BIOACCUMULATION OF HEAVY METALS IN TISSUES AND ORGANS OF ALBURNOIDES BIPUNKTATUS BLOCH IN ISKAR RIVER, BULGARIA

Assoc. Prof. Galerida Raikova-Petrova¹ PhD,

Assist. Prof. Desislav Rozdina1 PhD, Assoc. Prof. Ivan Petrov² PhD,

Assist. Prof. Emiliya Nikolova³ PhD

1.Department of General and Applied Hydrobiology, Faculty of Biology, Sofia University "St. Kl. Ohridski", tel. 028167315, galerida@abv.bg, tel. 028167396, d_rozdina@abv.bg;

2.Forest Research Institute, Bulgarian Academy of Sciences, 029620442, ivkrest@bv.bg,

3.Institute for Nuclear Research and Nuclear Energy, Bulgarian Academy of Sciences, tel. 029795595, eminik@inrne.bas.bg

ABSTRACT

The accumulation of heavy metals in bleak (*Alburnoides bipunctatus*) from the middle stream of the Iskar River has been studied. The deposition of heavy metals in tissues and organs was determined on 28 specimens of the species. An x-ray flouriscent analysys was used. The highest concentration of elements was found in the fins, gill caps and gills. The consumption of the bleak is potentially dangerous for heavy metal intoxication of the consumables. The analysis showed higher than the maximum allowed levels of Pb, Zn, Cd and Cu in certain tissues and organs. The highest levels of the elements Cd (4 mg/kg for 3 - 4 year old specimens) and Pb (12 mg/kg in 1 - 2 year olds and 15 mg/kg in 3 - 4 year olds) were established in fish muscles. Based on the bio-accumulation coefficient A. bipunctatus is classified as "macro-concentrator" in relation to heavy metals. A recommendation is given to the anglers to limit consumption of fish from the middle stream of Iskar River until the ecological status of the river improves.

Key words: heavy metals, Alburnoides bipunktatus, Iskar River, bioaccumulation

EX VITRO ADAPTATION OF IN VITRO CULTURES OF THE BALKAN ENDEMIC SPECIES STACHYS THRACICA DAVIDOV

Mariya Rogova*, Zhenia Yordanova, Antoniya Paskaleva, Daniela Marcheva, Veneta Kapchina-Toteva

Department of Plant Physiology, Faculty of Biology, Sofia University "St. Kliment Ohridski"

* corresponding author

ABSTRACT

The genus *Stachys* is one of the largest genera in the family Lamiaceae. It is presented globally by more than 400 species, with 22 naturally distributed species in Bulgaria. *Stachys thracica* Dav. (Thracian woundwort) is a Balkan endemic plant included in the Red Data

Book of Bulgaria with the national conservation status: rare. There is no available data on in vitro propagation and ex situ conservation of Thracian woundwort and scarce information about its chemical composition was available.

The present study aims to develop a protocol for successful *ex vitro* adaption of *in vitro* shoot cultures of S. thracica.

Thracian woundwort shoot culture was induced by sterilization of 100 ripe dry seeds with 70% ethanol and subsequent washing with 96% ethanol. After 14 days, 10% of 50 seeds cultivated on 0.7% water agar germinated, while on ½ MS medium no germination was observed. The sprouting seedlings were then transferred on basal MS medium, supplemented with 3% (w/v) sucrose and 0.7 g/L agar and cultivated under controlled environmental conditions. Regenerated plants with well-developed root system and plentiful leaf biomass were fell under *ex vitro* adaption in a growth camera under controlled environmental conditions. After 21 days, 100% of all used plants were successfully *ex vitro* acclimatised. Comparative analysis of total phenols, flavonoids and total antioxidant activities of in situ grown, *in vitro* and *ex vitro* cultivated *S. thracica* plants also were done. A collection from *ex vitro* adapted plants, which is an approach for preservation of *S. thracica* has been established.

Acknowledgements: This work was financially supported by the grant 141/2016 Sofia University, Bulgaria

Key words: Stachys thracica, ex vitro cultivation, ex situ conservation

HEAVY METAL INFLUENCE ON THE PIGMENT CONTENT OF PEA IN THE COINCUBATION SYSTEM PISUM/SCENEDESMUS

Dimitar Danchev¹, Gergana Marinova², Plamen Pilarski², Ganka Chaneva¹*

* corresponding author

1. Department of Plant Physiology, Faculty of Biology, Sofia University "St. Kliment Ohridski"

2. Laboratory "Experimental Algology" Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences

ABSTRACT

Co-cultivation of pea (*Pisum sativum L.*) and unicellular green alga Scenedesmus incrassatulus (*Chlorophyceae*) had a pronounced beneficial effects on the higher plant exposed to heavy metals' treatment. Adding of Cu, Cd and Pb to the nutrition medium strongly inhibited growth and pigment content of pea plants. The most significant toxic effect was provoked by Pb and Cd, which caused 45% - 60% growth inhibition. The amounts of chlorophyll a and chlorophyll b were also greatly reduced, while carotenoids were affected to a lesser degree. However, after Cu2+ adding to the medium, the growth and pigment content of pea plants remained less influenced.

Coincubation system *Pisum/Scenedesmus* significantly modulated the toxic effects of heavy metals on the higher plant - growth rate and pigment composition were affected to a

much lesser extent. That effect was particularly well expressed in the variants treated with copper ions. The most pronounced positive effect was found regarding carotenoids, whose concentration was slightly reduced in comparison to the control plants.

These results gave us a reason to assume that the toxic influences of heavy metals were accomplished through different mechanisms, and the most likely, the mode of action of Cu differs from that one of Cd and Pb. *Co-cultivation* of higher plant with Sc. *incrassatulus*, which strongly accumulated metals from the environment, resulted in a pronounced protective effect on the growth rate and pigment content of pea.

Key words: Pisum sativum, Scenedesmus incrassatulus, heavy metals, pigments

VIABILITY AND SURVIVAL OF LACTIC ACID MICROFLORA IN NEW SYNBIOTIC FREEZE-DRIED FOODS FROM SHEEP'S AND COW'S MILK

ABSTRACT

Have been developed new functional synbiotic food with a high percentage of live, active cells of L.bulgaricus 1381, Str.thermophilus 1374 and L.acidophilus 1379. The combination of strains of lactic acid cultures e selected experimentally on physiological and biochemical characteristics, resistant to the conditions in the gastrointestinal tract of humans. As starting raw materials used sheep's milk breed Rhodope Gypsies and cow's milk by the Bulgarian Rhodopes cattle. Lactic acid bacteria are involved in the gel of chitosan, which increases their stability in model conditions of digestion and provide an efficient cryoprotection during cryogenic treatment. The composition of new foods is enriched with natural ingredients - grains (quinoa, whose), fruits (elderberry, mango) walnuts, tahini, vegetable oils (linseed oil) and sweeteners (fructose and stevia). Through thorough determination it found that the new synbiotic foods processed by the method of freeze-drying contain a wide range of biologically active substances. Studies were conducted on the viability of lactic microflora in the synbiotic product before and after lyophilization. As a control used fresh milk, fermented with selected starter culture. Conducted microbiological studies indicate that strains streptococcus and lactobacillus in synbiotic products after lyophilization have higher percentage of survival (77.3% and 73% of Str. Thermophilus in synbiotic food from sheep's and cow's milk and 72.1% and 65% lactobacillus in the same foods) as compared with the lactic acid bacteria of the controls after lyophilization. This suggests that the new functional lyophilized foods are with a higher titre of useful microflora strain L.bulgaricus 1381, Str.thermophilus 1374 and 1379 L.acidophilus compared with lyophilized milks controls.

Key words: Synbiotic foods, lactic microflora, survival.

CARCINOGENIC METABOLITE OF THE PESTICIDE MANCOZEB, IN PRODUCTS OF RAINBOW TROUT, REARED IN CONDITIONS OF PERMISSIBLE UNDER EUROPEAN LAW NORM

Milena Tzanova¹, Vasil Atanasov¹, Botjo Zaharinov², Georgi Beev¹, Toncho Dinev¹, Elica Valkova¹

1.Trakia University, Stara Zagora, Faculty of Agriculture, Department "Biochemistry, Microbiology and Physic"

2.New Bulgarian University, Sofia, Department "Natural sciences"

ABSTRACT

Dithiocarbamate Mancozeb is used in plant protection as fungicide. In recent years his amount used in Europe including Bulgaria significantly increased. Carcinogenicity of dithiocarbamates due to their metabolite - ethylene thiourea (ETU), which causes thyroid and pituitary tumors. According to Regulation (EC) No 396/2005 the allowable maximum residue level of ETU in food products is 50 μ g/kg. The purpose of this study is to determine the accumulated amount of ETU in products of fish, exposed on mancozeb in surround water in the allowable concentration limit. Seeking an answer to the question: is this concentration limit really safe for the end user?

The biological material used in this study was eggs and skeletal muscle tissue derived from rainbow trout (*Oncorhynchus mykiss*) reared in a Bulgarian fish farm. Daily in the pond while eating, mancozeb is added in quantities which could lead to the final concentration of 0.5 μ g/L, meeting the requirements of Regulation 9/16.03.2001 of the Bulgarian ministry of environment and water and Directive 2000/60/EC. The biological material was sampled at the end of the 60 day trial period of three batches "two summers old" fish. They were taken daily and two water samples (each in 2 L) from the pools, where fish are cultivated - one immediately before and the other 30 minutes after the addition of the fungicide.

For the determination of ETU in eggs and muscle of the fish has been developed a new method which comprises two steps - selective extraction and accurate quantification by HPLC. The method was validated by grading its linearity, accuracy, precision, recovery and limit of detection and quantification limit. The assays in all analysed samples (fish and water) are below the limit of quantification of the method, 50 μ g/L. The resulting conclusions of this research are: the maximal residue level of mancozeb is really safe for the the end user. Fish cannot accumulate its carcinogenic degradation product, ETU.

Key words: Mancozeb; Dithiocarbamates; Ethylene thiourea; Rainbow trout (*Oncorhynchus mykiss*); *HPLC*

EFFECT OF DIETARY VITAMIN E SUPPLEMENTATION IN PIGS ON THE FATTY ACID PROFILE OF MEAT DURING STORAGE

Assoc. prof. Teodora Popova*, PhD, Prof. M. Ignatova, PhD, Prof. P. Marinova, DSc

> Institute of Animal Science, 2232, Kostinbrod e-mail: tlpopova@yahoo.com;

ABSTRACT

The effect of vitamin E supplementation in the diet on the fatty acid composition of pork (m. Longissimus dorsi) during frozen storage was studied in 20 hybrid pigs. The animals were divided in two groups (n=10), one receiving a control diet and the other - a diet containing vitamin E in amount of 400 mg/kg feed during the finishing period. After slaughter, samples of the muscle were taken, frozen and stored at -20°C.

The fatty acid composition was analysed in the fresh tissue and after frozen storage for a period of 3 and 6 months. Data were statistically assessed by two-way ANOVA, the model including fixed effects ascribed to the treatment, storage duration and their interaction on the fatty acid profile of m. Longissimus dorsi. Vitamin E influenced considerably the fatty acid composition of the muscle. The contents of C14:0, C16:0 and C16:1 were significantly increased (P<0.001) while that of C18:0 was decreased (P<0.01) in the pigs of the supplemented group. In regard to the polyunsaturated fatty acids, vitamin E in the diet decreased considerably the proportions of C18:2n-6, C18:3n-6, C20:4n-6 (P<0.01). The duration of the storage had less influence on the fatty acid composition in the pork. The content of C14:0 decreased over time (P<0.001), while significant increase was observed for C18:0 (P<0.001) and the total amount of the saturated fatty acids (P<0.05) in the pigs from both control and supplemented group.

Key words: pigs, vitamin E, meat, fatty acid composition, storage

BIOLOGICAL EFFECTS OF SELENIUM SUPPLEMENTATION IN FEEDING SHEEPS, GROWN IN AN ENDEMIC MOUNTAIN REGIONS IN THE MIDDLE RHODOPES ON SELENIUM CON- TENT IN MILK AND DAIRY PRODUCTS

Borislav Blazhev¹

1.Central Laboratory for Chemical Testing and Control, Sofia 1330, bul. "Nikola Mushanov" 120,

E-mail: B.Blazhev@bfsa.bg

ABSTRACT

The large number of different sheep species, their wide adaptability to forage quality and high genetic potential, makes possible their wide- spread dissemination. However, an imbalance of the main biogenic com- ponents (selenium, iodine) in mountainous areas could adversely affect the milk production and the daily secretion of essential trace and macro ele- ments through the milk produced during lactation.

This study was conducted with 16 lactating sheeps from Rhodope Cigay breed and 16 sheeps from Karachan breed, grown in two different regions of the Middle Rhodopes village Borino and village Smilian. The animals were divided into two groups - a control and a deficient group. Feed was prepared according ARC-norms with different amounts of se-lenium during the experimental period. During the lactating period, the sheep in the control group were further supplemented "per OS" with 0.25 mg selenium / day in form of NaHSe03. Milk samples from both breeds were taken on the 30th and 60th day after the addition of selenium and were compared with the content of selenium at the beginning of the experiment. Low concentrations of selenium in feed plant species (Se $<60-70\mu g$ / kg feed) during lactation (April-June) affect negatively the concentration of Se in sheep milk. The concentrations in the ewe's milk increased reliably as a result of supplementation. Within 60 days, the levels of the element has reached normal values in the two control groups, while in the deficient animals has established critical values in the milk (20-30 µgSe / l). The effect of Se-supplementation on average daily secretion of macro and trace nutrients was studied, as well as the changes in the milk production during the survey period. Recommendations for the inclusion of selenium supplements for small ruminants, bred in the grazing period in endemic areas has proposed.

Key words: selenium deficiency, supplementation, ewe's milk, Se-mineral supplements

ECOPHYSIOLOGICAL METHOD FOR INVESTIGATION OF LETTUCE (*LACTUCA SATIVA L.*) VAR. ROMANA AND VAR. CAPITATA RESPONSE TO ABIOTIC STRESS FACTORS

Svetoslav Anev, Cvetelina Vasileva, Paola Dimitrova, Nikolina Tzvetkova, Nidal Shaban

University of Forestry, Sofia

ABSTRACT

Lettuce (*Lactuca sativa L.*) is the preferred vegetable for consumers in late winter and early spring. Therefore, the production of lettuce is done during unfavorable for life processes in plants autumn-winter season. Although growing of lettuce at this period is in a greenhouse, the plants are subjected to stressful abiotic factors such as air temperature, irrigation regimes and living space, whose influence is often crucial to product quality. The presented method involved a set of eco-physiological parameters to assess the impact of environmental stressors on biological productivity and the quality of the lettuce. The rate of photosynthesis and transpiration, the concentration of chlorophyll and of nitrates in leaves of lettuce were measured periodically. These measurements make it possible to be compared the potential of different varieties of vegetables to provide quality agricultural production under different growing conditions.

Key words: Lettuce, stress, photosynthesis, transpiration

INFLUENCE OF ABIOTIC STRESS FACTORS IN THE OCCURRENCE OF FUNGAL PATHOGENS ON LETTUCE (LACTUCA SATIVA L.) VAR. ROMANA AND VAR. CAPITATA IN SOFIA VALLEY

Zhelyu Avramov, Paola Dimitrova, Nidal Shaban, Milena Yordanova, Emilia Dudaklieva, Milena Radeva*, Sergey Bistrichanov

> University of Forestry, Sofia * Agrokonsult - RADI RADEV ET, Novi Pazar

ABSTRACT

The lettuce (Lactuca sativa L.) is one of the oldest vegetable crops and attacked by a large number of phytopathogens causing economically significant diseases and their degree of manifestation and assault are greatly influenced by climatic growing conditions. The aim of the current study was to detect diseases in lettuce induced by abiotic stress factors. Studies were conducted on 19 lettuce varieties in the Educational experimental field of the University of Forestry, located in the region of Sofia field. The plants were planted at three different conditions with three repetitions without treatments (three types of greenhouses - two unheated greenhouses and low tunnel covered with a polymer foil). The observations were conducted in the fall of 2015 and twice at the beginning and at the end of spring 2016. All plants which have shown symptoms of the disease were examined in the laboratory and the number of the loss was statistically recorded. Samples of plants with suspected viral infection were subjected to a serological analysis and fungal pathogens were isolated on nutrient media and a morphology (microscopic) identification was carried. Up to date, the results of the studies indicate that the viral infection has not been established yet. The fungal pathogens Verticilium spp., Botritis spp. and Septoria spp., causaing plant diseases were established and identified. The largest attack and manifestation of gray mould was found in three varieties and the degree of collapse of the plants reached more than 90%. Next year this study will continue with the support of the Agrokonsult company and will be funded by The Scientific Research Centre of the University of Forestry in Sofia.

Key words: Lettuce, stress, photosynthesis, transpiration

DDT RESIDUES STILL IN OUR FOOD: MYTH OR REALITY?

Iskra Stoykova, Rumyana Dimitrova, Ruslan Dimitrov, Todorka Yankovska-Stefanova

Central Laboratory Of Veterinary Control And Ecology, Bulgarian Food Safety Agency

ABSTRACT

Pesticides are the only compounds made advisedly to be toxic. However, less than 5% of applied pesticides reach the target organisms. The other part remains in the environment, afterwards incorporates into the food chain affecting ecosystems and all human beings. It is

proved, that pesticides cause cancer and are toxic to the nervous system, reproductive organs, and endocrine system.

The use of pesticides starts with the development of dichloro-diphenyl-trichloroethane (DDT) as an insecticide in 1939. Because of it's high toxicity, bioaccumulation and a half-life of nearly 100 years, DDT is further included to the group of persistent organic pollutants (POPs) and was banned with the Stockholm convention. Eventhough it has not been used in Bulgaria for 50 years, DDT residues are still found in the food. Moreover, large amounts of unused organochlorine pesticides are left for storage at different locations in the country, for future disposal.

Herein, we report data about the determined residues of DDT in meat, fish and diary samples. The submitted results were analyzed at Central Laboratory of Veterinary Control and Ecology in the period 2014-2016.

The highest concentrations of DDT were found in fish samples- 0.26mg/kg on average, but the lowest values were found in the pork meat- 0,001mg/ kg. 100% of the samples from free-grazing animals (sheep and goats) contain DDT residues, unlike the farm animals (poultry and swine) where DDT residues were found respectively at 20% and 12% of the analyzed samples. All the results were below the maximum residue levels set in Regulation 396/2005, which means they are edible.

Nowadays, DDT residues are still present in the food. It's persistence and bioaccumulation in the environment are potential risk to the consumer's health. Therefore, food safety is an integral part of the Bulgarian policy, by means of the National Pesticide Monitoring Program, performed by Bulgarian Food Safety Agency.

FEEDINF VALUE OF MAIN BULGARIAN FEEDSTUFFS – CURRENT DATA AND SCIENTIFIC OPINION

Prof. Assoc. Mariana Petkova

Institute of Animal Science Kostinbrod, +359 887 148 754; m_petkova2002@abv.bg

ABSTRACT

Feeds have been proposed as a main factor for modern animal production and final outcome from animal husbandry. Importance of the optimal nutritional conditions coupled with reduced production costs and low risk for animal health have led many to predict that all feeds produced must be safe and its chemical characterization and feeding value should not mislead the user. However, for this to become a reality, the quality of feeds at a relevant manner must be evaluate regularly. In this paper, the actual data on nutritive value as evaluated by chemical characterization of main feeds in Bulgaria are summarized and analyzed. The total number of the feeds included in the study is 238 and period of the sampled is 2008-2011. Chemical analyses were made at DAIRYLAND LABORATORIES INC, Arcadia, Wisconsin in USA. Analytical and calculated data for: concentrate feeds (33); hays (41); haylages (24); silages (83); straws (3); meals (13); brewer grains (3); DDG (5); TMRs (30); sugar beets (3) etc were present. The comparison between main indicators for feeds

before and after this study is made and a scientific opinion on this subject was express. In conclusion, we offer the "Catalogue of Bulgarian feeds material" to be compiled as well as to complete with other feeds produced in Bulgaria.

Key words: feedstuffs, composition, nutritional value, Bulgaria

НЕУТРОФИЛИ И СЪОТНОШЕНИЕТО НЕУТРОФИЛИ- ЛИМФОЦИТИ В КРЪВ ЗА ОЦЕНКА НАТОКСИЧНОСТТА НА ХРАНИТЕЛНИ ДОБАВКИ КАТО ЕВТИН И НАДЕЖДЕН МЕТОД

Румяна Тодорова

Институт по биофизика и биомедицинско инженерство, БАН

РЕЗЮМЕ

Neutrophil–Lymphocyte Ratio (NLR) was estimated to be an important predictor of vascular diseases such as coronary artery disease, acute coronary syndromes and heart failure, diabetic nephropathy, inflammation and other conditions. Here is proposed the prognostic value of NLR for estimation the eligibility of new proposed food additives as a marker linked to immunity. Neutrophil count and NLR could be predictive and reliable tests for determination of toxicity of new proposed food additives as well as for long-time effect of already approved food additives in the market. The NLR values of patients using plant additives for different disorders or as stimulants could be easily calculated because NLR is a cheap, predictive and prognostic marker for early-stage diseases as well for toxicity (as immediately expressed or long-time effect).

SAFETY ASSUARNCE OF CANNED FOODS BY ADEQUATE PROGRAMMES FOR INTERNAL PRODUCTION CONTROL

Svetoslav Alexandrov^{*1}, Milena Ruskova¹, Todorka Petrova¹, Velitchka Gotcheva^{**2}, Angel Angelov², Nikolay Penov²

1.Food Research and Development Institute,154 Vasil Aprilov Blvd., 4000 Plovdiv, Bulgaria

2. University of Food Technologies, 26 Maritza Blvd., 4000 Plovdiv, Bulgaria

ABSTRACT

Canned foods are a large group of products presented by the definition "commercially sterile food in hermetically sealed containers". The wide variety of raw materials, the complexity and number of technological operations for the production of canned foods determine a large number of hazards that must be controlled. The present review presents safety criteria for raw materials for the production of canned foods, control of production hygiene, process control and regimes for ensuring industrial sterility of the end products. The article discusses development of adequate internal programmes for control aiming to assess raw materials and product compliance with legislative criteria, and to ensure control of production hygiene. The programmes are also an important element for verification of the monitoring of critical control points (CCPs) and of the efficiency of the implemented critical limits

Key words: canned foods, commercial sterility, safety criteria, programmes for production control, verification

PRACTICAL GUIDELINES FOR SCHEMES TO DETERMINE THE DATE OF MINIMUM DURABILITY OR THE 'USE BY' DATE OF MILK PRODUCTS

Assist. prof. Totyo Kolev¹, assoc. prof. Nikolina Naydenova, PhD², Galina Dicheva³

1. Agricultural institute-Stara Zagora, +359 877 436 441,e-mail: totyo.kolev@abv.bg

2. Faculty of Agriculture, Trakia University, town of Stara Zagora, +359 (0) 42 699 354, e-mail: n_naidenva@uni-sz.bg

3. Agricultural institute-Stara Zagora, +359 (0) 42 606 991

ABSTRACT

Normally, each consumer see the label, when buy food and dairy products, where it is recorded the expiry date or date of minimum durability. When buy food and dairy products, normally, each consumer see the label, where is recorded the expiry date or date of minimum durability. Producers have to record specific expiration date or date of minimum durability on the label according to Community law. They have to analyze milk and milk products to determine to what point under specified conditions of storage, respectively kind of container would be safe and quality for consumption by the users.

These guidelines are intended to give details about the stages and indicators that manufacturers should carry out of milk and milk products in determining the expiry date or date of minimum durability.

Key words: milk products, date of minimum durability, 'use by' date, food safety and quality

WHY DO WE NEED COBALT AND VITAMIN B12?

Radostina Alexandrova¹, Tanya Zhivkova¹, Desislav Dinev¹, Radoslav Todorov¹,², Abedulkadir Abudalleh¹, Daniela-Cristina Culita³, Gabriela Marinescu³, Ramona Tudose⁴, Otilia Costisor⁴, Luminita Patron³, Orlin Alexandrov⁵

1.Institute of Experimental Morphology, Pathology and Anthropology with Museum, Bulgarian Academy of Sciences

2. Faculty of Medicine, Sofia University "St. Kliment Ohridski", Sofia, Bulgaria

3.Institute of Physical Chemistry "Ilie Murgulescu", Romanian Academy, Bucharest, Romania

> 4.Institute of Chemistry, Romanian Academy, Timisoara, Romania 5.Health Service, Gorna Malina, Bulgaria

ABSTRACT

Cobalt is one of the most important trace elements in the world of animals and humans. In the form of vitamin B12 (Cobalamin), this metal plays a number of crucial roles in many biological functions: DNA synthesis, formation of red blood cells, maintenance of the nervous system, growth and development of children.

The human daily dietary intake of cobalt has been estimated to be about 40-50 ng. An adult person contains up to 1 mg of cobalt which means that this metal does not accumulate. Because the absorption of cobalt is coupled with that of iron, in the case of iron deficiency cobalt is absorbed to a greater extent.

Cobalamin occurs in substantial amounts only in foods derived from animals like liver, kidney, meat, eggs, dairy products, and its deficiency is highly prevalent in vegetarians. In humans, the vitamin is required in trace amounts – approximately 1 microgram/day.

Vitamin B12 deficiency (<148 pmol/L) induces different pathological states such as anemia and neuropsychiatric disorders. It is associated with adverse maternal and neonatal outcomes, including developmental anomalies, spontaneous abortions, preeclampsia, and low birth weight (<2500 g). Cobalt deficiency symptoms in ruminant animals include a loss of appetite, emaciation, weakness, anemia, and decreased production. There is evidence to support the role of this metal in immune processes. Some cobalt containing compounds are proved to possess anti-neoplastic activity.

The cobalt and its compounds have relatively low acute toxicity. The acute toxicity of water-soluble salts is higher than that of insoluble ones. The carbonyls of cobalt are the most toxic cobalt compounds. The increased intake of cobalt can lead to hypothyroidism and thyroid hyperplasia, because Co(II) ions are inhibiting the uptake of iodine into the thyroid gland.

Key words: trace elements, cobalt, vitamin B12/Cobalamin, nutrition, deficiency, toxicity

Acknowledgement: This study was supported by the National Fund "Scientific Research", Bulgarian Ministry of Education and Science - Grant N_{D} DFNI 5 02/12.12.2014 and a bilateral project between Bulgarian Academy of Sciences and Romanian Academy. R. Todorov was a volunteer in the Laboratory.

THE CHALLENGES OF CADMIUM

Radostina Alexandrova¹, Lora Dyakova², Vladimir Veselinov¹,³, Boyka Andonova-Lilova¹, Milena Glabcheva¹, Orlin Alexandrov⁴

1.Institute of Experimental Morphology, Pathology and Anthropology with Museum – Bulgarian Academy of Sciences, Acad. Georgi Bonchev Str., Block 25, Sofia 1113, Bulgaria

2.Institute of Neurobiology, Bulgarian Academy of Sciences, Acad. Georgi Bonchev Str., Block 23, Sofia 1113, Bulgaria

3.Faculty of Medicine, Sofia University "St. Kliment Ohridski", Sofia, Bulgaria 4.Health Service, Gorna Malina, Bulgaria

ABSTRACT

Cadmium (Cd) is an environmental contaminant that cannot be destroyed in nature. Cd exposure is wide spread in the general population and occurs through inhalation or ingestion. Intestinal absorption is greater in persons with iron, calcium, or zinc deficiency.

The biological half-life of this metal is long mainly due to its low rate of excretion from the body. It accumulates in a variety of tissues, including kidneys, liver, central nervous system and peripheral nervous system.

Cd is toxic for humans and animals – in can cause numerous cell and tissue injuries through various mechanisms such as oxidative stress, epigenetic changes, impairment in transport pathways, disorders in membrane structures and behaviour, inhibition of respiration, etc.

Clinical symptoms of Cd toxicity depend on route, quantity, and rate of exposure. The age, sex, diet, lifestyle and state of health are also important.

The chief organ of toxic impact in human is the kidney. In addition this metal affects cardiovascular system, hematopoesis, immune response. Cd has been reported to possess endocrine disruptor capacity and to be e metalloestrogen. Cd exposure has been connected also with the development of insulin resistance. The United States Environmental Protection Agency considered Cd to be a class B1 carcinogen. The metal has been proved to induce genetic and epigenetic changes, to act as a mitogen, to inhibit DNA repair and cell death. The epidemiologic data show causal association between cadmium and prostate, breast, and lung cancer.

Detoxification of cadmium with EDTA and other chelators is possible. There are data about protective effects of essential metals, vitamins, edible plants, phytochemicals, probiotics and other dietary supplements against Cd toxicity.

Key words: Cadmium, Food, Toxicity, Protective and detoxification strategies

DNA BARCODING ANALYSIS OF YEASTS FROM BULGARIAN FOOD PRODUCTS

Associate professor, dr. Dilnora Gouliamova¹, dr. Roumen Dimitrov²

1.The Stephan Angeloff Institute of Microbiology, Bulgarian Academy of Sciences, tel: +(3592) 979 31 82; e-mail: dilnorag@gmail.com;

2.Sofia University "St. Kliment Ohridski", Department of Physics

ABSTRACT

Food is the most studied ecosystem because food industry has a considerable economical value. The same time spoilage of food has a huge impact on consumer's health. The major food spoilage species is *Zygosaccharomyces bailii*. It is characterized by resistance to preservatives, osmotolerance and high fermentative ability. Other yeasts frequently occurring in food products are *Candida parapsilosis, Pichia membranifaciens, Debaryomyces* hansenii and *Rhodotorula*. During a yeast biodiversity survey in selected Bulgarian ecosystems conducted in 2008-2011 we have isolated yeast strains from boza, bialo salamureno sirene and kiselo mliako. Yeast strains were identified using DNA barcoding analysis. Strains that were isolated from three Bulgarian boza products were identified as – Isatchenkia orientalis, Candida inconspicua and Pichia fermentans. DNA analysis demonstrated that five bialo salamureno cheeses contained yeasts *Kluyveromyces marxianus, Saccharomyces serevisiae and Rhodotorula mucilaginosa*. The analysis showed that three kiselo mliako products contained yeasts Kluyveromyces marxianus and Rhodotorula mucilaginosa. Moreover, DNA barcoding analysis showed that two yeast strains represent novel yeast species.

Key words: DNA barcoding analysis, Food associated yeasts

GLADIOLUS – AN ALTERNATIVE CROP OF VEGETEBLES IN GLASS GREENHOUSE

PhD student Yordan Atanasov, Assoc. Prof. Dr. Ivanka Ivanova, Prof. Dr. Bistra Atanassova

Institute of Ornamental Plants – Sofia

tel.: +359-2-9366044, e-mail: iop_sofia@abv.bg

ABSTRACT

Gladiolus (*Gladiolus L.*) is an ornamental species resistant to herbicides commonly used to combat annual weeds at the production of early vegetables in unheated glass greenhouses. Its cultivation is an alternative to overcome the post-effect of these pesticides after harvest of the vegetable crops in early spring /March and April/. In this study gladiolus was planted following the gradual release of areas from green onions (*Allium cepa L.*). In order to control annual weeds, the onions were treated with the herbicide linuron, introduced after planting and before germination of plants in the month of October. The aim of the work was to establish the possible residual effect of this chemical on the growth and development

of gladiolus. The experiments were conducted with corms of variety "White Prosperity", in the period 2015-2016, in a greenhouse of the Institute of ornamental plants - Sofia. The plants were grown in two planting dates - 25.03 and 10.04.

In both years of the study, signs of phytotoxicity as well as abnormal growth and development of gladiolus due to post-influence of the applied to the green onions herbicide were not detected. It was found that plant height and length of cut flowers in the first year exceeded the same indicators in the second year by 7.66 cm and 5.41 cm and 7.96 cm and 2.2 cm respectively. The number of formed flower buds per plant and the diameter of flowers were greater in the second year, respectively 0.74 numbers and 0.54 cm and 0.53 numbers and 0.52 cm. The observed differences are not substantial for the ornamental quality of the plants. Taken together, the obtained results showed resistance of gladiolus to suggested post-effect of linuron and give us a reason to recommend this ornamental crop for growing in a greenhouse after application of linuron for the predecessor green onion.

Key words: gladiolus, cv. "White Prosperity", green onion, date planting, linuron

NOTIFIABLE BEE DISEASES

Delka Salkova, Kalinka Gurgulova

Institute of Experimental Morphology, Pathology and Anthropology with Museum, Bulgarian Academy of Sciences

National Diagnostic and Research Veterinary medical Institute "prof. Dr. Georgi Pavlov"

ABSTRACT

The honey bee colony is a super-organism with features in the epidemiology of infectious diseases where disease transmission should be taken into account both the individual and colony level. Bees are susceptible to many pests and pathogens that can affect their health. Serious infections may lead to bee mortality. Bee colonies may also be affected by various pests, predators and unfavorable environmental factors that aggravate the course of infectious diseases.

A number of parasites and diseases of honey bee are notifiable. This means that legislation at national and European level require notification to the competent authority in case of infection. Diseases of honey bees included in the list of the World Health Organization (OIE) are discussed. In different Member States, including in Bulgaria, are notified various diseases according to their prevalence and the risk of introduction. This concerns American and European foulbrood, parasitic fungus *Nosema apis and Nosema ceranae, tracheal mite Acarapis woodi, exotic parasite Aethina tumida* (small hive beetle), the mite Varroa destructor and Asian bee mites Tropilaelaps spp. The programs for disease surveillance and legislation for the protection of bees at European and local level are discussed.

In Europe from 2014 has been established the small hive beetle Aethina tumida. In Bulgaria are not found Acarapis woodi, *Aethina tumida and Tropilaelaps spp*.

Mite Varroa destructor is registered in Bulgaria in 1971. This exotic pest is a serious threat to honey bees worldwide. If this parasite is not controlled, the colonies have little chance to survive. Therefore, it is one of the best known examples of the negative effects of imported exotic pests and diseases.

Key words: Honey bees, (Apis mellifera), diseases, notifiable

PLANT OILS AS GREEN PESTICIDES – POTENTIAL POSSIBILITY TO CONTROL DISEASES AND PESTS INCUCUMBER GROWN IN GREENHOUSES

Vinelina Yankova, Dima Markova, Stoyka Masheva, Georgi Velichkov

Maritsa Vegetable Crops Research Institute (MVCRI), Plovdiv

ABSTRACT

Botanical pesticides are an alternative of synthetic chemical pesticides for pest control in modern ecological technologies. These products are not a threat for the environment and human health. Plant products have a number of advantages that make them preferable in modern organic agriculture. The range of these products is constantly expanding, which requires the mechanism of their action to be well known. During the period 2013-2014 a number of studies were conducted for establishment of the effectiveness of plant oils from mustard (Sinapis alba L.), hemp (Cannabis sativa L.) and varrow (Achillea millefolium L.) in concentration 0,5% and 1% against the powdery mildew (Podosphaera xanthii U. Braun & N. Snish. Comb. Nov. Syn. Sphaerotheca fuliginea) and cotton aphid (Aphis gossypii Glov.) in cucumber variety Kiara F1, grown in greenhouses. The effectiveness of plant oils in powdery mildew is compared with a standard Bayfidan 250 EC 0,02% (a.i. triadimenol). It was established a high effectiveness of the studied products compared to that of the standard. The 1% plant oils from hemp and yarrow demonstrate a good effectiveness (over 90%) to cotton aphid close to that of the standard Mospilan 20 SP 0,0125% (a. i. acetamiprid). The highest values of biological activity of the plant oils, included in the study were observed at 5th-7th day after treatment. The good effectiveness shown by the plant oils, gives us another alternative to control diseases and pests in greenhouse cucumbers.

Key words: Aphis gossypii, Podosphaera xanthii, cucumber, plant oils, effectiveness

RAPID ALERT SYSTEM FOR FOOD AND FEED (RASFF)

Branimir Kanakidis

Risk assessment center on food chain

ABSTRACT

Regulation (EC) No 178/2002 of the European Parliament and of the Council of 28 January 2002 laying down the general principles and requirements of legislation on food to create a European authority for food safety and laying down procedures on safety food. In the event that a member of the network has any information relating to the existence of a serious

direct or indirect risk to human health deriving from food or feed, this information is immediately notified in the Rapid Alert System for Food and Feed (RASFF). The information is immediately transmitted to the members in the network.

2.Legislation

Regulation (EU) No 16/2011 of 10 January 2011 laying down implementing measures for the Rapid Alert System for Food and Feed.

3. Types of notifications:

3.1. Alerts - message serious risk that requires immediate action or risk product already on the market.

3.2. Notifications of border rejection - concerns shipments of food, feed or raw materials which have been denied entry into the Community because of the risk to human health, animal health or the environment.

3.3. Notifications to follow up - do not require rapid response.

3.4. Notifications for attention - provide interesting information control authorities.

OBSOLETE PESTICIDES – CAUSES, CONSEQUENCES AND MEASURES TO SOLVE THE PROBLEM

Irena Bogoeva

Risk assessment center on food chain

ABSTRACT

Pesticides are chemical weapons, designed to fight diseases and plant pests. Despite the undeniable usefulness, their widespread use carries a number of risks and consequences for human health and the environment. A not inconsiderable problem with pesticides is the presence of large amounts of unused substances, so-called "obsolete pesticides", whose packages are damaged due to improper or prolonged storage. Following the weather, this has led to the entry of pesticides in the environment – through runoff and leaching into groundwater and farmland. This movement causes the penetration of these chemicals in agricultural production, in food for human consumption and feed, hence causing irreversible consequences for human health. Much of obsolete substances belong to the so-called persistent organic pollutants, which is the reason for their long-term accumulation in the environment.

Obsolete pesticides are unusable because they have suffered physical and/or chemical changes, resulting in the manifestation of phytotoxic effects on the target species or in an unacceptable risk to human health or the environment; products have suffered an unacceptable loss of biological efficacy, due to the dissolution of the active substances and/or other chemical or physical changes; their physical properties have changed to such an extent so that no longer can be administered by standard equipment. Most developing countries have accumulated outdated and deteriorated stocks of pesticides, that can no longer be used as

prescribed. The problem also relates to our country. In view of the dangerous nature of these pesticides, they must be disposed of in a manner which is safe for the environment. Long-term solution for the obsolete stocks in the application of preventive approaches, improved management and reduced stockpiles by appropriate and environmentally friendly measures and facilities.

VECTOR – BORNE DISEASES AND THE ROLE OF ENVIRONMENT TO OCCUR EMERGING RISKS

Svetoslav Stoev Risk assessment center on food chain

ABSTRACT

The emergence of new diseases have a wide range of predisposing factors. Over the past 60 years, more than 300 vector-borne diseases have emerged in humans, of which two thirds are of zoonotic origin (transmitted between humans and animals). About a quarter of the diseases that came out during the period 1990-2000 are vector transmitted. In fact, vector-borne diseases are particularly sensitive to environmental changes, in terms of changes in the susceptibility of the host, climate, plant cover and the movement of people and animals. The models describing changes in these factors serve to explain the mechanisms of transmission of these diseases and aim to draw up a proper assessment of the significance of various factors, scenarios for testing and monitoring and anticipate the future.

Vector-borne diseases are as transmiting only among animals (bluetongue in ruminants, lumpy skin disease in cattle) and zooantroponoses (Rift Valley Fever, heartworm).

Keywords: vector diseases, factors of transmission, role of the environment, emerging risks, Bluetongue, LSD, Rift Valley fever, heartworm

THE ORGANIC FARMING - A FACTOR FOR ENVIRONMENTAL PROTECTION

Miglena Peychinova1, Nevena Petrova1, Botyo Zaharinov2 1. Risk assessment center on food chain 2. New Bulgarian University

ABSTRACT

The transition from conventional to organic farming is a difficult time for every farmer. In recent years it became clear that modern conventional (intensive) farming is in crisis, because of the high use of chemicals and their impact on the environment (soil, water, landscape, biodiversity) and human health. The issues related to organic farming started to put more and more in society and the importance of the quality of food products is increasing constantly. There has been in recent years a steady trend of growth in the variety and range of organic products in food chain stores.

Key words: conventional farming; organic farming; GMOs; organic food and beverages, sustainable development strategy