Резюмета на научните публикации
на гл. ас. д-р Юлияна Милкова Иванова-Тумбева,
представени за участие в конкурса (извадка от общия списък)

Ju. Ivanova, G. Salama, R.M. Clancy, N.F. Shor, K.D. Nylander, D.A. Stoyanovsky, Formation of nitroxyl and hydroxyl radical in solutions of sodium trioxodinitrate: effects of pH and cytotoxicity, *J. Biol. Chem.* **278 (44)** (2003) 42761-42768. (IF_{2003} = 6,482)

Abstract. Despite its negative redox potential, nitroxyl (HNO) can trigger reactions of oxidation. Mechanistically, these reactions were suggested to occur with the intermediate formation of either hydroxyl radical ($^{\cdot}OH$) or peroxynitrite ($ONOO^-$). In this work, we present further experimental evidence that HNO can generate $^{\cdot}OH$. Sodium trioxodinitrate ($Na_2N_2O_3$), a commonly used donor of HNO, oxidized phenol and Me_2SO to benzene diols and $^{\cdot}CH3$, respectively. The oxidation of Me_2SO was O_2-independent, suggesting that this process reflected neither the intermediate formation of $ONOO^-$ nor a redox cycling of transition metal ions that could initiate Fenton-like reactions. In solutions of phenol, $Na_2N_2O_3$ yielded benzene-1,2-diol and benzene-1,4-diol at a ratio of 2:1, which is consistent with the generation of free $^{\cdot}OH$. Ethanol and Me_2SO, which are efficient scavengers of $^{\cdot}OH$, impeded the hydroxylation of phenol. A mechanism for the hydrolysis of $Na_2N_2O_3$ is proposed that includes dimerization of HNO to *cis*-hyponitrous acid (HO-N=N-OH) with a concomitant azo-type hemolytic fission of the latter to N_2 and $^{\cdot}OH$. The HNO-dependent production of $^{\cdot}OH$ was with 1 order of magnitude higher at pH 6.0 than at pH 7.4. Hence, we hypothesized that HNO can exert selective toxicity to cells subjected to acidosis. In support of this thesis, $Na_2N_2O_3$ was markedly more toxic to human fibroblasts and SK-N-SH neuroblastoma cells at pH 6.2 than at pH 7.4. Scavengers of $^{\cdot}OH$ impeded the cytotoxicity of $Na_2N_2O_3$. These results suggest that the formation of HNO may be viewed as a toxicological event in tissues subjected to acidosis.

D. Stoyanovsky, Y. Tyrina, V. A. Tyurin, D. Anand, D. N. Mandavia, D. Gius, **Ju. Ivanova**, B. Pitt, T.R. Billiar, V. Kagan, Thioredoxin and lipoic acid catalyse the denitrosation of low molecular weight and protein S-nitrosothiols, *J. Am. Chem. Soc.* **127(45)** (2005) 15815-15823. (IF_{2005} = 7,419)

Abstract: The nitrosation of cellular thiols has attracted much interest as a regulatory mechanism that mediates some of the pathophysiological effects of nitric oxide (NO). In cells, virtually all enzymes contain cysteine residues that can be subjected to S-nitrosation, whereby this process often acts as an activity switch. Nitrosation of biological thiols is believed to be mediated by N_2O_3, metal-nitrosyl complexes, and peroxynitrite. To date, however, enzymatic pathways for S-denitrosation of proteins have not been identified. Herein, we present experimental evidence that two ubiquitous cellular dithiols, thioredoxin and dihydrolipoic acid, catalyze the denitrosation of S-nitrosoglutathione, S-nitrosocaspase 3, S-nitrosoalbumin, and S-nitrosometallothionein to their reduced state with concomitant generation of nitroxyl (HNO), the one electron reduction product of NO. In these reactions, formation of NO and HNO was assessed by ESR spectrometry, potentiometric measurements, and quantification of hydroxylamine and sodium nitrite as end reaction products. Nitrosation and denitrosation of caspase 3 was correlated with its proteolytic activity. We also report that thioredoxin-deficient HeLa cells with mutated thioredoxin reductase denitrosate S-nitrosothiols less efficiently. We conclude that both thioredoxin and dihydrolipoic acid may be involved in the regulation of cellular S-nitrosothiols.

Pantcheva, **Ju. Ivanova**, R. Zhorova, M. Mitewa, S. Simova, H. Mayer-Figge, W.S. Sheldrick, Nickel(II) and Zinc(II) dimonensinates: single crystal X-ray structure, spectral properties and bactericidal activity, *Inorg. Chim. Acta* **363** (2010) 1879–1886. (IF_{2010} = 2,322)

Abstract. Mononuclear transition metal complexes of the polyether ionophorous antibiotic monensin (monensic acid, MonH) with nickel(II) and zinc(II) were prepared and characterized using single crystal X-ray diffraction and spectral methods. Monensin complexes crystallize as $[Ni(Mon)_2(H_2O)_2].H_2O.5MeCN$ (1) and $[Zn(Mon)_2(H_2O)_2].H_2O.4.5MeCN$ (2), respectively, in the monoclinic space group P21. Compounds 1 and 2 consist of two monoanionic monensic acid ligands (monensinates) bound in a

bidentate coordination mode to the transition metal ion. The metal center is placed in an octahedral environment with monensin anions occupying the equatorial plane of complexes via oxygens of carboxylate and hydroxyl groups located at the both ends of ionophore molecule. The strongly folded antibiotic anions are supported by intramolecular H-bonds, most of them originating from the two aqua ligands attached to the metal(II) ions in axial positions and completing their 6-fold coordination. The bioactivity assay reveals that the presence of divalent metal ion in the monensin complexes influences the biological properties of the ligand and should be taken into account when discussing its mode of action.

Ju. Ivanova*, I.N. Pantcheva*, M. Mitewa, S. Simova, H. Mayer-Figge, W.S. Sheldrick, Crystal structures and spectral properties of new Cd(II) and Hg(II) complexes of monensic acid with different coordination modes of the ligand, *Cent. Eur. J. Chem*. **8(4)** (2010) 852–860. (IF_{2010} = 1,065)

Abstract. The single crystal X-ray structures and the spectroscopic properties of complexes of monensic acid ($C_{36}H_{62}O_{11}.H_2O$) with toxic metal ions of Cd(II) and Hg(II) are discussed. The cadmium(II) complex (**1**) is of composition $[Cd(C_{36}H_{61}O_{11})_2(H_2O)_2]$ and crystallizes in the monoclinic system (space group P2(1), Z = 2) with a = 12.4090(8), b = 24.7688(16), c = 14.4358(11) Å, β = 91.979(7)°. Two ligand monoanions are bound in a bidentate coordination mode to Cd(II) via the carboxylate and the primary hydroxyl oxygens occupying the equatorial plane of the complex. The axial positions of the inner coordination sphere of Cd(II) are filled by two water molecules additionally engaged in intramolecular hydrogen bonds. The Hg(II) complex (**2**), $[Hg(C_{36}H_{60}O_{11})(H_2O)]$, crystallizes in the orthorhombic system (space group P2(1)2(1)2(1), Z = 4) with a = 12.7316(2), b = 16.4379(3), c = 18.7184(4) Å. The monensic acid reacts with Hg(II) in a tetradentate coordination manner via both oxygen atoms of the carboxylate function and oxygens of two hydroxyl groups. The twofold negative charge of the ligand is achieved by deprotonation of carboxylic and secondary hydroxyl groups located at the opposite ends of the molecule. Hg(II) is surrounded by five oxygen atoms in a distorted square pyramidal molecular geometry.

Y. Gluhcheva, V. Atanasov, R. Zhorova, M. Madzharova, **Ju. Ivanova,** M. Mitewa, Cobalt bioaccumulation in mouse blood plasma and liver, *Biothechnol. Biotechnol. Equip.* **24/2010/SE** (2010) 311-314. (IF_{2010} = 0,291)

Abstract. Heavy metals such as cobalt are shown to accumulate in various organs of humans and animals. Oral exposure of immature mice to cobalt compounds (cobalt chloride and cobalt-EDTA) led to significant increase in cobalt (II) concentration in blood plasma and liver. Pregnant balb/c mice in late gestation were subjected to cobalt chloride ($CoCl_2.6H_2O$) or cobalt-EDTA (Co-EDTA) treatment at daily doses of 75 mg/kg or 125 mg/kg which continued until day 30 of the newborn mice. Cobalt salts were dissolved and obtained from drinking tap water. Pure tap water was used as control. Mice were maintained in individual standard hard bottom polypropylene cages to ensure that all experimental animals obtained the required dose of cobalt salts. The newborn pups were sacrificed on days 18, 25 and 30 which correspond to different stages of development. Mice were weighed weekly and the experimental cobalt concentration was adjusted accordingly. Blood plasma and liver were used for measuring cobalt bioaccumulation. Cobalt (II) compounds showed differential bioaccumulation: higher concentrations were measured in the plasma compared to those measured in the liver. The effect depended on the type of compound used, dose, time duration as well as on the age of the experimental animals. Higher metal concentrations were detected in samples of mice treated with cobalt chloride compared to the samples exposed to Co-EDTA. The results indicate that 18-day-old mice are more sensitive to chronic exposure to cobalt compounds in high doses. Cobalt(II) concentrations in blood plasma may be used as a useful marker for diagnosing chronic exposure to cobalt compounds.

Y. Gluhcheva, M. Madzharova, V. Atanasov, R. Zhorova, M. Mitewa, E. Pavlova, **Ju. Ivanova**, The affect of cobalt salts on some weight indices in developing mice. *Acta Morphol. et Antropol.* **17** (2011) 89-98.

Although cobalt is an essential trace element long-term exposure and large amounts of its salts can have deleterious effects on humans and animals. Pregnant balb/c mice in late gestation were subjected to cobalt chloride ($CoCl_2.6H_2O$) or cobalt EDTA (Co-EDTA) treatment at daily doses of 75 mg/kg or 125 mg/kg. Cobalt compounds were dissolved and obtained from drinking tap water. Sodium EDTA (Na-EDTA) and pure tap water were used as controls. The newborn pups were sacrificed on days 18, 25 and 30 which correspond to different stages of development. Mice were weighed weekly and the experimental cobalt concentration was adjusted accordingly. Preliminary results showed that mice treated with cobalt salts ($CoCl_2$ and Co-EDTA) have smaller body weight compared to the control group. Liver weight was increased in the Co-EDTA-treated mice for both doses in all experimental groups. Spleen and liver weight was increased in case of high dose $CoCl_2$-treated mice. Spleen weight was the largest in high dose $CoCl_2$ –treated mice compared to all other groups. Liver weight of mice treated with Co-EDTA was the largest in all experimental groups compared to that induced by the other substances and in the control. The experimental results show that organic and inorganic cobalt salts affect body and organ weight.

Ju. Ivanova, I.N. Pantcheva, S. Simova, M. Mitewa, M. Tanabe, K. Osakada, Cd(II) and Pb(II) complexes of the polyether ionophorous antibiotic salinomycin, *Chem. Cent. J.* **5** (2011) art N 52. (IF_{2011}=1,65)

Background: The natural polyether ionophorous antibiotics are used for the treatment of coccidiosis in poultry and ruminants. They are effective agents against infections caused by Gram-positive microorganisms. On the other hand, it was found that some of these compounds selectively bind lead(II) ions in *in vivo* experiments, despite so far no Pb(II)-containing compounds of defined composition have been isolated and characterized. To assess the potential of polyether ionophores as possible antidotes in the agriculture, a detailed study on their *in vitro* complexation with toxic metal ions is required. In the present paper we report for the first time the preparation and the structure elucidation of salinomycin complexes with ions of cadmium(II) and lead(II).

Results: New metal(II) complexes of the polyether ionophorous antibiotic salinomycin with Cd(II) and Pb(II) ions were prepared and structurally characterized by IR, FAB-MS and NMR techniques. The spectroscopic information and elemental analysis data reveal that sodium salinomycin (SalNa) undergoes a reaction with heavy metal(II) ions to form $[Cd(Sal)_2(H_2O)_2]$ (1) and $[Pb(Sal)(NO_3)]$ (2), respectively. Abstraction of sodium ions from the cavity of the antibiotic is occurring during the complexation reaction. Salinomycin coordinates with cadmium(II) ions as a bidentate monoanionic ligand through the deprotonated carboxylic moiety and one of the hydroxyl groups to yield 1. Two salinomycin anions occupy the equatorial plane of the Cd(II) center, while two water molecules take the axial positions of the inner coordination sphere of the metal(II) cation. Complex 2 consists of monoanionic salinomycin acting in polydentate coordination mode in a molar ratio of 1: 1 to the metal ion with one nitrate ion for charge compensation.

Conclusion: The formation of the salinomycin heavy metal(II) complexes indicates a possible antidote activity of the ligand in case of chronic/acute intoxications likely to occur in the stock farming.

Y. Gluhcheva, I. Ivanov, V. Atanasov, N. Antonova, **Ju. Ivanova**, M. Mitewa, Hematological changes in case of chronic cadmium intoxication and monensin detoxication. Relationship with rheological variables, *Clin. Hemorheol. Micro*. **49** (2011) 417-422. (IF_{2011} = 2.838)

Abstract. The study evaluated the affect of chronic cadmium (Cd) and monensin treatment on some hematological parameters and its relationship with the rheological variables. Adult male mice were subjected to chronic treatment with cadmium acetate $[Cd(CH_3COO)_2 \times 2H_2O]$ (group 1), $Cd(CH_3COO)_2 \times 2H_2O$ followed by treatment with low dose monensin (group 2) and $Cd(CH_3COO)_2 \times 2H_2O$ followed by high dose monensin treatment (group 3). $Cd(CH_3COO)_2 \times 2H_2O$ and deprotonated monensin

were dissolved in distilled water and given daily to the experimental animals. Mice drinking distilled water served as a control group (group 4). Hematological parameters and erythrocyte morphology were evaluated in parallel with whole blood viscosity (WBV). Cd treatment reduced Hb and increased RDW. The addition of high dose monensin significantly improved erythrocytic indices compared to the control. Erythrocyte anisocytosis was observed in blood smears of Cd-treated mice corresponding to the increased RDW. WBV was significantly elevated in the experimental groups in the whole range of shear rates compared to the control group and in groups 2 and 3 was lower than in group 1 but remained higher compared to group 4. Correlations were found between WBV and RBC, Hb, Hct, MCV and RDW. The results suggest that hemorheological parameters such as WBV should be monitored in parallel with the hematological parameters when monensin is applied and heavy metal intoxication is suspected.

Ju. Ivanova*, I.N. Pantcheva*, R. Zhorova, G. Momekov, S. Simova, R. Stoyanova, E. Zhecheva, S. Ivanova, M Mitewa, Synthesis, spectral properties, antibacterial and antitumor activity of Salinomycin complexes with Co(II), Ni(II), Cu(II) and Zn(II) transition metal ions, *J. Chem. Chem. Eng.* **6** (2012) 551-562. (IF_{2012} = 0.95, CEPS)

Abstract. SalNa (sodium salinomycin) reacts with divalent transition metal ions of Co(II), Ni(II), Cu(II) and Zn(II) to produce novel compounds characterized by various spectroscopic methods. The interaction of metal (II) ions with SalNa results in the formation of mononuclear complexes of a general composition of $[M(Sal)_2 \cdot (H_2O)_2]$ nH_2O (n = 0 or 2) where the divalent cations replace Na^+ ions from the cavity of initial compound. The new compounds (disalinomycinates) possess an enhanced antibacterial activity against Gram-positive microorganisms as compared to both SalNa and SalH (salinomycinic acid), respectively. The metal (II) complexes manifest strong concentration dependent cytotoxic effect in experiments using human leukemia cell lines. The complexes of Co(II) and Cu(II) proved to exert superior activity as compared to the Ni(II) and Zn(II) analogues and are much more cytotoxic than SalNa and SalH. Further studies should be conducted to determine the therapeutic indexes of the new compounds.

Y. Gluhcheva, V. Atanasov, **Ju. Ivanova**, M. Mitewa, Cobalt-induced changes in the spleen of mice from different stages of development. *J. Toxicol. Environ. Health* A **75 (22-23)** (2012) 1418-1422. (IF_{2012} = 1.83)

Abstract. Cobalt(II) accumulates in organs such as spleen, kidneys, heart, and liver. The aim of the present study was to investigate the effects of cobalt ethylenediamine tetraacetic acid (Co-EDTA) on spleen of developing mice. Pregnant BALB/*c* mice in late gestation were subjected to Co-EDTA treatment at daily doses of 75 or 125 mg/kg in drinking water, which continued until d 90 of the newborn pups. The newborn pups were sacrificed on d 18, 25, 30, 45, 60, and 90, which correspond to different stages of development. Spleens were excised, weighed, and processed for histological analysis. Spleen index (SI) was calculated as a ratio of spleen weight to body weight. Cobalt(II) bioaccumulation in spleen was determined using flame atomic absorption spectrometry (FAAS). Preliminary results showed that chronic treatment of mice with low- or high-dose Co-EDTA disturbed extramedullary hematopoiesis in the spleen. The number of megakaryocytes was reduced compared to controls. SI was also reduced in d 18 mice treated with low- or high-dose Co-EDTA. However, exposure to 75 mg/kg led to an increase of SI in all other experimental groups. FAAS analysis revealed significant cobalt(II) accumulation in spleen of treated mice. The Co(II) levels in spleens of d 18 mice were highest compared to other experimental groups, indicating that at this period mice are more sensitive to treatment. Exposure to cobalt-EDTA resulted in accumulation of Co(II) in spleen, altered SI, and hematopoiesis. Immature mice appear to be more sensitive to chronic treatment than adults.

E. Pavlova, M. Madzharova, D. Dimova, Y. Gluhcheva, **Ju. Ivanova**, N. Atanassova**,** Effects of $CoCl_2$ and Co-EDTA on testicular morphology and sperm count in mouse, *Eur. Chem. Bull.,* **1(9)** (2012) 387-390.

Abstract. Cobalt (Co) is an essential trace element for mammals required for the synthesis of vitamin B12. When cobalt was applied chronically it tends to accumulate in different organs and tissues that can induce pathological alterations. We focused our present study on the comparative effects of chronic exposure to cobalt chloride ($CoCl_2 \cdot 6H_2O$) or cobalt EDTA (Co-EDTA) on testis and sperm count in mice of different ages. Both compounds were given in drinking water water with doses per day of 75 mg/ kg or 125 mg/ kg,

respectively. Treated animals were sacrificed at different time intervals (on days 18, 25, 45 and 60). The most obvious changes in testis morphology – depletion and retardations in germ cell development, disorganization of seminiferous epithelium and SCO (Sertoli-cell-only) tubules were observed on the studies of 45 and 60 days respectively. In early puberty, testis weight was reduced in similar extend (with 25%) after high doses of both Co compounds whereas in early maturity and adulthood this parameter was reduced significantly and more severe after exposure to high dose $CoCl_2$ as compared to Co-EDTA, probably due to the stability of the complex Co-EDTA and its weaker absorption. Concerning gonado-somatic index and sperm count in mature animals we found similar tendency of more adverse effects with high dose of $CoCl_2$. As a result of our work and data in literature we can conclude that the effects of chronic exposure to cobalt depend of the type of the compound used, dose, level, time duration as well as on the age of the animals under experiment. Cobalt could be considered as a possible risk factor for male fertility and health.

Y. Gluhcheva, E. Pavlova, V. Atanasov, **Ju. Ivanova**, Iv. Vladov, S. Ganeva, M. Mitewa, Effect of cobalt-EDTA on iron content in spleen and liver of immature mice. *Eur. Chem. Bull*., **1(10)** (2012) 442-444.

Abstract. Cobalt (Co) is an essential trace element and its accumulation affects the concentrations of other elements also. Co(II) is shown to compete with iron (Fe) for the transferrin receptor and to form a stable complex with haemoglobin thus affecting haematopoiesis. There are lack of data regarding the effect of chronic exposure to Co compounds on Fe content in spleen and liver of mice. The study deals with the effect of long-term treatment with cobalt-EDTA (Co-EDTA) on iron content in the spleen and liver of immature mice. Pregnant ICR mice were subjected to chronic treatment with daily dose of 75 mg/kg Co-EDTA which continued until day 25pnd of the newborn pups. Results show accumulation of Co(II) and altered Fe content in the spleen and liver of treated mice compared to age-matched controls with significantly increased Fe concentration in the livers of treated mice. The changes could explain impaired haematopoiesis and immune responses of exposed to Co(II) immature mice.

E. Pavlova, **Ju. Ivanova**, D. Dimova, Y. Gluhcheva, N. Atanassova. Alterations in adult mouse testis after subacute intoxication with cadmium and monensin detoxication. *Eur. Chem. Bull*. **1(11)** (2012) 463-465.

Abstract. Cadmium (Cd) is a heavy metal and a major environmental pollutant. The general population is exposed to Cd mainly via drinking water and food products. We have developed a mouse experimental model to investigate the *in vivo* effects of Cd and the chelating agent monensin on testis and sperm count during adulthood. Animals were divided into three groups: normal control (receiving distilled water and food): Cd group, exposed to 20 mg/kg b.w. Cd(II) acetate for the first 2 weeks of the experimental period and Cd+monensin group, receiving monensin (18 mg/kg) after Cd-intoxication (from 15^{th} to 28^{th} day). Histological observations of the testis demonstrated that Cd induced desquamation of germ cell and their assembly in the luminal region of the tubules. Areas in the testis without spermatides in latest steps of differentiation were also observed in this group. Monensin administration to Cd-treated animals restored histology of the testis to normal to a great extent (despite some Sertoli-cell-only tubules). Statistically significant changes in sperm count were not established for any of the experimental groups. Monensin can reduce injury of the testis and normalize its morphology after subacute exposure to Cd. The results of the present study demonstrated that monensin is a good compound in chelating therapy of some heavy metal intoxications.

Ju. Ivanova*, Y. Gluhcheva, K. Kamenova, S. Arpadjian, M. Mitewa. The tetraethyl ammonioum salt of monensic acid – an antidote for subacute cadmium intoxication: a study using an ICR mice model. *J. Trace Elem. Med. Biol.* **26(4)** (2012) 279-284, (IF_{2012} = 2.357)

Abstract. In this study, the ability of the chelating agent monensic acid (administered as the tetraethylammonium salt) to reduce the cadmium (Cd) concentration in the kidneys, liver, heart, lungs, spleen and testes of Cd-intoxicated mice was investigated. Chelation therapy with the tetraethylammonium salt of monensic acid led to a significant decrease of the Cd concentration in all of the organs of the Cd-treated mice. This effect varied from 50% in the kidneys to 90% in the hearts of the sacrificed animals (compared to the Cd-treated controls). No redistribution of the toxic metal ions to the brain of the animals

as a result of the detoxification with the chelating agent was observed. The detoxification of the animals with the antibiotic salt did not perturb the endogenous levels of copper (Cu) or zinc (Zn). The tetraethylammonium salt of monensic acid significantly ameliorated the Cd-induced total iron (Fe) depletion in the liver and spleen of Cd-treated mice. It also restored to control levels the values of transferrin-bound Fe and the total iron binding capacity (TIBC) of the plasma. These results imply that the tetraethylammonium salt of monensic acid could be an efficient antidote in cases of Cd-intoxication.

A. Gegova*, D. Mitkov, Y. Gluhcheva, S. Arpadjan, M. Mitewa, **Ju. Ivanova***. Effects of cadmium and monensin on the morphology of lung of mice, subjected to subacute cadmium intoxication. *Acta Morphol. et Antropol.* **19** (2012) 47-52.

The effects of cadmium (Cd) and monensin on the lungs of mice, subjected to subacute Cd intoxication were studied on ICR mouse model. The data demonstrated that Cd induced elevation of the lungs weight in Cd-intoxicated mice compared to the normal controls. The treatment of Cd-intoxicated mice with monensin recovered lungs weight to normal values, suggesting an ameliorative effect of the antibiotic on the lung function. Histopathological analysis of the lung tissue demonstrated that Cd induced circulatory and inflammatory alterations. Monensin administration to Cd-treated animals reduced the morphological alterations and restored histology of the lungs to normal in a great extent. These data were well correlated with the results from the atomic absorption that showed that monensin reduces the concentration of Cd in the lungs of the Cd-intoxicated animals by 30 % compared to the toxic control. Taken together the results presented in this study prove that monensin could be a promising chelating agent for the treatment of Cd-induced lung dysfunction.

Y. Gluhcheva, **Ju. Ivanova**, S. Arpadjan, M. Mitewa, Effects of cadmium and monensin on spleen of mice, subjected to subacute cadmium intoxication, *J. Toxicol. Environ. Health* A **76** (2013) 328-332. (IF_{2012} = 1.83)

Abstract. This study investigated the effects of cadmium (Cd) and monensin on spleen function in mice, subjected to subacute Cd-intoxication. Adult male ICR mice were divided into three groups (n = 6 per group) as follows: control group (received distilled water and food ad libitum); Cd-treated (20 mg/kg/b.w./day Cd(II) acetate for the first 2 weeks of the experimental protocol); monensin-treated mice (20 mg/kg/day Cd(II) acetate for the first 2 weeks followed by treatment with 16 mg/kg b.w./day monensin from days 15 to 28. On day 29, mice were sacrificed under light ether anesthesia. Exposure to Cd induced an increase in spleen index (SI). The treatment of Cd-intoxicated mice with monensin significantly reduced SI compared to Cd alone. The data from the atomic absorption analysis of spleen revealed a significant Cd accumulation in Cd-treated mice compared to controls, accompanied by a significant depletion of Fe concentration up to 30%. The treatment of the Cd-administered mice with monensin resulted in a significant decrease of Cd in spleen by 50% compared to Cd alone. Fe recovery occured in spleen of monensin-treated mice. Histopathological analysis of spleen showed that Cd significantly decreased the number of megakaryocytes and disturbed extramedullary hematopoiesis. The number of megakaryocytes increased when monensin was added. The data in this study suggest that monensin was able to reduce the effects of Cd on hematopoesis in mice.

Ju. Ivanova*, Y. Gluhcheva, D. Tsanova, A. Piskova, R. Djaleva, S. Mokresheva, D. Kamenova, M Mitewa, On the effect of chelating agents and antioxidants on Cd-induced organ toxicity. An overview, *Eur. J. Chem.* **4(1)** (2013) 74-84. (IF_{2012} = 0.64, data from the Webside of the Journal)

Abstract. Cadmium (Cd) has been classified as a human carcinogen. The World Health Organization (WHO) reported that the concentration of Cd in the environment has rapidly increased in the last few years. In many epidemiological studies, the correlation between environmental exposure of humans to Cd and diseases such as stroke, ischemia, renal and hepatic dysfunction, anemia, osteoporosis and diabetes has been discussed. For the treatment of heavy metal intoxications a therapy with chelating agents has been applied. A chelating agent is a compound that binds the toxic metal ion thus promoting its excretion by the living organisms. Recently, it has been found that Cd-induced toxicity is a result of formation of reactive oxygen species (ROS). These results increased the interest towards the antioxidants as possible agents for

the treatment of Cd-induced organ toxicity. Herein, we present summary and discussion of the literature data for the influence of chelating agents and antioxidants on Cd-induced pathological conditions in Cd-intoxicated animals.

D. Momekova, G. Momekov, **Ju. Ivanova**, I. Pantcheva, N. Stoyanov, M. Guenova, A Michova, K. Balashev, S. Arpadjan, M. Mitewa, S. Rangelov, N. Lambov, Sterically stabilized liposomes as a drug delivery platform for cytotoxic metal coordination compounds of salinomycin, *J. Drug Deliv. Sci. Tech.* (2013) in press. (IF_{2012} = 0.679)

Abstract. Sterically stabilized DPPC:CHOL:DSPE-PEG-2000 liposomal formulations of the lipophilic complexes of salinomycin with Na(I), K(I), Mn(II), Co(II), and Ni(II) ions were prepared by film-hydration method, at different drug-to-DPPC molar ratios. For the K(I) and Na(I) complexes optimal loading was established at a drug-to-DPPC molar ratio of 0.5:1, whereas for the Me(II) complexes it was encountered at 0.1:1. DLS revealed uniform LUV populations (130-160 nm) with monomodal size-distribution, further corroborated by AFM. Free and entrapped salinomycinates exhibited cytotoxicity in three human tumor cell lines, whereby the liposomal agents were superior *vs.* free complexes. DNA-fragmentation and flow cytometric assays showed that the cytotoxicity of free and liposomal salinomycinates is mediated by induction of apoptosis and G1 arrest. The ability of the carriers to retain the bio-activity of entrapped cargo gives us reason to conclude that the presented DPPC:CHOL:DSPE-PEG-2000 liposomes are suitable platforms for the salinomycin complexes, needing further evaluation and optimization.

Ju. Ivanova*, Y. Gluhcheva, K. Kamenova, S. Arpadjan, M. Mitewa. Monensin ammeliorates cadmium-induced hepatic injury in mice, subjected to subacute cadmium intoxication, *Biotechnol. Biotechnol. Equip.* (2013), accepted (IF_{2012} = 0.706)

Abstract. This research was designed to evaluate the potential application of monensin as an oral drug for the treatment of cadmium-induced hepatic dysfunction. The study was performed using ICR mouse model. 27 adult ICR male mice were divided into three groups, having nine animals in each group: control (received distilled water and food *ad libitum* for 28 days); Cd-intoxicated (treated orally with 20 mg/kg b.w. Cd(II) acetate from 1st to 14th days of the experimental protocol) and monensin treated group (intoxicated with Cd(II) acetate as described for the Cd-intoxicated group followed by an oral treatment with 16 mg/kg b.w. tetraethylammonium salt of monensic acid for 2 weeks). The results, presented in this paper, demonstrated that the treatment of Cd-intoxicated animals with monensin restored the liver weight/body weight index to normal values, decreased by 50 % the concentration of the toxic metal ion compared to the Cd-treated controls and recovered the homeostasis of Cu and Zn. Monensin reduced the activity of AST, ALP, ALT in the plasma of Cd-treated animals to the normal control levels and ameliorated the Cd-induced inflammation in the liver. Taken together these data demonstrated that monensin could be an effective chelating agent for the treatment of Cd-induced hepatotoxicity.

V. Atanasov, S. Stoykova, Y. Goranova, A. Nedzhib, L. Tancheva, **Ju. Ivanova**, I Pantcheva. Preliminary study on *in vivo* toxicity of monensin, salinomycin and their metal complexes. *Bulg. Chem. Commun.* (2013), accepted. (IF_{2012} = 0.171)

Abstract. The acute toxicity of polyether ionophores monensin, salinomycin and their metal complexes with ions of Na(I), Mg(II), Ca(II), Mn(II), Co(II), Zn(II) was evaluated on mice. The experimental data revealed that Ca(II) and Mg(II) complexes of salinomycin show the highest toxicity among the compounds tested with LD50 values of 20.5 mg/kg b.w (13 μmol/kg b.w.) and 25.8 mg/kg b.w (17 μmol/kg b.w.), respectively. The preliminary evaluation of biochemical indices of survived animals had shown that no significant changes occur within 72 h treatment with ionophorous antibiotics and their complexes.

*- показва статиите, в които гл. ас. Ю. Иванова-Тумбева е автор за кореспонденция

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