, characterized with constant occurrence - 5 points; 2. species, which are not spread in the majority of coenoses and ecotopes, but are distinguished with rather high occurrence frequency in the main formations and the most significant ecotopes - 4 points; 3. species, which do not have wide distribution, but are distinguished by the certain occurrence frequency in some ecotopes - 3 points; 4. species, characterized by sporadic distribution and low frequency of occurrence - 2 points; 5. extremely rare species - 1.

Analysis, performed in order to reveal the frequency of occurrence of moss species has shown, that the highest evaluation was conferred to 6% of the total number of bryoflora species or to only 9 species. The following taxons: *Hypnum cupressiforme*, *Leucodon* spec., *Brachythecium rutabulum*, *Brachythecium populeum*, *Neckera besseri*, *Anomodon attenuatus* are characterized by wide distribution and constant frequency of occurrence. The 17 % of the total number of bryoflora species were evaluated by 1 point. Despite the fact, that the material was taken several times, some of these taxons ware found only once. The species *Pottia truncata*, *Phasum cuspidatum*, *Pseudoscleropodium purum*, *Breidleria arcuata*, *Pleurochaete squarrosa* belong to rare species.

The rest species, comprising the bryoflora of forest belt, are positioned between these two extreme groups. The taxons, evaluated by 4 points are not everytypes of the lower forest belt, but are widely spread in favourable econiches. Their number makes 19% of the total number of species. Sometimes, within the limits of synusia, they are distinguished by significant development of the biomass. The epilyths, spread in humid forests of river ravines: *Thamnium alopecurum*, *Ctenidium molluscum*, *Mnium undulatum,* the species, characteristic to Fagetum nudum: *Isothecium myurum*, *Pterigynandrum filiforme;* and *Thuidium philibertii*, characteristic to hornbeam forests and some others belong to such species.

The species, evaluated by 3 and 4 points comprise 30% and 28% of the main list. Taken separately they exceed the number of taxons of other groups and together they make more than half of the bryoflora species. Species, evaluated by 3 points, found only in some coenoses and ecotopes, are represented by the following taxons: *Eurhynchium striatum*, *Euryinchium zetterstedtii*, *Rhynchostegium riparioides*, *Mnium stellare*, *Orthorhichum diaphanum* and others. Distribution of 2-point species is rather limited, though their total number reaches 42. *Weisia controversa*, *Mnium selligeri*, *Bryum bicolor* and others can be listed among such species.

Based on the mentioned data it can be supposed, that bryoflora of the lower forest belt of Lagodekhi Reserve is mainly presented by the species of medium and low frequency of occurrence. The quantity of species of high frequency of occurrence is the most limited. Despite the wide ecological amplitude of mosses, narrow ecological conditions of the environment are the main factors, determining their distribution. Our data are in agreement with those presented in the scientific literature [Maslovski, 1991].

Taxonomic structure of mosses allows to establish the pattern of bryoflora for the mentioned region. Acoording to Tolmachev [Tolmachev, 1974] the pattern of bryoflora is reflected in the best way by the specific composition of ten leading families, which hold the dominant position in bryoflora by the number of species. Composition of bryoflora of the studied region, presented by the leading 10 families is as follows: **Brachytheciaceae** - 22 species, **Mniaceae** - 14 species, **Amblystegiaceae** - 15 species, **Bryaceae** - 13 species, **Pottiaceae** - 11 species, **Dicranaceae** - 9 species, **Grimmiaceae** - 8 species, **Thuidiaceae** - 5 species, **Hypnaceae** - 6 species, **Trichostomaceae** - 4 species. The 104 species or more than half of total bryoflora are united in 10 leading families. The zonal-floristic peculiarities of the region are well reflected by systematical composition of bryoflora. The presence of **Brachytheciaceae**, **Mniaceae** and **Amblystegiaceae** among leading families points to the prevalence of forest landscapes in the mentioned region and its mesophilic character. The families **Grimmiaceae** and **Dicranaceae** serve as the evidence for its mountainous relief. Despite the physicogeographical and vegetative

contrasts, characteristic to the Caucasus, we made an attempt to compare bryoflora of forest belt of Lagodekhi Reserve with that of some regions of East Trans -Caucasus [Chikovani, 1965; Lubarskaya, 1974; Manakian, 1989] (Table 1). It turned out, that 8 families are common for 10 leading families of all regions. This points to the common botanical-geographical character of the mentioned regions.

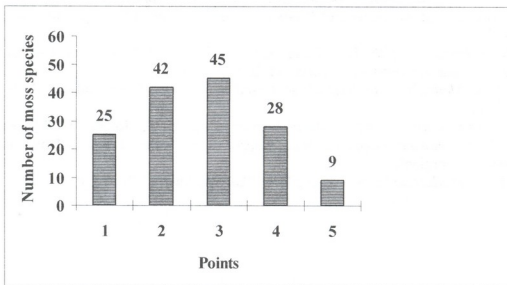


Fig. 1. Groups of Bryopsida species according to the frequency of occurrence.
1 point - 25 species (17% of total); 2 points - 42 species (28% of total); 3 points 45 species (45% of total); 4 points - 28 species (19% of total); 5 points 9 species (6% of total)

Table 1. Comparison of Lagodekhi Reserve forest belt bryoflora with that of some regions of East Trans-Caucasus.

Moss	Lagodekhi reserve		Nukha-Zaqatala		Gombori		NE Armenia		SE Armenia	
Family	Species number	Posi-tion	Species number	Position	Species number	Position	Species number	Position	Species number	Position
Brachytheciaceae	22	1	20	1	13	2	4	7	19	1
Mniaceae	14	3	6	8	6	7	4	8	5	8
Bryaceae	13	4	8	5	7	6	10	2	10	3
Amblystegiaceae	15	2	10	2	9	5	4	9	8	7
Dicranaceae	9	6	5	9	4	10	3	10	-	-
Grimmiaceae	8	7	-	-	5	8	5	5	9	6
Orthotrichaceae	7	-	-	-	10	4	6	4	10	4
Pottiaceae	11	5	9	4	14	-	7	3	9	5
Hypnaceae	6	8	10	3	-	-	-	-	4	9
Thuidiaceae	5	9	7	6	4	9	5	6	3	10
Trichostomaceae	4	10	6	7	13	3	11	1	11	2
Neckeraceae	-	-	5	10	-	-	-	-	-	-

References:

Chikovani N. *Checklist of bryophyte species of Georgia.* Braun-Blanquetia, vol. **34**, Camerino, 2004.

Lubarskaya L. B. *Bryopsida mosses of South-East Part of the Main Caucasus (Azerbaijan).* pp. 180-200, Baku, 1989 (in Russian).

Maslovski O. M. *Analysis of bryoflorotopological structure of Bryoflora complexes of Byelorussian Depression.* In: Advances and Perspectives in Bryology, L'vov, pp. 147-148, 1991 (in Russian).

Manakyan V. A. *Bryopsida of the Main Forest Massifs of Armenian Forest.* Collection of Works of the Armenian Academy of Sciences, **11**, Erevan, 1988.

Neshataev U. N. *Methods of Analysis of Geobotanical Material.* Leningrad, pp.53-98, 1984 (in Russian).

Reimers N. F. *Particularly Protected Natural Territories.* pp. 25-30, Moscow, 1978 (in Russian).

Tigishvili K. T. *Bryoflora of Lagodekhi State Reserve.* Report of the Project, Manuscript, Tbilisi, 1988 (in Georgian).

Tolmachev L. I. *Introduction to the Geography of Plants.* Leningrad, 243 p., 1974.

ღეროფოთლოვანი ხავსების ანალიზი შეხვედრილობის სიხშირის მიხედვით ლაგოდეხის ნაკრძლის ტყის სარტყელში

ტ. ციციშვილი

ნ. კეცხოველის ბოტანიკის ინსტიტუტი

(მიღებულია 08.05.2006)

რეზიუმე

ლაგოდეხის ნაკრძლის ტყის სარტყლის 125 სახეობის ღეროფოთლოვანი ხავსი, შეხვედრილობის სიხშირის მიხედვით, დაყოფილია 5 ჯგუფად. უმაღლესი შეფასება, 5 ბალი, მიიღო ხავსების საერთო რაოდენობის 6%, 4 ბალი - 19%, 3 ბალი - 28%, 2 ბალი - 30%, 1 ბალი -17%. მიღებული მონაცემებიდან ჩანს, რომ ტყის სარტყლის ბრიოფლორის ნახევარზე მეტი წარმოდგენილია საშუალო და უფრო დაბალი შეხვედრილობის სიხშირის მქონე სახეობებით. ფართოდ გავრცელებული სახეობების შეხვედრილობის სიხშირე კი ყველაზე მეტად არის შეზღუდული. აღნიშნული სარტყლის ბრიოფლორის სისტემატიკური სტრუქტურა, 10 წამყვანი ოჯახის მაგალითზე, შედარებულია აღმოსავლეთ ამერიკავკასიის ზოგიერთი რეგიონის ბრიოფლორასთან.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Ecology

THE ORIBATID MITES (ACARI, ORIBATIDA) OF GOMBORI RIDGE BEECH FOREST

ARABULI T.

Institute of Zoology, Chavchavadze ave. 31, 0179, Tbilisi, Georgia

(Received September 20, 2006)

Abstract

119 species of oribatid mites were registered in Gombori Ridge beech forests. Among them 3 species: *Phthiracarus balogi* (Feider, Suciu, 1957), *Tricheremaeus pilosus* Michael, 1988, and *Suctobelba granulate* Hammer, 1952, were recorded for the first time for Georgian fauna. According to faunal likeness distinct groupings of oribatid mites were formed, which is stipulated by distribution of beech forests at various heights. Fauna of Oribatid mites of soil of beech forests is richer than fauna of moss. Changes of number dynamics of oribatids are mutually opposed in those bitopes: while the number is high in moss, it decreases in soil, and visa versa, which is caused by temperature and moisture changes in ecosystem and migration ability of oribatids.

Introduction

Plant cover of Gombori Ridge is characterized with wide diversity and complicated floristic composition, which is caused by its ecological past. 9 zonal types are distinguished across the ridge [Sakhokia, 1960]. The basic component of Fagus forest is beech; beech forests represented by deadcovering and not evergreen subforests prevail. They occur on north exposition, forest edge at the west part of forest is relatively descended and begins from 800-900 m, and at the south exposition, where influence of dryness is higher it begins from 1000 m. On north exposition of south slopes, in humid places the beech comes to 900 m.

The number dynamics of oribatid mites in Georgia has been studied [Darejanashvili, 1965; Murvanidze, 1999; Arabuli, 2003], but number dynamics of oribatid mites inhabited simultaneously in two different stations was not studied yet. Hence, the goal of our work was to study number dynamics of oribatid mites of soil and moss of deadcovering beech forest.

Material and Methods

The material was collected during September 2001 – November 2003. Beech forests of both, south-west and north-east slopes were investigated, particularly: 1. deadcovering beech forest on Akhmeta-Tianeti pass (the utmost north-east of the ridge, near mount Shakhvetila), 2. deadcovering beech forest in Nagubrebi (on north exposition, village Tetrtsklebi), 3. beech forest with azalea subforest in central part of Gombori Ridge (Mount Tsivi, north exposition), 4. deadcovering beech forest (on north exposition of mount Tsivi), and 5. beech forest with deadcovering in Mariamjvari reserve (Table 1.).

Material was collected and worked out according to received methods of soil zoology [Krivolutski, 1973]. At each site 3 samples of soil and moss of 10 cm^3 area were taken. Extracted mites were fixed and temporary preparations were made [Balogh & Mahunka, 1983]. Mite number was counted on 1 m^2. Identification of mites was carried out by special guide-books.

The coefficient of faunal likeness was calculated by Jaccard's formula and the cluster was constructed according to the known method [Krebs, 1989] (Fig.1).

Results and Discussion

119 species united in 65 genera, 44 families and 24 upper families were registered in Gombori Ridge beech forests. Among them 3 species: *Phthiracarus balogi* (Feider, Suciu, 1957), *Tricheremaeus pilosus* Michael, 1988, and *Suctobelba granulate* Hammer, 1952, were recorded for the first time for Georgian fauna.

Our studies have shown that by species diversity of oribatid mites the soil is richer than moss. Among 119 species revealed in beech forests 100 ones were registered in soil, and 54 species – in moss. Characteristic species for every biotope were also studied; it was registered: 65 characteristic species in soil, and 10 – in moss, 44 - common for soil and moss (Table 1).

Table 1. Oribatid mites of Gombori Ridge beech forests

N	species	moss	soil					
			station	single samples				
				1	2	3	4	5
1	*Liochthonius lapponicus* (Tragardh, 1910,)		+					
2	*Hypochthonius rufulus* C.L. Koch, 1835		+					
3	*Hypochthoniella minutissima* (Berlese, 1904)		+	+				
4	*Mesoplophora pulchra* Sellnick, 1928		+		+			
5	*Epilohmannia gigantea* Berlese, 1917		+					
6	*Hoplophthiracarus vanderhammeni* Nied, 1991	+	+	+		+	+	+
7	*Phthiracarus ferrugineus* (C. L. Koch, 1841)	+	+	+		+	+	
8	*Phth. globosus* (C. L. Koch, 1841)		+				+	+
9	*Steganacarus csiszae* Balogh & Mahunka, 1979		+					
10	*St. striculus* (C. L. Koch, 1836)		+				+	
11	*St.serratus* (Feider & Suciu, 1957)		+					
12	*St. spinosus* (Sellnick, 1920)		+		+			
13	*St. (T) carinatus* (C. L. Koch, 1841)	+	+		+	+	+	+
14	*St. (T) phyllophorus* (Berlese, 1904)		+				+	
15	*St. balearicus* Perez-Inigo, 1969	+	+					
16	*St. bicarinatus* Jeleva,1970		+					
17	*Phthiracarus baloghi* (Feider, Suciu, 1957)		+					
18	*Archiphthiracarus. murphyi* (Harding, 1976)		+					
19	*A. lanatus* (Feider, Suciu, 1957)		+					
20	*A. ligneus* (Willmann, 1931)		+				+	+
21	*A.. clemens* (Aoki, 1963)		+					
22	*Rhysotritia ardua* (C.L. Koch, 1841)	+	+	+		+		+
23	*Oribotritia serrata* Feider et Suciu, 1958		+					
24	*Nothrus silvestris* Nicolet, 1855		+			+		
25	*Platynothrus grandjeani* Sitnikova, 1975		+					
26	*Camisia horrida* (Hermann, 1804)	+						
27	*Nanhermannia nana* (Nicolet, 1855)					+		
28	*Hermanniella granulata* (Nicolet, 1855)	+	+	+				

29	*H. punctulata* Berlese, 1908		+	+				
30	*Liodes theleproctus* (Hermann, 1804)	+						
31	*Arthrodamaeus femoratus* (C. L. Koch, 1840)	+	+	+				
32	*Metabelba filippova* Bul.-Zachvatkina, 1965					+		
33	*M. flagelliseta* Bulanova-Zachvatkina, 1965		+	+			+	
34	*M. pulverulenta* (C. L. Koch, 1839)	+	+			+		
35	*Metabelbella macerochaeta* Bul-Zach, 1967					+		
36	*Eupterotegeus ornatissimus* (Berlese, 1908)		+				+	
37	*Amerus troisii* (Berlese, 1883)		+					
38	*Amerobelba decedens* Berlese, 1908		+					
39	*Damaeolus ornatissimus* Csiszar, 1962	+	+				+	
40	*Eremobelba geographica* Berlese, 1908			+		+		
41	*Eremaeus hepaticus* C. L. Koch, 1836	+	+	+		+	+	
42	*E. oblongus* C. L. Koch, 1836	+	+					
43	*E. triglavensis* Tarman, 1958	+						
44	*Tricheremaeus pilosus* Michael, 1888		+					
45	*Zetorchestes microtychus* (Berlese, 1883)		+					
46	*Cultoribula bicultrata* Berlese, 1908		+					
47	*Gustavia microcephala* (Nicolet, 1855)		+					
48	*Adoristes ovatus* (C.L. Koch, 1840)		+					
49	*Liacarus brevilamellatus* Mihelcic, 1955						+	
50	*L. coracinus* (C. L. Koch, 1840)	+	+					
51	*L. tubifer* Djaparidze & Melamud, 1990		+	+		+		
52	*L. lencoranicus* Krivolutsky, 1967		+					
53	*Ceratoppia bipilis* (Hermann, 1804)	+		+				
54	*C. quadridentata* (Haller, 1882)	+						
55	*Carabodes femoralis* (Nicolet, 1855)	+	+					
56	*C. rugosior* Berlese, 1916		+			+	+	+
57	*C. procerus* Weigmann & Murvanidze 2003					+		
58	*Tectocepheus punctulatus* Djaparidze, 1985	+	+		+			
59	*T. sarekensis* (Tragardh, 1910)	+	+					
60	*T. velatus* (Michael, 1880)	+	+	+	+	+		+
61	*Berniniella bicarinata* Paoli, 1908						+	
62	*B. conjucta* (Strenzke, 1951)		+					
63	*B. exempta* (Mihelcic, 1959)		+	+				
64	*B. sigma* (Strenzke, 1951)	+	+	+				
65	*Microppia minus* (Paoli, 1908)		+					
66	*Oppiella maritima* (Willmann, 1928)		+					
67	*O. nasuta* (Moritz, 1965)		+			+		
68	*O. nova* (Oudemans, 1902)	+	+	+	+	+	+	+
69	*O. (R) hygrophila* (Mahunka, 1987)		+					
70	*O. obsoleta* (Paoli, 1908)		+					
71	*O. (R) simifallax* (Subias & Minguez, 1986)						+	
72	*O. (R) subpectinata* (Oudemans, 1900)	+	+	+	+	+	+	+
73	*O. unicarinata* (Paoli, 1908)	+						
74	*Oxyoppioides decipiens* (Paoli, 1908)	+	+					
75	*Ramusella insculpta* (Paoli, 1908)	+	+		+		+	
76	*R. mihelcici* (Perez-Inigo, 1964)			+				
77	*Quadroppia michaeli*, Mahunka, 1977	+	+		+	+		
78	*Q. quadricarinata* (Michael, 1885)	+	+					
79	*Suctobelba granulata* Hammer,1952		+		+	+		

80	*S. trigona* (Michael, 1888)	+	+					
81	*Suctobelbella acutidens* (Forsslund, 1941)	+	+					
82	*S. duplex* (Strenzke, 1950)	+	+					
83	*S. subcornigera* (Forsslund, 1941)		+				+	
84	*Banksinoma lanceolata* (Michael, 1888)					+		
85	*Cymbaeremaeus cymba* (Nicolet, 1885)	+	+			+		
86	*Eupelops acromios* (Hermann, 1804)		+					
87	*E. plicatus* (C. L. Koch, 1836)		+		+			
88	*E. torulosus* (C. L. Koch, 1840)	+		+		+		
89	*Achipteria coleoptrata* (Linne, 1746)		+					
90	*A. nitens* (Nicolet, 1855)	+						
91	*Parachipteria georgica* Murv., Weigm., 2003	+	+	+	+	+		
92	*P. punctata* (Nicolet, 1855)	+	+				+	
93	*P. nicoleti* (Berlese, 1883)						+	+
94	*Umbellozete fuscus* Krivolutsky, 1969	+	+	+		+		+
95	*Acrogalumna longipluma* (Berlese, 1904)		+					
96	*Pilogalumna tenuiclava* (Berlese, 1908)	+	+				+	+
97	*Ceratozetella sellnicki* (Rajski, 1958)	+	+					
98	*Ceratozetes gracilis* (Michael, 1884)	+	+	+	+	+	+	+
99	*C. laticuspidatus* Menke, 1964		+					
100	*C. longicuspidatus* Kulijev, 1962		+					
101	*C. mediocris* Berlese, 1908		+					
102	*Sphaerozetes piriformis* (Nicolet, 1855)	+	+					
103	*Trichoribates trimaculatus* (C.L.Koch, 1836)	+						
104	*T. caucasicus* Shaldybina, 1971	+						
105	*Chamobates caucasicus* Shaldybina, 1969			+				
106	*Ch. interpositus* Pschorn-Walcher, 1953	+						
107	*Ch. voigtsi* (Oudemans, 1902)		+					
108	*Euzetes globosus* (Nicolet, 1855)					+		
109	*Minunthozetes pseudofusiger* (Schwc, 1922)	+	+	+	+	+	+	
110	*Mycobates parmeliae* (Michael, 1884)	+						
111	*M. tridactylus* Willmann, 1929	+	+					
112	*Punctoribates punctum* (C. L. Koch, 1893)	+	+					
113	*Protoribates capucinus* (Berlese, 1908)	+	+					
114	*P. pannonicus* Willmann, 1951					+		
115	*Scheloribates laevigatus* (C. L. Koch, 1836)	+	+	+				
116	*Sch. latipes* (C. L. Koch, 1840)		+					
117	*Oribatula tibialis* (Nicolet, 1855)	+	+	+				+
118	*Phaulopi saakadzei* Djaparidze, 1985	+	+					
119	*Zygoribatula exilis* (Nicolet, 1855)	+	+		+			
	number of species	54	93	26	15	29	24	14

Beech forest with azalea subforest was distinguished by oribatid mite species diversity, where 29 species were recorded, beech forest with deadcovering (Akhmeta-Tianeti pass) – 26 species, and beech forest with deadcovering on north exposition of mount Tsivi – 24 species. It should be mentioned that beech forest with azalea subforest is rich in by species number, as well as by characteristic species (7 characteristic species were registered), which should be caused by diversity of plant cover.

In spite of studying of more or less similar ecosystems, while calculating the faunal likeness of Gombori Ridge oribatid mites, three distinct groupings have been formed (Fig. 1): the highest coefficient of likeness (35%) was noted between deadcovering beech forest located on

north exposition of mount Tsivi and Mariamjvari deadcovering beech forest, which is caused by location of those forests at the same heights. In spite of territorial distance, Akhmeta-Tianeti deadcovering beech forest and beech forest with azalea subforest of mount Tsivi located at higher regions of ridge were grouped together (34%), and beech forest of Nagubrebi placed at lower part of the ridge was absolutely isolated from them. Formation of distinct groupings should be stipulated by special sensitivity of oribatid mites to environmental conditions. As it was mentioned above beech forests on Gombori Ridge begin at various heights due to humidity, which affects the faunal composition of oribatid mites.

Number dynamics was studied in soil and moss simultaneously during 26 months. In September 2001 number of mites in moss consist of 22 500 specimens/m^2, while in soil 5159spec/m^2 were registered. In October insignificant increase of mite number was noted both, in soil and moss, but in November with temperature decrease number of specimens in moss decreased significantly, and at the expense of this their number increased up to 13 657 spec/m^2 in soil, which is caused by migration of mites from the moss to the soil at the beginning of adverse weather conditions. In December mite number was decreased in both biotopes and winter minimums were recorded.

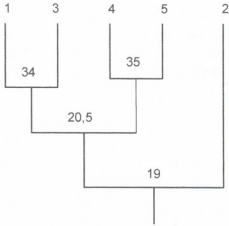


Fig.1. Cluster of faunal likeness of Oribatid mites in Fagus forest.

In January 2002 number of oribatid mites increased both, in soil and moss, and in February winter minimum (4 500 spec/m^2) was fixed in moss. In March and April together with the increase of humidity the minimal number was registered in soil. Just at the beginning of spring mite number increased in moss; increase was continued in April too, and in May maximum number was fixed – 37 000 spec/m^2. As for oribatid mites inhabited in soil, maximum number was revealed in June – 22 324 spec/m^2. In June and July, due to high temperature and dryness, mite number was decreased in moss, but in August it reached maximum – 41 500 spec/m^2.

In October 2002 the picture in soil and moss was contrary: in moss mite number minimum was recorded (4 500 spec/m^2), in soil – maximum (60 990 spec/m^2). In November situation appeared opposite: number increased up to 78 500 spec/m^2 in moss, but in soil – decreased to 16 999 spec/m^2.

In December 2002, as in 2001, number of oribatid mites decreased both in soil and moss, though mite number recorded in 2002 in soil was twice as much than that of moss.

In January 2003 the number of oribatid mites inhabited in soil increased, spring maximum (132 157 spec/m^2) was registered in March, but in moss - later, in May.

In spring 2003 soil mite number revealed considerably high, especially in July – 63 156 spec/m^2. In August and September and especially in October decrease of number was noted. As for oribatids inhabited in moss in June their number was grossly low. In the following months

intensive growth of their number was recorded and in September it reaches maximum – 337 000 spec/m^2.

Thus, as a result of our investigations it was found out that changes of number dynamics of oribatids are mutually opposed in soil and moss: while the number is high in moss, it decreases in soil, and visa versa, indicating the fact that oribatids respond rapidly to temperature and moisture changes and migrate actively towards optimal conditions.

References:

Arabuli T. *The quantiteitive dynamics of Oribatid Mites (Acari, Oribatei) in natural ecosystems (Hornbeam, (Carpinus caucasica Grossh) Forest).* Bulletin of the Georgian Academy of Sciences, **169**, 1, 169-171, 2004.

Balogh J. Mahunka S. *Primitiv Oribatids of the Palearctic region.* Akademiai Kiado. Budapest. p.372, 1983.

Darejanashvili Sh.D. *Seasonal and stational distribution of Acariformes, Oribatei in Tbilisi surroundings.* Bull.Acad.Sci.of Georgian SSR, **XXXVII**, 1, 175-179, 1965 (in Russian).

Krivolutski D.A. *Methods of complex investigation of soils on population density of Oribatid Mites.* In: Methods of Soil-Zoological Studies. Moscow, "Nauka", p. 44-48, 1973 (in Russian).

Krebs CH.J. *Ecological Methodology.* New York, Harper & Row Publisherrs, p.654, 1989.

Murvanidze M. *To the study of quantiteitive dynamics of Oribatid Mites (Acari, Oribatei) in urban conditions.* Bulletin of the Georgian Academy of Sciences, **160**, 2, 377-379, 1999.

გომბორის ქედის წიფლნარების ჯავშნიანი ტკიპები (Acari, Oribatida)

თ. არაბული

ზოოლოგიის ინსტიტუტი

(მიღებულია 20.09.2006)

რეზიუმე

გომბორის ქედზე წიფლნარების გამოკვლევის შედეგებად აღირიცხა ჯავშნიანი ტკიპების 119 სახეობა, რომელთაგან სამი: *Archiphthiracarus balogi, Tricheremaeus pilosus* და *Suctobelba granulata* პირველად იქნა რეგისტრირებული საქართველოს ფაუნისათვის. ჯავშნიანიან ტკიპებს შორის ფაუნისტური მსგავსების მიხედვით წარმოიქმნა განსხვავებული დაჯგუფებები, რაც განპირობებულია ქედზე წიფლნარების სხვადასხვა სიმაღლეზე გავრცელებით. დადგენილია, რომ წიფლნარების ნიადაგის ჯავშნიანი ტკიპების ფაუნა უფრო მდიდარია, ვიდრე ხავსის. ამ ორ ბიოტოპში ტკიპების რიცხოვნობის დინამიკა ურთიერთსაწინააღმდეგოდ იცვლება: როცა ტკიპების რიცხოვნობა მაღალია ხავსში, მაშინ მათი რაოდენობა დაბალია ნიადაგში და პირიქით, რაც ეკოსისტემაში ტენიანობის და ტემპერატურის ცვალებადობით და ორიბატიდების მიგრაციის უნარით არის განპირობებული.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Ecology

DYNAMICS OF CONDITION FACTOR OF VENDACE (*COREGONUS ALBULA* L.) IN THE LAKE PARAVANI

*JAPOSHVILI B.[1], KOKOSADZE T.[2], JAPOSHVILI O.[1]

[1] *Chavchavadze Tbilisi State Univeristy*
[2] *Institute of Zoology*
*Tel: (+ 995 99) 971827, *E-mail: bellaj99@yahoo.com*

(Received September 25, 2006)

Abstract

The dynamics of condition factor among both sexual groups of vendace has been studied for the first time. Condition factor of males and females were compared. The reason of differences is discussed. It was shown, that decrease of condition factor during last years is related with climate changes and global warming.

Key words: *Coregonus albula*, condition factor, Lake Paravani.

Introduction

Lake Paravani is the largest by its surface area among lakes of Georgia (37.5 km^2). It is situated in the Southern Part of Georgia on Javakheti upland on the 2080 m a.s.l. Volume of the lake is 90,8 mln m^3. Lake usually freezes in the second half of the December, while ice layer reaches its maximal thickness in March, very seldom it can be observed in the second half of February. In various years ice layer equaled to 47-73 cm, 80-90 cm, in very cold winter season it was even 1-1.2 m. Melting starts in the third decade of April. At the end of April or in the early May lake tends to be totally free from the ice cover [Barach, 1964, Apkhazava, 1975].

In 30s of 20th century vendace (*Coregonus albula* L.) was introduced in Paravani Lake from the Lagoda Lake (Volkhov hatchery). It was easily adapted to new environment and soon became object for commercial fishing [Demetrashvili, 1960, Japoshvili, 2002]. Data for condition factor for *Coregonus albula* is very poor and insufficient [Demetrashvili, 1960, Japoshvili, 2004, Kokhia, 1961, Peskova, 1960].

Materials and Methods

We have studied dynamics of condition factor for male and female *Coregonus albula* of Paravani Lake during 1999-2005. To calculate condition factor we have used Fulton's equation:

$K=(W/L^3)x100$

K is condition factor, W is the weight of the whole fish weight, L is total length of fish [Nikolskii, 1974, Murphy, Willis, 1996]

Age determinations were based on scales [Pravdin, 1966].

Results and Discussion

During the study males and females of vendace in Paravani Lake were represented by 4 age groups. Studies and calculations have shown that condition factor for females under the age group of 1+ reaches its peak in September. This indicator is increasing between May and September (from 0.65% to 0.81%), later on the indicator reduces and reaches the index observed in May (Fig. 1).

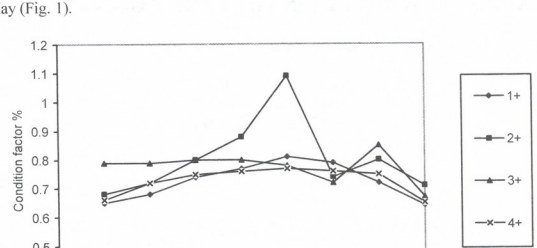


Fig. 1. Dynamics of condition factor of vendace females in Lake Paravani over the months.

In females under the age group of 2+, condition factor rises intensively and reaches its maximum in September -1.09%, while in October the curve is reduced and equals to 0.74%, however before the hatching it is risen a little bit achieving 0.80%, and falling again in December to 0.71%.

Female age group 3+ was observed in the period between May and September. They have shown condition factor of the same value approximately. It is well reflected on the curve that is almost linear. At the beginning of October coefficient equals to 0.72%, in November it reaches 0.85%, while in December it decreases to 0.67%.

In females of the age group 4+ condition factor tends to be 0.66% in May. In summer it rises to 0.77% and in December, following the hatching period the index decreases and equals to 0.65%.

Alteration of condition factor has been studied in males likewise (Fig. 2). Studies have shown that condition factor of males of age group 1+ reaches 0.58% in May, in July it rises to 0.78%, later on the value gradually falls down to 0.70% in September. In November the index increases for a while, equalling to 0.75% and decreases in December to 0.62%. In age group of 2+ coefficient is 0.60% in May. In summer period it increases dramatically and achieves to 0.84% in September. In October condition factor falls to 0.70%. At this period of time the curves are intercrossed for the age groups 1+ and 2+ in males. At the beginning of November it rises and reaches 0.78%, while in December it falls again and equals to 0.64%.

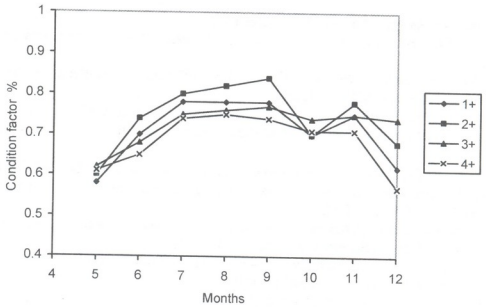


Fig. 2. Dynamics of condition factor of vendace males over the months in Lake Paravani.

Different results were found in males of the age group of 3+ and 4+. In both groups condition factor is rising from May to August. In the following months coefficient falls for the age group 4+, while 3+ age group preserves uniformity and the curve is almost linear.

Conclusions

Studies of females revealed that between May and August-September females of the age group of 1+ and 2+ are characterized with intensified diet. In addition in the age group 2+, before hatching, condition factor is decreased. It should be caused by falling of feeding rate due to the preparatory stage for hatching. This phenomenon for *Coregonus albula* is observed in other European lakes as well. In October-December species under the age group of 2+ and 3+ reflect similarity in nutrition curves caused by active involvement of those age groups in the hatching. Our studies have shown that in comparison with female species condition factor in males is less altered monthly. We suppose that condition factor is more exposed to seasonal changes due to generative synthesis in females.

We have recorded pretty low indices of the condition factor, which can be caused by several reasons, including: illegal catches intensified in the last period. As for discrepancy of our figures with previous data we suggest that differences are caused by altered terms of hatching, ice-cover formation, and warming of the lake as a result of global warming.

References:

Apkhazava I. *Lakes of Georgia.* Tbilisi, "Metsniereba", 1975.

Barach G. *Lakes of Georgia and their importance for fisheries.* Tbilisi, "Sabchota Sakartvelo", 1964.

Demetrashvili M. *For study of biology of lake Paravani vendace.* Bulletin of the Georgian Academy of sciences, **XXV**, N.1, 65-70, 1960.

Japoshvili B. *Results of visual observation on gonadogenesis of Vendace (Coregonus albula L.) in conditions of Paravani Lake.* Bull. of the Georgian Academy of sciences, **166**, 3, 591-594, 2002.

Japoshvili B. *Some Biological and morphometric characteristics of a vendace (Coregonus albul aL.) of the Lake Paravani.* Proceedings of the Georgian academy of Sciences, Biological series B, **2**, 1-2, 97-100, 2004.

Kokhia A. *Materials of diet of fishes of Paravani Lake.* Proceedings of the institute of zoology, Georgian Academy of Sciences, **XVIII,** 85-94, 1961.

Murphy B.R., Willis D. W. *Fisheries Techniques*. Second edition, American Fisheries society, Bethesda, Maryland, USA, XX, pp732, 1996.

Nikolski G. *Ecology of Fishes*. Moscow, "Visshaya Shkola", 1974.

Peskova O. Biology of vendace from Paravani and Sagamo lakes. Proceedings of scientific-research fishery station of Georgia, vol. **V**, 3-37, 1960.

Pravdin I. *Leading on fish study.* M., "Pishchevaya promishlennost", 373, 1966.

ევროპული ჭაფალას (*Coregonus albula* L.) ნაკვებობის კოეფიციენტის დინამიკა ფარავნის ტბაში

ჯაფოშვილი ბ.[1], ქოქოსაძე თ.[2], ჯაფოშვილი, ო.[1]

[1] ი. ჭავჭავაძის თბილისის სახელმწიფო უნივერსიტეტი
[2] ზოოლოგიის ინსტიტუტი

(მიღებულია 25.09.2006)

რეზიუმე

ევროპული ჭაფალას ორივე სქესის წარმომადგენლებში პირველადაა შესწავლილი ნაკვებობის კოეფიციენტის დინამიკა. შედარებულია მდედრებისა და მამრების ნაკვებობის კოეფიციენტი. განხილულია განსხვავებების მიზეზები. ნაჩვენებია, რომ ბოლო წლებში ნაკვებობის კოეფიციენტის დაცემა კლიმატურ ცვლილებებთან და გლობალურ დათბობასთანაა დაკავშირებული.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Entomology

NEW SPECIES OF MERMITHID *HEXAMERMIS DECEMLINEATAE* SP.N. (NEMATODA, MERMITHIDAE) FROM COLORADO BEETLE

GORGADZE O., LORTKIPANIDZE M., KOKHIA M., MELASHVILI N., KUCHAVA M.

Institute of Zoology

(Received October 9, 2006)

Abstract

The paper deals with the description of the new species of mermithid *H. decemlineatae* sp.n. The measurements of adult female, male and post parasitic larvae are given. Host: Colorado beetle (*Leptinotarsa decemlineata* Say). Parasitic and postparasitic mermithid larvae are revealed in the body of beetle, but adult females and males were recorded in the soil. The minimal number of mermithid larvae in each beetle was 1 specimen, and maximal - 30 specimens. In the host the number of parasitic larvae amounts to 1-3 specimens. In natural environment 47.5% of the beetles and 64.5% of larvae were infected with mermithid larvae. Nematode is localized in adipose tissue of the beetle and larvae.

Key words: parasitic and postparasitic nematodes, anatomical and morphological studies, *Hexamermis stepposis, Hexamermis angusta.*

Introduction

Colorado beetle *Leptinotarsa decemlineata Say* (Coleoptera) belongs to the Chrysomelidae family. It is harmful pest of potato. This pest was introduced from North America and was spread in most territories of Europe and Asia [Briantsev, 1966].

According to the studies carried out in biocenosis of potato sowings it was found out that those organisms (nematodes, bugs, carabus, ladybirds), which significantly reduce number of Colorado beetle, were consequently adapted on them. In this way entomopathogenic nematodes of the Mermithidae family are especially significant. Those nematodes in humid conditions can infest beetle, as well as larvae of Colorado beetle, and stimulate their death with 80-95% rate [Mishachkov, 1980]. Due to this fact mermithids appear to be perspective control agents against pests [Ipatieva, Pimenova, 1985; Rubtsov, 1978].

The goal of our work was to study nematodes of Colorado beetles distributed in some regions of East Georgia.

Materials and Methods

To study parasitic nematodes of Colorado beetle and its larvae potato sowings of private farms of mountain regions of East Georgia were researched. Places of collection were: villages Thesami, Ghulelebi, Trani, (Mtskheta-Mtianeti region).

665 specimens of the beetle and 1225 specimens of its larvae were dissected using Pavlovski method [Pavlovski, 1957]. 511 specimens of parasitic and postparasitic nematodes of one species were revealed in beetles and larvae, but the adult forms of the same species - in the soil of potato sowings. For anatomical and morphological study of collected nematodes temporary and long-term preparations were prepared [Poinar, 1975]. For nematode identification international index formulae of nematodology were used [De Man, 1884; Micoletzky, 1914]. It was established that recorded nematode belongs to the genus Hexamermis and family Mermithidae.

Genus diagnosis - Hexamermis Steiner, 1924 [Steiner, 1924].

Nematodes of this genus are of middle or big sizes (50-80 mm). Frontal part of the head of female, unlike male, is of mainly conic form. Tail end is rounded. Cuticle of the end parts of head and tail of parasitic and postparasitic larvae is thicker, than of the body middle part. Mouth opening in the frontal part of the head is placed symmetrically. Has 6 cephalic papillae; has no labial papillae; Amphids of small size. Vulva is straight and has significantly thickened stoma. Vagina is of pear-form; spicule - pair, straight and short. Has thick sexual papillae. Tail ends of parasitic and postparasitic larvae are rounded.

Typical species: *Hexamermis angusta* Rubzov, 1971 [Rubtsov, 1971].

Results and Discussion

Host: Colorado beetle (*Leptinotarsa decemlineata* Say).

Localization: in adipose tissue of the beetle and larvae.

The apical part of head of female nermithid is speculated, but of male – rounded. (Fig. 1 A, C). Neck gland is seen under cuticle. There are not protrudent tubers on the head. Amphids are small (4-6 μm) and oval. Their ducts are opened a bit lower of cephalic papillae. Cuticle oesophagus is not spread up to the mouth opening. Width of mouths opening walls is of 3-5 μm.

Female: n=7; L=42.3 (32.0-62.5) mm; a = 172.1 (155.3-203.6); V (%) = 55 (54-57);

Body width: near cephalic papillae consists of 70 (53-115) μm, near nerve ring – 162 (92-222) μm, near vulva – 250 (157-380) μm and near the end of trophosome – 158 (120-277) μm. Distance from the frontal part of the head to nerve ring is 269 (179-335) μm; up to vulva – 25.5 (17.3-30.8) mm; from the end of trophosome to the tail end – 168 (75-280) μm. Structure of vagina is not distinguished from that of species described by Rubtsov [Rubtsov, 1971]. Width of cuticle near mouth opening is 21 (19-32) μm, near vulva – 14 (9-18) μm, at the tail end – 25 (24-33) μm.

Male: n=23.5 (19.3-38.6) mm; a=135.1 (105-183.7); c=130.6 (83.8-159.4).

Body width: near cephalic papillae consists of 70 (56-93) μm; near nerve ring 115 (80-193) μm; at anus –155 (120-240) μm; the widest part of body - 198(153-322) μm. Distance from the frontal part of head to nerve ring is 305 (240-396) μm. Male has weakly bent pair spicule (Fig. 1. D), which length is 161 (103-250) μm, diameter – 20 (15-36) μm, but its end is acute. Cuticle width at head opening in the front part of the body is 18 (14-23) μm, in the middle part – 13 (10-16) μm and near tail - 13 (6-30) μm. Tail length is 200 (150-304) μm.

Postparasitic larvae

Female: n=1; L=25.3 mm.

Body width: at cephalic papillae – 98 μm, at nerve ring – 170 μm, at vagina – 335 μm, at the end of trophosome – 225 μm. Distance from the apical part of the head to nerve ring consists of 350 μm, from the end of trophosome to the end of tail – 345 μm. Distance from front of the head to vagina equals to 20.2 mm. Cuticle width is: near head opening in the front of the head – 41 μm, at vagina in the middle part of the body – 13 μm, and at the tail – 102 μm.

Differential diagnosis

By anatomical and morphological characteristics species described above resembles species *Hexamermis stepposis* Artyukhovsky et Khartschenko, (1965) [Artyukhovsky, Khartschenko, 1965], but is more similar to the species *Hexamermis angusta* Rubzov [Rubtsov, 1971], from which it is distinguished by the form of vulva lips, by form and size of amphids.

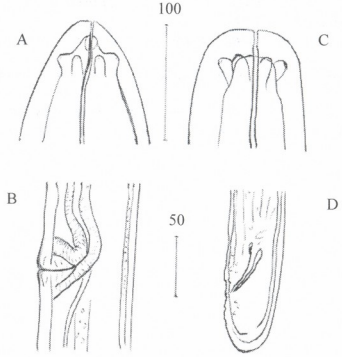


Fig. 1. Hexamermis decemlineatae sp. n
Female: A – frontal end of the body; B – vulva segment
Male: C – frontal end of the body; D – tail segment with spicule.

According to anatomical-morphological features *Hexamermis decemlineatae* sp.n. is considered as a new species for Georgia.

References:

Anderson R.S. *Methode pour I examen des nematodes en vue apicale.* Ann. Parasitol. Hum. Comp., **33**, 171-172, 1958.

Artiukhovski A.K., Kharchenko N.A. *Study of mermithids of Streletskaya Steppe.* Proceedings of Central Black Sea Reservoir, M., p.9, 1965 (in Russian).

Briantsev B.A. *Agricultural entomology.* L., "Kolos", pp.342, 1966 (in Rusian).

De Man J. G.. *Die frei in d. reinen Erde u. in sussen Wasser lebenden nematoden d. niederl. fauna Leiden.* p. 313-321, 1884.

Ipatieva G.V., Pimenova I.N. *Mermithids of the Altai Mountains.* Publ. Saratov University, p.1-149, 1985 (in Russian).

Micoletzky H. *Freilebende Subwasser-Nematoden der Ost- Alpen mit besonderer Beruckichtigung des Lunzer Seengebietes.* Zool. Jahrb. Abt., **1**, 36, 331-546, 1914.
Mishachkov G.A. *Biology and ecology of mermithid Hexamermis albicans - parasite of Colorado beetle.* In: Helminths of insect. M., "Nauka", 85-89, 1980 (in Russian).
Pavlovski E.N. *Methods of hand-anatomy of insect.* M., "Nauka", p.1-45, 1957 (in Russian).
Poinar G.O.jr. *Entomogenous nematodes. A manual and host list of insect – nematode associations.* Leiden E. J. Brill., p. 1-317, 1975.
Rubtsov I.A. *New species of Mermithidae from southern seaside.* Zool. J., **50**, 8, 1143-1152, 1971 (in Russian).
Rubtsov I.A. *Mermithids. Classification, significance, usage.* L., "Nauka", p.207, 1978 (in Russian).
Steiner G. *Beitrage zue Kenntnis der Mermithiden, 2. Mermithiden aus Paraguay in der Sammlung des Zoologischen Museum zu Berlin.* Zbl. Bacteriol., Parasitenkunde. **62**, 1-6.1924.

ახალი სახეობის ნემატოდა *Hexamermis decemlineatae* sp. n. (Nematoda, Mermithidae) კოლორადოს ხოჭოდან

გორგაძე ო., ლორთქიფანიძე მ., კოხია მ., მელაშვილი ნ., კუჭავა მ.

ზოოლოგიის ინსტიტუტი

(მიღებულია 09.10.2006)

რეზიუმე

აღწერილია მერმიტიდას *Hexamermis decemlineatae* sp.n. ახალი სახეობა. მოცემულია ზრდასრული ინდივიდების და პოსტპარაზიტული ლარვების განაზომები. მასპინძელი: კოლორადოს ხოჭო (*L. decemlineata* Say). ლოკალიზაცია: ხოჭოსა და ლარვების ცხიმოვანი ქსოვილი. ხოჭოს სხეულში გამოვლენილია მერმიტიდას პარაზიტული და პოსტპარაზიტული ლარვული ფორმები, ხოლო ზრდასრული ეგზემპლარები გამოვლენილია ნიადაგში. მერმიტიდების ლარვების მინიმალური რაოდენობა თითო ხოჭოში შეადგენდა 1 ეგზემპლარს, ხოლო მაქსიმალური - 30. მასპინძელში პარაზიტული ლარვების რაოდენობა აღწევდა 1-3 ეგზემპლარს. ბუნებრივ პირობებში მერმიტიდების ლარვების მიერ დაინვაზირებულია ხოჭოების 47,5%, ხოლო ლარვების 64,5%.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Genetics

GENE DRIFT IN THE MIRZAANI POPULATION OF WINE YEAST

SHATIRISHVILI A.[1], MENABDE M.[2], SADAGISHVILI T.[2], ZARNADZE T.[3]

[1]*I. Chavchavadze Tbilisi State University*
[2]*Iv. Javakhishvili Tbilisi State University*
[3]*A. Tsereteli Kutaisi State University*

(Received March 1, 2006)

Abstract

During 1996 – 2004 the antagonistic activity was studied in the Mirzaani (Kakheti) population of wine yeast. The population was found to be polymorphic by the feature. Three phenotypic classes were identified in the population: killer (K), neutral (N) and sensitive (S) classes. Gene drift resulting in periodic fluctuation of the phenotypic classes was revealed.

Key words: yeast population, antagonistic activity, killer-system.

Introduction

Among other factors affecting the gene pool of population the gene drift is of particular importance. As a result of its activity allele concentrations change in the gene pool. This process is especially intensified in the populations with great decrease in the number of members [Hedrick, 1999; Shatirishvili, 2002]. Such periodic (cyclic) number fluctuation in the wine yeast is due to abiotic, biotic or anthropogenic factors. In the regions of well-developed domestic wine production of Georgia fermentation of wine occurs spontaneously. The process is first prompted by a small group of yeasts, i.e. "the founder's principle" acts [Shatirishvili, 2002; Mayr, 1970]. Small numbers of yeasts give rise to tens of millions of new ones. The yeast gets into the grape juice from the philosphere, ripened grape berries and wine cellar implements, while the stream depends on the Drosophila [Ribero-Gasion et al., 1980].

Materials and Methods

In Saccharomyces, and particularly in the wine yeast, no traits identifying the population have been worked out yet. Its reproductive area depends on the Drosophila. Therefore, it occupies about 400-500 meters [Sadagishvili et al., 2001; Menabde et al., 2004]. Proceeding from that we considered the forms of wine yeast spread over village Mirzaani and nearby regions to be a population. The material (sediment) was taken from 10 different remote districts by the method described before [Menabde et al., 2004].

The antagonistic activity of strains was detected by means of the testing strains: K7 {KIL – K1}; S14 (sensitive to the system K1); Oxford genetic lines; the line M437 {KIL-K2} created at the Institute of Genetics of Russian Academy of Sciences; the line RA p192 (sensitive to the system K2), a Petergof genetic line. Special culture media were applied for studying the

antagonistic activity of the strains. The specificity of discovery of the killer systems was described before [Shatirishvili et al., 2001].

Results and Discussion

The alcoholic fermentation of grape juice represents a complex multi-stage process. Some microorganisms are involved in it and the yeast fungi join the process at the final stage. During the fermentation inter- and intra-specific competitions take place [Shatirishvili et al., 2001]. The inter-strain antagonism occurs in the wine yeast, that becomes apparent when the cells of sensitive strains are eliminated under the effect of a toxin released by another strain [Menabde et al., 2004].

In order to study the variability of gene frequencies in natural populations of wine yeast, in 1996-2004 we investigated the Mirzaani (Kakheti) population. With the interval of 2-3 years the strains were repeatedly isolated from 10 micropopulations (500 cultures in total) and analyzed genetically. The antagonistic activity was defined by introducing strokes of the strains under study to the nutrient media with a special loop. If the strain contains a killer system, the latter causes a lysis of sensitive test-cultures forming a sterile ring around the developed culture. If a strain is sensitive to the test culture the area of eliminated cells gets stained in dark blue color by Methylen – blue stain [Shatirishvili et al., 2001]. The strain that has a killer-system produces and releases the protein – a toxin that induces elimination of sensitive strain cells. The strains with neutral phenotype are sensitive to the toxin. The results obtained for the strains in 1996 are given as patterns in Tables 1 and 2. The other results have already been published [Sadagishvili et al., 2001; Menabde et al., 2004; Shatirishvili et al., 2001].

The strains constituting the population are arranged in three phenotype classes: killer (K), neutral (N) and sensitive (S) strains. The whole population as well as each micro population appeared to be polymorphic. When compared the structures of different populations studied in different years, we found that in 1998 and 2000 the rates of killer strains reached maximum (17,3% and 15,8 % respectively), while in 2004 the content of killer strains was minimal – 1,4% (see Fig. 1).

Table 1. Determination of antagonistic activity of Mirzaani "Rkatsiteli" population

Phenotype	Number of classes	Test-strains			
		M437	7A – P192	K7	S14
I	1	K	K	K	N
II	7	K	K	N	N
III	4	K	N	K	K
IV	2	K	N	K	N
V	9	N	K	K	N
VI	11	N	N	K	K
VII	3	N	N	K	N
VII	6	N	N	K	K
IX	451	N	N	N	N
X	2	N	N	N	S
XI	2	N	N	S	N
XII	1	S	N	N	S
XII	1	N	S	S	S

Changes in environmental conditions sharply affect the ratio of the strains with K, N and S phenotypes and cause variations in natural populations of the yeast. The reason for that is number fluctuation. Under the severe climate conditions in winter and spring the quantity of the yeast

dramatically decreases causing so called "bottleneck" effect. Thus, it comes the period when the number of strains in population and the density of the population become minimal [Shatirishvili, 2002].

In autumn (in vintage) in the sites of domestic wine production, where the wine fermentation is of spontaneous character, the number and density of the yeast within the population sharply rise. Thus, after passing the "bottleneck" the quantity of population members in the natural population reaches maximum. The small groups with occasionally survived yeasts give rise to the yeast population that means that "the founder's principle" works.

Table 2. Determination of the frequencies of phenotypes K, N and S in Mirzaani micropopulations

Micro-population	Number of analyzed strains	Killer		Neutral		Sensitive	
		Number	%	Number	%	Number	%
I	50	3	6	47	94	-	-
II	50	2	4	48	96	-	-
III	50	5	10	43	86	2	4
IV	50	-	-	50	100	-	-
V	50	1	2	48	96	1	2
VI	50	4	8	46	92	-	-
VII	50	5	10	43	86	2	4
VII	50	6	12	43	86	1	2
IX	50	12	24	38	76	-	-
X	50	5	10	45	90	-	-
Total	500	43	8.6	451	90.2	6	12

References:

Hedrick P. W. *Genetics of population*. Sones & Barlett Publishers, 1999.

Mayr E. *Populations, species and evolution.* Harvard university press, 1970.

Menabde M., Sadagishvili T., Kacharava M., Shatirishvili A. *Antagonistic activity in endemic population of wine yeast (Saccharomyces vini)*. Proc. Georgian Acad. Sci., Biol. Ser. B, **2**, 5-6, 57-60, 2004.

Ribero-Gasion J., Reino E., Ribero-Gasion P. *Theory and practice of wine-making*. v.3, M., 1980

Sadagishvili T., Zarnadze T., Menabde M., Shatirishvili A. *Killer systems of wine yeasts*. Proc.Georgian Acad.Sci., Biol.Ser. **27**, 1-3, 225-229, 2001.

Shatirishvili A., Sadagishvili T., Zarnadze T., Menabde M. *Antaginistic activity of natural saccharomyces*. Georgian Med. News, **70**, 1, 48-51, 2001.

Shatirishvili A. *Genetic mechanisms of evolution in natural population of wine yeasts.* In: Biology and contemporaneity, Tbilisi, p. 254-260, 2002.

ბენთა დრეიფის მოქმედება ღვინის საფუარის მირზაანის პოპულაციაში

შათირიშვილი ა.[1], მენაბდე მ.[2], სადაგიშვილი თ.[2], ზარნაძე თ.[3]

[1]ი. ჭავჭავაძის თბილისის სახელმწიფო უნივერსიტეტი
[2]ივ. ჯავახიშვილის თბილისის სახელმწიფო უნივერსიტეტი
[3]ა. წერეთლის ქუთაისის სახელმწიფო უნივერსიტეტი

(მიღებულია 01.03.2006)

რეზიუმე

შესწავლილია ანტაგონისტური აქტივობა ღვინის საფუარის მირზაანის პოპულაციაში. პოპულაციის სტრუქტურა პოლიმორფული აღმოჩნდა. იგი სამი ფენოტიპური კლასითაა წარმოდგენილი: კილერი (K), ნეიტრალური (N), მგრძნობიარე (S). მიკროევოლუციის მამოძრავებელი ფაქტორის ხემოქმედების შედეგად ფენოტიპური კლასები პერიოდულად ფლუქტუირებს.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Immunology

DISTRIBUTION FEATURES OF SOME ERYTHROCYTIC GROUP-SPECIFIC ANTIGENS SIGNIFICANT FOR CLINICAL MEDICINE IN ADJARA REGION

*NAGERVADZE M., DIASAMIDZE A., AKHVLEDIANI L., DUMBADZE G., KHUKHUNAISHVILI R., KORIDZE M.

Sh. Rustaveli Batumi State University
**E-mail: mari-na@nm.ru*

(Received September 20, 2006)

Abstract

Erythrocytic group antigens interesting in the viewpoint of transfusion, such as *c, K, D^u*, were studied in Adjara population. Those antigens are characterized with high immunogenicity but their screening is not conducted. It was shown that distribution frequency of *c* antigen in Adjara population is 90.2±3.03%. 4.2% of population is carrier of *K* factor. Distribution frequency of D^u antigen is 0.625±0.12%. Number of *CC* genotype carriers within the population appeared to be 8.0±2.7%.

Key words: immunoserological methods, *ABO* system, *Rh* system, *Kell* system.

Introduction

Erythrocytic group antigens stipulating for blood compatibility and appearing as the main reason of posttransfusion complication are significant for clinical medicine [Anstee et al., 1999]. Significance of those group antigens is associated with immune characteristics of the living organism. They take important role in transfusiology [Schonewille et al., 2006], epidemiology [Vojvodic, 2000] and transplantology [Bucin, 2006], in human genetics [Shubin, 1997], and especially in the studies of population genetics [Kucher, 2000]. Study of erythrocytic group systems is also important in ethnic anthropology [Shneider et al., 2002]. Heredity of those systems is so stable that their study for establishment of some ethnic group origin gives accurate data [Schmidt et al., 2003].

Today in Adjara two antigens (*A, B*) of *ABO* system and *D* antigen of *Rh* system are taken into account during blood transfusion. For the individuals where those antigens do not occur the theoretical risk of alloimmunosensitization is high [Judd et al., 1992]. In the viewpoint of transfusion *c* antigen, among rhesus system antigens, is also significant. Numerous data about alloimmunosensitization caused by this antigen are presented in scientific literature [Regan et al., 1997]. Distribution frequency of *c* antigen within world population is 80-82%. 18-20% of humans do not have this antigen and are revealed in *CC* state. Individuals with just this genotype belong to high-risk group of alloimmunosensitization.

Immune activity of *K* antigen is slightly minor than rhesus antigen (*RhD*) activity. Immunosensitization caused by *K* antigen is a frequent case [Donskov, 1996], which is certified by the numerous data of posttransfusion complications described in literature.

Today it is necessary to carry out screening of donors by *K* antigen. At present *Kell*-positive blood is not used in transfusion in many countries.

The majority of mistakes during rhesus system determination are related with weak variation of *D* antigen – D^u. Unlike *D* antigen, D^u antigen has latent antigen determinants. For their discovery, first, it is necessary to fix those determinants on the surface of erythrocytes, and further, to reveal them. Such study is carried out in all those cases when *Cde, cdE* phenotypes are revealed during the primary phenotyping of erythrocytes. Complex methods enable to delete individuals having CD^ue and cD^uE phenotypes from rhesus-negative donors.

Unfortunately, proceeding from high transfusion significance of abovementioned antigens, their screening is not conducted today.

The aim of our work was to study regularities of distribution of erythrocytic group antigens in Adjara region and to forecast theoretically expected alloimmunosensitization.

Materials and Methods

The study was carried out by immunoserological methods. Test-systems having anti -*c*, -*K* specificities, anti –*D* incomplete antibody, antiglobulin serum, standard group erythrocytes and standard serums, were used.

512 individuals of Adjara population were studied.

The obtained data-processing was carried out using statistical methods.

Results and Discussion

In the majority of studied individuals *c* antigen was registered (90.2±3.03%). Distribution frequency of *C* antigen was 53.0±5.3% [Nagervadze et al., 2006] (Fig.1). According to the research of allele concentrations it was revealed that concentration of *c* allele is high and equals to 0.74 (Fig. 2).

Forecasting of theoretically expected immunosensitization caused by *c* antigen was carried out. Carriers of *CC* genotype were separated out. Distribution frequency of *CC* genotype in Adjara population is equaled to 8.0±2.7% (*Cc* – 54.0±4.9 and *cc* – 38.0±4.8), implying that carriers of this genotype do not consist in *c* antigen and during transfusion in 92% cases incompatibility should be revealed (Fig. 3).

With low frequency, but nevertheless, D^u antigen was recorded in Adjara population. Distribution frequency of D^u antigen is 0.625±0.12% (Fig. 4).

Donors having D^u antigen should be belonged to rhesus-positive, but recipients – to rhesus-negative group and rhesus-negative blood should be transfused to them, because normal *D* antigen may cause immune response therein.

As a result of our studies it was revealed that 4.2% of Adjara population is carrier of *K* factor (Fig. 5).

Thus, it was established that distribution frequency of *c* antigen is high in Adjara population, and respectively theoretical risk of immunosensitization caused by this antigen is high. It is necessary to provide medical laboratories with information that erythrocyte screening of donor-recipients upon such antigens, as *c*, C^w, D^u, *K*, must be carried out. Such approach should

decrease cases of posttransfusion complications and the risk of immunosensitization should be brought to minimum.

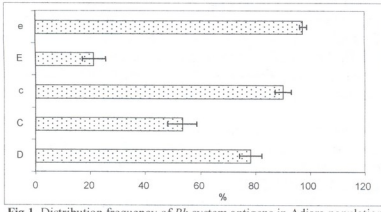


Fig.1. Distribution frequency of *Rh* system antigens in Adjara population

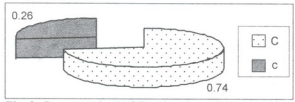


Fig. 2. Concentrations of *C* and *c* alleles in Adjara population

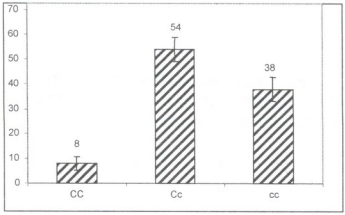


Fig. 3. Analysis of *Cc, CC* and *cc* genotypes in Adjara population

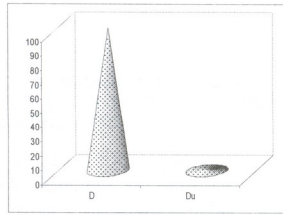


Fig. 4. Distribution frequency of *D* and *D*^{u} antigens in Adjara population

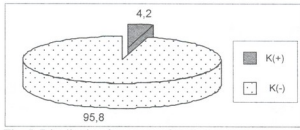


Fig. 5. Distribution frequency of *Kell* system phenotypes in Adjara population.

References:

Anstee D.J., Cartron J.P. *Toward an understanding of the red cell surface//Red cell immunohaemato logy towards its second century.* ESTM. TelAviv, p.12-44, 1999.

Bucin D., Johansson S., Malm T., Jogi P., Johansson J., Westrin P., et al. *Heart transplantation across the antibodies against HLA and ABO.* Transpl Int., **19**, 3, 239-44, 2006.

Kucher AN., Puzyrev VP., Chernetsov DB., Erdynieva LS., Sanchat NO. *Polymorphism of immunological and biochemical marker genes in rural populations of the Tuva Republic.* Genetika, **36**, 4, 562-9, 2000.

Nagervadze M., Akhvlediani L., Dumbadze G., Bagrationi K., Diasamidze A. *Distribution of Rh-Hr antigens in Adjara population.* Proc. Georgian Acad. Sci., Biol.Ser. B, **4**, 1, 48-51, 2006.

Schmidt HD, Scheil HG. *Blood group frequencies in Romania: microregional and ethnic differences.* Anthropol Anz., **61**, 4, 381-93, 2003.

Shneider Iu.V., Shilnikova I.N., Zhukova O.V. *Materials for studying the gene pools of Russia and neighboring countries: the Russian population of the Pskov region.* Genetika, **38**, 11, 1561-5, 2002.

Shubin PN., Morokov VA., Efimtseva EA., Chelpanova TI. *Ethnogenetics of the Komi-Zyrians according to data on the distribution of gene frequencies of erythrocyte and serum systems in the blood.* Genetika, **33**, 2, 235-42, 1997.

Vojvodic S. *Inhibitory activity of blood group antigens M and N in inhibition of virus hemagglutination reactions of influenza viruses.* Med Pregl., **53,** 1-2, 7-14, 2000.

კლინიკური მედიცინისათვის მნიშვნელოვანი ზოგიერთი ერითროციტური ჯგუფსპეციფიკური ანტიგენების გავრცელების თავისებურებანი აჭარის რეგიონში

ნაგერვაძე მ., დიასამიძე ა., ახვლედიანი ლ., დუმბაძე გ., ხუხუნაიშვილი რ., ქორიძე მ.

ჭ. რუსთაველის ბათუმის სახელმწიფო უნივერსიტეტი

(მიღებულია 20.09.2006)

რეზიუმე

აჭარის მკვიდრ მოსახლეობაში შესწავლილია ტრანსფუზიური თვალსაზრისით საინტერესო ერითროციტური ჯგუფური ანტიგენები, როგორიცაა *c, K, D^u*. აღნიშნული ანტიგენები საკმაოდ მაღალი იმუნოგენურობით ხასიათდებიან, მაგრამ არ ხდება მათი სკრინინგი. ნანახებია, რომ აჭარის მოსახლეობაში *c* ანტიგენის გავრცელების სიხშირე 90.20±3.03%-ია. აჭარის მოსახლეობის 4.2% ფაქტორის მტარებელია. *D^u* ანტიგენის გავრცელების სიხშირე – 0.625±0.12%-ია. გამოყოფილი იქნა *CC* გენოტიპების მტარებელი პირები; მათი გავრცელების სიხშირე 8.0 2.7%-ია.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

DETERMINATION OF ANTIMICROBIAL ACTIVITY OF NITROGEN FIXING BACTERIA

BAGALISHVILI M., BASILASHVILI L., KIKVIDZE M., GURIELIDZE M., NUTSUBIDZE N.

S. Durmishidze Institute of Biochemistry and Biotechnology

(Received October 6, 2006)

Abstract

Antagonistic features of rhizosphere of nitrogen-fixing microorganisms, which they reveal towards test-organisms, including phytopatogenic fungi and actinomyces have been studied. Most of investigated nitrogenfixers (28 cultures out of 30) display antimicrobial activity in different degree. Antimicrobial activity of rhizospere nitrogenfixing microorganisms together with other positive characteristics (fixation of molecular nitrogen, production of growth stimulators) enable to use these microorganisms in agriculture on the purpose of soil remediation and enhancing of productivity.

Key words: phytopatogenic fungi, actinomyces, nitrogen-fixing microorganisms

Introduction

Microorganisms are never found in isolated state in natural conditions. They can be obtained as pure cultures only artificially, but in nature, they represented in association, where different interactions are established between microbes. One of the most wide spread forms of interaction is antagonism - when one strain of organisms suppresses development of other organisms. Antagonistic properties of microorganisms are mostly manifested by formation of antimicrobial substances [Crawford, 1988].

28 pure cultures of nitrogen fixers were isolated from rhizosphere of cereals. These rhizospherous microorganisms use the energy of photoassimilates isolated by plant roots for the energy – consuming nitrogen fixation processes. It is supposed that these organisms obtain other advantages that make them competitive among various microbes around the plant roots [Shah et al., 1992].

That is the reason for study whether antimicrobial substances are isolated from rhizosphere of nitrogen fixing microorganisms and what the model is of action of these substances on other microbes.

Materials and Methods

30 nitrogen-fixing microorganisms were studied in all. As test-organisms were used actinomycetes: *Streptomices* 82; *Streptomices fradiae* 110, Phytopathogenic fungi: *Fusarium solani*, *Rhizoctonia* sp; yeasts: *Saccharomyces fragilis*, *Candida utilis*; gram-positive bacteria: *E.coli*, *Ps. fluorescens* (Table 1).

Antimicrobial activity of isolated by us and collectional nitrogen fixing microorganisms was studied by the method of agar blocks [Egorov, 1965] as follows: on Petri dishes with corresponding agar nutrient medium, the studied microorganisms were platted in a form of a lawn. After good development of a cultures and formation of antimicrobial substance, which is diffused in agar, we cut 10 mm diameter agar blocks with a special sterile lancet and replace them on other Petri dishes with previously platted test-organisms on a nutrient medium. After 18-20 h of incubation at the optimal for test-organisms temperature light zones were formed around agar blocks, indicating suppression of development of test-organisms. The results were registered 36- 48 h after incubation. According to sterile zones diameter around agar blocks we could judge about antimicrobial activity of microorganisms.

Results and Discussion

28 nitrogen fixing microorganisms, among studied by us 30 ones, reveal antimicrobial activity against 9 out of 11 test-organisms. The widest spectrum was characteristic for *A. brasilense* Г3 and *A. brasilense* G3 (7 test-organisms). Many microorganisms were found to have antimicrobial activity to *Fusarium solani* (19 microorganisms) and *Streptomyces fradiae* (17 microorganisms). The highest antimicrobial activity of microorganisms was manifested to *Fusarium solani*: G-41 - 19 mm, G111 - 19 mm and G71 - 18 mm (Table 1).

Table 1. Antimicrobial activity of nitrogen fixing microorganisms

Nitrogen fixing microorganisms	test-organisms (size of zones, mm)										
	Strept. fradiae 82	*Stept. fradiae 110*	*Fusarium solani*	*Rhizoctonia sp.*	*Sacch. fragilis*	*Candida utilis*	*Myc. phlei*	*Rhodococcus sp.*	*Staph. aureus*	*E. coli*	*Ps.fluorescens*
A.bras. ATCC 9825	11	12	-	14	-	-	13	11	14	-	-
A.brasilense Г3	11	14	13	14	-	15	12	16	-	-	-
A.brasilense G1	11	12	-	-	-	-	11	-	14	-	-
A.brasilense G2	12	13	-	11	-	13	12	-	-	-	-
A.brasilense G3	13	12	12	15	-	-	12	13	14	-	-
A.brasilense G4	12	-	11	15	-	11	13	-	-	-	-
A.brasilense G5	13	12	13	-	12	-	12	-	-	-	-
A.brasilense G12	11	-	11	-	-	-	-	-	-	-	-
A.brasilense G16	-	-	-	-	-	-	-	13	-	-	-
A.brasilense G20	-	-	12	-	-	11	-	-	-	-	-
G22	-	-	-	-	-	-	-	-	-	-	-
G23	-	12	-	-	-	-	13	11	-	-	-
G24	-	13	-	-	-	-	-	-	11	-	-
G26	11	16	17	-	11	-	-	-	-	-	-
G41	-	14	19	-	-	-	13	14	12	-	-
G43	-	15	14	-	-	-	-	-	-	-	-
G44	-	14	-	-	-	-	-	-	-	-	-

G45	-	13	11	11	-	-	-	-	-	-	-
G61	-	-	13	15	13	13	-	15	17	-	-
G62	-	-	12	-	12	-	-	-	11	-	-
G64	-	-	12	-	-	-	-	-	-	-	-
G66	-	12	13	-	-	-	-	14	-	-	-
G68	-	12	-	-	11	-	11	11	13	-	-
G70	-	-	14	-	11	11	-	-	-	-	-
G71	-	-	18	12	-	11	-	-	-	-	-
G72	-	-	12	-	11	-	-	-	-	-	-
G110	-	-	-	-	-	-	-	-	-	-	-
G111	-	11	19	-	-	-	-	-	-	-	-
G120	-	11	18	-	11	-	-	-	-	-	-
G121	-	-	17	-	11	-	-	-	11	-	-

Fusarium solani – is phytopathogenic fungi, it is characterized by high capability to infection and preserves in soil for a long time, as it suppresses development of useful microorganisms and simplifies the process of penetration into a plant tissue [Meyer et al 1998]. Issuing from the said above the fact that nitrogen fixing microorganisms and among them *Azospirillum* excrete antimicrobial substances towards such strong phytopathogenic organisms is very interesting (Fig.1, 2).

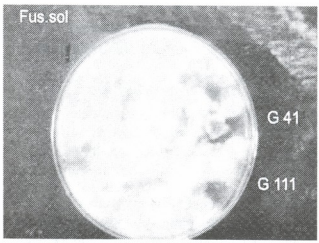


Fig. 1. Antagonism of nitrogen-fixing strains G41 and G111 towards *Fusarium solani*

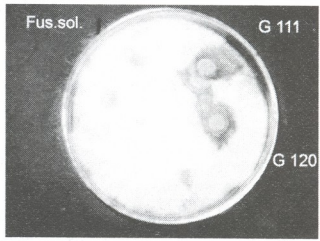


Fig. 2. Antagonism of nitrogen-fixing strains G111 and G120 towards *Fusarium solani*

Thus, most of the studied nitrogen fixers (28 cultures from 30) obtain antimicrobial activity to 9 from 11 test-organisms that gives additional advantages to rhizosphere to develop in microflora. Issuing from the said above introduction of active strains of nitrogen fixers into rhizosphere of agricultural cultures in our opinion will have positive effect on their growth and development.

References:

Egorov N. S. *Microbes – antagonists and biological methods of determination of antibiotic activity.* M., "Visshaya shkola", 1965.

Crawford D.L. *Development of recombinant Streptomyces for biotechnological and environmental uses.* Adv. Biotechnol., **6**, 183-206, 1988.

Meyer L., Weher F.C., Nel & Carling D.F. Characterization of the crater disease strain of *Rhizoctonia solani.* J. Phytopathology, **88**, 4, 366-371, 1998.

Shah S., Karkhanis V., Desai A. *Isolation and characterization of siderophore, with antimicrobial activity, from Azospirillum lipoferum M.* Curr. Microbiol., **25**, 34-35, 1992.

აზოტმაფიქსირებელი ბაქტერიების ანტიმიკრობული აქტივობის განსაზღვრა

ბაგალიშვილი მ., ბასილაშვილი ლ., კიკვიძე მ., გურიელიძე მ., ნუცუბიძე ნ.

ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 06.10.2006)

რეზიუმე

შესწავლილია რიზოსფერული აზოტმაფიქსირებელი მიკროორგანიზმების ანტაგონისტური თვისებები, რომლებსაც ისინი ავლენენ სხვადასხვა ტესტ-ორგანიზმების, მათ შორის ფიტოპათოგენური სოკოებისა და აქტინომიცეტების მიმართ. გამოკვლეულ აზოტფიქსატორთა უმრავლესობამ (30-დან 28 კულტურამ) გამოავლინა ანტიმიკრობული აქტივობა მეტ-ნაკლები ხარისხით. რიზოსფერული აზოტმაფიქსირებელი მიკროორგანიზმების აქტივობა სხვა დადებით მახასიათებლებთან ერთად (მოლეკულური აზოტის ფიქსაცია, ზრდის სტიმულატორების პროდუქციის უნარი) საშუალებას იძლევა ეს მიკროორგანიზმები გამოყენებულ იქნას სოფლის მეურნეობაში ნიადაგების გაჯანსაღებისა და პროდუქტიულობის გაზრდის მიზნით.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

THERMOPHILIC MICROSCOPIC FUNGI OF SOILS OF EAST GEORGIA

*DAUSHVILI L., BURDULI T., KUTATELADZE L., JOBAVA M., DZALAMIDZE I.

S. Durmishidze Institute of Biochemistry and Biotechnology. David Agmashenebeli Alley 10km, 0059 Tbilisi, Georgia
**E-mail: lela_d@mail.ru*

(Received August 14, 2006)

Abstract

70 cultures of microscopic fungi, isolated from dry subtropical-steppe zone of Sighnaghi region (east Georgia) have been investigated. 5 thermophilic, 5 thermotolerant and 4 psychrophilic (facultative) strains of micromycetes were revealed. The optimal ranges of growth and spore-formation of thermophilic micromycetes have been established.

Key words: thermophilic, thermotolerant, psychrophilic, *Aspergillus*.

Introduction

Thermophilic microorganisms, in particular thermophilic fungi are the subjects of intensive investigation. This interest is determined by preferred application of thermophilic fungi and their enzymes in different fields of industry, agriculture and medicine, compared with mesophilic microorganisms and their enzymes [Bilay, 1979].

Thermophilic microscopic fungi are potential sources of various industrially important thermostable enzymes, such as lipases, xylanases, proteases, amylases and pectinases. These enzymes have numerous applications in the detergent, starch, paper, food and pharmaceutical industries [Phutela et al., 2005]. Due to their increased thermostability, enzymes of thermophilic micromycetes are potentially useful in the starch industry for production of maltose and glucose. Thermostable amylase are generally preferred as their application minimizes contamination risk and reduces reaction time, thus providing considerable energy saving. Hydrolysis carried out at higher temperatures also minimizes polymerization of D-glucose to iso-maltose [Kunamneni et al., 2005]. Thermophilic fungi have a strong capacity to degrade polysaccharide constituents in plants, therefore having potential for biotechnological applications such as bioconversion of plant biomass into animal feed, plant fertilizers and chemicals for the food industry [De Faria et al., 2004].

The goal of the study was to reveal the extremophilic by temperature (thermopiles, thermotolerants and psychrophiles) strains of microscopic fungi among the collection of cultures, isolated from different type soils of dry subtropical-step zone of Sighnaghi (east Georgia); also to establish the extreme and optimal ranges of temperature for growth and spore-formation of micromycetes.

Materials and Methods

Cultures of microscopic fungi from the collection of the laboratory of biotechnology of S. Durmishidze Institute of biochemistry and biotechnology served as investigation objects.

The microscopic fungi were grown on the universal nutrient medium: wort (content of sugar 7.0%)-1.0l, agar-20.0.

Surface cultivation of cultures was performed on Petri dishes at temperature range -0°C - 60°C with 5°C intervals.

The growth of microscopic fungi was determined by means of measuring two parameters – the diameter of the colony in two perpendicular directions after 3-fold cultivation, 3, 5 and 7 days later. On the other hand the density of hyphae of the developing colonies in different parts was measured. The final sum of both parameters was appreciated via 3-point system.

Results and Discussion

Experiments were done on the cultures of microscopic fungi isolated from brown carbonate, chestnut and meadow-chernozem soils of Sighnaghi region. The collection consisted of 70 cultures representing 10 different genera of the 3 main classes of microscopic fungi, in particular: Zygomycetes (*Mortierella, Mucor, Rhizopus*), Ascomycetes (*Aspergillus, Penicillium, Chaetomium*) and Deuteromycetes (*Botrytis, Cladosporium, Trichoderma*) [Daushvili et al., 2004].

Microscopic fungi were grouped as thermophiles (obligative thermophiles), thermotolerants (facultative thermophiles) and psychrophiles (facultative psychrophiles) following Cooney and Emerson.

Microscopic fungi with not less than 20°C minimal growth temperature and with maximum at 50°C, were regarded as obligative thermophiles. The facultative thermophiles grew at lower than 20°C and at higher than 50°C. Facultative psychrophyles grew at 0°C and lower temperature and at the same time at 20-25°C too.

According to experimental results 20.0% of the studied fungi were extremophiles by temperature (Fig. 1). Since the tropical and subtropical climatic zones represent a favorable habitat for thermophilic fungi, 14.2% of obligate and facultative thermophiles is a natural phenomenon for Sighnaghi region soils [Bilay, 1985]. Presence of psychrotrophic micromycetes was less expectable here, while existence of facultative psychrophiles may be explained by their tolerance to moderate temperatures (20°-25°C).

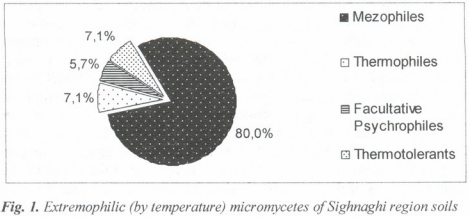


Fig. 1. *Extremophilic (by temperature) micromycetes of Sighnaghi region soils*

Majority of thermophilic and psychrophilic microscopic fungi were isolated from meadow-chernozem soils. This may be explained by the fact that this type of soil was distinguished with abundance and diversity of genera of microscopic fungi.

Among 10 studied genera of fungi thermophilic features were revealed only in genera: *Aspergillus, Chaetomium* and *Penicillium*. Psychrophiles were found among the genera: *Mortierella, Mucor* and *Cladosporium*. Majority of obligate and facultative thermophiles belonged to *Aspergillus* genus (Fig. 2).

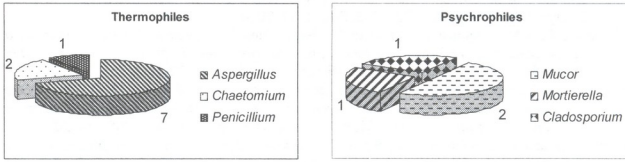


Fig. 2. *Genera of extremophilic fungi of Sighnaghi region soils*

The temperature amplitudes for optimal growth of thermophilic and thermotolerant microscopic fungi were established (Table 1). The temperature amplitude of optimal growth distinguished the thermotolerant representatives of *Aspergillus* genus. More over, the morphological changes, caused by the approaching the extreme temperature limit were less evident, while in genera *Chaetomium* and *Penicillium* the changes were clearly revealed. These morphological changes were mainly expressed in significant decline of spore-formation and transformation of colony color. In some cases changes of hypae length and surface consistence were observed.

Table 1. The optimal temperature ranges for growth of thermophilic fungi.
1 - *Chaetomium sp. S77,* 2 - *Aspergillus sp. S73,* 3 - *Aspergillus niger S60,* 4 - *Aspergillus niger S64,* 5 - *Aspergillus niger S65,* 6 – *Penicillium sp. S57,* 7 – *Chaetomium sp. S67,* 8 - *Aspergillus sp. S51,* 9 - *Aspergillus sp. S52,* 10 - *Aspergillus sp. S58.*

Minimum	Optimal temperature for growth							Maximum
22, 23				1				53, 55
23, 24				2				53, 55
17, 19				3				54, 55
17, 18				4				53, 54
23, 24				5				55
15, 16				6				54
15, 17				7				53
17, 19				8				55
22, 23				9				53, 54
23				10				51, 52
	17-19	28-30	38-40	41-42	44-45	47-48	51-52	

Temperature °C

References:

Bilay T. *Thermophilic fungi*. Kiev, 1985 (in Russian).

Bilai T. *Thermostable enzymes of fungi*. Kiev, 1979 (in Russian).

Daushvili L., Kutateladze L., Burduli T., Jobava M., Dzalamidze I., Aleksidze T., Tinikashvili L. *Isolation of microscopic fungi from soils of different regions of the Caucasus*. Proc. Georgian Acad. Sci., Biol. Ser. B. **2**, 5-6, 68-73, 2004.

De Faria F., Te'o V., Begquist P., Azevedo M., Nevalainen K. *Expression and processing of a major xylanase (XYN2) from the thermophilic fungus Humicola grisea var. thermoidea in Trichoderma reesei*. Letters in Appl. Microbiology, **34**, 119-123, 2002.

Kunamneni A., Kumar K., Singh S. *Response surface methodological approach to optimize the nutritional parameters for enhanced production of α-amylase in solid state fermentation by Thermomyces lanuginosus*. African Journal of Biotechnology, **4,** 7, 708-716, 2005.

Phutela U., Dhuna V., Sandhu Sh., Chadha B.S. *Pectinase and polygalacturonase production by a thermophilic Aspergillus fumigatus isolated from decomposting orange peels*. Brzailian Journal of Microbiology, **36**, 63-69, 2005.

სიღნაღის რეგიონის ნიადაგებში გავრცელებული თერმოფილური მიკროსკოპული სოკოები

დაუშვილი ლ., ბურდული თ., ქუთათელაძე ლ., ჯობავა მ., ძალამიძე ი.

ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 14.08.2006)

რეზიუმე

შესწავლილია სიღნაღის რეგიონის მშრალი სუბტროპიკული სტეპის ზონის ნიადაგებიდან გამოყოფილი მიკროსკოპული სოკოების 70 კულტურა. გამოვლენილია 5 თერმოფილური, 5 თერმოტოლერანტული და 4 ფსიქროფილური (ფაკულტატური) მიკრომიცეტი. დადგენილია თერმოფილური მიკრომიცეტების ზრდისა და სპორაწარმოქმნის ოპტიმალური ტემპერატურული ამპლიტუდები.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

STUDY OF ANTIBIOTIC AND PHAGE SENSITIVITY OF SOME AEROBIC PYOGENIC BACTERIA

GIORGADZE M.[1], NATIDZE T.[2]

[1]*Ltd "Doagnosis-90"*
[2]*Ltd "Immunogen"*

(Received May 15, 2006)

Abstract

It was shown that isolates of streptococcus, staphylococcus and Escherichia coli isolated from suppurative inflammation areas and from the blood of the dog diseased with sepsis are more sensitive to enco- and intestibacteriophages, as compared to antibiotics. At the same time lysis degree is mostly equaled to 4+ and 3+. The obtained results revealed outlook for usage of bacteriophages for treatment of infectious diseases of bacterial etiology in dogs, as preparation without side effects.

Key words: suppurative inflammation of skin, enco- and intestibacteriophages

Introduction

Forming of microbial populations resistant to antibiotics is the actual problem of medicine and veterinary. This phenomenon, which is mainly caused by specific plasmids occurring in bacteria, is connected with purposeless and sometimes uncontrolled usage of antibiotics. Usage of some antibiotics, due to resistance against them, is noneffective and improper.

Medical and veterinary practice demands to look for new antibiotics and to work out alternative preparations. Such preparation is a bacteriophage, which is successfully used in medical practice for treatment of infectious diseases, and among them for suppurative inflammation of skin. Priority of bacteriophages compared to antibiotics is impossibility of forming of resistant populations, absence of allergy, reproduction in the nidus of infection, rapidity of preparation, etc. [Barrow, Soothill, 1997; Carlton, 1999; Smith, Huggins, 1982; Marks, Sharp, 2000]

The aim of our work was comparative study of antibiotic and phage sensitivity of some aerobic pyogenic bacteria isolated from dogs sick with skin diseases (dermatitis, pyoderma), and from the dogs sick with sepsis.

Materials and Methods

In our experiments we used ten isolates of staphylococcus (*St. aurous* - 6, *St. epidermidis* – 4), six isolates of streptococcus (*Str. pyogenes* – 4, *Str.viridans* – 2), and seven isolates of Escherichia coli (*E.cili hemolytic* – 3, *E.coli nonhemolytic* – 4).

Antibiotic sensitivity was studied by disk-diffusion method, phage sensitivity – by Fisk modified method [Overturf et al., 1991].

In our study we used the following antibiotics: amoxacillin, ampicillin, ampicide, gentamicin, doxicicline, erythromycin, kanamycin, chloramphenicol, penicillin, streptomycin, tetracycline, triaxon, cipro-bai and ciprofloxacin.

Among bacteriophages were used the following ones: encophage, intestibacteriophage, pyophage, Sisphage, Fersisphage.

Results and Discussion

As a result of our studies it was established that the effect of antibiotics and phages on staphylococcus, streptococcus and Escherichia coli is different (Table 1). Some staphylococcus are polyresistant against antibiotics. For example, the majority among them revealed resistance against erythromycin, kanamycin, streptomycin, tetracycline, and partially against gentamicin and penicillin. Amoxicillin, ampicillin, ampicide, triaxon have high influence on staphylococcus. To those antibiotics *St.epidermis*-2 appeared to be resistant.

Table 1. Antibiotic and phage sensitivity of microbes

№	Microbes	Antibiotic													Bacteriophage					
		Amoxacillin	Ampicillin	Ampicide	Gentamicin	Doxicicline	Erythromycin	Kanamicin	Chloramphenicol	Penicillin	Tetracycline	Triaxon	Streptomycin	Cipro-bai	Ciprofloxacin	Encophage	Intestiphage	Pyophage	Sisphage	Fersisphage
1	*St. aureus - 1*	4+	4+	4+	4+	R	R	2+	4+	4+	R	R	3+	R	2+	4+	4+	4+	2+	2+
2	*St. aureus - 2*	4+	2+	2+	R	R	R	R	R	R	R	2+	2+	R	2+	3+	3+	2+	2+	2+
3	*St. aureus - 3*	4+	3+	4+	2+	3+	R	R	R	R	3+	4+	R	R	3+	2+	3+	2+	2+	R
4	*St. aureus - 4*	4+	4+	4+	3+	2+	R	2+	2+	4+	4+	4+	R	3+	3+	R	2+	2+	2+	2+
5	*St. aureus - 5*	4+	4+	4+	3+	4+	R	2+	4+	2+	R	3+	R	3+	4+	4+	4+	3+	R	3+
6	*St. aureus - 6*	4+	4+	4+	2+	2+	R	R	R	R	R	3+	R	3+	2+	2+	R	R	2+	2+
7	*St. epidermidis - 1*	3+	3+	2+	R	R	3+	3+	4+	2+	R	2+	2+	R	R	R	3+	2+	R	R
8	*St. epidermidis - 2*	R	R	R	R	R	R	R	3+	R	R	3+	R	R	2+	2+	2+	2+	2+	R
9	*St. epidermidis - 3*	4+	3+	-	3+	R	R	R	4+	4+	2+	4+	R	2+	4+	3+	2+	2+	2+	2+
10	*St. epidermidis - 4*	4+	4+	2+	2+	4+	R	R	R	3+	R	3+	R	3+	4+	2+	3+	3+	2+	2+
11	*St. pyogenes - 1*	3+	R	3+	4+	4+	-	2+	4+	2+	2+	4+	2+	4+	4+	3+	3+	2+	2+	2+
12	*St. pyogenes - 2*	4+	2+	4+	4+	4+	-	3+	4+	3+	3+	4+	3+	3+	4+	2+	3+	2+	R	2+
13	*St. pyogenes - 3*	4+	3+	4+	4+	4+	-	3+	4+	3+	3+	4+	3+	3+	4+	2+	2+	2+	R	R
14	*St. pyogenes - 4*	4+	3+	2+	3+	3+	2+	3+	4+	2+	3+	2+	2+	4+	3+	4+	4+	3+	2+	2+
15	*St. viridans - 1*	3+	3+	3+	3+	4+	2+	3+	4+	2+	4+	3+	3+	3+	3+	3+	4+	3+	2+	R
16	*St. viridans - 2*	3+	R	4+	3+	3+	2+	2+	3+	2+	4+	4+	2+	4+	4+	3+	3+	3+	2+	2+
17	*E. coli (H) - 1*	4+	4+	4+	3+	4+	R	2+	4+	R	4+	4+	2+	4+	4+	2+	3+	3+	2+	2+
18	*E. coli (H) - 2*	4+	4+	4+	3+	4+	R	4+	4+	R	4+	3+	3+	4+	3+	3+	3+	3+	3+	2+
19	*E. coli (H) - 3*	3+	3+	2+	4+	-	R	2+	4+	2+	3+	2+	3+	4+	2+	3+	2+	2+	2+	3+
20	*E. coli (NH) - 4*	4+	3+	2+	3+	-	R	3+	4+	3+	4+	3+	3+	3+	2+	2+	R	R	R	2+
21	*E. coli (NH) - 5*	4+	4+	2+	3+	-	2+	4+	3+	2+	4+	2+	3+	4+	3+	2+	3+	3+	2+	R
22	*E. coli (NH) - 6*	3+	2+	2+	4+	3+	3+	R	3+	3+	4+	3+	2+	4+	3+	2+	3+	3+	3+	2+
23	*E. coli (NH) - 7*	4+	4+	2+	3+	4+	4+	3+	3+	R	3+	3+	2+	3+	3+	3+	3+	2+	3+	2+

Note: R – resistant; "-" – not studied; H – hemolytic; NH – nonhemolytic.

Staphylococcus, as compared to antibiotics, lyses intensively encophage and intestibacteriophage. It should be mentioned that poly-antibiotic resistant staphylococcus (*St.aureus*-2, *St.aureus*-3, *St.epidermidis*-1, *St. epidermidis*-3, *St.epidermidis*-4) in most cases are lysed by bacteriophages in various degrees. Enco- and intestibacteriophage are especially characterized by this feature.

Unlike staphylococcus, streptococcus are more sensitive to the influence of antibiotics and bacteriphages. Among antibiotics with especially intensive effect are distinguished: amoxacillin, ampicillin, ciprofloxacin, triaxon and cipro-bai, (4+, 3+), and among bacteriophages – enco- and intestibacteriophages (4+, 3+, 2+).

Antibiotic- and phage-sensitivities of Escherichia coli are nearly similar. For example, *E.coli*-4, which is resistant against erythromycin, is sensitive against enco- and Fersisphage, and *E.coli-7*, which is resistant against penicillin, turned out to be sensitive against all bacteriophages. In other cases E.coli strains are sensitive to antibiotics (amoxacillin, ampicillin, ciprofloxacin, cipro-bai) and to bacteriophages, especially to enco- and intestiphages (3+, 2+).

Thus, Sensitivity of staphylococcus, streptococcus and Escherichia coli to antibiotics and phages is different. Staphylococcus shows distinct resistance to antibiotics. Strains are sensitive to enco- and intestibacteriophages that enables to use them as alternative agent.

References:

Barrow PA., Soothill JS. *Bacteriophage therapy and prophylaxis: rediscovery and renewed assessment of potential.* Trends Microbiol., **5**, 268-271, 1997.

Carlton RM. *Phage therapy: past history and future prospects.* Arch. Immunol. Ther. Exp., **47**, 267-274, 1999.

Marks T., Sharp R. *Bacteriophages and biotechnology.* J.Chem.Technolgy and Biotechnology, **75**, 1, 6-17, 2000.

Overtruf G.D., Talan D.A., Singer K. et al. *Phage Typing of Staphylococcus.* J.Clin.Microbiol., 29, 2, 373-375, 1991.

Smith HW., Huggins MB. *Successful treatment of experimental Escherichia coli infections in mice using phage: its general superiority over antibiotics.* J. Gen. Microbiol., **128**, 307-318, 1982.

ზოგიერთი აერობული ჩირქმბადი ბაქტერიის ანტიბიოტიკო- და ფაგომგრძნობელობის შესწავლა

გიორგაძე მ.[1], ნათიძე თ.[2]

[1]შპს „დიაგნოზი-90“;
[2]შპს „იმუნოვენი“

(მიღებულია 15.05.2006)

რეზიუმე

დადგენილია, რომ კანის ჩირქოვანი უბნებიდან და სეფსისით დაავადებული ძაღლის სისხლიდან გამოყოფილი სტრეპტოკოკების, სტაფილოკოკების და ეშერიხიების ი'ზოლატები ანტიბიოტიკებთან შედარებით გაცილებით მგრძნობიარეა ენკო- და ინტესტიბაქტერიოფაგების მიმართ. ამასთან, ლიზისის ხარისხი უმეტესად ტოლია 4+ და 3+. მიღებული შედეგები იძლევა ძაღლებში ბაქტერიოფაგის გამოყენების პერსპექტივას.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

DETERMINATION OF EFFECTIVE SCHEME OF ANTI-BRUCELLOSIS IMMUNIZATION OF HORNED CATTLE

GVINJILIA G., GVINJILIA M.

Laboratory of Ministry of Agriculture of Georgia

(Received August 10, 2006)

Abstract

Stability of immune response at vaccination of animals with strain-19 of anti-brucellosis vaccine was studied. Vaccination scheme of adult animals in stationary conditions was worked out. This regimen enables us to conduct animal vaccination by small dose once a year.

Key words: Rose-Bengal Reaction, Agglutination reaction, serological methods

Introduction

It is well known that Brucella is the microorganism causing worldwide spread disease – brucellosis. Brucella mainly infects horned cattle, sheep, goat, pig. Brucella infection can be also seen in wild species of animals [Callahan, 2006 a].

Brucellosis is a persistent disease conducted with elimination of microbe from reproductive system and mammary gland [Sigafoose, 2006]. On account of an economic impact connected with animal health and infection risk of humans the most countries have program of brucellosis control, which involves vaccination of young and adult animals by strain-19. Today, in epizootic and epidemiologic viewpoint the situation is alarm in Georgia [Callahan, 2006 b].

Thus, the goal of our work is to improve diagnostic methods, and special prophylactic agents, and schemes of their usage [Payeur, 2006]. We aimed to determine effectiveness of strain-19 and immune status of animal at various dosing regimen. One link of chain – recipient animal – must become as no recipient. It should be realized by usage of specific immunization; i.e. for extermination of brucellosis immune barrier should be used. Vaccinal prevention may directly set up the precondition of further irreversible liquidation of the disease.

Materials and Methods

88 calves of 4-6 months old and 2000 adult cows from safe on brucellosis farms were used in experiments. Their immunization with strain-19 was carried out in the following dosing regimes: 0.5 billions, 3 billions, 9 billions, and standard 80 billions of microbial cells.

The following standard serological methods were used:

1. Rose-Bengal Reaction (RBR),
2. Agglutination (Raite) reaction (AR)

Results and Discussion

The obtained data show that with the decrease of dose antigenic effect of preparation decreases, postvaccinal reactions disappear earlier than usual (Table 1.).

The obtained results reveal that calves, which were immunized with small dose of vaccine, maintain postvaccinal reactions during 4 months. The calves vaccinated with 9 and 80 billions of microbial cells maintain stable immune status during one year.

Among 22 calves immunized with 9 billion microbial cells 21 turned out to be positive. Animals immunized with standard dose show the same result.

On the second stage of study experiments were carried out on cows in order to determine postvaccinal state in adult cows. Experimental animals (2000 ones) were divided into two groups, vaccinated with 9 and 80 billion microbial cells, respectively.

As a result of serological investigations conducted after 4 months, as well as after 1 year it was revealed that postvaccinal reaction is positive during one year almost in all animals. Percentage of animals having positive reaction, which were vaccinated with 9 billion microbes consists of 92.7% at the end of year, but of animals vaccinated with 80 billion microbes – 93.3%.

Thus, we consider that high dosing regime is not needed, as post-immunization reactions are similar. The common scheme of anti-brucellosis vaccination of adult animals should be the following: cows being in stationary conditions must be vaccinated with the dose of 9 billion microbes once in a year. In that way we should prevent the problem of postvaccinal reaction without reduction of immunogenic characteristics of preparation.

Table 1. Results of serological studies carried out on 4-6 months old calves

Animal groups	Number of animals	Dose of immunization (billion microbes)	After 4 months		After year	
			positive	negative	positive	negative
1	22	0.5	12	19	-	22
2	22	3	15	7	2	20
3	22	9	21	1	21	1
4	22	80	22	-	21	21

Table 2. Results of serological studies carried out on adult caws

Number of animals	Dose of immunization (billion microbes)	After 4 month		After year	
		positive	negative	positive	negative
1000	9	990	10	927	73
1000	80	994	6	933	67

References:

Callahan J. *Brucellosis.* In: Collection of works in microbiology and virology. N24914-101, 2006a.

Callahan J. *Epidemiology of brucellosis.* In: Collection of works in microbiology and virology. N24914-102, 2006b.

Sigafoose T.M. *Brucellosis.* In: Collection of works in microbiology and virology. N24914-100, 2006.

Payeur J.B. *Laboratorial methods of brucellosis investigation.* In: Collection of works in microbiology and virology. N24915, 2006.

მსხვილფეხა რქოსანი პირუტყვის ბრუცელოზის საწინააღმდეგო იმუნიზაციის სქემის განსაზღვრა

ღვინჯილია გ., ღვინჯილია მ.

სხია სოფლის მეურნეობის სამინისტროს ლაბორატორია

(მიღებილია 10.08.2006)

რეზიუმე

შესწავლილია იმუნური პასუხის სტაბილურობა ცხოველის ორგანიზმში ბრუცელოზის საწინააღმდეგო ვაქცინის შტამი-19 მცირე დოზის შეყვანისას. შემუშავებულია ზრდასრული ცხოველების ვაქცინაციის სქემა სტაციონარულ პირობებში, რომელიც საშუალებას იძლევა განვახორციელოთ ცხოველთა ვაქცინაცია მცირე დოზით წელიწადში ერთხელ.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

STUDY OF THE EXTENT OF EXTREMOPHILICY OF HALOPHILIC MICROSCOPIC FUNGI FROM SALINE SOILS OF KAKHETI PLAIN

ZAKARIASHVILI N., *KUTATELADZE L., JOBAVA M., KHOKHASHVILI I., DZALAMIDZE I., KVESITADZE E., ALEKSIDZE T.

S. Durmishidze Institute of Biochemistry and Biotechnology
D. Agmashenebeli Alley 10km, 0059 Tbilisi, Georgia
**E-mail: kutateladze@geomail.ge*

(Received May 15, 2006)

Abstract

The extent of extremophilicy of halophilic microscopic fungi, isolated from saline soils of Kakheti plain has been investigated. The pH and temperature ranges of 44 cultures of fungi were established. The optimal pH and temperature of cultivation for each halophile has been selected. 23 extremophiles were revealed: 8 of them by temperature and 15 – by pH. 3 of them were thermophiles, 2 – psycrotolerants, 3 – thermotolerants, 10 – alkaliphiles, and 5 – pH-tolerants. 5 microscopic fungi – *Aspergillus sp.* A26, *Chaetomium sp.* A36, *Trichoderma sp.* A40, *Trichoderma sp.* A41 and *Fusarium sp.* A10 were regarded as extremophiles. They revealed extremophilic properties by three parameters simultaneously – temperature, pH and resistance to high concentrations of NaCl.

Key words: extremophiles, halophiles, thermophiles, pH-tolerant, microscopic fungi

Introduction

Determination of the limits of existence of living organisms is one of the central problems of present-days biology. Due to their high genetic and physiological adaptivity, microorganisms belong to the unique forms of life, managing to exist under the extreme environmental conditions (high and low temperatures, acid and alkali surrounding, high concentrations of salt, etc.) [Aguliar, 1996; Dix, 1995; Mouchacca, 1997; Stetter, 1999]. These types of microorganisms are named as extremophiles. They comprise 3 main groups: 1. thermophiles and psycrophiles 2. alkali- and acidophiles, 3. halophiles. Halophiles deserve a special interest among the extremophiles. Resistance to unfavorable environmental conditions, unique chemical structure and nonpathogenous properties of this group of microorganisms are responsible for involving them into the biotechnological processes. The halophilic cell is a "live laboratory", which makes possible to create several commercial products simultaneously (enzymes, nucleic acids, betonies, ectoines, carotinoids, etc.). The inhabitants of hyper-alkali soils are regarded as "rectifiers" of the environment polluted with organic toxins. In spite of the mentioned biopotential, the abilities of halophils are less used in large technological processes. From the practical experience it is clear that the stable enzymes, acting in extreme regimen (high temperature, alkali or acid surrounding)

possess great advantages in enzyme technology and biotechnology. Extremophiles are producers of this type of enzymes. According to this fact, selection of a high quality extremophiles among the halophilic microscopic fungi, isolated in our early experiments from saline soils of Kakheti plain [Laskhishvili et al., 2005], was of great interest.

Materials and Methods

44 microscopic fungi, isolated from saline soils of Kakheti plane served as objects for experiment. To select extremophiles, microscopic fungi were cultivated under the extreme conditions, on an universal, agar nutrient medium, containing the optimal concentration of NaCl. According to early experiments, the optimal concentrations of NaCl were established for particular halophile [Stetter, 1999].

Microscopic fungi were cultivated under the wide range of temperature (0-55°C, with intervals of 5°C), to reveal thermo- and psycrophiles. The wide amplitude of pH was tested to select acid and alkaliphiles (pH 2.0-10.0, with 0.5 intervals). Cultivation prolonged for 10 days. Values of temperature or pH, resulting in maximal increase, were regarded as optimal. Intensity of growth was evaluated using 3-mark system, taking into account the diameter and growth velocity of the colony of micromycetes.

While the thermophilicy was detected, the determinations offered by Quean and Emerson were used. In particular, the cultures of micromycetes with existence ranges from 20°C to 55°C were regarded as thermophiles. The cultures with maximal growth at ~50°C and able to develop at lower than 20°C were distinguished as thermotolerants.

Microscopic fungi growing at low temperature, but able to develop at 40°C too, were grouped as psycrotolerants.

Micromycetes well developing equally at pH ranging from 5 to 10 were regarded as alkaliphiles, while others, growing at a wide range of pH (2.0 - 10.0) were grouped as pH-tolerant.

Results and Discussion

While determining the extent of extremophilicy of halophilic microscopic fungi isolated from saline soils of Kakheti plane the existing ranges and optimal temperatures were established first of all. For this purpose halophiles were grown at universal agar nutrient medium with optimal concentrations of NaCl, at a wide range of temperature – from 0°C to 55°C, with 5°C intervals.

In table 1 the characteristics of microscopic fungi from saline soils of Kakheti plane are given following the temperature of cultivation. From the table it is clear that the majority of microflora consisted of mesophiles with optimal growing temperature 28-30°C. These meanings are in accordance with the literature data about the prevailing of mesophiles among the microorganisms in nature.

Some representatives of investigated strains sharply changed its morphological and cultural features while approaching the critical temperature. Representatives of different genera of fungi diversely reacted on a temperature fluctuations, e.g. degeneration of spores was mentioned in some species of *Fusarium* genus. The fluffy mycelium of several strains of the genus Mucor turned into skinny one. In some cultures of *Aspergillus* changes in colour or difficulties in spore merging was mentioned.

8 extremophiles by temperature has been selected among the microscopic fungi, isolated from saline soils of Kakheti plain. Among them 3 cultures – *Aspergillus sp.* A2, *A. sp.* A26, and *Chaetomium sp.* A36 were thermophiles, 2 – *Fusarium sp.* A10 and *F. sp.* A43 were psicrotolerants, and 3 – *Trichoderma sp.* A40, *T. sp.* A41 and *T. sp.* A42 were thermotolerants.

After arranging the extremophiles by temperature we aimed to reveal microscopic fungi, growing at extremal pH. For this purpose the cultures from the collection were grown on universal agar nutrient medium, with optimal concentration of NaCl and optimal temperature, changing the pH of the medium within wide range (from 2.0 to 11.0).

In Table 2 the halophilic microscopic fungi spread in saline soils of Kakheti plane are presented according to pH meanings. Among 44 strains of the collection, 15 turned to be halophiles. Between them 10 were alkaliphiles and 5 – pH-tolerant.

Table 1. The extent of extremophilicy of microscopic fungi from saline soils of Kakheti plane

Microscopic fungi	Temperature ranges of culture growth	Optimal temperature of growth, °C	Group of micromycetes by temperature	Relation of culture to NaCl concentrations
1. *Aspergillus sp. A-1*	15°C-40°C	30°C	mesophile	weak
2. *Aspergillus sp. A-2*	20°C-55°C	40°C-45°C	thermophile	weak
3. *Aspergillus sp. A-3*	15°C-45°C	28°C-32°C	mesophile	weak
4. *Aspergillus sp. A-4*	15°C-45°C	30°C	mesophile	weak
5. *Aspergillus sp. A-13*	15°C-40°C	28°C	mesophile	moderate
6. *Aspergillus sp. A-14*	15°C-45°C	25°C	mesophile	moderate
7. *Aspergillus sp. A-25*	15°C-40°C	30°C	mesophile	extreme
8. *Aspergillus sp. A-26*	20°C-55°C	40°C-45°C	thermophile	extreme
9. *Aspergillus sp. A-27*	15°C-40°C	30°C	mesophile	halotolerant
10. *Aspergillus sp. A-28*	15°C-45°C	30°C	mesophile	moderate
11. *Aspergillus sp. A-29*	15°C-45°C	30°C	mesophile	extreme
12. *Aspergillus sp. A-30*	15°C-45°C	30°C	mesophile	moderate
13. *Aspergillus sp. A-31*	15°C-45°C	25 °C -30°C	mesophile	halotolerant
14. *Penicillium sp.A-5*	15°C-45°C	25 °C -30°C	mesophile	weak
15. *Penicillium sp.A-6*	15°C-40°C	25 °C -30°C	mesophile	moderate
16. *Penicillium sp.A-15*	15°C-40°C	25 °C -30°C	mesophile	moderate
17. *Penicillium sp.A-16*	15°C-40°C	25 °C -30°C	mesophile	moderate
18. *Penicillium sp.A-17*	15°C-40°C	25 °C -30°C	mesophile	moderate
19. *Penicillium sp.A-18*	15°C-40°C	25 °C -30°C	mesophile	moderate
20. *Penicillium sp.A-19*	15°C-40°C	25 °C -30°C	mesophile	moderate
21. *Penicillium sp.A-20*	15°C-40°C	30°C	mesophile	halotolerant
22. *Penicillium sp.A-32*	15°C-45°C	30°C	mesophile	moderate
23. *Penicillium sp.A-33*	15°C-45°C	25 °C -30°C	mesophile	halotolerant
24. *Penicillium sp.A-34*	15°C-40°C	30°C	mesophile	halotolerant
25. *Penicillium sp.A-35*	15°C-40°C	30°C	mesophile	halotolerant
26. *Chaetomium sp. A-36*	20°C-55°C	40°C-45 °C	thermophile	halotolerant
27. *Chaetomium sp. A-37*	15°C-40°C	30°C	mesophile	halotolerant
28. *Chaetomium sp. A-38*	15°C-40°C	30°C	mesophile	moderate
29. *Chaetomium sp. A-39*	15°C-40°C	25 °C -30°C	mesophile	extreme
30. *Allescheria sp. A-11*	15°C-40°C	30°C	mesophile	weak
31. *Allescheria sp. A-12*	15°C-45°C	25 °C -30°C	mesophile	weak
32. *Cladosporium sp. A-21*	15°C-40°C	30°C	mesophile	moderate
33. *Cladosporium sp. A-22*	15°C-40°C	30°C	mesophile	moderate
34. *Fusarium sp. A-7*	15°C-40°C	25 °C -30°C	mesophile	weak
35. *Fusarium sp. A-8*	15°C-40°C	30°C	mesophile	weak
36. *Fusarium sp. A-9*	5°C-40°C	10°C-20 °C	mesophile	weak
37. *Fusarium sp. A-10*	5°C-40°C	10°C-20 °C	psicrotrophe (psicrotolerant)	halotolerant
38. *Fusarium sp. A-43*	15°C-45°C	30°C	mesophile	halotolerant
39. *Fusarium sp. A-44*	5°C-55°C	20°C-35 °C	thermotolerante	halotolerant
40. *Trichoderma sp. A-40*	5°C-55°C	20°C-35 °C	thermotolerante	halotolerant
41. *Trichoderma sp. A-41*	5°C-55°C	20°C-35 °C	thermotolerante	moderate
42. *Trichoderma sp. A-42*	15°C-45°C	20 °C -30°C	thermotolerant	halotolerant
43. *Mucor sp. A-23*	15°C-45°C	20°C -30°	mesophile	moderate
44. *Mucor sp. A-24*	15°C-45°C	20°C -30°	mesophile	moderate

Table 2. The extent of extremophilicy of microscopic fungi by pH

Microscopic fungi	Place of sampling	The range of living pH of the culture	Optimal pH	Characterization of culture
1. *A.sp.A25*	Soils with high salinity	5.0-10.0	9	alkaliphile
2. *A.sp.A26*		5.0-10.0	9	alkaliphile
3. *A.sp.A27*		4.5-8.5	5.0	-
4. *A.sp.A28*		4.5-7.5	5.0	-
5. *A.sp.A29*		2.5-10.0	4-8.0	pH-tolerant
6. *A.sp.A30*		4.0-7.5	6.0	-
7. *A.sp.A31*		2.5-10.0	4-8.0	pH-tolerant
8. *P.sp.A32*		4.5-7.5	6.0	-
9. *P.sp.A33*		2.5-10.0	3.5-7.5	pH-tolerant
10. *P.sp.A34*		4.5-10.0	9	alkaliphile
11. *P.sp.A35*		4.5-7.5	5.0	-
12. *Ch.sp.A37*		4.5-7.5	5.0	-
13. *Ch.sp.A36*		4.5-10.0	9	alkaliphile
14. *Ch.sp.A38*		5.0-8.0	6	-
15. *Ch.sp.A39*		4.5-7.5	6.0	-
16. *F.sp.A43*		5.0-7.5	6.0	-
17. *F. sp.A44*		4.5-7.5	5.5	-
18. *T. sp.A40*		4.5-10.0	9.0	alkaliphile
19. *T. sp.A41*		5.0-10.0	8.5	alkaliphile
20. *T. sp.A42*		4.5-8.0	6.0	-
21. *A. spA.13*	Soils with moderate salinity	5.0-8.0	6.0	-
22. *A. spA.14*		4.5-7.5	5.5	-
23. *P.sp.A15*		4.5-7.5	5.0	-
24. *P.sp.A16*		4.5-8.0	5.5	-
25. *P.sp.A17*		4.5-7.5	5.5	-
26. *P.sp.A18*		4.5-7.5	5.5	-
27. *P.sp.A19*		4.5-8.0	6.0	-
28. *P.sp.A20*		4.5-7.5	5.5	-
29. *Cl. sp.A21*		4.5-8.0	6.0	
30. *Cl. sp.A22*		4.5-7.5	6.0	-
31. *M. sp.A23*		4.5- 10.0	9.0	alkaliphile
32. *M. sp.A24*		4.5-10.0	9.0	alkaliphile
33. *A. sp.A1*	Soils with weak salinity	4.5-7.5	5.5	-
34. *A. sp.A2*		4.5-10.0	8.5	alkaliphile
35. *A. sp.A3*		4.5-7.5	5.5	-
36. *A. sp.A4*		4.5-7.5	6.0	-
37. *P.sp.A5*		4.5-7.5	5.5	-
38. *P.sp.A6*		4.5-8.0	6.0	-
39. *A. sp.A11*		5.0-8.0	6.0	-
40. *A.sp.A12*		4.5-7.5	6	-
41. *F. sp.A7*		2.5-10.0	4.5-8.0	pH-tolerant
42. *F. sp.A8*		2.5-10.0	4.5-8.0	pH-tolerant
43. *F. sp.A9*		4.5-7.5	6.0	-
44. *F. sp.A10*		4.5-10.0	8.5	alkaliphile

From the literature it is known that some representatives of *Aspergillus* and *Penicillium* genera are able to grow at a wide range of pH – from 2.0 to 10.0. The pH-tolerant microscopic fungi revealed in our experiments, belong also to these genera. Alkaliphils were found almost among all genera of fungi, except *Cladosporium* and *Allescheria* (Table 2). Extremophilic cultures by simultaneously three parameters (pH, temperature and NaCl concentration) were revealed on the base of selecting the extremophiles by pH and temperature. In particular, as high-quality extremophiles were evaluated: 1. thermophilic *Aspergillus sp.* A26, which is the extreme halophile and alkaliphile, at the same time. 2. *Chaetomium sp.* A36 – as halotolerant, alkaliphile and

thermotolerant. 3, 4. *Trichoderma sp.* A40 and *Trichoderma sp.* A41– as moderate halophile, alkaliphile and thermotolerant, and 5. *Fusarium sp.* A10 – as halotolerant, alkaliphile and psicrotolerant.

Selection of cultures, resistant to extreme conditions of temperature, pH and NaCl concentrations, and revealing the active producers of enzymes among them, is the perspective base for creating of new technologies.

References:

Aguliar A. *Extremophile research in the Europian Union: from fundamental aspects to industrial expectation.* FEMS Microbiol. Rev. 18, 89-92, 1996.

Dix N. I., Webster I. F*ungal ecology.* Chapman & Hall, London, U. K., 1995.

Laskhishvili M., Kutateladze L., Zakariashvili N., et al. *Izolation and identification of microscopic fungi from salty soils of Kakheti.* Proc. Georg. Acad. Sci. Biol. Ser. B, 3, 4, 60-63, 2005.

Mauchacca I. *Thermophilic fungi: biodiversity and taxonomic status.* Cryptogam. Mycol., 18, 19-69, 1997.

Stetter K. O. *Extremophiles and their adaptation to hot environments.* FEBS Lett., 452, 22-25, 1999.

კახეთის ვაკის მარილიან ნიადაგებში გავრცელებული ჰალოფილური მიკროსკოპული სოკოების ექსტრემოფილობის ხარისხის დადგენა

ზაქარიაშვილი ნ., ქუთათელაძე ლ., ჯობავა მ., ხოხაშვილი ი., ძალამიძე ი., კვესიტაძე ე. ალექსიძე თ.

ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 15.05.2006)

რეზიუმე

განსაზღვრულია კახეთის ვაკის მარილიანი ნიადაგებიდან გამოყოფილი ჰალოფილური მიკროსკოპული სოკოების ექსტრემოფილობის ხარისხი. დადგენილია კოლექციის 44 კულტურის სახიცოცხლო pH-ისა და ტემპერატურული დიაპაზონი. თითეული ჰალოფილისთვის შერჩეულია კულტივირების ოპტიმალური პირობები – pH და ტემპერატურა. გამოვლენილია 23 ექსტრემოფილი (მათ შორის 8 ტემპერატურის მიხედვით, 15 – pH-ის მიხედვით): 3 თერმოფილი, 2 – ფსიქროტოლერანტი, 10 – ალკალიფილი და 5 – pH-ტოლერანტი. მაღალი ხარისხის ექსტრემოფილებად შეფასებულია 5 მიკროსკოპული სოკო, რომელიც ერთდროულად სამი პარამეტრით (ტემპერატურა, pH და NaCl-ის მაღალი კონცენტრაციებისადმი მდგრადობა) აშკარავნებდა ექსტრემოფილობას: *Aspergillus sp.* A26, *Chaetomium sp.* A36, *Trichoderma sp. A*40, *Trichoderma sp.* A41 და *Fusarium sp.* A10

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Paleobiology

THE MAIN STAGES OF DEVELOPMENT OF *HAMAMELIDACEAE* ON THE TERRITORY OF EURASIA

SHATILOVA I., RUKHADZE L., MCHEDLISHVILI N.

Georgian National Museum, Davitashvili Institute of Paleobiology

(Received November 6, 2006)

Abstract

About the fossil *Hamamelidaceae* a big factual material is accumulated. In Europe and non-tropical Asia the findings of these plants are more often connected with the deposits of Oligocene, Early and Middle Miocene. In Georgia the greatest number of *Hamamelidaceae* were found in the Upper Miocene deposits. This material is of great interest, as it reflects the latest stage of florescence of the family in such region of Eurasia, where at present its representatives are fully absent.

Key words: *Hamamelidaceae*, development history, Western Georgia, Europe, non-tropical Asia.

Introduction

During the process of study of the palynological complexes of Sarmatian and Meotian deposits of Western Georgia our attention was attracted by the abundance of pollen grains of family *Hamamelidaceae.* That stimulated us to lead the monographycal investigation of this taxon and to trace its history on the territory of Eurasia. The big material was analyzed as nearly all Paleogene and Neogene floras described in literature contain one or another genus of *Hamamelidaceae*, except *Liquidambar*, which is the constant component of Cenozoic floras [Shatilova, Stuchlik, 2001].

Materials and Methods

In Georgia the earliest discoveries of *Hamamelidaceae* are dated by Paleogene. In the composition of Eocene, Oligocene, Early and Middle Miocene floras four genera (*Hamamelis, Corylopsis, Sycopsis, Liquidambar*) are known [Panova et al., 1984; Ramishvili, 1982].

In Late Miocene (Sarmatian, Meotian) the part of *Hamamelidaceae* in composition of flora increased and the family was represented by the following genera: *Hamamelis, Corylopsis, Eustigma, Fortunearia, Fothergilla, Parrotiopsis, Parrotia, Sycopsis, Distyliopsis, Distylium, Disanthus, Chunia, Liquidambar, Altingia.* The list is given by system of Endress [Endress, 1989].

All representatives of family *Hamamelidaceae* were probably the components of subtropical forests of plains and lower mountain belt (Fig.1.). In their composition both, evergreen and deciduous plants occur: *Carya, Lauraceae, Myrica, Quercus, Castanopsis, Araliaceae,* etc. [Kolakovsky, Shakryl, 1976; Shatilova et al., 1999].

After the Sarmatian the great number of subtropical forms died out. But this process didn't touch the family *Hamamelidaceae*, which in Meotian time continued to preserve the rich systematical composition. Between Sarmatian and Meotian some differences revealed in generic composition of family, but the number of genera was the same.

The main way of development of Pontian and Kimmerian vegetation was the widening of the area of deciduous cenosis and reduction of subtropical forests, which at the end of Middle Pliocene (Kimmerian) finished to exist as a separate formation. Subtropical plants preserved on the territory of Western Georgia after the Kimmerian became the components of warm-temperate forests. The single representatives of family *Hamamelidaceae (Liquidambar, Altingia, Corylopsis, Fortunearia, Parrotia)* were referred to such plants.

Results and Discussion

In the history of development of *Hamamelidaceae* on the territory of Georgia three stages can be distinguished. The initial one began after the first appearance of the taxon in geological chronology and continued till the time of its florescence. In Georgia it was the time of Paleogene, Lower and Middle Miocene. The second stage corresponded to the period of florescence of family. It was comparative short and embraced the Late Miocene (Sarmatian, Meotian). The third stage (time of decline of *Hamamelidaceae* and their extinction) was longer and corresponded to the whole Pliocene, Early and Middle Pleistocene.

The analysis of rich scientific literature shows that the first stage, during which the process of increasing of the systematical composition of *Hamamelidaceae* was going, finished in the Mesozoic in the non-tropical Asia. In Europe it was continued till the end of Eocene. During this time the representatives of *Hamamelidaceae* did not play significant role in plant communities, main components of which were palms and different *Lauraceae* [Sinitsin, 1980].

The second stage corresponded to the time of florescence of *Hamamelidaceae*, occurred at the end of Palaeogene and at the beginning of the Miocene in non-tropical Asia. In the Eocene, as a result of temperature fall, the flora of Early Palaeogene disappeared giving way to Turgaian flora. The plants forming the nucleus of this flora were concentrated in the southern mountainous regions on the border of Tethysian district. They revealed great tolerance spreading north and south along the mountain ranges and turned out to be capable to survive the drop of the temperature, which occurred at the boundary of Eocene and Oligocene [Meyen, 1987]. Representatives of *Hamamelidaceae* were among such plants. The great majority of them were shrubs ensuring their survival.

According to V. Sinitzin [1980], at the end of Late Oligocene all tropical forms (*Palmae, Proteaceae, Lauraceae, Myrtaceae*) were extinct in Asia, but warm-temperate trees as *Liquidambar, Liriodendron, Nyssa, Rhus, Magnolia*, characteristic for forests of Central-Chinese floristic province at present, continued to exist in the mesophylous forests of Siberia and in North-Eastern part of Asia.

By the end of Palaeogene and during the Miocene the Turgaian flora spread to the south and south-western (in Europe), replacing the retreating subtropical vegetation [Sinitsin, 1980]. After this time the florescence of *Hamamelidaceae* began on the territory of Europe. Here the typical polydominant forests, preserved after the Oligocene, were distributed. Judging from the localities of fossil remains of *Hamamelidaceae*, they were mainly connected with swamp forests. This formation was wide distributed in Central and South-Eastern Europe.

The third stage of development of *Hamamelidaceae*, which corresponds to the period of extinction, began in Early Miocene and proceeded rather rapidly in Asia. In Europe this process began in Late Miocene and proceeded gradually.

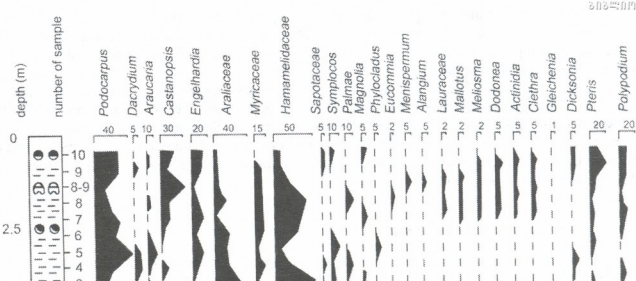


Fig.1. The percentage composition of subtropical plants of the components of lower mountain belt forests on the territory of Western Georgia in Late Miocene.

Conclusion

So, in the history of family *Hamamelidaceae* on the territory of Georgia, Europe and non-tropical Asia three main stages can be distinguished. In the several regions of Eurasia some phases of these stages were nonsynchronous and occupied different stretches of geological time. In Asia and Europe the evolution of *Hamamelidaceae* was closely related with the history of Turgaian flora. In Georgia the development of this family proceeded against the background of evolution of subtropical vegetation, determined the landscape of plain and lower mountain belt. Due to isolate position of Western Georgia, this formation preserved longer, than in other regions of Eurasia and should be traced till the end of Middle Pliocene.

References:

Endress P.K.. *A suprageneric taxonomic classification of the Hamamelidaceae.* Taxon, **38**, 3, 271-376, 1989.

Kolakovskiy A.A., Shakryl A.K. *Sarmatian floras of Abkhazeti.* Proceedings of Sokhumi Botanic Garden, **XXII**, 98-148, 1976 (in Russian).

Meyen S.V. *Basis of Paleobiology.* Moscow, 1987 (in Russian).

Panova L.A., Maligonova E.Yu., Tabachnukova I.P. *Miospore and nannoplankton of Eocene-Oligocene deposits of the north side of Akhaltsikhe depression. Spores and pollen in the deposits of phanerozoic.* Proceedings of VSEGEI, Nov. Ser., **327**, 74-93, 1984 (in Russian).

Ramishvili I.Sh. *Middle Miocene floras of Georgia according to the data of palynological analysis.* Tbilisi, 1982 (in Russian).

Shatilova I., Stuchlik L. *On the history of the development of family Hamamelidaceae.* Palaeontographica, Abt. **B**, Bd. **259**, 235-244, 2001.

Shatilova I.I., Rukhadze L.P., Mchedlishvili N.Sh., Makharadze N.V. *The main stages of development of vegetation and climate of Western Georgia.* Problems of Paleobiology, Tbilisi, **I**, 27-44, 1999 (in Russian).

Sinitsin V.L. *Natural conditions and climate of the territory of USSR in early and middle cenozoic.* Leningrad, 1980 (in Russian).

ოჯახ *Hamamelidaceae*-ს წარმომადგენლები დასავლეთ საქართველოს ნეოგენურში

შატილოვა ი., რუხაძე ლ., მჭედლიშვილი ნ.

საქართველოს ეროვნული მუზეუმი, ლ. დავითაშვილის პალეობიოლოგიის ინსტიტუტი

(მიღებულია 06.11.2006)

რეზიუმე

დღეისათვის დაგროვილია მდიდარი ფაქტიური მასალა ოჯახ *Hamamelidaceae*-ს შესახებ. ამ მცენარეთა ნამარხ ნაშთებს ევროპასა და არატროპიკულ აზიაში ყველაზე ხშირად პოულობენ ოლიგოცენურ, აღრეულ და შუა მიოცენურ ნალექებში. საქართველოში, ძირითადად, მას დასავლეთ ნაწილში, ე.წ. კოლხეთის რეფუგიუმში ამ ოჯახის სხვადასხვა გვარების წარმომადგენლები აღმოჩენილია გვიანმიოცენური (სარმატული, მეოტური) ფლორის შემადგენლობაში. აღნიშნული მასალა ძალზე მნიშვნელოვანია, რადგან იგი ახახავს ოჯახ *Hamamelidaceae*-ს ბატონობის ბოლო ეტაპს ევრაზიის იმ რეგიონში, სადაც დღეს ისინი სრულიად აღარ გვხვდება.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Plant Physiology

THE EFFECT OF BORON, ZINC AND MANGANESE ON THE ACTIVITY OF AMYLASES IN THE SEEDS OF *RAPHANUS SATIVUS, SPINACIA OLERACEA AND CORIANDRUM SATIVUM*

MANGALADZE N., TUTBERIDZE R., KILADZE N.

A. Tsereteli Kutaisi State University

(Received September 18, 2006)

Abstract

Seeds of *Raphanus sativus, Spinacia oleracea and Coriandrum sativum* were processed for 24 hours by 0.02% solutions of $ZnSO_4$, $KMnO_4$ and H_3BO_3. Control seeds were kept in distillate for the same time. To determine amylase activity on the second day of seed germination photocalorimetric method was used. At the seed processing with $ZnSO_4$, $KMnO_4$ and H_3BO_3 it was shown that the activity of α- and αβ-amylases were increased. Boron is suggested to have common positive effect on the amylase activity in seeds of all experimental species.

Key wards: microelements, amylase activity, photocalorimetric method

Introduction

It is well established that microelement deficiency may lead to alimentary diseases of plants [Braun et al., 1962; Katalimov, 1956; Marschner, 1995; Snowball et al., 1980]. Modern authors confirm that microelements are necessary for seed germination, development of vegetative organs, florescence and fruitage. There are some data indicating that microelements take significant role in the formation of anatomical structure of plant organs during the metabolic processes [Szpunar, 2004; Wang et al., 2003; Williams, 2001].

The present work was aimed to examine the role of microelements in the activity of enzymes in the plant cell. Particularly, the effect of boron, zinc and manganese on the activity of amylases in the seeds of *Raphanus sativus, Spinacia oleracea and Coriandrum sativum* was studied.

Materials and Methods

Changes of activities of amylases in the seeds of radish, spinach and coriander treated with solutions containing boron, zinc and manganese were examined. Seeds (300 of each plant) were placed on filtration paper treated with 0.02% solution of microelements for 24 hours. Particularly, seeds (100 ones from every plant) were processed in the $ZnSO_4$, $KMnO_4$ and H_3BO_3 solutions. Control seeds (100 ones from every plant) were set on filtration paper impregnated by distilled water for 24 hours. After processing seeds were placed on Petri dishes. On the second day after appearance of the sprout tips the activities of amylases were determined.

The enzymes were isolated in NaCl solution, incubated in standard starch solution; amount of starch unhydrolyzed by amylases was determined calorimetrically. As a result of hydrolysis and phosphorolysis starch is degraded to monosaccharides during seed germination. α and αβ-amylases, glycoamylase and aminopectin-1-6-glycosidase take part in hydrolysis. During swelling and germination of dry seeds hydrolysis activity of enzymes increases and as a result, starch content decreases and sugar content increases. Activity of amylases is evaluated by hydrolyzed starch in milligrams.

For isolation of amylases 4 g of germinated seeds were put in the mortar, 15 ml of 1% NaCl solution was added and crushed up to homogeneous material. Homogenate was put in the tubes cooled in fridge and centrifuged for 15 min. Substrate was prepared as follows: two tubes (each of 10 ml) were filled with 3 ml of acetate buffer (pH - 5.5) and 3 ml of 2% starch solution, mixed and heated up to 40°C. Further 0.5 ml enzyme preparation was added to one tube, and 5 ml of distilled water - to another one. Content of tubes was heated for 30 min. Then 2 ml of 0.1% of NaCl solution was added to the tubes, mixed and 0.5 ml of solution was taken from each one. Samples were brought into the flasks filled with 25 ml water. 1 ml of 0.1% of NaCl solution, 5 drops of 0.3% of iodine solution were added to the flasks and filled with water up to 50 ml and mixed. Solutions were examined on dyeing in the calorimeter. Red colour filter was used. Amylase activity was calculated by the formula: $A=E_c-E_0 / E_c \times 2.2/60$, where A enzyme activity, E_c and E_0 - optical density of control and experimental solutions, 2.2 coefficient per 1 ml of enzyme solution, 60 - coefficient per 1 mg starch (3 ml of 2% solution corresponds to 60 ml). The obtained data were processed by Student statistical method.

Results and Discussion

As is seen from Fig. 1 activities of α-amylase in the seeds of radish processed with boron and manganese were increased. Difference between activities of the enzyme of the seeds processed by zinc solution and the control is not statistically true. At the same tame processing by boron had stronger influence on the activity of α-amylase compared to manganese. Treatment of the radish seeds with all three experimental solutions increased activity of α-amylase as compared to the control. Processing with the boron caused the strongest effect.

Processing of spinach seeds with all three experimental solutions raised α-amylase activity, though difference with the control is reliable only in the cases of zinc and manganese. At the same time difference in the effects of zinc and manganese is not statistically true.

Processing of coriander seeds with experimental solutions caused statistically true increase in activity of α-amylase in cases of boron and zinc.

As a whole, treatment of radish seeds with H_3BO_3 showed the highest influence on the activity of α-amylase. The effect of $ZnSO_4$ was the biggest in case of spinach seeds, and $KMnO_4$ - in the seeds of radish and spinach.

While processing of radish seeds with experimental solutions activity of αβ-amylase in all three cases compared to control was increased. Differences between effects of boron, zinc and manganese solutions are unreliable. At the same time, activity of αβ-amylase increased much more than activity of α-amylase.

Activity of αβ-amylase in spinach seeds was increased in all three cases, though the strongest, statistically reliable effect was revealed in case of $KMnO_4$. Distinction between effects of H_3BO_3 and $ZnSO_4$ is not significant.

Activity of αβ-amylase in coriander seeds was increased in the cases of H_3BO_3 and $ZnSO_4$. Effect of $KMnO_4$ did not differ reliably from control. The highest influence had $ZnSO_4$.

Activities of amylases play significant role in the process of seed germination, as these enzymes promote degradation of starch and consumption of sugars by plant cells. Obtained results enable to consider that microelements, boron zinc and manganese make favour for increasing amylase activities. At the same time activity of αβ-amylase compared to α-amylase appeared to be more dependant from the presence of boron, zinc and manganese in the cells. Microelements turned to have species-specific character for amylase activities. α-amylase activities in radish and coriander are increased greatly at the effect of boron, but in spinach - at the effect of zinc. Activity of αβ-amylase in radish is equally dependant on boron, zinc and manganese, while in spinach the highest effect revealed - manganese, and in coriander - boron. Effect of boron on amylase activities is nearly similar to the effects of zinc and manganese. In a whole, for germination of seeds of radish, spinach and coriander presawing processing of seeds with the H_3BO_3 solutions should be considered as the most desirable.

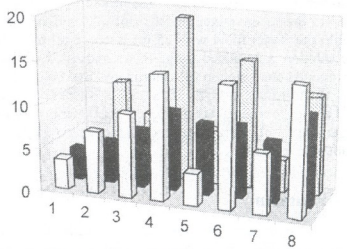


Fig. 1. Activities of amylases in the seeds of *Raphanus sativus, Spinacia oleracea* and *Coriandrum sativum.* 1st row - radish, 2nd row - spinach, 3rd row -coriander; 1 and 2 - control, 3 and 4 - with boron, 5 and 6 with zinc, 7 and 8 - with manganese; in all three cases the first column corresponds to α-amylase and the second column - to αβ-amylase.

References:

Braun T., Zister K., Tanner G. *Physiological consequences of manganese deficiency.* In: Microelemets. M.-L., "Nauka", 1962 (in Russian).

Katalimov M.V. *Microelement content in plants according to species characteristics and features.* In: Microelements in agriculture and medicine. Riga, 1956 (in Russian).

Marschner H. *Mineral nutrition of higher plants.* ed. 2nd, Academic Press, London, 1995.

Snowball L., Robson A.D. Lorenagan. *The effect of cooper on nitrogen fixation in subterranean clover (Trifolium subterraneum).* New Phytol., **85**, 63-72, 1980.

Szpunar J. *Metallomics: a new frontier in analytical chemistry.* Anal. Bioanal. Chem., **204**, 54-56, 2004.

Wang R., Okamoto M., Xing X., Crawford N.M. *Microarray analysis of the nitrate response in Arabidopsis roots and shoots reveal over 1000 rapidly responding genes and new*

linkages to glucose, trehalose-6-phosphate, iron, and sulfate metabolism. Plant Physiol., **132**, 556-567, 2003.

Williams R.J.P. *Chemical selection of elements by cells.* Coordin.Chem.Rev., **216-217**, 583-595, 2001.

ბორის, ცინკის და მანგანუმის ზეგავლენა ამილაზას აქტივობაზე *Raphanus sativus, Spinacia oleracea and Coriandrum sativum* თესლებში

მანგალაძე ნ., თუთბერიძე რ., კილაძე ნ.

ა. წერეთლის ქუთაისის სახელმწიფო უნივერსიტეტი

(მიღებულია 18.09.2006)

რეზიუმე

Raphanus sativus, Spinacia oleracea and Coriandrum sativum-ის თესლები დამუშავებულ იქნა 24 საათის განმავლობაში $ZnSO_4$, $KMnO_4$ და H_3BO_3 0.02% ხსნარებით. საკონტროლო თესლები იგივე დროით თავსდებოდა დისტილირებულ წყალში. განსაზღვრულია ამილაზას აქტივობა თესლის გაღივებიდან მეორე დღეს ფოტოკალორიმეტრული მეთოდით. ნაჩვენებია, რომ $ZnSO_4$, $KMnO_4$ და H_3BO_3 -ით დამუშავებული თესლების α- და αβ- ამილაზას აქტივობები იზრდება. გამოვლენილია, რომ ბორი დადებითად მოქმედებს ამილაზას აქტივობაზე სამივე სახეობის თესლში.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Plant Physiology

INFLUENCE OF SIMULATED ACID RAINS ON PHYSIOLOGICAL INDICES OF WHITE AND RED FORMS OF CABBAGE *(BRASSICA CAPITATA* L.)

RAPAVA L., CHANISHVILI SH., BADRIDZE G., BARBLISHVILI T., ABRAMIDZE S.

N. Ketskhoveli Institute of Botany

(Received October 10, 2006)

Abstract

Effect of pH2.5 water solution of sulphuric acid on seeds and leaves of red and white forms of cabbage (*Brassica capitata* L.) has been studied. Investigated physiological indices – photosynthetic activity of leaves, total activity of growth regulators, dry matter accumulation and water content have revealed higher sensitivity of white form of cabbage to simulated acid precipitations, while red form turned out to be more resistant to acid treatment. This may be due to the high content of anthocyanins in leaves of red cabbage.

Key words: acid rains, photosynthesis, dry matter accumulation, water content, cabbage

Introduction

Increasing of environmental acidity remains one of the most important ecological problems. The influence of acid rains has been mainly studied on woody plants for years [Eds. Muller C. et al., 1999]. During the last period scientists' attention has been focused on investigation of growth and productivity of cultivated plants under the influence of polluted environment [Evans et al., 1986; Hippeli, Elster, 1996].

Phytotoxic effect of polluted environment manifests itself through the changes in plant appearance and dry matter accumulation [Olson et al., 1987]. Therefore, it has been established that plants possess the ability of partial compensation of the primary effect of acid precipitation in the course of development, thus avoiding productivity decrease [Jay et al., 1987]. The opinion exists that plant organism is more resistant to acid pollution at early stages of development [Adams, Hutchinson, 1987].

Proceeding from the above mentioned the objective of our investigation was to study the influence of simulated acid precipitations on seeds and leaves of different varieties of the same species of cultivated plants.

Material and Methods

White and red forms of cabbage (*Brassica capitata* L.) were selected as test objects.

Anthocyanins are known to be one of the principal antioxidant substances, protecting plant organism against different environmental stresses like radiation, O_3, acid rains etc. [Filippovich, et al., 1975]. As the red form of cabbage is rich in substances of anthocyanic nature, presumably it

might be resistant to acid pollution. The comparative study of the effect of acid rains on two forms of cabbage served as a reason of their selection as test objects.

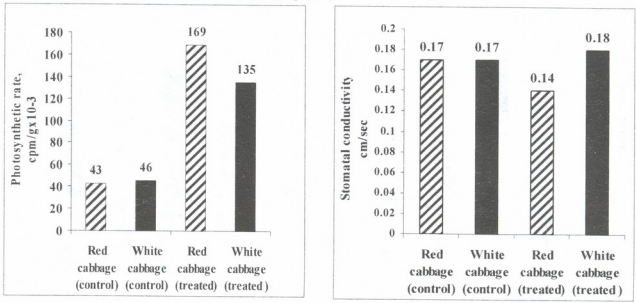
Water solution of sulphuric acid with pH2.5 was used for treating experimental seeds and plants. Cabbage seeds were soaked in acid solution for 24h. In plants, emerged from acid-soaked seeds photosynthetic activity [Voznesensky et al., 1965], stomatal conductivity and total activity of growth regulators (Kefeli, Turetskaya, 1966] were measured.

In other series of experiments leaves of red and white varieties of cabbage plants of the first year of vegetation were sprayed with acid three times with five days interval. Control plants were treated with the same amount of tap water. Material for analysis was taken 10 days after the last spraying.

In addition to the abovementioned indices (photosynthetic activity, stomatal conductivity, total activity of growth regulators) in cabbage plants sprayed with simulated acid rain some indices of water regime and dry matter accumulation were also determined.

Results and Discussion

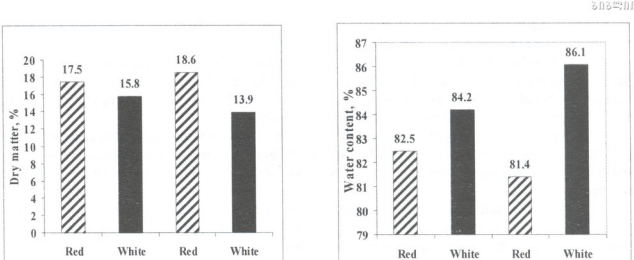
The obtained results show that significant intensification of photosynthetic rate took place in leaves of cabbage plants, emerged from acid-soaked seeds (Fig.1, a). The effect was more pronounced in red form of cabbage. No essential differences were found in stomatal conductivity of control and experimental variants of plant (Fig 1, b).



a b

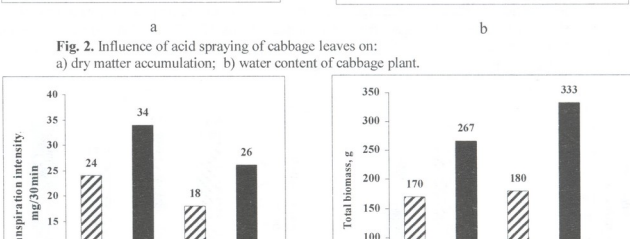
Fig. 1. Influence of acid treatment of cabbage seeds on:
a) photosynthetic activity of leaves; b) leaf stomatal conductivity.

Measurements of dry matter accumulation and water content have revealed that the red form of cabbage was distinguished with higher dry matter and less content of water compared with white form (Fig. 2, a, b). Spraying leaves with acid solution increased dry matter accumulation in red cabbage, while in white form the opposite results were mentioned (Fig 2, a, b). Transpiration index was higher in white cabbage. Spraying with acid solution diminished the index in both forms of cabbage but the effect was more apparent in white cabbage (Fig. 3, a). At the same time the total weight of plants increased (Fig. 3, b).



a b

Fig. 2. Influence of acid spraying of cabbage leaves on:
a) dry matter accumulation; b) water content of cabbage plant.

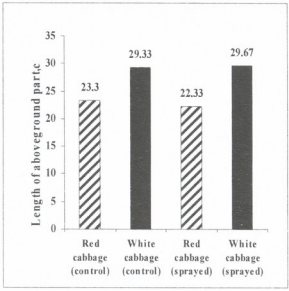


a b

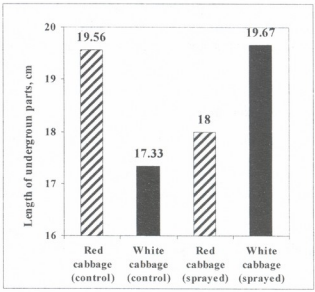
Fig. 3. Influence of acid spraying of cabbage leaves on:
a) transpiration intensity of leaves; b) plant total biomass.

Examination of above and under ground parts of tested plants has shown that as a result of leaf spraying in white cabbage the length of under ground parts increased, while no effect was mentioned on above ground parts (Fig. 4, a, b). In red cabbage spraying with acid caused diminishing of length of both above and under ground parts.

Testing of the total activity of growth regulators manifested essential reduction of the index of growth stimulators in leaves of plants emerged from acid-treated seed (Fig. 5 b; Fig. 6 b). Especially clear effect was mentioned in white cabbage. Opposite effect was revealed in case of plant spraying with acid solution: here significant activation of growth stimulators was detected, which was reflected on changes in biomass accumulation (Fig. 5 c, Fig. 6 c, Fig. 3 b).



a



b

Fig. 4. Influence of acid spraying of cabbage leaves on length of:
a) above- and b) underground parts of a plant.

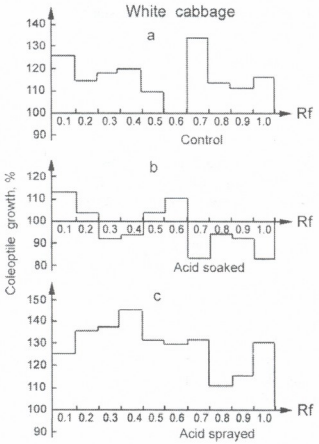


Fig. 5. Total activity of growth regulators in white cabbage leaves

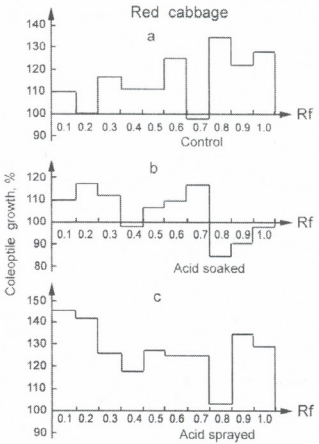


Fig. 6. Total activity of growth regulators in red cabbage leaves

According to the analysis of the obtained data white form of cabbage seems to be more sensitive to simulated acid rains, while red one is more resistant. This fact may be explained by high content of protective substances - anthocyanins in leaves of red cabbage.

References:

Adams C. M., Hutchinson T. C. *New Phytologist,* **106,** 3, 437-456, 1987.

Evans L. S., Sarrantonio M. J., Owen E. M. *New Phytologist,* **103**, 689-693, 1986.

Filippovich J. B., Egorova T. A., Sevast'yanova G. A. *Practical work in general biochemistry.*, M., "Prosveshchenye", 317p., 1975 (in Russian).

Hippeli S., Elster E. F. J. *Plant Physiology*, **148**, 249-257, 1996.

Jay J. S., Osmeloski J., Vamada K., Hellor L. *New Phytologist,* **105**, 1, 139-147, 1987.

Kefeli V.N., Turetskaya R.Kh. *Methods of determination of growth regulators and inhibitors.*, M., 1966 (in Russian).

Ten Years of Monitoring Forest Condition in Europe. Studies of Temporal Development, Spatial Distribution and Impact of Natural and Anthropogenic Stress Factors. Eds. **Muller C.** et al., Brussels: UN/ECE and EC. p.205-275, 1999.

Olson R. L., Winner W. E., Moore L. D. *Environment and Exp. Botany*, **27**, 2, 239-244, 1987.

Voznesensky V.L., Zalensky O.V. and Semikhatova O.A. *Methods of investigation of photosynthesis and respiration of plants.* Moscow, "Nauka", 1965(in Russian).

ხელოვნური მჟავე ნალექებით თესლისა და ფოთლების დამუშავების ბავლენა კომბოსტოს (*Brassica capitata* L.) წითელი და თეთრი ნაირსახეობების ფიზიოლოგიურ მაჩვენებლებზე

რაფავა ლ., ჭანიშვილი შ., ბადრიძე გ., ბარბალიშვილი თ., აბრამიძე ს.

(მიღებულია 10.10.2006)

რეზიუმე

შესწავლილია გოგირდმჟავას pH2.5 წყალხსნარით კომბოსტოს (Brassica capitata L.) წითელი და თეთრი ნაირსახეობების თესლისა და ფოთლების დამუშავების ეფექტი ფოთლების ფოტოსინთეზურ აქტივობაზე, ზრდის რეგულატორების აქტივობაზე, მშრალი ნივთიერების აკუმულაციასა და წყლის შემცველობაზე. კომბოსტოს თეთრი ნაირსახეობის როგორც თესლი, ისე ფოთლები უფრო მგრძნობიარე აღმოჩნდა მჟავე ნალექებით დამუშავებისადმი. კომბოსტოს წითელი ნაირსახეობის შედარებით მაღალი მდგრადობა მჟავათი დამუშავებისადმი საკარაუდოდ ამ ნაირსახეობის ფოთლებში დამცველობითი ფუნქციის მქონე ნივთიერების - ანთოციანების მაღალი შემცველობით უნდა იყოს განპირობებული.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Zoology

EFFICIENCY OF NEMATODE *STEINERNEMA CARPOCAPSAE* SAY AGAINST FALL-PLANTING CUTWORM (*AGRIOTIS SEGETUM* SCHIFF)

KAKULIA I.[1], KOKHIA M.[1], SHEKILADZE K.[2]

[1]*Institute of Zoology*
[2]*Agricultural State University*

(Received May 20, 2006)

Abstract

Bioecology of fall-planting cutworm - *Agriotis segetum* Schiff and the effect of the nematode *Steinernema carpocapsae* Say on the fall-planting cutworm was studied in Sachkhere-Chiatura district, West Georgia, for further usage of obtained results in pest biocontrol. In the field environment pest death rate consists of 68 % in the suspension of the concentration - 150 nematodes/ml, 66.5% - in case of 200 nem/ml, and 67.2% - in case of 250 nem/ml. Effect of entomopathogenic nematodes appeared the highest on the I plot, which was processed with suspension of 150 nem/ml concentration (68%) indicating that efficiency of the nematode *S. carpocapsae* against pests is high even at low concentration of suspension.

Key words: bioecological control, entomopathogenic nematode, nematode suspension.

Introduction

Damage to agriculture caused by pest is great. They obliterate significant part of the yield (nearly 25%) and decrease quality of product. Fall-planting cutworm *Agriotis segetum* Schiff is considered as the most dangerous pest of crops, and particularly of maize. As the maize is one of the main crops in Georgia to study the effect of entomopothogenic nematodes on fall-planting cutworm is the urgent problem.

Fall-planting cutworm is widespread pest in Georgia [Kanchaveli, Supatashvili, 1968]. Its larvae cause great damage to maize, horticultural crops; they injure not only seed germs, but root system too. Larvae cut aslant the root collar of saplings causing their death. Pest larvae of different ages hibernate in the soil. At the beginning of frosts young larvae die, but mature ones make soil bed in early spring and pupate there. Two weeks later nymph flies out of pupae, which generally occur in soil at daytime. Nymph blows 2500 eggs both on weed and cultural plants. Larvae are characterized with negative phototaxis and hence they are hidden in soil. Larvae stage lasts 28-38 days. In Georgia this pest gives 3 generations, and so, their number and respectively damage is great. Damage level caused by fall-planting cutworm belongs to high harmfulness categories.

Materials and Methods

Studies were carried out in the villages Kvatsikhe and Biga of Chiatura region. Plots of maize sowings were located on 10 m from river bank. For experiment 4 plots (3 - experimental and

1 - control) were chosen, each of them of $5m^2$ area. Experiments were carried out in autumn, 2005 (September) and spring, 2006 (May).

Experimental plots were treated with nematode (*Steinernema carpocapsae*) suspension of various concentrations. Suspensions were kept in thermos. Plots were treated early in the morning, at quiet whether conditions (air temperature – 16-20°C). Prepared nematode suspension was sprayed into plants by automax. 3, 8 and 10 days after live and dead larvae were counted both on experimental and control plots according to Franz method [Franz, 1968]. The obtained data are presented in the Table.

Results and Discussion

It was found that pest death rate in I plot was 68%, in II plot – 66.5%, and in III plot – 67.2% (see the table).

Table. Efficiency of nematoda *S. carpocapsae* against fall-planting cutworm (*Agriotis segetum*)

#	Nematode species	Concentration of nematode suspension nem/1ml	Number of larvae	Death rate of larvae			Total number of dead larvae	%
				3 days	8 days	10 days		
1.	*S.carpocapsae*	150 (I plot)	50	10	16	8	34	68
2.	"----------"	200 (II plot)	122	34	32	18	74	66,5
3.	"----------"	250.(III plot)	110	30	32	12	74	67,2
4.	control	clean water	100	-	-	1	1	1

As is seen from the table effect of entomopathogenic nematodes is a bit higher on I plot, which was processed with nematode suspension of 150nem/ml concentration (68%). Results of experiments carried out earlier in laboratory environment showed that while using nematode suspension of 200nem/ml concentration the death rate of larvae was 97%. We consider that the obtained results are caused by the closeness of I plot with the river, and its location in shadow.

Thus, efficiency of nematode *S. carpocapsae* against pests is rather high in spite of low concentration of suspension, and efficiency often depends not on the concentration of suspension but on the experimental conditions.

Obtained data confirm the literature data [Lortkipanidze et al., 2004; Hominick, Reid, 1990] according to which *S. capocapsae* is high-efficient agent for the control of fall-planting cutworm.

References:

Franz J.M. *Zur Berechung des Wirkunsrates einer Microbiologchen Bekamfung von Schädlinskten.* Planzensch., **80**, p. 65-71, 1968.

Hominick W.M., Reid A.P. *Perspectives on entomopathogenic nematology.* In: Entomopathogenic Nematodes in Biological Control (Gaugler R. and Kaya H.K. eds) CRC. Boca Raton FL. p.327-345, 1990.

Kanchaveli G., Supatashvili M. *Forest entomology.* Tbilisi. p. 1-375, 1968 (in Georgian).

Lortkipanidze M., Devdariani Ts., Maglakelidze L., Kokhia M. *Effect of nematodobacterial complex on some pests.* Proceeding of The Institute of Zoology, **XXII**, 45-48, 2004 (in Georgian).

ნემატოდა *STEINERNEMA CARPOCAPSAE* Say -ს ეფექტურობა შემოდგომის ნათესების ხვატარის – *AGRIOTES SEGETUM* Schiff მიმართ

კაკულია ი.[1], კოხია მ.[1], შეყილაძე ქ.[2]

[1]*ზოოლოგიის ინსტიტუტი*
[2]*საქართველოს სასოფლო-სამეურნეო უნივერსიტეტი*

(მიღებულია 20.05.2006)

რეზიუმე

შესწავლილია შემოდგომის ნათესების ხვატარის – *Agriotis segetum* Schiff ბიოეკოლოგია და ნემატოდა *Steinernema capocapsae* Say-ს მოქმედების ეფექტი შემოდგომის ნათესების ხვატარის მიმართ სახნერ-ჰიათურის რეგიონში, შემდგომ მავნე მწერების ბიოკონტროლში გამოსაყენებლად. საველე პირობებში მწერების სიკვდილიანობის პროცენტული მაჩვენებელი 150 ნემატოდა/მლ კონცენტრაციის ნემატოდურ სუსპენზიაში შეადგენდა 68%, 200 ნემ/მლ შემთხვევაში – 66.5%, ხოლო 250 ნემ/მლ-ში კი 67.2%-ია. ენტომოპათოგენური ნემატოდების მოქმედების ეფექტი ყველაზე მაღალი იყო I ნაკვეთზე, რომელიც დამუშავდა 150 ნემ/მლ კონცენტრაციის მქონე სუსპენზიით (68%), რაც იმის მაჩვენებელია, რომ ნემატოდა *S. carpocapsae*-ს ეფექტურობა მავნე მწერების წინააღმდეგ მაღალია მიუხედავად სუსპენზიის დაბალი კონცენტრაციისა.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Zoology

THE ORIBATID MITES (ACARI, ORIBATIDA) OF FLOOD PLAIN ALDER FORESTS OF CENTRAL COLCHIC LOWLAND

*MURVANIDZE M., KVAVADZE E.

Institute of Zoology
**E-mail: makam94@hotmail.com maka.murvanidze@gmail.com*

(Received October 16, 2006)

Abstract

The researches are provided in flooded ecosystems of Central Colchic Lowland on the territories of Colchic National Park, Kobuleti Reserve. These territories are absolutely unique and have great international importance. Oribatid mite fauna of flood plain alder forests in Central Colchic Lowland is studied. 79 species are registered. The ecological analysis shows that in biotops with frequent inundation similar oribatid communities are formed. The diversity of species and high density indicates on a leading role of oribatid mites in processes of decomposition in studied ecosystems.

Key words: Colchic Lowland, *Oribatida*, Ramsar Site, bioindication.

Introduction

The flood plain forests are widely distributed on Colchic Lowland. The main composer of these forests is alder (*Alnus barbata*). Here are also *Pterocarya pterocarpa, Populus canescens, Salix micans* and *S. alba* found. In understorey grow *Rhododendron luteum, Viburnum opulus, Clemantis vitalba, Crataegus pentagina* etc; from liana's group are found - *Smilax excelsa, Hedera colchica, Pericploca graeca* etc [Ketskhoveli, 1959]. The flood plain alder forests are important to maintain the biodiversity of not only Colchic Lowland, but of the whole Black Sea biogeographical region.

In XX century the major part of flood plain forests of Colchic Lowland were drought up and on meliorated soils citrus plantations were mainly cultivated. Currently the floodplain forests and bogs of Colchic Lowland have great international importance, as they make habitats for migrating, hibernating and nesting birds. The flooded habitats have great importance in maintaining the biodiversity of species on the concrete territories [Weigmann, 1997 b]. Currently part of the flooded ecosystems of Colchis are on the territories of Colchic National Park, Kobuleti Reserve and Ramsar Site and are protected by law.

It is known that oribatid mites are one of the main decomposers of organic matters [Ghilarov, Krivolutsky, 1975; Haq, 1987]. They are known as one of the best bioindicators of environment conditions as well [Klausnitzer, 1990, Weigmann, 1991, 1997 b]. Their diversity and density on the studied territory may indicate on condition of these ecosystems.

Information on oribatid mites of Colchic Lowland alder forests is very poor. Lagidze (1981) has registered 29 species of oribatids in bogs with alder forests. Three of them (*Nothrus*

palustris, Minunthozetes pseudofusiger and *Phthiracarus* sp.) were typical for swamped soils and were frequently found. 95 % of registered mites were found in 0-5 cm depth.

Within the animal researches regarding typical biocenoses of Colchic Lowland [Kurashvili, (edt.) 1984], 12 species of oribatid mites were registered on territory of Kulevi in the alder forest. Most of them were found in upper, 0-10 cm layer of soil.

Species that are registered in literature mainly coincide with our data, but this coincidence belongs to such wide distributed species as *Tectocepheus velatus, Platynothrus peltifer, Quadroppia quadricarinata, Scheloribates latipes* and *Minunthozetes pseudofusiger*.

Materials and Methods

Material was taken in June and July 2005. At each site three soil samples (10 cm^3, 0-10 cm depth from surface) were taken and animals were extracted by Tullgren–apparatus. On the studied territory 8 plots were investigated:

1. Anaklia. The left bank of riv. Tikori. Bog with alder forest. N = 42°2,793' E = 41° 37,017'; H = 5m;
2. Anaklia. The left bank of riv. "Didi Gali". Alder forest with *Buxus* understorey.
3. Parpala. The right bank of riv. Churia. Wet alder forest;
4. Imnati. Polydominant wooded bog. N = 42° 08, 283' E = 41° 57,299'; H= 5-7m;
5. Imnati. Kalamona forest. Bog with ash - alder forest. N = 42° 08,190'; E = 41° 96, 980';
6. Kulevi. Alder forest;
7. 6 km from Kulevi. Wet alder forest;
8. Partotskali Lake coast. Bog with alder forest.

As coefficient of faunal likeness between different plots, indicating species identity, Jaccard's coefficient was calculated [Chernov, 1975]. The calculation of community likeness was based on Renkonen's coefficient [Krebs, 1989]. The dominance identities and faunal likeness have been clustered to a dendrogram.

In this investigation only the adult mites were identified and counted.

For the identification of the oribatid mites mainly Ghilarov and Krivolutsky (1975), Balogh and Mahunka (1983), Niedbala (1983) Weigmann (2006) articles were used. For determination of the biogeographycal belonging of oribatid mites work of Subias (2004) was used.

Results and Discussion

79 species of oribatid mites united in 42 families and 50 genera were registered on studied territory (Tab. 1).

Great number of mesophyllic species is presented in the fauna and mainly found in humid ecosystems of Colchic Lowland. Such species are *Mesoplophora pulchra, Microtritia minima, Dissorhina ornata, Eupelops hygrophilus, Achipteria longisetosa, Pergalumna minor* and *Punctoribates mansanoensis*.

No species was registered in every plot. *Steganacarus personatus* was found everywhere except Kulevi (plot 6), and *Protoribates capucinus* was found everywhere except the bank of riv. Churia (plot 7). 34 species were found only in one plot (Tab. 1).

In fauna of oribatid mites predominate widely distributed mites: Palaearctic – 26 species (32 %), Holarctic – 20 species (25 %), Cosmopolits – 16 species (20 %) and European – 8 species (10 %). With less quantity are presented Mediterranean (5 species – 6 %), Caucasian, Endemic (2-2 species – 2-2 %) and Euro-Atlantic (1 species – 1 %) species (Tab. 1).

Table 1. The list of oribatid mites of floodplain alder forests in Central Colchic Lowland and their biogeographycal distribution (+ dominance %; dom. +: % < 1)

#	species	1	2	3	4	5	6	7	8	Distr.
1	*Hypochthonius rufulus* C. L. Koch, 1836	+	+						3	Cosm
2	*Mesoplophora pectinata* Mahunka, 1979				4					Pal
3	*M. pulchra* Sellnick, 1928	+							1	Pal
4	*Phthiracarus assimilis* Niedbala, 1983	+	+							Cauc
5	*Ph. crassus* Niedbala, 1983				+					Md
6	*Ph. ferrugineus* (C. L. Koch, 1841)	7	1	6			3		8	Pal
7	*Ph. globosus* (C. L. Koch, 1841)		11						+	Ho
8	*Ph. incertus* Niedbala, 1983								1	Hol
9	*Ph. lanatus* (Feider & Suciu, 1957)	+			+			9		Eu
10	*Ph. lentulus* (C. L. Koch, 1841)		3		4	16		36		Hol
11	*Ph. ligneus* Willmann, 1931		1				8		14	Hol
12	*Ph. nitens* (Nicolet, 1855)	1	1	3						Pal
13	*Hoplophthiracarus vanderhammeni* Niedbala, 1991	+	1				2	3		Cosm
14	*Steganacarus carinatus* (C. L. Koch, 1841)	+	+			17	2	3		Pal
15	*St. conjuctus* Niedbala, 1983								+	Md
16	St. *personatus* Niedbala, 1983	5	5	70	46	35		3	2	Eu
17	*St. striculus* (C. L. Koch, 1836)		+				4		2	Hol
18	*Microtritia minima* (Berlese, 1904)				+					Cosm
19	*Rhyzotritia ardua* (C. L. Koch, 1841)						3		1	Cosm
20	*Camisia horrida* (Hermann, 1804)								+	Hol
21	*Platinothrus peltifer* (C. L. Koch, 1839)	+	4		+		13	3		Cosm
22	*Nanhermannia nana* (Nicolet, 1855)	+	1			2	8	39	8	Cosm
23	*Belba sculpta* Mihelcic, 1957	+					1			Md
24	*Metabelba pulverulenta* (C. L. Koch, 1840)	+			+					Hol
25	*Arthrodamaeus femoratus* (C. L. Koch, 1840)								+	Pal
26	*Hypocepheus mirabilis* Krivolutski, 1971	1								Eu
27	*Amerobelba decedens*Berlese, 1908	1	2						+	Md
28	*Eremobelba geographica* Berlese, 1908	+	4						2	Eu
29	*Damaeolus ornatissimus* Csiszar, 1962		2							Pal
30	*Gustavia microcephala* (Nicolet, 1855)	2	+	3						Pal
31	*Liacarus brevilamellatus* Mihelcic, 1955	+								Md
32	*L. coracinus* (C. L. Koch, 1841)						1			Pal
33	*Xenillus tegeocranus* (Hermann, 1804)	+	+		+					Pal
34	*Ceratoppia quadridentata* (Haller, 1882)				+	5	1		1	Hol
35	*Carabodes femoralis* (Nicolet, 1855)				1					Pal
36	*C. rugosior* Berlese, 1916	+			3					Hol
37	*Tectocepheus velatus* (Michael, 1880)		+							Cosm
38	*Dissorhina ornata* (Oudemans, 1900)	4	1						1	Hol
39	*Oppia nitens* C. L Koch, 1836	28							7	Hol
40	*Oppiella (Rhinoppia) fallax* (Paoli, 1908)	2	6		+	2			2	Cosm
41	*O. obsoleta* (Paoli, 1908)	+	1							Pal
42	*O.neerlandica* (Oudemans, 1900)	+			+					Hol
43	*O. nova* (Oudemans, 1902)	+			+		16		6	Cosm
44	*O. tuberculata* (Bulanova-Zachvatkina, 1964)		15							Eu
45	*O. unicarnata* (Paoli, 1908)		+						+	Hol
46	*Ramusella insculpta* (Paoli, 1908)		2							Pal
47	*R. mihelcici* (Perez-Inigo, 1965)				1					Pal
48	*Quadroppia michaeli* Mahunka, 1977		+							Pal

49	*Q. quadricarinata* (Michael, 1885)	+								Hol
50	*Suctobelba trigona* (Michael, 1888)	+								Pal
51	*Suctobelbella duplex* (Strenzke, 1950)	+								Pal
52	*S. subcornigera* (Forsslund, 1941)		+							Cosm
53	*Autogneta tragardhi* (Forsslund, 1941)	+							1	Hol
54	*Eupelops acromios* (Hermann, 1804)				+					Pal
55	*E. hygrophilus* (Knuelle, 1954)									Eu
56	*Achipteria longisetosa* Murvanidze & Weigmann, 2003								1	End
57	*Parachipteria georgica* Murvanidze & Weigmann, 2003	13	+						42	End
58	*P. punctata* (Nicolet, 1855)				1					Hol
59	*P. nicoleti* (Berlese, 1883)						14			Hol
60	*Oribatella nigra* Kulijev, 1967		+							Cauc
61	*Galumna obvia* Berlese, 1914						2			Cosm
62	*Pergalumna minor* (Willmann, 1928)		28							Hol
63	*Ceratozetes gracilis* (Michael, 1884)		17	3						Cosm
64	*Edwardzetes edwardsi* (Nicolet, 1855)		+							Eu-Atl
65	*Latilamellobates incisellus* (Kramer, 1897					5				Pal
66	*Sphaerozetes piriformis* (Nicolet, 1885)					7				Pal
67	*S. tricuspidatus* Willmann, 1923						2			Pal
68	*Chamobates kieviensis* Shaldybina, 1980		1						1	Eu
69	*Ch. voigtsi* (Oudemans, 1902)	53	1		2					Pal
70	*Minunthozetes pseudofusiger* (Schweizer, 1956)	+	1		3	2			1	Pal
71	*Punctoribates mansanoensis* (Hammer, 1958)						1			Eu
72	*P. punctum* (C. L. Koch, 1908)		+		17	2				Cosm
73	*Protoribates capucinus* Berlese, 1908	+	2		1	5	20	3	2	Cosm
74	*Scheloribates laevigatus* (C. L. Koch, 1835)				+					Cosm
75	*Sch. latipes* (C. L. Koch, 1840)	+								Pal
76	*Dometorina plantivaga* (Berlese, 1895)				+					Cosm
77	*Oribatula tibialis* (Nicolet, 1855)	1	+		5				2	Hol
78	*Zygoribatula exilis* (Nicolet, 1855)				3	2				Hol
79	*Z. microporosa* (Bulanova-Zachvatkina, 1967)								+	Pal
	total	37	37	6	26	12	17	8	29	
	density 1000 ind/m^2	71	32	17	51	13	50	8	30	

The maximum density of oribatid mites was registered in Anaklia, on the bank of riv. Tikori (71 730 ind/m^2). The density was also high in polydominant wooded bog in Imnati (51 750 ind/m^2) and in alder forest in Kulevi (50 500 ind/m^2). The lower indexes were marked in wet alder forest 6 km from Kulevi (8 250 ind/m^2) (Tab. 1).

High dominance showed *Steganacarus personatus* (70 %, 46 % and 35 % in different sites), *Phthiracarus lentulus* (36%), *Nanhermannia nana* (39%), *Oppia nitens* (28 %), *Parachipteria georgica* (42%), *Pergalumna minor* (28%), *Chamobates voigtsi* (53%) and *Protoribates capucinus* (20%). Dominance value of other species is much less (Tab. 1).

Based on the calculations of faunal likeness and dominance analyses the ecological clusters were build. Three groups are distinguished in cluster of faunal likeness. In the first group there are united mites from Anaklia and Partotskali Lake (plots 1, 2, 8), what is caused by extreme wetness of these plots. Plots 1 and 2 are located on the banks of the river and inundate several times in the year. In the second group oribatid mites of Imnati and 6 km from Kulevi are united (plots 4, 5, 7). Those sites are distinguished by less humidity. The third plot (bank of riv. Churia) is located

a bit higher compared with other plots and inundates rarely. In this plot less number of oribatid mites was registered, what is also a reason for its low likeness with mites of other plots (Fig. 1).

Three groups were divided in cluster of dominance identities as well. The first group includes the dominant species of oribatid mites of riv. Churia and Imnati (plots 3, 4, 5). These plots are territorially close and ecologically similar. In the 3rd plot number of species was low, but their density was high (16 500 ind/m^2) and dominance identities were also high. The second group unites dominant species of Anaklia and Partotskali Lake (plots 1, 2, 8). As it was already mentioned, these plots are ecologically similar. Plots 6 and 7 are grouped together because they are close both, territorially and ecologically (Fig. 2).

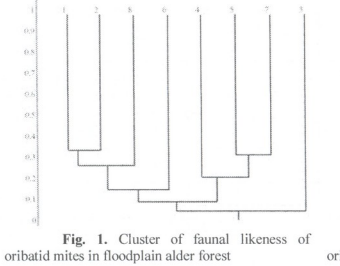


Fig. 1. Cluster of faunal likeness of oribatid mites in floodplain alder forest

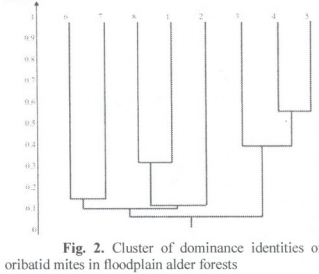


Fig. 2. Cluster of dominance identities of oribatid mites in floodplain alder forests

High diversity and density of oribatid mites are rather unexpected. It is known that mites prefer humid environment, but can not resist high humidity for long period and die because of invasion of microorganisms [Smrz, 1996]. The received results can be explained with low concurrence among the groups which is provoked by extreme conditions and only oribatid mites provide humification and decomposition processes. Low concurrence increases diversity of species of concrete group [Heaney, 2001].

Researches provided in flooded biotops of riv. Oder valley (Germany) showed that oribatid mites adapted to inundation during the winter period and hibernated in early stages; when inundation happened in summer, most of imagoes died [Weigmann, 2004]. In our case the studied territory inundates in spring, summer and autumn. The level of the water decreases in winter, but humidity remains high. We suppose that the main species of oribatid mites of Colchic Lowland alder forests are adapted to the several inundations and fluctuation of their quantity is less discernable during the year.

The species diversity and high density of oribatid mites on the studied territory indicates the unique flora of these ecosystems, active processes of decomposition and soil formation and proves necessity of its international protection.

References:

Balogh J., Mahunka. S. *Primitive Oribatis of Palaearctic Region.* Akademiai Kiado, Budapest,1983.

Chernov Yu. I. *The General Synecological Characteristic of Soil Invertebrates and Methods of Their Analysis.* In: Methods of soil-zoological investigations. Moscow, 160-216, 2002 (in Russian).

Ghilarov M. S., Krivolutsky D. A. *The Identification Keys of Soil Inhabiting Mites. Sarcoptiformes.* "Nauka", Moscow, 1975 (in Russian).

Heaney L. R. *Small Mammal Diversity Along Elevation Gradients in the Phillipines: an Assasment of Patterns and Hypotheses.* Global Ecology and Biogeography. **10,** 15-39, 2001.

Ketskhoveli N. *Vegetation Cover of Georgia.* Metsnierebata akademiis gamomcemloba, Tbilisi, 1959 (in Georgian).

Klausnitzer B. *Ecology of the City Fauna.* "Mir", Moscow, 1990 (in Russian).

Krebs Ch. J. *Ecological Methodology.* Harper & Row Publishers, New York, 1989.

Kurashvili B. (edt). In: *Animal Composition of Typical Biocenoses of Colchic Lowland.* "Mecniereba", pp. 38-43, 1985 (in Georgian).

Lagidze O. I. *Fauna of Podsol-Clay Soils of Colchic Lowland and Their Changes Within Drainage and Cultivating.* PhD abstract, 1981 (in Russian).

Niedbala W. *Les Noueaux Phthiracaridae (Acari, Oribatida) du Caucase.* Ann. Zool. **37,** 62, 1983.

Smrz J. *Some Aspects of the Strategy of Oribatid Mites* (*Oribatida*). Acarology. **IX**. Proceedings, 553-555, 1993.

Subias L. S. *Listado Sistematico, Sinonimico y Biogeografico de los Acaros Oribatidos (Acariformes, Oribatida) del Mundo.* (1758-2002). Graellsia, 60, 2004.

Weigmann G. *Oribatid Communities in Transtcts from Bogs to Forests in Berlin Indicating the Biotope Qualities.* In: F. Dusbabek and V. Bukva (Eds): Modern Acarology, vol. **1**. Academia Prague and SPB Academic Publishing bv. The Hague, 359-364, 1991.

Weigmann G. *Bioindication by means of Isovalent species Groups.* Abh. Ber. Naturkundemus. Goerlitz. **69,** 2, 59-65, 1997a.

Weigmann G. *Die Hornmilben-Fauna (Acari, Oribatida) in Auenböden des Unteren Odertals.* Faun. - Ökol. Mitt. **7,** 319-333, 1997b.

Weigmann G. *Recovery of the Oribatid Mite Community in a Flood Plain After Decline due to Long Time Inundation.* Phytophaga. **XIV,** 201-207, 2004.

Weigmann G. *Hornmilben (Oribatida). Die Tierwelt Deutschlands.* 76 Teil. Goecke, Evers, Kelter Publ. 2006.

კოლხეთის ცენტრალური დაბლობის ჭარბტენიანი მურქნარების ჯავშნიანი ტკიპები (ACARI, ORIBATIDA)

მურვანიძე მ., ყავაძე ე.

ზოოლოგიის ინსტიტუტი

(მიღებულია 16.10.2006)

რეზიუმე

გამოკვლევები ჩატარებულია კოლხეთის ცენტრალური დაბლობის ჭარბტენიან ეკოსისტემებში კოლხეთის ეროვნული პარკის, ქობულეთის ნაკრძალისა და რამსარის საიტის ტერიტორიებზე. ეს ტერიტორიები სრულიად უნიკალურია და აქვთ დიდი საერთაშორისო მნიშვნელობა. შესწავლილია ჯავშნიანი ტკიპების ფაუნა კოლხეთის ცენტრალური დაბლობის ჭარბტენიან მურქნარებში. რეგისტრირებულია 80 სახეობა. ეკოლოგიური ანალიზი გვიჩვენებს, რომ ბიოტოპებში, რომლებიც ხშირად იფარება წყლით, მსგავსი ორიბატიდული ფაუნა ყალიბდება. სახეობათა მრავალფეროვნება და მაღალი სიმჭიდროვე მიუთითებს გამოკვლეულ ეკოსისტემებში დეკომპოზიციის პროცესებში ჯავშნიანი ტკიპების წამყვან როლზე.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Letter to editor

NEW DATA ON FUNGAL DISEASES OF BULB ONION (*ALLIUM CEPA* L.)

*METREVELI I., KUPRASHVILI T.

L. Kanchaveli Institute of Plant Protection
**E-mail: metreveli_i@hotmail.com*

(Received November 6, 2006)

Abstract

During 2004-2006 the samples of bulb onion (*Allium cepa* L.), harvested in different regions of Georgia or imported from abroad have been examined on the presence of fungi. The list of the revealed fungi supplemented with short diagnoses, the information on sites and time of collection are presented. 5 species are registered for the first time on bulb onion in Georgia. One of them - *Epicoccum* sp. is not identified up to species. The descriptions of 4 new species of fungi are given. Among them 2 species was revealed on the local bulb onion cultivars, and 2 - on imported cultivars.

Bulb onion (*Allium cepa* L.), one of the significant vegetable crops of Liliaceae family, is widely cultivated in Georgia. Onion bulbs are used as food and in medicinal purposes. Onion skin is widely exploited as natural dye-staff. Obtaining big yield of high quality bulb onion is of great importance for the country agriculture. The yield and quality of bulb onion is significantly affected by fungal diseases, both in the open ground and during the storage.

The following most common and harmful fungal diseases have been registered on bulb onion – onion mildew (*Peronospora schleideni* Ung.), smut (*Urocystis cepulae* Frost.), onion rust (*P.mixta Fuckel porii* Wint.), grey rot (*Botrytis alli* Munn.) [Khazaradze, 1952; Shoshiashvili, Kirimelashvili, 1952; Zhvania, 1985; Nebulishvili, 1988]. The mentioned pathogens heavily reduce the yield of bulb onion and worsen its quality.

Active trade relations with foreign countries, uncontrolled situation at customs offices have had effect on agrocoenoses of cultivated crops. Reaction of microorganisms and saprophytic fungi, associated with the material introduced from abroad, undergoes changes in the process of competition in a new environment and the organisms often become pathogenic. This may cause wide expansion of the diseases in a new environment. This very phenomenon prompted us to investigate species composition of fungi associated with both local and imported onion bulbs.

The list of new species of fungi registered by us on the material obtained as a result of observations carried out during 2004-2006 in Georgia, is supplemented with short diagnoses and designations of collecting site and time. Information on collecting sites is appended to the species registered in Georgia and corresponding determination keys are cited. Species of fungi are arranged according to E. Muller and V. Lefler [Muller, Leffler, 1995].

The fungi which are registered for the first time in Georgia are indicated by *.

1. *Peronospora destructor* Berk. Casp [7:147] onion mildew.

Tbilisi, Central Market, green onion leaves brought from village Dzalisi Mtskheta District, 11.05.2004; Agrarian market, Marneuli, onion bulbs, 24.07.2005.

2. *Mortierella jenkini* (Smith) Naumov [9:14]

Tbilisi, Didi Dighomi, private commercial greengrocery. Onion bulbs, introduced from Akhalkalaki district, 17.07.2005.

3. *Choanephora conjuncta Couch. (9:88)

Hyphae of the colony are of filiform, of yellowish-grey color. Conidiophores 0.8 cm high and 10-35μm in diameter. Conidia oblong, ovate, pyriform, globose or elliptic, 8-20(24) x 6-10(12) μm. Brown sporangiospores are elliptic or spindle-shaped, 14-26 x 8-15 μm. Stretched mycelial mat of yellowish light grey color develops on onion skin.

Tbilisi, Saburtalo District, Vazha-Pshavela ave., private commercial greengrocery, 04.05.2004.

4. *Urocystis cepulae* Frost (6:517; 7:211; 10:319)

Gori District, village Kheltubani, 11.05.2004; Kareli district, village Khviti, 13.05.2004.

Tbilisi, Didi Dighomi, private vegetabl stall, 15.09.2006.

5. *Puccinia porii* G. Winter [11:161]

Agrarian market, gori, 13.09.2005; Telavi District, village Gulgula, 21.08.2004.

6. *Aspergillus niger v.* Tiegh. [12:547]

Kaspi District, village Kavtiskhevi, private plot, 16.09. 2005.

7. **Glioklad ium vermoeseni (*Biourge)Thom [13:39]

Aerial mycelium of white color later on becomes granular and turns white-pink. Thick twisted mycelial hyphae of 3-6 μm diameter with numerous vacuoles. Conidiophores 100-200 x 4-5 μm, sterigmas usually 8-12 μm, colorless ones usually 4-6 x 3-4 μm. Conidia elliptic in 1-2 mm long chains.

Tbilisi, Agrarian market, private commercial greengrocery, 18.07.2006.

8. *Verticillium lateritium* Berk. [13:79]

Lagodekhi District, village Ninigori., private greengrocery, 13.06.2005.

9. *Botrytis alli* Munn.. [12:179; 13:67; 10:485]

Telavi District, village Vardisubani, 16.08.2004; Tbilisi, Saburtalo District, private greengrocery, 23.10.2005.

10. *Cladosporium herbarum* (Pers.) Link. [12:313]

Kvareli District, village Shilda, Private plot 17.08.2005; Tbilisi, Didube District, Agrarian market, 23.10.2005.

11. *Periconia atra Corda* [12:349]

Samtredia District, Village Ivandidi, 11.07.2005; Tbilisi, agrarian market 01.05.2006.

12. *Alternaria porri* (Ellis) Cif. Ellis [13:177; 10:512]

Kutaisi, agrarian market, 17.03.2004.

13. *Macrosporium parasiticum* Thum. [11:161]

Samtredia, agrarian market, 17.08.2004.

14. *Stemphyllium allii* Oudem. [13:184; 10:537]

Samtredia, agrarian market, 16.08.2006.

15. *Cercospora duddiae* Welles [12:278; 13:112; 11:161; 10:517]

Tbilisi, Didi Dighomi, private plot 17.08.2005.

16. **Heterosporium alli-cepae* Ran. [13:144]

Conidiphores yellowish-brown, 200x7.5-20 μm wide. Conidia yellowish-grey, unicellular, conidia echinulate; pyriform with 1-2 septa. 31-78 x 8.5-18 μm ([13] 32-76 x 9.5-20 μm).

Lagodekhi District, village Ninigori, private plot, 09.2006; Rustavi, private greengrocery, 25.11.2005.

17. *Fusarium oxysporum* Schlecht [13:261]

Lagodekhi District, private plot, 22.12.2005.

18. **Fusarium avenaceum var. anguioides* (Sherb) Bilai [8:182].

Infected bulbs are darkened, in tissue constructing cells and intercellular spaces mycelial hyphae are developed. Mycelial scab of white color occurs between the bulb scales. Macroconida 20-38 x 3.9-5.3 µm. The fungus occurs in the storage conditions.

Gori, agrarian market, 18.06.2004.

19. **Epicoccum sp.*

Lagodekhi District, village Vardisubani, private plot, 13.06.2005.

20. *Colletotrichum circinans* (Berk.) Voglino. (13:196)

Tbilisi, Saburtalo District, agrarian market "Soplis nobati", 3.03.2004.

References:

Bilai V. I. *Fuzarii*, Kiev, 442 p., 1985 (in Russian).

Ellis M. B. *Dematiaceus Hyphomycetes.* Commonwealth Mycological Institute, Kew, Surrey, England, p.608, 1971.

Flora of spore plants of Georgia (Synopsis). Tbilisi, 1986 (in Russian).

Khazaradze E. Proceedings of Inst.Plant Prot. of Georg.SSR, **VIII**, 97-109, 1952 (Georgian).

Khokhryakov M. K., PotlaichukV. I., SemenovA. Ya, ElbakyanM. A. *Guide-book of diseases of agricultures.* Leningrad, p.303, 1984 (in Russian).

Muller E., Leffler V. *Mycology.* Moscow, p.344, 1995 (in Russian).

Nebulishvili E. Cand. (PhD) Thesis, Tbilisi, p.23, 1988 (in Russian).

Pidoplichko N. M., Milko A. A. *Atlas of mucoraceous molds.* Kiev, p.114, 1976 (in Russian).

Pidoplichko N.M. *Parasitic fungi of cultural plants. Imperfect fungi.* Guide-book, vol. **2**., Kiev, p.298, 1977 (in Russian).

Saccardo P. A. *Syllore Fungorum*, vol. **XIII**, Berlin, 1888.

Shoshiashvili I., Kirimelashvili N. Proceedings of Inst.Plant Prot. of Georg.SSR, **VIII**, 109-118, 1952 (in Georgian).

Yatchevskii A. A. *Fungi identification, imperfect fungi.* Vol. **2**, Petrograd, 580 p.1917, (Russian).

Zhvania N. Cand. (PhD) Thesis, Tbilisi, p.23, 1985 (in Russian).

ახალი მონაცემები ხახვის (*Allium cepa* L.) სოკოვანი დაავადებების შესახებ

მეტრეველი ი., კუპრაშვილი თ.

ლ. ყანჩაველის მცენარეთა დაცვის ინსტიტუტი

(მიღებულია 06.11.2006)

რეზიუმე

ნაშრომში წარმოდგენილია 2004-2006 წლებში საქართველოს სხვადასხვა რაიონებში კერძო პირთა და ფერმერთა ნაკვეთებზე მოყვანილი და საზღვარგარეთიდან იმპორტირებული სარეალიზაციო ხახვის ბოლქვებზე გამოვლენილი სოკოების სია მოკლე დიაგნოზით, მოპოვების ადგილისა და დროის ჩვენებით. გამოვლენილია 5 სახეობის სოკო, რომლებიც დღემდე არ იყო რეგისტრირებული ხახვზე საქართველოში. ერთერთი მათგანი - *Epicoccum sp.* არ არის იდენტიფიცირებული სახეობამდე. აღწერილია სოკოს 4 სახეობა, ამათგან 2 სახეობა გამოვლენილია ადგილობრივ, და 2 - სხვა ქვეყნებიდან შემოტანილი ხახვის მასალაზე.

ინსტრუქცია ავტორთათვის

სამეცნიერო ნაშრომი გამოიცემა 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.

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 ფრჩხილებში ჩასმული ავტორის გვარითა და წლით [Lernmark, Hagglof 1981].

მითითებული ლიტერატურა წარმოდგენილი უნდა იყოს შემდეგნაირად:

ჟურნალის შემთხვევაში

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.

წიგნის შემთხვევაში

Kuhn T.S. *The structure of scientific revolutions.* Chicago, IL, Chicago Press, 2000.

Brush S. *Flowing waters or teeming crowds.* In: Mental Models. D. Gentner (Ed.), Chicago IL., Chicago Press, 865-900, 2001.

მასალა რედაქციაში წარმოდგენილი უნდა იყოს ქაღალდზე ამობეჭდილი და დისკეტით (ან CD-ით). წერილი ერთი ფაილით უნდა იყოს შენახული (ცალკე ფაილად შეიძლება ილუსტრაციების წარმოდგენა), ხოლო ფაილის სახელწოდება წერილის პირველი ავტორის გვარს უნდა ატარებდეს.

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. წერილი 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.

ჟურნალის გამოცემა 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.

ნაშრომის 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.

ISSN 1512-2123

საქართველოს მეცნიერებათა აკადემიის

მაცნე

ბიოლოგიის სერია B
Biological series

2007
No. 1
Vol. 5

PROCEEDINGS

of the Georgian Academy of Sciences

ISSN 1512 – 2123

საქართველოს მეცნიერებათა აკადემიის

მაცნე

ბიოლოგიის სერია B
Biological series

2007
No. 1
Vol. 5

PROCEEDINGS

of the Georgian Academy of Sciences

Tbilisi. Georgia
2007

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

Published by NGO "Biomed", 14, L. Gotua st., Tbilisi 0160, Georgia

CONTENTS

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Biochemistry

THE IMPACT OF BIOACTIVE PREPARATIONS ON THE RESISTANCE OF RYEGRASS EXPOSED TO ORGANIC TOXICANTS – BENZENE, 3,4-BENZPYRENE AND TRINITROTOLUENE

*TSULUKIDZE N., BETSIASHVILI M., DZAMUKASHVILI N., SADUNISVILI T., KVESITADZE E.

Durmishidze Institute of Biochemistry and Biotechnology
**E-mail: ninuca_18@yahoo.com*

(Received October 16, 2006)

Abstract

Changes of the activities of glutathione S-transferase, phenoloxidase and peroxidase and of protein content in ryegrass, in response to the effect of organic pollutants – benzene, 3,4- benzpyrene (Bp), trinitrotoluene (TNT) and bioactive preparations, have been studied. It has been established that in most cases, during treatment of ryegrass seedlings with bioactive preparations – Fosnutren and Humiforte, enzyme activities and the protein amount have increased dramatically. Combined effects of benzene, Bp and TNT with bioactive preparations on enzymatic systems of ryegrass have been studied. It has been ascertained that treatment with bioactive preparations caused increase in the activities of glutathione S-transferase, phenoloxidase and peroxidase and protein amount in ryegrass that enhance the plant resistance to organic toxicants.

Key-words: Fosnutren, humiforte, glutathione S-transferase, ryegrass, organic pollutants

Introduction

The most effective remediation, perfect restoration and long-term preservation of chemically contaminated environment are possible by application of phytoremediation technologies. Phytoremediation involves clarification and restoration of chemically contaminated environment by means of plants and microorganisms, which are able to utilize and transform wide range of organic and inorganic toxicants. The plant (with its detoxification potential) capable to utilize the toxicants from – air, soil and water, all three elements of biosphere, is the most efficient mean for restoration of ecologically sound environment [Korte, 2000].

Detoxification process of toxic compounds in plant cell proceeds in three phases: reaction of activation, conjugation and compartmentation [Coleman, 1997]. In the first phase, hydrophilic group is formed in xenobiotic molecules at the expense of enzymatic transformation. As a result, the polarity and reactivity of toxicant molecule significantly increases. Various enzymes, including peroxidases and phenoloxidases, catalyze the activation reaction of xenobiotics [Kvesitadze et al., 2006].

Peroxidase (EC 1.11.1.7) is a widely spread enzyme, found in all green plants, fungi and aerobic bacteria. In a plant cell these enzymes display diverse functions, participate in a number of physiological and detoxification processes: hormonal regulation, lignification, and response to stress conditions, protection of a cell from infections and hydroperoxides. Free radical containing products formed as a result of reactions catalyzed by peroxidase are able to oxidize other compounds, including xenobiotics. Peroxidases of different plants are able to oxidize dimethylaniline, benzpyrene, phenol, aminofluorine and hydroxianysoles [Siegel, 1993].

Phenoloxidase (EC 1.14.18.1) is a copper containing metaloenzyme widely spread in microorganisms, plants, insects and animals. Phenoloxidase actively participate in oxidative degradation of organic toxicants. If the xenobiotic is of phenolic nature, then it is a substrate of phenoloxidase and oxidized in monophenolase and diphenolase reactions. In other cases, xenobiotic oxidation is carried out by co-oxidative mechanism by endogenous phenols [Papunidze et al., 2005].

In the second, conjugation phase xenobiotic or metabolite, activated in the first phase, connect with endogenous hydrophilic molecule. The obtained conjugate is polar and less toxic. In the third phase, compartmentation of inactive water-soluble conjugates takes place in vacuoles or cell wall.

Glutathione S-transferase (GST, EC. 2.5.1.18) is a representative of cytosolic enzymes. This dimmeric enzyme catalyzes bonding of tripeptide glutathione to electrophylic sites of various organic and inorganic molecules. Detoxification of various endo and xenobiotic compounds through this enzyme occurs as a result of covalent bonding between hydrophobic substrate and SH-group of cysteine residue in glutathione [Armstrong, 1997]. The obtained conjugate is less reactive and polar that simplifies its further compartmentation [Coskun, 2002]. In analogue with other detoxification enzymes, some isomers of this enzyme are inducible. Their intracellular level can increase as a result of effect of plant hormones, pathogens and xenobiotics (e.g. herbicides) [Lamoureux, 1989; Mars, 1996].

Frequently, under the effect of toxic compounds enhancement of protein biosynthesis processes in plant cells is observed. Increase of protein amount on one hand promotes balance of protein deficiency, found at conjugation with toxic compounds as a result of protein expenditure, and on the other hand is connected with induction of enzymes participating in detoxification processes [Kvesitadze et al., 2005].

At elaboration of new technologies, great importance is attached to the approaches that enable to regulate ecophysiological characteristics of plants without interfering their genome. In this point of view, the preparations of Spanish firm INAGROSA, particularly Fosnutren and Humiforte are of interest. These preparations are complex of synthesized free amino acids and microelements, applying of which significantly enhance plant productivity, and at the same time, their resistance to different toxicants effects. It should be mentioned that their absorption does not consume plant energy and depend on chlorophyll activity.

Ryegrass is widely applied for planting lawns in cities and along motorways. Consequently, evaluation of the plant resistance to different toxicants, prevalent in the environment and its detoxification capability is of importance.

Some compounds, widely spread in the environment and characterized by high toxicity have been chosen for the experiments. At present, motor transport takes the main place in environmental pollution in developed countries. Exhaust, together with different toxic compounds contains benzene and 3,4-benzpyrene. Compounds containing aromatic rings are extremely toxic and carcinogenic [Samoillloff, 1998; Curfs, 2003]. Among explosives, trinitrotoluene is the most toxic. Hundreds of hectares of contaminated soil remain after hostilities and military exercises [Robidoux, 1999].

The goal of our investigation was to estimate combined impact of bioactive preparations and organic pollutants on ryegrass seedlings in order to increase its phytoremediation capability.

Materials and Methods

The object of the study was ryegrass, seeds of which were swollen and after grown in tap water during 10 days. To induce enzymes the seedlings were placed in solutions containing benzene (0,1mM), 3,4-benzpyrene (0,1mM) and trinitrotoluene (0,1mM), during 5 days. Fosnutren and Humiforte, bioactive preparations were added together with toxicants in concentrations of 5 ml/l. After exposure, roots and leaves were cut and homogenized in a mortar in 0.05M phosphate buffer pH 7.5, in the ratio 1:3. After centrifugation at 12000g, 30 min, the obtained supernatant was studied for enzymes activity.

Glutathione S-transferase activity was determined spectrophotometrically at 340 nm, by measuring the rate of 1-chloro 2,4-dinitrobenzene (CDNB) conjugation with reduced glutathione [Habig, 1974]. Reaction mixture contained 1mM glutathione and 0.1ml enzyme preparation in 0.2M phosphate buffer, pH 6.5, final volume 3ml. Reaction started by addition of 1mM CDNB. Analogous mixture of the same content without enzyme preparations was used as a control.

As a unit of glutathione S-transferase activity the enzyme amount, which catalyzes conjugation of 1mM CDNB with glutathione in 1 min at 25°C is taken.

Peroxidase activity was determined spectrophotometrically at 470 nm according to the rate of guaiacol oxidation [Gregori 1972]. Reaction mixture contained 10mM guaiacol, 0.5 ml H_2O_2 solution (0.3%) and 0.01ml enzyme preparation in 0.05M phosphate buffer, pH 5.4, final volume 3ml.

As a unit of peroxidase activity the enzyme amount catalyzing oxidation of 1mM guaiacol in1 min at 25°C is taken.

Phenoloxidase activity was determined spectrophotometrically at 430 nm according to the rate of pyrocatechine oxidation [Lanzarini, 1972]. Reaction mixture contained 2mM pyrocatechine and 0.1ml enzyme preparation in citrate buffer, pH 4.7, final volume 3ml. Enzyme activity is expressed in ΔE/min.

The enzyme activities were calculated per g of fresh weight and expressed in percents in Tables. Protein was determined by Bradford's method [Bradford, 1974]. As a standard - bovine serum albumin was used.

Results and Discussion

Changes in the activities of glutathione S-transferase, peroxidase and phenoloxidase and protein content have been studied in roots and leaves of ryegrass, treated by biopreparations (Fig. 1). According to the obtained results, glutathione S-transferase and peroxidase activities were enhanced in roots and leaves of plants treated with biopreparations. Increase of peroxidase activity was observed only in plant roots, where the enzyme activity, in comparison with leaves, is higher, according to the literature data as well [Siegel, 1993]. Protein content in treated plants increases significantly both in roots and in leaves. Especially significant increase of protein amount, by 100% was observed in plants treated with Humiforte.

Changes in activities of glutathione S-transferase, peroxidase and phenoloxidase and in protein content were studied at exposure to different toxicants together with bioactive preparations in seedlings of ryegrass. At exposure of seedlings to benzene, significant increase in activities of enzymes and protein content was observed in experiments, when the plant was treated with toxicants together with bioactive preparation (Fig. 2). Different pictures were observed in roots and

leaves. Increase of activities in enzymes participating in detoxification was clearer; presumably, it is connected with treatment method of plants as roots are in direct contact with xenobiotics and with the ability of biopreparations to resist and limit transportation of toxicants to aboveground organs [Kvesitadze, 2005]. On such ability of Fosnutren and Humiforte indicates also significant increase of protein amount in roots, and decrease of protein content in leaves. In that case, obvious difference between stimulating effects of Fosnutren and Humiforte was not observed.

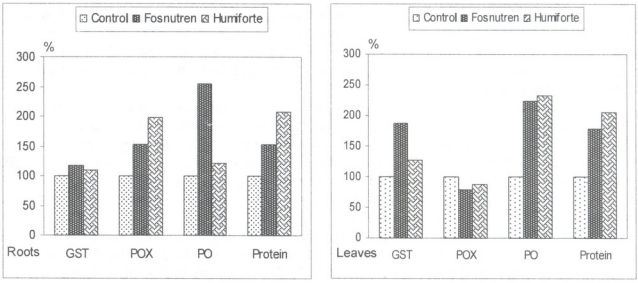


Fig. 1. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to Fosnutren and Humiforte. Exposure time – 5 days. Concentrations of Biopreparations – 5ml/l.

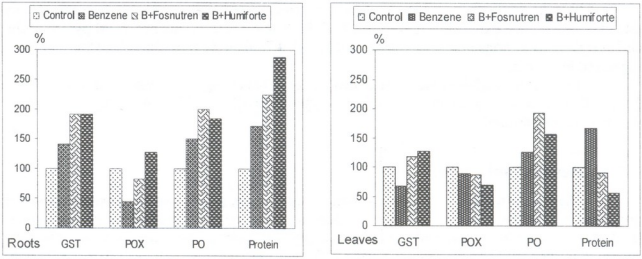


Fig. 2. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to benzene, Fosnutren and Humiforte. Exposure time – 5 days. Concentration of benzene – 0.1mM/l. Concentrations of Biopreparations – 5ml/l.

Changes in enzymes activities in response to exposure to 3,4-benzpyrene (Bp), organic toxicant, have been studied (Fig. 3). As a result of the experiments, it has been established that mainly, glutathione S-tranferase and oxidation enzymes activities increase in roots and leaves of ryegrass, in response to treatment with xenobiotic. Decrease of protein amount in seedlings, treated with toxicants only, indicates on higher toxicity of benzpyrene in comparison with that of benzene.

However, under the influence of bioactive preparations this toxic effect is significantly decreased, while protein content and enzyme activity increased.

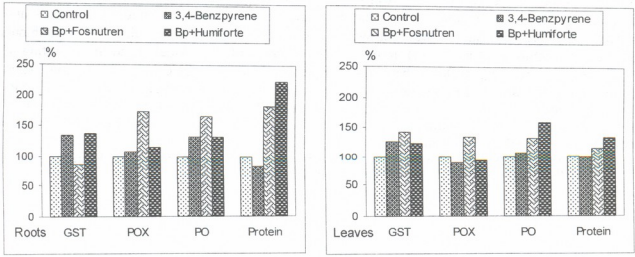


Fig. 3. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to 3,4-benzpyrene (Bp), Fosnutren and Humiforte. Exposure time – 5 days. Concentration of 3,4-benzpyrene – 01,mM/l. Concentrations of Biopreparations – 5ml/l.

The influence of trinitrotoluene (TNT) on seedlings of ryegrass has also been studied (Fig. 4). According to the obtained results, decrease in activity of oxidation enzymes is mainly observed in most cases, connected with another pathway of xenobiotic transformation in the studied plant. Increase of glutathione S-tranferase activity is observed in roots. Presumably, the enzyme renders safe active metabolites formed in the cell under toxicants exposure. Increase of protein content is found both in roots and leaves; it is especially sharp in the presence of bioactive preparations.

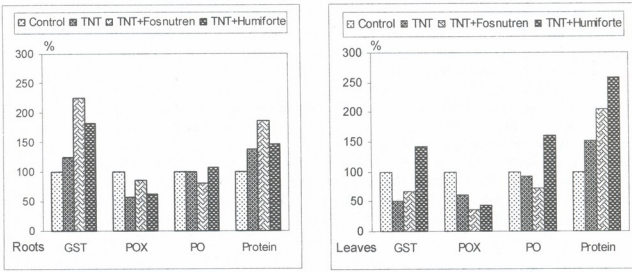


Fig. 4. Changes in the enzyme activities and protein content in roots and leaves of ryegrass exposed to TNT, Fosnutren and Humiforte. Exposure time – 5 days. Concentration of TNT – 0.1 mM/l. Concentrations of biopreparations – 5ml/l.

According to the obtained results, it might be concluded that at treatment with Fosnutren and Humiforte, in most of cases, activities of glutathione S-transferase, phenoloxidase, peroxidase, and protein content enhance dramatically in ryegrass. Application of bioactive preparations together with toxicants increases the plant resistance to xenobiotics as free amino acids and microelements are essential substrate for synthesis of enzymes and/or their substrates (glutathione in case of glutathione S-transferase). Ryegrass, as evergreen plant is widely used in decorating of cities and along motorways. On the base of our experiments, it might be concluded that application of the plant is desirable in combination with bioactive preparations, which significantly improve its phytoremediation capability. It should be also mentioned that the plant genome would not be affected; besides, it is safe for the environment. Phytoremediation itself is another method of nature protection from the dangerous impact of humans.

Acknowledgement: Biologically active preparations – Humiforte and Fosnutren were kindly provided by INAGROSA, Industrias Agrobiologicas, S.A.

References:

Armstrong R. *Structure, catalytic mechanism, and evolution of the glutathione transferases*. Chem. Res. Toxicol. **10**, 2-18, 1997.

Bradford M. *A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the triple of protein dye binding.* Anal. Biochem., **59**, 277-282, 1974.

Coleman J., Mechteld M., Emyr Davis T. *Detoxification of xenobiotics by plants: chemical modification and vacuolar compartmentation.* Trends in Plant Science, **2**, 141-151, 1997.

Coskun G., Zihnioglu F. *Effect of some biocides on glutathione-s-transferase in barley, wheat, lentil and chickpea plants.* Turk J. Biol., **26**, 89-94, 2002.

Cufts D., Beckers L., Godschalk R., Gijbels M., Van Schooten F. Environ. Mol. Mutagen. **42**, 243-249, 2003.

Gregory R. *A rapid assay for peroxidase activity*. Biochem. J., **101**, 582-583, 1966.

Habig W., Jakoby M. *Glutathione S-transferases – the first enzymatic step in mercapturic acid formation*, J. Biol. Chem., **22**, 7130-7139, 1974.

INAGROSA, Industrias Agrobiologicas, S.A.

Korte F., Kvesitadze G., Ugrekhelidze D., Gordeziani M., Khatisashvili G., Buadze O., Zaalishvili G., Coulton F. *Review: Organic toxicants and plants*. Ecotoxicology end Environmental Safety. **47**, 1-26, 2000.

Kvesitadze G., Khatisashvili G., Sadunishvili T., Evstigneeva Z. *Metabolism of Anthropogenic Toxicants in Higher Plants*. Moscow, "Nauka", 2005 (In Russian).

Kvesitadze G., Khatisashvili G., Sadunishvili T., Ramsden J. *Biochemical mechanisms of detoxification in higher plants. Basis of phytoremediation.* Springer, 2006.

Lamoureux G., Rusness D. *The role of glutathione and glutathione-S-transferases in pesticide metabolism, selectivity, and mode of action in plants and insects*. Wiley Interscience, 153-196, 1989.

Lanzarini G., Pifferi P., Samorani A. *Specificty of an o-diphenol oxidase from Punus avium fruits*. Phytochemistry, **11**, 89-94, 1972.

Marrs K.A. *The functions and regulation of glutathione S-transferases in plants.* Plant Mol. Biol. **47**, 127-158, 1996.

Papunidze V., Khatisashvili, G., Sadunishvili T. *Plant for a Healthy Environment*. 2005 (In Georgian).

Robidoux P., Hawari J., Thiboutot S., Ampleman G., Sunahara G. Ecotoxicol. Environ. Saf., **44**, 311-321, 1999.

Samoiloff M. *Benezene toxicity. Benzene and derivatives*. Organic chemistry **3**, 1998.

Siegel B. *Plant peroxidases - an organismic perspective*. J Plant Growth Regul., **12**, 303-312, 1993.

ბიოსასუქების გავლენა კოინდარის მდგრადობაზე ორგანული ტოქსიკანტების – ბენზოლის, 3,4-ბენზპირენისა და ტრინიტროტოლუოლის ზემოქმედებისას

წულუკიძე ნ., ბეციაშვილი მ., ძამუკაშვილი ნ., სადუნიშვილი თ., კვესიტაძე ე.

დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 16.10.2006)

შესწავლილია ფერმენტების – გლუტათიონ S-ტრანსფერაზას, ფენოლოქსიდაზას, პეროქსიდაზას აქტივობებისა და ცილის შემცველობის ცვლილება კოინდარში ორგანული ტოქსიკანტების – ბენზოლის, 3,4-ბენზპირენის, ტრინიტროტოლუოლისა (TNT) და ბიოსასუქების ზემოქმედების საპასუხოდ. დადგენილია, რომ კოინდარის ნაზარდების ბიოაქტიური პრეპარატებით – ფოსფუტრენითა და ჰუმიფორტეთი დამუშავებისას უმრავლეს შემთხვევაში მკვეთრად იმატებს ფერმენტების აქტივობა და ცილის რაოდენობა. შესწავლილია ბენზოლის, 3,4-ბენზპირენის, TNT-ს და ბიოსასუქების ერთობლივი გამოყენების გავლენა მცენარის ფერმენტულ სისტემებზე. დადგენილია, რომ ბიოაქტიური პრეპარატებით დამუშავება იწვევს კოინდარში გლუტათიონ S-ტრანსფერაზას, ფენოლოქსიდაზას, პეროქსიდაზას აქტივობისა და ცილის რაოდენობის მატებას, რაც ზრდის ამ მცენარის გამძლეობას ორგანული ტოქსიკანტების მიმართ.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Biotechnology

EFFECTIVE CONTROLLING OF BACTERIAL SPOT IN TOMATO WITH BACTERIOPHAGE

GHUDUMIDZE N.[1], ALAVIDZE Z.[2], CHKONIA I.[2], ELIASHVILI P.[3], GIORGOBIANI N.[3], SHAPOVALOVA N.[1], MEIPARIANI A.[2], SADUNISHVILI T.[1]

[1]*S. Durmishidze Institute of Biochemistry and Biotechnology*
[2]*G. Eliava Institute of Bacteriophages, Microbiology and Virology*
[3]*L. Kanchaveli Institute of plant protection*

(Received October 16, 2006)

Abstract

Tomato foliages were artificially contaminated with the culture washed out of 4 pathogenic strains of *Xanthomonas vesicatoria*. The disease developed on lower side as well as on upper side of foliages. The disease was caused by infection with pathogenic strains isolated from both damaged foliages and fruits. Spraying foliages with $7x10^6$ p.f.u. of phage just at the moment of infection or 24h later hindered the disease onset, but a week after affection suppressed the disease development. Unlike chemical substances phage did not require to be sprayed twice or several times. Phage does not damage the plant foliages and does not provoke the disease outbreak. Phage application is safe for a human as well as for a plant and it can be used as a salutary agent in dealing with damages caused by Bacterial Spot of sowing areas.

Keywords: Bacterial Spot, tomato foliages, chemical substances, bacteriophage.

Introduction

Bacterial Spot in tomato leaves is caused by phytopathogenic bacteria-*Xanthomonas vesicatoria*. Damage from these diseases may range from a light spotting of the foliage to almost complete defoliation of the plant, with corresponding impacts on the ability of photosynthesis and production potential [Leboeuf et al., 2005], and therefore decreases the yield of tomato crops. When the conditions are optimal for bacterial multiplication (high humidity, 28-32°C.) loss in tomato crops marketable yield can be great.

In order to control the Bacterial Spot the chemical substances are used. Treatment with acid or chlorine may be comparatively effective, if properly manage. But chlorine is inactivated by organic matter and its activity is affected by the pH of solution. Clearly, maintaining the accurate pH is a critical moment for successful disinfection with acid [Leboeuf et al., 2005].

Today the most effective bacteriocide against tomato Bacterial Spot in Georgia still has been copper sulfate spray. But copper sprays are less effective, when spray intervals are extended (7 days or less interval is required). Bacterial populations may show wide resistance to the given solution. Application of high concentrations of copper ions can damage plant tissue leading to a rapid bacteria reproduction. Copper spray may suppress the bacteria on the foliage surfaces, but bacteria located deeply may survive, multiply and cause an outbreak [Leboeuf et al., 2005].

Application of Cixomi and other copper-containing preparations including Kupxodat, Kuprophlo also are used for controlling the tomato Bacterial Spot.

All these compounds in certain amounts may penetrate into a human organism. The prevalence of the toxic substances can hinder such biological processes as growth, development, propagation and in some cases even stop them. Pesticides play a key role in xenobiotics, although, humans have to use pesticides, which finally get into biosphere and humans become a target of their action [Jurin, 2002].

At present the researchers try to study other desinfectants and alternative methods including treatment by microware, sonication and hydrostatic pressure.

One of the encouraging alternative ways in combating against Bacterial Spot in tomato is an application of bacteriophages for treatment purposes, as phages are a specific kind of viruses that attack suitable bacteria to kill pathogenic microorganisms.

The goal of our paper was to show the possibilities of phage application as an alternative tool to chemical substances and a natural biocontrolling agent against Bacterial Spot caused artificially on the tomato foliages.

Materials and Methods

In our experiment was used: *Lycopersicum esculentum* 45 day seedlings; 24h. the culture washed out of 4 various pathogenic strains of *Xanthomonas vesicatoria,* among them 3 strains isolated from the damaged tomato foliages and 1 - from damaged fruit; $7x10^6$ p.f.u. phage mixture of pure lines of mixture of phages isolated from sewages and damaged tomato materials.

The leaves selected for controlling were mechanically damaged by scratching the leaves with a needle. Infection was performed by dropping the bacterial culture onto the mechanically damaged plant leaves [Baltyukova et al., 1968].

The culture was dropped by micropipette. One strain infected some foliages located on various branches of one plant, which were mechanically damaged and added with drops of culture on both, lower and upper sides of foliages; the phage was sprayed by means of special sprayer.

Experiment procession. Tomato seedlings were planted separately into the pots and placed in the greenhouse. For trial 25 days later healthy plants were selected and divided into 5 groups. Control plants were included into I group - a total, 2 plants. The plant foliages of II group were infected with *Xanthomonas vesicatoria* culture - a total, 4 plants. This group was designed for bacterial controlling. The plants, foliages of which were sprayed with phage just at the moment of affection, were included into III group - a total, 4 plants. The plant foliages of IV group were sprayed with phage 24h after affection - a total, 4 plants. The plant foliages of V group were sprayed with phage after a week - a total, 4 plants.

The plants were placed in the thermostat room at constant temperature 28°C. The aeration was performed by airing the room. By day the light was switched on. High humidity was provided by frequent watering and natural evaporation of water from watery vessels.

Results and Discussion.

Observation was carried out within 4 week. It is remarkable, that the signs of bacterial spotting among the plants infected with bacteria were detected only 4 days after affection (II and IV groups) and only on the 5th day the disease developed in a shape of brownish spots on the lower and upper sides of the foliages, infected with pathogenic strains isolated from both damaged foliages and fruits.

During the whole experiment no signs of disease development were observed on the foliages of I group plants (Fig.1). As a result of the disease development on the plant foliages in II group, damaged leaves appear yellow (10 days) (Fig.2). Subsequently all leaves of the branches entirely yellowed. No signs of disease were observed on the plant foliages in III and IV groups within the whole period. After production of Bacterial Spot on the plant leaves in V group the disease was eliminated by phage spray - light spots remained on the infected sites (Fig.4), but the disease did not develop. Evidence of disease was not observed within 3 weeks. Thus, the results of the experiment demonstrated that bacteriophage application succeeded in dealing with bacterial spotting developed on the tomato foliages being infected with *Xanthomonas vesicatoria*. Phage hindered the disease onset, when it was sprayed just at the moment of bacteria affection (Fig.3); after 24h and even after 7 days. It is remarkable, that during the experiment in the case of each variation phage was used only once.

Fig.1. The control plant. (10th day of observation).

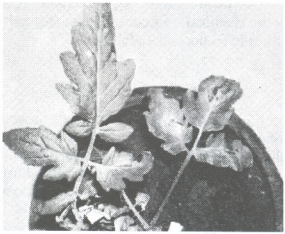


Fig.2. The bacterial-control plant (10th day of observation).

Fig.3. Phage was sprayed at the moment of infection (the 10-th day of observation).

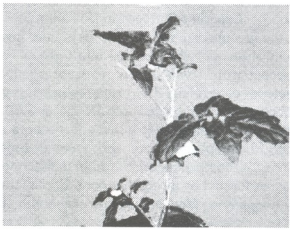


Fig.4. Phage was sprayed after a week of Infection (the 10-th day of observation).

Thus, phages can be used as an effective treatment remedy against Bacterial Spot produced on the tomato foliages. In comparison with chemical substances phages have some advantages: phage preparation is cheaper; when penetrating into any infected site the phage remains there until suitable bacterium exists; phage application does not damage plant organisms even in the case of a high concentration. When getting into a human organism through tomato the phages are safe unlike the chemical substances.

Acknowledgement: This work was carried out with financial assistance of ISTC within the framework of the project NG-1129.

References:

Baltjukova K., Matishevskaja M., Kulikovskaja M., Sudorenko C. *Methods for studying the causative against of plant bacterial disease*. Kiev. Nauka Dumkova. 1968.

Jurin V. *Basis of xenobiology*. Minsk. Novaja Znanie. 2002.

Leboeuf J., Cuppels D., Dick J., Pitblado R., Poewen St., Celetti M. *Bacterial diseases of tomato*; *Bacterial Spot*, *Bacterial Speck*, *Bacterial Canker*. Queen's Printer for Ontario. Factsheet ISSN 1198-712X, 363-365, 2005.

პამიდვრის ბაქტერიული სილაქავის ეფექტური მკურნალობა ბაქტერიოფაგით

დუდუმიძე ნ.[1], ალავიძე ზ.[2], ჭყონია ი.[2], ელიაშვილი ფ.[3], გიორგობიანი ნ.[3], შაპოვალოვა ნ.[1], მეიფარიანი ა.[2], სადუნიშვილი თ.[1]

[1]ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი
[2]გ. ელიავას ბაქტერიოფაგის, მიკრობიოლოგიისა და ვირუსოლოგიის ინსტიტუტი
[3]ლ. ყანჩაველის მცენარეთა დაცვის ინსტიტუტი

(მიღებულია 16.10.2006)

რეზიუმე

Xanthomonas vesicatoria-ს 4 პათოგენური შტამის ჩამონარეცხი კულტურით მოხდა პამიდვრის ფოთლების ხელოვნური ინფიცირება. დაავადება განვითარდა ფოთლების როგორც ქვედა, ასევე ზედა მხრიდან ინფიცირების შემთხვევაში. ფოთლებში დაავადება გამოიწვია როგორც დაავადებული ფოთლებიდან, ასევე ნაყოფიდან გამოყოფილი პათოგენური შტამებით დახნებოვნებამ. $7x10^6$ ტიტრის მქონე ფაგის ფოთლებზე შესხურებამ ინფიცირებისთანავე, ან 24 საათის შემდეგ ხელი შეუშალა დაავადების დაწყებას, ხოლო ინფიცირებიდან 1 კვირის შემდგომმა შესხურებამ შეაჩერა დაავადების განვითარება. ქიმიური საშუალებებისგან განსხვავებით, ფაგის მეორე ან მრავალჯერადი შესხურება არ იყო აუცილებელი, ფაგი არ აზიანებს მცენარის ქსოვილებს და ამით არ ახდენს დაავადების აფეთქების პროვოცირებას. ფაგის გამოყენება უსაფრთხოა როგორც თვით მცენარისთვის, ასევე ადამიანისთვის და შესაძლებელია გამოყენებული იყოს ნათესი ფართობების ბაქტერიული სილაქავისგან გამოწვეული დაზიანების საწინააღმდეგო სამკურნალო საშუალებად.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Botany

INTRASPECIFIC CHEMICAL DIFFERENTIATION OF *URTICA DIOICA* L. GROWING IN GEORGIA

KAVTARADZE N., GVINIASHVILI TS., ALANIA M., KUCHUKHIDZE J.

Kutateladze Institute of Pharmacochemistry

(Received May 15, 2006)

Abstract

Distinctive chemical and morphological features of *Urtica dioica* L. growing in Georgia have been revealed. *Utrica dioica* with red coloration was defined as a new variety - *Utrica dioica* L. var. rubescens Gviniaschvili et Kavtaradze – planta cum anthocyanea.

Key words: *Urtica dioica* L., flavonols, anthocyans.

Introduction

80 species of the genus Urtica L. are spread in temperate and tropical zones of both hemispheres [Mabberley, 1998]. Two species *Urtica dioica* L. and *Urtica urens* L. grow in Georgia [Shkhian, 1975]. They are cosmopolites, follow humans and are met in the conditions of violated natural plant cover. *Urtica dioica* L. is distributed everywhere. It grows in ruderal places, near housings, drafts and banks.

In folk and scientific medicine tincture and decoction of *Urtica dioica* L. is used as hemostatic agent [Budantsev et al., 1985]. Roots and rhizomes of this plant present a raw material for medicinal preparation used at prostate adenoma [Gide-book Vidal, 2002]. Data about phenolic composition of *Urtica dioica* occur in scientific literature. Flavonoids [Chaurasia, 1987], lignins [Kraus et al., 1990], coumarins [Wichtl, 1996] were isolated from *U. dioica*.

Chemical characteristics of *Urtica dioica* growing in Georgia were not studied yet. Phytochemical analysis revealed that qualitative chemical compositions of above-ground parts of *Urtica dioica* growing in various regions of Georgia are different.

The goal of our research was to find out differences of chemical compounds in correlation with morphological characteristics of the plant.

Materials and Methods

U. dioica was studied during 2000-2003. Herbarium material was collected in East Georgia (Kartli, Gori district) and West Georgia (Khobi district, village Alioni) during mass florescence of plant (July-August).

Traditional methods of chemical analysis were used [Alania et al., 2002]. Macromorphological studies of plant were also carried out (Table 2).

Results and Discussion

Our field investigations showed that plants of common *U. dioica* growing throughout the country develop green above-ground parts, but the specimens collected in West Georgia – red ones. Flavonoid glycosides were isolated from those plants and characterized (Table 1).

As is seen from the data given in the table *U. dioica* green is sharply distinct by qualitative flavonoid composition from that of *U. dioica* red. Standard green specimens synthesized only flavonol derivatives - kaempferol and quercetin, but red specimens – flavonols and in addition anthocyans, derivatives of pelargonidin [Kavtaradze et al., 2001; Alania et al., 2002; Kavtaradze et al., 2003].

In order to establish the relation of chemical heterogeneity of material with its morphology we have carried out comparative study of macromorphological characteristics of researched plants (Table 2).

As is seen from the data given in the table specimens are distinguished by the level of pubescence, by the size and level of lignification of stem, by coloration of rhizome, stem, footstalk, rib and lamina.

While field investigations it was noted that red coloration of edges and nodes of rhizomes, from which shoots of stems, footstalks, ribs and lamina are grown, is persisted during the whole vegetation and generational period, which is the very feature that distinguishing it. There are some data in scientific literature that among *U. dioica* occur some specimens, which stems may have coloration caused by presence of anthocyanin pigments changing into brown coloration at the moment of florescence [Medvedev, 1934].

It was revealed that in West Georgia red and green specimens of *U. dioica* could grow together maintaining their characteristic morphology and qualitative chemical composition (Tables 1, 2).

Table 1. Flavonoids isolated from above-ground parts of *Urtica dioica*.

Specimens	Isolated compounds	Empiric formula	Melting temperature, °C	Literature
Green specimens	Flavonols Quercetin (3,5,7,3',4'-pentahydroxiflavon)	$C_{15}H_{10}O_7$	303-306	Kavtaradze et al., 2001
	Isoquercitrin (quercetin-3-O-β-D-glucopyranside)	$C_{21}H_{20}O_{12}$	221-224	" "
	Hyperin (quercetin-3-O-β-D-galactopyranoside)	$C_{21}H_{20}O_{12}$	232-235	" "
	Rutin (quercetin-3-O-β-D-rutinoside)	$C_{27}H_{30}O_{16}$	187-189	" "
	Kaempferol-3-O-tri-galactoside	$C_{33}H_{40}O_{20}$	-	Alania et al., 2004
Red specimens	Anthocyans Pelargonidin-3-xyloside	$C_{20}H_{20}O_9$	260 (with decomposition)	Kavtaradze, Alania, 2003
	Pelargonidin-3-xylobioside	$C_{25}H_{28}O_{14}$	170 (with decomposition)	" "
	Pelargonidin-3-gluco-galactoside	-	-	" "
	Flavonols Nicotiflorin (kaempferol-3-0-β-D-rutinoside)	$C_{27}H_{28}O_{16}$	178-180	Alania et al., 2002
	Rutin (quercetin-3-O-β-D-rutinoside)	$C_{27}H_{30}O_{16}$	187-189	" "

Table 2. Comparative morphological characteristics of *Urtica dioica* L. specimens

Organs and the main morphological features		Characterization	
		Green specimens [Medvedev, 1934]	Red specimens
Rhizome	Form	Tetraquetrous, nodular	Tetraquetrous, nodular
	Coloration	Yellow (sapling) Brownish (mature)	Orange-yellow Edges and nodes of rhizome are red (both, saplings and mature ones)
	Pubescence	Not pubescent	Not pubescent
Stem	Form	Tetraquetrous, rather drooping, thin;	Tetraquetrous, rather drooping, thick
	Height	0.5-1.0 m, in average	1.2-1.8 m, in average
	Butt diameter	5-10 mm, in average	10-17 mm, in average
	Coloration	Green	Red
	Pubescence per 1 cm of the length Number of stinging hairs	10-15	25-35
	Number of usual hairs	300-340	430-460
Leaves	Form and average sizes	Bottom – cordiform-elongated, sharpened; Middle – cordiform-lancet. Upper – nearly lancet (male plants), spear-shaped (mother plants)	Bottom – cordiform-elongated, sharpened; Middle – cordiform-lancet. Upper – nearly lancet (male plants), spear-shaped (mother plants)
	Leaf edges of the bottom layers	Crenate, coarse-toothed	Crenate, coarse-toothed
	Leaf edges of the upper layers	Serrate	Serrate
	Leaf edges of the lateral shoots	Serrate	Serrate
	Coloration of the upper side	Succulent green or dark green	Succulent green with dark reddish coloration
	Coloration of the bottom side	Lighter compared to upper side of lamina	Saplings –succulent red Mature leaves - lighter
	Number of the main ribs	3	3
	Coloration of the ribs	Green	Red
	Pubescence per 1 cm Number of stinging hairs	15-22	38-46
	Number of usual hairs	370-395	525-540
Footstalk	Form	Round, from above - with deep sulcus lengthwise	Round, from above - with deep sulcus lengthwise
	Coloration	Green	Red during the whole vegetative and generational period
	Pubescence per 1 cm Number of stinging hairs	20-55	35-70
	Number of usual hairs	260-380	450-690

Thus, results of chemical analysis and comparative morphological study show morphological and chemical intraspecies heterogeneity of studied specimens of *U. dioica*. Hence,

the plant with red coloration is distinguished as a variety – *Urtica dioica* L., var. rubescens Gviniaschvili et Kavtaradze – planta cum anthocyanea (Georgia, Samegrelo, Khobi district, vil. Alioni, 07.08.2002, near private farm).

References:

Alania M.D., Gvazava L.N., et al. *Phenolic compounds of some plant species growing in Georgia.* Abstracts of the VI symposium on phenolic compounds. Moscow, p. 95, 2004 (in Russian).

Alania M.D., Kemertelidze E.P., Komissarenko H.F. *Flavonoids of some species of Astragalus L. of Georgian flora.* Tbilisi, 2002 (in Russian).

Alania M., Gvazava L., et al. *Polyphenol and cycloartane compounds from some genera of plants of Georgian flora.* Abstracts of the 1st International Pharmaceutical Congress. Tbilisi, p.4, 2002.

Budantsev A.L., Fokina G.A. *Urticaceae Family.* In: Plant resources of USSR: flowering plants, their chemical composition and usage. Leningrad, p. 137-140, 1985 (in Russian).

Chaurasia N., Wichtl M. *Flavonoglycoside aus U. dioica.* Planta Med., **53**, 432-434, 1987.

Gide-Book Vidal. Medicinal Preparations in Russia. Loviisa, "Prostavern Urtica Likvid", p. 710, 2002.

Kavtaradze N.Sh., Alania M.D., Aneli J.N. *Chemical components of U. dioica growing in Georgia.* Chemistry of Natural Compounds, **3**, p.244, 2001 (in Russian).

Kavtaradze N.Sh., Alania M.D. *Anthocyan glycosides of U. dioica.* Chemistry of Natural compounds, **3**, p. 249, 2003 (in Russian).

Kraus R., Spitteler G. *Phenolic compounds from roots of U. dioica.* Phytochem., **29**, 5, 1653-1659, 1990.

Marrberley D.J. *The Plant-Book.* Cambridge University Press, p.55, 1998.

Medvedev P.F. *Utrica of USSR.* Leningrad, 1934 (in Russian).

Shkhian A.S. *Flora of Georgia.* Tbilisi, p.84-85, 1975 (in Georgian).

Wichtl M. *Brennesselblatter. DAB-Kommentar. Wesenschaftliche Erlauterungen zum Deutschen Arzneibuch. Monographin A-C.* Stuttgart-Frankfurt a.m./Eschborn, Bd.II/1, 1996.

საქართველოში გავრცელებული *Urtica dioica* L.-ს შიდასახეობრივი ქიმიური დიფერენციაცია

ქავთარაძე ნ., ღვინიაშვილი ც., ალანია მ., კუჭუხიძე ჯ.

ი. ჭუთათელაძის ფარმაკოქიმიის ინსტიტუტი

(მიღებულია 15.05.2006)

რეზიუმე

შესწავლილია საქართველოს სხვადასხვა რაიონში გავრცელებული *U. dioica*–ის ქიმიური და მორფოლოგიური თავისებურებები. გამოვლენილია *U. dioica* – ის ახალი სახესხვაობა წითელი შეფერილობით - *Urtica dioica* L. var. rubescens Gviniaschvili et Kavtaradze.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Botany

ENDEMIC MEDICINAL PLANTS OF KHEVI (KAZBEGI REGION, GEORGIA)

KHAIKASHVILI KH.

Tbilisi Botanic Garden

(Received August 29, 2006)

Abstract

The paper deals with results of inventory of 18 endemic medicinal plant species, concerning their number, abundance, frequency, and life forms. Degree of threats with extinction of such rare species as *Thymus collinus, Valeriana cardanines* and *V. tilifolia* is in relation to their collection by local population.

Key words: endemic medicinal plants, endangered species, Khevi region, Georgia.

Introduction

Kazbegi region is located on the highest part of the Great Caucasus. Due to contrasting orographic, climate and edaphic conditions the region is characterized by its florocoenotic diversity and richness in endemic species. Thus, 26% of angiosperms of the Khevi flora are endemic, including 6 out of 11 Caucasian endemic genera.

Total number of endemic plants in Khevi flora accounts for 247 species, i.e. 51,1% of high-mountain endemic flora of the Greater Caucasus [Shetekauri, Gagnidze, 2000; Nakhutsrishvili et al., 2005].

It should be noted that among endemic plants of Khevi about 20 species are considered to be used in traditional and officinal medicine from which two species, *Betula raddeana* and *Senecio rhombifolius*, are included in Red Data Book of Georgia (1982). *B. raddeana* is also included in RDB of the former USSR (1984), and IUCN Red List (1997, 2000).

Materials and Methods

Inventory of the abundance of species was done according to Drude's 6-point scale [Drude, 1890]. Frequency has been evaluated after Braun-Blanquet (1951). Spectra of life forms were identified according to Raunkier (1934). Plot size for herbaceous plant was $10m^2$, for woody species - $20m^2$. Experimental plots were chosen randomly within the area of population investigated.

Results and Discussion

6 species (*Betula raddeana* Trautv., *Sorbus caucasigena* Kom. ex Gatsch., *Rosa buschiana* Chrshan., *R. didoensis* Boiss., *R. galuschkoi* Demurova, *R. oplisthes* Boiss.), out of

investigated 18 endemic medicinal plants, belong to woody plants. One species (*Thymus collinus* Bieb.) is semishrub. Herbaceous plants are represented by one bienial species, *Heracleum asperum* (Hoffm.) Bieb. and 10 perennial ones: *Cephalaria gigantea* (Ledeb.) Born., *Galanthus platyphyllus* Traub & Moldenke, *Galega orientalis* Lam., *Potentilla agrimonioides* Bieb., *P. caucasica* Juz., *Senecio rhombifolius* (Adams) Sch. Bip., *Thalictrum buschianum* Kem.-Nath., *Trifolium fontanum* Bobr., *Valeriana cardamines* Bieb., *V. tiliifolia* Troitzk.

The data on species inventory for plants under consideration are given in the following table.

Table. Data on medicinal plant species inventory for 10 m^2 and 20m^2 plots

Plants species	Elev.	Ex.	Steep.	Num.	Abun.	Freq.	Life forms
Betula raddeana	2000	N	45	4	Cop_2	2	Mezophanerophyte
Cephalaria gigantea	1700	-	15	8	Cop_2	1	Hemicryptophyte
Galanthus platyphyllus *	2350	E	20	41	Sp	4	Cryptophyte
Galega orientalis	2000	N	45	9	Cop_1	1	Hemicryptophyte
Heracleum asperum	1600	-	-	11	Cop_2	2	Hemicryptophyte
Potentilla agrimonioides	1800	S	40	-	Cop_2	2	Hemicryptophyte
P. caucasica	1700	-	-	16	Cop_2	2	Hemicryptophyte
Rosa buschiana	2000	N	40	3	Cop_1	2	Nanophanerophyte
R. didoensis	1700	W	20	-	Cop_1	2	Nanophanerophyte
R. galushkoi	1600	-	-	-	Cop_2	2	Nanophanerophyte
R. oplisthes	1900	N	45	-	Cop_2	2	Microphanerophyte
Senecio rhombifolius	1900	N	30	5	Sp	1	Hemicryptophyte
Sorbus caucasigena	2000	E	45	4	Cop_2	2	Microphanerophyte
Thalictrum buschianum	1900	E	40	12	Cop_2	2	Hemicryptophyte
Thymus collinus	1700	E	40	-	Cop_2	3	Chamaephytes
Trifolium fontanum	1700	-	-	-	Cop_2	3	Hemicryptophyte
Valeriana cardamines	2000	N	45	-	Cop_2	1	Hemicryptophyte
V. tiliifolia	2000	N	40	7	Cop_2	1	Hemicryptophyte

Abbreviations and sign: Elev. - Elevation m a.s.l., Ex. - Exposition, Steep. - Steepness in ^{0}, Num. - Number, Abun. - Abundance after Drude (1890), Freq. - Frequency after Braun-Blanquet (1951), Life forms after Raunkier (1934), *Endemic species of Georgia

As is shown in the table *Galanthus platyphyllus* growing on alpine meadows is very rare species. In spite of its rarity in Khevi region it is not utilized as a medicinal plants. Consequently this species is not under threat. According to Miller and others [Miller et al., 2006] *G. platyphyllus* is considered as IUCN Vulnerable category (VU) species.

Next rare endemic medicinal plant in the Khevi region is *Senecio rhombifolius* growing in subalpine tall herbaceous vegetation. Unlike other regions of Georgia in Khevi this wellknown and utilized medicinal plant is not collected for medicinal purposes. Consequently, in spite of scarce resources of this species it is not under threat in this region.

Comparatively abundant species is *Galega orientalis.* In Khevi it occurs in subalpine forests and forest margins, and not utilized by local population for medicinal purposes.

Rosa buschiana occupy dry rock and scree habitats in restricted area in subalpine and alpine belts. Next species, *R. didoensis* occurs in the forest margins and shrublands of upper mountain forests and subalpine belts. It must be noted that only fruits are used for medicinal purpose.

Valeriana cardamines and *V. tiliifolia* are more widely distributed and sufficiently abundant species. Their roots are intensively collected by local population for medicinal purpose. Consequently these species are under serious threat.

Thymus collinus is utilized by local population as medicinal and spice means. Consequently, its resources are gradually diminished.

As a result of cutting number of *Betula raddeana* as well as other components of subalpine forests, viz., *B.litwinowii, B. pendula, Acer trautvetteri* are also little by little diminished.

It is concluded that to maintain endangered medicinal and other species special protective measures need to be taken including strengthening the regime of Kazbegi State Reserve on that part of territory where medicinal plants are represented.

References:

Braun-Blanquet, J. *Planzensoziologie*, 2-e aufl. Wien, 1951, (After Jaroshenko P.A. Geobotany, Moscow, Leningrad, 1961 (in Russ.).

Drude, O. *Handbuch der Pflanzengeographie*, Stuttgart, 1890, (After Jaroshenko P.A. Geobotany, Moscow, Leningrad, 1961 (in Russ.).

IUCN Red List of Threatened Plants. Eddited by K. S. Walter, H.J. Cillet, World Conservation Monitoring Centre, 1997.

IUCN Red List of Threatened Species. Compiled by C. Hilton-Taylor, IUCN, Gland, Switzerland and Cambridge, UK, 2000.

Miller J.S., McCue K., Consiglio T., Stone J., Eristavi M., Sikharulidze Sh., Mikatadze-Pantsulaia Ts., Khutsishvili M. *Endemic medicinal plants of Georgia (Caucasus)*. Tbilisi, 2006.

Nakhutsrishvili G., Abdaladze O., Kikodze A. *Khevi, Kazbegi Region.* Tbilisi, 2005.

Raunkier C. *Life forms of plants and statistical plant geography.* N.Y., 1934 (After Jaroshenko P.A. Geobotany, Moscow, Leningrad, 1961 (in Russ.).

Red Data Book of Georgia. Tbilisi, 1982 (in Georg.).

Red Data Book of USSR, Moscow, 1984 (in Russ.).

Shetekauri Sh., Gagnidze R. *Diversity of high-mountain endemic flora of the Greater Caucasus.* In: Biological and landscape diversity of Georgia. Tbilisi, 151-158, 2000 (in Georg.).

ხეშო (ყაზბეგის რაიონი) გავრცელებული ენდემური სამკურნალო მცენარეები

ხაიკაშვილი ხ.

თბილისის ბოტანიკური ბაღი

(მიღებულია 29.08.2006)

რეზიუმე

სტატია ეხება ხეშის ფლორის 18 სამკურნალო ენდემურ სახეობას. მოცემულია მათი რაოდენობა, სიხშირე, შეხვედრილობა, სახიცოცხლო ფორმების სპექტრი. გამოვლენილია გადაშენების საფრთხის ქვეშ მყოფი ენდემური სახეობები.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Botany

ANALYSIS OF BRYOPSIDA SPECIES ACCORDING TO THE OCCURRENCE FREQUENCY IN THE FOREST BELT OF LAGODEKHI RESERVE

TIGISHVILI K.

N. Ketskhoveli Institute of Botany

(Received May 8, 2006)

Abstract

125 species of Bryopsida are divided into 5 groups according to the occurrence frequency. The highest mark, 5 points was conferred to the 6% of total number of mosses, 4 points - to the 19%, 3 points - to the 28%, 2 points- to the 30%, 1 point - to the 17 %. The obtained results show, that the species with medium and lower occurrence frequency comprise more than half of bryoflora of the lower forest belt. Widely spread species are characterized by the most limited occurrence frequency. Together with this, the comparison of systematical structure of bryoflora of the mentioned belt with the bryoflora of several regions of the East Trans-Caucasus is made on the example of 10 leading families.

Key words: East Trans-Caucasus, bryoflora, leading families.

Introduction

Mosses comprise a significant part of natural plant cover. Natural reserves are the objects for the protection of information resources and the investigation of untouched ecosystems in them is particularly important [Reimers, 1978].

The main objective of our investigation was thorough study of bryoflora of Lagodekhi Reserve. Revelation of the occurrence frequency of bryoflora is connected with many difficulties [Maslovski, 1991]. Despite this we made an attempt to solve the problem on the example of bryoflora of forest belt. The obtained results are of preliminary character.

Materials and Methods

Bryoflora of main ecosystems of forest belt of Lagodekhi Reserve has been studied using itinerary and semistationary methods. Material was taken and treated on sample plots according to the geobotanical method [Neshataev, 1987].

Results and Discussion

On the basis of rich bryological material, obtained in the forest belt of Lagodekhi Reserve (1200 samples) we made an attempt to establish the occurrence frequency of Bryopsida species. According to this feature Bryopsida species of the mentioned region have been divided into 5 main

groups: 1. species, spread in the most of coenoses and ecotopes, characterized with constant occurrence - 5 points; 2. species, which are not spread in the majority of coenoses and ecotopes, but are distinguished with rather high occurrence frequency in the main formations and the most significant ecotopes - 4 points; 3. species, which do not have wide distribution, but are distinguished by the certain occurrence frequency in some ecotopes - 3 points; 4. species, characterized by sporadic distribution and low frequency of occurrence - 2 points; 5. extremely rare species - 1.

Analysis, performed in order to reveal the frequency of occurrence of moss species has shown, that the highest evaluation was conferred to 6% of the total number of bryoflora species or to only 9 species. The following taxons: *Hypnum cupressiforme*, *Leucodon* spec., *Brachythecium rutabulum*, *Brachythecium populeum*, *Neckera besseri*, *Anomodon attenuatus* are characterized by wide distribution and constant frequency of occurrence. The 17 % of the total number of bryoflora species were evaluated by 1 point. Despite the fact, that the material was taken several times, some of these taxons ware found only once. The species *Pottia truncata*, *Phasum cuspidatum*, *Pseudoscleropodium purum*, *Breidleria arcuata*, *Pleurochaete squarrosa* belong to rare species.

The rest species, comprising the bryoflora of forest belt, are positioned between these two extreme groups. The taxons, evaluated by 4 points are not everytypes of the lower forest belt, but are widely spread in favourable econiches. Their number makes 19% of the total number of species. Sometimes, within the limits of synusia, they are distinguished by significant development of the biomass. The epilyths, spread in humid forests of river ravines: *Thamnium alopecurum*, *Ctenidium molluscum*, *Mnium undulatum,* the species, characteristic to Fagetum nudum: *Isothecium myurum*, *Pterigynandrum filiforme;* and *Thuidium philibertii*, characteristic to hornbeam forests and some others belong to such species.

The species, evaluated by 3 and 4 points comprise 30% and 28% of the main list. Taken separately they exceed the number of taxons of other groups and together they make more than half of the bryoflora species. Species, evaluated by 3 points, found only in some coenoses and ecotopes, are represented by the following taxons: *Eurhynchium striatum*, *Euryinchium zetterstedtii*, *Rhynchostegium riparioides*, *Mnium stellare*, *Orthorhichum diaphanum* and others. Distribution of 2-point species is rather limited, though their total number reaches 42. *Weisia controversa*, *Mnium selligeri*, *Bryum bicolor* and others can be listed among such species.

Based on the mentioned data it can be supposed, that bryoflora of the lower forest belt of Lagodekhi Reserve is mainly presented by the species of medium and low frequency of occurrence. The quantity of species of high frequency of occurrence is the most limited. Despite the wide ecological amplitude of mosses, narrow ecological conditions of the environment are the main factors, determining their distribution. Our data are in agreement with those presented in the scientific literature [Maslovski, 1991].

Taxonomic structure of mosses allows to establish the pattern of bryoflora for the mentioned region. Acoording to Tolmachev [Tolmachev, 1974] the pattern of bryoflora is reflected in the best way by the specific composition of ten leading families, which hold the dominant position in bryoflora by the number of species. Composition of bryoflora of the studied region, presented by the leading 10 families is as follows: **Brachytheciaceae** - 22 species, **Mniaceae** - 14 species, **Amblystegiaceae** - 15 species, **Bryaceae** - 13 species, **Pottiaceae** - 11 species, **Dicranaceae** - 9 species, **Grimmiaceae** - 8 species, **Thuidiaceae** - 5 species, **Hypnaceae** - 6 species, **Trichostomaceae** - 4 species. The 104 species or more than half of total bryoflora are united in 10 leading families. The zonal-floristic peculiarities of the region are well reflected by systematical composition of bryoflora. The presence of **Brachytheciaceae**, **Mniaceae** and **Amblystegiaceae** among leading families points to the prevalence of forest landscapes in the mentioned region and its mesophilic character. The families **Grimmiaceae** and **Dicranaceae** serve as the evidence for its mountainous relief. Despite the physicogeographical and vegetative

contrasts, characteristic to the Caucasus, we made an attempt to compare bryoflora of forest belt of Lagodekhi Reserve with that of some regions of East Trans -Caucasus [Chikovani, 1965; Lubarskaya, 1974; Manakian, 1989] (Table 1). It turned out, that 8 families are common for 10 leading families of all regions. This points to the common botanical-geographical character of the mentioned regions.

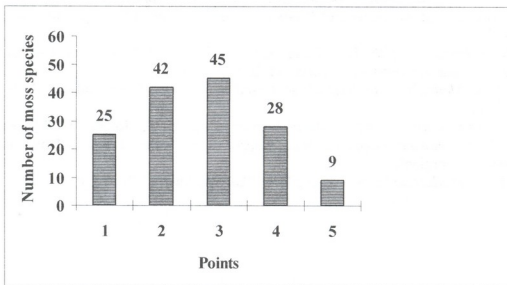


Fig. 1. Groups of Bryopsida species according to the frequency of occurrence.
1 point - 25 species (17% of total); 2 points - 42 species (28% of total); 3 points 45 species (45% of total); 4 points - 28 species (19% of total); 5 points 9 species (6% of total)

Table 1. Comparison of Lagodekhi Reserve forest belt bryoflora with that of some regions of East Trans-Caucasus.

Moss	Lagodekhi reserve		Nukha-Zaqatala		Gombori		NE Armenia		SE Armenia	
Family	Species number	Posi-tion	Species number	Position	Species number	Position	Species number	Position	Species number	Position
Brachytheciaceae	22	1	20	1	13	2	4	7	19	1
Mniaceae	14	3	6	8	6	7	4	8	5	8
Bryaceae	13	4	8	5	7	6	10	2	10	3
Amblystegiaceae	15	2	10	2	9	5	4	9	8	7
Dicranaceae	9	6	5	9	4	10	3	10	-	-
Grimmiaceae	8	7	-	-	5	8	5	5	9	6
Orthotrichaceae	7	-	-	-	10	4	6	4	10	4
Pottiaceae	11	5	9	4	14	-	7	3	9	5
Hypnaceae	6	8	10	3	-	-	-	-	4	9
Thuidiaceae	5	9	7	6	4	9	5	6	3	10
Trichostomaceae	4	10	6	7	13	3	11	1	11	2
Neckeraceae	-	-	5	10	-	-	-	-	-	-

References:

Chikovani N. *Checklist of bryophyte species of Georgia.* Braun-Blanquetia, vol. **34**, Camerino, 2004.

Lubarskaya L. B. *Bryopsida mosses of South-East Part of the Main Caucasus (Azerbaijan).* pp. 180-200, Baku, 1989 (in Russian).

Maslovski O. M. *Analysis of bryoflorotopological structure of Bryoflora complexes of Byelorussian Depression.* In: Advances and Perspectives in Bryology, L'vov, pp. 147-148, 1991 (in Russian).

Manakyan V. A. *Bryopsida of the Main Forest Massifs of Armenian Forest.* Collection of Works of the Armenian Academy of Sciences, **11**, Erevan, 1988.

Neshataev U. N. *Methods of Analysis of Geobotanical Material.* Leningrad, pp.53-98, 1984 (in Russian).

Reimers N. F. *Particularly Protected Natural Territories.* pp. 25-30, Moscow, 1978 (in Russian).

Tigishvili K. T. *Bryoflora of Lagodekhi State Reserve.* Report of the Project, Manuscript, Tbilisi, 1988 (in Georgian).

Tolmachev L. I. *Introduction to the Geography of Plants.* Leningrad, 243 p., 1974.

ღეროფოთლოვანი ხავსების ანალიზი შეხვედრილობის სიხშირის მიხედვით ლაგოდეხის ნაკრძლის ტყის სარტყელში

ტ. ციციშვილი

ნ. კეცხოველის ბოტანიკის ინსტიტუტი

(მიღებულია 08.05.2006)

რეზიუმე

ლაგოდეხის ნაკრძლის ტყის სარტყლის 125 სახეობის ღეროფოთლოვანი ხავსი, შეხვედრილობის სიხშირის მიხედვით, დაყოფილია 5 ჯგუფად. უმაღლესი შეფასება, 5 ბალი, მიიღო ხავსების საერთო რაოდენობის 6%, 4 ბალი - 19%, 3 ბალი - 28%, 2 ბალი - 30%, 1 ბალი -17%. მიღებული მონაცემებიდან ჩანს, რომ ტყის სარტყლის ბრიოფლორის ნახევარზე მეტი წარმოდგენილია საშუალო და უფრო დაბალი შეხვედრილობის სიხშირის მქონე სახეობებით. ფართოდ გავრცელებული სახეობების შეხვედრილობის სიხშირე კი ყველაზე მეტად არის შეზღუდული. აღნიშნული სარტყლის ბრიოფლორის სისტემატიკური სტრუქტურა, 10 წამყვანი ოჯახის მაგალითზე, შედარებულია აღმოსავლეთ ამერიკავკასიის ზოგიერთი რეგიონის ბრიოფლორასთან.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Ecology

THE ORIBATID MITES (ACARI, ORIBATIDA) OF GOMBORI RIDGE BEECH FOREST

ARABULI T.

Institute of Zoology, Chavchavadze ave. 31, 0179, Tbilisi, Georgia

(Received September 20, 2006)

Abstract

119 species of oribatid mites were registered in Gombori Ridge beech forests. Among them 3 species: *Phthiracarus balogi* (Feider, Suciu, 1957), *Tricheremaeus pilosus* Michael, 1988, and *Suctobelba granulate* Hammer, 1952, were recorded for the first time for Georgian fauna. According to faunal likeness distinct groupings of oribatid mites were formed, which is stipulated by distribution of beech forests at various heights. Fauna of Oribatid mites of soil of beech forests is richer than fauna of moss. Changes of number dynamics of oribatids are mutually opposed in those bitopes: while the number is high in moss, it decreases in soil, and visa versa, which is caused by temperature and moisture changes in ecosystem and migration ability of oribatids.

Introduction

Plant cover of Gombori Ridge is characterized with wide diversity and complicated floristic composition, which is caused by its ecological past. 9 zonal types are distinguished across the ridge [Sakhokia, 1960]. The basic component of Fagus forest is beech; beech forests represented by deadcovering and not evergreen subforests prevail. They occur on north exposition, forest edge at the west part of forest is relatively descended and begins from 800-900 m, and at the south exposition, where influence of dryness is higher it begins from 1000 m. On north exposition of south slopes, in humid places the beech comes to 900 m.

The number dynamics of oribatid mites in Georgia has been studied [Darejanashvili, 1965; Murvanidze, 1999; Arabuli, 2003], but number dynamics of oribatid mites inhabited simultaneously in two different stations was not studied yet. Hence, the goal of our work was to study number dynamics of oribatid mites of soil and moss of deadcovering beech forest.

Material and Methods

The material was collected during September 2001 – November 2003. Beech forests of both, south-west and north-east slopes were investigated, particularly: 1. deadcovering beech forest on Akhmeta-Tianeti pass (the utmost north-east of the ridge, near mount Shakhvetila), 2. deadcovering beech forest in Nagubrebi (on north exposition, village Tetrtsklebi), 3. beech forest with azalea subforest in central part of Gombori Ridge (Mount Tsivi, north exposition), 4. deadcovering beech forest (on north exposition of mount Tsivi), and 5. beech forest with deadcovering in Mariamjvari reserve (Table 1.).

Material was collected and worked out according to received methods of soil zoology [Krivolutski, 1973]. At each site 3 samples of soil and moss of 10 cm^3 area were taken. Extracted mites were fixed and temporary preparations were made [Balogh & Mahunka, 1983]. Mite number was counted on 1 m^2. Identification of mites was carried out by special guide-books.

The coefficient of faunal likeness was calculated by Jaccard's formula and the cluster was constructed according to the known method [Krebs, 1989] (Fig.1).

Results and Discussion

119 species united in 65 genera, 44 families and 24 upper families were registered in Gombori Ridge beech forests. Among them 3 species: *Phthiracarus balogi* (Feider, Suciu, 1957), *Tricheremaeus pilosus* Michael, 1988, and *Suctobelba granulate* Hammer, 1952, were recorded for the first time for Georgian fauna.

Our studies have shown that by species diversity of oribatid mites the soil is richer than moss. Among 119 species revealed in beech forests 100 ones were registered in soil, and 54 species – in moss. Characteristic species for every biotope were also studied; it was registered: 65 characteristic species in soil, and 10 – in moss, 44 - common for soil and moss (Table 1).

Table 1. Oribatid mites of Gombori Ridge beech forests

N	species	moss	soil					
			station	single samples				
				1	2	3	4	5
1	*Liochthonius lapponicus* (Tragardh, 1910,)		+					
2	*Hypochthonius rufulus* C.L. Koch, 1835		+					
3	*Hypochthoniella minutissima* (Berlese, 1904)		+	+				
4	*Mesoplophora pulchra* Sellnick, 1928		+		+			
5	*Epilohmannia gigantea* Berlese, 1917		+					
6	*Hoplophthiracarus vanderhammeni* Nied, 1991	+	+	+		+	+	+
7	*Phthiracarus ferrugineus* (C. L. Koch, 1841)	+	+	+		+	+	
8	*Phth. globosus* (C. L. Koch, 1841)		+				+	+
9	*Steganacarus csiszae* Balogh & Mahunka, 1979		+					
10	*St. striculus* (C. L. Koch, 1836)		+				+	
11	*St.serratus* (Feider & Suciu, 1957)		+					
12	*St. spinosus* (Sellnick, 1920)		+		+			
13	*St. (T) carinatus* (C. L. Koch, 1841)	+	+		+	+	+	+
14	*St. (T) phyllophorus* (Berlese, 1904)		+				+	
15	*St. balearicus* Perez-Inigo, 1969	+	+					
16	*St. bicarinatus* Jeleva,1970		+					
17	*Phthiracarus baloghi* (Feider, Suciu, 1957)		+					
18	*Archiphthiracarus. murphyi* (Harding, 1976)		+					
19	*A. lanatus* (Feider, Suciu, 1957)		+					
20	*A. ligneus* (Willmann, 1931)		+				+	+
21	*A.. clemens* (Aoki, 1963)		+					
22	*Rhysotritia ardua* (C.L. Koch, 1841)	+	+	+		+		+
23	*Oribotritia serrata* Feider et Suciu, 1958		+					
24	*Nothrus silvestris* Nicolet, 1855		+			+		
25	*Platynothrus grandjeani* Sitnikova, 1975		+					
26	*Camisia horrida* (Hermann, 1804)	+						
27	*Nanhermannia nana* (Nicolet, 1855)					+		
28	*Hermanniella granulata* (Nicolet, 1855)	+	+	+				

29	*H. punctulata* Berlese, 1908		+	+				
30	*Liodes theleproctus* (Hermann, 1804)	+						
31	*Arthrodamaeus femoratus* (C. L. Koch, 1840)	+	+	+				
32	*Metabelba filippova* Bul.-Zachvatkina, 1965					+		
33	*M. flagelliseta* Bulanova-Zachvatkina, 1965		+	+			+	
34	*M. pulverulenta* (C. L. Koch, 1839)	+	+			+		
35	*Metabelbella macerochaeta* Bul-Zach, 1967					+		
36	*Eupterotegeus ornatissimus* (Berlese, 1908)		+				+	
37	*Amerus troisii* (Berlese, 1883)		+					
38	*Amerobelba decedens* Berlese, 1908		+					
39	*Damaeolus ornatissimus* Csiszar, 1962	+	+				+	
40	*Eremobelba geographica* Berlese, 1908			+		+		
41	*Eremaeus hepaticus* C. L. Koch, 1836	+	+	+		+	+	
42	*E. oblongus* C. L. Koch, 1836	+	+					
43	*E. triglavensis* Tarman, 1958	+						
44	*Tricheremaeus pilosus* Michael, 1888		+					
45	*Zetorchestes microtychus* (Berlese, 1883)		+					
46	*Cultoribula bicultrata* Berlese, 1908		+					
47	*Gustavia microcephala* (Nicolet, 1855)		+					
48	*Adoristes ovatus* (C.L. Koch, 1840)		+					
49	*Liacarus brevilamellatus* Mihelcic, 1955						+	
50	*L. coracinus* (C. L. Koch, 1840)	+	+					
51	*L. tubifer* Djaparidze & Melamud, 1990		+	+		+		
52	*L. lencoranicus* Krivolutsky, 1967		+					
53	*Ceratoppia bipilis* (Hermann, 1804)	+		+				
54	*C. quadridentata* (Haller, 1882)	+						
55	*Carabodes femoralis* (Nicolet, 1855)	+	+					
56	*C. rugosior* Berlese, 1916		+			+	+	+
57	*C. procerus* Weigmann & Murvanidze 2003					+		
58	*Tectocepheus punctulatus* Djaparidze, 1985	+	+		+			
59	*T. sarekensis* (Tragardh, 1910)	+	+					
60	*T. velatus* (Michael, 1880)	+	+	+	+	+		+
61	*Berniniella bicarinata* Paoli, 1908						+	
62	*B. conjucta* (Strenzke, 1951)		+					
63	*B. exempta* (Mihelcic, 1959)		+	+				
64	*B. sigma* (Strenzke, 1951)	+	+	+				
65	*Microppia minus* (Paoli, 1908)		+					
66	*Oppiella maritima* (Willmann, 1928)		+					
67	*O. nasuta* (Moritz, 1965)		+			+		
68	*O. nova* (Oudemans, 1902)	+	+	+	+	+	+	+
69	*O. (R) hygrophila* (Mahunka, 1987)		+					
70	*O. obsoleta* (Paoli, 1908)		+					
71	*O. (R) simifallax* (Subias & Minguez, 1986)						+	
72	*O. (R) subpectinata* (Oudemans, 1900)	+	+	+	+	+	+	+
73	*O. unicarinata* (Paoli, 1908)	+						
74	*Oxyoppioides decipiens* (Paoli, 1908)	+	+					
75	*Ramusella insculpta* (Paoli, 1908)	+	+		+		+	
76	*R. mihelcici* (Perez-Inigo, 1964)			+				
77	*Quadroppia michaeli*, Mahunka, 1977	+	+		+	+		
78	*Q. quadricarinata* (Michael, 1885)	+	+					
79	*Suctobelba granulata* Hammer,1952		+		+	+		

80	*S. trigona* (Michael, 1888)	+	+					
81	*Suctobelbella acutidens* (Forsslund, 1941)	+	+					
82	*S. duplex* (Strenzke, 1950)	+	+					
83	*S. subcornigera* (Forsslund, 1941)		+				+	
84	*Banksinoma lanceolata* (Michael, 1888)					+		
85	*Cymbaeremaeus cymba* (Nicolet, 1885)	+	+			+		
86	*Eupelops acromios* (Hermann, 1804)		+					
87	*E. plicatus* (C. L. Koch, 1836)		+		+			
88	*E. torulosus* (C. L. Koch, 1840)	+		+		+		
89	*Achipteria coleoptrata* (Linne, 1746)		+					
90	*A. nitens* (Nicolet, 1855)	+						
91	*Parachipteria georgica* Murv., Weigm., 2003	+	+	+	+	+		
92	*P. punctata* (Nicolet, 1855)	+	+				+	
93	*P. nicoleti* (Berlese, 1883)						+	+
94	*Umbellozete fuscus* Krivolutsky, 1969	+	+	+		+		+
95	*Acrogalumna longipluma* (Berlese, 1904)		+					
96	*Pilogalumna tenuiclava* (Berlese, 1908)	+	+				+	+
97	*Ceratozetella sellnicki* (Rajski, 1958)	+	+					
98	*Ceratozetes gracilis* (Michael, 1884)	+	+	+	+	+	+	+
99	*C. laticuspidatus* Menke, 1964		+					
100	*C. longicuspidatus* Kulijev, 1962		+					
101	*C. mediocris* Berlese, 1908		+					
102	*Sphaerozetes piriformis* (Nicolet, 1855)	+	+					
103	*Trichoribates trimaculatus* (C.L.Koch, 1836)	+						
104	*T. caucasicus* Shaldybina, 1971	+						
105	*Chamobates caucasicus* Shaldybina, 1969			+				
106	*Ch. interpositus* Pschorn-Walcher, 1953	+						
107	*Ch. voigtsi* (Oudemans, 1902)		+					
108	*Euzetes globosus* (Nicolet, 1855)					+		
109	*Minunthozetes pseudofusiger* (Schwc, 1922)	+	+	+	+	+	+	
110	*Mycobates parmeliae* (Michael, 1884)	+						
111	*M. tridactylus* Willmann, 1929	+	+					
112	*Punctoribates punctum* (C. L. Koch, 1893)	+	+					
113	*Protoribates capucinus* (Berlese, 1908)	+	+					
114	*P. pannonicus* Willmann, 1951					+		
115	*Scheloribates laevigatus* (C. L. Koch, 1836)	+	+	+				
116	*Sch. latipes* (C. L. Koch, 1840)		+					
117	*Oribatula tibialis* (Nicolet, 1855)	+	+	+				+
118	*Phaulopi saakadzei* Djaparidze, 1985	+	+					
119	*Zygoribatula exilis* (Nicolet, 1855)	+	+		+			
	number of species	54	93	26	15	29	24	14

Beech forest with azalea subforest was distinguished by oribatid mite species diversity, where 29 species were recorded, beech forest with deadcovering (Akhmeta-Tianeti pass) – 26 species, and beech forest with deadcovering on north exposition of mount Tsivi – 24 species. It should be mentioned that beech forest with azalea subforest is rich in by species number, as well as by characteristic species (7 characteristic species were registered), which should be caused by diversity of plant cover.

In spite of studying of more or less similar ecosystems, while calculating the faunal likeness of Gombori Ridge oribatid mites, three distinct groupings have been formed (Fig. 1): the highest coefficient of likeness (35%) was noted between deadcovering beech forest located on

north exposition of mount Tsivi and Mariamjvari deadcovering beech forest, which is caused by location of those forests at the same heights. In spite of territorial distance, Akhmeta-Tianeti deadcovering beech forest and beech forest with azalea subforest of mount Tsivi located at higher regions of ridge were grouped together (34%), and beech forest of Nagubrebi placed at lower part of the ridge was absolutely isolated from them. Formation of distinct groupings should be stipulated by special sensitivity of oribatid mites to environmental conditions. As it was mentioned above beech forests on Gombori Ridge begin at various heights due to humidity, which affects the faunal composition of oribatid mites.

Number dynamics was studied in soil and moss simultaneously during 26 months. In September 2001 number of mites in moss consist of 22 500 specimens/m^2, while in soil 5159spec/m^2 were registered. In October insignificant increase of mite number was noted both, in soil and moss, but in November with temperature decrease number of specimens in moss decreased significantly, and at the expense of this their number increased up to 13 657 spec/m^2 in soil, which is caused by migration of mites from the moss to the soil at the beginning of adverse weather conditions. In December mite number was decreased in both biotopes and winter minimums were recorded.

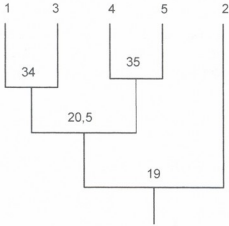


Fig.1. Cluster of faunal likeness of Oribatid mites in Fagus forest.

In January 2002 number of oribatid mites increased both, in soil and moss, and in February winter minimum (4 500 spec/m^2) was fixed in moss. In March and April together with the increase of humidity the minimal number was registered in soil. Just at the beginning of spring mite number increased in moss; increase was continued in April too, and in May maximum number was fixed – 37 000 spec/m^2. As for oribatid mites inhabited in soil, maximum number was revealed in June – 22 324 spec/m^2. In June and July, due to high temperature and dryness, mite number was decreased in moss, but in August it reached maximum – 41 500 spec/m^2.

In October 2002 the picture in soil and moss was contrary: in moss mite number minimum was recorded (4 500 spec/m^2), in soil – maximum (60 990 spec/m^2). In November situation appeared opposite: number increased up to 78 500 spec/m^2 in moss, but in soil – decreased to 16 999 spec/m^2.

In December 2002, as in 2001, number of oribatid mites decreased both in soil and moss, though mite number recorded in 2002 in soil was twice as much than that of moss.

In January 2003 the number of oribatid mites inhabited in soil increased, spring maximum (132 157 spec/m^2) was registered in March, but in moss - later, in May.

In spring 2003 soil mite number revealed considerably high, especially in July – 63 156 spec/m^2. In August and September and especially in October decrease of number was noted. As for oribatids inhabited in moss in June their number was grossly low. In the following months

intensive growth of their number was recorded and in September it reaches maximum – 337 000 spec/m^2.

Thus, as a result of our investigations it was found out that changes of number dynamics of oribatids are mutually opposed in soil and moss: while the number is high in moss, it decreases in soil, and visa versa, indicating the fact that oribatids respond rapidly to temperature and moisture changes and migrate actively towards optimal conditions.

References:

Arabuli T. *The quantiteitive dynamics of Oribatid Mites (Acari, Oribatei) in natural ecosystems (Hornbeam, (Carpinus caucasica Grossh) Forest).* Bulletin of the Georgian Academy of Sciences, **169**, 1, 169-171, 2004.

Balogh J. Mahunka S. *Primitiv Oribatids of the Palearctic region.* Akademiai Kiado. Budapest. p.372, 1983.

Darejanashvili Sh.D. *Seasonal and stational distribution of Acariformes, Oribatei in Tbilisi surroundings.* Bull.Acad.Sci.of Georgian SSR, **XXXVII**, 1, 175-179, 1965 (in Russian).

Krivolutski D.A. *Methods of complex investigation of soils on population density of Oribatid Mites.* In: Methods of Soil-Zoological Studies. Moscow, "Nauka", p. 44-48, 1973 (in Russian).

Krebs CH.J. *Ecological Methodology.* New York, Harper & Row Publisherrs, p.654, 1989.

Murvanidze M. *To the study of quantiteitive dynamics of Oribatid Mites (Acari, Oribatei) in urban conditions.* Bulletin of the Georgian Academy of Sciences, **160**, 2, 377-379, 1999.

გომბორის ქედის წიფლნარების ჯავშნიანი ტკიპები (Acari, Oribatida)

თ. არაბული

ზოოლოგიის ინსტიტუტი

(მიღებულია 20.09.2006)

რეზიუმე

გომბორის ქედზე წიფლნარების გამოკვლევის შედეგებად აღირიცხა ჯავშნიანი ტკიპების 119 სახეობა, რომელთაგან სამი: *Archiphthiracarus balogi, Tricheremaeus pilosus* და *Suctobelba granulata* პირველად იქნა რეგისტრირებული საქართველოს ფაუნისათვის. ჯავშნიანიან ტკიპებს შორის ფაუნისტური მსგავსების მიხედვით წარმოიქმნა განსხვავებული დაჯგუფებები, რაც განპირობებულია ქედზე წიფლნარების სხვადასხვა სიმაღლეზე გავრცელებით. დადგენილია, რომ წიფლნარების ნიადაგის ჯავშნიანი ტკიპების ფაუნა უფრო მდიდარია, ვიდრე ხავსის. ამ ორ ბიოტოპში ტკიპების რიცხოვნობის დინამიკა ურთიერთსაწინააღმდეგოდ იცვლება: როცა ტკიპების რიცხოვნობა მაღალია ხავსში, მაშინ მათი რაოდენობა დაბალია ნიადაგში და პირიქით, რაც ეკოსისტემაში ტენიანობის და ტემპერატურის ცვალებადობით და ორიბატიდების მიგრაციის უნარით არის განპირობებული.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Ecology

DYNAMICS OF CONDITION FACTOR OF VENDACE (*COREGONUS ALBULA* L.) IN THE LAKE PARAVANI

*JAPOSHVILI B.[1], KOKOSADZE T.[2], JAPOSHVILI O.[1]

[1] *Chavchavadze Tbilisi State Univeristy*
[2] *Institute of Zoology*
*Tel: (+ 995 99) 971827, *E-mail: bellaj99@yahoo.com*

(Received September 25, 2006)

Abstract

The dynamics of condition factor among both sexual groups of vendace has been studied for the first time. Condition factor of males and females were compared. The reason of differences is discussed. It was shown, that decrease of condition factor during last years is related with climate changes and global warming.

Key words: *Coregonus albula*, condition factor, Lake Paravani.

Introduction

Lake Paravani is the largest by its surface area among lakes of Georgia (37.5 km^2). It is situated in the Southern Part of Georgia on Javakheti upland on the 2080 m a.s.l. Volume of the lake is 90,8 mln m^3. Lake usually freezes in the second half of the December, while ice layer reaches its maximal thickness in March, very seldom it can be observed in the second half of February. In various years ice layer equaled to 47-73 cm, 80-90 cm, in very cold winter season it was even 1-1.2 m. Melting starts in the third decade of April. At the end of April or in the early May lake tends to be totally free from the ice cover [Barach, 1964, Apkhazava, 1975].

In 30s of 20th century vendace (*Coregonus albula* L.) was introduced in Paravani Lake from the Lagoda Lake (Volkhov hatchery). It was easily adapted to new environment and soon became object for commercial fishing [Demetrashvili, 1960, Japoshvili, 2002]. Data for condition factor for *Coregonus albula* is very poor and insufficient [Demetrashvili, 1960, Japoshvili, 2004, Kokhia, 1961, Peskova, 1960].

Materials and Methods

We have studied dynamics of condition factor for male and female *Coregonus albula* of Paravani Lake during 1999-2005. To calculate condition factor we have used Fulton's equation:

$K=(W/L^3)x100$

K is condition factor, W is the weight of the whole fish weight, L is total length of fish [Nikolskii, 1974, Murphy, Willis, 1996]

Age determinations were based on scales [Pravdin, 1966].

Results and Discussion

During the study males and females of vendace in Paravani Lake were represented by 4 age groups. Studies and calculations have shown that condition factor for females under the age group of 1+ reaches its peak in September. This indicator is increasing between May and September (from 0.65% to 0.81%), later on the indicator reduces and reaches the index observed in May (Fig. 1).

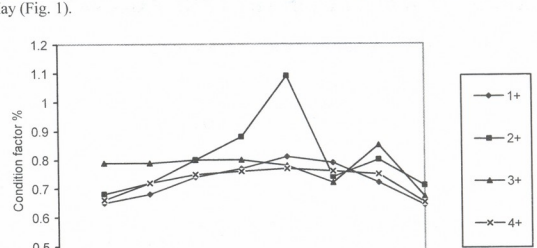


Fig. 1. Dynamics of condition factor of vendace females in Lake Paravani over the months.

In females under the age group of 2+, condition factor rises intensively and reaches its maximum in September -1.09%, while in October the curve is reduced and equals to 0.74%, however before the hatching it is risen a little bit achieving 0.80%, and falling again in December to 0.71%.

Female age group 3+ was observed in the period between May and September. They have shown condition factor of the same value approximately. It is well reflected on the curve that is almost linear. At the beginning of October coefficient equals to 0.72%, in November it reaches 0.85%, while in December it decreases to 0.67%.

In females of the age group 4+ condition factor tends to be 0.66% in May. In summer it rises to 0.77% and in December, following the hatching period the index decreases and equals to 0.65%.

Alteration of condition factor has been studied in males likewise (Fig. 2). Studies have shown that condition factor of males of age group 1+ reaches 0.58% in May, in July it rises to 0.78%, later on the value gradually falls down to 0.70% in September. In November the index increases for a while, equalling to 0.75% and decreases in December to 0.62%. In age group of 2+ coefficient is 0.60% in May. In summer period it increases dramatically and achieves to 0.84% in September. In October condition factor falls to 0.70%. At this period of time the curves are intercrossed for the age groups 1+ and 2+ in males. At the beginning of November it rises and reaches 0.78%, while in December it falls again and equals to 0.64%.

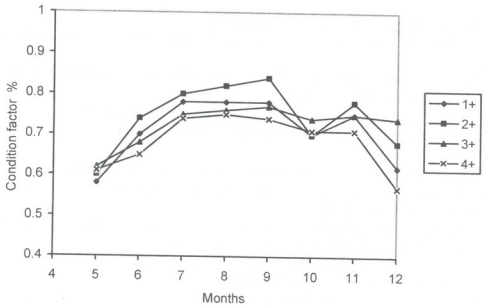


Fig. 2. Dynamics of condition factor of vendace males over the months in Lake Paravani.

Different results were found in males of the age group of 3+ and 4+. In both groups condition factor is rising from May to August. In the following months coefficient falls for the age group 4+, while 3+ age group preserves uniformity and the curve is almost linear.

Conclusions

Studies of females revealed that between May and August-September females of the age group of 1+ and 2+ are characterized with intensified diet. In addition in the age group 2+, before hatching, condition factor is decreased. It should be caused by falling of feeding rate due to the preparatory stage for hatching. This phenomenon for *Coregonus albula* is observed in other European lakes as well. In October-December species under the age group of 2+ and 3+ reflect similarity in nutrition curves caused by active involvement of those age groups in the hatching. Our studies have shown that in comparison with female species condition factor in males is less altered monthly. We suppose that condition factor is more exposed to seasonal changes due to generative synthesis in females.

We have recorded pretty low indices of the condition factor, which can be caused by several reasons, including: illegal catches intensified in the last period. As for discrepancy of our figures with previous data we suggest that differences are caused by altered terms of hatching, ice-cover formation, and warming of the lake as a result of global warming.

References:

Apkhazava I. *Lakes of Georgia.* Tbilisi, "Metsniereba", 1975.

Barach G. *Lakes of Georgia and their importance for fisheries.* Tbilisi, "Sabchota Sakartvelo", 1964.

Demetrashvili M. *For study of biology of lake Paravani vendace.* Bulletin of the Georgian Academy of sciences, **XXV**, N.1, 65-70, 1960.

Japoshvili B. *Results of visual observation on gonadogenesis of Vendace (Coregonus albula L.) in conditions of Paravani Lake.* Bull. of the Georgian Academy of sciences, **166**, 3, 591-594, 2002.

Japoshvili B. *Some Biological and morphometric characteristics of a vendace (Coregonus albul aL.) of the Lake Paravani.* Proceedings of the Georgian academy of Sciences, Biological series B, **2**, 1-2, 97-100, 2004.

Kokhia A. *Materials of diet of fishes of Paravani Lake.* Proceedings of the institute of zoology, Georgian Academy of Sciences, **XVIII,** 85-94, 1961.

Murphy B.R., Willis D. W. *Fisheries Techniques*. Second edition, American Fisheries society, Bethesda, Maryland, USA, XX, pp732, 1996.

Nikolski G. *Ecology of Fishes*. Moscow, "Visshaya Shkola", 1974.

Peskova O. Biology of vendace from Paravani and Sagamo lakes. Proceedings of scientific-research fishery station of Georgia, vol. **V**, 3-37, 1960.

Pravdin I. *Leading on fish study.* M., "Pishchevaya promishlennost", 373, 1966.

ევროპული ჭაფალას (*Coregonus albula* L.) ნაკვებობის კოეფიციენტის დინამიკა ფარავნის ტბაში

ჯაფოშვილი ბ.[1], ქოქოსაძე თ.[2], ჯაფოშვილი, ო.[1]

[1] ი. ჭავჭავაძის თბილისის სახელმწიფო უნივერსიტეტი
[2] ზოოლოგიის ინსტიტუტი

(მიღებულია 25.09.2006)

რეზიუმე

ევროპული ჭაფალას ორივე სქესის წარმომადგენლებში პირველადაა შესწავლილი ნაკვებობის კოეფიციენტის დინამიკა. შედარებულია მდედრებისა და მამრების ნაკვებობის კოეფიციენტი. განხილულია განსხვავებების მიზეზები. ნაჩვენებია, რომ ბოლო წლებში ნაკვებობის კოეფიციენტის დაცემა კლიმატურ ცვლილებებთან და გლობალურ დათბობასთანაა დაკავშირებული.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Entomology

NEW SPECIES OF MERMITHID *HEXAMERMIS DECEMLINEATAE* SP.N. (NEMATODA, MERMITHIDAE) FROM COLORADO BEETLE

GORGADZE O., LORTKIPANIDZE M., KOKHIA M., MELASHVILI N., KUCHAVA M.

Institute of Zoology

(Received October 9, 2006)

Abstract

The paper deals with the description of the new species of mermithid *H. decemlineatae* sp.n. The measurements of adult female, male and post parasitic larvae are given. Host: Colorado beetle (*Leptinotarsa decemlineata* Say). Parasitic and postparasitic mermithid larvae are revealed in the body of beetle, but adult females and males were recorded in the soil. The minimal number of mermithid larvae in each beetle was 1 specimen, and maximal - 30 specimens. In the host the number of parasitic larvae amounts to 1-3 specimens. In natural environment 47.5% of the beetles and 64.5% of larvae were infected with mermithid larvae. Nematode is localized in adipose tissue of the beetle and larvae.

Key words: parasitic and postparasitic nematodes, anatomical and morphological studies, *Hexamermis stepposis, Hexamermis angusta.*

Introduction

Colorado beetle *Leptinotarsa decemlineata Say* (Coleoptera) belongs to the Chrysomelidae family. It is harmful pest of potato. This pest was introduced from North America and was spread in most territories of Europe and Asia [Briantsev, 1966].

According to the studies carried out in biocenosis of potato sowings it was found out that those organisms (nematodes, bugs, carabus, ladybirds), which significantly reduce number of Colorado beetle, were consequently adapted on them. In this way entomopathogenic nematodes of the Mermithidae family are especially significant. Those nematodes in humid conditions can infest beetle, as well as larvae of Colorado beetle, and stimulate their death with 80-95% rate [Mishachkov, 1980]. Due to this fact mermithids appear to be perspective control agents against pests [Ipatieva, Pimenova, 1985; Rubtsov, 1978].

The goal of our work was to study nematodes of Colorado beetles distributed in some regions of East Georgia.

Materials and Methods

To study parasitic nematodes of Colorado beetle and its larvae potato sowings of private farms of mountain regions of East Georgia were researched. Places of collection were: villages Thesami, Ghulelebi, Trani, (Mtskheta-Mtianeti region).

665 specimens of the beetle and 1225 specimens of its larvae were dissected using Pavlovski method [Pavlovski, 1957]. 511 specimens of parasitic and postparasitic nematodes of one species were revealed in beetles and larvae, but the adult forms of the same species - in the soil of potato sowings. For anatomical and morphological study of collected nematodes temporary and long-term preparations were prepared [Poinar, 1975]. For nematode identification international index formulae of nematodology were used [De Man, 1884; Micoletzky, 1914]. It was established that recorded nematode belongs to the genus Hexamermis and family Mermithidae.

Genus diagnosis - Hexamermis Steiner, 1924 [Steiner, 1924].

Nematodes of this genus are of middle or big sizes (50-80 mm). Frontal part of the head of female, unlike male, is of mainly conic form. Tail end is rounded. Cuticle of the end parts of head and tail of parasitic and postparasitic larvae is thicker, than of the body middle part. Mouth opening in the frontal part of the head is placed symmetrically. Has 6 cephalic papillae; has no labial papillae; Amphids of small size. Vulva is straight and has significantly thickened stoma. Vagina is of pear-form; spicule - pair, straight and short. Has thick sexual papillae. Tail ends of parasitic and postparasitic larvae are rounded.

Typical species: *Hexamermis angusta* Rubzov, 1971 [Rubtsov, 1971].

Results and Discussion

Host: Colorado beetle (*Leptinotarsa decemlineata* Say).

Localization: in adipose tissue of the beetle and larvae.

The apical part of head of female nermithid is speculated, but of male – rounded. (Fig. 1 A, C). Neck gland is seen under cuticle. There are not protrudent tubers on the head. Amphids are small (4-6 μm) and oval. Their ducts are opened a bit lower of cephalic papillae. Cuticle oesophagus is not spread up to the mouth opening. Width of mouths opening walls is of 3-5 μm.

Female: n=7; L=42.3 (32.0-62.5) mm; a = 172.1 (155.3-203.6); V (%) = 55 (54-57);

Body width: near cephalic papillae consists of 70 (53-115) μm, near nerve ring – 162 (92-222) μm, near vulva – 250 (157-380) μm and near the end of trophosome – 158 (120-277) μm. Distance from the frontal part of the head to nerve ring is 269 (179-335) μm; up to vulva – 25.5 (17.3-30.8) mm; from the end of trophosome to the tail end – 168 (75-280) μm. Structure of vagina is not distinguished from that of species described by Rubtsov [Rubtsov, 1971]. Width of cuticle near mouth opening is 21 (19-32) μm, near vulva – 14 (9-18) μm, at the tail end – 25 (24-33) μm.

Male: n=23.5 (19.3-38.6) mm; a=135.1 (105-183.7); c=130.6 (83.8-159.4).

Body width: near cephalic papillae consists of 70 (56-93) μm; near nerve ring 115 (80-193) μm; at anus –155 (120-240) μm; the widest part of body - 198(153-322) μm. Distance from the frontal part of head to nerve ring is 305 (240-396) μm. Male has weakly bent pair spicule (Fig. 1. D), which length is 161 (103-250) μm, diameter – 20 (15-36) μm, but its end is acute. Cuticle width at head opening in the front part of the body is 18 (14-23) μm, in the middle part – 13 (10-16) μm and near tail - 13 (6-30) μm. Tail length is 200 (150-304) μm.

Postparasitic larvae

Female: n=1; L=25.3 mm.

Body width: at cephalic papillae – 98 μm, at nerve ring – 170 μm, at vagina – 335 μm, at the end of trophosome – 225 μm. Distance from the apical part of the head to nerve ring consists of 350 μm, from the end of trophosome to the end of tail – 345 μm. Distance from front of the head to vagina equals to 20.2 mm. Cuticle width is: near head opening in the front of the head – 41 μm, at vagina in the middle part of the body – 13 μm, and at the tail – 102 μm.

Differential diagnosis

By anatomical and morphological characteristics species described above resembles species *Hexamermis stepposis* Artyukhovsky et Khartschenko, (1965) [Artyukhovsky, Khartschenko, 1965], but is more similar to the species *Hexamermis angusta* Rubzov [Rubtsov, 1971], from which it is distinguished by the form of vulva lips, by form and size of amphids.

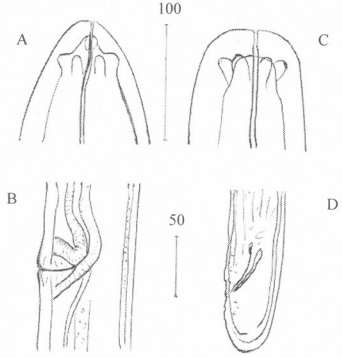


Fig. 1. Hexamermis decemlineatae sp. n
Female: A – frontal end of the body; B – vulva segment
Male: C – frontal end of the body; D – tail segment with spicule.

According to anatomical-morphological features *Hexamermis decemlineatae* sp.n. is considered as a new species for Georgia.

References:

Anderson R.S. *Methode pour I examen des nematodes en vue apicale.* Ann. Parasitol. Hum. Comp., **33**, 171-172, 1958.

Artiukhovski A.K., Kharchenko N.A. *Study of mermithids of Streletskaya Steppe.* Proceedings of Central Black Sea Reservoir, M., p.9, 1965 (in Russian).

Briantsev B.A. *Agricultural entomology.* L., "Kolos", pp.342, 1966 (in Rusian).

De Man J. G.. *Die frei in d. reinen Erde u. in sussen Wasser lebenden nematoden d. niederl. fauna Leiden.* p. 313-321, 1884.

Ipatieva G.V., Pimenova I.N. *Mermithids of the Altai Mountains.* Publ. Saratov University, p.1-149, 1985 (in Russian).

Micoletzky H. *Freilebende Subwasser-Nematoden der Ost- Alpen mit besonderer Beruckichtigung des Lunzer Seengebietes.* Zool. Jahrb. Abt., **1**, 36, 331-546, 1914.
Mishachkov G.A. *Biology and ecology of mermithid Hexamermis albicans - parasite of Colorado beetle.* In: Helminths of insect. M., "Nauka", 85-89, 1980 (in Russian).
Pavlovski E.N. *Methods of hand-anatomy of insect.* M., "Nauka", p.1-45, 1957 (in Russian).
Poinar G.O.jr. *Entomogenous nematodes. A manual and host list of insect – nematode associations.* Leiden E. J. Brill., p. 1-317, 1975.
Rubtsov I.A. *New species of Mermithidae from southern seaside.* Zool. J., **50**, 8, 1143-1152, 1971 (in Russian).
Rubtsov I.A. *Mermithids. Classification, significance, usage.* L., "Nauka", p.207, 1978 (in Russian).
Steiner G. *Beitrage zue Kenntnis der Mermithiden, 2. Mermithiden aus Paraguay in der Sammlung des Zoologischen Museum zu Berlin.* Zbl. Bacteriol., Parasitenkunde. **62**, 1-6.1924.

ახალი სახეობის ნემატოდა *Hexamermis decemlineatae* sp. n. (Nematoda, Mermithidae) კოლორადოს ხოჭოდან

გორგაძე ო., ლორთქიფანიძე მ., კოხია მ., მელაშვილი ნ., კუჭავა მ.

ზოოლოგიის ინსტიტუტი

(მიღებულია 09.10.2006)

რეზიუმე

აღწერილია მერმიტიდას *Hexamermis decemlineatae* sp.n. ახალი სახეობა. მოცემულია ზრდასრული ინდივიდების და პოსტპარაზიტული ლარვების განაზომები. მასპინძელი: კოლორადოს ხოჭო (*L. decemlineata* Say). ლოკალიზაცია: ხოჭოსა და ლარვების ცხიმოვანი ქსოვილი. ხოჭოს სხეულში გამოვლენილია მერმიტიდას პარაზიტული და პოსტპარაზიტული ლარვული ფორმები, ხოლო ზრდასრული ეგზემპლარები გამოვლენილია ნიადაგში. მერმიტიდების ლარვების მინიმალური რაოდენობა თითო ხოჭოში შეადგენდა 1 ეგზემპლარს, ხოლო მაქსიმალური - 30. მასპინძელში პარაზიტული ლარვების რაოდენობა აღწევდა 1-3 ეგზემპლარს. ბუნებრივ პირობებში მერმიტიდების ლარვების მიერ დაინვაზირებულია ხოჭოების 47,5%, ხოლო ლარვების 64,5%.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Genetics

GENE DRIFT IN THE MIRZAANI POPULATION OF WINE YEAST

SHATIRISHVILI A.[1], MENABDE M.[2], SADAGISHVILI T.[2], ZARNADZE T.[3]

[1]*I. Chavchavadze Tbilisi State University*
[2]*Iv. Javakhishvili Tbilisi State University*
[3]*A. Tsereteli Kutaisi State University*

(Received March 1, 2006)

Abstract

During 1996 – 2004 the antagonistic activity was studied in the Mirzaani (Kakheti) population of wine yeast. The population was found to be polymorphic by the feature. Three phenotypic classes were identified in the population: killer (K), neutral (N) and sensitive (S) classes. Gene drift resulting in periodic fluctuation of the phenotypic classes was revealed.

Key words: yeast population, antagonistic activity, killer-system.

Introduction

Among other factors affecting the gene pool of population the gene drift is of particular importance. As a result of its activity allele concentrations change in the gene pool. This process is especially intensified in the populations with great decrease in the number of members [Hedrick, 1999; Shatirishvili, 2002]. Such periodic (cyclic) number fluctuation in the wine yeast is due to abiotic, biotic or anthropogenic factors. In the regions of well-developed domestic wine production of Georgia fermentation of wine occurs spontaneously. The process is first prompted by a small group of yeasts, i.e. "the founder's principle" acts [Shatirishvili, 2002; Mayr, 1970]. Small numbers of yeasts give rise to tens of millions of new ones. The yeast gets into the grape juice from the philosphere, ripened grape berries and wine cellar implements, while the stream depends on the Drosophila [Ribero-Gasion et al., 1980].

Materials and Methods

In Saccharomyces, and particularly in the wine yeast, no traits identifying the population have been worked out yet. Its reproductive area depends on the Drosophila. Therefore, it occupies about 400-500 meters [Sadagishvili et al., 2001; Menabde et al., 2004]. Proceeding from that we considered the forms of wine yeast spread over village Mirzaani and nearby regions to be a population. The material (sediment) was taken from 10 different remote districts by the method described before [Menabde et al., 2004].

The antagonistic activity of strains was detected by means of the testing strains: K7 {KIL – K1}; S14 (sensitive to the system K1); Oxford genetic lines; the line M437 {KIL-K2} created at the Institute of Genetics of Russian Academy of Sciences; the line RA p192 (sensitive to the system K2), a Petergof genetic line. Special culture media were applied for studying the

antagonistic activity of the strains. The specificity of discovery of the killer systems was described before [Shatirishvili et al., 2001].

Results and Discussion

The alcoholic fermentation of grape juice represents a complex multi-stage process. Some microorganisms are involved in it and the yeast fungi join the process at the final stage. During the fermentation inter- and intra-specific competitions take place [Shatirishvili et al., 2001]. The inter-strain antagonism occurs in the wine yeast, that becomes apparent when the cells of sensitive strains are eliminated under the effect of a toxin released by another strain [Menabde et al., 2004].

In order to study the variability of gene frequencies in natural populations of wine yeast, in 1996-2004 we investigated the Mirzaani (Kakheti) population. With the interval of 2-3 years the strains were repeatedly isolated from 10 micropopulations (500 cultures in total) and analyzed genetically. The antagonistic activity was defined by introducing strokes of the strains under study to the nutrient media with a special loop. If the strain contains a killer system, the latter causes a lysis of sensitive test-cultures forming a sterile ring around the developed culture. If a strain is sensitive to the test culture the area of eliminated cells gets stained in dark blue color by Methylen – blue stain [Shatirishvili et al., 2001]. The strain that has a killer-system produces and releases the protein – a toxin that induces elimination of sensitive strain cells. The strains with neutral phenotype are sensitive to the toxin. The results obtained for the strains in 1996 are given as patterns in Tables 1 and 2. The other results have already been published [Sadagishvili et al., 2001; Menabde et al., 2004; Shatirishvili et al., 2001].

The strains constituting the population are arranged in three phenotype classes: killer (K), neutral (N) and sensitive (S) strains. The whole population as well as each micro population appeared to be polymorphic. When compared the structures of different populations studied in different years, we found that in 1998 and 2000 the rates of killer strains reached maximum (17,3% and 15,8 % respectively), while in 2004 the content of killer strains was minimal – 1,4% (see Fig. 1).

Table 1. Determination of antagonistic activity of Mirzaani "Rkatsiteli" population

Phenotype	Number of classes	Test-strains			
		M437	7A – P192	K7	S14
I	1	K	K	K	N
II	7	K	K	N	N
III	4	K	N	K	K
IV	2	K	N	K	N
V	9	N	K	K	N
VI	11	N	N	K	K
VII	3	N	N	K	N
VII	6	N	N	K	K
IX	451	N	N	N	N
X	2	N	N	N	S
XI	2	N	N	S	N
XII	1	S	N	N	S
XII	1	N	S	S	S

Changes in environmental conditions sharply affect the ratio of the strains with K, N and S phenotypes and cause variations in natural populations of the yeast. The reason for that is number fluctuation. Under the severe climate conditions in winter and spring the quantity of the yeast

dramatically decreases causing so called "bottleneck" effect. Thus, it comes the period when the number of strains in population and the density of the population become minimal [Shatirishvili, 2002].

In autumn (in vintage) in the sites of domestic wine production, where the wine fermentation is of spontaneous character, the number and density of the yeast within the population sharply rise. Thus, after passing the "bottleneck" the quantity of population members in the natural population reaches maximum. The small groups with occasionally survived yeasts give rise to the yeast population that means that "the founder's principle" works.

Table 2. Determination of the frequencies of phenotypes K, N and S in Mirzaani micropopulations

Micro-population	Number of analyzed strains	Killer		Neutral		Sensitive	
		Number	%	Number	%	Number	%
I	50	3	6	47	94	-	-
II	50	2	4	48	96	-	-
III	50	5	10	43	86	2	4
IV	50	-	-	50	100	-	-
V	50	1	2	48	96	1	2
VI	50	4	8	46	92	-	-
VII	50	5	10	43	86	2	4
VII	50	6	12	43	86	1	2
IX	50	12	24	38	76	-	-
X	50	5	10	45	90	-	-
Total	500	43	8.6	451	90.2	6	12

References:

Hedrick P. W. *Genetics of population*. Sones & Barlett Publishers, 1999.

Mayr E. *Populations, species and evolution.* Harvard university press, 1970.

Menabde M., Sadagishvili T., Kacharava M., Shatirishvili A. *Antagonistic activity in endemic population of wine yeast (Saccharomyces vini)*. Proc. Georgian Acad. Sci., Biol. Ser. B, **2**, 5-6, 57-60, 2004.

Ribero-Gasion J., Reino E., Ribero-Gasion P. *Theory and practice of wine-making*. v.3, M., 1980

Sadagishvili T., Zarnadze T., Menabde M., Shatirishvili A. *Killer systems of wine yeasts*. Proc.Georgian Acad.Sci., Biol.Ser. **27**, 1-3, 225-229, 2001.

Shatirishvili A., Sadagishvili T., Zarnadze T., Menabde M. *Antaginistic activity of natural saccharomyces*. Georgian Med. News, **70**, 1, 48-51, 2001.

Shatirishvili A. *Genetic mechanisms of evolution in natural population of wine yeasts.* In: Biology and contemporaneity, Tbilisi, p. 254-260, 2002.

ბენთა დრეიფის მოქმედება ღვინის საფუარის მირზაანის პოპულაციაში

შათირიშვილი ა.[1], მენაბდე მ.[2], სადაგიშვილი თ.[2], ზარნაძე თ.[3]

[1]ი. ჭავჭავაძის თბილისის სახელმწიფო უნივერსიტეტი
[2]ივ. ჯავახიშვილის თბილისის სახელმწიფო უნივერსიტეტი
[3]ა. წერეთლის ქუთაისის სახელმწიფო უნივერსიტეტი

(მიღებულია 01.03.2006)

რეზიუმე

შესწავლილია ანტაგონისტური აქტივობა ღვინის საფუარის მირზაანის პოპულაციაში. პოპულაციის სტრუქტურა პოლიმორფული აღმოჩნდა. იგი სამი ფენოტიპური კლასითაა წარმოდგენილი: კილერი (K), ნეიტრალური (N), მგრძნობიარე (S). მიკროევოლუციის მამოძრავებელი ფაქტორის ხემოქმედების შედეგად ფენოტიპური კლასები პერიოდულად ფლუქტუირებს.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Immunology

DISTRIBUTION FEATURES OF SOME ERYTHROCYTIC GROUP-SPECIFIC ANTIGENS SIGNIFICANT FOR CLINICAL MEDICINE IN ADJARA REGION

*NAGERVADZE M., DIASAMIDZE A., AKHVLEDIANI L., DUMBADZE G., KHUKHUNAISHVILI R., KORIDZE M.

Sh. Rustaveli Batumi State University
**E-mail: mari-na@nm.ru*

(Received September 20, 2006)

Abstract

Erythrocytic group antigens interesting in the viewpoint of transfusion, such as *c, K, D^u*, were studied in Adjara population. Those antigens are characterized with high immunogenicity but their screening is not conducted. It was shown that distribution frequency of *c* antigen in Adjara population is 90.2±3.03%. 4.2% of population is carrier of *K* factor. Distribution frequency of D^u antigen is 0.625±0.12%. Number of *CC* genotype carriers within the population appeared to be 8.0±2.7%.

Key words: immunoserological methods, *ABO* system, *Rh* system, *Kell* system.

Introduction

Erythrocytic group antigens stipulating for blood compatibility and appearing as the main reason of posttransfusion complication are significant for clinical medicine [Anstee et al., 1999]. Significance of those group antigens is associated with immune characteristics of the living organism. They take important role in transfusiology [Schonewille et al., 2006], epidemiology [Vojvodic, 2000] and transplantology [Bucin, 2006], in human genetics [Shubin, 1997], and especially in the studies of population genetics [Kucher, 2000]. Study of erythrocytic group systems is also important in ethnic anthropology [Shneider et al., 2002]. Heredity of those systems is so stable that their study for establishment of some ethnic group origin gives accurate data [Schmidt et al., 2003].

Today in Adjara two antigens (*A, B*) of *ABO* system and *D* antigen of *Rh* system are taken into account during blood transfusion. For the individuals where those antigens do not occur the theoretical risk of alloimmunosensitization is high [Judd et al., 1992]. In the viewpoint of transfusion *c* antigen, among rhesus system antigens, is also significant. Numerous data about alloimmunosensitization caused by this antigen are presented in scientific literature [Regan et al., 1997]. Distribution frequency of *c* antigen within world population is 80-82%. 18-20% of humans do not have this antigen and are revealed in *CC* state. Individuals with just this genotype belong to high-risk group of alloimmunosensitization.

Immune activity of *K* antigen is slightly minor than rhesus antigen (*RhD*) activity. Immunosensitization caused by *K* antigen is a frequent case [Donskov, 1996], which is certified by the numerous data of posttransfusion complications described in literature.

Today it is necessary to carry out screening of donors by *K* antigen. At present *Kell*-positive blood is not used in transfusion in many countries.

The majority of mistakes during rhesus system determination are related with weak variation of *D* antigen – D^u. Unlike *D* antigen, D^u antigen has latent antigen determinants. For their discovery, first, it is necessary to fix those determinants on the surface of erythrocytes, and further, to reveal them. Such study is carried out in all those cases when *Cde, cdE* phenotypes are revealed during the primary phenotyping of erythrocytes. Complex methods enable to delete individuals having CD^ue and cD^uE phenotypes from rhesus-negative donors.

Unfortunately, proceeding from high transfusion significance of abovementioned antigens, their screening is not conducted today.

The aim of our work was to study regularities of distribution of erythrocytic group antigens in Adjara region and to forecast theoretically expected alloimmunosensitization.

Materials and Methods

The study was carried out by immunoserological methods. Test-systems having anti -*c*, -*K* specificities, anti –*D* incomplete antibody, antiglobulin serum, standard group erythrocytes and standard serums, were used.

512 individuals of Adjara population were studied.

The obtained data-processing was carried out using statistical methods.

Results and Discussion

In the majority of studied individuals *c* antigen was registered (90.2±3.03%). Distribution frequency of *C* antigen was 53.0±5.3% [Nagervadze et al., 2006] (Fig.1). According to the research of allele concentrations it was revealed that concentration of *c* allele is high and equals to 0.74 (Fig. 2).

Forecasting of theoretically expected immunosensitization caused by *c* antigen was carried out. Carriers of *CC* genotype were separated out. Distribution frequency of *CC* genotype in Adjara population is equaled to 8.0±2.7% (*Cc* – 54.0±4.9 and *cc* – 38.0±4.8), implying that carriers of this genotype do not consist in *c* antigen and during transfusion in 92% cases incompatibility should be revealed (Fig. 3).

With low frequency, but nevertheless, D^u antigen was recorded in Adjara population. Distribution frequency of D^u antigen is 0.625±0.12% (Fig. 4).

Donors having D^u antigen should be belonged to rhesus-positive, but recipients – to rhesus-negative group and rhesus-negative blood should be transfused to them, because normal *D* antigen may cause immune response therein.

As a result of our studies it was revealed that 4.2% of Adjara population is carrier of *K* factor (Fig. 5).

Thus, it was established that distribution frequency of *c* antigen is high in Adjara population, and respectively theoretical risk of immunosensitization caused by this antigen is high. It is necessary to provide medical laboratories with information that erythrocyte screening of donor-recipients upon such antigens, as *c*, C^w, D^u, *K*, must be carried out. Such approach should

decrease cases of posttransfusion complications and the risk of immunosensitization should be brought to minimum.

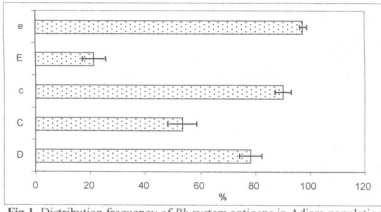


Fig.1. Distribution frequency of *Rh* system antigens in Adjara population

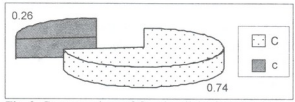


Fig. 2. Concentrations of *C* and *c* alleles in Adjara population

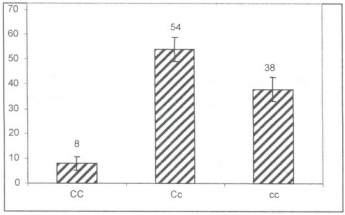


Fig. 3. Analysis of *Cc, CC* and *cc* genotypes in Adjara population

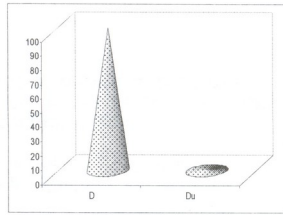


Fig. 4. Distribution frequency of *D* and *D*^{u} antigens in Adjara population

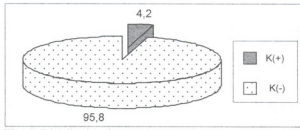


Fig. 5. Distribution frequency of *Kell* system phenotypes in Adjara population.

References:

Anstee D.J., Cartron J.P. *Toward an understanding of the red cell surface//Red cell immunohaemato logy towards its second century.* ESTM. TelAviv, p.12-44, 1999.

Bucin D., Johansson S., Malm T., Jogi P., Johansson J., Westrin P., et al. *Heart transplantation across the antibodies against HLA and ABO.* Transpl Int., **19**, 3, 239-44, 2006.

Kucher AN., Puzyrev VP., Chernetsov DB., Erdynieva LS., Sanchat NO. *Polymorphism of immunological and biochemical marker genes in rural populations of the Tuva Republic.* Genetika, **36**, 4, 562-9, 2000.

Nagervadze M., Akhvlediani L., Dumbadze G., Bagrationi K., Diasamidze A. *Distribution of Rh-Hr antigens in Adjara population.* Proc. Georgian Acad. Sci., Biol.Ser. B, **4**, 1, 48-51, 2006.

Schmidt HD, Scheil HG. *Blood group frequencies in Romania: microregional and ethnic differences.* Anthropol Anz., **61**, 4, 381-93, 2003.

Shneider Iu.V., Shilnikova I.N., Zhukova O.V. *Materials for studying the gene pools of Russia and neighboring countries: the Russian population of the Pskov region.* Genetika, **38**, 11, 1561-5, 2002.

Shubin PN., Morokov VA., Efimtseva EA., Chelpanova TI. *Ethnogenetics of the Komi-Zyrians according to data on the distribution of gene frequencies of erythrocyte and serum systems in the blood.* Genetika, **33**, 2, 235-42, 1997.

Vojvodic S. *Inhibitory activity of blood group antigens M and N in inhibition of virus hemagglutination reactions of influenza viruses.* Med Pregl., **53,** 1-2, 7-14, 2000.

კლინიკური მედიცინისათვის მნიშვნელოვანი ზოგიერთი ერითროციტური ჯგუფსპეციფიკური ანტიგენების გავრცელების თავისებურებანი აჭარის რეგიონში

ნაგერვაძე მ., დიასამიძე ა., ახვლედიანი ლ., დუმბაძე გ., ხუხუნაიშვილი რ., ქორიძე მ.

ჭ. რუსთაველის ბათუმის სახელმწიფო უნივერსიტეტი

(მიღებულია 20.09.2006)

რეზიუმე

აჭარის მკვიდრ მოსახლეობაში შესწავლილია ტრანსფუზიური თვალსაზრისით საინტერესო ერითროციტური ჯგუფური ანტიგენები, როგორიცაა *c, K, D^u*. აღნიშნული ანტიგენები საკმაოდ მაღალი იმუნოგენურობით ხასიათდებიან, მაგრამ არ ხდება მათი სკრინინგი. ნანახებია, რომ აჭარის მოსახლეობაში *c* ანტიგენის გავრცელების სიხშირე 90.20±3.03%-ია. აჭარის მოსახლეობის 4.2% ფაქტორის მტარებელია. *D^u* ანტიგენის გავრცელების სიხშირე – 0.625±0.12%-ია. გამოყოფილი იქნა *CC* გენოტიპების მტარებელი პირები; მათი გავრცელების სიხშირე 8.0 2.7%-ია.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

DETERMINATION OF ANTIMICROBIAL ACTIVITY OF NITROGEN FIXING BACTERIA

BAGALISHVILI M., BASILASHVILI L., KIKVIDZE M., GURIELIDZE M., NUTSUBIDZE N.

S. Durmishidze Institute of Biochemistry and Biotechnology

(Received October 6, 2006)

Abstract

Antagonistic features of rhizosphere of nitrogen-fixing microorganisms, which they reveal towards test-organisms, including phytopatogenic fungi and actinomyces have been studied. Most of investigated nitrogenfixers (28 cultures out of 30) display antimicrobial activity in different degree. Antimicrobial activity of rhizospere nitrogenfixing microorganisms together with other positive characteristics (fixation of molecular nitrogen, production of growth stimulators) enable to use these microorganisms in agriculture on the purpose of soil remediation and enhancing of productivity.

Key words: phytopatogenic fungi, actinomyces, nitrogen-fixing microorganisms

Introduction

Microorganisms are never found in isolated state in natural conditions. They can be obtained as pure cultures only artificially, but in nature, they represented in association, where different interactions are established between microbes. One of the most wide spread forms of interaction is antagonism - when one strain of organisms suppresses development of other organisms. Antagonistic properties of microorganisms are mostly manifested by formation of antimicrobial substances [Crawford, 1988].

28 pure cultures of nitrogen fixers were isolated from rhizosphere of cereals. These rhizospherous microorganisms use the energy of photoassimilates isolated by plant roots for the energy – consuming nitrogen fixation processes. It is supposed that these organisms obtain other advantages that make them competitive among various microbes around the plant roots [Shah et al., 1992].

That is the reason for study whether antimicrobial substances are isolated from rhizosphere of nitrogen fixing microorganisms and what the model is of action of these substances on other microbes.

Materials and Methods

30 nitrogen-fixing microorganisms were studied in all. As test-organisms were used actinomycetes: *Streptomices* 82; *Streptomices fradiae* 110, Phytopathogenic fungi: *Fusarium solani*, *Rhizoctonia* sp; yeasts: *Saccharomyces fragilis*, *Candida utilis*; gram-positive bacteria: *E.coli*, *Ps. fluorescens* (Table 1).

Antimicrobial activity of isolated by us and collectional nitrogen fixing microorganisms was studied by the method of agar blocks [Egorov, 1965] as follows: on Petri dishes with corresponding agar nutrient medium, the studied microorganisms were platted in a form of a lawn. After good development of a cultures and formation of antimicrobial substance, which is diffused in agar, we cut 10 mm diameter agar blocks with a special sterile lancet and replace them on other Petri dishes with previously platted test-organisms on a nutrient medium. After 18-20 h of incubation at the optimal for test-organisms temperature light zones were formed around agar blocks, indicating suppression of development of test-organisms. The results were registered 36- 48 h after incubation. According to sterile zones diameter around agar blocks we could judge about antimicrobial activity of microorganisms.

Results and Discussion

28 nitrogen fixing microorganisms, among studied by us 30 ones, reveal antimicrobial activity against 9 out of 11 test-organisms. The widest spectrum was characteristic for *A. brasilense* Г3 and *A. brasilense* G3 (7 test-organisms). Many microorganisms were found to have antimicrobial activity to *Fusarium solani* (19 microorganisms) and *Streptomyces fradiae* (17 microorganisms). The highest antimicrobial activity of microorganisms was manifested to *Fusarium solani*: G-41 - 19 mm, G111 - 19 mm and G71 - 18 mm (Table 1).

Table 1. Antimicrobial activity of nitrogen fixing microorganisms

Nitrogen fixing microorganisms	test-organisms (size of zones, mm)										
	Strept. fradiae 82	*Stept. fradiae 110*	*Fusarium solani*	*Rhizoctonia sp.*	*Sacch. fragilis*	*Candida utilis*	*Myc. phlei*	*Rhodococcus sp.*	*Staph. aureus*	*E. coli*	*Ps.fluorescens*
A.bras. ATCC 9825	11	12	-	14	-	-	13	11	14	-	-
A.brasilense Г3	11	14	13	14	-	15	12	16	-	-	-
A.brasilense G1	11	12	-	-	-	-	11	-	14	-	-
A.brasilense G2	12	13	-	11	-	13	12	-	-	-	-
A.brasilense G3	13	12	12	15	-	-	12	13	14	-	-
A.brasilense G4	12	-	11	15	-	11	13	-	-	-	-
A.brasilense G5	13	12	13	-	12	-	12	-	-	-	-
A.brasilense G12	11	-	11	-	-	-	-	-	-	-	-
A.brasilense G16	-	-	-	-	-	-	-	13	-	-	-
A.brasilense G20	-	-	12	-	-	11	-	-	-	-	-
G22	-	-	-	-	-	-	-	-	-	-	-
G23	-	12	-	-	-	-	13	11	-	-	-
G24	-	13	-	-	-	-	-	-	11	-	-
G26	11	16	17	-	11	-	-	-	-	-	-
G41	-	14	19	-	-	-	13	14	12	-	-
G43	-	15	14	-	-	-	-	-	-	-	-
G44	-	14	-	-	-	-	-	-	-	-	-

G45	-	13	11	11	-	-	-	-	-	-	-
G61	-	-	13	15	13	13	-	15	17	-	-
G62	-	-	12	-	12	-	-	-	11	-	-
G64	-	-	12	-	-	-	-	-	-	-	-
G66	-	12	13	-	-	-	-	14	-	-	-
G68	-	12	-	-	11	-	11	11	13	-	-
G70	-	-	14	-	11	11	-	-	-	-	-
G71	-	-	18	12	-	11	-	-	-	-	-
G72	-	-	12	-	11	-	-	-	-	-	-
G110	-	-	-	-	-	-	-	-	-	-	-
G111	-	11	19	-	-	-	-	-	-	-	-
G120	-	11	18	-	11	-	-	-	-	-	-
G121	-	-	17	-	11	-	-	-	11	-	-

Fusarium solani – is phytopathogenic fungi, it is characterized by high capability to infection and preserves in soil for a long time, as it suppresses development of useful microorganisms and simplifies the process of penetration into a plant tissue [Meyer et al 1998]. Issuing from the said above the fact that nitrogen fixing microorganisms and among them *Azospirillum* excrete antimicrobial substances towards such strong phytopathogenic organisms is very interesting (Fig.1, 2).

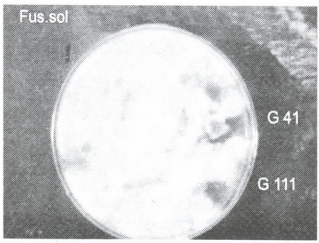


Fig. 1. Antagonism of nitrogen-fixing strains G41 and G111 towards *Fusarium solani*

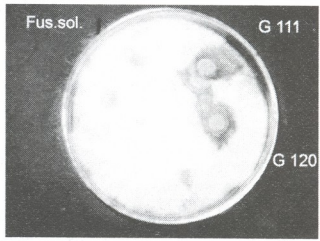


Fig. 2. Antagonism of nitrogen-fixing strains G111 and G120 towards *Fusarium solani*

Thus, most of the studied nitrogen fixers (28 cultures from 30) obtain antimicrobial activity to 9 from 11 test-organisms that gives additional advantages to rhizosphere to develop in microflora. Issuing from the said above introduction of active strains of nitrogen fixers into rhizosphere of agricultural cultures in our opinion will have positive effect on their growth and development.

References:

Egorov N. S. *Microbes – antagonists and biological methods of determination of antibiotic activity.* M., "Visshaya shkola", 1965.

Crawford D.L. *Development of recombinant Streptomyces for biotechnological and environmental uses.* Adv. Biotechnol., **6**, 183-206, 1988.

Meyer L., Weher F.C., Nel & Carling D.F. Characterization of the crater disease strain of *Rhizoctonia solani.* J. Phytopathology, **88**, 4, 366-371, 1998.

Shah S., Karkhanis V., Desai A. *Isolation and characterization of siderophore, with antimicrobial activity, from Azospirillum lipoferum M.* Curr. Microbiol., **25**, 34-35, 1992.

აზოტმაფიქსირებელი ბაქტერიების ანტიმიკრობული აქტივობის განსაზღვრა

ბაგალიშვილი მ., ბასილაშვილი ლ., კიკვიძე მ., გურიელიძე მ., ნუცუბიძე ნ.

ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 06.10.2006)

რეზიუმე

შესწავლილია რიზოსფერული აზოტმაფიქსირებელი მიკროორგანიზმების ანტაგონისტური თვისებები, რომლებსაც ისინი ავლენენ სხვადასხვა ტესტ-ორგანიზმების, მათ შორის ფიტოპათოგენური სოკოებისა და აქტინომიცეტების მიმართ. გამოკვლეულ აზოტფიქსატორთა უმრავლესობამ (30-დან 28 კულტურამ) გამოავლინა ანტიმიკრობული აქტივობა მეტ-ნაკლები ხარისხით. რიზოსფერული აზოტმაფიქსირებელი მიკროორგანიზმების აქტივობა სხვა დადებით მახასიათებლებთან ერთად (მოლეკულური აზოტის ფიქსაცია, ზრდის სტიმულატორების პროდუქციის უნარი) საშუალებას იძლევა ეს მიკროორგანიზმები გამოყენებულ იქნას სოფლის მეურნეობაში ნიადაგების გაჯანსაღებისა და პროდუქტიულობის გაზრდის მიზნით.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

THERMOPHILIC MICROSCOPIC FUNGI OF SOILS OF EAST GEORGIA

*DAUSHVILI L., BURDULI T., KUTATELADZE L., JOBAVA M., DZALAMIDZE I.

S. Durmishidze Institute of Biochemistry and Biotechnology. David Agmashenebeli Alley 10km, 0059 Tbilisi, Georgia
**E-mail: lela_d@mail.ru*

(Received August 14, 2006)

Abstract

70 cultures of microscopic fungi, isolated from dry subtropical-steppe zone of Sighnaghi region (east Georgia) have been investigated. 5 thermophilic, 5 thermotolerant and 4 psychrophilic (facultative) strains of micromycetes were revealed. The optimal ranges of growth and spore-formation of thermophilic micromycetes have been established.

Key words: thermophilic, thermotolerant, psychrophilic, *Aspergillus*.

Introduction

Thermophilic microorganisms, in particular thermophilic fungi are the subjects of intensive investigation. This interest is determined by preferred application of thermophilic fungi and their enzymes in different fields of industry, agriculture and medicine, compared with mesophilic microorganisms and their enzymes [Bilay, 1979].

Thermophilic microscopic fungi are potential sources of various industrially important thermostable enzymes, such as lipases, xylanases, proteases, amylases and pectinases. These enzymes have numerous applications in the detergent, starch, paper, food and pharmaceutical industries [Phutela et al., 2005]. Due to their increased thermostability, enzymes of thermophilic micromycetes are potentially useful in the starch industry for production of maltose and glucose. Thermostable amylase are generally preferred as their application minimizes contamination risk and reduces reaction time, thus providing considerable energy saving. Hydrolysis carried out at higher temperatures also minimizes polymerization of D-glucose to iso-maltose [Kunamneni et al., 2005]. Thermophilic fungi have a strong capacity to degrade polysaccharide constituents in plants, therefore having potential for biotechnological applications such as bioconversion of plant biomass into animal feed, plant fertilizers and chemicals for the food industry [De Faria et al., 2004].

The goal of the study was to reveal the extremophilic by temperature (thermopiles, thermotolerants and psychrophiles) strains of microscopic fungi among the collection of cultures, isolated from different type soils of dry subtropical-step zone of Sighnaghi (east Georgia); also to establish the extreme and optimal ranges of temperature for growth and spore-formation of micromycetes.

Materials and Methods

Cultures of microscopic fungi from the collection of the laboratory of biotechnology of S. Durmishidze Institute of biochemistry and biotechnology served as investigation objects.

The microscopic fungi were grown on the universal nutrient medium: wort (content of sugar 7.0%)-1.0l, agar-20.0.

Surface cultivation of cultures was performed on Petri dishes at temperature range -0°C - 60°C with 5°C intervals.

The growth of microscopic fungi was determined by means of measuring two parameters – the diameter of the colony in two perpendicular directions after 3-fold cultivation, 3, 5 and 7 days later. On the other hand the density of hyphae of the developing colonies in different parts was measured. The final sum of both parameters was appreciated via 3-point system.

Results and Discussion

Experiments were done on the cultures of microscopic fungi isolated from brown carbonate, chestnut and meadow-chernozem soils of Sighnaghi region. The collection consisted of 70 cultures representing 10 different genera of the 3 main classes of microscopic fungi, in particular: Zygomycetes (*Mortierella, Mucor, Rhizopus*), Ascomycetes (*Aspergillus, Penicillium, Chaetomium*) and Deuteromycetes (*Botrytis, Cladosporium, Trichoderma*) [Daushvili et al., 2004].

Microscopic fungi were grouped as thermophiles (obligative thermophiles), thermotolerants (facultative thermophiles) and psychrophiles (facultative psychrophiles) following Cooney and Emerson.

Microscopic fungi with not less than 20°C minimal growth temperature and with maximum at 50°C, were regarded as obligative thermophiles. The facultative thermophiles grew at lower than 20°C and at higher than 50°C. Facultative psychrophyles grew at 0°C and lower temperature and at the same time at 20-25°C too.

According to experimental results 20.0% of the studied fungi were extremophiles by temperature (Fig. 1). Since the tropical and subtropical climatic zones represent a favorable habitat for thermophilic fungi, 14.2% of obligate and facultative thermophiles is a natural phenomenon for Sighnaghi region soils [Bilay, 1985]. Presence of psychrotrophic micromycetes was less expectable here, while existence of facultative psychrophiles may be explained by their tolerance to moderate temperatures (20°-25°C).

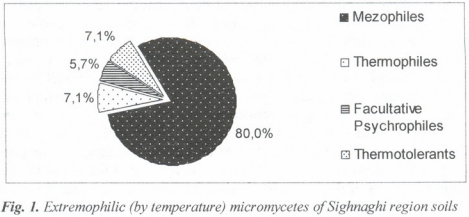


Fig. 1. *Extremophilic (by temperature) micromycetes of Sighnaghi region soils*

Majority of thermophilic and psychrophilic microscopic fungi were isolated from meadow-chernozem soils. This may be explained by the fact that this type of soil was distinguished with abundance and diversity of genera of microscopic fungi.

Among 10 studied genera of fungi thermophilic features were revealed only in genera: *Aspergillus, Chaetomium* and *Penicillium*. Psychrophiles were found among the genera: *Mortierella, Mucor* and *Cladosporium*. Majority of obligate and facultative thermophiles belonged to *Aspergillus* genus (Fig. 2).

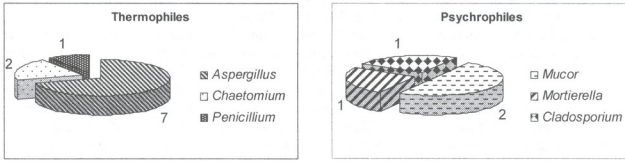


Fig. 2. *Genera of extremophilic fungi of Sighnaghi region soils*

The temperature amplitudes for optimal growth of thermophilic and thermotolerant microscopic fungi were established (Table 1). The temperature amplitude of optimal growth distinguished the thermotolerant representatives of *Aspergillus* genus. More over, the morphological changes, caused by the approaching the extreme temperature limit were less evident, while in genera *Chaetomium* and *Penicillium* the changes were clearly revealed. These morphological changes were mainly expressed in significant decline of spore-formation and transformation of colony color. In some cases changes of hypae length and surface consistence were observed.

Table 1. The optimal temperature ranges for growth of thermophilic fungi.
1 - *Chaetomium sp. S77,* 2 - *Aspergillus sp. S73,* 3 - *Aspergillus niger S60,* 4 - *Aspergillus niger S64,* 5 - *Aspergillus niger S65,* 6 – *Penicillium sp. S57,* 7 – *Chaetomium sp. S67,* 8 - *Aspergillus sp. S51,* 9 - *Aspergillus sp. S52,* 10 - *Aspergillus sp. S58.*

Minimum	Optimal temperature for growth							Maximum
22, 23				1				53, 55
23, 24				2				53, 55
17, 19				3				54, 55
17, 18				4				53, 54
23, 24				5				55
15, 16				6				54
15, 17				7				53
17, 19				8				55
22, 23				9				53, 54
23				10				51, 52
	17-19	28-30	38-40	41-42	44-45	47-48	51-52	

Temperature °C

References:

Bilay T. *Thermophilic fungi*. Kiev, 1985 (in Russian).

Bilai T. *Thermostable enzymes of fungi*. Kiev, 1979 (in Russian).

Daushvili L., Kutateladze L., Burduli T., Jobava M., Dzalamidze I., Aleksidze T., Tinikashvili L. *Isolation of microscopic fungi from soils of different regions of the Caucasus*. Proc. Georgian Acad. Sci., Biol. Ser. B. **2**, 5-6, 68-73, 2004.

De Faria F., Te'o V., Begquist P., Azevedo M., Nevalainen K. *Expression and processing of a major xylanase (XYN2) from the thermophilic fungus Humicola grisea var. thermoidea in Trichoderma reesei*. Letters in Appl. Microbiology, **34**, 119-123, 2002.

Kunamneni A., Kumar K., Singh S. *Response surface methodological approach to optimize the nutritional parameters for enhanced production of α-amylase in solid state fermentation by Thermomyces lanuginosus*. African Journal of Biotechnology, **4,** 7, 708-716, 2005.

Phutela U., Dhuna V., Sandhu Sh., Chadha B.S. *Pectinase and polygalacturonase production by a thermophilic Aspergillus fumigatus isolated from decomposting orange peels*. Brzailian Journal of Microbiology, **36**, 63-69, 2005.

სიღნაღის რეგიონის ნიადაგებში გავრცელებული თერმოფილური მიკროსკოპული სოკოები

დაუშვილი ლ., ბურდული თ., ქუთათელაძე ლ., ჯობავა მ., ძალამიძე ი.

ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 14.08.2006)

რეზიუმე

შესწავლილია სიღნაღის რეგიონის მშრალი სუბტროპიკული სტეპის ზონის ნიადაგებიდან გამოყოფილი მიკროსკოპული სოკოების 70 კულტურა. გამოვლენილია 5 თერმოფილური, 5 თერმოტოლერანტული და 4 ფსიქროფილური (ფაკულტატური) მიკრომიცეტი. დადგენილია თერმოფილური მიკრომიცეტების ზრდისა და სპორაწარმოქმნის ოპტიმალური ტემპერატურული ამპლიტუდები.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

STUDY OF ANTIBIOTIC AND PHAGE SENSITIVITY OF SOME AEROBIC PYOGENIC BACTERIA

GIORGADZE M.[1], NATIDZE T.[2]

[1]*Ltd "Doagnosis-90"*
[2]*Ltd "Immunogen"*

(Received May 15, 2006)

Abstract

It was shown that isolates of streptococcus, staphylococcus and Escherichia coli isolated from suppurative inflammation areas and from the blood of the dog diseased with sepsis are more sensitive to enco- and intestibacteriophages, as compared to antibiotics. At the same time lysis degree is mostly equaled to 4+ and 3+. The obtained results revealed outlook for usage of bacteriophages for treatment of infectious diseases of bacterial etiology in dogs, as preparation without side effects.

Key words: suppurative inflammation of skin, enco- and intestibacteriophages

Introduction

Forming of microbial populations resistant to antibiotics is the actual problem of medicine and veterinary. This phenomenon, which is mainly caused by specific plasmids occurring in bacteria, is connected with purposeless and sometimes uncontrolled usage of antibiotics. Usage of some antibiotics, due to resistance against them, is noneffective and improper.

Medical and veterinary practice demands to look for new antibiotics and to work out alternative preparations. Such preparation is a bacteriophage, which is successfully used in medical practice for treatment of infectious diseases, and among them for suppurative inflammation of skin. Priority of bacteriophages compared to antibiotics is impossibility of forming of resistant populations, absence of allergy, reproduction in the nidus of infection, rapidity of preparation, etc. [Barrow, Soothill, 1997; Carlton, 1999; Smith, Huggins, 1982; Marks, Sharp, 2000]

The aim of our work was comparative study of antibiotic and phage sensitivity of some aerobic pyogenic bacteria isolated from dogs sick with skin diseases (dermatitis, pyoderma), and from the dogs sick with sepsis.

Materials and Methods

In our experiments we used ten isolates of staphylococcus (*St. aurous* - 6, *St. epidermidis* – 4), six isolates of streptococcus (*Str. pyogenes* – 4, *Str.viridans* – 2), and seven isolates of Escherichia coli (*E.cili hemolytic* – 3, *E.coli nonhemolytic* – 4).

Antibiotic sensitivity was studied by disk-diffusion method, phage sensitivity – by Fisk modified method [Overturf et al., 1991].

In our study we used the following antibiotics: amoxacillin, ampicillin, ampicide, gentamicin, doxicicline, erythromycin, kanamycin, chloramphenicol, penicillin, streptomycin, tetracycline, triaxon, cipro-bai and ciprofloxacin.

Among bacteriophages were used the following ones: encophage, intestibacteriophage, pyophage, Sisphage, Fersisphage.

Results and Discussion

As a result of our studies it was established that the effect of antibiotics and phages on staphylococcus, streptococcus and Escherichia coli is different (Table 1). Some staphylococcus are polyresistant against antibiotics. For example, the majority among them revealed resistance against erythromycin, kanamycin, streptomycin, tetracycline, and partially against gentamicin and penicillin. Amoxicillin, ampicillin, ampicide, triaxon have high influence on staphylococcus. To those antibiotics *St.epidermis*-2 appeared to be resistant.

Table 1. Antibiotic and phage sensitivity of microbes

№	Microbes	Antibiotic													Bacteriophage					
		Amoxacillin	Ampicillin	Ampicide	Gentamicin	Doxicicline	Erythromycin	Kanamicin	Chloramphenicol	Penicillin	Tetracycline	Triaxon	Streptomycin	Cipro-bai	Ciprofloxacin	Encophage	Intestiphage	Pyophage	Sisphage	Fersisphage
1	*St. aureus - 1*	4+	4+	4+	4+	R	R	2+	4+	4+	R	R	3+	R	2+	4+	4+	4+	2+	2+
2	*St. aureus - 2*	4+	2+	2+	R	R	R	R	R	R	R	2+	2+	R	2+	3+	3+	2+	2+	2+
3	*St. aureus - 3*	4+	3+	4+	2+	3+	R	R	R	R	3+	4+	R	R	3+	2+	3+	2+	2+	R
4	*St. aureus - 4*	4+	4+	4+	3+	2+	R	2+	2+	4+	4+	4+	R	3+	3+	R	2+	2+	2+	2+
5	*St. aureus - 5*	4+	4+	4+	3+	4+	R	2+	4+	2+	R	3+	R	3+	4+	4+	4+	3+	R	3+
6	*St. aureus - 6*	4+	4+	4+	2+	2+	R	R	R	R	R	3+	R	3+	2+	2+	R	R	2+	2+
7	*St. epidermidis - 1*	3+	3+	2+	R	R	3+	3+	4+	2+	R	2+	2+	R	R	R	3+	2+	R	R
8	*St. epidermidis - 2*	R	R	R	R	R	R	R	3+	R	R	3+	R	R	2+	2+	2+	2+	2+	R
9	*St. epidermidis - 3*	4+	3+	-	3+	R	R	R	4+	4+	2+	4+	R	2+	4+	3+	2+	2+	2+	2+
10	*St. epidermidis - 4*	4+	4+	2+	2+	4+	R	R	R	3+	R	3+	R	3+	4+	2+	3+	3+	2+	2+
11	*St. pyogenes - 1*	3+	R	3+	4+	4+	-	2+	4+	2+	2+	4+	2+	4+	4+	3+	3+	2+	2+	2+
12	*St. pyogenes - 2*	4+	2+	4+	4+	4+	-	3+	4+	3+	3+	4+	3+	3+	4+	2+	3+	2+	R	2+
13	*St. pyogenes - 3*	4+	3+	4+	4+	4+	-	3+	4+	3+	3+	4+	3+	3+	4+	2+	2+	2+	R	R
14	*St. pyogenes - 4*	4+	3+	2+	3+	3+	2+	3+	4+	2+	3+	2+	2+	4+	3+	4+	4+	3+	2+	2+
15	*St. viridans - 1*	3+	3+	3+	3+	4+	2+	3+	4+	2+	4+	3+	3+	3+	3+	3+	4+	3+	2+	R
16	*St. viridans - 2*	3+	R	4+	3+	3+	2+	2+	3+	2+	4+	4+	2+	4+	4+	3+	3+	3+	2+	2+
17	*E. coli (H) - 1*	4+	4+	4+	3+	4+	R	2+	4+	R	4+	4+	2+	4+	4+	2+	3+	3+	2+	2+
18	*E. coli (H) - 2*	4+	4+	4+	3+	4+	R	4+	4+	R	4+	3+	3+	4+	3+	3+	3+	3+	3+	2+
19	*E. coli (H) - 3*	3+	3+	2+	4+	-	R	2+	4+	2+	3+	2+	3+	4+	2+	3+	2+	2+	2+	3+
20	*E. coli (NH) - 4*	4+	3+	2+	3+	-	R	3+	4+	3+	4+	3+	3+	3+	2+	2+	R	R	R	2+
21	*E. coli (NH) - 5*	4+	4+	2+	3+	-	2+	4+	3+	2+	4+	2+	3+	4+	3+	2+	3+	3+	2+	R
22	*E. coli (NH) - 6*	3+	2+	2+	4+	3+	3+	R	3+	3+	4+	3+	2+	4+	3+	2+	3+	3+	3+	2+
23	*E. coli (NH) - 7*	4+	4+	2+	3+	4+	4+	3+	3+	R	3+	3+	2+	3+	3+	3+	3+	2+	3+	2+

Note: R – resistant; "-" – not studied; H – hemolytic; NH – nonhemolytic.

Staphylococcus, as compared to antibiotics, lyses intensively encophage and intestibacteriophage. It should be mentioned that poly-antibiotic resistant staphylococcus (*St.aureus*-2, *St.aureus*-3, *St.epidermidis*-1, *St. epidermidis*-3, *St.epidermidis*-4) in most cases are lysed by bacteriophages in various degrees. Enco- and intestibacteriophage are especially characterized by this feature.

Unlike staphylococcus, streptococcus are more sensitive to the influence of antibiotics and bacteriphages. Among antibiotics with especially intensive effect are distinguished: amoxacillin, ampicillin, ciprofloxacin, triaxon and cipro-bai, (4+, 3+), and among bacteriophages – enco- and intestibacteriophages (4+, 3+, 2+).

Antibiotic- and phage-sensitivities of Escherichia coli are nearly similar. For example, *E.coli*-4, which is resistant against erythromycin, is sensitive against enco- and Fersisphage, and *E.coli-7*, which is resistant against penicillin, turned out to be sensitive against all bacteriophages. In other cases E.coli strains are sensitive to antibiotics (amoxacillin, ampicillin, ciprofloxacin, cipro-bai) and to bacteriophages, especially to enco- and intestiphages (3+, 2+).

Thus, Sensitivity of staphylococcus, streptococcus and Escherichia coli to antibiotics and phages is different. Staphylococcus shows distinct resistance to antibiotics. Strains are sensitive to enco- and intestibacteriophages that enables to use them as alternative agent.

References:

Barrow PA., Soothill JS. *Bacteriophage therapy and prophylaxis: rediscovery and renewed assessment of potential.* Trends Microbiol., **5**, 268-271, 1997.

Carlton RM. *Phage therapy: past history and future prospects.* Arch. Immunol. Ther. Exp., **47**, 267-274, 1999.

Marks T., Sharp R. *Bacteriophages and biotechnology.* J.Chem.Technolgy and Biotechnology, **75**, 1, 6-17, 2000.

Overtruf G.D., Talan D.A., Singer K. et al. *Phage Typing of Staphylococcus.* J.Clin.Microbiol., 29, 2, 373-375, 1991.

Smith HW., Huggins MB. *Successful treatment of experimental Escherichia coli infections in mice using phage: its general superiority over antibiotics.* J. Gen. Microbiol., **128**, 307-318, 1982.

ზოგიერთი აერობული ჩირქმბადი ბაქტერიის ანტიბიოტიკო- და ფაგომგრძნობელობის შესწავლა

გიორგაძე მ.[1], ნათიძე თ.[2]

[1]შპს „დიაგნოზი-90“;
[2]შპს „იმუნოვენი“

(მიღებულია 15.05.2006)

რეზიუმე

დადგენილია, რომ კანის ჩირქოვანი უბნებიდან და სეფსისით დაავადებული ძაღლის სისხლიდან გამოყოფილი სტრეპტოკოკების, სტაფილოკოკების და ეშერიხიების ი'ზოლატები ანტიბიოტიკებთან შედარებით გაცილებით მგრძნობიარეა ენკო- და ინტესტიბაქტერიოფაგების მიმართ. ამასთან, ლიზისის ხარისხი უმეტესად ტოლია 4+ და 3+. მიღებული შედეგები იძლევა ძაღლებში ბაქტერიოფაგის გამოყენების პერსპექტივას.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

DETERMINATION OF EFFECTIVE SCHEME OF ANTI-BRUCELLOSIS IMMUNIZATION OF HORNED CATTLE

GVINJILIA G., GVINJILIA M.

Laboratory of Ministry of Agriculture of Georgia

(Received August 10, 2006)

Abstract

Stability of immune response at vaccination of animals with strain-19 of anti-brucellosis vaccine was studied. Vaccination scheme of adult animals in stationary conditions was worked out. This regimen enables us to conduct animal vaccination by small dose once a year.

Key words: Rose-Bengal Reaction, Agglutination reaction, serological methods

Introduction

It is well known that Brucella is the microorganism causing worldwide spread disease – brucellosis. Brucella mainly infects horned cattle, sheep, goat, pig. Brucella infection can be also seen in wild species of animals [Callahan, 2006 a].

Brucellosis is a persistent disease conducted with elimination of microbe from reproductive system and mammary gland [Sigafoose, 2006]. On account of an economic impact connected with animal health and infection risk of humans the most countries have program of brucellosis control, which involves vaccination of young and adult animals by strain-19. Today, in epizootic and epidemiologic viewpoint the situation is alarm in Georgia [Callahan, 2006 b].

Thus, the goal of our work is to improve diagnostic methods, and special prophylactic agents, and schemes of their usage [Payeur, 2006]. We aimed to determine effectiveness of strain-19 and immune status of animal at various dosing regimen. One link of chain – recipient animal – must become as no recipient. It should be realized by usage of specific immunization; i.e. for extermination of brucellosis immune barrier should be used. Vaccinal prevention may directly set up the precondition of further irreversible liquidation of the disease.

Materials and Methods

88 calves of 4-6 months old and 2000 adult cows from safe on brucellosis farms were used in experiments. Their immunization with strain-19 was carried out in the following dosing regimes: 0.5 billions, 3 billions, 9 billions, and standard 80 billions of microbial cells.

The following standard serological methods were used:

1. Rose-Bengal Reaction (RBR),
2. Agglutination (Raite) reaction (AR)

Results and Discussion

The obtained data show that with the decrease of dose antigenic effect of preparation decreases, postvaccinal reactions disappear earlier than usual (Table 1.).

The obtained results reveal that calves, which were immunized with small dose of vaccine, maintain postvaccinal reactions during 4 months. The calves vaccinated with 9 and 80 billions of microbial cells maintain stable immune status during one year.

Among 22 calves immunized with 9 billion microbial cells 21 turned out to be positive. Animals immunized with standard dose show the same result.

On the second stage of study experiments were carried out on cows in order to determine postvaccinal state in adult cows. Experimental animals (2000 ones) were divided into two groups, vaccinated with 9 and 80 billion microbial cells, respectively.

As a result of serological investigations conducted after 4 months, as well as after 1 year it was revealed that postvaccinal reaction is positive during one year almost in all animals. Percentage of animals having positive reaction, which were vaccinated with 9 billion microbes consists of 92.7% at the end of year, but of animals vaccinated with 80 billion microbes – 93.3%.

Thus, we consider that high dosing regime is not needed, as post-immunization reactions are similar. The common scheme of anti-brucellosis vaccination of adult animals should be the following: cows being in stationary conditions must be vaccinated with the dose of 9 billion microbes once in a year. In that way we should prevent the problem of postvaccinal reaction without reduction of immunogenic characteristics of preparation.

Table 1. Results of serological studies carried out on 4-6 months old calves

Animal groups	Number of animals	Dose of immunization (billion microbes)	After 4 months		After year	
			positive	negative	positive	negative
1	22	0.5	12	19	-	22
2	22	3	15	7	2	20
3	22	9	21	1	21	1
4	22	80	22	-	21	21

Table 2. Results of serological studies carried out on adult caws

Number of animals	Dose of immunization (billion microbes)	After 4 month		After year	
		positive	negative	positive	negative
1000	9	990	10	927	73
1000	80	994	6	933	67

References:

Callahan J. *Brucellosis.* In: Collection of works in microbiology and virology. N24914-101, 2006a.

Callahan J. *Epidemiology of brucellosis.* In: Collection of works in microbiology and virology. N24914-102, 2006b.

Sigafoose T.M. *Brucellosis.* In: Collection of works in microbiology and virology. N24914-100, 2006.

Payeur J.B. *Laboratorial methods of brucellosis investigation.* In: Collection of works in microbiology and virology. N24915, 2006.

მსხვილფეხა რქოსანი პირუტყვის ბრუცელოზის საწინააღმდეგო იმუნიზაციის სქემის განსაზღვრა

ღვინჯილია გ., ღვინჯილია მ.

სხია სოფლის მეურნეობის სამინისტროს ლაბორატორია

(მიღებილია 10.08.2006)

რეზიუმე

შესწავლილია იმუნური პასუხის სტაბილურობა ცხოველის ორგანიზმში ბრუცელოზის საწინააღმდეგო ვაქცინის შტამი-19 მცირე დოზის შეყვანისას. შემუშავებულია ზრდასრული ცხოველების ვაქცინაციის სქემა სტაციონარულ პირობებში, რომელიც საშუალებას იძლევა განვახორციელოთ ცხოველთა ვაქცინაცია მცირე დოზით წელიწადში ერთხელ.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Microbiology

STUDY OF THE EXTENT OF EXTREMOPHILICY OF HALOPHILIC MICROSCOPIC FUNGI FROM SALINE SOILS OF KAKHETI PLAIN

ZAKARIASHVILI N., *KUTATELADZE L., JOBAVA M., KHOKHASHVILI I., DZALAMIDZE I., KVESITADZE E., ALEKSIDZE T.

S. Durmishidze Institute of Biochemistry and Biotechnology
D. Agmashenebeli Alley 10km, 0059 Tbilisi, Georgia
**E-mail: kutateladze@geomail.ge*

(Received May 15, 2006)

Abstract

The extent of extremophilicy of halophilic microscopic fungi, isolated from saline soils of Kakheti plain has been investigated. The pH and temperature ranges of 44 cultures of fungi were established. The optimal pH and temperature of cultivation for each halophile has been selected. 23 extremophiles were revealed: 8 of them by temperature and 15 – by pH. 3 of them were thermophiles, 2 – psycrotolerants, 3 – thermotolerants, 10 – alkaliphiles, and 5 – pH-tolerants. 5 microscopic fungi – *Aspergillus sp.* A26, *Chaetomium sp.* A36, *Trichoderma sp.* A40, *Trichoderma sp.* A41 and *Fusarium sp.* A10 were regarded as extremophiles. They revealed extremophilic properties by three parameters simultaneously – temperature, pH and resistance to high concentrations of NaCl.

Key words: extremophiles, halophiles, thermophiles, pH-tolerant, microscopic fungi

Introduction

Determination of the limits of existence of living organisms is one of the central problems of present-days biology. Due to their high genetic and physiological adaptivity, microorganisms belong to the unique forms of life, managing to exist under the extreme environmental conditions (high and low temperatures, acid and alkali surrounding, high concentrations of salt, etc.) [Aguliar, 1996; Dix, 1995; Mouchacca, 1997; Stetter, 1999]. These types of microorganisms are named as extremophiles. They comprise 3 main groups: 1. thermophiles and psycrophiles 2. alkali- and acidophiles, 3. halophiles. Halophiles deserve a special interest among the extremophiles. Resistance to unfavorable environmental conditions, unique chemical structure and nonpathogenous properties of this group of microorganisms are responsible for involving them into the biotechnological processes. The halophilic cell is a "live laboratory", which makes possible to create several commercial products simultaneously (enzymes, nucleic acids, betonies, ectoines, carotinoids, etc.). The inhabitants of hyper-alkali soils are regarded as "rectifiers" of the environment polluted with organic toxins. In spite of the mentioned biopotential, the abilities of halophils are less used in large technological processes. From the practical experience it is clear that the stable enzymes, acting in extreme regimen (high temperature, alkali or acid surrounding)

possess great advantages in enzyme technology and biotechnology. Extremophiles are producers of this type of enzymes. According to this fact, selection of a high quality extremophiles among the halophilic microscopic fungi, isolated in our early experiments from saline soils of Kakheti plain [Laskhishvili et al., 2005], was of great interest.

Materials and Methods

44 microscopic fungi, isolated from saline soils of Kakheti plane served as objects for experiment. To select extremophiles, microscopic fungi were cultivated under the extreme conditions, on an universal, agar nutrient medium, containing the optimal concentration of NaCl. According to early experiments, the optimal concentrations of NaCl were established for particular halophile [Stetter, 1999].

Microscopic fungi were cultivated under the wide range of temperature (0-55°C, with intervals of 5°C), to reveal thermo- and psycrophiles. The wide amplitude of pH was tested to select acid and alkaliphiles (pH 2.0-10.0, with 0.5 intervals). Cultivation prolonged for 10 days. Values of temperature or pH, resulting in maximal increase, were regarded as optimal. Intensity of growth was evaluated using 3-mark system, taking into account the diameter and growth velocity of the colony of micromycetes.

While the thermophilicy was detected, the determinations offered by Quean and Emerson were used. In particular, the cultures of micromycetes with existence ranges from 20°C to 55°C were regarded as thermophiles. The cultures with maximal growth at ~50°C and able to develop at lower than 20°C were distinguished as thermotolerants.

Microscopic fungi growing at low temperature, but able to develop at 40°C too, were grouped as psycrotolerants.

Micromycetes well developing equally at pH ranging from 5 to 10 were regarded as alkaliphiles, while others, growing at a wide range of pH (2.0 - 10.0) were grouped as pH-tolerant.

Results and Discussion

While determining the extent of extremophilicy of halophilic microscopic fungi isolated from saline soils of Kakheti plane the existing ranges and optimal temperatures were established first of all. For this purpose halophiles were grown at universal agar nutrient medium with optimal concentrations of NaCl, at a wide range of temperature – from 0°C to 55°C, with 5°C intervals.

In table 1 the characteristics of microscopic fungi from saline soils of Kakheti plane are given following the temperature of cultivation. From the table it is clear that the majority of microflora consisted of mesophiles with optimal growing temperature 28-30°C. These meanings are in accordance with the literature data about the prevailing of mesophiles among the microorganisms in nature.

Some representatives of investigated strains sharply changed its morphological and cultural features while approaching the critical temperature. Representatives of different genera of fungi diversely reacted on a temperature fluctuations, e.g. degeneration of spores was mentioned in some species of *Fusarium* genus. The fluffy mycelium of several strains of the genus Mucor turned into skinny one. In some cultures of *Aspergillus* changes in colour or difficulties in spore merging was mentioned.

8 extremophiles by temperature has been selected among the microscopic fungi, isolated from saline soils of Kakheti plain. Among them 3 cultures – *Aspergillus sp.* A2, *A. sp.* A26, and *Chaetomium sp.* A36 were thermophiles, 2 – *Fusarium sp.* A10 and *F. sp.* A43 were psicrotolerants, and 3 – *Trichoderma sp.* A40, *T. sp.* A41 and *T. sp.* A42 were thermotolerants.

After arranging the extremophiles by temperature we aimed to reveal microscopic fungi, growing at extremal pH. For this purpose the cultures from the collection were grown on universal agar nutrient medium, with optimal concentration of NaCl and optimal temperature, changing the pH of the medium within wide range (from 2.0 to 11.0).

In Table 2 the halophilic microscopic fungi spread in saline soils of Kakheti plane are presented according to pH meanings. Among 44 strains of the collection, 15 turned to be halophiles. Between them 10 were alkaliphiles and 5 – pH-tolerant.

Table 1. The extent of extremophilicy of microscopic fungi from saline soils of Kakheti plane

Microscopic fungi	Temperature ranges of culture growth	Optimal temperature of growth, °C	Group of micromycetes by temperature	Relation of culture to NaCl concentrations
1. *Aspergillus sp. A-1*	15°C-40°C	30°C	mesophile	weak
2. *Aspergillus sp. A-2*	20°C-55°C	40°C-45°C	thermophile	weak
3. *Aspergillus sp. A-3*	15°C-45°C	28°C-32°C	mesophile	weak
4. *Aspergillus sp. A-4*	15°C-45°C	30°C	mesophile	weak
5. *Aspergillus sp. A-13*	15°C-40°C	28°C	mesophile	moderate
6. *Aspergillus sp. A-14*	15°C-45°C	25°C	mesophile	moderate
7. *Aspergillus sp. A-25*	15°C-40°C	30°C	mesophile	extreme
8. *Aspergillus sp. A-26*	20°C-55°C	40°C-45°C	thermophile	extreme
9. *Aspergillus sp. A-27*	15°C-40°C	30°C	mesophile	halotolerant
10. *Aspergillus sp. A-28*	15°C-45°C	30°C	mesophile	moderate
11. *Aspergillus sp. A-29*	15°C-45°C	30°C	mesophile	extreme
12. *Aspergillus sp. A-30*	15°C-45°C	30°C	mesophile	moderate
13. *Aspergillus sp. A-31*	15°C-45°C	25 °C -30°C	mesophile	halotolerant
14. *Penicillium sp.A-5*	15°C-45°C	25 °C -30°C	mesophile	weak
15. *Penicillium sp.A-6*	15°C-40°C	25 °C -30°C	mesophile	moderate
16. *Penicillium sp.A-15*	15°C-40°C	25 °C -30°C	mesophile	moderate
17. *Penicillium sp.A-16*	15°C-40°C	25 °C -30°C	mesophile	moderate
18. *Penicillium sp.A-17*	15°C-40°C	25 °C -30°C	mesophile	moderate
19. *Penicillium sp.A-18*	15°C-40°C	25 °C -30°C	mesophile	moderate
20. *Penicillium sp.A-19*	15°C-40°C	25 °C -30°C	mesophile	moderate
21. *Penicillium sp.A-20*	15°C-40°C	30°C	mesophile	halotolerant
22. *Penicillium sp.A-32*	15°C-45°C	30°C	mesophile	moderate
23. *Penicillium sp.A-33*	15°C-45°C	25 °C -30°C	mesophile	halotolerant
24. *Penicillium sp.A-34*	15°C-40°C	30°C	mesophile	halotolerant
25. *Penicillium sp.A-35*	15°C-40°C	30°C	mesophile	halotolerant
26. *Chaetomium sp. A-36*	20°C-55°C	40°C-45 °C	thermophile	halotolerant
27. *Chaetomium sp. A-37*	15°C-40°C	30°C	mesophile	halotolerant
28. *Chaetomium sp. A-38*	15°C-40°C	30°C	mesophile	moderate
29. *Chaetomium sp. A-39*	15°C-40°C	25 °C -30°C	mesophile	extreme
30. *Allescheria sp. A-11*	15°C-40°C	30°C	mesophile	weak
31. *Allescheria sp. A-12*	15°C-45°C	25 °C -30°C	mesophile	weak
32. *Cladosporium sp. A-21*	15°C-40°C	30°C	mesophile	moderate
33. *Cladosporium sp. A-22*	15°C-40°C	30°C	mesophile	moderate
34. *Fusarium sp. A-7*	15°C-40°C	25 °C -30°C	mesophile	weak
35. *Fusarium sp. A-8*	15°C-40°C	30°C	mesophile	weak
36. *Fusarium sp. A-9*	5°C-40°C	10°C-20 °C	mesophile	weak
37. *Fusarium sp. A-10*	5°C-40°C	10°C-20 °C	psicrotrophe (psicrotolerant)	halotolerant
38. *Fusarium sp. A-43*	15°C-45°C	30°C	mesophile	halotolerant
39. *Fusarium sp. A-44*	5°C-55°C	20°C-35 °C	thermotolerante	halotolerant
40. *Trichoderma sp. A-40*	5°C-55°C	20°C-35 °C	thermotolerante	halotolerant
41. *Trichoderma sp. A-41*	5°C-55°C	20°C-35 °C	thermotolerante	moderate
42. *Trichoderma sp. A-42*	15°C-45°C	20 °C -30°C	thermotolerant	halotolerant
43. *Mucor sp. A-23*	15°C-45°C	20°C -30°	mesophile	moderate
44. *Mucor sp. A-24*	15°C-45°C	20°C -30°	mesophile	moderate

Table 2. The extent of extremophilicy of microscopic fungi by pH

Microscopic fungi	Place of sampling	The range of living pH of the culture	Optimal pH	Characterization of culture
1. *A.sp.A25*	Soils with high salinity	5.0-10.0	9	alkaliphile
2. *A.sp.A26*		5.0-10.0	9	alkaliphile
3. *A.sp.A27*		4.5-8.5	5.0	-
4. *A.sp.A28*		4.5-7.5	5.0	-
5. *A.sp.A29*		2.5-10.0	4-8.0	pH-tolerant
6. *A.sp.A30*		4.0-7.5	6.0	-
7. *A.sp.A31*		2.5-10.0	4-8.0	pH-tolerant
8. *P.sp.A32*		4.5-7.5	6.0	-
9. *P.sp.A33*		2.5-10.0	3.5-7.5	pH-tolerant
10. *P.sp.A34*		4.5-10.0	9	alkaliphile
11. *P.sp.A35*		4.5-7.5	5.0	-
12. *Ch.sp.A37*		4.5-7.5	5.0	-
13. *Ch.sp.A36*		4.5-10.0	9	alkaliphile
14. *Ch.sp.A38*		5.0-8.0	6	-
15. *Ch.sp.A39*		4.5-7.5	6.0	-
16. *F.sp.A43*		5.0-7.5	6.0	-
17. *F. sp.A44*		4.5-7.5	5.5	-
18. *T. sp.A40*		4.5-10.0	9.0	alkaliphile
19. *T. sp.A41*		5.0-10.0	8.5	alkaliphile
20. *T. sp.A42*		4.5-8.0	6.0	-
21. *A. spA.13*	Soils with moderate salinity	5.0-8.0	6.0	-
22. *A. spA.14*		4.5-7.5	5.5	-
23. *P.sp.A15*		4.5-7.5	5.0	-
24. *P.sp.A16*		4.5-8.0	5.5	-
25. *P.sp.A17*		4.5-7.5	5.5	-
26. *P.sp.A18*		4.5-7.5	5.5	-
27. *P.sp.A19*		4.5-8.0	6.0	-
28. *P.sp.A20*		4.5-7.5	5.5	-
29. *Cl. sp.A21*		4.5-8.0	6.0	
30. *Cl. sp.A22*		4.5-7.5	6.0	-
31. *M. sp.A23*		4.5- 10.0	9.0	alkaliphile
32. *M. sp.A24*		4.5-10.0	9.0	alkaliphile
33. *A. sp.A1*	Soils with weak salinity	4.5-7.5	5.5	-
34. *A. sp.A2*		4.5-10.0	8.5	alkaliphile
35. *A. sp.A3*		4.5-7.5	5.5	-
36. *A. sp.A4*		4.5-7.5	6.0	-
37. *P.sp.A5*		4.5-7.5	5.5	-
38. *P.sp.A6*		4.5-8.0	6.0	-
39. *A. sp.A11*		5.0-8.0	6.0	-
40. *A.sp.A12*		4.5-7.5	6	-
41. *F. sp.A7*		2.5-10.0	4.5-8.0	pH-tolerant
42. *F. sp.A8*		2.5-10.0	4.5-8.0	pH-tolerant
43. *F. sp.A9*		4.5-7.5	6.0	-
44. *F. sp.A10*		4.5-10.0	8.5	alkaliphile

From the literature it is known that some representatives of *Aspergillus* and *Penicillium* genera are able to grow at a wide range of pH – from 2.0 to 10.0. The pH-tolerant microscopic fungi revealed in our experiments, belong also to these genera. Alkaliphils were found almost among all genera of fungi, except *Cladosporium* and *Allescheria* (Table 2). Extremophilic cultures by simultaneously three parameters (pH, temperature and NaCl concentration) were revealed on the base of selecting the extremophiles by pH and temperature. In particular, as high-quality extremophiles were evaluated: 1. thermophilic *Aspergillus sp.* A26, which is the extreme halophile and alkaliphile, at the same time. 2. *Chaetomium sp.* A36 – as halotolerant, alkaliphile and

thermotolerant. 3, 4. *Trichoderma sp.* A40 and *Trichoderma sp.* A41– as moderate halophile, alkaliphile and thermotolerant, and 5. *Fusarium sp.* A10 – as halotolerant, alkaliphile and psicrotolerant.

Selection of cultures, resistant to extreme conditions of temperature, pH and NaCl concentrations, and revealing the active producers of enzymes among them, is the perspective base for creating of new technologies.

References:

Aguliar A. *Extremophile research in the Europian Union: from fundamental aspects to industrial expectation.* FEMS Microbiol. Rev. 18, 89-92, 1996.

Dix N. I., Webster I. F*ungal ecology.* Chapman & Hall, London, U. K., 1995.

Laskhishvili M., Kutateladze L., Zakariashvili N., et al. *Izolation and identification of microscopic fungi from salty soils of Kakheti.* Proc. Georg. Acad. Sci. Biol. Ser. B, 3, 4, 60-63, 2005.

Mauchacca I. *Thermophilic fungi: biodiversity and taxonomic status.* Cryptogam. Mycol., 18, 19-69, 1997.

Stetter K. O. *Extremophiles and their adaptation to hot environments.* FEBS Lett., 452, 22-25, 1999.

კახეთის ვაკის მარილიან ნიადაგებში გავრცელებული ჰალოფილური მიკროსკოპული სოკოების ექსტრემოფილობის ხარისხის დადგენა

ზაქარიაშვილი ნ., ქუთათელაძე ლ., ჯობავა მ., ხოხაშვილი ი., ძალამიძე ი., კვესიტაძე ე. ალექსიძე თ.

ს. დურმიშიძის ბიოქიმიისა და ბიოტექნოლოგიის ინსტიტუტი

(მიღებულია 15.05.2006)

რეზიუმე

განსაზღვრულია კახეთის ვაკის მარილიანი ნიადაგებიდან გამოყოფილი ჰალოფილური მიკროსკოპული სოკოების ექსტრემოფილობის ხარისხი. დადგენილია კოლექციის 44 კულტურის სახიცოცხლო pH-ისა და ტემპერატურული დიაპაზონი. თითეული ჰალოფილისთვის შერჩეულია კულტივირების ოპტიმალური პირობები – pH და ტემპერატურა. გამოვლენილია 23 ექსტრემოფილი (მათ შორის 8 ტემპერატურის მიხედვით, 15 – pH-ის მიხედვით): 3 თერმოფილი, 2 – ფსიქროტოლერანტი, 10 – ალკალიფილი და 5 – pH-ტოლერანტი. მაღალი ხარისხის ექსტრემოფილებად შეფასებულია 5 მიკროსკოპული სოკო, რომელიც ერთდროულად სამი პარამეტრით (ტემპერატურა, pH და NaCl-ის მაღალი კონცენტრაციებისადმი მდგრადობა) აშკარავნებდა ექსტრემოფილობას: *Aspergillus sp.* A26, *Chaetomium sp.* A36, *Trichoderma sp. A*40, *Trichoderma sp.* A41 და *Fusarium sp.* A10

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Paleobiology

THE MAIN STAGES OF DEVELOPMENT OF *HAMAMELIDACEAE* ON THE TERRITORY OF EURASIA

SHATILOVA I., RUKHADZE L., MCHEDLISHVILI N.

Georgian National Museum, Davitashvili Institute of Paleobiology

(Received November 6, 2006)

Abstract

About the fossil *Hamamelidaceae* a big factual material is accumulated. In Europe and non-tropical Asia the findings of these plants are more often connected with the deposits of Oligocene, Early and Middle Miocene. In Georgia the greatest number of *Hamamelidaceae* were found in the Upper Miocene deposits. This material is of great interest, as it reflects the latest stage of florescence of the family in such region of Eurasia, where at present its representatives are fully absent.

Key words: *Hamamelidaceae*, development history, Western Georgia, Europe, non-tropical Asia.

Introduction

During the process of study of the palynological complexes of Sarmatian and Meotian deposits of Western Georgia our attention was attracted by the abundance of pollen grains of family *Hamamelidaceae.* That stimulated us to lead the monographycal investigation of this taxon and to trace its history on the territory of Eurasia. The big material was analyzed as nearly all Paleogene and Neogene floras described in literature contain one or another genus of *Hamamelidaceae*, except *Liquidambar*, which is the constant component of Cenozoic floras [Shatilova, Stuchlik, 2001].

Materials and Methods

In Georgia the earliest discoveries of *Hamamelidaceae* are dated by Paleogene. In the composition of Eocene, Oligocene, Early and Middle Miocene floras four genera (*Hamamelis, Corylopsis, Sycopsis, Liquidambar*) are known [Panova et al., 1984; Ramishvili, 1982].

In Late Miocene (Sarmatian, Meotian) the part of *Hamamelidaceae* in composition of flora increased and the family was represented by the following genera: *Hamamelis, Corylopsis, Eustigma, Fortunearia, Fothergilla, Parrotiopsis, Parrotia, Sycopsis, Distyliopsis, Distylium, Disanthus, Chunia, Liquidambar, Altingia.* The list is given by system of Endress [Endress, 1989].

All representatives of family *Hamamelidaceae* were probably the components of subtropical forests of plains and lower mountain belt (Fig.1.). In their composition both, evergreen and deciduous plants occur: *Carya, Lauraceae, Myrica, Quercus, Castanopsis, Araliaceae,* etc. [Kolakovsky, Shakryl, 1976; Shatilova et al., 1999].

After the Sarmatian the great number of subtropical forms died out. But this process didn't touch the family *Hamamelidaceae*, which in Meotian time continued to preserve the rich systematical composition. Between Sarmatian and Meotian some differences revealed in generic composition of family, but the number of genera was the same.

The main way of development of Pontian and Kimmerian vegetation was the widening of the area of deciduous cenosis and reduction of subtropical forests, which at the end of Middle Pliocene (Kimmerian) finished to exist as a separate formation. Subtropical plants preserved on the territory of Western Georgia after the Kimmerian became the components of warm-temperate forests. The single representatives of family *Hamamelidaceae (Liquidambar, Altingia, Corylopsis, Fortunearia, Parrotia)* were referred to such plants.

Results and Discussion

In the history of development of *Hamamelidaceae* on the territory of Georgia three stages can be distinguished. The initial one began after the first appearance of the taxon in geological chronology and continued till the time of its florescence. In Georgia it was the time of Paleogene, Lower and Middle Miocene. The second stage corresponded to the period of florescence of family. It was comparative short and embraced the Late Miocene (Sarmatian, Meotian). The third stage (time of decline of *Hamamelidaceae* and their extinction) was longer and corresponded to the whole Pliocene, Early and Middle Pleistocene.

The analysis of rich scientific literature shows that the first stage, during which the process of increasing of the systematical composition of *Hamamelidaceae* was going, finished in the Mesozoic in the non-tropical Asia. In Europe it was continued till the end of Eocene. During this time the representatives of *Hamamelidaceae* did not play significant role in plant communities, main components of which were palms and different *Lauraceae* [Sinitsin, 1980].

The second stage corresponded to the time of florescence of *Hamamelidaceae*, occurred at the end of Palaeogene and at the beginning of the Miocene in non-tropical Asia. In the Eocene, as a result of temperature fall, the flora of Early Palaeogene disappeared giving way to Turgaian flora. The plants forming the nucleus of this flora were concentrated in the southern mountainous regions on the border of Tethysian district. They revealed great tolerance spreading north and south along the mountain ranges and turned out to be capable to survive the drop of the temperature, which occurred at the boundary of Eocene and Oligocene [Meyen, 1987]. Representatives of *Hamamelidaceae* were among such plants. The great majority of them were shrubs ensuring their survival.

According to V. Sinitzin [1980], at the end of Late Oligocene all tropical forms (*Palmae, Proteaceae, Lauraceae, Myrtaceae*) were extinct in Asia, but warm-temperate trees as *Liquidambar, Liriodendron, Nyssa, Rhus, Magnolia*, characteristic for forests of Central-Chinese floristic province at present, continued to exist in the mesophylous forests of Siberia and in North-Eastern part of Asia.

By the end of Palaeogene and during the Miocene the Turgaian flora spread to the south and south-western (in Europe), replacing the retreating subtropical vegetation [Sinitsin, 1980]. After this time the florescence of *Hamamelidaceae* began on the territory of Europe. Here the typical polydominant forests, preserved after the Oligocene, were distributed. Judging from the localities of fossil remains of *Hamamelidaceae*, they were mainly connected with swamp forests. This formation was wide distributed in Central and South-Eastern Europe.

The third stage of development of *Hamamelidaceae*, which corresponds to the period of extinction, began in Early Miocene and proceeded rather rapidly in Asia. In Europe this process began in Late Miocene and proceeded gradually.

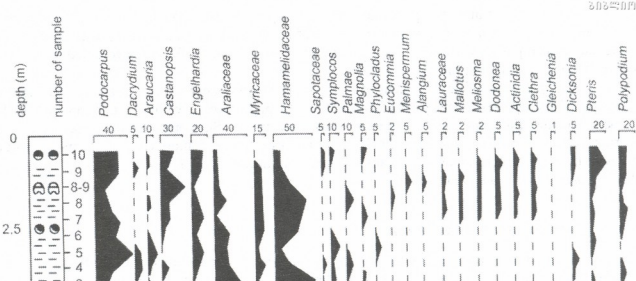


Fig.1. The percentage composition of subtropical plants of the components of lower mountain belt forests on the territory of Western Georgia in Late Miocene.

Conclusion

So, in the history of family *Hamamelidaceae* on the territory of Georgia, Europe and non-tropical Asia three main stages can be distinguished. In the several regions of Eurasia some phases of these stages were nonsynchronous and occupied different stretches of geological time. In Asia and Europe the evolution of *Hamamelidaceae* was closely related with the history of Turgaian flora. In Georgia the development of this family proceeded against the background of evolution of subtropical vegetation, determined the landscape of plain and lower mountain belt. Due to isolate position of Western Georgia, this formation preserved longer, than in other regions of Eurasia and should be traced till the end of Middle Pliocene.

References:

Endress P.K.. *A suprageneric taxonomic classification of the Hamamelidaceae.* Taxon, **38**, 3, 271-376, 1989.

Kolakovskiy A.A., Shakryl A.K. *Sarmatian floras of Abkhazeti.* Proceedings of Sokhumi Botanic Garden, **XXII**, 98-148, 1976 (in Russian).

Meyen S.V. *Basis of Paleobiology.* Moscow, 1987 (in Russian).

Panova L.A., Maligonova E.Yu., Tabachnukova I.P. *Miospore and nannoplankton of Eocene-Oligocene deposits of the north side of Akhaltsikhe depression. Spores and pollen in the deposits of phanerozoic.* Proceedings of VSEGEI, Nov. Ser., **327**, 74-93, 1984 (in Russian).

Ramishvili I.Sh. *Middle Miocene floras of Georgia according to the data of palynological analysis.* Tbilisi, 1982 (in Russian).

Shatilova I., Stuchlik L. *On the history of the development of family Hamamelidaceae.* Palaeontographica, Abt. **B**, Bd. **259**, 235-244, 2001.

Shatilova I.I., Rukhadze L.P., Mchedlishvili N.Sh., Makharadze N.V. *The main stages of development of vegetation and climate of Western Georgia.* Problems of Paleobiology, Tbilisi, **I**, 27-44, 1999 (in Russian).

Sinitsin V.L. *Natural conditions and climate of the territory of USSR in early and middle cenozoic.* Leningrad, 1980 (in Russian).

ოჯახ *Hamamelidaceae*-ს წარმომადგენლები დასავლეთ საქართველოს ნეოგენურში

შატილოვა ი., რუხაძე ლ., მჭედლიშვილი ნ.

საქართველოს ეროვნული მუზეუმი, ლ. დავითაშვილის პალეობიოლოგიის ინსტიტუტი

(მიღებულია 06.11.2006)

რეზიუმე

დღეისათვის დაგროვილია მდიდარი ფაქტიური მასალა ოჯახ *Hamamelidaceae*-ს შესახებ. ამ მცენარეთა ნამარხ ნაშთებს ევროპასა და არატროპიკულ აზიაში ყველაზე ხშირად პოულობენ ოლიგოცენურ, აღრეულ და შუა მიოცენურ ნალექებში. საქართველოში, ძირითადად, მას დასავლეთ ნაწილში, ე.წ. კოლხეთის რეფუგიუმში ამ ოჯახის სხვადასხვა გვარების წარმომადგენლები აღმოჩენილია გვიანმიოცენური (სარმატული, მეოტური) ფლორის შემადგენლობაში. აღნიშნული მასალა ძალზე მნიშვნელოვანია, რადგან იგი ახახავს ოჯახ *Hamamelidaceae*-ს ბატონობის ბოლო ეტაპს ევრაზიის იმ რეგიონში, სადაც დღეს ისინი სრულიად აღარ გვხვდება.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Plant Physiology

THE EFFECT OF BORON, ZINC AND MANGANESE ON THE ACTIVITY OF AMYLASES IN THE SEEDS OF *RAPHANUS SATIVUS, SPINACIA OLERACEA AND CORIANDRUM SATIVUM*

MANGALADZE N., TUTBERIDZE R., KILADZE N.

A. Tsereteli Kutaisi State University

(Received September 18, 2006)

Abstract

Seeds of *Raphanus sativus, Spinacia oleracea and Coriandrum sativum* were processed for 24 hours by 0.02% solutions of $ZnSO_4$, $KMnO_4$ and H_3BO_3. Control seeds were kept in distillate for the same time. To determine amylase activity on the second day of seed germination photocalorimetric method was used. At the seed processing with $ZnSO_4$, $KMnO_4$ and H_3BO_3 it was shown that the activity of α- and αβ-amylases were increased. Boron is suggested to have common positive effect on the amylase activity in seeds of all experimental species.

Key wards: microelements, amylase activity, photocalorimetric method

Introduction

It is well established that microelement deficiency may lead to alimentary diseases of plants [Braun et al., 1962; Katalimov, 1956; Marschner, 1995; Snowball et al., 1980]. Modern authors confirm that microelements are necessary for seed germination, development of vegetative organs, florescence and fruitage. There are some data indicating that microelements take significant role in the formation of anatomical structure of plant organs during the metabolic processes [Szpunar, 2004; Wang et al., 2003; Williams, 2001].

The present work was aimed to examine the role of microelements in the activity of enzymes in the plant cell. Particularly, the effect of boron, zinc and manganese on the activity of amylases in the seeds of *Raphanus sativus, Spinacia oleracea and Coriandrum sativum* was studied.

Materials and Methods

Changes of activities of amylases in the seeds of radish, spinach and coriander treated with solutions containing boron, zinc and manganese were examined. Seeds (300 of each plant) were placed on filtration paper treated with 0.02% solution of microelements for 24 hours. Particularly, seeds (100 ones from every plant) were processed in the $ZnSO_4$, $KMnO_4$ and H_3BO_3 solutions. Control seeds (100 ones from every plant) were set on filtration paper impregnated by distilled water for 24 hours. After processing seeds were placed on Petri dishes. On the second day after appearance of the sprout tips the activities of amylases were determined.

The enzymes were isolated in NaCl solution, incubated in standard starch solution; amount of starch unhydrolyzed by amylases was determined calorimetrically. As a result of hydrolysis and phosphorolysis starch is degraded to monosaccharides during seed germination. α and αβ-amylases, glycoamylase and aminopectin-1-6-glycosidase take part in hydrolysis. During swelling and germination of dry seeds hydrolysis activity of enzymes increases and as a result, starch content decreases and sugar content increases. Activity of amylases is evaluated by hydrolyzed starch in milligrams.

For isolation of amylases 4 g of germinated seeds were put in the mortar, 15 ml of 1% NaCl solution was added and crushed up to homogeneous material. Homogenate was put in the tubes cooled in fridge and centrifuged for 15 min. Substrate was prepared as follows: two tubes (each of 10 ml) were filled with 3 ml of acetate buffer (pH - 5.5) and 3 ml of 2% starch solution, mixed and heated up to 40°C. Further 0.5 ml enzyme preparation was added to one tube, and 5 ml of distilled water - to another one. Content of tubes was heated for 30 min. Then 2 ml of 0.1% of NaCl solution was added to the tubes, mixed and 0.5 ml of solution was taken from each one. Samples were brought into the flasks filled with 25 ml water. 1 ml of 0.1% of NaCl solution, 5 drops of 0.3% of iodine solution were added to the flasks and filled with water up to 50 ml and mixed. Solutions were examined on dyeing in the calorimeter. Red colour filter was used. Amylase activity was calculated by the formula: $A=E_c-E_0 / E_c \times 2.2/60$, where A enzyme activity, E_c and E_0 - optical density of control and experimental solutions, 2.2 coefficient per 1 ml of enzyme solution, 60 - coefficient per 1 mg starch (3 ml of 2% solution corresponds to 60 ml). The obtained data were processed by Student statistical method.

Results and Discussion

As is seen from Fig. 1 activities of α-amylase in the seeds of radish processed with boron and manganese were increased. Difference between activities of the enzyme of the seeds processed by zinc solution and the control is not statistically true. At the same tame processing by boron had stronger influence on the activity of α-amylase compared to manganese. Treatment of the radish seeds with all three experimental solutions increased activity of α-amylase as compared to the control. Processing with the boron caused the strongest effect.

Processing of spinach seeds with all three experimental solutions raised α-amylase activity, though difference with the control is reliable only in the cases of zinc and manganese. At the same time difference in the effects of zinc and manganese is not statistically true.

Processing of coriander seeds with experimental solutions caused statistically true increase in activity of α-amylase in cases of boron and zinc.

As a whole, treatment of radish seeds with H_3BO_3 showed the highest influence on the activity of α-amylase. The effect of $ZnSO_4$ was the biggest in case of spinach seeds, and $KMnO_4$ - in the seeds of radish and spinach.

While processing of radish seeds with experimental solutions activity of αβ-amylase in all three cases compared to control was increased. Differences between effects of boron, zinc and manganese solutions are unreliable. At the same time, activity of αβ-amylase increased much more than activity of α-amylase.

Activity of αβ-amylase in spinach seeds was increased in all three cases, though the strongest, statistically reliable effect was revealed in case of $KMnO_4$. Distinction between effects of H_3BO_3 and $ZnSO_4$ is not significant.

Activity of αβ-amylase in coriander seeds was increased in the cases of H_3BO_3 and $ZnSO_4$. Effect of $KMnO_4$ did not differ reliably from control. The highest influence had $ZnSO_4$.

Activities of amylases play significant role in the process of seed germination, as these enzymes promote degradation of starch and consumption of sugars by plant cells. Obtained results enable to consider that microelements, boron zinc and manganese make favour for increasing amylase activities. At the same time activity of αβ-amylase compared to α-amylase appeared to be more dependant from the presence of boron, zinc and manganese in the cells. Microelements turned to have species-specific character for amylase activities. α-amylase activities in radish and coriander are increased greatly at the effect of boron, but in spinach - at the effect of zinc. Activity of αβ-amylase in radish is equally dependant on boron, zinc and manganese, while in spinach the highest effect revealed - manganese, and in coriander - boron. Effect of boron on amylase activities is nearly similar to the effects of zinc and manganese. In a whole, for germination of seeds of radish, spinach and coriander presawing processing of seeds with the H_3BO_3 solutions should be considered as the most desirable.

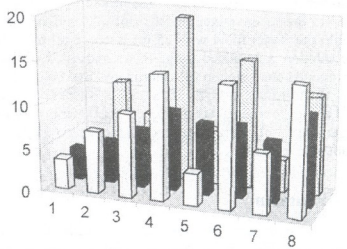


Fig. 1. Activities of amylases in the seeds of *Raphanus sativus, Spinacia oleracea* and *Coriandrum sativum.* 1st row - radish, 2nd row - spinach, 3rd row -coriander; 1 and 2 - control, 3 and 4 - with boron, 5 and 6 with zinc, 7 and 8 - with manganese; in all three cases the first column corresponds to α-amylase and the second column - to αβ-amylase.

References:

Braun T., Zister K., Tanner G. *Physiological consequences of manganese deficiency.* In: Microelemets. M.-L., "Nauka", 1962 (in Russian).

Katalimov M.V. *Microelement content in plants according to species characteristics and features.* In: Microelements in agriculture and medicine. Riga, 1956 (in Russian).

Marschner H. *Mineral nutrition of higher plants.* ed. 2nd, Academic Press, London, 1995.

Snowball L., Robson A.D. Lorenagan. *The effect of cooper on nitrogen fixation in subterranean clover (Trifolium subterraneum).* New Phytol., **85**, 63-72, 1980.

Szpunar J. *Metallomics: a new frontier in analytical chemistry.* Anal. Bioanal. Chem., **204**, 54-56, 2004.

Wang R., Okamoto M., Xing X., Crawford N.M. *Microarray analysis of the nitrate response in Arabidopsis roots and shoots reveal over 1000 rapidly responding genes and new*

linkages to glucose, trehalose-6-phosphate, iron, and sulfate metabolism. Plant Physiol., **132**, 556-567, 2003.

Williams R.J.P. *Chemical selection of elements by cells.* Coordin.Chem.Rev., **216-217**, 583-595, 2001.

ბორის, ცინკის და მანგანუმის ზეგავლენა ამილაზას აქტივობაზე *Raphanus sativus, Spinacia oleracea and Coriandrum sativum* თესლებში

მანგალაძე ნ., თუთბერიძე რ., კილაძე ნ.

ა. წერეთლის ქუთაისის სახელმწიფო უნივერსიტეტი

(მიღებულია 18.09.2006)

რეზიუმე

Raphanus sativus, Spinacia oleracea and Coriandrum sativum-ის თესლები დამუშავებულ იქნა 24 საათის განმავლობაში $ZnSO_4$, $KMnO_4$ და H_3BO_3 0.02% ხსნარებით. საკონტროლო თესლები იგივე დროით თავსდებოდა დისტილირებულ წყალში. განსაზღვრულია ამილაზას აქტივობა თესლის გაღივებიდან მეორე დღეს ფოტოკალორიმეტრული მეთოდით. ნაჩვენებია, რომ $ZnSO_4$, $KMnO_4$ და H_3BO_3 -ით დამუშავებული თესლების α- და αβ- ამილაზას აქტივობები იზრდება. გამოვლენილია, რომ ბორი დადებითად მოქმედებს ამილაზას აქტივობაზე სამივე სახეობის თესლში.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Plant Physiology

INFLUENCE OF SIMULATED ACID RAINS ON PHYSIOLOGICAL INDICES OF WHITE AND RED FORMS OF CABBAGE *(BRASSICA CAPITATA* L.)

RAPAVA L., CHANISHVILI SH., BADRIDZE G., BARBLISHVILI T., ABRAMIDZE S.

N. Ketskhoveli Institute of Botany

(Received October 10, 2006)

Abstract

Effect of pH2.5 water solution of sulphuric acid on seeds and leaves of red and white forms of cabbage (*Brassica capitata* L.) has been studied. Investigated physiological indices – photosynthetic activity of leaves, total activity of growth regulators, dry matter accumulation and water content have revealed higher sensitivity of white form of cabbage to simulated acid precipitations, while red form turned out to be more resistant to acid treatment. This may be due to the high content of anthocyanins in leaves of red cabbage.

Key words: acid rains, photosynthesis, dry matter accumulation, water content, cabbage

Introduction

Increasing of environmental acidity remains one of the most important ecological problems. The influence of acid rains has been mainly studied on woody plants for years [Eds. Muller C. et al., 1999]. During the last period scientists' attention has been focused on investigation of growth and productivity of cultivated plants under the influence of polluted environment [Evans et al., 1986; Hippeli, Elster, 1996].

Phytotoxic effect of polluted environment manifests itself through the changes in plant appearance and dry matter accumulation [Olson et al., 1987]. Therefore, it has been established that plants possess the ability of partial compensation of the primary effect of acid precipitation in the course of development, thus avoiding productivity decrease [Jay et al., 1987]. The opinion exists that plant organism is more resistant to acid pollution at early stages of development [Adams, Hutchinson, 1987].

Proceeding from the above mentioned the objective of our investigation was to study the influence of simulated acid precipitations on seeds and leaves of different varieties of the same species of cultivated plants.

Material and Methods

White and red forms of cabbage (*Brassica capitata* L.) were selected as test objects.

Anthocyanins are known to be one of the principal antioxidant substances, protecting plant organism against different environmental stresses like radiation, O_3, acid rains etc. [Filippovich, et al., 1975]. As the red form of cabbage is rich in substances of anthocyanic nature, presumably it

might be resistant to acid pollution. The comparative study of the effect of acid rains on two forms of cabbage served as a reason of their selection as test objects.

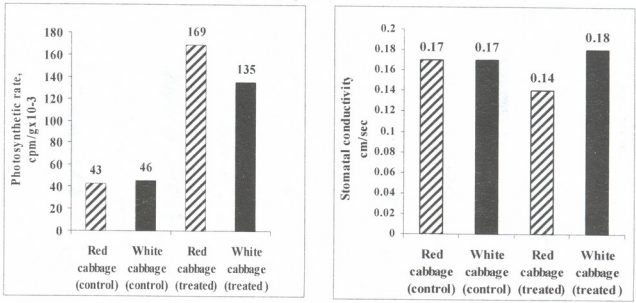
Water solution of sulphuric acid with pH2.5 was used for treating experimental seeds and plants. Cabbage seeds were soaked in acid solution for 24h. In plants, emerged from acid-soaked seeds photosynthetic activity [Voznesensky et al., 1965], stomatal conductivity and total activity of growth regulators (Kefeli, Turetskaya, 1966] were measured.

In other series of experiments leaves of red and white varieties of cabbage plants of the first year of vegetation were sprayed with acid three times with five days interval. Control plants were treated with the same amount of tap water. Material for analysis was taken 10 days after the last spraying.

In addition to the abovementioned indices (photosynthetic activity, stomatal conductivity, total activity of growth regulators) in cabbage plants sprayed with simulated acid rain some indices of water regime and dry matter accumulation were also determined.

Results and Discussion

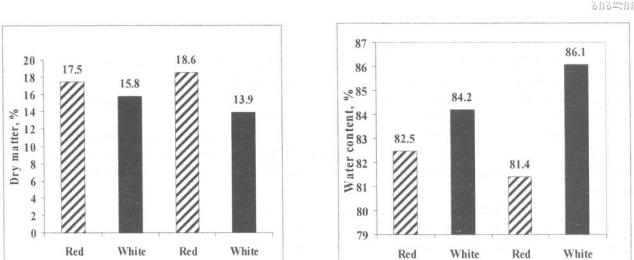
The obtained results show that significant intensification of photosynthetic rate took place in leaves of cabbage plants, emerged from acid-soaked seeds (Fig.1, a). The effect was more pronounced in red form of cabbage. No essential differences were found in stomatal conductivity of control and experimental variants of plant (Fig 1, b).



a b

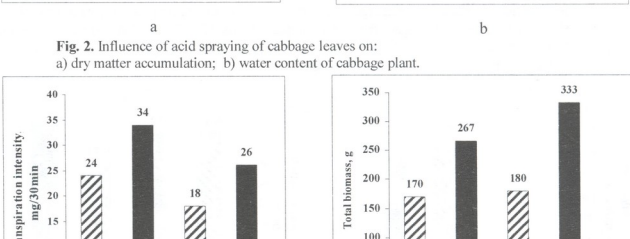
Fig. 1. Influence of acid treatment of cabbage seeds on:
a) photosynthetic activity of leaves; b) leaf stomatal conductivity.

Measurements of dry matter accumulation and water content have revealed that the red form of cabbage was distinguished with higher dry matter and less content of water compared with white form (Fig. 2, a, b). Spraying leaves with acid solution increased dry matter accumulation in red cabbage, while in white form the opposite results were mentioned (Fig 2, a, b). Transpiration index was higher in white cabbage. Spraying with acid solution diminished the index in both forms of cabbage but the effect was more apparent in white cabbage (Fig. 3, a). At the same time the total weight of plants increased (Fig. 3, b).



a b

Fig. 2. Influence of acid spraying of cabbage leaves on:
a) dry matter accumulation; b) water content of cabbage plant.

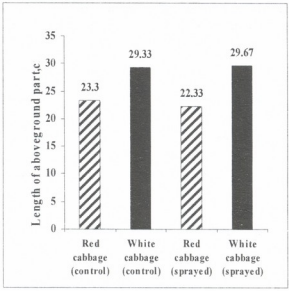


a b

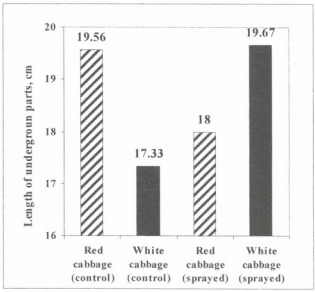
Fig. 3. Influence of acid spraying of cabbage leaves on:
a) transpiration intensity of leaves; b) plant total biomass.

Examination of above and under ground parts of tested plants has shown that as a result of leaf spraying in white cabbage the length of under ground parts increased, while no effect was mentioned on above ground parts (Fig. 4, a, b). In red cabbage spraying with acid caused diminishing of length of both above and under ground parts.

Testing of the total activity of growth regulators manifested essential reduction of the index of growth stimulators in leaves of plants emerged from acid-treated seed (Fig. 5 b; Fig. 6 b). Especially clear effect was mentioned in white cabbage. Opposite effect was revealed in case of plant spraying with acid solution: here significant activation of growth stimulators was detected, which was reflected on changes in biomass accumulation (Fig. 5 c, Fig. 6 c, Fig. 3 b).



a



b

Fig. 4. Influence of acid spraying of cabbage leaves on length of:
a) above- and b) underground parts of a plant.

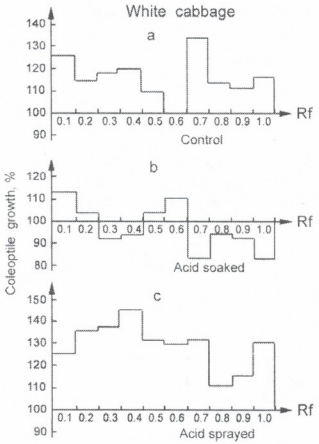


Fig. 5. Total activity of growth regulators in white cabbage leaves

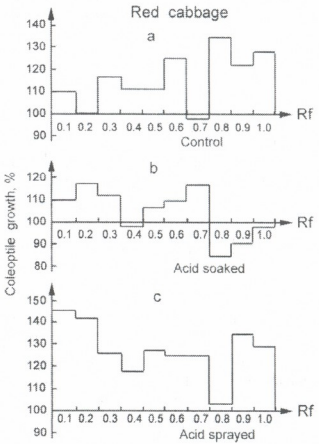


Fig. 6. Total activity of growth regulators in red cabbage leaves

According to the analysis of the obtained data white form of cabbage seems to be more sensitive to simulated acid rains, while red one is more resistant. This fact may be explained by high content of protective substances - anthocyanins in leaves of red cabbage.

References:

Adams C. M., Hutchinson T. C. *New Phytologist,* **106,** 3, 437-456, 1987.

Evans L. S., Sarrantonio M. J., Owen E. M. *New Phytologist,* **103**, 689-693, 1986.

Filippovich J. B., Egorova T. A., Sevast'yanova G. A. *Practical work in general biochemistry.*, M., "Prosveshchenye", 317p., 1975 (in Russian).

Hippeli S., Elster E. F. J. *Plant Physiology*, **148**, 249-257, 1996.

Jay J. S., Osmeloski J., Vamada K., Hellor L. *New Phytologist,* **105**, 1, 139-147, 1987.

Kefeli V.N., Turetskaya R.Kh. *Methods of determination of growth regulators and inhibitors.*, M., 1966 (in Russian).

Ten Years of Monitoring Forest Condition in Europe. Studies of Temporal Development, Spatial Distribution and Impact of Natural and Anthropogenic Stress Factors. Eds. **Muller C.** et al., Brussels: UN/ECE and EC. p.205-275, 1999.

Olson R. L., Winner W. E., Moore L. D. *Environment and Exp. Botany*, **27**, 2, 239-244, 1987.

Voznesensky V.L., Zalensky O.V. and Semikhatova O.A. *Methods of investigation of photosynthesis and respiration of plants.* Moscow, "Nauka", 1965(in Russian).

ხელოვნური მჟავე ნალექებით თესლისა და ფოთლების დამუშავების ბავლენა კომბოსტოს (*Brassica capitata* L.) წითელი და თეთრი ნაირსახეობების ფიზიოლოგიურ მაჩვენებლებზე

რაფავა ლ., ჭანიშვილი შ., ბადრიძე გ., ბარბალიშვილი თ., აბრამიძე ს.

(მიღებულია 10.10.2006)

რეზიუმე

შესწავლილია გოგირდმჟავას pH2.5 წყალხსნარით კომბოსტოს (Brassica capitata L.) წითელი და თეთრი ნაირსახეობების თესლისა და ფოთლების დამუშავების ეფექტი ფოთლების ფოტოსინთეზურ აქტივობაზე, ზრდის რეგულატორების აქტივობაზე, მშრალი ნივთიერების აკუმულაციასა და წყლის შემცველობაზე. კომბოსტოს თეთრი ნაირსახეობის როგორც თესლი, ისე ფოთლები უფრო მგრძნობიარე აღმოჩნდა მჟავე ნალექებით დამუშავებისადმი. კომბოსტოს წითელი ნაირსახეობის შედარებით მაღალი მდგრადობა მჟავათი დამუშავებისადმი საკარაუდოდ ამ ნაირსახეობის ფოთლებში დამცველობითი ფუნქციის მქონე ნივთიერების - ანთოციანების მაღალი შემცველობით უნდა იყოს განპირობებული.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Zoology

EFFICIENCY OF NEMATODE *STEINERNEMA CARPOCAPSAE* SAY AGAINST FALL-PLANTING CUTWORM (*AGRIOTIS SEGETUM* SCHIFF)

KAKULIA I.[1], KOKHIA M.[1], SHEKILADZE K.[2]

[1]*Institute of Zoology*
[2]*Agricultural State University*

(Received May 20, 2006)

Abstract

Bioecology of fall-planting cutworm - *Agriotis segetum* Schiff and the effect of the nematode *Steinernema carpocapsae* Say on the fall-planting cutworm was studied in Sachkhere-Chiatura district, West Georgia, for further usage of obtained results in pest biocontrol. In the field environment pest death rate consists of 68 % in the suspension of the concentration - 150 nematodes/ml, 66.5% - in case of 200 nem/ml, and 67.2% - in case of 250 nem/ml. Effect of entomopathogenic nematodes appeared the highest on the I plot, which was processed with suspension of 150 nem/ml concentration (68%) indicating that efficiency of the nematode *S. carpocapsae* against pests is high even at low concentration of suspension.

Key words: bioecological control, entomopathogenic nematode, nematode suspension.

Introduction

Damage to agriculture caused by pest is great. They obliterate significant part of the yield (nearly 25%) and decrease quality of product. Fall-planting cutworm *Agriotis segetum* Schiff is considered as the most dangerous pest of crops, and particularly of maize. As the maize is one of the main crops in Georgia to study the effect of entomopothogenic nematodes on fall-planting cutworm is the urgent problem.

Fall-planting cutworm is widespread pest in Georgia [Kanchaveli, Supatashvili, 1968]. Its larvae cause great damage to maize, horticultural crops; they injure not only seed germs, but root system too. Larvae cut aslant the root collar of saplings causing their death. Pest larvae of different ages hibernate in the soil. At the beginning of frosts young larvae die, but mature ones make soil bed in early spring and pupate there. Two weeks later nymph flies out of pupae, which generally occur in soil at daytime. Nymph blows 2500 eggs both on weed and cultural plants. Larvae are characterized with negative phototaxis and hence they are hidden in soil. Larvae stage lasts 28-38 days. In Georgia this pest gives 3 generations, and so, their number and respectively damage is great. Damage level caused by fall-planting cutworm belongs to high harmfulness categories.

Materials and Methods

Studies were carried out in the villages Kvatsikhe and Biga of Chiatura region. Plots of maize sowings were located on 10 m from river bank. For experiment 4 plots (3 - experimental and

1 - control) were chosen, each of them of $5m^2$ area. Experiments were carried out in autumn, 2005 (September) and spring, 2006 (May).

Experimental plots were treated with nematode (*Steinernema carpocapsae*) suspension of various concentrations. Suspensions were kept in thermos. Plots were treated early in the morning, at quiet whether conditions (air temperature – 16-20°C). Prepared nematode suspension was sprayed into plants by automax. 3, 8 and 10 days after live and dead larvae were counted both on experimental and control plots according to Franz method [Franz, 1968]. The obtained data are presented in the Table.

Results and Discussion

It was found that pest death rate in I plot was 68%, in II plot – 66.5%, and in III plot – 67.2% (see the table).

Table. Efficiency of nematoda *S. carpocapsae* against fall-planting cutworm (*Agriotis segetum*)

#	Nematode species	Concentration of nematode suspension nem/1ml	Number of larvae	Death rate of larvae			Total number of dead larvae	%
				3 days	8 days	10 days		
1.	*S.carpocapsae*	150 (I plot)	50	10	16	8	34	68
2.	"----------"	200 (II plot)	122	34	32	18	74	66,5
3.	"----------"	250.(III plot)	110	30	32	12	74	67,2
4.	control	clean water	100	-	-	1	1	1

As is seen from the table effect of entomopathogenic nematodes is a bit higher on I plot, which was processed with nematode suspension of 150nem/ml concentration (68%). Results of experiments carried out earlier in laboratory environment showed that while using nematode suspension of 200nem/ml concentration the death rate of larvae was 97%. We consider that the obtained results are caused by the closeness of I plot with the river, and its location in shadow.

Thus, efficiency of nematode *S. carpocapsae* against pests is rather high in spite of low concentration of suspension, and efficiency often depends not on the concentration of suspension but on the experimental conditions.

Obtained data confirm the literature data [Lortkipanidze et al., 2004; Hominick, Reid, 1990] according to which *S. capocapsae* is high-efficient agent for the control of fall-planting cutworm.

References:

Franz J.M. *Zur Berechung des Wirkunsrates einer Microbiologchen Bekamfung von Schädlinskten.* Planzensch., **80**, p. 65-71, 1968.

Hominick W.M., Reid A.P. *Perspectives on entomopathogenic nematology.* In: Entomopathogenic Nematodes in Biological Control (Gaugler R. and Kaya H.K. eds) CRC. Boca Raton FL. p.327-345, 1990.

Kanchaveli G., Supatashvili M. *Forest entomology.* Tbilisi. p. 1-375, 1968 (in Georgian).

Lortkipanidze M., Devdariani Ts., Maglakelidze L., Kokhia M. *Effect of nematodobacterial complex on some pests.* Proceeding of The Institute of Zoology, **XXII**, 45-48, 2004 (in Georgian).

ნემატოდა *STEINERNEMA CARPOCAPSAE* Say -ს ეფექტურობა შემოდგომის ნათესების ხვატარის – *AGRIOTES SEGETUM* Schiff მიმართ

კაკულია ი.[1], კოხია მ.[1], შეყილაძე ქ.[2]

[1]*ზოოლოგიის ინსტიტუტი*
[2]*საქართველოს სასოფლო-სამეურნეო უნივერსიტეტი*

(მიღებულია 20.05.2006)

რეზიუმე

შესწავლილია შემოდგომის ნათესების ხვატარის – *Agriotis segetum* Schiff ბიოეკოლოგია და ნემატოდა *Steinernema capocapsae* Say-ს მოქმედების ეფექტი შემოდგომის ნათესების ხვატარის მიმართ სახნერ-ჰიათურის რეგიონში, შემდგომ მავნე მწერების ბიოკონტროლში გამოსაყენებლად. საველე პირობებში მწერების სიკვდილიანობის პროცენტული მაჩვენებელი 150 ნემატოდა/მლ კონცენტრაციის ნემატოდურ სუსპენზიაში შეადგენდა 68%, 200 ნემ/მლ შემთხვევაში – 66.5%, ხოლო 250 ნემ/მლ-ში კი 67.2%-ია. ენტომოპათოგენური ნემატოდების მოქმედების ეფექტი ყველაზე მაღალი იყო I ნაკვეთზე, რომელიც დამუშავდა 150 ნემ/მლ კონცენტრაციის მქონე სუსპენზიით (68%), რაც იმის მაჩვენებელია, რომ ნემატოდა *S. carpocapsae*-ს ეფექტურობა მავნე მწერების წინააღმდეგ მაღალია მიუხედავად სუსპენზიის დაბალი კონცენტრაციისა.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Zoology

THE ORIBATID MITES (ACARI, ORIBATIDA) OF FLOOD PLAIN ALDER FORESTS OF CENTRAL COLCHIC LOWLAND

*MURVANIDZE M., KVAVADZE E.

Institute of Zoology
**E-mail: makam94@hotmail.com maka.murvanidze@gmail.com*

(Received October 16, 2006)

Abstract

The researches are provided in flooded ecosystems of Central Colchic Lowland on the territories of Colchic National Park, Kobuleti Reserve. These territories are absolutely unique and have great international importance. Oribatid mite fauna of flood plain alder forests in Central Colchic Lowland is studied. 79 species are registered. The ecological analysis shows that in biotops with frequent inundation similar oribatid communities are formed. The diversity of species and high density indicates on a leading role of oribatid mites in processes of decomposition in studied ecosystems.

Key words: Colchic Lowland, *Oribatida*, Ramsar Site, bioindication.

Introduction

The flood plain forests are widely distributed on Colchic Lowland. The main composer of these forests is alder (*Alnus barbata*). Here are also *Pterocarya pterocarpa, Populus canescens, Salix micans* and *S. alba* found. In understorey grow *Rhododendron luteum, Viburnum opulus, Clemantis vitalba, Crataegus pentagina* etc; from liana's group are found - *Smilax excelsa, Hedera colchica, Pericploca graeca* etc [Ketskhoveli, 1959]. The flood plain alder forests are important to maintain the biodiversity of not only Colchic Lowland, but of the whole Black Sea biogeographical region.

In XX century the major part of flood plain forests of Colchic Lowland were drought up and on meliorated soils citrus plantations were mainly cultivated. Currently the floodplain forests and bogs of Colchic Lowland have great international importance, as they make habitats for migrating, hibernating and nesting birds. The flooded habitats have great importance in maintaining the biodiversity of species on the concrete territories [Weigmann, 1997 b]. Currently part of the flooded ecosystems of Colchis are on the territories of Colchic National Park, Kobuleti Reserve and Ramsar Site and are protected by law.

It is known that oribatid mites are one of the main decomposers of organic matters [Ghilarov, Krivolutsky, 1975; Haq, 1987]. They are known as one of the best bioindicators of environment conditions as well [Klausnitzer, 1990, Weigmann, 1991, 1997 b]. Their diversity and density on the studied territory may indicate on condition of these ecosystems.

Information on oribatid mites of Colchic Lowland alder forests is very poor. Lagidze (1981) has registered 29 species of oribatids in bogs with alder forests. Three of them (*Nothrus*

palustris, Minunthozetes pseudofusiger and *Phthiracarus* sp.) were typical for swamped soils and were frequently found. 95 % of registered mites were found in 0-5 cm depth.

Within the animal researches regarding typical biocenoses of Colchic Lowland [Kurashvili, (edt.) 1984], 12 species of oribatid mites were registered on territory of Kulevi in the alder forest. Most of them were found in upper, 0-10 cm layer of soil.

Species that are registered in literature mainly coincide with our data, but this coincidence belongs to such wide distributed species as *Tectocepheus velatus, Platynothrus peltifer, Quadroppia quadricarinata, Scheloribates latipes* and *Minunthozetes pseudofusiger*.

Materials and Methods

Material was taken in June and July 2005. At each site three soil samples (10 cm^3, 0-10 cm depth from surface) were taken and animals were extracted by Tullgren–apparatus. On the studied territory 8 plots were investigated:

1. Anaklia. The left bank of riv. Tikori. Bog with alder forest. N = 42°2,793' E = 41° 37,017'; H = 5m;
2. Anaklia. The left bank of riv. "Didi Gali". Alder forest with *Buxus* understorey.
3. Parpala. The right bank of riv. Churia. Wet alder forest;
4. Imnati. Polydominant wooded bog. N = 42° 08, 283' E = 41° 57,299'; H= 5-7m;
5. Imnati. Kalamona forest. Bog with ash - alder forest. N = 42° 08,190'; E = 41° 96, 980';
6. Kulevi. Alder forest;
7. 6 km from Kulevi. Wet alder forest;
8. Partotskali Lake coast. Bog with alder forest.

As coefficient of faunal likeness between different plots, indicating species identity, Jaccard's coefficient was calculated [Chernov, 1975]. The calculation of community likeness was based on Renkonen's coefficient [Krebs, 1989]. The dominance identities and faunal likeness have been clustered to a dendrogram.

In this investigation only the adult mites were identified and counted.

For the identification of the oribatid mites mainly Ghilarov and Krivolutsky (1975), Balogh and Mahunka (1983), Niedbala (1983) Weigmann (2006) articles were used. For determination of the biogeographycal belonging of oribatid mites work of Subias (2004) was used.

Results and Discussion

79 species of oribatid mites united in 42 families and 50 genera were registered on studied territory (Tab. 1).

Great number of mesophyllic species is presented in the fauna and mainly found in humid ecosystems of Colchic Lowland. Such species are *Mesoplophora pulchra, Microtritia minima, Dissorhina ornata, Eupelops hygrophilus, Achipteria longisetosa, Pergalumna minor* and *Punctoribates mansanoensis*.

No species was registered in every plot. *Steganacarus personatus* was found everywhere except Kulevi (plot 6), and *Protoribates capucinus* was found everywhere except the bank of riv. Churia (plot 7). 34 species were found only in one plot (Tab. 1).

In fauna of oribatid mites predominate widely distributed mites: Palaearctic – 26 species (32 %), Holarctic – 20 species (25 %), Cosmopolits – 16 species (20 %) and European – 8 species (10 %). With less quantity are presented Mediterranean (5 species – 6 %), Caucasian, Endemic (2-2 species – 2-2 %) and Euro-Atlantic (1 species – 1 %) species (Tab. 1).

Table 1. The list of oribatid mites of floodplain alder forests in Central Colchic Lowland and their biogeographycal distribution (+ dominance %; dom. +: % < 1)

#	species	1	2	3	4	5	6	7	8	Distr.
1	*Hypochthonius rufulus* C. L. Koch, 1836	+	+						3	Cosm
2	*Mesoplophora pectinata* Mahunka, 1979				4					Pal
3	*M. pulchra* Sellnick, 1928	+							1	Pal
4	*Phthiracarus assimilis* Niedbala, 1983	+	+							Cauc
5	*Ph. crassus* Niedbala, 1983				+					Md
6	*Ph. ferrugineus* (C. L. Koch, 1841)	7	1	6			3		8	Pal
7	*Ph. globosus* (C. L. Koch, 1841)		11						+	Ho
8	*Ph. incertus* Niedbala, 1983								1	Hol
9	*Ph. lanatus* (Feider & Suciu, 1957)	+			+			9		Eu
10	*Ph. lentulus* (C. L. Koch, 1841)		3		4	16		36		Hol
11	*Ph. ligneus* Willmann, 1931		1				8		14	Hol
12	*Ph. nitens* (Nicolet, 1855)	1	1	3						Pal
13	*Hoplophthiracarus vanderhammeni* Niedbala, 1991	+	1				2	3		Cosm
14	*Steganacarus carinatus* (C. L. Koch, 1841)	+	+			17	2	3		Pal
15	*St. conjuctus* Niedbala, 1983								+	Md
16	St. *personatus* Niedbala, 1983	5	5	70	46	35		3	2	Eu
17	*St. striculus* (C. L. Koch, 1836)		+				4		2	Hol
18	*Microtritia minima* (Berlese, 1904)				+					Cosm
19	*Rhyzotritia ardua* (C. L. Koch, 1841)						3		1	Cosm
20	*Camisia horrida* (Hermann, 1804)								+	Hol
21	*Platinothrus peltifer* (C. L. Koch, 1839)	+	4		+		13	3		Cosm
22	*Nanhermannia nana* (Nicolet, 1855)	+	1			2	8	39	8	Cosm
23	*Belba sculpta* Mihelcic, 1957	+					1			Md
24	*Metabelba pulverulenta* (C. L. Koch, 1840)	+			+					Hol
25	*Arthrodamaeus femoratus* (C. L. Koch, 1840)								+	Pal
26	*Hypocepheus mirabilis* Krivolutski, 1971	1								Eu
27	*Amerobelba decedens*Berlese, 1908	1	2						+	Md
28	*Eremobelba geographica* Berlese, 1908	+	4						2	Eu
29	*Damaeolus ornatissimus* Csiszar, 1962		2							Pal
30	*Gustavia microcephala* (Nicolet, 1855)	2	+	3						Pal
31	*Liacarus brevilamellatus* Mihelcic, 1955	+								Md
32	*L. coracinus* (C. L. Koch, 1841)						1			Pal
33	*Xenillus tegeocranus* (Hermann, 1804)	+	+		+					Pal
34	*Ceratoppia quadridentata* (Haller, 1882)				+	5	1		1	Hol
35	*Carabodes femoralis* (Nicolet, 1855)				1					Pal
36	*C. rugosior* Berlese, 1916	+			3					Hol
37	*Tectocepheus velatus* (Michael, 1880)		+							Cosm
38	*Dissorhina ornata* (Oudemans, 1900)	4	1						1	Hol
39	*Oppia nitens* C. L Koch, 1836	28							7	Hol
40	*Oppiella (Rhinoppia) fallax* (Paoli, 1908)	2	6		+	2			2	Cosm
41	*O. obsoleta* (Paoli, 1908)	+	1							Pal
42	*O.neerlandica* (Oudemans, 1900)	+			+					Hol
43	*O. nova* (Oudemans, 1902)	+			+		16		6	Cosm
44	*O. tuberculata* (Bulanova-Zachvatkina, 1964)		15							Eu
45	*O. unicarnata* (Paoli, 1908)		+						+	Hol
46	*Ramusella insculpta* (Paoli, 1908)		2							Pal
47	*R. mihelcici* (Perez-Inigo, 1965)				1					Pal
48	*Quadroppia michaeli* Mahunka, 1977		+							Pal

49	*Q. quadricarinata* (Michael, 1885)	+								Hol
50	*Suctobelba trigona* (Michael, 1888)	+								Pal
51	*Suctobelbella duplex* (Strenzke, 1950)	+								Pal
52	*S. subcornigera* (Forsslund, 1941)		+							Cosm
53	*Autogneta tragardhi* (Forsslund, 1941)	+							1	Hol
54	*Eupelops acromios* (Hermann, 1804)				+					Pal
55	*E. hygrophilus* (Knuelle, 1954)									Eu
56	*Achipteria longisetosa* Murvanidze & Weigmann, 2003								1	End
57	*Parachipteria georgica* Murvanidze & Weigmann, 2003	13	+						42	End
58	*P. punctata* (Nicolet, 1855)				1					Hol
59	*P. nicoleti* (Berlese, 1883)						14			Hol
60	*Oribatella nigra* Kulijev, 1967		+							Cauc
61	*Galumna obvia* Berlese, 1914						2			Cosm
62	*Pergalumna minor* (Willmann, 1928)		28							Hol
63	*Ceratozetes gracilis* (Michael, 1884)		17	3						Cosm
64	*Edwardzetes edwardsi* (Nicolet, 1855)		+							Eu-Atl
65	*Latilamellobates incisellus* (Kramer, 1897					5				Pal
66	*Sphaerozetes piriformis* (Nicolet, 1885)					7				Pal
67	*S. tricuspidatus* Willmann, 1923						2			Pal
68	*Chamobates kieviensis* Shaldybina, 1980		1						1	Eu
69	*Ch. voigtsi* (Oudemans, 1902)	53	1		2					Pal
70	*Minunthozetes pseudofusiger* (Schweizer, 1956)	+	1		3	2			1	Pal
71	*Punctoribates mansanoensis* (Hammer, 1958)						1			Eu
72	*P. punctum* (C. L. Koch, 1908)		+		17	2				Cosm
73	*Protoribates capucinus* Berlese, 1908	+	2		1	5	20	3	2	Cosm
74	*Scheloribates laevigatus* (C. L. Koch, 1835)				+					Cosm
75	*Sch. latipes* (C. L. Koch, 1840)	+								Pal
76	*Dometorina plantivaga* (Berlese, 1895)				+					Cosm
77	*Oribatula tibialis* (Nicolet, 1855)	1	+		5				2	Hol
78	*Zygoribatula exilis* (Nicolet, 1855)				3	2				Hol
79	*Z. microporosa* (Bulanova-Zachvatkina, 1967)								+	Pal
	total	37	37	6	26	12	17	8	29	
	density 1000 ind/m^2	71	32	17	51	13	50	8	30	

The maximum density of oribatid mites was registered in Anaklia, on the bank of riv. Tikori (71 730 ind/m^2). The density was also high in polydominant wooded bog in Imnati (51 750 ind/m^2) and in alder forest in Kulevi (50 500 ind/m^2). The lower indexes were marked in wet alder forest 6 km from Kulevi (8 250 ind/m^2) (Tab. 1).

High dominance showed *Steganacarus personatus* (70 %, 46 % and 35 % in different sites), *Phthiracarus lentulus* (36%), *Nanhermannia nana* (39%), *Oppia nitens* (28 %), *Parachipteria georgica* (42%), *Pergalumna minor* (28%), *Chamobates voigtsi* (53%) and *Protoribates capucinus* (20%). Dominance value of other species is much less (Tab. 1).

Based on the calculations of faunal likeness and dominance analyses the ecological clusters were build. Three groups are distinguished in cluster of faunal likeness. In the first group there are united mites from Anaklia and Partotskali Lake (plots 1, 2, 8), what is caused by extreme wetness of these plots. Plots 1 and 2 are located on the banks of the river and inundate several times in the year. In the second group oribatid mites of Imnati and 6 km from Kulevi are united (plots 4, 5, 7). Those sites are distinguished by less humidity. The third plot (bank of riv. Churia) is located

a bit higher compared with other plots and inundates rarely. In this plot less number of oribatid mites was registered, what is also a reason for its low likeness with mites of other plots (Fig. 1).

Three groups were divided in cluster of dominance identities as well. The first group includes the dominant species of oribatid mites of riv. Churia and Imnati (plots 3, 4, 5). These plots are territorially close and ecologically similar. In the 3rd plot number of species was low, but their density was high (16 500 ind/m^2) and dominance identities were also high. The second group unites dominant species of Anaklia and Partotskali Lake (plots 1, 2, 8). As it was already mentioned, these plots are ecologically similar. Plots 6 and 7 are grouped together because they are close both, territorially and ecologically (Fig. 2).

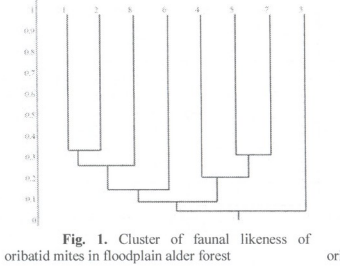


Fig. 1. Cluster of faunal likeness of oribatid mites in floodplain alder forest

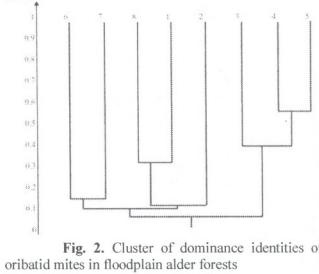


Fig. 2. Cluster of dominance identities of oribatid mites in floodplain alder forests

High diversity and density of oribatid mites are rather unexpected. It is known that mites prefer humid environment, but can not resist high humidity for long period and die because of invasion of microorganisms [Smrz, 1996]. The received results can be explained with low concurrence among the groups which is provoked by extreme conditions and only oribatid mites provide humification and decomposition processes. Low concurrence increases diversity of species of concrete group [Heaney, 2001].

Researches provided in flooded biotops of riv. Oder valley (Germany) showed that oribatid mites adapted to inundation during the winter period and hibernated in early stages; when inundation happened in summer, most of imagoes died [Weigmann, 2004]. In our case the studied territory inundates in spring, summer and autumn. The level of the water decreases in winter, but humidity remains high. We suppose that the main species of oribatid mites of Colchic Lowland alder forests are adapted to the several inundations and fluctuation of their quantity is less discernable during the year.

The species diversity and high density of oribatid mites on the studied territory indicates the unique flora of these ecosystems, active processes of decomposition and soil formation and proves necessity of its international protection.

References:

Balogh J., Mahunka. S. *Primitive Oribatis of Palaearctic Region.* Akademiai Kiado, Budapest,1983.

Chernov Yu. I. *The General Synecological Characteristic of Soil Invertebrates and Methods of Their Analysis.* In: Methods of soil-zoological investigations. Moscow, 160-216, 2002 (in Russian).

Ghilarov M. S., Krivolutsky D. A. *The Identification Keys of Soil Inhabiting Mites. Sarcoptiformes.* "Nauka", Moscow, 1975 (in Russian).

Heaney L. R. *Small Mammal Diversity Along Elevation Gradients in the Phillipines: an Assasment of Patterns and Hypotheses.* Global Ecology and Biogeography. **10,** 15-39, 2001.

Ketskhoveli N. *Vegetation Cover of Georgia.* Metsnierebata akademiis gamomcemloba, Tbilisi, 1959 (in Georgian).

Klausnitzer B. *Ecology of the City Fauna.* "Mir", Moscow, 1990 (in Russian).

Krebs Ch. J. *Ecological Methodology.* Harper & Row Publishers, New York, 1989.

Kurashvili B. (edt). In: *Animal Composition of Typical Biocenoses of Colchic Lowland.* "Mecniereba", pp. 38-43, 1985 (in Georgian).

Lagidze O. I. *Fauna of Podsol-Clay Soils of Colchic Lowland and Their Changes Within Drainage and Cultivating.* PhD abstract, 1981 (in Russian).

Niedbala W. *Les Noueaux Phthiracaridae (Acari, Oribatida) du Caucase.* Ann. Zool. **37,** 62, 1983.

Smrz J. *Some Aspects of the Strategy of Oribatid Mites* (*Oribatida*). Acarology. **IX**. Proceedings, 553-555, 1993.

Subias L. S. *Listado Sistematico, Sinonimico y Biogeografico de los Acaros Oribatidos (Acariformes, Oribatida) del Mundo.* (1758-2002). Graellsia, 60, 2004.

Weigmann G. *Oribatid Communities in Transtcts from Bogs to Forests in Berlin Indicating the Biotope Qualities.* In: F. Dusbabek and V. Bukva (Eds): Modern Acarology, vol. **1**. Academia Prague and SPB Academic Publishing bv. The Hague, 359-364, 1991.

Weigmann G. *Bioindication by means of Isovalent species Groups.* Abh. Ber. Naturkundemus. Goerlitz. **69,** 2, 59-65, 1997a.

Weigmann G. *Die Hornmilben-Fauna (Acari, Oribatida) in Auenböden des Unteren Odertals.* Faun. - Ökol. Mitt. **7,** 319-333, 1997b.

Weigmann G. *Recovery of the Oribatid Mite Community in a Flood Plain After Decline due to Long Time Inundation.* Phytophaga. **XIV,** 201-207, 2004.

Weigmann G. *Hornmilben (Oribatida). Die Tierwelt Deutschlands.* 76 Teil. Goecke, Evers, Kelter Publ. 2006.

კოლხეთის ცენტრალური დაბლობის ჭარბტენიანი მურქნარების ჯავშნიანი ტკიპები (ACARI, ORIBATIDA)

მურვანიძე მ., ყავაძე ე.

ზოოლოგიის ინსტიტუტი

(მიღებულია 16.10.2006)

რეზიუმე

გამოკვლევები ჩატარებულია კოლხეთის ცენტრალური დაბლობის ჭარბტენიან ეკოსისტემებში კოლხეთის ეროვნული პარკის, ქობულეთის ნაკრძალისა და რამსარის საიტის ტერიტორიებზე. ეს ტერიტორიები სრულიად უნიკალურია და აქვთ დიდი საერთაშორისო მნიშვნელობა. შესწავლილია ჯავშნიანი ტკიპების ფაუნა კოლხეთის ცენტრალური დაბლობის ჭარბტენიან მურქნარებში. რეგისტრირებულია 80 სახეობა. ეკოლოგიური ანალიზი გვიჩვენებს, რომ ბიოტოპებში, რომლებიც ხშირად იფარება წყლით, მსგავსი ორიბატიდული ფაუნა ყალიბდება. სახეობათა მრავალფეროვნება და მაღალი სიმჭიდროვე მიუთითებს გამოკვლეულ ეკოსისტემებში დეკომპოზიციის პროცესებში ჯავშნიანი ტკიპების წამყვან როლზე.

Proc. Georgian Acad. Sci., Biol. Ser. B
Vol. 5, No. 1, 2007.

Letter to editor

NEW DATA ON FUNGAL DISEASES OF BULB ONION (*ALLIUM CEPA* L.)

*METREVELI I., KUPRASHVILI T.

L. Kanchaveli Institute of Plant Protection
**E-mail: metreveli_i@hotmail.com*

(Received November 6, 2006)

Abstract

During 2004-2006 the samples of bulb onion (*Allium cepa* L.), harvested in different regions of Georgia or imported from abroad have been examined on the presence of fungi. The list of the revealed fungi supplemented with short diagnoses, the information on sites and time of collection are presented. 5 species are registered for the first time on bulb onion in Georgia. One of them - *Epicoccum* sp. is not identified up to species. The descriptions of 4 new species of fungi are given. Among them 2 species was revealed on the local bulb onion cultivars, and 2 - on imported cultivars.

Bulb onion (*Allium cepa* L.), one of the significant vegetable crops of Liliaceae family, is widely cultivated in Georgia. Onion bulbs are used as food and in medicinal purposes. Onion skin is widely exploited as natural dye-staff. Obtaining big yield of high quality bulb onion is of great importance for the country agriculture. The yield and quality of bulb onion is significantly affected by fungal diseases, both in the open ground and during the storage.

The following most common and harmful fungal diseases have been registered on bulb onion – onion mildew (*Peronospora schleideni* Ung.), smut (*Urocystis cepulae* Frost.), onion rust (*P.mixta Fuckel porii* Wint.), grey rot (*Botrytis alli* Munn.) [Khazaradze, 1952; Shoshiashvili, Kirimelashvili, 1952; Zhvania, 1985; Nebulishvili, 1988]. The mentioned pathogens heavily reduce the yield of bulb onion and worsen its quality.

Active trade relations with foreign countries, uncontrolled situation at customs offices have had effect on agrocoenoses of cultivated crops. Reaction of microorganisms and saprophytic fungi, associated with the material introduced from abroad, undergoes changes in the process of competition in a new environment and the organisms often become pathogenic. This may cause wide expansion of the diseases in a new environment. This very phenomenon prompted us to investigate species composition of fungi associated with both local and imported onion bulbs.

The list of new species of fungi registered by us on the material obtained as a result of observations carried out during 2004-2006 in Georgia, is supplemented with short diagnoses and designations of collecting site and time. Information on collecting sites is appended to the species registered in Georgia and corresponding determination keys are cited. Species of fungi are arranged according to E. Muller and V. Lefler [Muller, Leffler, 1995].

The fungi which are registered for the first time in Georgia are indicated by *.

1. *Peronospora destructor* Berk. Casp [7:147] onion mildew.

Tbilisi, Central Market, green onion leaves brought from village Dzalisi Mtskheta District, 11.05.2004; Agrarian market, Marneuli, onion bulbs, 24.07.2005.

2. *Mortierella jenkini* (Smith) Naumov [9:14]

Tbilisi, Didi Dighomi, private commercial greengrocery. Onion bulbs, introduced from Akhalkalaki district, 17.07.2005.

3. *Choanephora conjuncta Couch. (9:88)

Hyphae of the colony are of filiform, of yellowish-grey color. Conidiophores 0.8 cm high and 10-35μm in diameter. Conidia oblong, ovate, pyriform, globose or elliptic, 8-20(24) x 6-10(12) μm. Brown sporangiospores are elliptic or spindle-shaped, 14-26 x 8-15 μm. Stretched mycelial mat of yellowish light grey color develops on onion skin.

Tbilisi, Saburtalo District, Vazha-Pshavela ave., private commercial greengrocery, 04.05.2004.

4. *Urocystis cepulae* Frost (6:517; 7:211; 10:319)

Gori District, village Kheltubani, 11.05.2004; Kareli district, village Khviti, 13.05.2004.

Tbilisi, Didi Dighomi, private vegetabl stall, 15.09.2006.

5. *Puccinia porii* G. Winter [11:161]

Agrarian market, gori, 13.09.2005; Telavi District, village Gulgula, 21.08.2004.

6. *Aspergillus niger v.* Tiegh. [12:547]

Kaspi District, village Kavtiskhevi, private plot, 16.09. 2005.

7. **Glioklad ium vermoeseni (*Biourge)Thom [13:39]

Aerial mycelium of white color later on becomes granular and turns white-pink. Thick twisted mycelial hyphae of 3-6 μm diameter with numerous vacuoles. Conidiophores 100-200 x 4-5 μm, sterigmas usually 8-12 μm, colorless ones usually 4-6 x 3-4 μm. Conidia elliptic in 1-2 mm long chains.

Tbilisi, Agrarian market, private commercial greengrocery, 18.07.2006.

8. *Verticillium lateritium* Berk. [13:79]

Lagodekhi District, village Ninigori., private greengrocery, 13.06.2005.

9. *Botrytis alli* Munn.. [12:179; 13:67; 10:485]

Telavi District, village Vardisubani, 16.08.2004; Tbilisi, Saburtalo District, private greengrocery, 23.10.2005.

10. *Cladosporium herbarum* (Pers.) Link. [12:313]

Kvareli District, village Shilda, Private plot 17.08.2005; Tbilisi, Didube District, Agrarian market, 23.10.2005.

11. *Periconia atra Corda* [12:349]

Samtredia District, Village Ivandidi, 11.07.2005; Tbilisi, agrarian market 01.05.2006.

12. *Alternaria porri* (Ellis) Cif. Ellis [13:177; 10:512]

Kutaisi, agrarian market, 17.03.2004.

13. *Macrosporium parasiticum* Thum. [11:161]

Samtredia, agrarian market, 17.08.2004.

14. *Stemphyllium allii* Oudem. [13:184; 10:537]

Samtredia, agrarian market, 16.08.2006.

15. *Cercospora duddiae* Welles [12:278; 13:112; 11:161; 10:517]

Tbilisi, Didi Dighomi, private plot 17.08.2005.

16. **Heterosporium alli-cepae* Ran. [13:144]

Conidiphores yellowish-brown, 200x7.5-20 μm wide. Conidia yellowish-grey, unicellular, conidia echinulate; pyriform with 1-2 septa. 31-78 x 8.5-18 μm ([13] 32-76 x 9.5-20 μm).

Lagodekhi District, village Ninigori, private plot, 09.2006; Rustavi, private greengrocery, 25.11.2005.

17. *Fusarium oxysporum* Schlecht [13:261]

Lagodekhi District, private plot, 22.12.2005.

18. **Fusarium avenaceum var. anguioides* (Sherb) Bilai [8:182].

Infected bulbs are darkened, in tissue constructing cells and intercellular spaces mycelial hyphae are developed. Mycelial scab of white color occurs between the bulb scales. Macroconida 20-38 x 3.9-5.3 µm. The fungus occurs in the storage conditions.

Gori, agrarian market, 18.06.2004.

19. **Epicoccum sp.*

Lagodekhi District, village Vardisubani, private plot, 13.06.2005.

20. *Colletotrichum circinans* (Berk.) Voglino. (13:196)

Tbilisi, Saburtalo District, agrarian market "Soplis nobati", 3.03.2004.

References:

Bilai V. I. *Fuzarii*, Kiev, 442 p., 1985 (in Russian).

Ellis M. B. *Dematiaceus Hyphomycetes.* Commonwealth Mycological Institute, Kew, Surrey, England, p.608, 1971.

Flora of spore plants of Georgia (Synopsis). Tbilisi, 1986 (in Russian).

Khazaradze E. Proceedings of Inst.Plant Prot. of Georg.SSR, **VIII**, 97-109, 1952 (Georgian).

Khokhryakov M. K., PotlaichukV. I., SemenovA. Ya, ElbakyanM. A. *Guide-book of diseases of agricultures.* Leningrad, p.303, 1984 (in Russian).

Muller E., Leffler V. *Mycology.* Moscow, p.344, 1995 (in Russian).

Nebulishvili E. Cand. (PhD) Thesis, Tbilisi, p.23, 1988 (in Russian).

Pidoplichko N. M., Milko A. A. *Atlas of mucoraceous molds.* Kiev, p.114, 1976 (in Russian).

Pidoplichko N.M. *Parasitic fungi of cultural plants. Imperfect fungi.* Guide-book, vol. **2**., Kiev, p.298, 1977 (in Russian).

Saccardo P. A. *Syllore Fungorum*, vol. **XIII**, Berlin, 1888.

Shoshiashvili I., Kirimelashvili N. Proceedings of Inst.Plant Prot. of Georg.SSR, **VIII**, 109-118, 1952 (in Georgian).

Yatchevskii A. A. *Fungi identification, imperfect fungi.* Vol. **2**, Petrograd, 580 p.1917, (Russian).

Zhvania N. Cand. (PhD) Thesis, Tbilisi, p.23, 1985 (in Russian).

ახალი მონაცემები ხახვის (*Allium cepa* L.) სოკოვანი დაავადებების შესახებ

მეტრეველი ი., კუპრაშვილი თ.

ლ. ყანჩაველის მცენარეთა დაცვის ინსტიტუტი

(მიღებულია 06.11.2006)

რეზიუმე

ნაშრომში წარმოდგენილია 2004-2006 წლებში საქართველოს სხვადასხვა რაიონებში კერძო პირთა და ფერმერთა ნაკვეთებზე მოყვანილი და საზღვარგარეთიდან იმპორტირებული სარეალიზაციო ხახვის ბოლქვებზე გამოვლენილი სოკოების სია მოკლე დიაგნოზით, მოპოვების ადგილისა და დროის ჩვენებით. გამოვლენილია 5 სახეობის სოკო, რომლებიც დღემდე არ იყო რეგისტრირებული ხახვზე საქართველოში. ერთერთი მათგანი - *Epicoccum sp.* არ არის იდენტიფიცირებული სახეობამდე. აღწერილია სოკოს 4 სახეობა, ამათგან 2 სახეობა გამოვლენილია ადგილობრივ, და 2 - სხვა ქვეყნებიდან შემოტანილი ხახვის მასალაზე.

ინსტრუქცია ავტორთათვის

სამეცნიერო ნაშრომი გამოიცემა 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.

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 ფრჩხილებში ჩასმული ავტორის გვარითა და წლით [Lernmark, Hagglof 1981].

მითითებული ლიტერატურა წარმოდგენილი უნდა იყოს შემდეგნაირად:

ჟურნალის შემთხვევაში

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.

წიგნის შემთხვევაში

Kuhn T.S. *The structure of scientific revolutions.* Chicago, IL, Chicago Press, 2000.

Brush S. *Flowing waters or teeming crowds.* In: Mental Models. D. Gentner (Ed.), Chicago IL., Chicago Press, 865-900, 2001.

მასალა რედაქციაში წარმოდგენილი უნდა იყოს ქაღალდზე ამობეჭდილი და დისკეტით (ან CD-ით). წერილი ერთი ფაილით უნდა იყოს შენახული (ცალკე ფაილად შეიძლება ილუსტრაციების წარმოდგენა), ხოლო ფაილის სახელწოდება წერილის პირველი ავტორის გვარს უნდა ატარებდეს.

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. წერილი 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.

ჟურნალის გამოცემა 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.

ნაშრომის 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.