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Invited speakers

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Humic substances: from structure to application through plant action mechanism

Baigorri R.^{1,2}, Urrutia O.², Erro J.², San Francisco S.¹,García-Mina J.M.² ¹Timac Agro, Orkoien, Spain. roberto.baigorri@timacagro.es ²University of Navarra, Pamplona, Spain

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Humic substances (HS) are the Organic Matter component in soils that connect the mineral phase with the vegetation cover. Like an interphase connecting the no-living and the living world through a huge quantity of chemical and biochemical reactions. And this marvelous ecological role is possible because of the high versatility of HS.

We previously describe the HS adaptability to different soils (acidic, alkaline, sodic, saline...) in relation to the structural complexity in function to these soil characteristics. Thus Gray, Brown or Fulvic fraction, showed different molecular arrangement and structure. And how humification is an ordered and no randomness process in which composting is a very early step. In a biological point of view, we described the hormonal mechanism in which HS are involved when are in contact to plants promoting development and growth in both root and shoot.

Finally, here we show the applicability of HS in the manufacturing of different products:

1. a solid granulated fertilizer, in which a humic phase was applied to a sulphatephosphate matrix yielding the previously anion-metal humic complexes described in soils,

2. a humic based iron chelate protected by a second organic molecule (Fig. 1) to prevent the collapse of iron humate and,

3. livestock humic-feed in two different phosphate phases as monocalcium and dicalcium are, yielding phosphate-calcium-humate bounds in function of their different pH and crystallographic cell.



Figure 1. Humic-iron chelate protected by a second organic molecule.

Key roles of humic substances in global biogeochemical cycles

Cervantes F.J.

Engineering Institute, Universidad Nacional Autónoma de México (UNAM), Unidad Juriquilla, Querétaro, Mexico, E-mail: fcervantesc@iingen.unam.mx

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Microbial reduction of humic substances (HS) is a respiratory process involving extracellular electron transfer, which has significant impact on the transformation and fate of inorganic and organic contaminants in organic rich environments. The present work aims to provide an overview on the new scientific advances revealing novel roles of HS in key biogeochemical cycles. Anaerobic methane oxidation (AOM) linked to microbial reduction of HS [1] will be discussed. Also, the new findings indicating the potential role of HS on mitigating emissions of both methane and carbon dioxide in wetlands will also be examined [2]. Regarding the N cycle, the novel humic dependent-anaerobic ammonium oxidation [3] process will be presented and its potential role in marine N cycle will be discussed. Moreover, recent findings indicating the potential role of HS on mitigating the missions of nitrous oxide from wetlands will also be presented [4].

References

- 1. Valenzuela et al. Appl. Environ. Microbiol. 2017. 83, e00645-17.
- 2. Valenzuela et al. Sci. Total Environ. 2019. 650, 2674-2684.
- 3. Rios-Del Toro et al. Environ. Sci. Technol. Lett. 2018. 5, 571-577.
- 4. Valenzuela et al. Unpublished data.

Regulation of methanogenesis over introducing of humic compounds to biocatalytic systems with complicated content

<u>Efremenko E</u>.^{1,2}, Senko O.^{1,2}, Stepanov N.^{1,2}, Volikov A.B.¹, Mareev N.V.¹, Perminova I.V.¹ ¹Lomonosov Moscow State University, Moscow, Russia, elena_efremenko@gmail.com ²Emanuel Institute of biochemical physics RAS, Moscow, Russia

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Landfills of solid household waste pose a serious long-term risk to the environment and public health. This is due to the active production of landfill gas, which mainly contains CH_4 and CO_2 with a low addition of H_2S , H_2 , and NH_3 . For this reason, in many developed countries, special procedures are used to minimize landfill gas emission. The search for new solutions to reduce the intensity of landfill gas production and the proportion of CH_4 in its composition is highly relevant.

CH₄ is formed as a result of the metabolic activity of methanogenic microbial consortia that are formed spontaneously under natural conditions and consist of hydrolytic and acetogenic bacteria as well as methanogenic archaea [1].

Recently, the inhibition of the methanogenic activity of anaerobic consortia by various humic compounds (HCs) containing various functional (carboxyl, phenolic, hydroxyl, etc.) groups, has been actively studied [2]. At the same time, it should be noted that under real conditions (in landfills), microorganisms involved in the formation of biogas are in a stable state due to both the formation of a stable consortia, high cell density, and immobilization (sorption predominantly) on various materials. In this regard, it seemed appropriate to study the effect of HCs on the methanogenic consortia and the methanogenesis process itself in the presence of naturally and artificially immobilized forms of anaerobic microorganisms [1].

The purpose of this work was to evaluate the activity of various natural methanogenic consortia using the bioluminescent luciferin–luciferase method of ATP determination and to compare this method with the traditionally used approach based on studying the characteristics of methanogenesis: the process efficiency and the methane content in the accumulated biogas. In particular, the new technique was applied to assess the effect of different concentrations of HCs. This approach was used to analyze the effect of introducing potassium humate and fulvic acids (1–10 g/L) into media with different methanogenic consortia producing biogas.

The increase in the concentration of potassium humate led to a decrease in the overall energy status of the cells and reduced methanogenesis efficiency. However, fulvic acids introduced into the media stimulated methanogenesis in half of the tested consortia, which was accompanied by an increase in ATP concentration in cell samples. So, a positive correlation between the metabolic activity of cells in biogas formation and the concentration of ATP was observed. ATP concentration control appears to be an attractive tool for finding compounds that suppress methanogenesis in landfills.

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References

- 1. Senko et al. // Catalysts. 2019. 9(4):326.
- 2. Stepanov et al. // Sustainability. 2019. 11:3158-11.

Living with microbes and stars: the discontinuous universe of natural organic matter

Hertkorn N.¹, Harir M.¹, Gonsior M.², Schmitt-Kopplin Ph.¹

¹Helmholtz-Center Munich - HMGU, German Research Center for Environmental Health, Research Unit Analytical Biogeochemistry (BGC), 85758 Neuherberg, Germany; hertkorn@helmholtz-muenchen.de

²University of Maryland Center for Environmental Science, Chesapeake Biological Laboratory, Solomons, USA

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Various astrophysical and astrochemical environments and conditions readily generate mixtures of complex molecules, commonly in the temperature range from 2500 -2.725 K (the temperature of the cosmic microwave background). The compositional diversity of these molecules exceeds that of our current terrestrial biomolecules by a substantial margin. In the solar system, carbonaceous chondrites which show carbon contents up to 5% are remnants from the initial times of solar system formation 4.54 billion years (Ga) ago; very few other geological traces from the Hadean epoch (>4 Ga) remain on Earth. Recently discovered organomagnesium compounds in meteorites (CHOMg) are indicators of thermal and shock stress during meteorite formation, i.e. they are the first identified geomolecules to relate organic molecular diversity and conditions of meteorite formation. CHOMg compounds in meteorites can be seen as conceptual soil analogues which existed at the time of the formation of the earth and even several millions of years before that. They connect the previously known set of extraterrestrial abiotic CHNOS molecules and the mineral phase. Thermostable CHOMg compounds might have selectively preserved specific organic molecules through organo-mineral-interaction over geologic time scales and are therefore relevant in origin of life studies.

Current terrestrial extreme environments such as Yellowstone National Park are organic chemo-diversity hot spots and reveal an unique chemo-diversity that has not yet been observed anywhere in aquatic surface environments on Earth. The formation of NOM preceded the evolution of life and at any time in Earth's history, coevolution occurred between prebiotic/abiotic molecules, NOM and primitive and higher forms of life. The near continuum of binding sites available to ions and organic molecules acts to buffer against environmental and chemical extremes in the geo- and biosphere, which could damage life because of their potent reactivity. This key supportive role of NOM in life processes is sustained by strong interactions between biological and geochemical cycles.

Modern organic structural spectroscopy enables a thorough description of NOM molecular features which reflect ecosystem conditions and formation history. Hence, NOM from terrestrial (soil), freshwater (lakes, rivers, groundwater), marine (estuarine, surface and deep ocean), atmospheric (secondary organic aerosols) and extraterrestrial sources show distinct individual characteristics which in conjunction will enable a lively representation of our boundless carbon and all other element cycles. This contribution will use recent examples of molecular characterization of NOM from various sources to illustrate avenues to a holistic contemplation of our Earth in an evolving universe.

Humic substances in the healthcare industry

Mirza M.A.

Department of Pharmaceutics, SPER, Jamia Hamdard, New Delhi-110062

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Currently humic substances are being marketed as non-prescription products in a range of categories across the globe. Different functionalities, potentials, abundance and low cost raw material of HS augur a future in therapeutic product category or in pharmaceutical excipient. But the way forward is not as easy as in case of non-prescription products (majorly safety and quality data needed). The associated impediments are,

- requirement of full battery of safety and efficacy data (depending upon claims and categories of the products);

- availability of consistent quality raw material;

- full chemical and analytical characterization;

- low yield and consumption of by-product;

- not well recognized by regulatory agencies.

So, it requires a consorted effort by different stake holders of product development. If planned meticulously, it can also be dealt in a collaborative fashion with experts of specific fields in fine tuning with regulatory requirements. The invited talk would touch upon these nuances in details.

Biodegradation of a petroleum-derived groundwater plume reveals the compositional continuum of dissolved organic matter

Podgorski D.C.¹, Zito P.², Smith D.F.², Cao X.³, Schmidt-Rohr K.³, Wagner S.⁴,

Stubbins A.⁵, Cozzarelli I.M.⁶, Bekins B.A.⁶, Spencer R.G.M.²

¹University of New Orleans, New Orleans, USA, dcpodgor@uno.edu

²National High Magnetic Field Laboratory, Florida State University, Tallahassee, USA

³Brandeis University, Waltham, USA

⁴Rensselaer Polytechnic Institute, Troy, USA

⁵Northeastern University, Boston, USA

⁶United States Geological Survey, Reston and Menlo Park, USA

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Quantitative and qualitative analytical techniques including dissolved organic carbon (DOC), excitation-emission matrix spectroscopy (EEMS), ultrahigh resolution mass spectrometry (UHR-MS), benzene polycarboxylic acid (BPCA), and nuclear magnetic resonance (NMR) spectroscopy were utilized to measure the molecular-level composition, structural, and reactivity continua of a dissolved organic matter (DOM) plume located at the National Crude Oil Spill Fate and Natural Attenuation Research Site, near Bemidji, Minnesota, USA. All analytical measurements indicate a smooth and continuous change in DOM composition along the centerline of the plume transect. Regardless of methodological selectivity or limitations, results from each technique tracked the same path along the DOM continuum. Moreover, we show that the reactivity of the DOM pool is dependent on the composition observed along the gradient. A detailed understanding of the relationship between the composition and reactivity continua of DOM will aid in our development of miniaturized (lab-on-a-chip) assays that provide quantitative field measurements of DOM reactivity in aquatic ecosystems.



Figure 1. PCA plot of the compositional continuum that results from biodegradation of DOM produced from an aliphatic organic carbon source in a confined aquifer. Changes in fluorescence, molecular-level composition, and structure across the DOM plume transect are smooth and continuous, corresponding with a simultaneous decrease in microbial reactivity. The discontinuity or gap in composition is an indicator of DOM from different source material and corresponding reactivity potential.

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Methodological approaches in assessing the effectiveness of the use of feed additives of humic nature in productive livestock

Stepchenko L.M.

Dnipro state agrarian and economic university, Dnipro, Ukraine, stepchenko2@gmail.com

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The spectrum of biological effects of feed additives and preparations of humic nature in industrial animal husbandry has a multi-vector dependence. At the same time, the selection of humic-containing raw materials (peat, brown coal, leonardite, sapropel, and also the products of the vital activity of worms) in terms of physicochemical, biological, and environmental characteristics can be put in first place. Only after the first stage it is possible to evaluate the biological effect of humic substances on the animal organism. The report gives a retrospective of methodological approaches for the selection of humic feed additives for various types of farm animals as adaptogens, immunomodulators and regulators of metabolic processes of biological production synthesis. In our studies, environmentally friendly peat was used from 3 different deposits of Ukraine, which were selected after analysis of peat samples from 18 deposits using the screening system which we developed for their assessment (Stepchenko L.M., Sedykh N.I. 2011). More than 30 years of experience in this area research allowed us to offer a certain sequence of experiments stages to assess the effectiveness of humic nature feed additives in animal husbandry. 1. Determination of biological characteristics of the studied feed additives and determination of the presence of acute and chronic toxicity. 2. Conducting one or more experiments on laboratory animals in order to determine the dose-effect relationship. 3. Determination in laboratory animals (mice and rats), the level of antioxidant, hepatoprotective and immunomodulating activities of humic nature food additives can also be established by biological markers of indicators of homeostasis. 4. The transition to checking the effectiveness of the use of feed additives for farm animals in an environment of livestock. Initially, 2-3 dosages are used, the effectiveness of which was detected in laboratory animals, taking into account the metabolic rate of the corresponding species of productive animals and taking into account the duration of their use and compatibility with veterinary measures that are carried out in the conditions of the economy. 5. Assessment of the animals' productive qualities, qualitative and quantitative characteristics of biological products. The report will also present methodological approaches to determine the degree of influence of humic nature feed additives on digestion processes, the degree of metabolic restructuring of the liver function, the main indicators of homeostasis, as well as quantitative and qualitative characteristics of the resulting biological products. Experiments on the use of feed additives obtained from peat were carried out on the basis of a research humic substances laboratory named after professor L.A. Christeva of the Dnipro State Agrarian and Economic University and agricultural enterprises of Ukraine. As a result of applying the proposed research approaches, we developed technological schemes for the application of such feed additives as "Huminat", "Hydrogumat" and "Humilid" for broiler chickens, laying hens, ostriches, pheasants, turkeys, pigs of various technological groups, as well as for cattle.

The colonic bioreactor – a forerunner model for future biotechnology (function, role, products & management)

Swidsinski A. Humboldt University, Charite Hospital, Chariteplatz 1, 10098, Berlin, Germany, e-mail: alexander.swidsinski@charite.de

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Biotechnology is a key to our future. Using a bioreactor as a tool, men produce already nowadays gas, food, enzymes, insulin and a diverse set of biologicals on a very large scale. Adapting a biotechnology only on its own purpose, mankind is borrowing the wisdom of life, originating from four billion years of evolution. Actually, our earth itself is a large bioreactor, with a highly complex composition of different subsystems, each of them again specialized for a distinct purpose. Colon, is one of the most sophisticated natural bioreactors and as an organ indispensable for all vertebrates. Colonic bioreactor produces energy, vitamins, hormones, neuronal active substances and is important for antigen presentation purposes using a complex polymicrobial community. These functions are not a random result of accidentally crossing microbiota, but a result of deliberate selection, steering and facilitation of complex biomasses. We are just at the beginning of unravelling advanced morphologic and functional solutions.

The lecture describes the principles by which our colonic bioreactor is maintained, its relation to our health, functional disorders, inflammation, the interaction with antibiotics, probiotics and – last but not least – humic acids. Different to antibiotics, and probiotics, humic acids are extremely complex and still chemically poorly characterized. That is due to the complexity of the microbiota and substrates that humic acids are originating from. However, exactly this complexity stresses their highly differentiated role in microbe mediated organic synthesis and degradation, which can be potentially used for the specific regulation of polymicrobial metabolism within our bioreactor.

Immunotoxicity and allergenic properties of humic acids isolated from peloid

<u>Zhernov Yu.V.</u>¹, Litovkina A.O.¹, Perminova I.V.², Khaitov M.R.¹ ¹National Research Center – Institute of Immunology FMBA of Russia, Moscow, Russia, y.zhernov@nrcii.ru ²Lomonosov MSU, Department of Chemistry, Moscow, Russia

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Currently, biologically active food additives (BAFA) based on humic acids (HA) are widely distributed, which leads to the appearance of clinical cases associated with development of atypical reactions after ingestion of these supplements. HA are known to possess a broad spectrum of pharmaceutical activity, including pronounced antiinflammatory and immunomodulatory properties. This fact stimulated us to study a complex evaluation of the immunotoxicity and allergenic properties of HA.

The low-mineralized silt sulfide mud (peloid) samples were collected during spring, 2017 from the Molochka Lake of the Samara region, Russia. HA were isolated from peloid samples using extraction with 0.5 M NaOH as described in [1]. The immunotoxicity and allergenic properties of HA were evaluated according to the methodological recommendations of the European Medicines Agency and Research center for medical application, Russia. [2]. The effect of HA on the humoral immune response was evaluated by the change in the number of antibody-forming cells (AFC) in the spleen of mice when they were immunized with sheep red blood cells (SRBC) [3]. The effect of HA on the cell-mediated immune response was evaluated on the model of induction of the type IV hypersensitivity reaction to hapten reagent 2,4,6-trinitrobenzenesulfonic acid (TNBS) (Sigma-Aldrich) [4]. The effect of HA on neutrophil activation was evaluated by phorbol 12-myristate 13-acetate (PMA) and zymosan was assessed by luminol-dependent chemiluminescence (CL) [5]. The serum total IgE level after intraperitoneal administration of HA to BALB/c mice was studied by the solid-phase ELISA.

HA extract was studied in complex tests on a female F1 (CBA x C57BL 6) hybrid mice and BALB/c mice weighing 18-24 g six weeks of age. Administered per orally in doses of 100 and 1000 mg/kg (p.o.), HA exhibited neither immunotoxic nor allergenic effects. In CL tests, HA administered in mice in a dose of 100 mg/kg (p.o.) during 14 days inhibited the production of reactive oxygen species by neutrophils. HA administered to BALB/c mice in doses of 100 and 1000 mg/kg reduced the intensity of systemic anaphylaxis.

In conclusion, the results of our experiments indicate that HA from peloids does not exhibit undesired immunotoxicity and allergenic properties and however pose questions for the further investigation of BAFA based on HA as a potential allergenic agent.

References

- 1. Agapov A.I., Avvakumova N.P. // Patent RU 2028149, July 20, 1999.
- 2. Neuman D.A. // Fd Chem Toxic. 1995. 33(10):887-894.
- 3. Ladics G.S. // Methods. 2007. 41(1):9–19.
- 4. Wirtz S. et al. // Nat Protoc. 2017. 12(7):1295–1309.
- 5. Lieberman M.M. et al. // Clin Diagn Lab Immunol. 1996. 3(6):654–662.

Methodology for immunotropic activity study of humic acids

Zykova M.V.¹, Trofimova E.S.², Danilets M.G.², Ligacheva A.A.², Mikhalev D.A.¹, Belousov M.V.¹

¹Siberian State Medical University, Tomsk, Russia, huminolog@mail.ru

²Goldberg Research Institute of Pharmacology and Regenerative Medicine, Tomsk, Russia

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Investigation of the humic acids (HAs) effect on various factors of cellular and humoral immunity is a currently important scientific field in immunopharmacology. We studied of immunomodulating properties for peat HAs. For HAs isolation, nine representative types of peat were taken from major peat bogs in the Tomsk region (Russia). The nine peat samples analyzed varied in the botanical composition, the degree of decomposition (from 5-10% to 40-45%), and the ash content (from 2.6% to 16.3%). The HA fractions isolated as described earlier [Zykova, 2018] using basic (NaOH) and pyrophosphate (Na₄P₂O₇) extractions, were designated as HAb and HAp, respectively. Methodology for immunotropic activity study of HAs consisted of several stages.

First step was the screening of HAs NO-stimulating properties (18 samples) of various concentrations (0.1; 1; 10; 50 and 100 µg/mL) in murine peritoneal macrophages cultures. All tested HAs exhibited stimulation of NO production. It was stated that HAp samples have higher activity than HAb samples. The second step consisted of two experiments that were carried out in murine peritoneal macrophages culture in comparison with lipopolysaccharide (LPS) as stimulated control and the standard LPS inhibitor antibiotic Polymyxinum B that blocks stimulating activity of LPS by binding with the endotoxin. Results showed that HAp samples are more preferable in activation of macrophages than HAb samples due to their immunotropic properties being independent from endotoxin presence. Absence of endotoxin in natural substance is preferable in development of immunotropic medications. The HAp samples of raised peats induced cells activation by classical path, increased NO production and reduced arginase expression. Results allowed to determining the most active HAp sample from the raised pine-cotton-grass peat (aHAp) as with addition of the LPS inhibitor in the cell culture its NO-stimulating activity did not decrease, but increased, arginase activity decreased. This indicates that aHAp sample have potential immunotropic activity mediated by Th1-dependent immune response. The third step was a study of the cytokine profile for aHAp sample that was carried out using key cytokines: IL-2, IL-4, IL-10, IL-12, TNF-α and INF-y. The results showed that the aHAp sample reduced the production of IL-10 against the background of stimulation of the production of key proinflammatory cytokines IL-2, IL-12, TNF- α and INF- γ by antigen-presenting cells. Thus, the aHAp sample showed the properties of a classic M1 activator, capable of supporting and enhancing the Th1 immune response. The fourth step was to study the effect of an aHAp sample on the reactions of the Th1 and Th2-dependent immune response in vivo. To assess the Th1-immune response, a mice immunization model was used by ram erythrocytes. The effect of the aHAp sample on the humoral immunity was evaluated by the number of antibody-forming cells in the spleen and the synthesis of hemacolutinins. The effect of the aHAp sample on the cellular immunity was evaluated by the delayed-type hypersensitivity reaction (this is a Th1-marker reaction). The course of administration of the aHAp sample reduced the intensity of the cellular immunity reaction and enhanced the indices of humoral immunity. This means that the aHAp sample really showed the properties of a classic M1 activator, meaning it can stimulate the immune response in infectious and inflammatory processes, chronic and oncological diseases. To evaluate the Th2-dependent immune response, an ovalbumin model of animal immunization was used. We investigated the level of IgG1 and IgE, degranulation reaction of mast cells and anaphylactic index by Weigle. The aHAp sample was found to the active inhibition of the synthesis of IgG1 and IgE, stabilization of mast cell membranes and suppress Th2-dependent immune response. This means that the aHAp sample has an antiallergic effect. Thus, HAs are promising immunomodulators.

Section 1 Humic substances as complex systems, their composition and properties

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Micromorphological features of humus plasma formation in ornitogenic soils of Maritime Antarctica (North-West part of Antarctic peninsula region)

Abakumov E.

Department of Applied Ecology, Saint-Petersburg State University, Saint-Petersburg, Russia, e_abakumov@mail.ru, e.abakumov@spbu.ru

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Soil cover of the Antarctic characterizes by relatively low thickness and spatial discontinuity of the superficial organo-mineral bodies and the absence of the most of the features that are specific for the "normal" soils (even for the high latitudes of the northern hemisphere). That is why micromorphological method is considered as most suitable for investigation of pedogenic features in the soils of Antarctica [1, 2, 3]. Soils, located on the Ardley Island, Fildes Peninsula were selected for fine sections monoliths sampling. One soil is normal organo-mineral soils under organic remnants of cereal plant - Deshampsia antarctica (62-14-385 S, 58-58-486 W). Second soil was sampled under recent penguin rockeries, which is now abandoned and faced to colonization of algae - Prasiola crispa and cereal plant - Deshampsia antarctica (62-14-391 S, 58-580549 W). Soil samples were air-dried and being passed through a 1-mm sieve for further chemical analyses. Fine sections of soil material were prepared form micro monoliths of soils, sampled in field. Samples were dried and saturated with resin. Thin sections were investigated with use of polarization microscope Leica DM 750 p in transmitted light and crossed nicols. The following soil micromorphological indexes have been investigated: soil microfabric, spatial arrangements of fabric units, soil particles distribution, elements of microstructure and character of organic matter. Soil formed under pure plant remnants without admixture of guano showed formation of humus plasma on the surface of primary minerals. This plasma forms the films or stains of organic matter, coating the mineral substrate. These morphological formations can be considered as initial signs of pedogenesis. The more developed stage of biogenic-abiogenic interactions is formation of soil aggregates, cemented by humic materials. This type of interactions is more typical for superficial layers of soils, formed under the sod horizons in locations of Deshampsia antarctica. Soils, formed under guano showed formation of specific organic plasma, derived from organic remnant of guano. This plasma plays an important role in aggregation of silt and sandy particles and their stabilization on the surface of soils, which important in conditions of stormy winds, and finally results in formation of stable soil cover on the territory of penguin rockeries. Moreover, the guano plasma coatings penetrate into the deepest soil horizons and result in enriching of all soil horizons by organic matter and mineral nutrients. This is a reason of initiation of humification in all horizons of soil, affected by ornitogenic effect on the topsoil layer. Thus, organic matter which is invisible on the macroscale, exist in soils of various environment of Antarctica and can be fruitfully investigated with the use of modern microscopic methods.

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References

- 1. Ilieva R. et al. // Bulgarian J. Ecolog. Sci. 2003. 2(304):52-54.
- 2. Kubiena W.L. // Antarctic J. 1970. 5(4):105-106.
- 3. Mergelov et al. // Sci. Reports. 2018. 8(1): paper № 3367.

To the question of the molecular structure of humic acids of soil humus

Bezuglova O.

Southern Federal University, Rostov-on-Don, Russia, Iola314@mail.ru

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There are still no clear molecular formulas for humic substances (HSs), all the suggested options have a schematic hypothetical character. In the second half of the 20th century, the concept of HSs of different origin was developed: they have a common principle of structure with a nucleus composed of an aromatic carbon skeleton substituted by alkyl and functional groups and a peripheral part enriched with polysaccharide and polypeptide fragments. However, some researchers believed that the HA molecule does not contain repetitive subunits and consists of randomized polymers, including various aromatic fragments. This concept is consistent with the hypothesis advanced by Piccolo [1], who suggested that HSs have a supramolecular structure. The supramolecular nature of humus acids could be considered proven by a number of instrumental techniques.

Of interest are studies focusing on the structural organization and parameters of these structures in soils of different genesis and in different soil horizons. The objects of our study were HAs (FAs) extracted from soils in Rostov region. The X-ray structural analysis of HAs was conducted using X-ray.

It has been established that the supramolecular organization of HAs in Chernozems and Chestnut soils can be described as a spatial structure composed of 2–4 layers of condensed aromatic systems supplemented by a network of chain fragments with different degrees of regularity and lengths. Structural parameters vary among soils of different genesis and genetic horizons of the same soil. Interplanar distances are distinctly registered; they vary from 0.349 to 0.371 nm. Down the profile an increase in the interplanar distances and a decrease in the thickness of sets of chain fragments are observed. It seems reasonable to assume that these primary particles of HSs consist of formations that represent spatial structures of several layers of condensed aromatic systems supplemented by a network comprised of chain fragments with different degrees of regularity and lengths, as is schematically represented in Figure.



Figure. Hypothetical structure of a humic acid structural fragment

Thus, our results suggest that humus compounds are organized as several oligomer molecules associated into more or less condensed systems forming the basis for mobile and steady supramolecular formations.

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References

1. Piccolo A. // Soil Sci. 2001. 166(11):810-832.

How does the complex structure and size of Boom Clay natural dissolved organic matter affect its reactivity towards radionuclides?

Buchatskaya Yu.^{1,2}, Salah S.², Durce D.², Devillers M.¹, Steudtner R.³ ¹Université catholique de Louvain, Louvain-la-Neuve, Belgium, ybuchats@sckcen.be ²SCK•CEN, Mol, Belgium ³Helmholtz-Zentrum Dresden-Rossendorf, Dresden, Germany

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In the context of radioactive waste disposal, Boom Clay (BC) is considered as one of the potential host rock formations for the storage of high-level and long-lived (HLW-LL) radioactive waste in Belgium. Boom Clay was deposited in seawater approximately 30 million years ago and is mainly composed of siliciclastic minerals, carbonates and organic matter. Significant part of the BC organic matter (up to 250 mg C/L) occurs in dissolved form, meaning it passes – by definition - a 0.45 µm filter. This dissolved organic matter (DOM) differs in size (from hundreds to thousands of Da, Fig. 1), solubility at different pH-values (humic and fulvic acids), structure (aliphatic versus aromatic) and reactivity (functional groups). Previous studies [1] showed that DOM can form complexes with uranium and thus, influence the geochemical behavior of uranium, however the complex structure of natural DOM was not taken into account so far.



Figure. 1. Mw distributions of BC DOM obtained by size-exclusion chromatography (SEC) coupled to UV-Vis 280 for DOM obtained by piezometer sampling (pore water DOM) and for DOM leached from BC sample from HADES underground laboratory in Mol, Belgium.

Therefore, two fractions of natural DOM were chosen to investigate their binding properties towards uranium: a "small" (< 1 kDa) and a "colloidal" (0.45 μ m – 100 kDa) fraction. The small fraction was separated using ultrafiltration with 1 kDa PES membranes from BC pore water sampled under in-situ conditions in the underground HADES facility (~-223m). The colloidal fraction (Ex-045-100) was separated by leaching DOM with UP water from a Boom Clay core, filtering it and recuperating the residue from 100 kDa PES membranes. The current study is focused on the isolation, purification, and characterization of the colloidal DOM fraction using UV-Vis, SEC, X-ray Photoelectron Spectroscopy, chemical and elemental analysis and potentiometric titrations. Besides this, the binding properties of the colloidal fraction constants using TRLFS and UC experiments.

References

1. Dierckx A. et al. Transport of radionuclides due to complexation with organic matter in clay formations(Trancom-Clay). EUR(Luxembourg) 2000.

Lignin in the humic acids of Chernozems of Transbaikalia

Chimitdorzhieva E.O., Chimitdorzhieva G.D.

Institute of General and Experimental Biology SB RAS, 6 Sakhyanovoi st., Ulan-Ude, 670047, Russia, erzhena_ch@mail.ru, galdorj@gmail.com

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Lignin structures are known to be included to the aromatic core of humic acids (HA), which form the organic part of soils (Fleig, 1971; Manskaya, Kodina, 1975; Tuev, 1989; Orlov, 1990; Chukov, 2001; Zavarzin, 2004). Lignin of different plant species has a varied chemical structure and molecular weight (Fang et al., 2017). Features of the climate of Transbaikalia: negative mean annual temperature (-4⁰ C), soils for a long time (5-7 months) and deep (up to 3-4m) frozen, short vegetation period, summer is short but hot, high solar radiation level (up to 3000 hours / year), low precipitation (200-300 mm / year), large fluctuations in the mean daily and monthly air temperatures. Such extreme conditions promote a high accumulation of stable carbon compounds in the form of lignin (up to 35%) in herbaceous plants as adaptation reaction. Grasses of Transbaikalia are peculiar, due to the imposition of latitudinal and altitudinal zonality. Hence, the aim of the study was to identify the quantity and quality of lignin phenols in herbs and their transformation into humic acids. The objects of the study are grassy vegetation and humic acids (HA) from the disperse-carbonate Chernozems of the Tugnuy Hollow of the Selenginsky Middle Mountain and guasigley Chernozems of the South part of the Vitim Plateau. Lignin was determined by mild alkaline oxidation of copper oxide in a nitrogen medium (Ertel, Hedges, 1984) at the Hanover University, Germany, with the advice of professors Gugenberger, and W. Zech from Bayreuth University. While determining lignin parameters in grasses from two subtypes of Chernozems, we obtained: 1) vanillin phenols are accumulated more intensive in herbs on permafrost Chernozems compared to steppe ones; 2) lilac phenols content is higher in vegetation from permafrost Chernozems too; 3) compositional ratio V: S: C of phenols in plants on permafrost soils was following: for forbs 5: 5: 1; for legumes 5: 3: 1; for cereals 1: 1: 1, and from steppe Chernozems, respectively: for forbs 2: 3: 1; for legumes 3: 2: 1; for cereals 2: 1: 1. It would seem that soil hydromorphism is more conducive to the accumulation of natural phenols in vegetation, but this is not noticeable in a 0-10 cm layer of guasigley Chernozems, where V: S: C = 2: 1: 1 versus from steppe one: 3: 3: 1. HA from permafrost soils contain significant amount of both vanillin and lilac structures, where V: S: C = 5: 4: 1, while in HA from steppe soils this ratio is 3: 4: 1. HA from both soils contain the same number of lilac structures that requires additional research. It should be assumed that initially high lignin content in vegetation should have contributed to a high accumulation of lignin in soil and, accordingly, in HA from permafrost soils. But the results show that there are more lignin phenols in HA than in soil. It should be assumed: 1) part of the aromatic fragments of lignin is included in the structure of the HA molecules; 2) most part of lignin transfer to humine, which reaches up to 71% of the total carbon in guasigley Chernozems; 3) other part remains in soil detritus (Chimitdorzhieva, 2016).

Preliminary data do not allow to explain the results because of a large complex of factors (climatic conditions, physical and chemical properties of soils, unstable redox potential, cryoarid microflora, grass species diversity, high solar radiation, and many others).

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Molecular mechanisms of humic substances stabilization

Chukov S.N.

Saint Petersburg State University, Russia, S.Chukov@spbu.ru

Keywords: humification, humic acids, 13C-NMR, polyconjugated aromatic structures

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Humic substances (HS) are one of the largest reservoirs of carbon accumulation in the biosphere. This becomes possible due to their high biochemical stability, which is acquired in the process of humification. The study of the molecular mechanisms of stabilization of HS in soils and other natural sites is one the fundamental problems of natural science.

An important factor in the physical and biochemical stability of HS is the presence in their composition of the cyclic type fragments and especially polyconjugated (condensed) aromatic structures. From the point of view of biothermodynamic and biochemical stability, cyclic structures (aromatic and heterocyclic) possess the greatest stability. Their content relatively increases with the depth of humification.

The mechanism of the formation and accumulation of polyconjugate aromatic structures in the soils HS molecules is still unclear. In the work of Lishtvan et al. (2012) revealed the formation of aromatic polyconjugation systems by destroying molecular fragments of an aliphatic structure and polymerization of monoaromatic fragments. At the stage of the genesis of brown coal, when there is a sharp decrease in the content of monoaromatic fragments and an increase in the content of polyconjugation systems, there is also an increase in the size of "planar" graphite-like structures (Lishtvan et al. 2012).

In our studies, it was shown by solid-state 13C-NMR spectroscopy in humic acids (HAs) of soils with a large degree of humification (chernozems and gray forest soils), there are poly-conjugated aromatic structures of the "coal" type. But we did not find any poly-conjugated aromatic structures in the soils HA of the taiga zone (the aromatic part is represented mainly by "scattered" nuclei). Apparently, in the HA of chernozems, we are dealing with purely soil biochemical processes of the formation of "humus coal". Many researchers attribute this stable organic substance to the fraction of humins associated with clay minerals (Krasilnikov, 2016; Wang e.a., 2014). But the discovery of such structures in HA preparations isolated from humus horizons by pyrolysis mass spectrometry (Schulten and Schnitzer, 1995) and NMR (Chukov, 1998, 2018; Nicole DiDonato, EA, 2017, etc.) indicates that the processes of formation of such stable compounds occur during humification during the formation of the "nuclear" part of HA macromolecules.

To decipher the molecular mechanisms of the formation of polyconjugate aromatic structures, it is necessary to study the HS in both natural and experimental conditions. The study of the HS of soils contrasting in the biohydrothermal conditions of humus formation, as well as the study of the "further" humification and decarbonization of HS in model laboratory experiments.

Chemical structure peculiarities of different organic matter pools in Haplic Chernozem of the Streleskaya steppe

Danchenko N.N.¹, Artemyeva Z.S.¹, Kolyagin Yu.G.², Kogut B.M.¹ ¹Dokuchaev Soil Science Institute, Moscow, Russia, nataly_danch@mail.ru ²Lomonosov Moscow State University, Moscow, Russia

Keywords: SOM, granulo-densitometric fractionation, CPMAS ¹³C NMR, structural features

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Soil organic matter (SOM) is heterogeneous in composition, properties, structure, functions, turnover and mean residence times of its components. Traditionally, the chemical nature of humic substances (humic and fulvic acids) extracted from the soil sample as a whole has been most thoroughly studied. Last decades, there has been increasing interest in the chemical structure of the organic matter of different localization in the soil matrix. In this context, the method of CPMAS ¹³C NMR spectroscopy is becoming increasingly popular due to the possibility of chemical characterization of low soluble organic substances without destruction.

The aim of our work was to study the chemical composition of OM of different localization in the soil matrix using the CPMAS ¹³C NMR method. The research subject was the virgin Haplic Chernozem (medium loamy) on the loamy loesses (Streleskaya steppe plot of the Central Chernozemic State Biospheric Reserve, Kursk region).

SOM pools were isolated by a modified variant of granulo-densitometric fractionation, which allows subdividing SOM into four pools: free OM (LFfr), occluded OM (LFocc), OM of clay (Clay) and OM of the residue of soil after the removal of LF and clay (Res).

It is shown that OM pools with different localization in the soil matrix differ significantly in their chemical structure. The data confirm the plant origin of free OM, its chemical structure is the closest to that of plant residues: ~ 50% of C contributes by O-Alkfragments, mainly from carbohydrates with various degrees of polymerization. Aromatic fragments making up 21.8% of C, are presented predominantly by guaiacyl, syringyl and cumaryl structural units of lignin. The decrease in O-alkyl C and the increase in aromatic C in passing from free to occluded OM, indicates a deeper degree of microbiological processing of organic material in the occluded OM, including such resistant to degradation components as cellulose, lignin and tannins. OM in clay is characterized by higher contribution of O-alkyl C and alkyl C as compared with occluded OM but lower proportion of aromatic fragments (by 2.1 times) and carbonyl groups (by 5.5 times). These structural features indicate the predominantly aliphatic nature of C associated with clay. Moreover, long-chain structures - the most hydrophobic component of OM predominate among alkyl fragments of clay: the signal intensity at 30 ppm was very high. A sharp increase in the contribution of peptides (1.5 times) along with the alkyl C in clay is apparently due to the presence of microbial products and cellular debris with a high content of alkyl fragments. The spectra of the Res are close to those of Clay, but have noticeable differences: the share of aromatic C in Res increases by 1.4 times, and alkyl, on the contrary, decreases by 1.4 times. Apparently, these differences are due to a greater degree of Res heterogeneity. In addition, the increase in the share of aromatic C may be due to the presence of black carbon components in this fraction.

Revealed structure peculiarities of the different OM pools, isolated by granulodensitometric fractionation, indicate the specificity of OM biodegradation processes occurred in various components of soil matrix.

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Metal-organic complexes as a major sink for rare earth elements in soils

Fedotov P.S.^{1,2}, <u>Rogova O.B.³</u>, Dzhenloda R.Kh.^{1,2}, Karandashev V.K.^{1,4} ¹National University of Science and Technology "MISiS", 4 Leninsky Prospect, Moscow 119049, Russia

²Vernadsky Institute of Geochemistry and Analytical Chemistry, Russian Academy of Sciences, 19 Kosygin Street, Moscow 119991, Russia

³Dokuchaev Soil Science Institute, 7 bld 2 Pyzhevskiy Per, Moscow 119017, Russia

⁴The Institute of Microelectronics Technology and High-Purity Materials, Russian Academy of Sciences, 6 Institution Street, Chernogolovka 142432, Russia

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The role of rare earth elements (REEs) in soil-plant systems has attracted an increasing attention but still remains somewhat unclear. Apart from in-vivo studies on the uptake of REEs by plants, in-vitro chemical fractionation of REEs in soil according to their physicochemical mobility can offer additional insights into the behavior of REEs. In the present work the fractionation of REEs has been studied by the example of background, aerially and hydrogenically contaminated soil samples using dynamic (continuous flow) extraction, which allows one to mimic natural conditions and minimize artefacts. The eluents applied addressed exchangeable, specifically sorbed, bound to Mn oxides, bound to metal-organic complexes, and bound to amorphous and poorly ordered Fe/Al oxides fractions extractable by 0.05 M Ca(NO₃)₂, 0.43 M CH₃COOH, 0.1 M NH₂OH·HCl, 0.1 M K₄P₂O₇ at pH 11, and 0.1 M (NH₄)₂C₂O₄ at pH 3.2, respectively. The distribution of trace metals (such as Pb, Cu, Zn, Ni) between separated fractions varies with sample and is dependent on the type of contamination. However, for all studied samples, the recoveries of REEs by pyrophosphate are surprisingly high, up to 40-45% of their total concentrations in background and anthropogenic transformed floodplain soils. The contents of REEs bound to Fe/Al oxides are relatively low, no more than 12%. REEs in other fractions may be taken into consideration only for aerially contaminated soil. To the best of our knowledge, such predominant association of REEs and metal-organic complexes (e.g. humic and fulvic compounds) independently on the type of soil has not been reported before.

To the best of our knowledge, the predominant association of REEs and metalorganic complexes in soils regardless the sample type and contamination is reported for the first time. This finding may offer a novel insight into the fate and behavior of REEs in soil-plant systems. The results should be extended to a variety of soil samples of different origin. Besides, it may be beneficial to apply another SEP (Mittermüller et al, 2016) under dynamic conditions and to compare the data obtained.

For the risk assessment, it would extremely important to study processes of adaptation of REEs entering the soil with various carriers such as phosphorus fertilizers, coal and phosphate dust, industrial wastes, etc.

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Spectral characteristics of water-soluble and alkaline-soluble humic substances of fallow soils

Giniyatullin K., Smirnova E., Okunev R.

Kazan (Volga region) Federal University, Russian Federation, ginijatullin@mail.ru

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Solutions of colored natural compounds isolated from soils, bottom sediments, brown coals, "crude oils" and asphaltenes are characterized by a continuous uniform increase in the absorption intensity with a decrease of wavelength in UV and visible spectral regions. The similar nature of the spectra can be explained in different ways: high molecular weight, wide polydispersity, random molecular structure, wide variety of chromophores, the effect of the "total spectrum" of the ensemble of molecules, as well as the combined effect of all the above reasons. Characterization of UV and visible spectra of uncertain composition compounds can be successfully used to quickly assess the direction of changes in natural organic matters (OM) in dynamic natural systems. The criteria SUVA₂₅₄ and Sr. are usually used to characterize the spectra. It is believed that the SUVA₂₅₄ criterion has a close correlation with the aromaticity of OM, and Sr with their molecular masses [1, 2].

The content of organic matter and its individual fractions in layered samples isolated from the old arable horizon were studied in different age (7 and 75-80 years) fallow soils. It was shown that with age of deposits there is a significant increase in the content of OM in the upper layers of the old arable horizon and the content of water-soluble and alkalinesoluble fractions. The qualitative composition of the labile OM fractions was evaluated according to the SUVA₂₅₄ and Sr criteria, which were calculated from the absorption spectra in the UV range. It was shown that in the upper part of the old arable horizon (0-5 and 5-10 cm) there is accumulation of water-soluble and alkali-soluble humic substances (HS) with a lower degree of aromaticity and higher molecular weight than the HS in the total thickness of this horizon, apparently due to the predominant accumulation of younger more aliphatic compounds under the fallow vegetation. With the age of the deposit, the aromaticity of water-soluble OM significantly increases, which may indicate a preferential selection of more water-soluble molecules that are more resistant to destruction. In the case of alkaline-soluble OM, this tendency is practically does not appear. It can be concluded that the factor of selective conservation of stable compounds for this fraction is not so relevant, and the stability of alkaline-soluble HS in soils is most likely due to their spatial inaccessibility to heterotrophic microorganisms due to the formation of supramolecular aggregates, as well as interactions with minerals and metal ions.

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References

- 1. Weishaar J.L. et al. // Environ. Sci. Technol. 2003. 37: 4702–4708.
- 2. Helms J.R. et al. // Limnol. Oceanogr. 2008. 53(3):955–969.

Soluble organic matter in the urban soils of Rostov agglomeration

<u>Gorbov S.</u>, Skripnikov P., Bezuglova O., Tischenko S. Southern Federal University, Rostov-on-Don, Russia, sngorbov@sfedu.ru

Keywords: Urbic Technosol, Chernozem, soluble organic matter

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The soil organic matter investigation is a necessary stage for solving many problems related to the effective soil resources exploitation and forecasting the techno-pedogenesis effects. Humus plays the major role in soil formation. On the one hand, it is very dynamic and sensitive to changes in the environment and/or anthropogenic impacts. On the other hand, it is a conservative part of a soil that has a certain tolerance towards disturbing impacts and can preserve its ecological status in the city conditions.

The aim of this paper was to discuss the specifics of accumulation and migration of soluble organic matter (SOM) through soil profile. The most mobile fraction of soil humus was isolated by cold and hot distilled water extraction according [1]. The SOM fraction was determined on a TOC-L CPN Shimadzu carbon analyzer with the solid sample combustion unit SSM-5000A.

The most typical soils were selected from seven soil profiles to identify patterns. All studied soils were divided into two groups depending on the level of transformation: natural soils (Haplic Chernozems (Calcic)) and anthropogenic soils (Urbic Technosols).

Haplic Chernozems (Calcic) are characterized by high humus content, but large amount of humic acids is strongly associated with calcium. As a result, SOM takes only a small part of soil organic matter and usually do not exceeding 0.14% for soil in whole, which corresponds to 4.0% in terms of C org. These values are much lower for Urbic horizons of anthropogenic soils and amounts to 0.04% to soil or about 2.0% in terms of C org. Natural soils are characterized by a smooth decline absolute and relative SOM values down the profile. In individual cases there is a slight accumulation of SOM was observed for carbonate barrier in horizon B. These processes are noted for soils with high content of total humus in surface humus accumulative horizons under forest vegetation where migration organic matter in the soil profile is most pronounced. Anthropogenic soils are characterized by a morphological two-fold profile. The upper part is represented by Urbic horizons, lower part is buried Haplic Chernozems, often deprived of upper horizon A to a depth of 20-30 cm. As a result, Urbic Technosols is characterized by the chaotic distribution of SOM in anthropogenic thickness and a smooth decline in the buried chernozem thickness. Besides, long-term conservation of soil profile under impermeable coatings contributed to lack accumulation of SOM into the carbonate barrier in the lower part of the profile.

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References

1. Gregorich E.G. et al. // Geoderma. 2003. 113, 237–252.

Molecular recognition in supramolecular systems of humic substances

Jorobekova Sh. Institute of Chemistry and Phytotechnologies, National Academy of Sciences, Bishkek, Kyrgyzstan; jorobekova@mail.ru

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The key processes in biology and medicine are molecular recognition. In these processes, there is the formation of non-covalent complexes due to ionic, van der Waals, hydrogen and coordination bonds, as well as hydrophilic and hydrophobic interactions. The strength of the non-covalent interaction between the two molecules is characterized by the dissociation constant Kd, which is related to the standard free energy by the equation ΔG^{o}_{free} = RT In Kd. Humic substances (HS) as supramolecular receptors bind various ligands (metal ions, hydrophobic organic compounds, cationic and anionic polyelectrolytes, etc.) with the formation of non-covalent complexes characterized by the corresponding values of dissociation constants. In particular, according to the results of our studies, the Kd for humic acids (HA) with various metal ions vary from 10^{-3} M to 10^{-10} M in dependence on the reaction conditions. Complexes of HA with hydrophobic organic compounds are characterized by Kd values from 10⁻³ M to 10⁻⁵ M. The strength of interpolyelectrolyte complexes (IPEC) is expressed by Kd values from 10⁻¹ M to 10⁻⁶ M. Corresponding to these Kd values, the free binding energy also changes. A characteristic feature of the molecular recognition by the humic receptors of the abovementioned ligands is their low specificity that can be explained by the dissipative nature of HS. Moreover, HS belong to systems with disordered bonds of elements that freely change their relationship to each other under various factors influence.

The conformational variability of HS has a great influence on the affinity for ligands. If necessary, part of the internal free energy of the HS goes to the establishment of conformation favorable for ligand binding and then ΔG_{free} decreases. Of particular importance are the hydrophilic/hydrophobic properties of HS. As a rule, the attachment of polar ligands to the HS occurs with the formation of the same hydrogen bonds that exist in the HS molecule. Hydrogen binding is a very important determinant of the specificity of molecular recognition. But, hydrogen binding does not always affect the change of free energy, since the energy that increases upon complexation represents a balance between the energy of hydrogen bonding in the complexes and the energy of cleavage of hydrogen bonds with the polar groups' ligands and HS. Hydrophobic compounds bind to HS differently depending on difference in their structure and provide a more accurate determination of the free binding energy in intermolecular interactions.
Determination of metals in humic acid salts by ICP-AES

<u>Karpukhina E.A.</u>, Vlasova E.A., Volkov D.S., Proskurnin M.A., Perminova I.V. Lomonosov Moscow State University, Moscow, Russia, karpukhina.evgeniya@gmail.com

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Humic substances (HS) consist of heterogeneous, difficult to separate mixtures of closely related organic macromolecules with a wide molecular weight distribution and a rich variety of functional groups. Due to many ways they interact with organic and inorganic substances in the environment, HS are of great importance for the biosphere. In particular, the high complexing ability of HS with metal ions has a significant effect on transport, deposition, and the presence of heavy metals in soils and water systems.

As humic substances are widely used as fertilizers in agriculture, the ability to determine the amount of heavy metals associated with HS in commercial samples is of great importance for predicting and accurately assessing the amount of certain metals introduced into the soil, as it is necessary to be able to evaluate both the toxic properties of the fertilizer and its nutritional properties. For example, humates of coal origins often contain arsenic, which is contained in the feedstock.

The best method for determining the metal content in humic samples is atomic emission spectroscopy with inductively coupled plasma (ICP–AES). It is widely used in biological, medical, and environmental studies, in particular for the determination of metals in waters, soils, and other natural materials. ICP–AES is a highly sensitive method, provides the results with high accuracy in the range from ultralow to macroscopic amounts, and simultaneously determines more than 70 elements.

As the objects of study, the salts of "Life Force" (sodium humate, Russia), "Sakhalin" (potassium humate, Russia) and "Powhumus" (potassium humate, Germany) humic acids were selected. An ICP-AES Agilent 720ES spectrometer with axial plasma view was used.

The following methods for the decomposition of humates were selected: direct introduction of aqueous solutions with and without preliminary centrifugation, nitric acid extract, fusion with lithium metaborate, and microwave decomposition with nitric acid. The following metals were determined: Ag, Al, As, B, Ba, Be, Ca, Cd, Co, Cr, Cu, Fe, K, Li, Mg, Mn, Mo, Na, Ni, P, Pb, S, Sb, Se, Si, Sn, Sr, Ti, V, W, and Zn.

The study of humic preparations of coal origin using ICP–AES showed that all the studied samples have the same elemental composition and differ in the ratio of components: in Powhumus S, Ti, Li, Cr and As prevail; in Sakhalin, Sr, Ba, Co, W and Cu. The content of microimpurities was calculated in the samples (Sakhalin, about 1440 mg/kg; Powhumus, about 470 mg/kg; and Life Force, about 290 mg/kg) and macroimpurities excluding potassium and sodium (Sakhalin, about 450 g/kg, of which 350 g/kg is silicon; "Powhumus" is about 360 g/kg, of which 315 g/kg is silicon; and the Life Force is about 163 g/kg, of which 142 g / kg is silicon).

Comparison of decomposition methods showed that the most complete extraction of elements (except Cd, As, and S) occurs by fusion with lithium metaborate. For Ca, S, Ni, and Be, sample preparation method was not of great importance; for other decomposition methods, the content of elements increased in the series of an aqueous solution — nitric acid decomposition — fusion.

If necessary, rapid assessment of the metal content in humic samples can be limited to the analysis of aqueous extracts without subsequent centrifugation.

Changes in molecular composition of slow and passive pools of Haplic Chernozems during restoration organic matter storage

<u>Kholodov V.A.</u>¹, Farkhodov Yu.R.^{1,2}, Yaroslavtseva N.V.¹ ¹Dokuchaev Soil Science Institute, Moscow, Russia, vkholod@mail.ru ²Lomonosov Moscow State University

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An active, slow and passive pools of soil organic matter (SOM) plays different roles in soil media. Wherein their closely link each other by degradation and restoration processes in soils. The behavior of active pool of SOM during soil restoration more or less is known. Herewith features of restoration slow and passive pools not clear enough.

Thermolabile organic matter is a part of SOM, which volatilized at heating up to 350°C. Content of thermolabile organic matter in soil closely correlated with active C pool. In contrast thermostable one are volatilized at temperature from 350 to 600 °C and can be attributed to slow and passive pools of SOM. In this connection molecular structure both thermostable and thermostable parts of SOM can be studied by double-shoot pyrolysis combine with gas chromatography mass-spectrometry (double-shot Py-GC MS).

Goal of this work was to investigation of changes of molecular composition of slow and passive pools of Haplic Chernozems (Loamic, Pachic) during restoration after total degradation of organic matter without the receipt of plant residues.

Objects of this work were Haplic Chernozems of long-term field experiments: permanent tilled bare fallow since 1964 (total degradation of SOM), unmanaged fallow since 1998 after permanent tilled bare fallow since 1964 (reclaimed soil) and annually mown steppe (undisturbed soil).

Molecular set of slow pool was estimated by second step of pyrolysis Py-GC MS of humic acids are isolated from Chernozems. The passive pool composition was evaluated by second step of Py-GC MS of whole soil.

Slow and passive pools of degraded Chernozems have height abundance of not available for plants nitrogen compounds. In process of restoration of Chernozems in slow pools polysaccharides accumulate. In passive pool increased content of alkanes and aromatic structures.

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Composition and dynamics of volatile and semi-volatile organic contaminants in potable water transported via extended water distribution networks of a metropolis

Konstantinov A.I.¹, Vozhdaeva M.Yu.^{2,3}, Kholova A.R.², Wagner E.V.², Trukhanova N.V.², Melnitskiy I.A.², Perminova I.V.¹, Kantor E.A.³, Beloliptsev I.I.⁴

¹Lomonosov MSU, Department of Chemistry, Moscow, Russia, konstant@med.chem.msu.ru

²Ufavodokanal municipal unitary enterprise, Center of analytical water quality control, Ufa, Russia ³Ufa State Petroleum Technical University, Department of Physics, Ufa, Russia

⁴Financial University under the Government of the Russian Federation, Ufa branch, Ufa, Russia

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With the objective of identifying degree of water quality change at a long-time transportation of potable water on vast city networks, as well as in dead-lock zones, potable water quality of the surface water intake of Ufa city was analyzed with a use of different GC and HPLC techniques. Forty-six volatile and semi-volatile organic compounds were detected in potable water according to an obligatory water quality control program, and more than 250 organic compounds were identified under supplementary monitoring program. It was shown that halogenated by-products were formed during the first hours of contact between residual organic matter present in water and chlorinating agent, which was added at the city water treatment plant. Additional formation of trihalomethanes (up to 40%) was caused by the presence of residual chlorine during transportation of water via the distribution system. The content of haloacetic acids and semi-volatile halogenated organic compounds in potable water dropped down by 70-90% of the initial one after water exited from the plant. At the same time, these zones were characterized with enhanced concentration of squalene that was proposed as an indicator of intensity of biotransformation processes occurring in water (Figure 1).



Figure 1. The scheme of the city water distribution networks and the content of the main classes of organic water pollutants.

Carcinogenic and non-carcinogenic risks reduction was demonstrated in case of transportation of potable water from the surface water intake to remote and dead-lock zones of the city water network. In case of the ground water intake the risks mentioned were shown to increase. The latter is constrained by increasing of content of metals and chlorination by-products.

Lignin phenols content and distribution in gran-size fractions of soils

Kovalev I.V., Kovaleva N.O.

Lomonosov Moscow State University, Soil Science Faculty, Russia, kovalevmsu@mail.ru

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The complex aromatic structure and hydrophobic properties of lignin, as well as its high biochemical stability determine the unique role of this biopolymer in the processes of humification, organomineral interactions and structure formation. Typical structural fragments of lignin in soils are also recognizable for a long time, because fragments of partially decomposed lignin unavailable for microorganisms are stabilized on the external surfaces of metal oxides or clay minerals.

Quantile distribution of lignin (lignin oxidation products (VSC), mg/g Corg.), the degree of oxidation (ac/al)_v, the degree of lignin transformation with respect to the original plant tissue (T, %) in gran-size fractions of soils of the North Caucasus and gray soils of the Moscow region showed that with a decrease of the fraction size (<2 μ m, 2-20 μ m), the amount of lignin in them is significantly reduced – 10 times compared with large fractions (20-250 μ m, >250 μ m). Most of the oxidation products of lignin phenols (up to 50 %) are associated with the coarse sand fraction (>250 μ m). However, maximum oxidation level (ac/al)v and the degree of biopolymer transformation (T,%) and low values of S/V-relations is observed in the most shallow silty and silty fractions, which are due to as a selective loss of syringil aromatic structures, and demethylation of metaxylene groups.

Adsorption of lignin phenols on the mineral surface is facilitated by the high specific surface area of the mineral phase and its positive charge. Biopolymer molecules are encrusted with clay minerals and become inaccessible to microorganisms. This is probably the reason for the high correlation between lignin content (VSC) and specific soil surface values. High values of specific surface area are caused by the presence of mixed-layer minerals in the studied soils. Layered silicates are represented by illite (polytype 2M1), kaolinite (low-temperature polytype 1TC), and vermiculite (polytype 2M) and illite-montmorillonite. It can be assumed that oxidized lignin monomers are fixed by clay minerals. The "ideal" matrix for adsorption of lignin phenols and enzymes is amorphous Fe and Al hydroxides. They are widely distributed in the soil in the free form or in the form of Fe-Mn nodules.

Soil (mm), fractions, (µm)	Lignin (VSC), mg g ⁻¹ Corg	Syringic phenols/ vanillic phenols (S/V)	Vanillic acids/ vanillin (ac/al) _v	Syringic acids/ syringic aldehydes (ac/al) _s	Amino sugar, mg g ⁻¹ N
Soil, <2 mm	11.6	0.80	0.27	0.40	70.9
Clay, 0-2 µm	6.7	1.09	0.33	0.45	38.3
Silt, 2-20 µm	18.9	0.75	0.29	0.41	48.9

Table 1. Lignin parameters of soil gran-size fractions

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References

1. Kovalev I.V., Kovaleva N.O. // In: 19th International Conference of Humus Substances and their Contribution to the Climate Change Mitigation, 16-21 September, 2018, Albena Resort, Bulgaria, pp. 119–122.

n-Alkane distribution in buried soils: implication for paleoecology

<u>Kovaleva N.O.</u>¹, Stolpnikova E.M.², Kovalev I.V.¹ ¹Lomonosov Moscow State University, Moscow, Russia, natalia_kovaleva@mail.ru ²Severtsov Institute of Ecology and Evolution RAS, Moscow, Russia

Keywords: n-alkanes, palaeosols, Pleistocene, isotope composition, past vegetation, paleoecology

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Organic matter of palaeosols is an important resource of information about paleoclimate, which makes it possible to judge the type of past vegetation. Carbon isotopic and n-alkane composition are the most informative methods for study of palaeosols and sediments with low humus content. Long-chain unbranched alkanes can persist for a long time in soils and sediments and save information about past vegetation cover, algae and bacteria pools.

The object of study is the Early Paleolithic multi-layered site Mukhkai II located in the eastern part of the North Caucasus (Dagestan, Russia) at an altitude of 1629 m above sea level. Due to its location on the plateau-like surface, Early Pleistocene sediments have been preserved here. This lagoon-sea sequence contains cultural layers with multiple finds of Pliocene-Early Pleistocene fauna bones and stone tools of Olduvay culture. Paleomagnetic analysis dates all discovered deposits to the Matuyama epoch and indicates two positive excursions: Jaramillo (1.07-0.99 Ma) and Olduvai (1.77-1.95 Ma) subhrons. The age of these sites is estimated at 2.1-1.7 Ma.

Determination of the isotope composition of carbon organic matter of soils and sediments was carried out using a Thermo-Finnigan Delta V Plus IRMS mass spectrometer (data are expressed in relation to the PDB standard). Thermo Flash1112 element analyzer was used to measure the nitrogen and carbon content. Qualitative and quantitative determination of n-alkanes was carried out by capillary gas-liquid chromatography. The analysis was carried out on an Agilent 6890 gas chromatograph with a flame ionization detector and a column DB-1ms. The standard mixture of n-alkanes C12-C36 was used for calibration. Magnetic susceptibility (χ , cm3/g, CGS) was measured by kappametr KT-5.

The isotope composition of organic matter corresponds to the C3 vegetation and ranges from -26.1 - -27.2‰. It becomes heavier (-24.6 - -20.3‰) in modern humus horizons, which has formed under alpine mountain grasses. The samples have enriched with biomarkers of woody vegetation (n-alkanes C27 and C29), especially in the horizon below the cultural layer. At the same time, the dominance of medium chain alkanes and the excess of even alkanes over odd ones are recorded. This suggests that formation of sediments possibly occurred in flooded conditions with contribution of algae and bacteria.

Thus, despite the low content of organic matter, there are the stages of its accumulation distinguished in the Early Pleistocene sequence and these stages are not always associated with cultural layers. These pedolithic sediments were formed in stable hydromorphic conditions without abrupt changes of plant populations throughout the Early Pleistocene. Such uniformity of conditions may be related to position of the ancient site on a lagoon shore that was a part of Akchagyl-Apsheron Caspian basin.

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Photoacoustics of soil organic matter: comparison of various IR modalities for soil fractions of various agrogenesis

<u>Krivoshein P.</u>¹, Volkov D.^{1,2}, Mikheev I.¹, Rogova O.², Proskurnin M.¹ ¹Chemistry Department of M.V. Lomonosov Moscow State University, Moscow, Russia, 119991 ²Department of Chemistry and Physical Chemistry of Soils, Dokuchaev Soil Science Institute, Moscow 119017, Russia

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The studies of anthropogenesis influence on physical and physicochemical properties of soils experience a notable shift. Apart from traditional approaches to assessing the bulk properties, the new information level is to determine soil fractions and aggregate structures including the changes at meso- and microaggregate levels. IR photoacoustic (PA) spectroscopy is an instrument for delicate measurement and modeling of the dependence of soil properties on its physical and elemental composition, moisture, porosity, and density. The PA effect is based on the absorption of IR radiation by the sample followed by the heat transfer to the environment. Pressure waves in surrounding gas are detected by a sensitive microphone providing high sensitivity of measurements [1]. FTIR PA spectrometry has several features like simpler sample preparation, small amounts of the test sample, and the ability to monitor the penetration of radiation in the sample.

In this study, the dependence of mid-IR PA spectra of chernozem soils of various agrogenesis conditions was studied and compared with ATR-FTIR spectroscopy. PA modality provides measurements of soil aggregates up to 2–5 mm without sample decomposition, which is a unique property of this modality. The spectra quality does not change for various size fractions. PA spectra clearly show the organic matter region from 1800 to 1300 cm⁻¹ with some differences in the composition. Below 1300 cm⁻¹, the bands are weak in comparison, which is different from PA measurements of other types of samples like engineered nanomaterials. In our opinion, it requires theoretical modeling and experimentally studying model systems. For soil, PA and ATR modalities are complementary — while ATR provides high sensitivity for the mineral part of soil, the organic part is rather poorly revealed, and vice versa for PA measurements. ATR spectra of soil fractions in the mineral part are different, and FTIR PA and ATR measurements may be used together for studying soil degradation and recultivation processes.

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Aggregation of soil clay particles by organic polyelectrolytes

Lasareva E.V.¹, Parfenova A.M.¹, Azovtseva N.A.² ¹Lomonosov Moscow State University, Chemical Faculty, Moscow, Russia, parf@colloid.chem.msu.ru ²Dokuchaev Soil Science Institute, RAS, Moscow, Russia Keywords: chitosan, humic substances, clay, flocculation, soil structure

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The aggregation of soil clay particles is the basis of soil structure formation. However, mechanisms of that aggregation are still debatable. Our approach is that combine action of two types of soil organic polyelectrolytes (PE) – labile ones released by soil biota in situ and highly transformed organic PE (Humic substances (HS)) may be responsible for such aggregation. Labile polyelectrolyte (in our experiment it is chitosan) behaves as a flocculant linking mineral particles modified by HS. To confirm our hypothesis turbidimetric and rheological studies of model clays (montmorillonite and kaolinite) and carbonate systems modified with HS were carried out. It was shown, that the modification of clay by humic acid does not affect rheological behavior of their pastes but the strength of such systems enhanced considerably after the interaction of modified clay with a flocculant.

The action of polyelectrolyte mixture has a synergetic effect on the aggregation. Synergism of PE action is explained by the structure of mixed PE layers on the surface of clay particles. The strongly adsorbing cationic polymer (flocculant) having a big number of contacts with the surface of clay particles, forms a thin layer due to charge neutralization and decreases the electrostatic repulsion between particles. They can approach to closer distances that favors the "bridging" flocculation via extended macrochains of anionic polymer (HS) located on the surface. Adsorption of phenols with one, two and three hydroxyl groups that may be incorporated in the HS structure was measured on different soil minerals. It was shown that increasing number of phenolic hydroxyl groups leads to the increase of phenol adsorption on minerals. Clays were found to be better phenol sorbents compared with carbonate and different micas (biotite, muscovite and vermiculite). The modification of montmorillonite with glucose increases adsorption of all phenols.

The proposed adsorption-flocculation mechanism of soil formation due to combine action of different types of organic polyelectrolytes may be applied to other fields of investigation, for instance, the study of aggregation of colloid and particulate riverine matter in the mixing zone river-sea (delta, estuary) where settled aggregates may be a beginning of soil formation [1], water purification and preparation of sand-clay pastes for various applications.

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References

1. Lasareva E.V. et al. // Geophysical Research: Biogeosciences. 2019. 124:1–14. https://doi.org/10.1029/2019JG005026.

Complexation of heavy metal ions with peat humic acids

Lodygin E.D.¹, Alekseev I.I.², Vasilevich R.S.¹, Abakumov E.V.² ¹Institute of Biology, Komi Science Center, Ural Branch, Russian Academy of Sciences, Syktyvkar, Russia, lodigin@ib.komisc.ru ²Saint Petersburg State University, Saint Petersburg, Russia

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Based on a model experiment, the binding ability of humic acids (HAs) of Hemic Folic Cryic Histosol (peat with vegetated plots) to lead and cadmium ions was revealed. It has been shown that the process of adsorption of metal ions by suspended humic acids is similar to the process of complexation of metal ions with certain functional groups of dissolved HAs. This trend follows the Irving-Williams series, which describes the stability order of metal complexes. For humic acids, the stability of complexes with dipositive metal ions is of the following order: $Pb^{2+} > Cd^{2+}$, which is partly due to a decrease in the ionic size and the effects of the ligand field. The thermodynamic characteristics (limiting specific adsorption, constant of sorption equilibrium, Gibbs energy, entropy and thermal effect of sorption) of the adsorption process of selected heavy metal ions are calculated. It was found that the critical specific adsorption of Pb²⁺ ions is almost an order of magnitude higher than that of Cd²⁺ ions and amounts to 0.0078-0.034 mmol / L for Cd²⁺ ions and 0.16-0.29 mmol / L for Pb²⁺ ions. The changes in the sorption enthalpy are 48.4 kJ/mol for Cd²⁺ ions and 22.6 kJ/mol for Pb²⁺ ions indicate the endothermic nature of adsorption on HAs preparations. This is due to the chemisorption processes. A large amount of heat is spent on the destruction of the hydration shell of metal ions, which is confirmed by the positive values of the change in sorption entropy. It was shown that the limiting stage of adsorption for Pb²⁺ ions is the stage of ion diffusion to the sorbent, and chemosorption itself proceeds quite quickly. Values of $\Delta S > -10 \text{ J}$ / (mol \cdot K) indicate a dissociative adsorption mechanism for both metals, i.e. non-adsorbed ions in solution are in a more ordered state than after adsorption. Negative values of the Gibbs energy change for Cd²⁺ and Pb²⁺ ions indicate that the adsorption of Cd²⁺ and Pb²⁺ ions by humic acid preparations of peat soils is a spontaneous process with a complex mechanism including complexation and ion exchange processes.

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Role of humus substances in the formation of the soil geochemical fluxes during deposit exploitation in Okhotsk region

Makhinova A.F., Makhinov A.N.

Institute of Water and Ecology Problems, Far Eastern Branch of the Russian Academy of Sciences, Khabarovsk, Russia, mahinova@ivep.as.khb.ru

Keywords: humus, soils geochemical fluxes, elements, mining

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The mechanisms involved in the formation of the soil geochemical fluxes are dependent on the physical state of the solid, liquid and organic soil phases. The synthesis and decomposition of organic matter and the formation of the finely dispersed and soluble humic acid fractions play an important role. The geochemical processes in soils occur as a result of three forces: (1) the adhesion or the mutual molecular attraction between fine particles less than 2-3 μ m in size, thus resulting in their coagulation and aggregation into the <1 μ m fractions (the effect of sorption); (2) the molecular absorption of water onto the surface of fine particles of the solid and organic soil phases, which causes soil swelling (the effect of water intake); (3) the molecular interaction between the soil phases and gravity moisture promoting the dissolution and removal of weathering products.

Excess water in soil pores will tend to mix with elements and their compounds to form soil solution, which move through the pore space due to gravity formation the soil geochemical fluxes. The interaction between the aggressive humic acid fraction and the soil microorganisms results in the formation of complex cations ([FeHSO₄]⁺, ([FeHSO₄]²⁺, ([CuHSO₄]⁺ and protons ([H⁺]) in soil pores. With increasing pH in the solution (<3.5), complex salts dissociate to give high-charge cations (Fe²⁺⁽³⁺⁾, Cu²⁺, Zn²⁺, Pb²⁺) [1]. In the presence of free oxygen, the oxidizing properties of manganese effect the dissociation of Co²⁺ and Co³⁺. Such components are indicative of hypergene conditions within the Okhotsk region. The concentrations of elements in the soils correlate with the organic matter content and particle-size distribution.

Two types of geochemical migration fluxes were identified in the background: (1) N – natural and (2) A – anthropogenic. The natural migration fluxes are directed vertically downward the soil profile and assist the subsurface exchange of substances.

The anthropogenic migration fluxes are related to areas of mining operations. The waste produced during open-pit mining and other mining and smelting operations in a source of soil contamination with heavy metals. After being exposed to air, the oxidation of sulfide-bearing minerals takes places according to the following reaction: $2FeS_2$ (pyrite) + $2H_2O + 7O_2 = 2FeSO_4 + 2H_2SO_4$, with the formation of unstable iron sulfates [2]. The products of the mineral degradation and the fine particles of weakly oxidized mineral substances are transported down the slopes with the rain water runoff and form the anthropogenic migration fluxes.

The concentrations of heavy metals are generally high in the vicinity of each mining site, which represents the zone affected by mining lying within a 200-500 m radius depending on the natural conditions.

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References

- 1. Senesi N., Poffredo E. // In: "Chemical Processes in Soil", Soil Sci. Soc. Amer., Madison, 2005, 563–617.
- 2. Makhinova A.F. et al. // J. Geochem. Exploration. 2013. 132:140–148.

Characterization of peat humic acids modification during the peat formation process using the HSQC NMR

<u>Malgina E.</u>¹, Bogolitsyn K.¹, Faleva A.¹, Kozevnokov A.¹, Kosheleva A.¹, Sobolev N.¹ ¹Northern (Arctic) Federal University named after M. V. Lomonosov, Arkhangelsk, Russia, n.sobolev@narfu.ru

Keywords: humic acids, HSQC, peat, NMR, humification

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1D NMR spectrometry is a powerful tool for characterization of the humification process occurring in the bog ecosystem and for analysis of its dynamic. However, there are some limitations of the mentioned method in analysis of the randomized polymeric mixture such as humic acids (HA). The peak overlapping is one of the main problems. To avoid this limitation 2D NMR spectrometry method is nowadays widely used. Application of this technique to the HA analysis enhance the information value of the analysis results.

In the present study 2D NMR experiment was conducted for the characterization of the peat HA modification through the peat core. For this purpose, HA were isolated from each of 7 layers (from 0 to 375 cm depth) of peat according to [1]. Then, 50 mg of the extracted HA was dissolved in 600 μ L of d6-DMSO and further analyzed using 600 MHz Bruker Avance III NMR spectrometer. 1H–13C heteronuclear single quantum coherence (HSQC) spectra were collected.

An example of HSQC spectrum (one of the 7) presented on Fig. 1. All obtained spectra can be separated on 4 main regions. For region A decreasing of the fatty and amino acids signal was observed with the increasing of the layer's depth. In aliphatic region B the dynamic of carbohydrate increasing up to the maximum on the layer 35-85 cm was found. Also, in region B tyrosine and phenylalanine amino acids were determined. We faced with difficulties in the interpretation of the results in aliphatic region C due to the intense signal of the internal standard, which will be removed in further experiments. For aromatic region D the decreasing of the well resolved signal of the aromatic compounds was observed.



Figure 1. HSQC spectrum of HA isolated from peat layer 35-85 cm

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References

1. Lamar R. // J. AOAC Int. 2014. 97(3):721–730. doi:10.5740/jaoacint.13-393.

Possibilities of determination of soil organic matter content and its quality using near infrared spectroscopy in different ecosystems

<u>Menšík L.¹</u>, Kunzová E.¹, Nerušil P.¹, Pospíšilová L.², Hlisnikovský L.¹, Kulhavý J.³ ¹Division of Crop Management Systems, Crop Research Institute, Prague, Czech Republic, ladislav.mensik@vurv.cz

²Dept. of Agrochemistry, Soil Science, Microbiology and Plant Nutrition, Mendel University in Brno, Czech Republic

³Dept. of Forest Ecology, Mendel University in Brno, Czech Republic

Keywords: near-infrared spectroscopy techniques, soil organic matter, humic substances, humic acids, arable soil, grassland, forest

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Soil organic matter (SOM) plays an important role in terrestrial ecosystems (agroecosystems, grasslands and forests). SOM is very important in maintaining fertility and in improving physical, biochemical and biological soil properties. Classical analytical (laboratory) methods of determination SOM (carbon /SOC/, humic substances /HS/, humic acids /HA/ etc.), which are accurate and sufficiently reproducible, but at the same time very often demanding on staff working in a specialized laboratory, further on time (relatively long time to determine the result), but also on financial means. The aim of the study is to present new possibilities of determination of SOM content and its quality /HS, HA/ using NIRS.

Identical soil samples from different ecosystems (arable land, grassland, forest) were measured using classical laboratory methods and on a FOSS NIRSystems 6500 instrument dispersive spectrometer (Company NIRSystems, Inc., Silver Spring, USA). Sample scanning was set in reflectance mode for the 400–2500 nm range. WinISI II software (Infrasoft International, Inc., USA), version 1.50, Partial Least Squares (PLS) and modified PLS were used to develop calibration equations and graphical outputs.



Figure 1. Tightness of the relationship between laboratory analysis and NIRS prediction of SOC in agriculture soil /arable land/ (left) and HA in forest soils (right).

The precision of the determination, expressed as the value of the determination coefficient (R^2) of the calibration set (n = 130-550) for the SOM parameters (Corg, HS, HA), is in the range of 0.85-0.99 (0.80-0.90 applicable) for common agricultural or forestry practice (0.90 or more excellent). Measurement (determination) is sufficiently accurate, safe to work and has no negative impact on the environment.

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References

 Menšík L. et al. // In: Jambor V., Malá S. 18th International Symposium Forage Conservation, Mendel Univerity in Brno, August 13-16, 2019, Brno, Czech Republic, pp. 190-197. ISBN 978-80-7509-670-8.

Amphiphilic components of humic substances of Transbaikalia soils

Milkheev E.Yu., Chimitdorzhieva G.D., Davydova T.V.

Institute of General and Experimental Biology SB RAS, Ulan-Ude, Russia, evgmilh@gmail.com

Keywords: humic acids, fulvic acids, amphiphilic components

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Humic substances (HS) are supramolecular systems of natural organic compounds with a high degree of structural heterogeneity, which determines their resistance to biodegradation. HS consist of aromatic rings and aliphatic chains and contain both polar groups and non-polar fragments, which gives them amphiphilicity, that is, the ability to exhibit both hydrophobic and hydrophilic properties [Milanovsky, 2000]. An integrated approach consisting in studying the composition and properties of HS using the method of liquid chromatography of hydrophobic interaction allows us to obtain valuable information on the structural and functional parameters of humic (HA) and fulvic acids (FA) of various genesis isolated from chernozem-permafrost soils and Transbaikalia Burozem.



Figure 1. Chromatograms of humic (a) and fulvic acids (b) Meadow Chernozem permafrost soils.

Chromatographic analysis of the HS of soil genetic horizons (Fig. 1) showed the main patterns of differentiation of the amphiphilic components of HA and FA according to the profile. The composition of both HA and FA contains components with pronounced hydrophilic properties. In fulvic acids, the hydrophilic fraction is predominant and accounts for approximately 80–90% of the total area of the chromatogram. Hydrophilic components of HA make up 28–41% of the total area of chromatograms. HA of Burozem and meadow chernozem soil, respectively. The relative content of hydrophobic components in the composition of HS increases in the series: FC (Burozem) –FK (Meadow Chernozemic) –HC (Burozem) –HC (Meadow Chernozemic). The ability of humic acids to coagulate with calcium salts, their carbonization, degree of aromaticity, and other basic chemical and physico-chemical parameters changes in a similar way.

Due to the predominance of lateral radicals in their molecules, humic acids of Burozem have a higher hydrophilicity than HA from meadow chernozem soils, in whose molecules aromatic carbon networks predominate. Therefore, HA Burozem are prone to peptization, which is why their high resistance to electrolytes and mobility in the soil profile are related to that of meadow black-chernozem soil. Most likely, the more pronounced hydrophilic properties are in HV, the more these substances will be mobile in the soil profile, and to a greater extent, they will act as agents for acid hydrolysis of minerals and elute. Hydrophobic HSs, on the contrary, will be fixed at the place of formation, forming the accumulative characteristics of the profile. Apparently, this is the meaning and role of the amphiphilic property in the formation of the humus profile of soils.

The effect of cyclic cryogenic exposure on the dispersion parameters of humic substances

<u>Orlov A.S.</u>, Kuznetsova I.A., Mironenko K.A., Ponomareva T.I., Zubov I.N. N. Laverov Federal Center for Integrated Arctic Research Russian Academy of Science, Arkhangelsk, Russia, alseror@yandex.ru

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Humic substances are one of the main structure-forming components of soils. These are responsible for its fertility and resistance to erosion processes, which can occur under the influence of both natural and anthropogenic factors. Such factors in the cold climate of the European North of the Russian Federation include, first of all, the cyclic cryogenic exposure. Its influence on the state of the humus component of soils is currently scarcely investigated. It is known that one of the forms of the presence of humic acids in the soils is the colloidal (semi-colloidal) form [1-3]. At the same time, it should be noted the extremely low stability and regenerative capacity of soils of the Arctic regions.

The goal of this research is to investigate the effect of cyclic cryogenic exposure on colloid-dispersion parameters of the humic substances.

An analysis of the data obtained by the method of quantum correlation spectroscopy allows saying that a rather high polydispersity characterizes the studied sample of humates isolated from the coarse humus horizon of forest podzolic soil. It should be noted that even in the region of low concentrations (100 mg / I), supramolecular formations D = 80-3000 nm are fixed in the solution of humic substances, which is apparently due to their functional nature and indicates a high ability to intermolecular interactions. In this case, the contribution of groups of particles of each dimension to the dynamic light scattering is directly proportional to both their numerical value and diameter.

The obtained results indicate that cyclic cryogenic exposure contributes to the gradual partial destruction of supramolecular formations of the humic substances. It is confirmed both by a decrease in the contribution to the light scattering of a group of large associates with D = 500-3000 nm, whose contribution to the total number of particles is not significant, and the total redistribution of particles by size. It is seen that even with a four-time freeze-thaw cycle, particles are redistributed in size from D = 80-500 nm to D = 20-100 nm.

Thus, the results obtained during this study indicate the sensitivity of the hydrodynamic characteristics of humic substances to cyclic cryogenic exposure. It is shown that humates isolated from the coarse humus horizon of forest podzolic soil, even with significant dilutions, are predominantly in the form of supramolecular formations of an associative nature. It has been established that cyclic freezing contributes to a decrease in colloid-dispersion characteristics (particle sizes and contribution to light scattering) in aqueous solutions of humates. That is associated, apparently, with the partial destruction of their macromolecular formations and requires further investigation.

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References

- 1. Orlov. D.S. // Moscow: MSU. 1990. p. 325 (In Russian).
- 2. Popov A.I. // Saint-Petersburg: St. Petersburg University Press. 2004. p. 248 (In Russian).
- 3. Tate R. // Moscow: Mir. 1991. p. 400 (In Russian).

Application of Mössbauer spectroscopy for the diagnostics of functional groups of the humic substances

Pankratov D.A., Zimbovskaya M.M.

Lomonosov Moscow State University, Moscow, Russia, pankratov@radio.chem.msu.ru

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The waters of open reservoirs, soils and organogenic rocks contain high-molecular organic compounds of natural origin - humic substances (HS). HS, having a large number of diverse functional groups, is capable of entering into redox, sorption, and complexing interactions with iron, as one of the most common elements in the Earth's crust.

Earlier, we showed that, HS of different origins and fractional compositions are adsorbed selectively on macroporous weakly basic anion-exchange resin (MWBAER) [1]. So, HS of coal origin are sorbed on MWBAER by predominantly carboxyl groups of aromatic fragments, while lignosulfonate derivatives, by means of strongly acidic sulfonic groups. Thus, in the process of HS sorption on MWBAER, fractionation of individual functional groups of natural polyelectrolytes is carried out, which allows them to be excluded from further interactions, for example, with transition metal ions. It is known that MWBAER by immobilization of HS can be used for sorption of iron ions for natural water treatment. Therefore, it is obvious that 57-iron, being a Mössbauer nuclide, can be used as a probe for structural fragments of HS capable of forming coordination bonds with iron atom.

To demonstrate the possibilities of this approach for studying the fractionation of HS functional groups using MWBAER, MWBAER immobilized by HS after adsorption of 57iron chloride solutions was studied by Mössbauer spectroscopy. Two HS preparations was immobilized in MWBAER: Sakhalinskii potassium humate (OOO Biomir-2000, Moscow, Russia) of coal origin and sodium lignohumate (OOO NPO RET, St. Petersburg, Russia), which is produced via the oxidation of lignin sulfonate. The conditions for sorption of 57iron ions were determined based on previously obtained isotherms of adsorption of iron salts on these sorbents. Mössbauer spectra were obtained both for wet adsorption products frozen to the temperature of liquid nitrogen, and for dried samples at room temperature. The Mössbauer spectra are broadened, asymmetric doublets (Fig. 1), which can be satisfactorily described by a superposition of two or three subspectra of symmetric doublets corresponding to different sites of probe atoms.





Thus, the method of fractionation of the functional groups of HS using MWBAER was tested, which allows us to study the nature of the coordination "HS - iron" interaction. It is shown that coordinated HS iron can occupy two or three different positions at the formation of bonds with fractionated by functional groups HS.

References

1. Pankratov D.A. et al. // Russ. J. Phys. Chem. A. 2017. 91(6):1109–1115.

Humic substances: new approaches for their isolation from natural objects

<u>Popov A.I.</u>¹, Simonova J.V.¹, Tcivka K.I.², Song Ge¹, Birilko D.A.¹, Kholostov G.D.¹, Sazanova E.V.¹

¹St. Petersburg State University, St. Petersburg, Russian Federation, paihumic@gmail.com ²St. Petersburg State Agrarian University, St. Petersburg, Russian Federation

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Organic matter of some natural objects (soils, brown coals, peats, bottom sediments etc.) is a complex of various organic compounds. In addition to humic substances (HS) in composition includes: lipids (waxes, fats, gums and et al.), pigments its (oxyanthraquinones and related substances, tetrapyrole compounds and carotenoids), kerogen, glomalin and so on. In the traditional assessment of qualitative composition of natural object organic matter, all the listed substances, as well as the hydrolytic degradation products of particulate organic matter, pass into the alkaline solution (Figure 1). The resulting mixture, which has both specific and individual organic compounds, is usually separated into fulvic acids, hymatomelanic acids, and humic acids only.



Figure 1. Scheme of humic-like compounds isolation by alkaline solution and their separation.

We propose new approaches to isolate clean HS from natural objects. They includes: (i) preliminary extraction of individual organic compounds, which are soluble in 90 % acetone solution (specifically tetrapyrole compounds and carotenoids); (ii) HS and glomalin isolation by organic solvent after a preceding protonation; (iii) HS separation from glomalin by a certain sorbent; (iv) isolation of water-soluble organic compounds, as well as fresh and semi-decomposed postmortem residues and excreta of plants and soil biota; (v) lipid isolation by benzene-ethanol mixture; (vi) kerogen determination by thermal method (based on of the loss on ignition).

These approaches were experimentally tested on samples of some soils. Note that the biological activity of HS, which were isolated by an organic solvent, was higher activity of HS, which were isolated from same soils by an alkaline sodium pyrophosphate solution.

The authors understand that the new scheme of HS isolation from natural objects is not yet fully worked out, that it is only the first approximation for decision of problems, which are connected with HS chemistry.

Binding of organic ions to humic acids: spectroscopic and thermodynamic analyses of the ruling molecular interactions

<u>Sedláček P.</u>¹, Smilek J.¹, Kalina M.¹, Enev V.¹

¹Institute of Physical and Applied Chemistry, Faculty of Chemistry, Brno University of Technology, Brno, Czech Republic, sedlacek-p@fch.vut.cz

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Combination of aromatic or aliphatic structural moleties with polar functional groups dissociable in an aqueous solution represents a structural motif abundant in anthropogenic environmental solutes such as pesticides, surfactants or antibiotics. The amphipathic structure provides these compounds with versatility of possible molecular interactions that they can be involved in. We have performed an extensive experimental study focused on binding of various model organic ions to humic substances of diverse origin. For this purpose, we have combined various analytical strategies, including spectroscopic assays UV-VIS steady-state and (differential spectrometry. time-resolved fluorescence spectroscopy), originally developed diffusion techniques (experimental evaluation of the solute molecular transport in a medium with homogenously dispersed humic substances), and examination of the binding thermodynamics based on (isothermal titration) calorimetry and equilibrium dialysis (see Fig. 1). This contribution evaluates the particular merits of the individual experimental techniques and summarizes the main findings on what type of molecular interactions plays a principal role in binding charged organic compounds by humic substances.





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The interactions of organic species with humic acids studied by dialysis techniques. Comparison of selectively methylated with "natural" humic acids

Smilek J.^{1,2}, Sedláček P.^{1,2}, Kalina M.^{1,2} ¹Institute of Physical and Applied Chemistry, Faculty of Chemistry, Brno University of Technology, Brno, Czech Republic, smilek@fch.vut.cz

²Materials Research Centre, Brno University of Technology, Faculty of Chemistry, Brno, Czech republic

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The interactions of standard humic acids provided from International Humic Substances Society (IHSS) specifically Leonardite Humic Acid Standard 1S0104H have been deeply studied with organic ionic compounds (such as surface active agents and organic dyes). The unconventional diffusion and dialysis techniques (originally developed on our institution) have been used for the study on binding of organic species to humic acids. Two types of humic acids have been compared - humic acids with selectively blocking carboxylic functional groups (-COOH) in comparison with humic acids in their native form.

The results of dialysis experiments confirm the strong affinity of both types of humic acids towards to ionic species. Surprisingly, the effect humic acids modification (blocking of carboxyls) is minor than it could be expected. The modified equivalents of standard humic acids had almost the similar affinity towards to ionic compounds with non-modified (native) humic acids.



Figure 1. Schematic diagram of the equilibrium dialysis experiment

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Spectroscopic characterization of different samples of humic acids

<u>Sokolova I.</u>¹, Kudlatskaya A.¹, Selyanina S.², Naumova L.¹, Tchaikovskaya O.¹ ¹National Research Tomsk State University, Tomsk, Russia, sokolova@phys.tsu.ru ²Federal Scientific Research Centrum for Complex Learning of the Arctic, Arkhangelsk, Russia

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Humic substances are the products of extremely complex chemical and microbial processes of the decomposition and secondary synthesis of plant and animal residues in soil and water. Humic substances (including humic and fulvic acids) are naturally photoactive components, which are widely present in environment. Humic substances representing the main fraction of organic matter receive increased attention because of their reactivity as light absorbers. Depending on their origin and structure, humic substances have a remarkable ability to absorb light and transfer this energy to other substrates and in some cases strongly affect photolysis of xenobiotics.

The absorption and fluorescence spectra of different samples of humic acids (HAs) are investigated. The samples of HAs were prepared from peat of Arkhangelsk Region. The comparison of this HAs with the samples of humic acids obtained from Aldrich Chemical Co is carried out.



Figure 1. Dependence of the optical density of the absorption spectra of various HAs samples on the drying time.

The longer is the duration of drying of the samples, the lower the optical density of the absorption spectra. In the process of humification and with the duration of peat drying, the fluorescence intensity of the HAs increases. The dependence of the fluorescence spectra of the investigated samples on the fluorescence excitation wavelength was found.

The effect of UV radiation from excilamps on the spectroscopic properties of humic acids has been investigated.

An important point to emphasize is the significance and prospects of optical and photochemical methods in determining the characteristics and functions of humic acids as extremely sensible and informative methods.

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Features of molecular-mass distribution of humic acids from permafrost peats of Russian Arctic

<u>Vasilevich R.S.</u>¹, Vezhov K.S.¹, Lodygin E.D.¹, Abakumov E.V.² ¹Institute of Biology of Komi Science Centre of the Ural Branch of the Russian Academy of Sciences, Syktyvkar, Russia, vasilevich.r.s@ib.komisc.ru ²Saint-Petersburg State University, Saint-Petersburg, Russia

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The peatlands investigated are in the ecoton south tundra – forest-tundra (Komi Republic, Russia, Plots 1-1, 1-2) and ecoton north tundra - south tundra (Nenets Autonomous Okrug, Russia, Plots 2-1, 2-2). Humic acids (HAs) from the two permafrost peat soils - Hemic Folic Cryic Histosol (Plot 1-1, Plot 2-1) and Hemic Folic Cryic Histosol (Turbic) (Plot 1-2, Plot 2-2) have been studied from peat plateau-thermokarst complexes. A quantitative assessment of the content of functional groups and molecular fragments of studied HAs (¹³C-NMR CP/MAS data) indicates the difference in their molecular composition in seasonal thawed (STL) and permafrost layers (PL) of peat. Specifically, the aromatic fragments and methoxy groups show increasing trends and paraffin groups show decreasing trends with respect to depth for all soils. The obtained values of the number average molecular weights (M_n) of the studied HA preparations have a significant variation (6.9–23.1 kDa) in peat profiles (Fig.). The highest M_p of HAs are confined to the upper horizons of the STL, due to the greater contribution of long chain paraffin structures (up to 35%) and polysaccharide fragments (up to 27%), which leads to an increase in the Stokes radius of HA molecules. Increasing the degree of peat decomposition down the profile and the associated greater humification of the organic material, cause a systematic decrease in M_{n} . Profile reduction of M_{n} is due to decrease of proportion of high and medium molecular weight fractions and increase of proportion of low molecular weight fraction. The M_p increment in the lower horizons of plots 1-1, 1-2, 2-2 is likely due to the coagulation of HAs on the mineral matrix of peat deposits (the peat of these horizons has high ash content) to form high molecular weight HAs. Thus, it has been shown that the humification of organic peat material is associated with the transformation of high molecular weight HAs structures enriched in paraffin and carbohydrate fragments to condensed structures with lower molecular weights.





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Emergent functional behavior of humic substances as result of chemical characteristics

Vialykh E.A.^{1,2} ¹University of Colorado, Boulder, CO, USA, elena.vialykh@colorado.edu ²BioHumaNetics Inc. Gilbert, AZ, USA

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The structural organization of humic substances (HS) has been a central question of earth sciences for several decades. Recently, HS were recognized as complex mixtures of small molecules and oligomers. We investigate the correlation between the chemical composition of HS, perceived as labile aggregates, and their emergent functions (EF).

Complex organic mixtures can be considered within the context of complexity theory. The study is done through investigations of how interactions between parts creates an emergent collective system behavior and how the system interacts and forms relationships with its environment [1]. In order to create a model of HS, which illustrates the connection between chemical composition and EF, the theoretical model should encompass properties that relate to H-bonding, hydrophobic and charge-transfer interactions and depend on conformations. Therefore, we developed a computational approach that combines with ideas of supramolecular structural organization and dynamic combinatorial chemistry [2]. The focus of the work was on the labile (reversible and changeable), connection between molecular units with bonds (interactions) that form and break on elementary step timescales. The important reactions are dissociation, proton transfer, hydrogen bond interactions, which occurs in a timescale range of pico- to nanoseconds. Therefore, the composition of a system will depend on its environment.

Computational modelling was used to help to understand the processes and mechanisms on the molecular scale that occur in different fractions of the HS, fulvic acids and humic acids, as they interact with metal ions and organic pollutants. The conformational changes in different solvents were also investigated. The importance of non-covalent interactions in the EF of HS is highlighted. H-bonding, hydrophilic/hydrophobic surface areas and π -stacking interactions play a significant role in aggregation processes as well as in the sorption of environmental pollutants. The interaction of a polar pollutant, phenol, with HS occurs through H-bonding, whereas non-polar benzene interacts through hydrophobic and π -stacking interactions. The non-polar pollutant results in a much stronger sorption by HS and causes an additional structural rearrangement of the aggregates. The interactions of HS with metal ions revealed a variation of the studied properties of HS, including H-bonding, structural conformation, the hydrophilic/hydrophobic surface areas and the potential energy, with the change of environmental conditions. Thus, a decrease of water content or an increase of the potential energy, i.e. formation of more stable aggregates.

The significance of this work is in shifting the focus from a search for the exact "unique" structure of HS to the correlation of chemical characteristics of the sample with its emergent functional behavior. The new approach could be beneficial in solving ongoing disputes in the field of HS research, since it reveals how different properties of HS emerge from their chemical composition. Overall in this work we showed that recognition of HS as labile supramolecular aggregates of different molecular fragments, allows one to consider the lability of the system when it interacts with environmental pollutants.

References

- 1. Bar-Yam Y. https://www.researchgate.Net/Publication/246294756_General_ Features_of_Complex_Systems (2002).
- 2. Corbett P.T. et al. // Chem. Rev. 2006. 106:3652.

Thermal stability of soil organic matter of Typical Chernozems under different land uses

Volkov D.S.^{1,2}, Rogova O.B.², Proskurnin M.A.¹, Farkhodov Yu.R.²

¹Chemistry Department of M.V. Lomonosov Moscow State University, Moscow, Russia, 119991 ²Department of Chemistry and Physical Chemistry of Soils, V.V. Dokuchaev Soil Science Institute, Moscow 119017, Russia

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Chernozems are among the most fertile soils relevant for global food production and the main part of productive agricultural land in Russia is confined to Chernozems. Soil organic matter (SOM) of bulk samples and size fractions (by dry screening) of Chernozems with a rather wide agricultural use history (from intact steppe to permanent bare fallow) was studied by thermogravimetry (TGA) under pyrolysis conditions in an inert atmosphere to approach its transformation depending on the type of use. The data basis is a 55-year-long field experiment of the Kursk Research Institute of Agricultural Production and V.V. Alekhin Tsentralno-Chernozemny Nature Reserve of Russia. It was shown that the SOM of the formed deciduous tree shelterbelt soil is the least thermally stable over the whole decomposition temperature range and contains the maximum amount of thermally unstable compounds. Among the aggregates of this soil, the aggregates with a size of 1-2 mm are the most thermally stable, the least stable are 7–10 mm in size and less than 0.25 mm, which is confirmed by the whole set of applied statistical criteria. Steppe soil stands out for its highest thermal stability formed over the centuries of lack of ploughing under predominant grassy vegetation. Moreover, all its aggregates contain predominant amounts of thermally unstable SOM, the most thermally stable are aggregates of 1-2 mm. SOM recovery processes that occur in soil of the overgrown fallow and no-tillage farming if compared with the initial (bare fallow and cropland, respectively) are not the same. Predominant SOM accumulation occurs in aggregates of 0.5–1 and 1–2 mm.

With systematic observation of agricultural land, a statistically significant increase in T50 in the range of 170–600 °C. A new parameter to assess the degree of organic matter degradation, the ratio of the mass loss in the 380–600 to 170–380 °C temperature range that is referred to as two-range thermal stability ratio. It is shown that this relative index discriminates soil samples depending on the land use type and does not require the knowledge on carbon content. As well as a decrease in two-range thermal stability ratio, the same range may indicate the manifestation of SOM degradation processes and the need for measures to compensate for a decrease in thermally unstable SOM components. At the same time, state-of-the-art thermogravimetry is a highly automated method of analysis, which almost does not require consumables and operator time, and thus can be used in small laboratories.

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On the origin of optical properties of natural organic matter: the role of excitation energy transfer and its experimental observation

Yakimov B.P.¹, Rubekina A.A.¹, Zherebker A.Ya.^{1,2}, Perminova I.V.¹, <u>Shirshin E.A.</u>¹ ¹Lomonosov Moscow State University, Moscow, Russia, shirshin@lid.phys.msu.ru ²Skolkovo Institute of Science and Technology, Moscow, Russia

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Despite being extensively studied for decades, the origin of natural organic matter (NOM) optical properties remains debatable. While the possibility to describe as a superposition of individual chromophores seems the most obvious approach, several facts contradict this hypothesis. As a result, speculations on the dominating role of chromophores interaction in the formation of NOM spectral properties are widely discussed in the literature. However, although it is quite obvious that electronic interaction must occur in such a complex molecular ensemble as NOM, a direct experimental study of this phenomenon has not been reported yet. In this work we aim at demonstration of the role of electronic interaction in the formation of NOM optical properties. Making use of time-resolved spectroscopy techniques in the subpicosecond to nanosecond ranges, we uncover the chain of events which lead to the formation of the red edge in NOM optical spectra. Then, we burn a hole in the NOM spectra and show how this affects the energy transfer chain. Based on these experiments, a semi-quantitative model of photophysical processes in NOM was developed.

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Structures of individual components of biologically active oxidized lignin material determined by ultra-high resolution mass-spectrometric techniques

<u>Zherebker A.</u>¹, Kharybin O.¹, Orlov A.¹, Lechtenfeld O.², Fedoros E.I.³, Perminova I.V.⁴, Nikolaev E.N.¹

¹Skolkovo Institute of Science and Technology, Moscow, Russia, a.zherebker@skoltech.ru ²Helmholtz-Centre for Environmental Research, Leipzig, Germany

³Petrov National Medical Research Center of Oncology, Saint-Petersburg, Russia

⁴Lomonosov Moscow State University, Moscow, Russia

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Oxidized lignin materials possess a wide range of biological activity including hepatoprotection [1]. However, molecular complexity prevents a determination of active species and, consequently, a description of the molecular mechanism of their action. At this regard FTICR MS in combination with isotopic tags provide a unique opportunity to obtain structural information about individual molecular components, which can be used for in silico search of molecular targets. Here, we report on the first combination of several selective labeling techniques to enumerate functional groups and structural fragments of water-soluble product of oxidative degradation of lignin BP-Cx-1.

Carboxylic, carbonyl and phenolic moieties were determined by selective incorporation of deuterium tags via esterification, reduction and acetylation, respectively. In most cases, the numbers of oxygen atoms were covered by these functional groups. Additionally, we observe electrophilic addition of deuterium, which clearly indicates the presence of α , β -unsaturated carboxyls and carbonyls in the BP-Cx-1 structures. Application of acid-base catalyzed H/D exchange enabled to determine the substitutional pattern of aromatic rings. The obtained results were used to extract structures from the curated database of biological activity ChEMBL, which matched results of FTICR MS analysis and isotopic labeling. The developed approach can be considered as a first step in the understanding of the BP-Cx-1 biological action.



Figure 1. Work-flow for the BP-Cx-1 individual components determination using FTICR MS, isotopic labeling and ChEMBL data-mining.

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References

1. Fedoros E.I. et al. // Oncotarget. 2018. 9:18578-18593.

Synthesis of hydroxide iron(III), which stabilized by fulvic acid. Comparison and characteristic iron binding capacity

<u>Zhirkova A.M.</u>¹, Kydralieva K.A.^{1,2}, Sobolev P.S.¹, Lebedev V.A.¹, Volikov A.B.¹, Zykova M.V.³, Perminova I.V.¹

¹Lomonosov Moscow State University, Moscow, Russia nastya12349@mail.ru

²Moscow Aviation Institute, Moscow, Russia

³Siberian State Medical University, Tomsk, Russia

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Iron Deficiency Anemia is the global health problem both for humans, animals, and plants. Iron replacement therapy is the most adopted method of iron deficiency correction. Iron(III) hydroxide stabilized with polysaccharides is among the most prospective medications. However, it may induce specific adverse reactions (e.g., anaphylactic shock). The alternative medications are needed. In our research, we used fulvic acids instead as polymaltosate for stabilization of iron(III) hydroxide. Fulvic acids are natural macroligands with high binding affinity for iron(III). We hypothesized that they can serve as prospective ligands for stabilization of Fe(III) hydroxide. To achieve this goal, we carried out syntheses with polymaltosate and fulvic acid.

Firstly, we prepared iron(III) hydroxide by mixing solutions of FeCl₃ and NaHCO₃ at pH 7. The precipitate of Fe(OH)₃ was collected and added with the heated-up solution of maltodextrin. The precipitate was centrifuged and the product was designated as PMFe1. The similar synthesis was carried out with fulvic acid and the obtained product was designated FAFe2. The obtained samples were analyzed using X-ray diffraction analysis (XRD), TEM and Moessbauer spectroscopy. It was found that the both samples contained ferrihydrite nanoparticles. Moessbauer spectroscopy confirmed that iron was located in octahedral oxygen package.

The obtained samples and the parent FA were analyzed for iron-binding capacity using ferrozine test. Ferrozine and Fe^{2+} form a complex with violet color. Addition of both FA, FAFe2 and PMFe1 caused a decrease in color intensity. It is indicative of inhibitory activity and binding capacity of these samples. The control experiment was carried out with EDTA. The results are presented in Fig. 1.



Figure 1. Concentration dependence of inhibition activity of the samples tested

The highest activity was characteristic of PMFe1 and FA, who should very high activity in binding Fe(II). Activity of FAFe2 was similar to control, indicative of high activity of all samples tested. The obtained results may open a way for preparing FA-based iron-containing compounds for iron replacement therapy. They can be also used for designing biocatalytic systems for facilitating microbial redox-reactions.

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The effect of electrolysis on size dispersion of humic substances

Zubov I.N.¹, Gavrilov S.V.², Kanarskiy A.V.², Khabarov Y.G.³, Orlov A.S.¹, Selyanina S.B.¹ ¹Laverov Federal Center for Integrated Arctic Research Russian Academy of Science, Arkhangelsk, Russia, gumin@fciarctic.ru

²Kazan National Research Technological University, Kazan, Russia

³Northern (Arctic) Federal University named after M.V. Lomonosov, Arkhangelsk, Russia

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The chemical and biological activity of humic substances (HS) determines their relevance in agriculture, production of sorbents and nanocarriers of various ions, etc. At that, they are characterized by complexity and inconstancy of the composition, which limits the possibility of application. There are various ways to modify the HS. Along with a change in the ionic composition, a significant part of them is aimed at stabilizing particle sizes in solutions, because this largely determines the properties (in particular, bioavailability) of the products obtained [1]. For the purification of such preparations, an electrolysis method is effective, which can also affect the processes of the association of macromolecules. Therefore, it is of interest to consider the effect of electrolysis on inert electrodes on the dispersion of HS molecules size in solution.

Humic substances were isolated and fractionated according to [2]. The electrolysis was carried out at a current density of $250 \text{ mA}/\text{m}^2$ on iron electrodes in an alkaline medium at a concentration of HS 1 g/I for 6 hours. The dispersion parameters were measured by quantum correlation spectroscopy (HORIBA LB-550) at an initial concentration of HS (1 g/I) and after dilution 200 times.

The obtained data confirm that during electrolysis in an alkaline medium, anodic dissolution of iron practically does not occur. Two groups of particles were detected in the initial preparations of HS: 2-12 nm in diameter (individual macromolecules and small associates) and 200-900 nm (larger supramolecular formations). With the increase in the concentration, an equilibrium shift towards large particles is observed. After electrolysis, the polydisperse bimodal character of the particle size distribution is preserved. At the same time, the system becomes more homogeneous: HS macromolecules form associates of a smaller size (50-400 nm), and the monomolecular fraction is not determined. The revealed changes are probably due to electromagnetic and thermal effects and are explained by an increase in the number of active centers, as a result of which the tendency of HS to association increases, but smaller aggregates are formed. This allows one to expect an increase in the bioavailability of HS as a result of electrolysis.

Thus, the analysis of molecules size dispersion showed that the HS particles are in the form of agglomerates in solutions. A significant contribution of associative processes to the formation of HS properties is revealed. During electrolytic exposure, the dispersion of the particle size changes: it becomes more uniform, and the particle size decreases. This can positively affect the bioavailability of humic preparations.

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References

- 1. Canellas L.P. et al. // Scientia Horticulturae. 2015. 196:15-27.
- 2. Zavarzina A.G. et al. // Soil Science. 2008. 12:1–9 (In Russian).

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Tritium label in studying composites of humic substances with nanodiamonds

<u>Abmetko I.V.</u>¹, Chernysheva M.G.¹, Kulikova N.A.^{1,2}, Badun G.A.¹, Perminova I.V.¹ ¹Lomonosov Moscow State University, Moscow, Russia, ira_abmetko@mail.ru ²Bach Institute of Biochemistry of RAS, Moscow, Russia

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Tritium labeled compounds can be used as a tracers of compound concentration in different systems including biological objects [1]. The method is characterized with high sensitivity and the detection level is determined by the specific radioactivity of the labeled compound.

In present study the approach was applied for characterization of adsorption composites of detonation nanodiamonds and humic substances (HS). We used nanodiamonds (Aldrich) as received and subjected to air annealing that results in the reversion of zeta potential of nanoparticles in the aqueous suspension from positive to negative value. Coal humic acids and peat fulvic acids were used. Tritium label was introduced in the HS using tritium thermal activation method according to the procedure [2].

Composites were purified from non-bound HS and final composites were characterized by dynamic light scattering to determine particles size and zeta potential in the aqueous suspensions and liquid scintillation spectrometry to direct determination of HS surface concentration. To analyze the composites uptake and biodistribution in plants adsorption complexes of non-radioactive HS and tritium labeled nanodiamonds were obtained [1, 3].

In the presentation we are going to discuss the peculiarities of HS adsorption on nanodiamonds, the influence of HS type and surface properties of nanodiamonds. It was found that equilibrium adsorption of fulvic acids on positively charged nanodiamonds riches 80 mg/g and decreases to 30 mg/g after washing the composite from poorly bound HS. In the case of nanodiamonds subjected to air annealing adsorption was as much as 3 times lower.

Moreover, both initial and annealed nanodiamonds, as well as its adsorption composites were tested in the matter of how it can be uptake by wheat seedlings. In this experiments tritium labeled nanodiamonds [3] were used in both in the control experiments with plants and preparation of adsorption composites with HS following by interaction with wheat seedlings. It was shown that adsorption on roots was less for nanodiamonds subjected to air annealing, while penetration into above-ground parts was similar for both types of nanodiamonds. The detail results will be shown in the presentation.

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References

- 1. Chernysheva et al. // J. Soils Sediments. 2018. 18:1335–1346.
- 2. Badun et al. // Radiochim. Acta. 2010. 98:161-166.
- 3. Badun et al. // Radiochim. Acta. 2014. 102:941-946.

Lipid biomarkers under broad-leaved forest plant community as the proxies for the revealing stabilization mechanisms of the huminomineral soil matrix

Bazhanova A.¹, Zavgorodnyaya Yu.¹

¹Lomonosov Moscow State University / Department of Soil Science, Moscow, Russia, abazh.msu@gmail.ru

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n-Alkanes attribute among the most hydrophobic organic molecules of soil lipid fraction that allows them to be associate with an organo-mineral matrix forming resistant complexes. n-Methyl ketones are produced during first steps of n-alkanes degradation. Annual dynamic of main alkane homologues ratios – CPI, OEP, LSR, calculated for soil density fractions allows to estimate the nature of the organo-mineral associations in the natural systems should not only open up a new road towards improved understanding of organic C and N dynamics in soils and sediments, but also facilitate the modeling of the fate and transport of contaminants [1].

The test plot was selected under lime tree plant community where typical Albic Retisols are developed. Sampling was made from leaf litter, roots of topsoil, soil of horizons AY, AYel, EL, BT, twice in autumn and once in summer. Soil density separation was performed using flotation method in heavy liquid ZnCl2 (ρ = 1.60 g cm-3), the two fractions was separated – the free light fraction and the heavy one. In these samples total C and N concentrations were determined with a CHNS analyzer (Elementar). Lipid components were extracted by the method of pressurized fluid extraction in ASE 200 (Dionex). Sample purification and fractionation were carried out in an adsorption column with aluminum oxide. The qualitative identification and quantitative determination of target components were performed by GC/MS Agilent 6890N (Agilent Technologies) with a quadrupole mass-selective detector MSD5973N (Agilent Technologies).

The relative content of n-alkanes in SOM increases for the bulk samples down the soil profile up to 2 times and reaches maximum quantity in the EL horizon -725 μ g/gCorg. Analysis showed significant seasonal changes in the content and compositions of n-alkanes. In summer their amount in light fraction is about 1-2% of total carbon, and increased up to 4-8% during the autumn. n-Alkane content increases from 195,3 to 360,6 μ g/gC in bulk soil with the dominance in the upper part of the profile of long-chain plant origin alkanes (C27, C29) and an increase of the part related to medium-chain in underlying horizons as a result of a greater contribution of bacterial biomass (LSR increases from 8.5 to 10.5).

The maximum content of even n-alkane homologues in the soil was observed during the leaf and root residual inflow and the process of its subsequent microbial degradation, which were detected by the shifting of homologs composition ratio. The content of ketones in the free OM fraction is low, because ketones formed during the oxidation of detritus of alkanes quickly undergo further biodegradation. At the same period, there was a relative increase of the content of n-methyl-ketones associated with the mineral part of the soil. This is possible during the mineralization of the corresponding n-alkanes, strongly bound in the layers in organo-mineral complexes of soil particles, but available to microorganism action. n-Alkanes become vulnerable to fermentative oxidation, which contributes to surface n-methyl-ketones enrichment [2] that slows down the process of n-methyl-ketones mineralization.

References

1. Kleber M. et al. // Biogeochemistry. 2007. 85(1):9-24.

2. Lehmann J., Kleber M. // Nature. 2015. 528: 60–68.

Study of the effect of liquid humic preparations from vermicompost on the germination of soybean seeds of the "Zhansaya" variety

Boguspayev K.K., Titov I.N., Faleyev D.G., Azimova E.D., Akilbekova A., Nusupov A.A. al-Farabi Kazakh National University, Almaty, Kazakhstan, kboguspayev@yandex.ru

Keywords: vermicompost, alkaline extraction, soybean seeds, germination

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The technology for producing humic biological products from vermicomposts (biohumus) by alkaline treatment in the liquid phase allows one to obtain concentrated preparations containing almost all components of vermicomposts, including humic acids in high concentrations. In the experiments, for the production of liquid humic products used vermicompost obtained in "comperemedia" piles of cattle manure, with compost worms *Eisenia fetida*. Vermicompost (humidity 60-70%) was loaded in the vessel was added 0.1 M solution of potassium pyrophosphate in 0.1 N. the solution of KOH. Alkaline extraction was performed with constant stirring of the mixture at 80°C for 30 min. The resulting mixture was sedimented for 30-60 minutes; the supernatant (alkaline extract) was poured into the storage tank. The alkaline extract was cooled to room temperature and, to obtain the finished product, neutralized with a solution of nitric acid HNO₃ to a pH of 7.0–8.0 [1]. Sterile "Zhansaya" soybean seeds were placed in Petri dishes with a diluted preparation (1:100) and germinated at room temperature in a germination chamber. Distilled water was used as a control. The duration of the experiment is 7-8 days. The experiment was carried out in three agility.

In fig. 1 presents the results of experiments on the germination of soybean seeds of the variety "Zhansaya" using a liquid humic preparation from vermicompost at a dilution of 1: 100



Figure 1. Zhansaya soybean seedlings treated with a liquid humic preparation in a 1: 100 dilution. Left - control, without treatment (distilled water).

Morphometric indicators were used to evaluate the effect of the drug: the length of the main root (mm), the number of lateral roots of the main root (pcs). As a result, it was found that the average length of the main root in the experiment was 195 mm, in the control - 130 mm.), The number of lateral roots of the main root in the experiment was 19 pcs., In control 7, which clearly indicates the positive effect of liquid humic preparations on the germination of soybean seeds.

References

1. Titov I.N. // In: Materials of the IX International Scientific and Practical Conference daRostim 2013.

The influence of fulvic acid on seed germination and plant productivity

Brazienė Z., Paltanavičius V., Avižienytė D.

LRCAF Rumokai Experimental Station, Klausuciai, Lithuania, zita.braziene@lammc.lt

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Intensive field cultivation technologies cause a degradation of the soil. One of the ways to increase the activity of the soil and the productivity of plants is to use biological preparations and bioorganic fertilizers [1]. Fulvic acid application increase root activity [3], improve growth of plants and yield quantity and quality, limiting the development of some pathogens [2]. Also, fulvic acids improve soil physicochemical conditions [4].

The vegetation and field experiments were conducted in 2017-2018 at the Rumokai Experimental Station of the Lithuanian Research Centre for Agriculture and Forestry. The preparation Mineral Ful (bioactive fulvic acid complex) was used in our experiments.

Fulvic acid increased spring wheat seed germination 6.1 % (Table 1), height of shoots -2.9 %, air-dry mass of shoots -12.5 %, air-dry mass of roots -22.7 % (Table 2) compared to control. Fulvic acid increased the spring wheat grain yield 14.7 %, the sugar beet roots yield -7.9 % (Table 3).

Table 1. The influence of fulvic acid and chemical stain on wheat seed germination

Treatment	Amount (%) of germinated seeds		Amount (%) of seeds damaged by		
	After 3 days	After 7 days	Fusarium spp.	Monographella nivalis	
Control	79.5	90.3	3.8	2.7	
Fulvic acid	87.0	95.8	1.8	2.0	
Stain	43.0	92.2	1.2	1.0	

Table 2. The influence of fulvic acid and chemical stain on wheat shoots and roots

Treatment	Shoots	Mass of shoots, g		Mass of roots, g	
	height, cm	Fresh	Air-dry	Fresh	Air-dry
Control	20.7	8.3	1.6	8.2	2.2
Fulvic acid	21.3	9.7	1.8	11.6	2.7
Stain	15.7	8.0	1.4	9.1	2.5

Table 3. The influence of fulvic acid on spring wheat and sugar beets yield

Treatment		Spring wheat		Sugar beet		
	Yield,	Amount of crude protein,	Yield,	Sugar content,		
	t ha⁻¹	%	t ha⁻¹	%		
Control	2.281	9.8	89.54	15.06		
Fulvic acid	2.616	10.5	96.58	15.30		

References

1. Jakienė E., Spruogis V. // Žemės ūkio mokslai. 2015. 22(3):107–120 (In Lithuanian).

2. Moradi P. et al. // J. Centr. Europ. Agric. 2017. 18(3): 584-597.

3. Schmidt W. et al. // In: Hartmann A., Schmid M., Wenzel W., Hinnsinger P. Rhizosphere – Perspectives and Challenges, Elsevier. 2004. 1171–1178.

4. Yang et al. Journal of Soil Science and Plant Nutrition. 2013. 13(3):526–533.

The effect of biohumus on the growth of seedlings of *Scorzonera tau-saghyz* Lipsch. et Bosse in the laboratory

Faleyev D.G., Boguspaev K.-K.K.-K., Faleyev E.G., Myrzagaliev J.J. Scientific-Research Institute of Biology and Biotechnology, Almaty, Kazakhstan, ex-eko@yandex.ru

Key words: Scorzonera tau-saghyz, Eisenia fetida, biohumus, vermekulturs, rubber

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Scorzonera tau-saghyz is an endemic of Kazakhstan, capable of accumulating up to 40% of rubber in dry roots in quality not inferior to rubber of hevea. Currently, research is underway to restore the abundance of this species, as well as to develop effective and cost-effective biotechnologies for producing commercial rubber from *S. tau-saghyz* roots [1, 2].



Figure 1. Appearance of the root system of *S. tau-saghyz* seedlings grown without biohumus (1) and with the addition of 20% biohumus (2) (on day 172 of the experiment).

Laboratory studies have shown that the application of vermicompost in the amount of 20% contributes to the improvement of growth parameters and survival of seedlings *S. tau-saghyz*. So, when you make vermicompost average height and number of leaves were almost 1.5 times, the dry weight of the aboveground part of seedlings *S. tau-sagyz* more than 2 times, the survival rate of seedlings by 7.1% higher than in the controls. Most of the studied morphological parameters of the root system *S. tau-saghyz* did not differ significantly, while the number of lateral roots of the main root, and the maximum length of lateral roots adventitious roots 1st order when making vermicompost were, respectively, 2 and 4 times higher than in controls. The use of vermicompost can substantially contribute to the development of cost-effective modern biotechnology aimed at restoring the strength of nature and getting a commercial rubber root *S. tau-saghyz*.

The data obtained can be used in the development of cost-effective modern biotechnology to restore the number of naturally rare and endangered species, rubber tau-sagyz and a commercial rubber from roots *S. tau-saghyz*.

References

- 1. Omo-Ikerodah E.E. et al. //African J. Biotechnol. 2009. 8(25):7303–7307.
- 2. Богуспаев и др. // Вестник КазНУ, серия экологическая. 2013. 2/2(38): 64-70.

Beneficial effect of "EldORost" on seed germination energy of wheat, mung beans and radish

Filippova O.I.¹, Kulikova N.A.¹, Konstantinov A.I.¹, Grigoryeva I.O.¹, <u>Zhilkibayev O.T.²</u>, Perminova I.V.¹

¹Lomonosov Moscow State University, Moscow, Russia

²Al-Farabi Kazakh National University, Almaty, Kazakhstan, zhilkibaevoral@mail.ru

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"EldORost" is a universal organic fertilizer and natural plant growth regulator. It consist of salts of humic and fulvic acids (12%), a complex of minerals and trace elements, amino acids, flavonoids and etc. Among the numerous hypotheses about the nature of the physiological activity of humic substances (HS), the hypothesis about their hormone-like action, which is primarily understood as an auxin-like effect, has recently become the most popular [1]. Therefore, HS is widely used in agriculture as a stimulator for plant growth at the early stages of their development. Our study was aimed to estimate effect of "EldORost" on germination energy of crops belonging three agricultural groups: grains (on the example of wheat *Triticum aestivum* L., cv. L1), legumes (on the example of mung bean *Vigna radiata* (L.) R.Wilczek. cv. "Pobeda-104") and small-seeded vegetables (on the example of radish *Raphanus sativus* L. var. *sativus* cv. Krasnyi Velikan). To reach this goal, plant seeds were germinated in the presence of "EldORost" of different concentration; distilled water was used as a control.

The content of organic carbon (OC), humic and fulvic acids in "EldORost" measured using TOC analyzers Shimadzu (Japan) were 68, 59 and 9 g OC/I respectively. Meanweighted molecular mass was 7.4 kDa, and polydispersity was 3.4. Carbon distribution among structural fragments determined using 13C NMR spectroscopy demonstrated relatively high content of carboxylic groups (13%) and aromatic structures (44%). "EldORost" increased seed germination energy for all the studied plants (Figure 1).



Figure 1. Effect of "EldORost" on germination energy.

Our results demonstrated that maximum beneficial effect of "EldORost" was observed when initial solution of plant growth stimulator was diluted 150 times, whereas more concentrated solution were less efficient. So, "EldORost" is a promising product for pre-sowing seed treatment.

References

1. Olaetxea M. et al. // Appl. Soil Ecol. 2018. 123:521–537.

Testing of drugs used for pre-sowing seed treatment

<u>Grekhova I.V.</u>, Gilmanova M.V., Bazhutina L.A. GAU of Northern Trans-Urals, Tyumen, E-mail: grehova-rostok@mail.ru

Keywords: pre-sowing seed treatment, humic preparations, seedling method.

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To obtain high crop yields, the intensity of the initial processes of plant growth is of great importance. Scientific studies have established that pesticides can have a depressing effect on the physiological processes of cultivated plants. Some protectants have a negative effect on the linear growth of seedlings and germination. The effectiveness of pre-sowing treatment of seeds can be increased by the introduction of regulators or fertilizers into the solution of the protectants.

Research objective: evaluation of preparations for pre-sowing seed treatment by the method of seedlings.

The method of seedlings is based on the reaction of the test culture and allows you to determine the effect of drugs on the growth and development of plants. The following drugs were studied: Rostok – a humic preparation from peat (manufacturer of LLC «SPC "Evrika", Russia), Gumisol – a humic preparation from vermicompost (manufacturer of LLC «Agrofirm «Hermes», Ukraine), PreCedeTM – an aqueous mixture of nutrients (manufacturer ATR Nutrition, Canada). The seeds of the test culture (spring wheat) were soaked in water and preparations in a concentration according to the recommendations of the manufacturers. In glasses (500 ml) filled with dark gray forest soil, 15 pieces were sown per vessel. The vessels were placed in an air conditioner at t = 20°C with a changeable program of day and night. Four repetitions. In the course of the experiment, the germination energy (on day 4), germination (on day 7), the length and mass of the root system and the terrestrial part of plants (on day 14) were recorded.

The seed germination energy in the control was 32%, the tested drugs increased it: Rostok – 2.1 times, $PreCede^{TM}$ – 1.8 times, Gumisol – 1.6 times. Germination was significantly increased by Rostok and $PreCede^{TM}$ preparations by 17 and 15%, respectively. The drug Humisol reduced germination by 3%; the difference with the control is not significant.

The development of the root system was positively affected by all the drugs studied, but to a different extent. The number of roots was most formed during seed treatment with Rostok, an increase of 18% to the control. $PreCede^{TM}$ and Gumisol increased the number of roots by 9 and 3%, respectively, but the increase in the use of the last drug is not significant. According to the length of the root system, the difference between the preparations is not significant, an increase relative to the control: Rostok – 27%, $PreCede^{TM} - 27\%$, Gumisol – 24%. The mass of the root system exceeded the control: $PreCede^{TM} - 27\%$, Rostok – by 53%, Gumisol – by 19%. The difference between the first two drugs in this indicator is not significant; the last drug is significantly inferior to the previous ones.

According to the length and weight of the plants, the greatest increase in control was obtained from the use of the Rostok preparation – 38 and 93%, respectively. PreCedeTM had an average effect on these parameters, an increase of 17 and 24%. The lowest increases from the drug Gumisol – 10 and 2%, respectively, the increase in plant mass is not significant.

Thus, we recommend adding the Rostok humic preparation to the protectant mixture.

Effects of lignohumate and biochar on plant responses in humus-poor and humus-rich soils polluted with metals

Kiryushina A.P.¹, Paramonova A.I.^{1,2}, Korolev P.A.^{1,2}, Uchanov P.V.¹, <u>Terekhova V.A.²</u> ¹Institute of Ecology and Evolution, Russian Academy of Sciences, Moscow, Russia ²Lomonosov Moscow State University, Moscow, Russia, vterekhova@gmail.com

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Various organic materials are widely used in agriculture with the aim of improving soil quality, plant nutrients regime, and plant production. Both lignohumate and biochar, coproducts of thermochemical conversion of lignocellulosic materials, are marketed as universal soil improvers [1,2]. Their sorption properties are especially relevant in case of polymetallic soil contamination, since these substances have a sufficiently high exchange capacity with respect to metals. However, rather conflicting data on the effectiveness of these products as well as ambiguous effects of their combined and separate applications are published.

We investigated the impact of biochar (70-80% pure carbon, Company "Metakom", Russia) and lignohumate (C total 36 %, Company "RET", Russia) on plant responses in two typical agricultural topsoils (0-20 cm): Chernozem (Voronezh region, organic carbon 5.4%) and Agrozem (rice fields of Kalmykia, organic carbon 1.5%). Natural and metal-spiked (650 mg/kg Pb + 1100 mg/kg Zn + 660 mg/kg Cu) soil samples were analyzed. One week after metal spiking, biochar (5%) and lignohumate (0.25 %) were added. The pots with soil samples were incubated for 120 days at 22-24°C and 60% relative humidity. We analysed growth parameters (seeds germination, roots and sprouts length) of seedlings of three species: *Sinapis alba* (mustard), *Raphanus sativus* (radish), *Avena sativa* (oat). Each experimental had 3 replicates.

The results indicate that: 1) effects of exogenous organic matter application on plant responses varied greatly between humus-poor and humus-rich soils; biochar and lignohumate amendments significantly increased plant root and spout length in chernozem, whereas their effect was slight in the humus-poor soil; 2) effect of lignohumate and biochar mixture was significantly higher than that of lignohumate amendments (radish and oat - roots length more for 39-36% and roots length - 25-28%); 3) biochar added to humus-poor soil caused a strong inhibitory effect on seed germination of all plant species, sprout length by 35%, and root length by 15%; the combined introduction of lignohumate and biochar had a small positive effect only on oats. Interestingly, on unpolluted samples test parameters of *A. sativa* were more informative, while in the presence of heavy metals in the samples, plant reactions to biochar and lignohumate were better seen on mustard *S. alba* which is used as green fertilizer ("green manure crops") or "siderites".

Thus, the results indicate that effectiveness of biochar and lignohumate amendments (both alone and in mixtures) have the potential to substantially improve the quality and fertility status of a certain type of agricultural soils polluted with high concentrations Cu, Pb and Zn. No significant plant-stimulating effects were detected in humus-poor soil neither in polluted any un-polluted samples.

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References

- 1. Perminova I.V. et al. // In: Viable Methods of Soil and Water Pollution Monitoring, Protection and Remediation, Springer Netherlands, 69:249–274.
- 2. Pospíšlová Ľ. et al. // J. Life Sciences. 2016. 10:153–160.
The effect of HA preparations of different soils on the physiological processes of the *Chlorella vulgaris* algae

<u>Toropkina M.A.</u>, Rumin A.G., Chukov S.N. Saint Petersburg State University, Russia, S.Chukov@spbu.ru

Keywords: humic acids, physiological activity, 13C-NMR, aromatic structures

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Physiological activity is one of the important environmental functions of humic acids (HA). We are studying the physiological effect of HA preparations on the unicellular green alga *Chlorella vulgaris*. To assess the effect of HA preparations on algae, indicators such as photosynthesis, respiration, cell numbers and sizes were used.

The structural and functional properties of HAs isolated from contrasting soils vary greatly. This is due to the specific bio-hydrothermal conditions of humification, which determines the molecular structure of the preparations. The content of aromatic structures in HA molecules was determined by 13C-NMR. The content of free radicals was evaluated by method ESR.

The study of the rate of photosynthesis and respiration of Chlorella vulgaris was carried out by the oxygen content in the light and in the dark. The duration of the interaction of algae cells with HA ranged from 1 to 5 days. The size and abundance of the algal cell culture were estimated by direct counting under a microscope using a computer program for analyzing graphic images (in Python), as well as a Shimadzu SALD-2201 laser diffractometer.

During the research, it was found that HA preparations favorably affect the physiological processes of the culture of the alga *Chlorella vulgaris*. In the range of concentrations from 0.001 to 0.007%, HA concentrations of up to 0.003% are optimal. At HA concentrations of more than 0.003%, a negative effect of HA is observed for all preparations: photosynthesis decreases in *Chlorella vulgaris* cells and destructive processes increase sharply, causing an increase in oxygen consumption.

The positive effect of the aromaticity of the preparations and the concentration of free radicals of chernozem HA on the acceleration of photosynthesis of *Chlorella vulgaris* was shown. HA of sod-podzolic soils with low aromaticity and low content of free radicals stimulate mainly the processes of reproduction.

As a result of the studies, a tendency of a positive effect of aromaticity and concentration of free radicals of HA preparations on the acceleration of photosynthesis of *Chlorella vulgaris* was noted. On the contrary, the stimulation of the processes of the *Chlorella vulgaris* culture reproduction can be associated with a low concentration of free radicals and a small fraction of aromatic fragments in the HA molecules.

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Effects of some humic substance(s) based plant activators on growth, yield and quality parameters of cherry tomato cultivar under regular and stressed soil conditions

<u>Turan M.</u>¹, Yıldırım E.², Ekinci M.², Kılıç M.¹, Bolouri P.¹, Hijazi N.¹ ¹University of Yeditepe, İstanbul, Turkey, m_turan25@hotmail.com ²University of Atatürk, Erzurum, Turkey

Keywords: plant activator, cherry tomato, growth, yield, mineral content, stressed soil

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In this study, the effects of some plant activators on plant growth characteristics, yield and biochemical properties of cherry tomatoes grown in two different soil media (stressed and regular soil) were investigated under controlled greenhouse conditions. Studies were conducted in pots. Powhumus (PH), seed covering (SC), root enhancer (RE) and fulvic acid (FA) were treated as seed and drench applications. Treatments were grouped as follows: Control 0: no treatments, PH 1: PH seed coating, SC 1: SC seed coating, PH2: PH seed coating +PH, SC 2: SC seed coating +PH, RE: RE seed coating +RE, FA: FA seed coating +FA. In order to determine the effects of the applications on plant growth parameters were examined, such as dry plant and root weight, plant height, stem diameter, leaf number; yield parameters such as fruit number per cluster, fruit height, fruit diameter, fruit weight, cluster weight, cluster number per plant and yield per plant; mineral content of leaf, root and fruit; chlorophyll reading value (SPAD); and some fruit characteristics such as total soluble solids (TSS), fruit dry matter (DM), and vitamin C. Furthermore, some physiological and biochemical characteristics of tomato leaf such as malondialdehyde (MDA), hydrogen peroxide (H_2O_2), proline and sucrose, and antioxidant enzyme activities (catalase-CAT, peroxidase-POD and superoxide dismutase-SOD) were also determined. All plant activators improved plant growth and yield of tomato under both soil conditions. Plant activator applications increased the plant height, stem diameter, leaf number, SPAD, plant dry weight and root dry weight, and the highest effect were determined as 59% (FA), 47% (RE), 54% (SC2), 38% (FA), 60% (RE) and 70% (RE) in regular soil, and as 79% (FA), 50%(RE), 60%(FA), 42%(FA), 78%(FA), and 72% (FA) in stressed soil, respectively, as compared to the control 0 treatment. With the applications, increases in DM, vitamin C, TSS, fruit height, fruit diameter, fruit weight, cluster weight, cluster number per plant, fruit per cluster, yield per plant were 12%(RE), 33% (SC1), 29% (SC1), 15% (SC2), 23% (SC2), 43% (SC2), 45% (SC2), 21% (RE), 26% (SC1) and 56% (RE) in regular soil and 41% (SC2), 40% (SC1), 38% (SC1), 21% (FA), 26% (FA), 53% (FA), 54% (FA), 58% (FA), 58% (FA) and 81 % (FA) in stressed soil, respectively, as compared to the control 0. According to the study result, FA generally had more positive effects than the other treatments under stressed soil conditions. Applications were more effective on plant growth, fruit characters and yield parameters in stressed soil plants than those in regular soil. RE was the most effective application for mineral content (leaf, root and fruit) in regular soil, whereas the most effective application in stressed soil was FA. N. P, K, Ca, Mg, Mn, Na, Zn, B, Fe and Cl content (leaf, root and fruit) in regular soil was increased by 23-90% as compared to control, whereas this was ranged from 40 to 92% in stressed soil. CAT, POD and SOD activity, H₂O₂, MDA and proline content were decreased in regular and stressed soil with treatments as compared to control, but POD activity was more in SC 1 application in stressed soil. The content of sucrose elevated with especially RE treatment (57% increase in regular soil, 65% increase in stressed compared to control). In conclusion, plant activator treatments could be suggested to be used especially in cherry tomato cultivation in problematic soil areas.

The biological activity of native and modified humic acids

Zherebtsov S.I.¹, Votolin K.S.¹, Malyshenko N.V.¹, Ismagilov Z.R.^{1,2} ¹Institute of Coal Chemistry and Materials Science FRC CCC SB RAS, Kemerovo, Russia, sizh@yandex.ru

²Institute of Catalysis, SB RAS, Novosibirsk, Russia

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Chemical modification with the aim of changing the composition of humic acids (HA) and introducing new functional groups into their structure is a promising direction for improving and obtaining the necessary properties of humic substances. The greatest attention is drawn to their biological activity, which is associated with the ability of HA to participate in redox reactions in a plant cell, depending on the content of various structural groups (quinoid, phenolic), the degree of aromaticity, and the ratio of hydrophilic and hydrophobic fragments [1,2].

The purpose of the study is to evaluate the biological activity of native and modified humic acids isolated from brown coal of the Tisulsky deposit of the Kansk-Achinsk basin (TC) and its naturally oxidized form (TCO). The humic acids were modified with hydrogen peroxide and n-butanol. The change in the functional and structural composition during the modification was studied using EPR, ¹³C NMR CPMAS.

Tests were conducted to determine the biological activity of HA in the form of sodium humates (0.005% aqueous solutions) obtained from native and modified humic acids using "Iren" wheat seeds. Biological activity was evaluated by three test functions - seed germination energy (GE), root length (RL), seedling height (SH) and the phytoactivity index (FI) was calculated, which reflects the deviation of the test function from the control (seed germination in water).

Initial HA showed bioactivity in relation to wheat seeds. FI humic acids TC exceeded the control experience by 12%, series TCO by 20%. There is a tendency for indicators of all test functions to exceed control. For example: SH excess by 17% (HA TC) and 33% (HA TCO). When using HA modified with hydrogen peroxide of various concentrations, the dependence of the FI on the concentration of paramagnetic centers (PMC) in humic acids was found. Modification with hydrogen peroxide leads to a decrease in the content of PMC and a decrease in the biological activity of HA. When using HA modified by n-butanol alkylation, FI was 1.14 and 1.24; SH exceeded control by 29 and 37% for TC group and TCO group respectively. Humic preparations obtained from modified by n-butanol alkylation of HA can be considered promising for subsequent tests in field experiments.

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- 1. Kuharenko T.A. // SFC. 1976. 2:24–31.
- 2. Dobbs L.V. et al. // J. of Agric. Food Chem. 2010. 58(6): 3681–3688.

Autoradiographic study of the distribution of 55-iron(III)-EDTA chelate at the presence of humic substances in *Triticum aestivum L.*

<u>Zimbovskaya M.M.</u>, Kulikova N.A., Filippova O.I., Pankratov D.A. Lomonosov Moscow State University, Moscow, Russia, pankratov@radio.chem.msu.ru

Keywords: autoradiography, iron(III)-EDTA chelate, wheat

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EDTA is one of the widely used ligands for delivering minerals to plants. It is known that EDTA-ion forms stable, highly soluble compounds with many metals in a wide pH range. This provides easy transport of mineral nutrients to plant tissue. Therefore, EDTA complexes, for example, with iron are often used in laboratory practice as a control when studying the absorption of other iron-containing preparations by plants or in agrochemistry production for the operational prevention of plant diseases. In the first case, sometimes, iron compounds in the presence of humic substances (HS) are considered as alternative iron-containing preparations [1]. In the second case, the contact of the iron(III)-EDTA chelate with the HS of the soil or substrate on which the cultivated plant handled is inevitable. In any case, participation and possible role of HS in the distribution of iron-containing preparations general and, in particular, iron(III)-EDTA chelate are of interest.

Therefore, the aim of this work was to study the possible participation of HS coal in the distribution of iron(III)-EDTA chelate in the parts of wheat Triticum aestivum L. at root and foliar uptake. The autoradiographic method is studying the distribution of nuclides in a plant directly. It is based on the use of the sensitivity of some materials to radionuclide radiation. In the present work are used, BaFBr:Eu²⁺ luminescent screens in the Cyclone Storage System (Perkin Elmer) digital radiography system were used as a detector. As the detected radionuclide, 55-iron was used, which is characterized by optimal characteristics both in terms of radiation energy (mainly β -radiation - 231.4 keV) and half-life (2.73 y). With this radionuclide label iron(III)-EDTA chelate was synthesized, and the purity of which was confirmed by thin-layer chromatography. This radiation-emitting material was used to prepare four solutions: two for the experiment with root absorption, and two for the experiment with leaf absorption, differing between the pairs by the concentration of iron(III)-EDTA chelate (0.558 and 0.055 mg/l, accordingly). In each pair, one solution contained only iron(III)-EDTA chelate, and in another potassium humate was additionally added to a concentration of 1000 mg/l. The root uptake experiment was use 18 days of age wheat seedlings, which were placed for one day in vessels with a labeled solution. The leaf absorption experiment was carried out with seedlings of wheat, which were sprayed with a labeled solution and kept for 7 days. After germination, the plants were washed, dried and subjected to autoradiography. In total, 18 plants at root uptake and 16 plants at foliar uptake were studied. From the data obtained it follows that the presence of HS affects the distribution of iron(III)-EDTA chelate. Sic, the formation of "films" was observed the roots at accumulating the iron-containing preparation. HS contribute is evenly distributed iron contained preparation in the shoots at root uptake. In the absence of HS, the 55-iron is predominantly located at the points of the most intensive plant growth - at the tips of the roots, at the tips of the shoots, and also in the thickened lower part of the shoot. During foliar uptake of iron concentration is observed in the at root part of shoots, and in the presence of HS, the iron content is much higher. Thus, in the work the role of HS in the distribution of iron in such a strong complex compound as iron(III)-EDTA chelate is demonstrated.

^{1.} Kulikova N.A. et al. // J. Agric. Food Chem. 2017. V. 65, № 51. P. 11157–11169. DOI: 10.1021/acs.jafc.7b03955.

Section 3 Humic substances and microorganisms

Book of Abstracts Fifth International Conference of CIS IHSS on Humic Innovative Technologies «Humic substances and living systems» (HIT-2019) October 19–23, 2019, Moscow, Russia

Stabilization of soil organic matter and development microbial community in chronoseries of soils formation of south taiga ecosystem (bars of Ladoga Lake, Russian North-West)

Abakumov E.¹, Polyakov V.¹, Pershina E.¹, Ivanova E.¹

¹Department of Applied Ecology, Saint-Petersburg State University, Saint-Petersburg, Russia, e abakumov@mail.ru, e.abakumov@spbu.ru

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This abstract presents the result of the studies of soil formation on the different aged coastal bars in transgression zone of the Ladoga Lake in the Nizhnesvirsky Nature Reserve (Leningrad region, North-West of Russian Federation). The Ladoga Holocene transgression is one of the most informative and applicable models for pedogenesis, successional processes occurring in young and mature areas can be traced here. Local processes of soil formation such as podsolization, gleyfication, peat formation and humus accumulation were identified. Physical, physic-chemical and biological soil properties with a detailed description of the morphology of soil of different aged coastal bars are presented. The investigation presents the data on the soil chronoseries, located on the four Ladoga coastal bars with different ages from 70±25 to 1590±25 years BP. The trends of accumulation and transformation of organic matter, elemental composition of humic acids (HAs), development of plant communities and the influence of the soil formation factors on the rate of formation of soil horizons were estimated. The degree of soils organic matter stabilization has been assessed with the use of modern instrumental methods (spectroscopy of nuclear magnetic resonance (CP/MAS 13C-NMR). Analysis of the molecular composition of HAs showed that aliphatic groups accumulate in the soils. This is due to the high level of hydromorphic in this environment. Due to evolution of podzols, aliphatic groups and the accumulation of humic substances in the soil are increased, with increase of aliphatic groups in molecular composition of HAs it makes HAs more aggressive and leads to increase migration ability. The molecular composition of HAs of young soils is enriched with aromatic fragments and oxygen-containing functional groups, which ensures their high thermodynamic stability. Also the taxonomy composition of microbial communities of different-aged soil has been investigated. Different groups of microorganisms are considered as responsible drivers of various soil formation processes. Copiotrophic microorganisms (belonging to Proteobacteria. Actinobacteria and Bacteroidetes phyla) dominated in microbial communities of the topsoil (organic) horizons. Podzolic (E) horizons were characterized by the increased abundance of Mycobacterium sp. (Actinobacteria). In deeper horizons, the percentage of copiotrophic bacterial groups decreased providing the growth of oligotrophs, as well as archaeal lineages. The deepest (gleyic, G) horizons were characterized by the presence of anaerobic, methane-producing, bacteria.

Thus, the connection between microbial community structure and the continuous development of the soil profile was revealed. The change in physic-chemical parameters was the key driving factor causing the adaptation of the prokaryotic community. The older coastal bar expressed the clearer signs of podzol formation, the greater thickness of the E horizon, and the more obvious dissimilarity between the microbiomes in different genetic horizons. This indicates that microbial community can be considered as a driving mechanism, which regulates solum vertical differentiation within the time of podzol ontogenesis.

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The properties of humic acid – cytotoxic study

Bratishko K.A.^{1,2}, Buyko E.E.^{1,2}, Ivanov V.V.², Zykova M.V.² ¹Tomsk Polytechnic University, Tomsk, Russian Federation, https://tpu.ru ²Siberian State Medical University, Tomsk, Russian Federation, https://ssmu.ru/ru/

Keywords: humic acids, cytotoxicity, MTS

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Humic acids (HA) are applied in alternative medicine but for their implication in the official medicine a wide-range preclinical and clinical studies are needed. Cell-viability cytotoxicity test is necessary to evaluate potential safety profile of HA and to choose the proper concentrations for the future investigations. To conduct the cytotoxicity-test the peat HA sample has been chosen. Isolation of HA sample was performed by the pyrophosphate $(Na_4P_2O_7)$ extraction as described earlier [Zykova, 2018]. The raised pine-cotton-grass peat (R=30-35 %, 10-50 cm) was taken from peat oligotrophic bog in Tomsk region (Bakchar bog area, Russia). For this sample promising antiradical and antioxidant activities have been shown during the previous studies [Zykova, 2018].

The cytotoxic properties of HA were evaluated on 3T3-I1 cell-line using the standard MTS-assay (3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium). MTS-assay was performed after achieving 100% confluence of cells using the CellTiter 96® Aqueous Cell Proliferation Assay (Promega). Briefly, DMEM/F12 (100 μ l) was supplemented with MTS solution 20 μ l/well, incubated for 1 h, and then the absorbance was recorded at 490 nm with a 96-well plate reader Sunrise RC4 (Tecan).



Figure 1. Viability test of HA with MTS (3T3-L1, 24 h of incubation). Viability is expressed as % of control (mean). The results are given as the mean ± SD (n=6). * indicates a significant difference (multiple t-test) from control (p < 0.05).

Figure 1 shows the results of a cell mitochondrial activity viability test (MTS-test) after 24 h of incubation of 3T3-L1 fibroblastic cells with eleven different concentrations of HA from 3.125 to 400 μ g/ml. At 25, 50 and 100 μ g/ml HA there was no influence on the cell viability. However, a concentration-dependent toxic effect of HA was revealed on the cells cultivated with the 150 - 400 μ g/ml HA (for 13.0, 12.5, 12.9, 11.05, 11.2 % respectively). Alternatively, cell viability was increased in presence of 3.125, 6.25, 12.5 μ g/ml HA (for 18.5, 14.3, 7.5% respectively). This pronounced effect may have been caused by the increasing of cell proliferation at less concentrations of HA. The cytotoxicity at the higher doses of HA can be explained by the osmotic effect and ability of HA to penetrate nuclei and to influence on the DNA binding affinity of the transcriptional factors [Yang et al., 2002]. Eventually, HA doesn't demonstrate strong cytotoxic properties and allows to conduct investigations in wide range of concentrations.

Effect of peat humic acid on the composition of cytosolic soluble carbohydrates and lipids in *Alternaria alternata*

<u>Fedoseeva E.</u>¹, Tereshina V.², Danilova O.², Prudnikova E.³, Terekhova V.³ ¹Pirogov Russian National Research Medical University, Moscow, Russia, elenfedoseeva@gmail.com

² Winogradsky Institute of Microbiology, Research Center of Biotechnology of the Russian Academy of Sciences, Moscow, Russia

³Lomonosov Moscow State University, Moscow, Russia

Keywords: humic acid, micromycetes, Alternaria, stress, osmolytes, lipids, sterols, trehalose

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Humic substances can influence on various plant and microbial cell components such as plasma membrane, mitochondria, DNA containing components, and cytosol [3]. Such changes can be considered both positive and stressful for organisms. It has been known that fungi can respond to stress changing the composition of membrane lipids and synthesizing protective osmolytes [1] Melanin-synthesizing fungi, as a rule, are characterized by high resistance to external influences. The aim was to study the effect of humic acid (HA) on cytosolic soluble carbohydrates and lipids in melanin-synthesizing fungus *Alternaria alternata* to understand whether the action of HA is stressful.

Fungal culture was cultivated on a liquid Chapek medium with the addition of peat HA (100-200 mg/L) and without it as a control. Mycelium was investigated in trophophase. The method of lipid extraction inhibiting phospholipases was used to analyze the lipid composition of fungal cells. Quantitative analysis of the membrane lipids was performed using two-dimensional thin-layer chromatography. The method of gas–liquid chromatography (GLC) was used to study the composition of fatty acids in fractions of polar or individual lipids. The osmolytes were determined by the method of GLC trimethylsilyl sugar derivatives with an internal standard after extraction and purification.

Mannitol and trehalose dominated within the composition of osmolytes. Under the action of peat HA level of mannitol reduced, while the level of trehalose increased (by 1.5-2 times). Within membrane lipids, phosphatidylcholines, phosphatidylethanolamines, and phosphatidic acids dominated. However, more distinct effect of HA was observed in the structure of other lipid groups, namely an increase in the proportion of sterols and sphingolipids. It has been known that an increase in trehalose is a response to heat shock, in sterols - to a microbial attack; and a high proportion of phosphatidic acids in the membranes can indicate an overall adverse effect on living organisms [1, 2]. Thus the increase in the proportions of sterols and trehalose in our experimental research can be considered as stress response of fungal cells on peat HA.

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- 1. lanutsevich E.A. et al. // Microbiology (SGM). 2016. 162(6):989-99.
- 2. Mantil E. et al. // Colloids and Surfaces B: Biointerfaces. 2019. 178:94–102.
- 3. Zandonadi D.B. et al. // Planta. 2007. 225: 1583-1595.

Influence of humic substances on the decomposition of insecticides in rice fields of Bali Island

<u>Gasanov M.E.</u>¹, Zavgorodnyaya Yu.A.² ¹Skoltech CDISE, Moscow, Russia, gasanov.msu@yandex.ru ²Lomonosov MSU, Moscow, Russia

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The need to intensify agriculture in densely populated areas of South-East Asia leads to the active use of fertilizers and pesticides. For pest management of rice farmers often use insecticides of the group of neonicotinoids. It is known that humic substances affect the behavior of organic pollutants in the environment. Humic substances are able to bind toxicants, changing their migration ability, as well as to have a stimulating effect on the microbial community of the soil, accelerating the process of decomposition of organic pollutants.

The aim of our work was to assess the effect of humic product on the transformation and migration of pesticides in soils. The object of the study was widely used on the island of Bali drugs based on neonicotinoid insecticides: thiamethoxam, Imidacloprid sand fipronil. During the model experiment, soil samples with insecticides and humic preparation were incubated for 14, 28, 56 days at a temperature of 25°C. Humic preparation was introduced as a solution with a concentration of 0.25% (5 ml per 25 g of soil). During the experiment, the indicators of the microbial state of the soil (basal respiration, nitrogen fixation, methane formation) were measured. The method for determining pesticides in soil is based on acetonitrile extraction and subsequent determination on a chromato-mass spectrometer (HPLC-Q-TOF).

The results show an increase in methane formation in the soil on day 7, which can be associated with a significant number of methanogenic bacteria in flooded rice cakes. As a result of the experiment, after 14 days, a decrease in the content of Imidacloprid in the variant with the introduction of humic substances by 30% was revealed. The decrease in the detectable pesticide in soil may be due to the specific effect of humic preparation on certain groups of microorganisms.

Peculiarities of antioxidant activity of humic substances

<u>Kudryasheva N.S.</u>^{1,2}, Kovel E.S.¹, Sachkova A.S.³, Rozhko T.V.⁴ ¹Institute of Biophysics FRC KSC SB RAS, Krasnoyarsk, Russia, n_qdr@yahoo.com ²Siberian Federal University, Krasnoyarsk, Russia ³National Research Tomsk Polytechnic University, Tomsk, Russia ⁴Krasnoyarsk Medical Academy, Krasnoyarsk, Russia

Keywords: antioxidant activity, enzymatic bioassay, humic substances, bioluminescence, reactive oxygen species

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Humic substances (HS) are known to function as natural toxicity attenuators in natural solutions, involving solutions of redox-active compounds. In our study we monitored toxicity of oxidizer solutions under addition of HS.

Bioluminescence bacteria-based and enzyme-based bioassays were used to monitor toxic and antioxidant effects at cellular and biochemical levels, respectively. The bioluminescence assays are widely used to monitor toxicity of media due to high rates of the assay procedures and simple registration of the luminescent signal. These features provide a large number of measurements under comparable conditions, which is extremely important for statistical processing. A suppression (or activation) of maximal bioluminescence intensity is a quantitative characteristic of general toxicity (GT) in the environment. A particular feature of the enzymatic bioassay is specificity to the group of toxic agents of oxidative nature: additionally to reducing maximum luminescence intensity, a bioluminescence delay period occurs; it depends non-additively on the oxidizer' concentration and redox potential. This feature is a basis for monitoring the oxidative toxicity (OxT) of the environment. Hence, the bioluminescent enzymatic bioassay can be used to determine both: changes of GT in high-concentration solutions of HS and ability of HS to change OxT of media, i.e. antioxidant activity of HS.

Toxicity and antioxidant activity of HS was studied in a wide range of their concentrations. The toxicity of HS solutions was evaluated with effective concentrations EC-50; detoxification coefficients D_{OxT} and D_{GT} were used to evaluate antioxidant activity in solutions of organic and inorganic oxidizers. The toxicity and antioxidant activity of HS were compared to those of the other bioactive compound, fullerenol-60 (F). Differences in the effects of HS and F were related to (A) the mobility of functional groups and fragments of HS, (B) their higher ability to reduce the content of reactive oxygen species (ROS) in the solutions. The peculiarities of the HS antioxidant effect were: the low active concentrations of HS, dependence of their antioxidant activity on time and oxidizer' hydrophobicity.

Change of ROS content under addition of HS was studied in tritiated water (Tritium is a beta-emitting radionuclide of low-specific radioactivity, product of a lot of radiochemical processes, which is intensively accumulated in environment now). Tritium and HS demonstrated synergetic activation effect on the bioluminescence enzymatic bioassay. Content of ROS was evaluated by chemiluminescence luminol assay. Correlations between bioluminescence response and ROS content were found in solutions of HS+ tritiated water, revealing molecular aspect of HS antioxidant function in diluted solutions. Additionally, statistical processing revealed a decrease of stochasticity in tritium low-dose effects under exposure to HS.

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Effect of humic compounds on adsorption of virulent bacteriophages on host cells: case study for *Escherichia coli* and C600-9g, and *Staphylococcus aureus* A515 – bacteriophage phA515

Letarova M.A.¹, Perminova I.V.², Letarov A.V.^{1,2} ¹Federal Center of Biotechnology, Vinogradsky Institute of Microbiology, RAS, Moscow, Russia,

letarova.maria@gmail.com

²Lomonosov Moscow State University, Department of Chemistry, Moscow, Russia

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Bacteria and bacteriophages successfully coexist for more than two billion years. In addition to biological mechanisms, a significant contribution to this phenomenon is made by such a factor as the presence of humic substances (HS). It is particular importance for high-density ecosystems. The aim of this work was to assess contribution of HS into adsorption of bacteriophage on the surface of the host cell. We used two laboratory models of the host and virulent bacteriophage, nominally: Escherichia coli (str. C600) and bacteriophage 9g, and clinical isolate Staphylococcus aureus (str. A515) and bacteriophage phA515. E. coli C600 is a representative of Gram-negative bacteria, whereas St. aureus A515 is a representative of Gram-positive microorganisms. Both host strains were first grown in LB medium overnight, diluted, was grown to logarithmic phase and then washed with M9 minimum medium. The cultures of bacteriophages were added to the cells with multiplicity of one phage particle per hundred cells. Then, the content of free phage was measured. Three HS samples were tested in this study: aqueous HA (SRHA), peat HA and coal HA. They were added to test media in concentrations of 100 µg/mL. For *E. coli C600 / 9g*, an increase in the bacteriophage single burst was observed: up to 1200 particles per cell compared to 800 in the control. In the presence of peat HA, the phage unit cycle did not change, but the yield of phage particles was five times larger than for SRHA. The presence of coal HA caused an increase in the amount of adsorbed phage particles and a decrease in the half-adsorption time of phage particles (from 5 to 2.5 min). For the St. aureus A515 / phA515, the presence of all HS samples caused an increase in the time of half-adsorption from 4 to 8 min, which might be associated with enhanced aggregation of the cells as compared to control. The presence of SRHA has also caused an increase in the release time of phage particles without an increase in the page yield per cell. In case of SRHA, it can be concluded that we observed an increase in the single burst time. Conclusion can be made that HS affect all most important environmental characteristics of bacteriophage. This may be important for explanation of the persistence of virulent bacteriophages and of their hosts in natural ecosystems. Some ways for a use of HS in controlling phage cycle might be suggested.

Synthesis and redox properties of quinonoid-enriched humic substances prepared with a use of Fenton reaction

<u>Mareev N.V.</u>, <u>Molodykh A.A.</u>, Melnikova S.V., Bil I.V., Volikov A.B., Perminova I.V. Lomonosov Moscow State University, Moscow, Russia, mr.n.mareev@yandex.ru, alex-molod@yandex.ru

Keywords: humic, landfill, redox, capacity, quinone, methanogen, inhibitor

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Gas emissions from the landfills pose a threat to human health and provide a large source of greenhouse gases. Of particular concern is methane emission. The existing solutions to suppression of methane emission are costly and not efficient. New biocatalytic systems are needed. It has been reported in the literature that humic substances (HS) are capable of inhibiting methane synthesis by methanogenic bacteria. HS are natural polymolecular systems with numerous hydroxy- and carboxy-aromatic units. For enhancing inhibition ability of HS, it is necessary to increase their acceptor properties determined by the content of quinone units in the structure. Previously, we described a use of phenol formaldehyde condensation as the synthetic approach for preparing quinonoid-enriched HS [1]. In this work, we use Fenton oxidation for developing the green synthesis of quinonoid-enriched HS. Potassium humate from leonardite (CHP) and fulvic acids (FAP) were used for modification. Structure of the prepared products was studied using FTIR-and NMR- spectroscopy. Quinonoid centers used in this study are presented in Figure 1.



Figure 1. Quinonoid centers incorporated into the humic materials used in this study: hydroquinone (1), 2-methylhydroquinone (2), 1,4-naphtoquinone (3), 2-hydroxy-1,4-naphtoquinone (4), 1,4-dihydroxy-9,10-anthraquinone (5).

The redox capacity of all derivatives was determined as described in [2]. The method is based on reduction of $K_3Fe(CN)_6$ to $K_4Fe(CN)_6$. To get information on distribution of quinonoid-centers between molecular components of HS, we fractionated the CHP-HQ250 derivative on SPE PPL cartridge at pH 2 and 7.

According to the redox measurements, the reducing capacity of CHP was very low – 0.3 mmol/g, the values for its derivatives with anthraquinone (AQ) and naphthoquinone (NQ) were close to 0. Much higher reducing capacity (1.3 mmol/g) was obtained for hydroquinone derivative (CHP-HQ250). The fraction of this derivative isolated at pH 7 had the highest redox capacity of all the CHP-derivatives (1.8 mmol/g). The obtained results are indicative of high accepting properties of CHP and its NQ and anthraquinone derivatives, which are most suitable for suppressing methanogenic activity of microbial consortia. At the same time, fulvic acids and CHP-HQ possessed high donor capacity which is indicative of their substantial antioxidant properties. Our results show that directed modification is a promising tool for tailoring redox properties of HS.

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- 1. Perminova et al. // Environ. Sci. Technol. 2005. 39(21):8518-8524.
- 2. Matthiessen A. // Vom Vasser. 1995. 84:229.

The effect of humic substances on increasing the viability of hydrocarbon-oxidizing bacteria

<u>Nikolaev Yu.A.</u>¹, Demkina E.V.¹, Perminova I.V.², Loiko N.G.¹, Borzenkov I.A.¹, Ivanova A.E.¹, Konstantinov A.I.², El-Registan G.I.¹

¹Federal Research Center "Fundamentals of Biotechnology" of the Russian Academy of Sciences, Moscow, Russian Federation, NikolaevYA@mail.ru

²Lomonosov Moscow State University, Moscow, Russian Federation

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The aim of the work was to investigate the effect of humic substances (HS) on the viability of bacteria destructing hydrocarbons. It was shown that the introduction of HS (0.03–3.0 g/L) into the *Acinetobacter junii* culture of the stationary growth phase resulted to an increase in the number of viable cells (VC) up to 15 times compared to the control without HS during 4 months storage (Fig. 1A). The increase in the number of VC is associated both with an increase in the formation of persistent cells (P) [1], which mature into resting cells (RC) [2], and stress-protective (antioxidant [3]) activity of HS. The introduction of HS into the pre-stationary bacterial culture of *Rhodococcus erytrhropolis* led to an increase in the number of VC up to 11 times during storage for 1 month (Fig. 1B).



Fig. 1. A. Change in the number of viable cells of *A. junii* (% of control) during storage for 30 and 120 days in the control (without HS) and experimental variants with the addition of Humikom: 1 - control, 2 - 0.03, 3 - 0.15, 4 - 0.6, 5 - 1.5, 6 - 3.0 g/L.

B. The number of viable cells of *R. erytrhropolis* after 1 month of incubations with different concentrations of HS (g/L): 1 - 0.15; 2 - 0.5; 3 - 1.5, compared with the control without HS (100%). HS are: HS - Sakhalin, G7 - Humate+7, S - sapropelic, HI - Irkutsk, HT -

Technoexport, LH - Lignohumate, F - fulvic acids, PH - Powhumus.

The formation of persistent cells is increased by the most oxidized hydrophilic HS (increased content of alkyl, alkoxy, carboxyl and carbonyl groups), and the stress-protective activity of HS is determined by the contribution of less oxidized humates with higher antioxidant activity (increased content of aromatic carbon, both substituted and not substituted heteroatoms).

- 1. Balaban N.Q. et al. // Nature Rev. Microbiol. 2019. 17:441–448.
- 2. Loyko N.G. et al. // Microbiol. 2015. 84(5): 595–609.
- 3. Menshchikova E.B. et al. // LAP Lambert Academic Publishing. 2012. Saarbruchen. P. 495.

Bioluminescent monitoring of detoxification processes

<u>Rozhko T.V.</u>¹, Kudryasheva N.S.^{2,3}

¹Krasnoyarsk Medical Academy, Krasnoyarsk, Russia, gutniktv72@mail.ru
²Institute of Biophysics FRC KSC SB RAS, Krasnoyarsk, Russia
³Siberian Federal University, Krasnoyarsk, Russia

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Bioluminescence of marine bacteria is sensitive to toxic compounds. That is why the bacterial bioluminescent assay systems are used as biosensors in ecology and medicine.

Bioluminescent assays in vivo and in vitro were used to monitor detoxification of solutions of radionuclides of low activity by humic substances (HS).

Effects of low-level radiation on bioluminescent assay systems in vivo and in vitro were studied. Bioluminescent assay systems of different levels of organization were used: intact bacteria and bioluminescent enzyme reactions. Solutions of 241Am and Tritiated water (HTO) were used as sources of radiation.

Detoxification of the radioactive solutions by HS was under study. It was shown that HS change bioluminescence kinetics in 241Am solutions approaching it to control.

HTO and HS demonstrated synergetic activation effect on the bioluminescence enzymatic bioassay.

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Influence of humic compounds on bioluminescence of immobilized photobacteria

<u>Senko O.</u>^{1,2}, Stepanov N.^{1,2}, Efremenko E.^{1,2} ¹Lomonosov Moscow State University, Moscow, Russia, elena_efremenko@list.ru ²Emanuel Institute of Biochemical Physics, RAS, Moscow, Russia

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An analytical test based on the use of bioluminescent bacteria is characterized by a quick response and qualitative analysis. Additionally, it is possible to adapt existing portable devices for its use [1,2]. Particular attention is paid to the use of biosensitive elements in the form of immobilized bioluminescent bacterial cells [3,4]. It was previously shown for a biosensitive element developed on the basis of the bacterial *Photobacterium phosphoreum* cells immobilized in macroporous poly(vinyl alcohol) cryogel (PVA cryogel). It was found that such a biosensitive element can be successfully used for analyzing the toxicity of media containing heavy metal ions, phenol derivatives, and organophosphorous pesticides, both in a discrete and flow-through mode [3,4]. The PVA cryogel as a carrier positively influences the stability of the cell bioluminescent signal in the absence of toxins and does not affect the bioluminescent registration with the modern devices used in the experiments.

The use of immobilized bacterial cells of *P. phosphoreum* for primary screening of humic compounds (HCs), proposed for use to suppress gas evolution from landfills, from the point of view of their toxic effect on cells of microorganism, was proposed in this work.

It was shown that in the studied range of concentrations of HCs (0.1-10 g/I) there is a notable direct correlation between the concentration of HCs and the residual luminescence intensity of immobilized *P. phosphoreum* cells. Moreover, those HCs that had the maximum effect in the case of suppression of the methanogenesis process [5], maximally inhibited the luminescence of photobacteria

Thus, it can be argued that the biosensitive element in the form of immobilized *P. phosphoreum* cells can be used a primary screening tool for assessing the effect of HCs on cells of microorganisms.

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- 1. Michelini E. et al. // Anal. Bioanal. Chem. 2013. 405:6155–6163.
- 2. Roggo C. et al. // Curr. Opin. Biotechnol. 2017. 45:24–33.
- 3. Efremenko E.N. et al. // Luminescence. 2016. 31:1283-1289.
- 4. Efremenko E.N. et al. // Appl. Biochem. Microbiol. 2014. 50:477-482.
- 5. Stepanov N. et al. // Sustainability. 2019. 11:3158–11p.

The influence of old oil pollution on the composition of organic matter and the microbiological activity of peat soils in the permafrost zone

Solovyova A.A., Rozanova M.S.

Lomonosov Moscow State University, Moscow, Russia, solovyova.a.a@mail.ru

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Cryolithozone biogeocenosis are characterized by low biological turnover capacity, mosaic of soil and vegetation cover, poor species composition of phytocenoses and their low bioproductivity.

Samples of clean and oil-contaminated oligotrophic peat of undisturbed structure (monoliths) were taken on hilly peatlands of the northern taiga of Western Siberia, developing in permafrost conditions. Peat was maintained throughout the experiment at natural humidity, for which the monoliths were regularly weighed (once every three days) and, if necessary, sprayed with water. The thawing periods (temperature + 7°C) and freezing (-10°C) lasted 14 days, the duration of the experiment was 4 months.

To assess the effect of pollution on the composition of organic matter and the change in the microbial community, the following parameters were determined: the content and composition of labile humic substances (Clhs) (labile humic (Clha) and fulvic acids (Clfa), total carbon content (Ctot), water-extractable carbon content (Cw), basal respiration (BR), microbial biomass carbon (MB), petroleum content in samples (OiIP). Results are shown in Table 1.

				-	-	-							
	Depth, cm	Clhs, %	Clha, %	Clfa, %	Cha/ Cfa	Cw, %	Ctot, %	Cw/ Ctot,%	Cw/ Clhs,%	Clhs/ Ctot,%	BR, µgC C- CO₂/g×h	MB, µgC g	OilP, mg kg
Oil	0	12.18	3.42	8.77	0.39	1.46	42.57	3.44	12.02	28.62	12.90	1088.6	2247
Oil	15	18.02	7.08	10.94	0.74	1.29	44.38	2.90	7.13	40.61	23.76	1821.6	9581
Oil	20	18.00	6.58	11.42	0.58	0.96	50.45	1.91	5.36	35.68	17.57	1528.6	13300
Oil	25	21.56	7.58	13.98	0.54	0.80	50.32	1.58	3.69	42.84	6.60	503.1	25707
Oil	30	20.83	6.75	14.08	0.48	0.57	53.08	1.07	2.73	39.24	4.63	197.6	35240
Clear	0	15.45	2.90	12.54	0.26	1.67	43.09	3.88	10.83	35.84	109.69	8876.8	1161
Clear	15	21.70	6.76	14.94	0.45	1.12	44.85	2.50	5.17	48.38	69.21	5475.2	5064
Clear	23	21.69	7.19	14.49	0.50	0.89	50.73	1.76	4.12	42.75	11.52	1357.4	5940
Clear	30	17.42	6.22	11.20	0.54	0.65	51.79	1.25	3.71	33.64	5.43	260.3	6162
Clear	40	20.51	6.71	13.80	0.50	0.63	51.22	1.23	3.07	40.04	4.42	213.4	5785

Table 1. The content of oil products, various fractions of organic matter and indicators of microbiological activity of peat

The content of the Clhs Clear variant is relatively evenly distributed over the profile, decreasing in its lower part. In the Oil variant, an increase in the content of Clhs occurs at the bottom of the profile; this is probably due to the increased hydrophobicity of peat due to the content of Oil, and soluble fractions of organic substances accumulate in the lower part of the profile contaminated with oil products. Low biological activity in the upper horizons contributes to this, since bioavailable organic matter is not consumed by microorganisms.

Differences in the indicators of microbiological activity between the Clear and Oil variants are maximal in the surface layers of peat. In the upper layer of the BR in the Clear variant, it is almost 8 times higher than in the same peat layer in the Oil variant, the carbon content of the microbial biomass was 8900 μ gC/g in the Clear variant and 1090 μ g/g in the Oil variant.

ATP-measurements in estimation of effect possessing by humic compounds on various cells of microorganisms

Stepanov N., Senko O., Efremenko E.

Lomonosov Moscow State University, Moscow, Russia, elena_efremenko@gmail.com Emanuel Institute of biochemical physics RAS, Moscow, Russia

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The effectiveness of using the estimation of adenosine triphosphate (ATP) concentration in cells to control the cell viability of microorganisms in a wide variety of biocatalytic processes has been repeatedly demonstrated [1-4]. The ATP concentration can be quickly and easily determined using bioluminescence with great accuracy, even with an ultralow content of the substance in the sample (as low as $10^{-14} \div 10^{-12}$ M), using luciferin–luciferase reagents [4,5]. However, when using ATP-analysis to determine the energy status of cells in any new biocatalytic process, it is necessary to evaluate the possible effect of components of the culture media on the activity of the applied samples of luciferase, since it is known that some substances can inhibit this enzyme [5].

In a study of the use of humic compounds (HCs) for suppression of biocatalytic gas evolution from landfills, it was proposed to use the ATP-analysis to assess the viability of cells of anaerobic microorganisms involved in the process of gas formation [6]. For this, the inhibitory effect of HCs on the luciferase reagent was previously evaluated.

It was found that HCs in a concentration of more than 0.1 g /L in the cultural media have a notable inhibitory effect on luciferase, and the concentrations of HCs more than 5 g/L completely inhibit the luminescence of this enzyme.

Guided by these results, in order to adequately assess the concentration of intracellular ATP in cells exposed to presence of HCs, the analyzed samples of cultural media with HCs were diluted an appropriate number of times to avoid inhibition of luciferase within analysis. A positive correlation between the metabolic activity of cells in biogas formation and the concentration of ATP in cells was observed. So, the control of ATP concentration appeared to be an attractive and simple tool for finding compounds that can suppress methanogenesis in landfills.

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- 1. Efremenko E.N., Tatarinova N.Y. // Microbiology. 2007. 76(3):336–341.
- 2. Stepanov N., Efremenko E. // New Biotechnology. 2017. 34:54–58.
- 3. Efremenko E.N. et al. // Int. Biodeterioration Biodegradation. 2005. 56:94–100.
- 4. Ismayilov I.T. et al. // Moscow University Chemistry Bulletin. 2015. 70(4):197–201.
- 5. Lomakina G.Y. et al. // Biochemistry. 2015. 80:701–713.
- 6. Stepanov N. et al. // Sustainability. 2019. 11:3158–11p.

Humic product enhances the effect of a biological agent: combined application of *Trichoderma sp.* and Lignohumate® to suppress a pathogen of cabbage clubroot

Suada I.K.¹, <u>Yakimenko O.S.</u>², Stepanov A.L.², Schegolkova N.M.² ¹Faculty of Agriculture, Udayana University, Bali, Indonesia ²Soil Science Faculty, Lomonosov Moscow State University, Moscow, Russia, iakim@soil.msu.ru

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Possibility of humic products to enhance the effect of biological agents to suppress plant pathogens is often reported. Development of new formulations of environmentally friendly products based on both humics and biological products is highly relevant, especially for tropical regions, where plants are susceptible to numerous fungi diseases.

The objective of this study were: (1) to evaluate how effective is the combined application of *Trichoderma sp.* and humic product Lignohumate® (LH) to control cabbage clubroot, a disease caused by the soil-borne fungi *Plasmodiophora brassicae* and (2) to estimate the best ratios for a combined humic-biological formulation.

The field experiment was conducted in Bali (Indonesia) using cabbage (*Brassica oleracea* L.) susceptible to clubroot disease. *Trichoderma asperellum* isolated from cabbage plantation was applied in levels: 0 (control), $1x10^6$, $2x10^6$, and $3x10^6$ spores per plant. LH was used in concentrations 0, 0.01, 0.02, 0.05, 0.1, 0.5, 1.0, and 2% (w/v) to pour planted cabbage. Leaf area and chlorophyll content were measured after planting. After harvest the disease incidence, total amount of galls on roots, root and canopy dry weight, and microbial population (bacteria, fungi, and *Trichoderma*) were determined.

Results. Experimental field was heavily infected with clubroot: incidence of disease achieved 88% at the control treatment. When added separately, both tested products contributed to a decrease in the incidence of 1.3-1.8 times (up to 66 and 49%). Effect of *Trichoderma* alone was visible, but statistically insignificant, whether LH alone at highest doses (1 and 2%) was able to decrease the pathogen infection. At combined application of *Trichoderma* with LH at rates higher than 0.02% the positive effect was much more pronounced, and in some cases the incidence of disease decreased up to 42-58%. The combination of treatments that gave the best effect was the *T. asperellum* treatment on population of $3x10^6$ spores with LH-dose 0.5% which provided the least disease incidence (7.1%).

Besides of disease incidence, both products affected also the root galls formation, reducing their amount on cabbage roots in 2-3 times. LH enhanced the effect of *Trichoderma* and at the most effective treatment (*T. asperellum* population of $3x10^6$ CFU with 0.5% LH) the minimum amount of galls was observed: 1 against 19 in control. At LH rates 1% and 2% amount of galls on roots was higher than at 0.05%. The lowest root weight, indicating minimum level of root infection, was also observed for the same treatment. The highest content of chlorophyll in leaves and canopy dry yield was recorded for 0.5% concentration of LH. Application of LH also stimulated the microbial population in soil, and the concentration of 0.5% provided the highest increment of bacterial and fungi population.

Conclusion. Humic product LH demonstrated the capacity to enhance the effect of antagonistic fungi *Trichoderma* to suppress the pathogen of clubroot disease *Plasmodiophora brassicae*. The best combination of humic and biological products was $3x10^6$ spores of *T. asperellum* and 0.5% LH. The higher concentrations of LH did not provide further positive effects.

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Participation of micromycetes and bacteria in joint humification waste of wood breed

Sviridova O.V.¹, Vorobyov N.I.¹, <u>Puhalsky Y.V.^{1,2}</u>, Orlova O.V.¹, Popov A.A.¹, Mityukov A.S.⁴, Laktionov Yu.V.¹, Pishchik V.N.^{1,3}

¹All-Russian Research Institute for Agricultural Microbiology, Saint-Petersburg, Russian Federation, juan_diego@humic.edu

²Scientific Production Association «BioEcoTech LTD», Saint-Petersburg, Russian Federation ³Agrophysical Research Institute, Saint-Petersburg, Russian Federation

⁴Limnology Institute of Russian Academy of Sciences, Saint-Petersburg, Russian Federation

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The processes of microbiological humification of bark of coniferous trees at two levels of mineral fertilizers with using of biological preparation Barkon developed in the ARRIAM (Saint Petersburg, Russia) were studied. The dominated microorganisms in Barkon were mycromycetes *Penicillium chrysogenum* (Thom 1910) OH 4 RCAM 00741 and bacteria *Pseudomonas fluorescens* 7RCAM 00537. The composition of the biopreparation included micromycetes, cellulolytic and nitrogen-fixing bacteria, and lignin decomposers bacteria (Sviridova et al., 1993). The biochemical conversion processes in compost occurred with the close interaction of micromycetes from genus *Pennicilium* and *Pseudomonas fluorescens* bacteria in a self-organizing biosystem (Fig.1). As it was shown by factor analysis, the micromycetes in this biosystem mainly destructed of polymer organic molecules of lignocellulose, due to the presence in them of oxidoreductases (Vedenyapina et al. 2010). The absence of an optimal organization of humification processes was revealed without the use of a biopreparation indicates

The optimal technological conditions for composting wastes of coniferous wood using nitrogen fertilizers and biopreparation – Barkon have been identified. As a result of composting obtained an organic fertilizer. This fertilizer was tested on phytotoxicity. It was fixed that the fertilizers studied do not possess phytotoxic activity and inhibit the development of phytopathogenic micromycetes *Fusarium oxysporum*.



Figure 1. A graph of the maximum correlation coefficients between the biochemical characteristics of compost and the microbiological composition of the biosystem during the destruction of sawdust conifers trees. Note: Numbers correspond the designations: 1 – micromycetes (cells), 2 - proteolytic bacteria (cells), 3 - amylolytic bacteria (cells), 4 – lignin decomposers bacteria(cells), 5 - cellulolytic bacteria (cells), 6 - N_{total}, 7 - N-NO₃, 8 - C_{org}, 9 - level of humification of compost, 10 - C-CO₂, 11 – using/unusing of N (1/0), 12 – using/unusing of a biopreparation (1/0).

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Effects of lignohumate and biochar on microbial communities in agricultural soils differing in organic matter content

<u>Terekhova V.A.</u>^{1,2}, Fedoseeva E.³, Pukalchik M.A.⁴, Ivanova A.E.¹, Verkhovtseva N.V.¹, Pozdnyakov L.A.¹, Kulachkova S.A.¹, Gorlenko M.V.¹, Karpukhin M.M.¹, Yakimenko O.S.¹ Lomonosov Moscow State University, Moscow, Russia, vterekhova@gmail.com

²Severtsov Institute of Ecology and Evolution, Moscow, Russia

³Pirogov Russian National Research Medical University, Moscow, Russia

⁴Skolkovo Institute of Science and Technology, Moscow, Russia

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Structural and functional microbial characteristics are considered as sensitive indicators of soil health. The abundance and diversity of microorganisms determine high metabolic potential of soils. In different countries, a specific set of indices — determined by the structure and composition of microbial communities—is used to normalize soil quality and to assess environmental risk. However, stability and reliability of microbial indices in soils that differ in humus status have not been sufficiently studied.

The aim of this study was to compare the influence of lignohumate and biochar on microbiological properties of soils polluted by several metals, differing in organic matter content.

Responses of soil microorganisms (fungi and bacteria) on lignohumate (dose of 0.25%, total C content of 36%, Company "RET", Russia) and biochar (dose of 5%, total C content of 70-80%, Company "Metakom", Russia) were evaluated in four typical agricultural topsoils (0-20 cm) under controlled conditions using pots experiments. Functional and structural characteristics of microbial communities were analyzed by microbiological, chromatographic and biochemical procedures, including analysis of microbial lipid profiles (GC-MS method), PCR diagnostics, multi-substrate testing (MST).

The results showed that lignohumate and biochar (separately and in mixtures) affected both fungal and bacterial communities. The effect of organic amendments on the composition and structure of soil microbial communities was remarkably stronger than that of Cu, Pd, and Zn addition.

Lignohumate and biochar application resulted in higher microbial carbon biomass (Cmic), microbial quotient (qCO₂), soil enzyme and MST activities. Humus-rich soils were more markedly affected than humus-poor soils.

The change in fungal biodiversity was stronger than in bacterial biodiversity. Biochar alone affected more severely the structural and functional microbial characteristics than in mixture with lignohumate. The most remarkable effects were in metal-polluted, humuspoor soils.

Biochar and lignohumate resulted in higher soil pH and nutrient content.

The effect of humus content in natural samples of investigated soils was stronger than lignohumate and biochar addition and metal contamination on all parameters. This fact should be taken into account for the efficient use of nature-friendly technologies.

Our results highlight additional research need for a better understanding of the effect of humic substances and biochar on microbial communities of different soils and their further effects on ecosystem functions.

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Differential oxidation of phenolic acids by laccase in homogeneous and heterogeneous systems during humification reactions

Zavarzina A.¹, Demin V.¹, Belova O.², Lisov A.^{1,2} ¹Faculty of Soil Science, Moscow State University, Moscow, Russia, zavarzina@mail.ru ²Institute of Biochemistry and Physiology of Microorganisms, Pushchino, Moscow Region, Russia

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Oxidation and condensation of low molecular weight phenolic compounds (PC) in the presence of biocatalysts is considered an important pathway of humic substances (HS) formation and organic C accumulation in soils [1]. Recently we have found occurrence of phenolic acids in lichens: p-hydroxybenzoic acid (HDB), syringic acid (SYR) and vanillic acid (VAN) were most abundant, while protocatechuic acid (PCAT) and caffeic acid (CAF) occur in trace amounts [2]. We hypothesized that phenolic acids detected in lichens may serve as precursors of HS at the pre-vascular stage, however their oxidative reactivities require investigation. In this work we studied oxidation of phenolic acids by laccase of lichen *Solorina crocea* both in homogeneous and heterogeneous systems. We have prepared solutions of individual phenolic acids as well as mixtures of SYR and VAN acids with *p*-HDB acid (mixture 1), PCAT acid (mixture 2), gallic (GAL) acid (mixture 3) and CAF acid (mixture 4). Concentration of each acid in the mixture was 2 mM. Reactions were carried out in 20mM Na-acetate buffer (pH 4.5) within 24 hours with both free laccase and laccase immobilized on hydroxyaluminum-kaolinite. Phenolic acids were analyzed by RP-HPLC as described in [2].



Fig. 1. Oxidation of phenolic acids mixtures by laccase in homogeneous (A) and heterogeneous (B) systems.

We have found that oxidative reactivity of individual phenolic acids varied considerably and formed the order: GAL≥CAF>PCAT>SYR>>VAN. Hydroxybenzoic acid was not oxidized under the reaction conditions used. When present in mixtures, reactivity of OH-substituted phenolic acids formed the order CAF>GAL>PCAT (Fig 1A). Methoxy-substituted VAN acid was oxidized only in the presence of GAL and SYR acids (mixture 3), while SYR acid was actively oxidized only in the absence of GAL and CAF acids (mixtures 1 and 2). Transformation of PCAT, GAL and CAF acids by laccase considerably increased in the presence of the mineral phase (Fig 1B) which is important finding in relation to aromatic C accumulation in soils.

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- 1. Stevenson F.J. 1994. Humus chemistry: genesis, composition, reactions. J. Wiley and Sons, New York.
- 2. Zavarzina A.G. et al. // European J. Soil Sci. 2019. 1–15. doi: 10.1111/ejss.12822.

Section 4 Humic substances, animals and human beings

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Relative analysis of functional groups of humic acids of peloids

<u>Avvakumova N.P.</u>, Krivopalova M.N., Glubokova M.N., Katunina E.E., Zhdanova A.V. Samara State Medical University, Samara, Russia

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Nowadays there have been established a large number of positive effects of humic substances on living systems. An adaptogenic, regenerative, anti-toxic, immunotropic effect of humic acids of different origin has been detected in several studies. As natural environmentally friendly substances, humic acids of peloids (HAs) are characterized by non-specific universal pharmacological action. The probabilistic character of substance organization, its heterogeneity, heterofunctionality and polymolecularity create considerable obstacles for using humic peloid products in medical practice.

The purpose of this work is a comparative analysis of infrared and ultraviolet spectra of humic acids (HAs) of peloids and magnesium, calcium, zinc and silver (I) humicates, as well as a qualitative analysis of the products of HA hydrolysis using the method of high performance liquid chromatography (HPLC).

In order to justify the interpretation of the results of the spectral analysis and to identify individual components, an HA solution with 1.0 % (mass) concentration was exposed to acid hydrolysis. The products of hydrolysis were extracted with the help of chloroform, carbon tetrachloride, ethyl acetate and diethyl ether. Derived eluents were exposed to HPLC with the preliminary derivation using the liquid chromatograph Diotronic.

The products of acid hydrolysis of HAs contain a considerable amount of components, which differ slightly depending on the extractant. According to the time of sample emergence, the following compounds can be identified: caprylic, octanoic, pelargonic, vanillic, myristic, azelaic, palmitic, oleic acids, dehydroabietic acids, cyclohexanol, glycerol, 18-methyl-nonadecanol, hexadecanol, octadecanol, 2-octyldodecanol, tetracosane, heptacosane, cholesterol and squalane.

The infrared absorption spectrum of free humic acids of peloids has a profile typical of the class of compounds under study. The positions of intensity peaks of the absorption bands prove that the substance under study is an HA. In order to justify the assignment of bands, the spectra of metal humicates which have a similar look were obtained. A considerable increase of the band intensity within the frequency interval of 1623-1605 cm⁻¹ and the band transformation at 1709 cm⁻¹ into a small shoulder at the peak of a fluctuation band of a free group -COOH is observed in humicate spectra. The noted changes allow to state that the peaks within the frequency interval of 1623-1605 cm⁻¹ are mostly conditioned by fluctuations of the bonds of carboxylate ion. The spectra were analyzed by calculating the ratio of relative intensity of bandwidths at 1615 and 1710 cm⁻¹. The results of calculations showed the inverse ratio of optical density of the bands with the mentioned frequencies. Based on these calculations, a relative quantitative assessment of the level of free and ionized carboxylic group in the samples under study was made.

The obtained experimental results of spectral analysis and the component structure of the products of hydrolysis allow assessing the functional structure of HA peloids with more precision.

Humic substances of peloid as natural adaptogens

Avvakumova N.P.¹, Shurygina O.V.², Krivopalova M.A.¹, Rusakov D.A.², Zhernov Yu.V.³, <u>Glubokova M.N.¹</u>, Katunina E.E.¹

¹Samara State Medical University, Samara, Russia

²IDK Medical Company - MD Medical Group "Mother and Child", Moscow, Russia

³National Research Center – Institute of Immunology FMBA of Russia, Moscow, Russia

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Today the fact that the mobility of male germ cells is the key factor influencing delivery of fatherly genetic material to an ovum does not raise doubts. Kinetic characteristics define quality, viability of spermatozoids and its cryotolerance. Decrease of spermatozoids mobility can lead to an astenozoospermia and be the reason of male infertility. Correction of spermatogenesis is most often carried out by use of hormonal therapy which possesses side effect breaking physiological parameters of an organism. The low efficiency of therapy is caused by insufficiency of knowledge of the factors defining mobility of gametes. In this regard the great interest is attracted by drugs on the basis of peloid. Components of peloid have antioxidant, reparative, cytoprotective, proliferative effect. Maintaining constancy of structure of bio-systems, humic substances (HS) of peloid promote restoration of biological norm at disease states.

The aim of this work was a research of impact of HS of peloid on mobility of men's gametes.

The experiment was conducted on volunteers at the age of 25-30. Fractions of 0.1% solutions of humic substances of peloids were used: humic acids (HA), hymatomelanic acids (HMA), and initial HS extract (HFA). In the series of experiments a buffer solution FertiCult flushing (HEPES, 4 g/L HSA) (FertiPro, Belgium) and a specimen solution were added to the target volume of the purified ejaculation sample. A series of experiments with the different content of peloid drugs was carried out to determine the optimal concentration. Changes in sperm motility were observed after 10, 30, 60, 120 and 240 minutes of incubation at room temperature. Ejaculation analysis was carried out using Nikon Eclipse E200 microscope (Japan) and computer sperm analysis Sperm Class accordance Microptic SL (Barcelona, Spain) in Analvzer from with WHO recommendations. The results were evaluated using the statistical software package SPSS 12.0.2 of the same name company (or Statistica 6.0 of Statsoft). Based on the results of data allocation checking for compliance with the normal allocation, nonparametric methods of comparing the parameters in the test groups (Mann-Whitney criterion) were used. The data obtained by us prove adaptogenic properties of humic peloid drugs, which do not have a negative impact on the life of biotest. We have identified differences in the kinetic characteristics of the tests depending on the type of drug. The results of sperm motility measuring in samples with the addition of HS show that in 10 minutes of incubation there is an improvement in the kinetic properties of sperm.

Thus, the humic components of the peloids have the ability to increase human sperm motility in vitro. The intensity of the effect depends on the type of peloid drug, concentration, incubation time. The obtained results determine the perspective of studies of the effect of humic peloid drugs on male fertility function.

The effect of potassium humate on the body of animals in the form of a feed additive

Bailina G.E.¹, Ermagambet B.T.¹, Kassenova J.M.¹, Kuchar E.V.² ¹TOO Institute of Coal Chemistry and Technology, Nur-Sultan, Kazakhstan ²Kazakh Agro Technical University named after S. Seifullin

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Experiments on the use of humic preparations as a feed additive for farm animals were started in the 60s and are currently ongoing. Extensive experimental material has been accumulated, proving that the use of humates leads to an acceleration of animal growth, a decrease in the incidence and mortality rate, and an increase in their body's resistance to adverse environmental conditions. The consequence is an increase in animal productivity. Based on this, the purpose of scientific research is to develop a technology for producing a feed additive based on potassium humate. The test of a feed additive based on potassium humate was carried out on laboratory white outbred mice, with appropriate feeding and keeping conditions.

Groups of animals were selected on the basis of analogues. Before setting up the experiment, the animals were kept in quarantine for 7 days. The drug was administered on an empty stomach after a 12-hour fasting diet. Observation of animals was carried out during the entire period of the experiment.

Adult white mongrel mice with an average body weight of 23 g were used to determine the growth and development properties of the preparation. The study drug was administered using vodka. During the experiment, 3 groups of animals were formed (3 experiments and 1 control), 3 mice in each group. Animals received the study drug in dosages: group 1 and 2 (experience) received a 0.1% solution of the drug per 1 liter. The animals in the control group were given drinking water in similar volumes. The animals were constantly monitored, paid attention to behavior, physical activity.

The negative effect of liquid humates on the animal organism has not been identified. The animals of the experimental group, compared with the animals of the control group, were more active, mobile, well-fed food, hairline had a natural shine.

It was noted that the fluctuations in the gain in live weight between control animals and animals receiving liquid humates in the age group of animals from 1 to 3 years were 53–55 grams per day for different animals, and 154 grams per day for one to three months old day.

Thus, the test results showed that the introduction of liquid humates in the diet in addition to the main feed did not negatively affect the behavior and condition of the animals.

Fulvic acids separated from Suwannee River behavior in the aqueousxylene system: influence of ionic strength and interaction with proteins

Chernysheva M.G., Badun G.A.

Lomonosov Moscow State University, Moscow, Russia, chernysheva@radio.chem.msu.ru

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The behavior of compounds in the aqueous can be considered as a model of the interaction of the compounds with cellular membrane. The result of the interaction strongly depends on the conditions including pH, salts concentration, presence of other compounds that can form composites and etc.

In present work we studied the behavior of fulvic acids separated from Suwannee River (SRFA, IHSS standard) in the aqueous-xylene system. We used tritium labeled SRFA to determine its concentration in the bulk of organic phase and at the liquid-liquid interface by means of scintillation phase method [1]. Two important conditions that can significantly affect humic substances were considered. They are ionic strength of the aqueous phase and the presence of proteins that can be encapsulated by humic materials.

The experiment was carried out in 0.028 M phosphate buffer (pH 6.8) as well as in phosphate buffered saline (0.16 M, pH 7.3) to analyze the influence of ionic strength on SRFA behavior. It was found that the increase in the ionic strength doesn't change the distribution ratio, while results in the increase of the adsorption as much as one order of magnitude.

In the studying the interference of SRFA and globular proteins during the adsorption at the liquid-liquid interface and distribution in the aqueous-xylene system hen egg white lysozyme and human serum albumin were used as model proteins. In these experiments' tritium label was contained either in protein or in SRFA.

It was found that both proteins reduce adsorption of SRFA, while adsorption of albumin in presence of SRFA was close to the adsorption of free protein, and it depended on the SRFA concentration in the case of lysozyme: at low SRFA concentrations it was lower than for free protein and exceeds this value for high SRFA concentration. Moreover, when SRFA was mixed with lysozyme the formation of insoluble complexes was observed as it was shown previously for coal humic acids [2]. Using tritium labeled materials we have determined the composition of the complex for different concentrations of SRFA.

The details of SRFA interaction with lysozyme and albumin will be discussed in the presentation.

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- 1. Chernysheva M.G. et al. // Chemosphere. 2020. 238: 124646. doi:10.1016/j.chemosphere.2019.124646.
- 2. Chernysheva M.G., Badun G.A. // In: Funct. Nat. Org. Matter Chang. Environ., Springer Netherlands, Dordrecht, 2013:280–291. doi:10.1007/978-94-007-5634-2_12.

Novel polyphenolic ligand of BP-Cx family drugs: cell distribution and mechanism of action

<u>Fedoros E.I.</u>^{1,2}, Badun G.A.³, Grozdova I.D.³, Melik-Nubarov N.S.³, Perminova I.V.³, Baldueva I.A.², Danilova A.B.², Nehaeva T.L.², Ryakhovskiy A.A.⁴, Pigarev S.E.^{1,2}, Panchenko A.V.², Anisimov V.N.²

¹Nobel LTD, Saint-Petersburg, Russia, efedoros@scioco.com

²Petrov National Medical Research Center of Oncology, Saint Petersburg, Russia

³Lomonosov Moscow State University, Moscow, Russia

⁴CHEMDIV LTD, Moscow District, Khimki, Russia

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BP-Cx-1, novel polyphenolic ligand, is a water-soluble fraction of wood lignin and is the platform for a portfolio of innovative pharmacological products such as antineoplastic agent BP-C1, radiomitigator BP-C2 and geroprotective composition BP-C3 [1]. In our previous study, a number of polyphenolic components of BP-Cx-1 (flavonoids, sapogenins, phenathrenes etc.) were identified as the major carriers of biological activity of BP-Cx drug family. *In vitro* and *in silico* target screening yielded overlapping lists of target proteins: adenosine receptors A1, A2A; dopamine receptor DRD4; glucocorticoid receptor GR; serotonin receptor 5-HT1; prostaglandin receptors PGI2, EP2; muscarinic cholinergic receptor, GABAA receptor [2]. Most of them are involved in cancer and/or inflammation signaling pathways.

In the present study, the IC50 of BP-Cx-1 was measured by radioligand method and a range of IC50 values between 22.8 and 40.3 μ g/ml were obtained for A1, A2A, BZD, EP2 and IP (PGI2) receptors. IC50 for 5-HT1 and for GR were 3.0 μ g/ml and 12.6 μ g/ml, respectively, both being within the range of BP-Cx-1 concentrations detectable in *in vivo* models (see *Panchenko et al* abstract in this conference book).

Further, distribution of [3H] labelled BP-Cx-1 in murine fibroblasts NIH3T3 and MCF7/R carcinoma cells was studied by autoradiography method. [3H]-BP-Cx-1 (marked by silver grains formed under tritium β -irradiation) was mainly localized along the cell membrane, in the perinuclear region and in the nucleus, suggesting ability of BP-Cx-1 to penetrate cells and bind to membrane or cytosol receptors. It is hypothesized that [3H]-BP-Cx-1 detectable in the nucleus is part of an activated GR complex, known to be involved in regulation of transcription of genes responsible for the anti-inflammatory response.

It is reported, that glucocorticoids may act at the very first step of the immune response by modulating differentiation, maturation and function of dendritic cells (DCs) [3]. In our experiment, we observed similar effect of BP-Cx-1 on DCs: downregulation of expression of the lipid-presentation molecule CD1a, co-stimulatory molecules CD80, CD86 and CD 40, decreased production of pro-inflammatory cytokines IL-4 and TNFa and increased production of anti-inflammatory cytokine IL-10. Observed production of G-CSF and GM-CSF by DCs in response to treatment with BP-Cx-1 may explain the radiomitigating effect of BP-C2.

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- 1. Anisimov V.N. et al. // Oncotarget. 2017. 8(59):100951-100956.
- 2. Fedoros E.I. et al. // Oncotarget. 2018. 9(26):18578–18593.
- 3. Cain D.W., Cidlowski J.A. // Nature Rev. Immunology. 2017. 17(4):233–247.

Adsorption of ciprofloxacin and diclofenac using humics and silicacoated Fe₃O₄ nanoparticles

<u>Gorlov N.</u>¹, Privar Y.², Bondarenko L.¹, Gorbunova N.³, Bratskaya S.², Kydralieva K.^{1,3} ¹Moscow Aviation Institute, Moscow, Russia, kamila.kydralieva@gmail.com ²Institute of Chemistry, Far-Eastern Branch, Russian Academy of Science ³Institute of Applied Biochemistry and Machine-Building, Moscow, Russia

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The emerging pollutants such as non-steroidal anti-inflammatory drugs (diclofenac sodium, DCF) and antibiotics (ciprofloxacin, CIP) have aroused increasing concerns, and the efficient removal of pharmaceuticals from wastewater is becoming an urgent problem. An eco-friendly composites consisting of Fe_3O_4 nanoparticles modified by humic acids (HA) and/or 3-aminopropyl-triethoxysilane (APTES) have been fabricated via an in-situ/solgel method, characterized and used for CIP and DCF adsorptive removal. XRD, SEM, Mossbauer spectroscopy, zeta potential (DLS), N₂ adsorption-desorption measurements and elemental analysis were employed to characterize all synthesized composites: $Fe_3O_4/APTES$, $Fe_3O_4/APTES$, $Fe_3O_4/APTES/HA$. The adsorption kinetics, isotherms as well as various influence factors, e.g., pH, ionic speciation and strength on the CIP and DCF removal were systematically investigated. The CIP and DCF recovery was studied from 0.1 mM solutions at solid:liquid ratio 1:1000, pH 7.5, and the contact time 24 h. The equilibrium concentrations were determined spectrophotometrically.

 Fe_3O_4 conjugation with humic acids lead to increase specific surface from 117 to 142 m²/g for the Fe_3O_4 and the Fe_3O_4 -HA20 (20 wt% of HA), respectively. The sorbents under study are characterized by a high sorption capacity regarding to ciprofloxacin. The results obtained indicate that the adsorption of CIP by the Fe_3O_4 -HA20 is largely enhanced in comparison with the bare Fe_3O_4 .

High uptakes of diclofenac reaching 178 and 256 mg g⁻¹ for Fe₃O₄/APTES/HA and Fe₃O₄/APTES composites respectively proved that the obtained xerogels are potential candidates for removal of drugs from waters and wastewaters. The isotherm of DCF adsorption was described well by the Freundlich model.

The results showed that the presence of HA weakened the sorption capacity of Fe₃O₄/APTES, but decreased its toxicity towards *Paramecium caudatum* and *Sinapis alba*.

To maximize the removal degree for DCF, ultrasonic (US) treatment in the absence or presence of Fe₃O₄/APTES and HA was investigated. Two frequencies, three time and three pH conditions were tested (30 and 100 kHz; 5, 10, and 30 min and 3, 7, and 9, respectively). The adsorption of DCF has increased when Fe₃O₄/APTES in the presence HA was sonicated at 100 kHz, pH 3 (58%, respectively) compared with 30 kHz, pH 3 (32%, respectively) within 30 min. These facts suggested that the fabricated composites have great potential for CIP and DCF-contaminated water treatment.

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Study of structure – anti-HIV activity relationship of shilajit samples with a use of cell-based assays and solution state ¹³C NMR spectroscopy

Konstantinov A.I.¹, Zhernov Yu.V.², Perminova I.V.¹ ¹Lomonosov MSU, Department of Chemistry, Moscow, Russia, konstant@med.chem.msu.ru ²NRC Institute of Immunology FMBA of Russia, Moscow

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Shilajit that emerges as a resinous exudation from rocks of mountains is considered to be a complex product of biotransformation of plant residues [1]. Antiviral activity of shilajit resulting from its biological activity was repeatedly reported [1-2]. However, structural composition of its active components remains unknown. In this study we investigated inhibitory effect of dried water extractions of 11 raw shilajit samples originated from different geographic regions on HIV in cell-based assays. Structural analysis of the samples were conducted using quantitative solution state ¹³C NMR spectroscopy. The HIV-1 899A was used in this study to infect TZM-bl cells (NIH AIDS Reagent Program No. 8129, USA) obtained by genetic engineering of HeLa cells. EC₅₀ values of dried shilajit water extractions were calculated using the Reed and Muench method. To obtain structural composition data, recorded ¹³C NMR spectra of shilajit samples were divided into intervals corresponding to typical structural units of natural organic matter (Figure 1) and integrated.

A strong direct correlation (R=0.82; p-value=0.02) between EC_{50} values measured for shilajit samples and percentages of non-phenolic aromatic carbon was observed. Thus, aromatic structures were found to reduce antiviral activity of shilajit, and this was drastically different from the results obtained later for humic substances [3].

Antiviral activity of shilajit samples indicates that these substances hold significant promise as safe and efficacious antiviral drugs for treatment of HIV infection. Further studies of their antiviral activity and identification of antiviral structural components should include preliminary sample fractionation and separation of low-molecular aromatic compounds.





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- 1. Agarwal S.P. et al. // Phytother. Res. 2007. 21:401–405.
- 2. Cagno V. et al. // J. Ethnopharmacol. 2015. 166:129–134.
- 3. Zhernov Yu.V. et al. // New J. Chem. 2017. 41:212–224.

The influence of the feed material Reasil[®] Humic Health on the elimination rate of antibacterials from the body of broilers

Korsakov K.V., Vasiliev A.A., Kozlov S.V.

Saratov State Vavilov Agrarian University, Saratov, Russia, korsakov@lifeforce.pro

Keywords: broiler chickens, feeding, supplement, humic acids, Reasil[®] Humic Health, elimination, antibiotics, coccidiostats

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In the first experiment on the broiler chickens in 2019 at the Slavinskaya poultry farm in the Krasnodar region 4 buildings were involved. The building number 22 (38 788 heads) and building number 16 (40 040 heads) were two control groups. The building number 23 (38 800 heads) and building number 20 (45 310 heads) were experimental groups. The broilers in the experimental groups received Reasil[®] Humic Health feed material additionally as part of the premix at the dose 0.5 kg of the feed material per 1 ton of feed. Reasil[®] Humic Health is based on unmodified microporous humic acids from Leonardite. The conditions and therapeutical preventive treatment schemes for all chickens were the same.

At 18 days of age a control slaughter of chickens was carried out in the quantity of 10 heads from each group. The muscle tissue was investigated. The analysis by the Premi test system for the presence of a residual amount of antibiotics and coccidiostatics in the broiler meat of all control groups showed a positive result, and the analysis of the meat of the experimental group showed the negative result. A repeat test at 21 days of age showed the same results.

In the second experiment broilers of the COBB 500 cross were reared at the vivarium of Saratov State Vavilov Agrarian University. From 21 day of age, all chickens were given the drug "Florfenicol" orally for 7 days, in dosages according to the instructions for use. During the experiment chickens of the 1st, 2nd and 3d experimental groups in addition to the main diet were fed Reasil[®] Humic Health in the amount of 1.0, 1.5 and 2.0 g per 1 kg of feed respectively. Poultry slaughter was carried out 24 hours after the start of antibiotic drinking and after 24 hours, 72 hours, 7 days and 10 days after stopping the antibiotic drinking. As a result, it was found that the combined use of the antibiotic Florfenicol and Reasil[®] Humic Health in doses of 1.0-2.0 g / kg of feed does not prevent the accumulation of the antibiotic Florfenicol and Reasil[®] Humic Health in a dose of 1.0 g / kg accelerates the elimination of the antibacterial drug from broiler's body by 2 times, and in doses of 1.5 and 2.0 g / kg of feed – by 4 times after discontinuation of antimicrobial therapy.

The results of two experiments show that the feed material Reasil[®] Humic Health based on unmodified microporous humic acids from Leonardite accelerates the elimination of antibiotics and coccidiostatics from the muscle tissue of broiler chickens.

Humic substances potentiate inhibitory activity of sulbactam with respect to β -lactamase TEM-1

<u>Kravtsova D.</u>, Cherkasova T., Rubtsova M., Grigorenko V., Perminova I. Lomonosov Moscow State University, Department of Chemistry, Moscow, Russia, d.k.97@mail.ru

Keywords: antibiotic resistance, beta-lactams, synergy, inhibition, humic substances

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Antibiotic resistance became a global problem, while microorganisms developed several adaptive tools to escape the action of antibiotics. The most efficient tool, which is used by gram-negative bacteria, is enzymatic degradation of antibiotics. It is realized via release of beta-lactamases which are capable of degrading beta-lactam ring of the most powerful beta-lactam antibiotics. To overcome this problem, inhibitors of beta-lactamases are widely used in clinical practice. However, they are guickly loosing activity due to efficient molecular recognition developed by microorganisms. The aim of this work was to test humic substances (HS) as a new type of beta-lactamase inhibitors. The ideas behind were: firstly, HS are capable of interaction with a broad range of enzymes and this might reduce activity of beta-lactamases; secondly, molecular ensemble of HS has dynamic constitution which might hinder molecular recognition of HS or embedded antibiotics by bacteria. For achieving the goal of this study, we have synthesized CENTA, which is a chromogenic analogue of beta-lactam antibiotic cephalothine as described in [1]. We used CENTA as a chromogenic substrate for studying enzymatic activity of β-lactamase TEM-1 in the presence of sulbactam (clinically approved beta-lactam inhibitor) and humic materials used in this study. A set of humic materials used for testing was assembled from the samples differing substantially in hydrophobicity and functional group composition. It included coal hymatomelanic acid (CHM-Irk), coal humic acid (CHA-Irk), coal humic acid (CHP), hydroquinone derivative of CHP (CHP-HQ), and peat fulvic acid (PFA-HT).

The parameters of enzymatic kinetics of CENTA hydrolysis by the TEM-1 β -lactamase were determined. The values of KM_{eff} and V_{max} accounted for (89 ± 1) μ M and (1.9 ± 0.1) μ M/s, respectively. The inhibition constant (KI) of the enzymatic hydrolysis of CENTA by subactam accounted for (12.7 ± 2.5) μ M. A competitive type of inhibition was established. According to the data obtained, the most active was a mixture of subactam and CHA-Irk, which reduced initial hydrolysis rate by 42%. The presence of CHA-Irk enhanced activity of subactam two times.

The second place in potentiating inhibition efficiency of sulbactam was taken by CHP-HQ, whose presence inhibited the reaction by 39%. It was followed by CHA-Pow and CHM-Irk, which reduced the hydrolysis rate of CENTA by 31% and 30%, respectively. Fulvic acids (PFA-HT) had the lowest synergetic activity – the resulting inhibitory activity accounted for 23%, which is very close to the activity of pure sulbactam (21%).

The results obtained are indicative of synergetic action between humic substances and low molecular weight β -lactamase inhibitors. The value of the found synergetic effect was the highest for the most hydrophobic coal humic materials, and the lowest one – for the hydrophilic fulvic acids.

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References

1. Bebrone C. et al. // Antimicrob Agents Chemother. 2001. 45:1868–1871.

Actoprotector activity of humic acids low-mire wood-grass peat

Logvinova L.A.¹, Zykova M.V.¹, Gostyukhina A.A.², Zamoshchina T.A.^{1,2}, Tsupko A.V.¹, Belousov M.V.¹

¹Siberian State Medical University, Tomsk, Russia, ludmila_logvinova@mail.ru

²Siberian Federal science-clinical center of Federal microbiological agency, Tomsk, Russia

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Humic acids (HAs) positively affect many body systems: excretory, digestive, nervous. In a few studies, the effect of HAs on the activity of the cardiovascular system is proved [Zykova, 2017; Lasukova, 2018]. With pathology of the heart and blood vessels, which annually take the lives of about 17 million people, there is a decrease in exercise tolerance due to the development of hypoxia. Due to the high mortality from cardiovascular disease, it is important to investigate the actoprotective activity of HAs.

Materials and methods. HAs were isolated by alkaline extraction from sample of lowland wood-grass peats collected from «Tagan» deposit of Tomsk region [Zykova, 2013]. Physicochemical study of HAs carried out [Zykova, 2013]. Bioactivity of HAs was tested on 40 utbred male rats "Wistar" 250-300 g in autumn. The animals were divided on 4 groups of 10 animals each (Table 1). Solution of NaCl and HAs was administered intragastrically to experimental animals before 30 min swimming at the rate of 0.5 ml per 100 g of mass. As forced exercise used a forced swimming test [Pat. 2617206]. Upon completion of the five-day testing, all animals were withdrawn from the experiment by simultaneous decapitation under CO₂-anesthesia. Decapitated animals collected blood to obtain serum. To assess the level of glycolytic processes during exercise, the rat serum lactate content was determined [Brancaccio, 2010; Brooks, 1999]. The concentration of lactate in the serum of the experimental and intact groups was determined by the colorimetric method [Dolgova, 2012; Kamyshnikov, 2000].

Group of animals	Living	0.9 % NaCl	1 % HAs	Swimming test	
-	conditions	intragastricall		-	
Intact	daylight	-	-	-	
Control No 1	daylight	-	-	+	
Control No 2	daylight	+	-	+	
Experimental group	daylight	-	+	+	

Table 1. The distribution of animals in groups

Results and discussion. The use of the HAs solution and the presentation of physical exercise to rats in the form of a swimming test until they were completely tired during the 5 days of the experiment contributed to their performance (Table 2). Determining the degree of change in endurance showed an increase of 24-39 % by the last day of testing.

Table 2. The effect of the HAs on the performance of rats in a swimming test

Group of animals/Swimming days			Intact duration	ı (s)	
	1	2	3	4	5
Intact	Did not swim				
Control No 1	38	46	53	52	47
Control No 2	64	61	65	69	63
Experimental group	70	76	101	97	97

The use of HAs before the start of physical activity led to the maintenance of the level of lactate in the blood (Table 3) at the level of reference values.

Table 3. The effect of HAs on serum lactate levels

Group of animals	Intact	Control group No 1	Control group No 2	Experimental group
Lactate level (mmol/l)	3.7	4.5	3.1	3.5

Influence of different oxidants on toxicity level of aqueous humic substances assayed by chlorophyll fluorescence of *Scenedesmus quadricauda* (Turp.)

Mikhaylov G.S., Zimbovskaya M.M., Timofeev N.P., Matorin D.N., <u>Pankratov D.A.</u> Lomonosov Moscow State University, Moscow, Russia, pankratov@radio.chem.msu.ru

Keywords: ferrate, ozone, toxicity, water treatment

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Current list of water pollutants is long and includes heavy metals, organic compounds, microorganisms, etc. A common organic pollutant is humic substances (HSs), known surfactants, which creates a problem of higher metal content in water. Furthermore, HSs are polydisperse which leads to premature sorbent saturation and filter clogging, shortening their lifespan. In addition to that, humic acid salts are highly soluble in water, changing its organoleptic parameters and making it importable. Therefore, HSs removal from water is an important task for water quality improvement. An important part of water purification process intended to reduce organic compound content is its treatment with oxidizing agents.

The task of this work was to assay the process of potassium humate (concentration ranging from 0 to 250 mg I-1) treatment by two oxidizing agents of different nature. Oxygen-ozone mixture was a control oxidant as it is commonly used for water purification. Aqueous ferrate-ion was used as an alternative, formed by leaching the product of self-propagating high-temperature synthesis (SHS) between iron and potassium nitrate [1]. Compared to ozone water treatment, ferrate as an oxidizing agent offers many advantages. As such, ferrates have high redox potential. Also, their reduction results in iron (III) hydroxide formation suspension able to absorb remaining pollutants making it easier to remove them by filtration.

HSs content after the oxidation was monitored by spectrophotometry and permanganate index method. It was shown that the nature of oxidizing agent affects to reaction mechanism and HSs removal effectiveness from water. Comparative assessment of aqueous HSs toxicity towards *Scenedesmus quadricauda* (Turp.) algae before and after oxidation was carried out by chlorophyll fluorescence measurement after one day long exposure of algae to the HSs solutions. It was shown that in the high HSs concentration interval (above 100 mg I-1) the ozonized oxygen treatment decreases the toxicity level while ferrate-ion treatment increases the toxicity of HSs solution. In the concentration interval below 100 mg I-1 both oxidants decrease the toxicity level after treatment. It was also shown that oxidation using ferrate decreases Fv/Fm ratio which indicates the decrease in the number of active photosystems. This number shows that such conditions as exposition to ferrate oxidized HSs are extreme for algae and cause it to become dormant. Therefore, as shown in the two examples of oxidants, using oxidizing agents for water treatment requires further analysis of possible consequences of HSs harsh oxidation.

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References

1. Pankratov D.A. et al. // Patent #RU 2600346.

Assessment of pharmacokinetics and biological effects of ligninderived polyphenolic compositions BP-C3 and BP-C2 in in vivo models

Panchenko A.V.¹, Fedoros E.I.^{1,2}, Pigarev S.E.^{1,2}, Maydin M.A.¹, Gubareva E.A.¹, Bykov V.N.¹, Drachev I.S.¹, Anisimov V.N.¹, Chernysheva M.G.³, Badun G.A.³, Perminova I.V.³

¹Petrov National Medical Research Center of Oncology, Saint Petersburg, Russia, ando.pan@gmail.com

²Nobel, Ltd., Saint Petersburg, Russia

³Lomonosov Moscow State University, Moscow, Russia

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BP-Cx-1 is a water-soluble multicomponent composition of polyphenolic compounds derived from lignin-based enterosorbent - Polyphepan® treated in the alkaline medium at the elevated pressure and temperature with continuous supply of oxygen [1]. BP-Cx-1 is used as base for a number of compositions with different activities. BP-C3 is a formulation, which comprises BP-Cx-1 with iron complex, selenium, ascorbic acid and retinol. Longterm treatment with BP-C3 had anticarcinogenic and geroprotective activity in female SHR mice [2]. BP-C3 was found to reduce the toxic effect of 5-fluorouracil against hematopoiesis and intestinal epithelium which was evident by preserved organ/body ratios for the lymphopoietic organs, anemia reduction and intestinal crypt survival [3]. BP-C2 is a composition which comprises BP-Cx-1 with ammonium molybdate, developed as radioprotector/radiomitigator [4]. BP-C2 was tested in two total body gamma-irradiation models (CBA and C57BL/6 mice) and exhibited a radiomitigating effect in the midlethal range of radiation doses. BP-C2 improved animal survival, protected intestinal crypts and Lgr5+ stem cells and enhanced extramedullar hematopoiesis in the spleen. Neither BP-C2 nor BP-C3 administered to intact animals has effects on blood count or biochemistry, body weight or animal behavior. For pharmacokinetics, the labeled [3H]-BP-Cx-1 was administered intravenously or by gavage once to BALB/c mice. In case of i.v. administration the primary organs of accumulation were the lungs, liver, kidneys and spleen (50-5000 µg/g tissue). BP-Cx-1 has low oral absolute bioavailability, its concentration in animal organs is several orders of magnitude lower than when administered i.v. (2-18 µg/g tissue). The present study demonstrated the feasibility of using a lignin derived polyphenolic compositions to diminish the toxic effects of chemotherapy and radiation injury. Both studied compositions provide significant protection effect against intestinal injury caused by chemical (BP-C3) or radiation (BP-C2) exposure. The extensive therapeutic potential of lignin derived polyphenolic compositions is yet to be explored.

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- 1. Fedoros E.I. et al. // Oncotarget. 2018. 9(26):18578-18593.
- 2. Anisimov V.N. et al. // Aging. 2016. 8(9):1866–1875.
- 3. Panchenko A.V. et al. // Exp Ther Med. 2018. 15(3):3124–3132.
- 4. Bykov V.N. et al. // Int J Radiat Biol. 2018. 94(2):114–123.
BP-C2 (complexes of molybdenum with polymer of benzene polycarboxylic acids derived from lignin): Evaluation of antigenotoxic potential in COMET assay

<u>Pigarev S.E.</u>^{1,2}, Trashkov A.P.³, Anisimov V.N.², Fedoros E.I.^{1,2}, Panchenko A.V.² ¹Nobel LTD, Saint-Petersburg, Russia, spigarev@scioco.com

²Petrov National Medical Research Center of Oncology, Saint Petersburg, Russia

³Petersburg Nuclear Physics Institute named by B.P. Konstantinov of NRC «Kurchatov Institute», Gatchina, Russia

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BP-C2 is a drug candidate developed as a radiomitigator for civic applications and as a medical countermeasure for radiation emergencies [1]. In rodents exposed to gammaradiation BP-C2 has demonstrated radiomitigative potential believed to be mediated through hematopoietic and intestinal progenitor stem cells [2].

Objective of the study presented here was to evaluate antigenotoxic potential of BP-C2 using in-vivo alkaline COMET assay. 35 male albino rats (*Rattus norvegicus*) were randomized to one of the 5 experimental groups (n=7). 0.9% sodium chloride was used as the negative and methyl methanesulfonate (MMS) as the positive control. MMS (40 mg/kg) or NaCl 0.9% were administered intraperitoneally once. BP-C2 was administered at 20 mg/kg, 200 mg/kg or 2000 mg/kg by gavage once 1 hour before administration of MMS.

Effects of MMS and BP-C2 were assessed by estimating the percentage of DNA in comet tail in three cell subpopulations: hepatocytes, red bone marrow cells and peripheral blood leucocytes (Table 1).

Target	Time	NaCI 0.9% (Negative control)	MMS (Positive control)	MMS+BPC2 20mg/kg	MMS+BPC2 100mg/kg	MMS+BPC2 2000mg/kg
Hepatocytes	1 hr.	5.5±0.8	14.2±3.6 ¹	11.1 ± 1.5^{1}	9.8±1.0 ^{1,2}	7.9±1.3 ²
	6 hrs.	6.8±0.8	24.9±4.0 ¹	18.1 ± 3.2^{1}	9.0±0.1 ^{1,2}	8.1±1.7 ²
	18 hrs.	5.6±0.7	31.3±5.2 ¹	17.9±4.0 ^{1,2}	10.2±1.4 ^{1,2}	9.0 ± 3.0^2
Leucocytes	1 hr.	3.8±0.7	8.8±1.0 ¹	8.0±1.0 ¹	4.6±1.4 ²	5.0 ± 0.8^2
	6 hrs.	3.1±0.6	13.5±1.0 ¹	15.5 ± 2.0^{1}	3.8±0.6 ²	9.7±2.0 ¹
	18 hrs.	4.0±0.6	17.2±2.8 ¹	13.5 ± 2.0^{1}	5.1±2.3 ²	10.3±1.1 ^{1,2}
Red bone marrow cells	1 hr.	11.9±2.4	23.3±3.4 ¹	19.1±3.4 ¹	18.8±6.6 ¹	19.3±2.0 ¹
	6 hrs.	9.8±1.4	33.1 ± 2.5^{1}	25.1±3.3 ^{1,2}	19.5±3.2 ^{1,2}	21.0±2.9 ^{1,2}
	18 hrs.	8.1±1.5	30.7 ± 1.9^{1}	20.4±2.5 ^{1,2}	$13.5 \pm 1.4^{1,2}$	10.1 ± 1.2^2
1		. 2				

Table 1. Amount of DNA in the comet tail, %

 $^{1} - p < 0.05$ vs Control; $^{2} - p < 0.05$ vs MMS

In animals treated with MMS, at the lowest tested dose (20 mg/kg) BP-C2 significantly reduces percentage of DNA in comet tail in all three cell types, though not reversing it to the level observed in the control group. At the higher doses (100 or 2000 mg/kg) BP-C2 almost completely reverses DNA damage in all three cell types.

References

1. Anisimov V.N. et al. // Oncotarget. 2017. 8(59):100951–100956.

2. Bykov V.N. et al. // Int J Radiat Biol. 2018. 94(2):114–123.

Influence of feed additives of humic nature on the formation of nonspecific adaptive response of the rats' hemostasis under the influence of stressors

Stepchenko L., Diachenko L.

Dnipro state agrarian and economic university, Dnipro, Ukraine, stepchenko2@gmail.com, linadyach@ukr.net

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The promising area for today is the searching for natural substances that can reduce the negative impact of hypoxia and oxidative stress on the body and thus can increase nonspecific adaptive resistance and affect the lability of the hemostasis system. It is known, that humic substances do not have a toxic effect that can increase the level of resistance of the animal body [1–3], but their effect on the system of hemostasis under the conditions of stress factors of different genesis has not been studied. Therefore, the aim of the study was to investigate the effect of feed additives of humic nature (FAHN) Humilid and Eco-Impulse Animal, which were obtained in different ways from the same peat on the state of the system of hemostasis of rats against the background of stress factors.

The studies were performed on white sexually mature young male rats weighing 180-200 g based on vivarium. The animals were divided into 5 groups of 8 animals each. Group 1 (control) - intact animals. Rats 2, 3, 4 and 5 groups modeled combined stress. The model of water-immobilization stress in combination with elements of emotional stress (lengthening of daylight) was taken as a basis, due to which the effect of combined stress was achieved. The comparative analysis of the effect of the FAHN Humilid (acid-alkaline hydrolysis) and Eco-Impulse Animal (alkaline hydrolysis of peat using electric shock) on the overall parameters of the hemostasis system. Results were statistically analyzed using the onefactor dispersion analysis ANOVA. For all statistical calculations, the significance was considered as the value of P < 0.05. It has been established that due to waterimmobilization combined stress in rats, a significant prolongation of prothrombinase complex formation time, prothrombin activation time, and an increase in fibrinogen content compared to intact animals. In rats that received the FAHN Humilid after a stressful period, normalization of the formation time of the prothrombinase complex was observed, both externally and internally. In addition, in these rats, the time of prothrombin activation and the amount of fibrinogen did not differ from the control values. In turn, rats treated with Eco-Impulse Animal FAHN showed stabilization of prothrombinase complex formation externally, whereas indicators characterizing the intrinsic pathway of prothrombinase formation, prothrombin activation time, and fibrinogen count did not differ from that of animals who did not receive any feed additives. Therefore, humic substances in the composition of feed additives Humidil can act as activators of faster formation of nonspecific adaptation of the hemostasis system to the action of stressors. The Eco-Impulse Animal, obtained by the specific method, exerts its influence only at the first stage of coagulation hemostasis. It is possible that upon receipt of this FAHN, substances that have not been used in the processes of plasma factor synthesis or that have an inhibitory effect on the course of hemostasis reactions may have been formed.

- 1. Stepchenko L.M. Biologically active substances humic nature as a regulator of homeostasis of poultry. Collection of conference materials. 2011. 1:164–166.
- 2. Diachenko L.M., Stepchenko L.M. // Theor. Appl. Veter. Med. 2018. 6(3):34-38.
- 3. Paronik V.A. et al. // Animal Biology. 2015. 17(4):109–114.

Investigation of the antioxidant activity of the humic substances and their quinone derivatives before and after reduction by NaBH₄

Volikov A.B., Perminova I.V.

Lomonosov Moscow State University, Russia, ab.volikov@gmail.com

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Humic substances exhibit antioxidant activity due to phenolic and other groups in their structure. At the same time, some of the groups in their structure are in the oxidized form, for example, quinones, converting them to the reduced form, it is possible to increase the antioxidant activity of humic substances. Based on the difference in antioxidant activity, amount of oxidized groups in the structure can be estimated.

Two humics samples were used in this work: humic substances of leonardite - HS (Humintech, Germany) and fulvic acids of groundwater - FA (Humintech, Germany). To study the more oxidized form of humic substances, quinone derivatives of hydroquinone (HS-HQ, FA-HQ) and pyrocatechol (HA-PQ, FA-PQ) were obtained by Fenton reagent (Fe²⁺, H₂O₂) treatment in alkaline condition. The reduced form of humics was obtained by NaBH₄ treatment; for this, 5 ml of a 5 g/l solution of NaBH₄ was added to 5 ml of a 200 mg/l solution of humics sample, kept for 24 hours, after which excess NaBH₄ was removed by adding 1M HCl to pH=7. Determination of antioxidant activity was carried out using the ABTS radical according to the widely used method, Trolox was used as a standard, the measurement results of Trolox equivalent antioxidant capacity (TEAC) are shown in the figure below.



Figure 1. TEAC of humic substances and their quinone derivatives before NaBH₄ treatment (white columns) and after (gray columns). (P=0.95, n=3).

As can be seen from the results, TEAC of HS sample are noticeably lower than FA, which can be explained by the large contribution of phenolic groups to the structure of fulvic acids. But after NaBH₄ treatment the value of TEAC is compared amounts to 3 μ mol/mg, which is caused by the transition of quinone groups to hydroquinone form in HS. Quinone derivatives show an average TEAC activity – about 2 μ mol/mg, and their reduction by NaBH₄ leads to an increase TEAC in HS derivatives and a decrease in FA derivatives, which may be associated with partial destruction of individual groups in relatively small fulvic acid molecules. If we pay attention to the difference between the initial and reduced forms, the initial sample of humic substances - HS, as well as their pyrocatechol derivatives are of the greatest values, which indicates the greatest contribution of oxidized fragments to their structure.

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Modification of humic substances of coal by ferrocene

Zimbovskaya M.M., Reshetova M.D., Konstantinov A.I., Shatalova T.B., Farat O.K., Pankratov D.A.

Lomonosov Moscow State University, Moscow, Russia, pankratov@radio.chem.msu.ru

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The high reactivity of humic substances (HS) is caused by the presence of a large number of functional groups in the composition of their macromolecules: primarily carboxyl, hydroxyl, carbonyl, nitrogen and sulfur containing groups. Functional groups of HS are inclined towards selective coordination, especially in relation to transition metals. Several methods are known for the synthesis of preparations of HS and iron: by the interaction of aqueous solutions of HS and iron salts; by means of mechanical dispersion (dry grinding of HS powder and magnetite in a ball mill); by chemical coprecipitation in the presence of HS (the Massart method), and by the interaction of metallic iron with aqueous HS solutions by oxidative in situ synthesis [1]. However, there is no direct evidence of the existence in these compounds of coordination bonds between iron ions and functional groups of HS. Perhaps this is due to the limited number of accessible functional groups of HS capable of forming strong covalent bonds with iron ions, as well as steric hindrances. It is obvious that the modification of HS using ligands that are prone to the formation of coordination bonds with iron will make it possible to obtain HS compounds with a high content of fixed iron. This may be of practical interest, for example, for agrochemical purposes. Such a modification of the HS is possible as a result of the esterification of the cyclopentadienyl derivative with the hydroxyl groups of the HS. It is known that C_5H_6 easily forms cyclopentadienyl π -complexes with iron - as ferrocene.

Ferrocene has great sustainability, chemical and thermal stability, lability in redox processes, has non-toxic effects. Ferrocene can be used as initial compound for number of derivatives with various substituents. Therefore, ferrocene carboxylic acid chloride was used as the initial reagent for the modification of HS. The initial ferrocene was acylated with acetic anhydride for obtain acetylferrocene. Then ferrocene carboxylic acid chloride was obtained by oxidation of acetylferrocene. The ferrocene carboxylic acid chloride was obtained at interaction of ferrocene carboxylic acid with thionyl chloride.

Modification of HS of coal by hydroxyl groups with ferrocene carboxylic acid chloride was carried out with benzoyl chloride under the conditions of inverse phase-transfer catalysis [2]. The product synthesized in the work is insoluble in water, but it is well soluble in some organic solvents. HS modified by ferrocene obtained as a result of described operations was characterized by such physico-chemical methods as: Mössbauer and IR spectroscopy, DTA, TGA with analysis of evolved gases and size-exclusion chromatography. It has been shown that the physico-chemical properties of the resulting HS modified by ferrocene contain characteristic features of both HS and ferrocene. Average molecular weight of HS after modification has decreased by half. Iron in the preparation is in three coordination positions, and 7% of iron (II) in one of them. In the samples is not confirmed present initial ferrocene or it derivatives used in the synthesis. Thus, the work carried out the modification of the HS by ferrocene and showed that the obtained compound has unique properties that differ from the initial HS.

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References

1. Pankratov D.A., Anuchina M.M. // Materials Chemistry and Physics. 2019. 231:216–224.

2. Khil'ko S.L. et al. // Solid Fuel Chemistry. 2015. 49(4):206–212.

Section 5 Humic raw material, humic products and their application

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Comparative effects of emamectin-benzoate and vermicompost tea on fall armyworm

<u>Adejumo G.D.</u>¹, Adesanwo O.O.¹, Soyelu O.J.²

¹Department of Soil Science and Land Resources Management, Faculty of Agriculture, Obafemi Awolowo University, Ile-Ife, Nigeria, gbengadaniel289@gmail.com

²Department of Crop Production and Protection, Faculty of Agriculture, Obafemi Awolowo University, Ile-Ife, Nigeria

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The rate of application of emamacin–benzoate to control fall armyworm (Spodoptera frugiperda) – an invasive species on maize plant is increasingly alarming in Nigeria and little or no attention is paid to its residual effect on soil properties and soil fauna. This study was conducted to test the insecticidal effect of vermicompost tea as a viable and sustainable option to emammacin-benzoate on fall armyworm. A comparative study was carried out to test the effectiveness of vermicompost tea over emamectin-benzoate. Vermicompost tea was produced from the mixture of organic materials, characterized and screened in the laboratory to determine the lethal concentration and time. The vermicompost and emamectin-benzoate were subjected to spectroscopic analysis to identify their functional groups using Fourier Transform Infra-Red (FTIR). Elemental composition of the vermicompost was determined using ICP. Data obtained were subjected to analysis of variance and mean values was separated using Fischer's LSD0.05. The FTIR result of vermicompost and emamectin-benzoate revealed similar functional groups like amides and haloalkanes which could be responsible for their insecticidal efficacy against fall armyworm. The insecticidal effect of vermicompost tea at 100 % concentration against fall armyworm was not significantly different from effect of emamectin-benzoate on maize plant tissues. Vermicompost tea applied at all levels effectively reduced number of damaged leaves and improved maize yield, similar to the effect of emamectin-benzoate. No significant effect of vermicompost tea and emamectinbenzoate was observed on soil fauna and soil properties except soil pH. However, vermicompost tea should be subjected to more instrumental analyses to establish its structure.

Platinum-containing nanocomposites based on humic substances from therapeutic mud

<u>Aleksandrova G.P.</u>¹, Lesnichaya M.V.¹, Dolmaa G.², Sukhov B.G.¹, Regdel D.², Trofimov B.A.¹

¹Favorsky Irkutsk Institute of Chemistry of SB RAS, Irkutsk, Russia, alexa@irioch.irk.ru ²Institute of Chemistry and Chemical Technology of Mongolian AS, Ulaan-Baatar, Mongolia

Keywords: humic substances, nanoparticles, platinum, therapeutic mud

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The search for new functional materials is one of the defining characteristics of modern science and technology. Metal nanoparticles, in particular, platinum nanoparticles can possess a wide range of properties that can be used for many practical applications. Platinum has high corrosion resistance and numerous catalytic applications including automotive catalytic converters and petrochemical cracking catalysts.

Previously, first time we studied characteristic of water-soluble silver and gold containing nanocomposites with a variable amount of the metal component based on reducing and stabilizing matrix of humic substances isolated from various deposits in Mongolia [1-2]. The humic substances of therapeutic mud have the greatest reducing and stabilizing ability among the investigated objects [3-4].

Due to potential technological interests of platinum nanoparticles, the synthesis and study of nanoparticles was a very active field of research during last years. In particularly, these studies of nanostructured materials show a strong dependence of their properties on size and shape. For example, the size effect on the catalytical efficiency is known, and the perspective effect on catalysis by the shapes of metal nanoparticles is anticipated and under investigation.

In the present, we report on the result of synthesized Pt-containing nanocomposites based on humic substances. The composition and properties of humic substances isolated from therapeutic mud in Mongolian deposits are established. The methodology of obtaining platinum-containing nanobiocomposites using a matrix of humic substances is described and their composition and structure are characterized by a complex of modern physicochemical methods, such as X-ray energy dispersive spectral microanalysis, infrared spectroscopy, ultraviolet spectroscopy, X-ray diffraction analysis, scanning electron microscopy.

- 1. Lesnichaya M.V. et al. // Doklady Chemistry. 2015. 460(1):13-16.
- 2. Aleksandrova G.P. et al. // Russian Chemical Bulletin. 2017. 66(1):143-149.
- 3. Khutsishvili S.S. et al. // J. Thermal Anal. Calorimetry. 2019. 137(4):1181–1188.
- 4. Dolmaa G., Aleksandrova G.P. Biological activity of humic substances and their nanocomposites. Saarbrücken, 2017.

Increase of efficiency of liquid humic fertilizers based on the use of combined feedstock

Arynov K.T., Auyeshov A.P., Faskhutdinov M.F., <u>Zhilkibayev O.T.</u> AspanTau LTD LLP, 255 Auyezov Street, Kalkaman-2, 050006 Almaty, Kazakhstan, tau_aspan@mail.ru

Keywords: brown coal, humic fertilizers, peat, vermicompost, extraction.

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Liquid fertilizers produced from peat and vermicompost become more common in agriculture, especially in organic farming. As far as it is known, extracts of peat and vermicompost contain a complex of bioactive substances: vitamins, phytohormones, amino acids, humic acids and so on acting as important nutrient and growth-stimulating substances used for treatment of agricultural crops. The use of these components in liquid fertilizer improves efficiency.

However, liquid fertilizers have a number of disadvantages, the most significant of which is a high content of ballast substance – water. Long-distance transportation of liquid fertilizer increases transportation costs and the need in increase of concentration of liquid fertilizers or, in other words, reduction of product cost per hectare of land is crucial for research works for countries with vast areas such as Russian Federation and Republic of Kazakhstan.

We are working on production of new liquid humic fertilizers using combined feedstock such as brown coal, peat and vermicompost. We have produced samples of liquid fertilizers with an increased content of humic acids (25 times) preserving useful properties of peat and vermicompost extracts. This has been achieved by mixing two different extracts of brown coal one of which is rich with salts of humic acid and the second one as the source of a number of bioactive substances.

The produced samples were studied using a screening electron microscope JSM-6490LV, systems of energy-disperse microanalysis INCA Energy and structural analysis HKL-Basic. It was shown that the samples contain useful elements such as K, Ca, Mg and Fe. IR Fourier spectra of samples were made using spectrometer Shimadzu IR Prestige-21 with attenuated total reflection Miracle by Pike Technologies.

Experimental researching of new products are still in progress.

The effect of humic acids adsorption on pH-dependent surface charging and aggregation of Fe_3O_4 -APTES nanoparticles

Bondarenko L.¹, Illes I.², Tombácz E.², Magomedov I.¹, Rabinskiy L.¹, Kydralieva K.¹ ¹Moscow Aviation Institute, Moscow, Russia, I.s.bondarenko92@gmail.com ²University of Szeged, Szeged, Hungary

Keywords: magnetite, 3-aminopropyl-triethoxysilane, humic acids, isoelectric point, zeta-potential

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Humic acids (HA) are often used as a stabilizer for magnetite nanoparticles (Fe₃O₄) and as a functional component for magnetic nanocomposites. Despite the protective shell of HA the oxidative degradation of Fe₃O₄ to maghemite Fe₂O₃ nanoparticles is observed. This led to a change in the biological activity of the composite (*Terekhova et al., 2018*). To improve chemical stability of Fe₃O₄ the surface charge control was used through pretreatment using silanes. In this study, effect of HA adsorption on pH-dependent surface charging and aggregation of the Fe₃O₄ functionalized by 3-aminopropyl-triethoxysilane (APTES) was studied. A set of the Fe₃O₄-APTES samples were formulated: under argon (Ar) or air atmosphere and/or further dialysis (D) and subsequent treatment of the samples: in harsh (US-treatment) and mild (magnetic stirrer, M) conditions. As a total, five samples were obtained: Fe₃O₄, Fe₃O₄-APTES (Ar), Fe₃O₄-APTES (Ar-D-M), Fe₃O₄-APTES (Ar-D-US), and Fe₃O₄-APTES (air).

Magnitude of isoelectric points (iep, the pH of $\xi = 0$), zeta-potential and hydrodynamic size were measured at 25 ± 0.1 °C in a disposable zeta cell (DTS 1060) of NanoZS (Malvern, UK) apparatus [2]. The range of pH was between ~3 and ~10. According to the magnitude of the shift of the pH-dependent zeta potential functions due to the sorption of increasing amount of HA in case of all samples, it is possible to formulate a series of samples in which the sorption capacity decreases: Fe₃O₄-APTES (Ar-D-M)≥ Fe₃O₄-APTES(air) > Fe₃O₄≥Fe₃O₄-APTES(Ar-D-US) >> Fe₃O₄-APTES (Ar).

Treatment way has a great effect on the sorption capacity of the samples. Dialysis of the sample Fe₃O₄-APTES (Ar) led to enhance adsorbing of HA. The sample exposed to the magnetic stirrer has a larger sorption capacity than the sample after the US-treatment: Fe₃O₄-APTES (Ar-D-M)> Fe₃O₄-APTES (Ar-D-US) >> Fe₃O₄-APTES (Ar). It is likely that harsh conditions during sonication (extreme high temperature and pressure in cavities forming at interfaces) led to destroy Fe₃O₄-APTES-HA bonds and reduce the sorption capacity.

The hydrodynamic diameter increases near the isoelectric point for the samples Fe_3O_4 -APTES without HA. The hydrodynamic size for the samples Fe_3O_4 -APTES at pH < 5 and pH> 9 varied from 700 nm to 300 nm. In the range 5<pH<9, the hydrodynamic size is about 100-300 nm. But, the hydrodynamic diameter for Fe_3O_4 -APTES the increases near the isoelectric point (from 200 to 750 nm), when not fully coverage with HA. In case of full coverage with HA, the hydrodynamic size remains unchanged at all pH range (around 150 nm for all of samples).

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- 1. Terekhova V. et al. // In: The 19th International Conference of International Humic Substances Society, 16-21 September 2018, Bulgaria. P. 205.
- 2. Tombácz E. et al. // Colloids and Surfaces A: Physicochemical and Engineering Aspects. 2013. 435:91–96.

Humic acids impact on bioactivity and aging of iron ions or nanoparticles

Bondarenko L.¹, Uchanov P.², Terekhova V.³, Kydralieva K.^{1,3} ¹Moscow Aviation Institute, Moscow, Russia, e-mail I.s.bondarenko92@gmail.com ²Institute of Ecology and Evolution RAS, Moscow, Russia ³Moscow State University, Moscow, Russia

Keywords: iron ions, magnetite nanoparticles, humic acids, aging, toxicity

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At present the question still remains whether the toxicity of iron oxide nanoparticles (NPs) from the released ions or NPs itself arises and how humic acids (HA) can influence on toxicity of NPs. In this study effect of humic acids, an natural organic polyligand that easily coordinate to iron (II, III) ions and graft to Fe_3O_4 NPs surfaces, on behavior of Fe_3O_4 -NPs were investigated in the aspects of their toxicity using standard test systems with higher plants *Sinapis alba* and invertebrates *Paramecium caudatum*. The Fe(II, III)-humic complexes were prepared by reaction of weighed amounts of the HA and the iron source with HA:Fe ratio of 1:0.150 (w/w). Colloidal Fe_3O_4 -NPs were synthesized by a chemical precipitation method *in situ* when the magnetic particles are grown within the HA matrix that provides steric stabilization of the colloidal system.

Several treatments of the test cultures were made: a) control A – tap water without Fe and HA; b) control B – tap water with Fe (II) or/and Fe(III), c) control C – tap water with HA, d) five treatments with the following substances: Fe(II, III)-HA: Fe(II)-HA, Fe(III)-HA, Fe(II)-HA, Fe(II)-HA > Fe(II), Fe(II, III) > Fe(III) > HA > Fe_3O_4-NPs-HA > Fe(III)-HA > Fe(II)-HA > Fe(II)

For higher plants *S. alba*, the addition of HA to Fe(II) and Fe(III) and their mixtures reduces their toxicity. However, when HA were added to Fe₃O₄-NPs, the toxicity of NPs increases significantly. In relation to infusoria *P. caudatum*, the addition of humic acids to Fe(II) ions and Fe(II, III) reduces their toxicity (in relation to the mixture - significantly). However, in the case of Fe (III) the toxicity of the complex increases after the addition of HA. The toxicity of the nanocomposite Fe₃O₄-NPs-HA decreases slightly compared to nanoparticles Fe₃O₄-NPs.

Three months after storage, the toxicity of humic acids and the composite Fe_3O_4 -NPs-HA decreased in relation to higher plants *S. alba*. In relation to infusoria *P. caudatum*, complexes with HA also remained less toxic than ions Fe (II), Fe(III) and a Fe(II, III) of ions. However, the composite Fe_3O_4 -NPs-HA turned out to be more toxic than HA and nanoparticles Fe_3O_4 -NPs and this nanocomposite before storage.

Thus, the addition of humic acids reduces the toxicity of iron ions but increases the toxicity of Fe_3O_4 nanoparticles. After three months of storage humic acids become less toxic, however, nanocomposites Fe_3O_4 -NPs-HA retain their toxic effect.

Interpreting the results was performed by a two-way ANOVA with interactions using XLSTAT Software, 2014. The treatment method (single HA, single NPs or their mixture) and the treatment concentration have a significant effect on acute toxicity samples. For both Fe3O4-NPs and Fe3O4-NPs-HA the toxicity more largely depend on the treatment concentration.

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The content and composition of organic matter in bottom sediments of the Yenisei River near Mining-Chemical Combine (MCC) Rosatom

Bondareva L.G. Federal Scientific Center of Hygiene named after F.F. Erisman, Mytischi, Russia Iydiabondareva@gmail.com

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More than 50 years Krasnoyarsk Mining-Chemical Combine (MCC) manufactures weapon plutonium in single-pass production reactors cooled with water of the Yenisei River. Water discharge from these reactors is the major source of radioactive contamination of the Yenisei River. Sediment composition influences the level of radioactivity to a high degree. The total concentration of measured radionuclides (excluding ⁴⁰K) increases as the sediment decreases in size and becomes less well sorted and as the textural distribution of the sediment becomes more skewed toward fine material. Organic matter has important influences on the fate of environmental pollution in water dam reservoirs. The aim of the studies was to assess content of organic matter fractions in Yenisei River's bottom sediments, and to determine their influence on the content of heavy metals and PAHs. In sediment samples, the content of C organic (Corg), C extracted (Cex), C humic acid (Cha), C fulvic acid (Cfa) and C non-hydrolyzed (Cnh), buffer capacity, and content of heavy metals and PAHs was analyzed. The formation of radioactive flood-land deposits is provided by rapid deposition of suspended material in stagnant zones during periodical flood. Humus compounds contribute significantly to accumulation of radionuclides in the flood-land deposits and bottom sediments.

Radionuclides can also form stable, insoluble complexes with humic materials that tend to reduce radionuclide mobility. These insoluble complexes may be radionuclide-humate colloids that subsequently precipitate from solution or complexes of radionuclides and humic substances that sorb to clay minerals or other soil particulates strongly enough to immobilize the radionuclides. Colloid formation appears to be favored by increased radionuclide concentration and lowered pH; however, the conditions that favor formation of insoluble complexes that sorb to particulates are still poorly understood.

Radiochemical analysis of separate fractions showed that about 20% of Pu and Am are associated with the organic fraction: Pu is nearly equally distributed between Cha and Cfa fractions, whereas Am is preferentially associated with the Cfa fraction (the most mobile fraction of humus matter). It was demonstrated in model experiments that the calciumhydrocarbonate type of water of the Yenisei River causes suppression of formation of mobile fulvate complexes of hydrolyzable radionuclides and, therefore, their transfer into the aqueous phase. In combination with the observed very high distribution coefficients of the radionuclides and low content of their mobile geochemical forms in flood-land deposits of the Yenisei River this suggest that they cannot contribute somewhat significantly to the secondary radioactive contamination of the river water by all mechanisms except migration by mechanical transfer.

The possibility of using hydrolytic lignin processing waste as a humic fertilizer

Brykovskaya N.N.¹, Romanenkov V.A.¹, Pashkevich E.B.¹ ¹Department of Soil Science, Lomonosov MSU, Moscow, Russia, nn_br@mail.ru

Keywords: lignohumate, humic substances, salad (*Latuca sativa* L.)

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Hydrolytic lignin is a valuable raw material used in various industries. After chemical processing of hydrolytic lignin, waste products containing organic compounds, including humic substances (HS), are formed. Biostimulants based on humate-containing raw materials are widely used in agriculture. HS can reduce the influx of heavy metals into the plant, linking them to the HS-metal complexes, which limits the bioavailability of metals [Koukal, 2003; Vereshchagin, Reyzvich, 2005]. The scientific literature provides information on the use of lignohumates (LH) in soils with polymetallic pollution [Izosimov, 2016].

The possibility of using hydrolysis lignin processing waste as a humic fertilizer is a promising direction in addressing waste management issues. This study examined three types of hydrolysis lignin processing waste. Results revealed the presets of organic matter in waste, including HS. The possibility of using these wastes as LH is assessed. Based on the growing experience, biological activity, fertilizer value, and remediation potential of LH were studied when applied at various concentrations. The effect of different concentrations of LH obtained during the processing of hydrolytic lignin on the value of biomass and quality indicators of salad «Vitaminnyy» (*Latuca sativa* L.) was evaluated. LH was added in two concentrations - 0.05 and 0.1 mg C per 1 kg of soil. The comparison was carried out with absolute control and control with the introduction of full mineral fertilizer. The addition of salad biomass to the control with full mineral fertilizer for both doses of LH1 was 38-39%, for LH2 17-38%, which indicates the high efficiency of the tested LH (Fig. 1), while the high concentration of LH turned out to be more effective.

When full mineral fertilizer was applied, the content of nitrates in the lettuce exceeded the maximum concentration limit and amounted to 4782.5 mg per kg of crude material. The use of all LH test subjects made it possible to obtain environmentally friendly products, the nitrate content of which was lower than the maximum concentration limit (MCL), while the absolute nitrate content was more than halved compared to the control with full mineral fertilizer (Fig. 2). The effect of LH1 on an increase in the level of chlorophyll in salad and LH3 on an increase in the content of carotenoids was traced.



Fig.1. It is the biomass of salad «Vitaminnyy»



Fig.2. The nitrate content in the green mass of salad «Vitaminnyy»

On soils with a soil pollution level of copper 1 the MCL produced by the introduction of copper chelate, the responsiveness of salad plants to the use of three tested LH was analyzed. When copper was introduced into the soil, its content in salad leaves on the variant without LH reached the MCL. When LH1 was added at a high dose and LH2 at a low dose, the copper content in plants was significantly lower than the MCL, all other doses of LH did not allow lowering the copper content below the MCL. The effect of the changes made in the content of total chlorophyll, carotenoids, cellular proteins, and protein, compared with a positive result, was weakly expressed in comparison with the experiment with the original soil and more often turned out to be unreliable.

Priority soil micro-aggregate and mezo-aggregate structure synthesis for humic substances better functioning via Biogeosystem Technique soil processing

Chernenko V.V.¹, Glinushkin A.P.², Zinchenko V.E.³, <u>Kalinitchenko V.P.^{2,4}</u>, Minkina T.M.⁵, Makarenkov D.A.⁶, Mandzhieva S.S.⁵, Sushkova S.N.⁵, Il'ina L.P.⁷, Motasova E.D.² ¹Russian Research Institute of Viticulture and Winemaking, Novocherkassk, Federal Rostov Agrarian Research Center, Rassvet, Rostov Region, Russia, tchernencko2011@yandex.ru ²Russian Research Institute of Phytopathology, Bolschije Wjasjomy, Russia

³Federal Rostov Agrarian Research Center, Rassvet, Rostov Region, Russia

⁴Institute of Fertility of Soils of South Russia, Persianovka, Russia

⁵Southern Federal University, Rostov-on-Don, Russia

⁶Institute of Chemical Reagents and High Purity Chemical Substances of National Research Centre "Kurchatov Institute", Moscow, Russia

⁷Southern Scientific Center of Russian Academy of Sciences, Rostov-on-Don, Russia

Keywords: Biogeosystem Technique, intra-soil milling, soil illuvial horizon micro-aggregate and mezoaggregate structure, humic substances better functioning

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"Soil organic matter – soil mineral matter – humic substances" interfaces are crucial for soil organic matter pool, soil structure and productivity. Agronomy technical means and technologies for humic substances apply to soil are outdated. The technological platform of land use is imitative and leads to improper humic substances management, and humic substances potential for soil full scale improving is restrained.

Standard soil tillage, as well as reclamation tillage fails to transform the soil aggregate system adverse properties. Copying Nature in standard soil processing system leads to upper soil layer excessive dispersion, dead-end porosity, soil compaction and heavy block structure. In result, soil aggregates composition responsible for the "mineral matter – water – organic matter" interface become deteriorated and in its turn deteriorates humic substances functioning in soil.

For breaking through the fundamental shortcoming of current industrial technological platform in the field of "agrarian soil mineral matter – humic substances" interface and better result of humic substances apply, in the framework of Biogeosystem Technique (BGT*) methodology we propose a transcendental (uncommon for Nature – not fully imitating the natural phenomena) approach to soil ameliorative processing – BGT* soil improvement – device and technology for illuvial horizon intra-soil milling. Milling working body is fully immersed into the soil. Transcendental artificial soil weathering is applied – not the soil upper layer, but internal soil illuvial layer. Intra-soil milling of a 20-45 cm layer is forming the soil illuvial horizon micro- and mezo-aggregate geophysical system structure. Machines were developed, field trials fulfilled.

The "mineral matter – water – organic matter" interfaces become more controllable. Favorable conditions for humic substances soil functioning, rhizosphere and plants growth are creating. 36 years after one-time 20-45 cm layer intra-soil milling, the soil micro-aggregates prevail, water penetrates the soil freely. This provides the multilevel "soil mineral matter – water – organic matter" interfaces, promotes flocculation of the soil particles and dissolved organic matter. Internal soil geophysical and organic-mineral structure surface become more stable.

The plants consume less energy and water, simultaneously use more nutrients, sequestering more CO_2 and releasing more ionized O_2 most useful for CH_4 and other air pollutants oxidizing and stability of the Earth atmosphere. Soil microbiological process important for soil "mineral matter – water – organic matter" stable interfaces and humic substances soil functioning is optimized, C content is increased: in 0-20 cm layer for 25%, in 20-40 cm layer for 60%.

Study of humic acids and preparations based on them

Dugarjav J.¹, Avid B.¹, Zherebtsov S.²

¹Institute of Chemistry and Chemical Technology, Mongolian Academy of Sciences, Ulaanbaatar, Mongolia, dugar21mn@yahoo.com 2Federal Research Center of Coal and Coal Chemistry of Russian Academy of Sciences,

Kemerovo, Russia

Keywords: humic acids, humate, lymphocyte, fertilizer, Humaton preparation, humic mud, desertification, structural parameters, biological activity.

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In this work have been presented the results of the investigation of humic acids obtained from Mongolian and Russian naturally-oxidized form of brown coal. Humic acids and their soluble salt (humates) have been characterized for elemental content, functional groups, biological activity and solution properties, like optical density, threshold of aggregation and viscosity. Several structural parameters were determined by FT-IR spectroscopy, gel chromatography. We found that soluble salt of the humic acids were able to stimulate the immune system of sick animals, especially on stimulation of transfer of 0 – lymphocyte to T and B – lymphocytes. Antioxidant properties of humic acid in the form of sodium salt relatively to living systems was investigated in vitro and in vivo condition. It was found antioxidant properties of humic substances consists in decreasing synthesis of malondialdehyde that is intermediate product of animal fat peroxidation. As a result, we could develop three commercial products to be used as plant growth stimulator (humic fertilizer): feed humic acid preparation named Humaton for animals and humic mud preparation for treatment of human skin disease. All these products were standardized and have been used. In cooperation with Federal Research Center of Coal and Coal Chemistry of Russian Academy of Sciences is also being carried out evaluation of the biological activity of native and modified humic substance (HP) in order to build a humic preparation to counteracting desertification. Studies have shown that the destructive alkylation and subsequent debituminating of initial coals changes the structural group composition of humic acids, increases the degree of aromaticity. It was established that the biological activity of HP is directly proportional to the following structural parameters: the degree of aromaticity fa, the hydrophilic-hydrophobic parameter fhh, and the parameter reflecting the ratio of aromatic and aliphatic fragments of organic mass of HP (aromaticity / aliphaticity) far / al.

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- 1. Dugarjav J., Avid B. // In: Fourth international conference on chemical investigation and utilization of natural resources: 93, July 8-0, 2016, Ulaanbaatar, Mongolia.
- 2. Dugarjav J. et al. // In: Mezhvuz. Scientific and Practical Conference, Saint Petersburg, 1991.
- 3. Dugarjav J. et al. // Patent of Mongolia, №710,1992.
- 4. Byambasuren N. et al. // In: "Diagnose erudition and progressive methodologies", p. 132–135, State central veterinary laboratory, Ulaanbaatar, 2017.
- 5. Zherebtsov S. et al. // Solid Fuel Chemistry. 2019. 53(5):253–261.
- 6. Zherebtsov S. et al. // Solid Fuel Chemistry, 2019. 53(3):145–151.

Stabilizing ability of humic acids towards Fe₃O₄ nanoparticles under various oxidation conditions

Dzeranov A.¹, Bondarenko L.¹, Pankratov D.², Terekhova V.², Kydralieva K.^{1,2} ¹Moscow Aviation Institute, Moscow, Russia, I.s.bondarenko92@gmail.com ²Lomonosov Moscow State University, Moscow, Russia

Keywords: magnetite nanoparticles, oxidation, humic acids effect, Mössbauer spectroscopy

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Humic acids (HA), natural polyelectrolytes and strong chelates can effectively prevent the aggregation of magnetite nanoparticles, from one side, and from another side, HA as antioxidants can affect the iron species state. The increasing application of iron-based nanoparticles has raised concerns regarding their environmental behavior and potential ecological effects. After release into the environment, iron-based NPs undergo various transformations via the interactions with different geochemical and biological components, which ultimately influence their behavior and potential toxicity. In this study, the oxidation of HA-coated magnetite nanoparticles (Fe₃O₄-HA NPs) is investigated under mild and harsh conditions in order to understand the oxidation behavior and the chemical stability of transition forms in the presence of humic acids. XRD analysis and Mössbauer spectroscopy were used to identify the phase composition of NPs and the change in the oxygen environment of iron atoms.

The chemical precipitation *in situ* method with in the HA matrix or the self-assembly method was used to formulate the Fe₃O₄ NPs from Fe(II) and Fe(III) salts by NH₄OH addition in argon atmosphere. To simulate the oxidation process of the Fe₃O₄-HA three treatment ways were used: mild (storage in tap water during 24 hours, st) and two huge conditions (influence of concentrated HNO₃ or mechanical treatment in the grinder at 1400 rpm in 10 min in air atmosphere, mill). As a total, the following samples were analyzed: Fe₃O₄-HA (ini), Fe₃O₄-HA (st), Fe₃O₄-HA (HNO₃) and Fe₃O₄-HA (mill) and NPs of Fe₃O₄ (ini), Fe₃O₄ (HNO₃) as a control.

According to the XRD data, main phase for all samples tested before and after oxidation is a magnetite Fe₃O₄ NPs. Complete phase transformation to maghemite Fe₂O₃ for all samples under treatment (storage in H₂O, nitric acid and mechanical treatment) was evidenced by Mössbauer spectroscopic measurements. Thus, XRD technique suffers from serious limitations and ineffective in discriminating magnetite. The oxidative degradation of Fe₃O₄ to Fe₂O₃ nanoparticles changed in the order: Fe₃O₄ (HNO₃) < Fe₃O₄-HA (st) < Fe₃O₄-HA (HNO₃) < Fe₃O₄-HA (mill), and indicates oxidative effect of HA towards Fe²⁺ ions.

Mitigating effect of HA on bioactivity of two types of iron oxide nanoparticles of Fe₃O₄ and γ -Fe₂O₃ to the green algae *Scenedesmus quadricauda* and the higher plant (seeds of white mustard *Sinapis alba*) a higher plant (white mustard) were investigated with a focus on the effect of oxidation state. Results show that the growth inhibition by iron oxides nanoparticles (NPs) to both test-species increased with oxidation of the NPs with an order Fe₃O₄ (ini) < Fe₃O₄-HA (ini) < Fe₂O₃-HA (above Fe₃O₄-HA (mill)). The main toxic mechanism, which could explain the difference in toxicity of the NPs is probably Fe²⁺ and Fe³⁺ ions release. These findings will be helpful for the understanding of HA role on the fate and toxicity of iron-based NPs in the environment.

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Separation of oil-water mixtures using humic acids-coated magnetic nanoparticles

Dzhusupkalieva R.¹, Kydralieva K.²

¹Zhangir Khan West Kazakhstan Agrarian and Technical University, Uralsk, Kazakhstan ²Moscow Aviation Institute, Moscow, Russia

Keywords: oil recovery, magnetic nanoparticles, humic acids, composites

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In this study, magnetic nanoparticles (NPs) coated by humic acids (HA) was used to separate a reference VD2 oil from oil-water mixture. Iron oxide NPs are attractive for their inherent low toxicity and magnetism, which allows easy separation of NPs from fluids. Iron oxide NPs were synthesized using a hydrothermal method in air atmosphere. Ferric chloride and ferrous chloride along with HA were used as precursors, and ammonium hydroxide was used as a precipitation agent. AFM images for NPs before and after oil absorption were performed. DLS and zeta potential measurement were performed on a Malvern Nanosizer instrument. Samples were characterized by FTIR spectrometer (FT-IR-200, ThermoNicolet). XRD analysis was performed on DRON-UM-2. Oil samples were prepared by mixing crude oil with pure distilled water in vial via sonication (40 kHz, ambient conditions) for 10-60 minutes. Following the preparation of an oil-water mixture, different amounts of NPS were added to the oil-water and mixed for 5-10 min via sonication to obtain optimum oil:NPs ratio. The NPs concentration for the oil experiment was in the range 4-70 ppm. Then, NPs were separated with Nd magnet (0.3 Tc) for specific periods of time. The remaining oil-water solution in each vial was collected for measurement by UV-vis (Shimadzu UV-2600) and fluorescence spectroscopy (Solar CM2203).

The XRD spectrum of the synthesized NPs showed that magnetite (Fe_3O_4) is the dominant phase of NPs, although the presence of some maghemite cannot be discounted. The average crystal size of 18 and 11 nm for Fe_3O_4 and Fe_3O_4 /HA respectively.

DLS plot for Fe_3O_4 and Fe_3O_4 /HA NPs before oil separation experiment had one single peak and the z-average was 140 and 230 nm respectively, while after oil experiment z-average increases to 310 and 520 nm. Increases in the size of NPs indicate that oil is adsorbed by the NPs and shows the presence of a second layer on the surface of NPs.

To quantify the oil removal, the fluorescence spectrum at excitation wavelength of 337 nm was recorded. Fluorescence spectroscopy results showed excellent oil removal from oil-water mixture in the pure water in experiments with different concentrations of NPs after 30 minutes of separation. By increasing the NPs concentration from 4 to 40 ppm, the oil removal percentage increased from 42 to near 74 %. Fluorescence intensity decreased indicating greater removal of aromatic hydrocarbons. These results are in agreement with UV-vis data. Increasing of NPs concentration to 70 ppm lead to NPs aggregation, reducing surface area and absorption capacity. It resulted in oil removal decreasing.

Fluorescence results in the experiments with effect of separation time showed that by increasing the separation time from 10 minutes to 60 minutes, oil removal increased from 32 to near 100%.

In the presence of humic substances, reduction in NPs aggregation and disaggregation has been observed. Moreover, in case of Fe_3O_4 /HA NPs the percentage of the oil removal was higher than for bare Fe_3O_4 .

Future experiments with simulation of environmental conditions (salinity, pH, ions, natural organic matter, oil grade and concentration) need to be developed.

Esterification of peat by mechanochemical method

<u>Efanov M.V.</u>¹, Konshin V.V.², Sartakov M.P.³ ¹LLC "MIP "Yugra – Biotechnology", Khanty-Mansiysk, Russia, efanov_1973@mail.ru ²Altay state technical university, Barnaul, Russia ³Yugra state university, Khanty-Mansiysk, Russia

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Peat is a unique natural composite, a source of humic substances and raw materials for industry and agriculture. The aim of this work is to develop a new acylated thermoplastic binder from peat to study the possibility of its application in the industry of plate materials [1]. The esterification of peat was carried out in the processing of lowland peat acetic anhydride by mechanochemical method with different molar ratio of reagents from 1:1:0.5 to 1:1:2.5 for 30 min at 25 ^oC. The chemical composition of products of esterification of peat given in the table 1.

Table 1. The influence of the molar ratio of OH:Ac₂O in the acetylation of peat in conditions of intensive mechanical grinding on the properties of the obtained products*

Sample	Mole ratio OH:Ac ₂ O	Content of acetyl groups, %	Solubility in acetone, %	Conversion OH- groups, %
Peat	-	-	5	-
1	1:0,5	10.5	20	42
2	1:1	11.7	30	47
3	1:2	15.2	47	61
4	1:3	17.8	60	71

*the mass of peat is 10.0 g, the temperature of the acetylation – 50 $^{\circ}$ C, the duration of the acetylation – 60 min.

It was found that with an increase in the molar ratio there is an increase in the content of bound acetyl groups in products and their solubility in acetone (table 1).

The influence of duration mechanochemical treatment of peat on the process of xanthogenation under the action of carbon disulphide in the presence of NaOH (table 2) [2].

Table 2. Xanthogenation of peat

Duration xanthogenation of peat, min	Content CS ₂ , %	Solubility in water, %
10	3.6	25
20	5.0	37
30	6.9	49
40	9.0	61
50	11.2	72
60	15.4	89

It is shown that the increase in the duration xanthogenation of peat is the increase in the content and solubility products in water (table 2).

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- 1. Efanov M.V. Method of producing the binder from peat. // Patent of Russian Federation № 2614671. Published 28.03.2017. Bulletin № 10.
- 2. Efanov M.V., Filina E.S. // Chemistry of natural compounds. 2004. 2:151–153.

Electrophysical characteristics of an activated adsorbent from humic acid

Ermagambet B.T.¹, Kasenov B.K.², Kazankapova M.K.¹, Kassenova Zh.M.¹, Nurgaliev N.U.¹, Kuanyshbekov E.E.²

¹TOO "Institute of Coal Chemistry and Technology", Nur-Sultan, Kazakhstan, coaltech@bk.ru ²Chemical and Metallurgical Institute named after J. Abishev, Karaganda, Kazakhstan

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Electrophysical properties are the main indicator of carbon materials used in electrothermal processes, as well as for the manufacture of superconducting materials, capacitors and fuel cells from them [1]. Measurements of electrophysical properties were carried out according to the method [2].

The electrophysical characteristics of an activated adsorbent from humic acid based on coal from the Maykuben deposit (Kazakhstan) were investigated. Porous-carbon material was obtained by carbonization and activation in argon and water vapor. The study of electrophysical properties (dielectric constant (ϵ), electrical resistance (R)) was carried out by measuring the electrical intensity of the samples on a LCR-800 serial device (Taiwan) at an operating frequency of 1 kHz, 5 kHz and 10 kHz continuously in dry air in thermostatic mode with exposure time at each fixed temperature (293–483 K). Based on the data obtained, the band gap (ΔE) of the samples was calculated.

When comparing the electrophysical characteristics of the activated adsorbent from humic acid at various frequencies, it was found that a change in frequency from 1 kHz to 10 kHz does not strongly affect the value of electrical resistance in the range 293-483 K. For example, the lgR values at 293 K and at 1 kHz are 4, 03 ohms, at 5 kHz - 3.99 ohms and at 10 kHz - 3.97 ohms, and at 483 K and at 1 kHz - 2.93 ohms, at 5 kHz - 2.92 ohms and at 10 kHz - 2, 92 ohms. The values of dielectric constant (ϵ) at various frequencies varies with decreasing transition from 1 kHz to 10 kHz. For example, the value of ϵ at 293 K and 5 kHz - 3.295 \cdot 10⁵, at 293 K and 10 kHz - 1.462 \cdot 10⁵, and at 483 K and 1 kHz - 4.336 \cdot 10⁸, at 483 K and 5 kHz - 5.474 \cdot 10⁷ and at 483 K and 10 kHz - 2.013 \cdot 10⁷. The above data show that the maximum values of ϵ are obtained by measuring a frequency of 1 kHz.

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- 1. Yermagambet B.T. et al. // Industry of Kazakhstan. 2019. 1(105):70–72.
- 2. Zhumadilov E.K. et al. // Bulletin of the NAS of the Republic of Kazakhstan. 2004. 5:114–118.

Evaluation of the effectiveness of the use of humic substances as washing agents of oil-contaminated soils

<u>Grechischeva N.Yu.¹</u>, Fakhretdinova D.R.¹, Murygina V.P.², Gaydamaka S.N.² ¹National University of oil and gas "Gubkin University", Moscow, Russia, yanat2@mail.ru ²Lomonosov Moscow State University, Moscow, Russia

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A huge amount of oil polluted soils are located in Russia. The solvent/chemical surfactant washing technique is widely applied for the oil-contaminated soils, but use of it leads to soil recontamination. Natural surfactants based on humic substances (HS), the active matrix of organic matter of soils, peats, natural waters, can be an alternative to chemical surfactants.

The aim of this research is evaluation of the prospects for the use of humic substances as washing agents compare to chemical surfactants in the remediation of oil-contaminated soils.

Gravimetric analysis determination of oil content showed that coal HS "Powhumus" is the most active washing agent towards both the native oil-contaminated soil and to model systems. The use of *Powhumus* reduced the content of oil hydrocarbons from 5.00 to 1.81% in the case of model systems and from 28.96% to 12.79% for native oilcontaminated soil. The action of *Powhumus* is comparable to the well-known synthetic surfactant (sodium lauryl sulfate). It is shown that the effectiveness of the Powhumus increases by 20% upon condition of oil-degradation preparation "Rhoder" use. The introduction of oil degradation microorganisms contributes to the formation of more favorable conditions for the development of soil microorganisms and the degradation of soil contamination. The analysis of group composition of residual oil contamination of soil washed various surfactants and subsequent application of the biological product showed that Powhumus is the most effective agent for the removal of light and heavy oil hydrocarbons of contaminated soils. The coal humic substances "Sakhalin" are more effective for removal of oil polyaromatic hydrocarbons from contaminated soil. In use of water and synthetic surfactants as washing agents of oil-contaminated soil, the natural microbiota of the contaminated soil was suppressed. In use of the natural surfactants based on humic substances led to the development of *Pseudomonadaceae*. Rhodococcus, Bacillaceae, Sarcina soil bacterial community.

Fluorescence quenching of humic substances by engineering silver nanoparticles

<u>Grigoryeva I.O.</u>, Konstantinov A.I., Bagdasaryan A., Volkov D.S. Perminova I.V. Lomonosov Moscow State University, Russian Federation, irastarkova@mail.ru

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Silver nanoparticles (Ag-NPs) enter the environment as a result of their intensive use to impart bactericidal properties to the products. In addition, Ag-NPs are used as broadacting antiseptics as an alternative to antibiotics. At the same time, it is of great interest to study the interaction of Ag NPs with the most reactive components of natural ecosystems humic substances. The aim of this work was to evaluate the binding properties of humic substances of coal with respect to silver nanoparticles using the method of quenching of fluorescence.

As HS, we used coal HA (CHP) and their derivatives modified with hydroquinone (CHP-HQ). Quenching experiments used citrated Ag NPs and ionic silver in the form of silver nitrate. Fluorescence spectra were obtained at pH 12 and a carbon concentration of 5 mg/L to exclude the effect of the internal filter. The silver content ranged from 0 to 5×10^{-5} M. The quenching constant was calculated using the Stern-Volmer equation.

It was found that for all HS samples, in the presence of silver ions, fluorescence quenching is observed. The dependence of F_0/F on $[Ag^+]$ was linear, which allowed the use of the Stern-Volmer equation to calculate the fluorescence quenching constant:

$$F_0/F=1+K[Q]$$

where F_0 and F are the fluorescence intensities in the absence and in the presence of a quencher, respectively, [Q] is the quencher concentration.

The quenching constants for HA coal and HA coal modified with hydroquinone were 2.12*10⁴ L/mol and 2.25*10⁴ L/mol, respectively.

When quenching the fluorescence of Ag-NPs, the dependence of F_0/F on [AgNPs] is characteristic of two types of fluorophores, one of which is not available for the quencher [1]. In this case, a modified form of the Stern-Volmer equation was used to calculate the fluorescence quenching constant:

$F_0/F=1/f_1K[Q]+1/f_1$,

where f_1 is the fraction of the initial fluorescence available for quenching.

The quenching constants for HA coal and HA coal modified with hydroquinone were 3.02*10⁶ L/mol and 2.10*10⁶ L/mol, respectively. Thus, the values of the quenching constant of the Ag fluorescence of HS nanoparticles exceeded by two orders of magnitude the values of the quenching constant of the fluorescence of HS by ion silver. Nonlinear quenching as a result of adsorption of HV on Ag-NPs.

References

1. Guocheng Zh., Jun Y. // J. Fluorescence. 2017. 27(4):1233–1243.

Biogeosystem Technique methodology and technological solutions for priority soil humic substances synthesis and healthy soil, water, and environment

<u>Kalinitchenko V.P.</u>^{1,4}, Meshalkin V.P.², Makarenkov D.A.³, Glinushkin A.P.⁴, Minkina T.M.⁵, Zinchenko V.E.⁶, Chernenko V.V.⁷, Mandzhieva S.S.⁵, Sushkova S.N.⁵, Il'ina L.P.⁸, Rykhlik A.E.¹, Larin G.S.¹, Grishina E.V.⁴

¹Institute of Fertility of Soils of South Russia, Persianovka, Russia, kalinitch@mail.ru ²Mendeleev University of Chemical Technology of Russia, Moscow, Russia

³Institute of Chemical Reagents and High Purity Chemical Substances of National Research Centre "Kurchatov Institute", Moscow, Russia

⁴Russian Research Institute of Phytopathology, Bolschije Wjasjomy, Russia

⁵Southern Federal University, Rostov-on-Don, Russia

⁶Federal Rostov Agrarian Research Center, Rassvet, Rostov Region, Russia

⁷Russian Research Institute of Viticulture and Winemaking, Federal Rostov Agrarian Research Center, Rassvet, Rostov Region, Russia

⁸Southern Scientific Center of Russian Academy of Sciences, Rostov-on-Don, Russia

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Humic substances interfaces in the "environment – soil – soil organic matter – soil mineral matter – technical means and technologies of agronomy, land reclamation, and waste utilization" are the great challenge concerning the current "Ecosphere – Technology" conflict caused by the current outdated imitative technological platform of land use. Copying Nature leads to soil degradation, water scarcity, improper waste management. This interfere the humic substances soil improving and plant fertilizing function. Mechanisms of the fundamental processes occurring at the "humic substances – mineral – water interfaces" remain elusive. The problem of humic substances is to be discussed understanding critically the larger-scale processes occurring at the mineral-water interfaces concerning the existing level of land use, assessing as a whole the Humankind technologies suitability for geospheres.

Technological platform of Noosphere is to be developed concerning a current multialternative business development model as becoming biosphere restricted. This fact is an adverse circumstance for the functioning of humic substances in soil as an agent of soil structure, nutrients turnover, and fertility.

For breaking through the fundamental shortcomings of current industrial technological platform and better result of humic substances apply we propose a new field of science and technology for world strategic development – Biogeosystem Technique (BGT*) – as the only opportunity to face the large-scale calls: sustainable evolution of healthy soil, water saving, high soil fertility, high quality environment to reduce the "organic matter – mineral – water" interfaces elusiveness and increase the Biosphere stability.

BGT* – interdisciplinary institutional innovative integrated solutions, environmentally and economically sound, increasing the Earth biogeochemical flux and biogeosystems stability.

BGT* – implements a transcendental (uncommon for Nature) technical approach.

BGT* – is proven in practice providing the synergy of theory and experiment.

BGT* provides: micro-aggregate soil illuvial horizon structure via intra-soil milling; reduced fresh water consumption via intra-soil pulse continuous-discrete watering; environmentally safe dispersed waste recycling for soil structuring and plant nutrition via intra-soil milling and intra-soil pulse continuous-discrete watering-chemigation; and priority conditions for humic substances function as an agent of soil organic matter synthesis and stability, soil health and high productivity.

Formulation of multilayer magnetic Cu²⁺ and Ni²⁺ – imprinted sorbents based on humic acids

<u>Kasymova E.J.</u>¹, Kydralieva K.A.², Jorobekova Sh.J.³ ¹Kyrgyz-Russian Slavic University, Bishkek, Kyrgyzstan, kasymova_elvira@mail.ru ²Moscow Aviation Institute, Moscow, Russia ³Institute of Chemistry and Phytotechnology NAN KR

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The technique for formulation of magnetic ion-imprinted sorbents aimed at specific metal ions seems to be the most attractive for the design of materials with various sizes, shapes, compositions and morphology, which provide important opportunities for adjusting and controlling the physicochemical properties of the collected materials and create an important perspective for them application. To obtain magnetic selective ion-imprinted sorbents, preliminary encapsulation of Fe₃O₄ nanoparticles into the HA matrix was used, on the surface of which by successive "crosslinking" it is possible to create several layers that add rigidity to the structure and also protect magnetite from the destruction of aggressive substances. Fe₃O₄ nanoparticles were obtained by chemical precipitation by the addition of FeCl₂ and FeCl₃ solutions in the ratio 2:3 in NH₄OH medium. The optimal reaction parameters for the formation of the Fe₃O₄/HA nanocomposite were determined, where the concentration was 0.2 g of HA per 1 g of Fe₃O₄. The time of introducing HA into magnetite medium was determined, which amounted to 20 sec.

A technique has been developed for producing magnetic multilayer selective ionimprinted sorbents with the formation of a crosslinked 3D structure of the composite based on ammonium humate, phenylene diamine (FDA), solutions of target metal ions (Cu²⁺, Ni²⁺). The resulting magnetic multilayer ion-imprinted materials (IIM) are selective with respect to the target metals. The multilayer sorbents provide the integrity of the structure of magnetite. Such multilayer magnetic ions-imprinted sorbents can be used repeatedly in the technique of magnetic separation. The magnetic solid phase sorption characteristics were studied. The effects of various parameters on the extraction efficiency such as pH of solution, the amount of adsorbent, extraction time, the type and concentration of eluent were systematically investigated. Furthermore, the thermodynamic and kinetic properties of the adsorption process were studied to explore the internal adsorption mechanism.

Removal of the Ni(II), Cu(II) from the imprinted material and creation of cavities for hosting Cu(II) was proved by comparison of the response of IIM/HA/FDA to template with that of non-imprinted material (NIM/HA/FDA). The relative selectivity coefficients of Me-IIM for Cu(II)/Co(II), Cu(II)/Cd(II), Cu(II)/Zn(II), and Cu(II)/Pb(II) were determined. The experimental data fit well with the Langmuir adsorption isotherm. The max Cu/Ni-adsorption capacity obtained from the Langmuir isotherm is 256 and 167 mg/g for Cu(II)/Ni(II) IIM, respectively.

Spectral properties of LIGNOHUMATE® as affected by the duration of the technological process

<u>Khundzhua D.A.</u>¹, Yakimenko O.², Stepanov A.², Gladkov O.³, Patsaeva S.¹ ¹Faculty of Physics, Lomonosov Moscow State University, Moscow, Russia, dasha.ok@list.ru, ²Faculty of Soil Sciences, Lomonosov Moscow State University, Moscow, Russia ³LTD RET, St-Petersburg, Russia

Keywords: himic-like substances, absorption, fluorescence

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The key factors for obtaining high-quality humic products are organic matter (OM) source and technological regimes of OM treatment. LTD RET company has developed and patented an industrial technology for the conversion of technical lignosulfonate with following synthesis of a number of humic-like biologically active substances (HLS) [1], which finally compose the commercial humic product Lignohumate® (LH). Yield, composition and biological activity of the synthesized HLS depend on the technological regimes used (alkali content, changes in temperature, pressure, oxidation rate and time) and change with the course of technological process.

The objectives of the study are: (1) to characterize the product in a number of LH samples taken at different times during the technological process using spectroscopic techniques with further perspective to corroborate them with chemical composition and biological activity of LH samples; (2) to reveal qualitative differences in LH spectral features as a function of time. Samples of LH were taken from the heated working mixture at certain time intervals from the start of the oxidation process: at 0, 15, 30, 45, 60 and 120 min. Spectroscopic measurements were performed to analyze LH samples dissolved in water. Absorption spectra were measured with photometer Solar PB2201, fluorescence spectra were detected with luminescence spectrometer Solar CM2203 with several wavelengths of excitation.

Absorption and fluorescence properties of untreated lignosulfonate differ from that for other humic products [2]. However, this study revealed characteristic spectral features caused by samples treatment. Absorption spectra of all the samples demonstrated a local maximum peak at 285 nm and a shoulder at 235 nm, which indicated the presence of phenolic compounds. The intensity of peak at 285 nm in absorption spectra was apparently decreasing along with duration of the technological process. Fluorescence spectra of all samples upon excitation at 235 or 270 nm showed two overlapping emission bands: with maximum at 320 nm (phenolic fluorescence) and within 400...420 nm (humiclike fluorescence). The intensities ratio of those bands has been drastically changed during technological processing, after 15 min of treatment the UV emission band peaked at 320 nm (excited at 235 nm) became prevailing in spectra and the maximum of the blue band shifted from 400 to 420 nm. Fluorescence quantum yield of treated samples was sufficiently lower for treated samples than that for untreated, and was found slightly decreasing along with duration of the process. This makes possible to distinguish untreated and treated LH samples even at short duration of process. This knowledge will help to determine the optimum duration of the process for obtaining a high-quality product.

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- 1. US Patent No. 7198805 B2. Method for Producing Humic Acid Salts. April 3, 2007.
- 2. Yakimenko O. et al. //J. Soils and Sediments. 2018. 18(4):1279–1291.

Complex humic product LIGNOHUMATE: application efficiency on garden crops

Kokhan S.K. LIGNOGUMAT LLC, St-Petersburg, Russia, ksk@lignohumate.ru

Keywords: agriculture, apple, peach

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Humic products as an independent type of agrochemicals are increasingly expanding their presence in modern agricultural technologies, including gardening. They have long proved their effectiveness not only for territories with a favorable climate and high intensity of agriculture, but also under less favorable conditions, in particular in areas of arid steppes and semi-deserts with semi-intensive and extensive farming. And this interest is continuously growing up due to the widespread climate aridization in recent decades.

For over 25 years, our Group of Companies conducted numerous researches on the use of our product "Lignohumate" on a variety of crops in various soil and climatic conditions, including areas with insufficient moisture and high air temperatures.

Being of a practical use in agriculture for more than twenty years, "Lignohumate" has proved its effectiveness. Application in combination with plant protection products provides reliable yield increases. Although the main focus of Group of Companies' products application for decades has been and remains field crops, but recently agricultural producers show more interest in gardening as well. Therefore we initiated the test program for Lignohumate on pome and stone fruit crops, namely, apple and peach. These crops grow in certain climatic zones. For this long-term study, the Crimean Peninsula and the corresponding farms were selected.

This research is carried out under the supervision of the All-Russian National Scientific Research Institute of Viticulture and Enology "Magarach" Russian Academy of Science. The results showed, that when growing fruits, the additional yield reaches 5-6 t/ha. The effect of Lignohumate on the quality of the obtained production is even more pronounced (increase in content of sugar and vitamins on 10-15%).

Due to the low cost per hectare, the use of Lignohumate as a component of a tank mixture is highly cost-effective in of crop protection technics.

The changes in hydrophobic-hydrophilic properties of soil organic matter under the action and aftereffect of mineral fertilizers

<u>Matveeva N.V.</u>^{1,2}, Rogova O.B.¹, Milanovsky E.Yu.^{1,2} ¹Dokuchaev Soil Science Institute, Moscow, Russia ²Lomonosov Moscow State University, Moscow Russia

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The contact angle of wetting (CA) of the solid phase of Chernozems under the action and aftereffect of mineral fertilizers NPK (4 years after the abolition of fertilizers) was measured. The results of this study show that CA can serve as an integral indicator of soil properties and reflect their change under anthropogenic influence. The use of mineral fertilizers NPK increases the hydrophobicity of the surface of the solid phase of the Chernozem. Three particle size and density fractions (M.S. Shaimukhametov method) were identified, different in composition: silt fraction with particles less than 1 µm, light fraction (LF) with a density less than 2 g/cm³ and the residue after the separation of the first two fractions. Alkaline extracts of humus substances (HS) were prepared from the initial soil samples, as well as silt and LF fractions using an alkaline solution of pyrophosphate Na. Carried out the chromatographic fractionation of alkaline extractions from soils and soil fractions of topsoil. The analysis of the obtained results shows that in the areas of application of mineral fertilizers NPK there is an increase in the content of hydrophobic components in the soil, which is consistent with the dynamics of the contact angle of wetting. Analyzing chromatograms of the soil fractions can talk about the redistribution of hydrophobic-hydrophilic components between them. More hydrophobic fragments of HS are concentrated in the LF, more hydrophilic - in the silt fraction. At the same time, the use of mineral fertilizers increases the degree of hydrophobicity of both the LF and the silt fraction. The increase in the content of hydrophobic components in the silt fraction in the application of fertilizers is more expressed than in the LF.

The site of aftereffect of mineral fertilizers is characterized by a smaller spread of the above indicators, which indicates the alignment of the hydrophobic hydrophilic properties of the soil and their fractions composition with the abolition of one of the factors of anthropogenic load. We believe that liquid chromatography of the hydrophobic interaction HS of soils and their fractions can be used in monitoring the state of the soil cover of agricultural lands.

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Soil solution calcium carbonate equilibrium as a driver of soil organic matter and heavy metals transfer and turnover in focus of humic substances soil fertility effect

<u>Minkina T.M.</u>¹, Glinushkin A.P.², Kalinitchenko V.P.^{2,3}, Makarenkov D.A.⁴, Zinchenko V.E.⁵, Chernenko V.V.⁶, Chelnokov V.V.⁷, Mandzhieva S.S.¹, Sushkova S.N.¹, Il'ina L.P.⁸, Chaika V.A.⁴

¹Southern Federal University, Rostov-on-Don, Russia, tminkina@mail.ru

²Russian Research Institute of Phytopathology, Bolschije Wjasjomy, Russia

³Institute of Fertility of Soils of South Russia, Persianovka, Russia

⁴Institute of Chemical Reagents and High Purity Chemical Substances of National Research Centre "Kurchatov Institute", Moscow, Russia

⁵Federal Rostov Agrarian Research Center, Rassvet, Rostov Region, Russia

⁶Russian Research Institute of Viticulture and Winemaking, Novocherkassk, Federal Rostov Agrarian Research Center, Rassvet, Rostov Region, Russia

⁷Mendeleev University of Chemical Technology of Russia, Moscow, Russia

⁸Southern Scientific Center of Russian Academy of Sciences, Rostov-on-Don, Russia

Keywords: Soil organic matter, C, N, P, and heavy metals turnover, soil solution calcium carbonate equilibrium, humic substances management success

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The knowledge problem of development is to identify critical boundary conditions of ambiguity, complexity, equivocality, and uncertainty as an ecological analytical construct, and apply new innovative possibilities for strategic improved landscape management in focus of biogeochemical cycling and resources growth in every part of biosphere, in particular in vast low productive terrains of dry steppe. It is pertinent that the uptake capacity of land-based soil carbon (C) sinks be enhanced through targeted management. However, the best management practices should not be focused on C sequestration from the atmosphere, or irreversible subsoil and geological C burial. Simultaneous C global balance stimulation and C biological phase enrichment are strongly needed. An attempt to have a carbon fixation effect for the sake of net hydrogen production and negative balances of CO₂ emissions will turn to the atmosphere water vapor excess after hydrogen burning. Water vapor is more dangerous greenhouse gas than CO₂. Soil organic matter (SOM) is declining in most agricultural ecosystems, impacting multiple ecosystem services. The nature of C, nitrogen (N), phosphorus (P), and heavy metals (HMs) turnover is critical for the global biogeochemical cycling. Currently, the stability of existing artificial steppe ecosystems is extremely weak, and therefore the dryland C balance is switching from soil C uptake to soil C efflux. The uncontrolled water, mineral and organic matter transfer is an adverse consequence of the standard ecosystems management. Temporal dynamics of soil structure alteration is deemed to be important for essential ecosystem functions but very little is known about it. The state and transfer of SOM, P and other nutrients, and microelements including HMs are closely linked to the content and state of carbonates in soil and in soil solution. Carbonates, as a part of soil minerals, live and decaying organic matter cause the formation of associated ions and complexes of carbonates, SOM, P, microelements, and HMs. Organic matter forms films on the primary carbonates surface, reducing carbonate sedimentation. Thus, the range of mobility, transfer, and bioavailability of organic matter, P, microelements, and HMs is vast and uncertain. The cause of soil solution excess saturation with calcium carbonate, as well as the transfer and accumulation of organic matter, and HMs is the ions association and formation of mineral and organic complexes in soil solution. The properties, structure, and soil solution calcium carbonate equilibrium (CCE) is under the influence of ions activity and determines the dynamics of material composition, migration, and accumulation of mineral and organic matter into the soil continuum aggregate system, determining the humic substances management success under the influence of CCE.

On the effect of autumn treatment of soils with humic acid preparations on soil biological fertility and yield in the German agriculture

<u>Nowick W.¹</u>, Semeniuk I.², Karpenko E.², lutynska G.³, Yamborko N.³ ¹daRostim Private Institute of Applied Biotechnology, Waldheim, Germany, info@darostim.de ²Department of Physical Chemistry of Fossil Fuels InPOCC, NAS Ukraine, Lviv, igorsem777@gmail.com

³Institute of Microbiology and Virology, NAS Ukraine, Kiew, iutynska@mail.ru

Keywords: humic preparations, phytohormone-humic compositions, biological indicators of the soil

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As part of the international long-term program Tandem^{12/21} (2012-2021) we investigated changes in the microbiological parameters of the soil at different locations (soil index 27 to 67) through the application of various phytohormone-humic acid (PHC) composites. PHCs applied in the spring during the spring stimulated photosynthesis and contributed to the improvement of the biological parameters of the soil. In order to intensify the conversion of organic material into humus and to stabilize these indicators in winter, the experimental fields were treated after harvest with PHCs (autumn treatment - AT) [1].

The results of the monitoring of soil biology and yield increases showed additional effects of a systematic AT. The averages were compared between 2012-2018 with additional AT with the 2006-2011 control period. The AT resulted in a new biological equilibrium in the soil with a 47 to 89% higher concentration of the air nitrogen-binding (N) and phosphorus-mobilizing (P) bacteria relevant for biological plant nutrition. Yield increases of 6.6 to 7.7 GE (10 to 22%) were achieved with the same or even lower N fertilizer use. The efficiency of the N fertilizer used in relation to the yield (Y / N) increased by 10 to 40%.



Figure 1, 2. Effect of autumn treatment on biological soil fertility and yield at different sites (average values: control: 2006-2011, AT: 2012-2018)

References

1. Nowick W. et al. // In: XII International scientific-applied conference daRostim, Odessa 2016, Ukraine, Proceedings, p.173–177.

On the long-term effect of Novihum in the cultivation of wine

<u>Nowick W.¹</u>, Nowick E.¹, Sorge R.², Maslak D.³, Skakun T.³, Lomonosova V.³, Kuleshova Y.³, lutynska G.⁴, Yamborko N.⁴

¹daRostim Private Institute of Applied Biotechnology, Waldheim, Germany, info@darostim.de ²Novihum Technologies GmbH, Dresden, Germany, r.sorge@novihum.com

³Belarusian State University, Biological Faculty, Minsk, Republic of Belarus, feklistova@bsu.by ⁴Institute of Microbiology and Virology, NAS Ukraine, Kiew, iutynska@mail.ru

Keywords: Novihum, wine, soil, photosynthesis, phytopathogenic bacteria and fungi

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The dynamics of the long-term effect of the soil additive Novihum on a wine plantation, which was established in 2013 on a lignite open pit recultivation area, were investigated. The additional plant physiological effect of Novihum, which correlates with faster shoot growth, better vitality, greater photosynthetic performance and higher yields, ends after about 5-8 years. The annual measurements of the chlorophyll fluorescence dynamics (CFD) on the vine leaves proved to be particularly efficient, showing a linear decrease in Novihum activity in terms of additional photosynthetic performance over the entire period. In addition, we report on the geochemical initial state of the soil and results from biological soil analyzes after the 5th year of cultivation. The latter, however, are only snapshots and provide only initial hypotheses on the assumed role of the biological, phytosanitary and physico-chemical initial state of the soil on the long-term effect of Novihum. The influence of these soil parameters should therefore be investigated more precisely in new experimental settings.





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References

1. Nowick W. et al. // In: XIV International scientific-applied conference daRostim, Minsk 2018, Belarus, Proceedings, p.147–148.

Design of humic-based iron nanofertilizers: iron (hydr)oxide chemistry, nanoscale benefits, and multilevel impact of humic substances

Polyakov A.Yu.^{1,2}, Cieschi M.T.³, Sorkina T.A.⁴, Zimbovskaya M.M.², Lebedev V.A.^{2,5}, Volkov D.S.², Pankratov D.A.², Kulikova N.A.², Perminova I.V.²

¹Kurnakov Institute of General and Inorganic Chemistry, Russian Academy of Sciences, Moscow, Russia, a.yu.polyakov@gmail.com

²Lomonosov Moscow State University, Moscow, Russia

³Autonomous University of Madrid, Madrid, Spain

⁴Management Company RUSNANO, LLC, Moscow, Russia

⁵Bernal Institute, University of Limerick, Limerick, Ireland

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Iron deficiency, also known as chlorosis disease, is a frequent issue for numerous crops and strongly impacts agriculture in the countries rich in calcareous soils. This is due to the low availability of microscale iron(III) (hydr)oxides abundant in soils with pH > 7. Typical treatment of lime-induced chlorosis is the application of synthetic iron chelates such as Fe-EDTA, Fe-DTPA, Fe-EDDHA, etc. While these compounds rapidly provide plants with bioavailable iron, chelating agents can accumulate in soils, contaminate groundwater, and increase the mobility of heavy metals and radionuclides. The high cost is another important drawback of the iron chelate preparations. At the same time, nanosized (<5-20 nm) iron (hydr)oxides exhibit good availability to plants and within the recent decades became a versatile platform for the design of advanced iron nanofertilizers (Fe-NF). Because of high affinity to the iron oxide surface and rich beneficial effects on plants, humic substances (natural eco-friendly polyelectrolytes) are considered as the best stabilizing agent for Fe-NF. However, for mass production and effective utilization of the nanofertilizers, several fundamental and technological issues still should be solved, including identification of the most bioavailable iron (hydr)oxide nanoparticles (in terms of both phase composition and size), long-term stabilization of hybrid iron-humic colloids, and development of the most effective application routes considering the nanoparticle uptake and translocation in the target plants.

In the present conference contribution, we will discuss the most important features of Fe-NF production based on the extensive studies pursued by our group. We comprehensively characterize iron (hydr)oxide nanoparticles - the main part of the nanofertilizers – using state-of-the-art materials science techniques: analytical transmission electron microscopy (TEM), electron energy loss spectroscopy (EELS), extended X-ray absorption fine structure spectroscopy (EXAFS), and small-angle X-ray scattering (SAXS). We also use a great gift coming directly from the nuclei of Fe atoms: the ability to analyze iron speciation by Mössbauer spectroscopy including analysis of the frozen aqueous reaction mixtures and ⁵⁷Fe-enriched samples. These techniques allowed a deep understanding of nanoparticulate iron (hydr)oxide chemistry and important ironhumic interactions. Our bioassays include root application of Fe-NF to cucumber, wheat, and soybean plants grown in Hoagland / Knop nutrient solutions or model calcareous soil. Content of iron and chlorophyll *a* in the fertilized crops and their photosynthetic activity were monitored. Mass spectrometry chemical analysis of soybean shoots, pods, and roots fertilized with ⁵⁷Fe-enriched preparations was used as well to track Fe-NF translocation. Recent results on foliar spray application of the nanofertilizers would also be discussed.

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Comparison of soil humic acids and lignohumates

Pospíšilová L.¹, Horáková E.¹, Vlček V.¹, Menšík L.²

¹Dept. of Agrochemistry, Soil Science, Microbiology and Plant Nutrition, Mendel University in Brno, Czech Republic, pospisi1@mendelu.cz

²Research team Sustainable Management and Utilization of Permanent Grassland, Division of Crop Management Systems, Crop Research Institute, Prague, Czech Republic

Key words: humic acids, structure, spectroscopic techniques

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Humic acids are among the most important constituents of soil humic substances. because of relating to soil yield. They are of great importance not only in soil, but also for solving environmental problems, in industry, and medicine. Considerable effort has been made in their research. In spite of that, there is still up-to-date improving of their identification methods. Determination of their structure is guite complicated and usually elemental composition and non-destructive spectral techniques are applied. In this work, SFS, FTIR, and ¹³C NMR spectroscopy were used for detailed characterization of soil HA and lignohumates. ¹³C NMR spectroscopy help us quantitatively detect different types of carbon (e.g. carbonyl, carboxyl, aromatic, olefinic, anomers, and aliphatic carbon). Percentage of aromatic and aliphatic carbons allows us to calculate aromaticity degree. We came to the conclusion that soil HA contained higher amount of aromatic carbon and less aliphatic carbon to compare with lignohumate. ¹³C NMR spectrum of lignohumates (Fig. 1) indicated presence of phenolic carbon (C=O) at 143-157 ppm; sp3 carbon (C-O) at 43-87 ppm; aliphatic and methoxylic groups at 50-60 ppm; long ring -O-alkyl and -CH2groups at 30 ppm. Less content of aromatic carbon (region 106 - 157 ppm) and more aliphatic carbon (region 15-106 ppm) was found in lignohumates. Indexes calculated from different spectral measurements are considered as a very important indicators of HA quality and origin.



Figure 1. ¹³C NMR spectrum of lignohumate.

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References

1. Pospíšilová L. et al. // Acta Agriculture Scandinavica, Section B – Soil and Plant Science. 2015. 65/6:517-528. ISSN 0906-4710.

The change of soil humic substance (HS) and heavy carbon isotope $(\delta^{13}C)$ in soil and soil microorganisms over *in vitro* bioremediation of oil-spilled soil

Shkidchenko A.¹, Akhmetov L.¹, Andreev A.²

¹Institute of Biochemistry and Physiology of Microorganisms RAS, Pushchino, Russia, shkidchenko@gmail.com

²Institute for Cell Biophysics RAS, Pushchino, Russia

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The problem of complete (100%) restoration of affected soils after oil hydrocarbons spills is still relevant, despite the efforts of researchers working in the field of environmental engineering. The laboratory experiment focuses on studying the kinetics of soil HS fractions, isotopic composition δ^{13} C (soil, oil hydrocarbons and microbial biomass) and heterotrophs number.

Bioremediation of oil-pollited soil at 6°C by introducing a consortium of active oil-degrading bacteria has failed in crude oil full biodegradation till the end of experiment (400 days), even with additional inoculations of the consortium. After adding crude oil the HS content in the soil has grown, with the content of soluble HS being increased two-fold, and the one of insoluble HS – three-fold, and remaining the parameters the same till the 400th day (Figure 1).



Figure 1. The kinetics of heterotrophs number, HS and crude oil content during *in vitro* soil experiment.

Application of mass-spectrometry has made possible defining fractionation of carbon isotopes of crude oil hydrocarbons by microorganisms. Heavy isotopes (δ^{13} C) are picked for the microbial biomass but the light ones (δ^{12} C) are turned out as CO₂ [1]. Isotopic composition of soil carbon has changed from δ^{13} C -29,94 to -29,18‰, and the fact confirms the effect of carbon isotopes fractionation by microorganisms which possibly is a reason of incomplete oil biodestruction in soil.

^{1.} Zyakun A.M. et al. // Mikrobiologiya. 1986. 6:953–957.

Mechanical and mechanochemical activation of West Siberian peat

<u>Skripkina T.</u>^{1,2}, Naymushina O.¹, Tikhova V.³, Podgorbunskikh E.², Zubakova E.¹ ¹Sobolev Institute of Geology and Mineralogy, SB RAS, Novosibirsk, Russia ²Institute of solid state chemistry and mechanochemistry SB RAS, Novosibirsk, Russia, urazovatanya@mail.ru

³Vorozhtsov Novosibirsk Institute of Organic Chemistry SB RAS, Novosibirsk, Russia

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It is known that mechanochemical treatment in the presence of reagents of various nature (acid-base, redox, enzymes) leads to changes in the quantitative contents, physical, chemical properties and reactivity of the main components of peat, including humic acids [1]. The aim of this work was to study the effect of mechanical and mechanochemical activation of the West Siberian peat from forest steppe on its copper ions bind capacity. The peat under study contains 2.7% humic acids, 2.8% fulvic acids and 2.17% of ash.

The mechanical activation of peat was carried out in a planetary mill AGO-2. The mechanochemical activation of peat was carried out under the same conditions with 5% wt. dry sodium percarbonate $Na_2CO_3 \cdot 1,5H_2O_2$, which previously showed its effectiveness in the mechanochemical oxidation of brown coal humic acids [2].

The surface properties of peat before and after mechanochemical treatment were studied by thermal desorption of nitrogen. It was found that C_{BET} increases from 6.26 for the initial peat to 18.18 for mechanically activated (MA) peat and to 27.83 for mechanochemically activated (MCA) peat. The capacity of the monolayer for the initial peat is 0.957 mg/l, for MA peat it decreases to 0.858 mg/l, for MCA it increases to 1.092 mg/l. The specific surface area calculated by the Brunauer-Emmett-Teller equation for the initial peat is of $4.2 \pm 0.3 \text{ m}^2/\text{g}$, for MA peat is of $3.7 \pm 0.2 \text{ m}^2/\text{g}$, for MCA peat it increases to $4.8 \pm 0.3 \text{ m}^2/\text{g}$.

The results of reverse potentiometric titration and IR spectrometry indicate that the mechanical and mechanochemical activation of this type of peat under the conditions used does not increase the content of phenolic and carboxyl groups. The IR spectra of MA and initial peat are similar, while the IR spectrum of the MCA peat shows a decrease in absorption in the regions of 1725 and 1260 cm⁻¹, with a simultaneous increase in the regions of 1613 and 1418 cm⁻¹, which indicates the passage of a chemical reaction neutralization of the acid groups of humic acids and their transformation into the form of sodium humate.

Cu (II) ions sorption experiments were carried out. The peat under investigation could be efficiently used for removal of cooper from low-level liquid waste. Graphical solution of the linear Langmuir equation for sorption on peat gives the following values $Q = 2.5 \cdot 10^{-3}$ mmol/g, and a constant = $0.76 \cdot 10^2$. A comparison of the IR spectra does not show a significant difference in the content of the main functional groups in the studied peat samples. The spectra for mechanically activated peat before and after copper sorption is almost identical, which may indicate the physical nature of the sorption of copper ions on this peat. Sorption characteristics can be improved by changing the surface properties, for example, by mechanochemical treatment under the described conditions.

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- 1. Savel'eva A.V. et al. // Russ. J. Appl. Chem. 2013. 86(4):552–557.
- 2. Skripkina T.S. et al. // Solid Fuel Chem. 2018. 52(6):356–360.

Characterization of humic-like substances of linden pyrochar as stimulators of the intensity of soil respiration

Smirnova E.V., <u>Okunev R.V.</u>, Giniyatullin K.G., Guseva I.A., Gordeeva K.A. Kazan (Volga region) Federal University, Kazan, Russian Federation, tutinkaz@yandex.ru

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Pyrochar is a promising ameliorant obtained by pyrolysis of organic material under oxygen deficiency conditions, the use of which is aimed at solving two of the most important problems of our time: long-term improvement of soil fertility and the need for atmospheric carbon sequestration [1]. The perspectives for atmospheric carbon dioxide sequestration are associated with the fact that the carbon content in pyrochar, which is usually resistant to oxidation, can be stored in soils from several hundred to thousands of years [2]. Thus, the introduction of pyrochar, prepared and applied to soils in an industrial scale, makes it possible to remove up to 50% of the carbon of the initial plant residues from the biological circuit [3].

Along with inert components (benzene polycarboxylic acids), a significant part of the carbon in the pyrolysis products of organic materials can be represented by humic substances, glycolipids, phospholipids, etc. [2]. They will be characterized by different resistance to microbial destruction, however, the introduction of these substances with pyrochar can change the availability of available organic food for soil microorganisms. As a result, after the introduction of pyrochar, the CO₂ release can increase due to the rapid mineralization of the labile fraction of the pyrogenic material. In our work, using different temperatures of pyrolysis, pyrochars were obtained from the remains of linden wood. Humic-like substances were extracted with an alkaline solution [4]. Their qualitative characteristics were studied by UV spectrometry. To study the effect of humic-like substances on substrate-induced (SIR) and basal respiration of soil, model experiments with the introduction in the soil of native pyrochar and pyrochar treated with an alkaline solution were carried out. The intensity of soil respiration was determined after 3 months and 6 months of incubation.

The results of the study showed that native linden pyrochars added to the soil cause a short-term increase in soil SIR. The addition of pyrochar to the soil modified by treatment with an alkaline solution leads to an additional increase in the emission of CO_2 from the soil, which decreases slightly over time.

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- 1. Lehmann J., Joseph S. Biochar for Environmental Management: Science and Technology. London: Earthscan Publ., 2009. P.1–12.
- 2. Kuzyakov Y. et al. // Soil Biol. Biochem. 2014. 70:229–236.
- 3. Lehmann J. et al. // Mitig. Adapt. Strat. Glob. Chang. 2006. 11:395–419.
- 4. Kononova M.M., Belchikova N.P. // Sov. Soil Sci. (Pochvovedenie). 1961. 10:75-87.

Comparative analysis of industrially produced and reference humic substances

Upska K.¹, Klavins M.², Viksna A.¹

¹Faculty of Chemistry, University of Latvia, Riga, Latvia, karina.upska@lu.lv, arturs.viksna@lu.lv ²Department of Environmental Science, University of Latvia, Riga, Latvia, maris.klavins@lu.lv

Keywords: humic substances, authentification, stable isotopes

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Humic substances have a wide spectrum of biological activity and therefore have found applications in agriculture, veterinary, medicine, environmental technologies. Large quantities of humic substances are produced in many countries around the world and their use in various areas of the economy is developing. However, it should be noted that the structure and properties of humic substances depend on their origin. However, as humic substances can be obtained from diversity of different sources using different extraction, concentration and purification methods, their properties and structure have high variability. All these factors can influence properties and thus also application areas. Some manufactures do not always correctly identify the sources of the humic substances, and their use is not always justified. The aim of the study is to compare properties of industrially produced humic substances with properties of humic substances of known origin (soil, peat, water etc.). Another aim of the study is to develop authentication methods of humic products, based on identification of their origin. In the study were used potassium humates, humic acids and fulvic acids isolated from well characterized and known sources: low rank coal, peat of different origin (low moor, raised bog peat), soil, compost, vermicompost and others. The methods used for characterization includes elemental analysis, spectroscopic characterization, UV, FTIR, fluorescence) and stable isotope (δC , δN , δO) ratio analysis. The results of the study indicate possibilities to identify sources of industrially produced humic substances.

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Universal organic fertilizers "EldORost" based on humic substances

Zhilkibayev O.T.¹, Shoinbekova S.S.¹, Sarsenbaeva G.B.², Aitbayev T.E.³, Tukenova Z.A.⁴ ¹AI-Farabi Kazakh National University, Almaty, Kazakhstan, zhilkibaevoral@mail.ru ²Kazakh Research Institute of Plant Protection and Quarantine named after Zh. Zhiyembaev, Almaty, Kazakhstan

³Kazakh National Agrarian University, Almaty, Kazakhstan

⁴Kazakh Research Institute of Soil Science and Agrochemistry named after U.U.Uspanov, Almaty, Kazakhstan

Keywords: humic substances, organic fertilizer, agriculture, crops, yield

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Seventy percent of Kazakhstan land is subject to desertification. Today some 14 mil ha out of 182 mil ha of Kazakhstan pasture lands are completely non-usable, and the total degradation area exceeds 50 mil ha, which is explained by intensive and very intensive desertification. One of the most efficient methods used to increase soil fertility and crop vield is the use of organic fertilizers. They help increasing the content of humus in soil, soil structure and avoiding many adverse consequences caused by the use of chemicals. In general, environmental balance cannot be maintained without the use of organic fertilizers for farming purposes. Therefore, development of new high-efficiency and low-cost multipurpose organic fertilizers with complex features (regulating, antistress, immunestimulating, moisture-retaining, etc.) on the basis of humic acids becomes very urgent since the need in high-efficiency phytoregulators is growing day in and day out. The purpose of this work is the creation and introduction of a new domestic integrated highperformance organic fertilizer for modern technologies to restore soil fertility, forests, reduce land degradation in arid areas of Kazakhstan and increase the yield of agricultural cultures. We have produced an innovative domestic plant growth regulator from highly oxidized brown coal and lowland peat by extraction using alkali solutions adding a set of amino acids, natural phytohormones and micro- and macroelements. The distinguishing feature and scientific novelty of the proposed project as compared to the traditional ones is the use of natural amino acids, phytoharmones extracted from vegetable raw materials and addition of micro- and macroelements in certain proportions. In-depth laboratory and demonstration field (microplot) comparative tests on cereals, vegetables and other crops demonstrated a high efficiency of the new domestic multipurpose organic plant growth regulator.

"EldoRost" is a natural regulator of plant growth and development and intended for all types of crops in any soil and climatic zones. It promotes the cultivation of environmentally friendly agricultural products and reducing the content of heavy metals, radionuclides and nitrates in products. It is allowed to produce better and more environmentally pure products (with a high content of carbohydrates, proteins, lipids and other valuable substances). The national universal organic plant growth regulator is a natural product what has valuable antioxidant properties. It is actively involved in the neutralization and elimination of toxins.

The study the effect of a new organic fertilizer codenamed EldORost on sowing qualities, yield, and also on the fungal and bacterial microflora of grain seeds (wheat, rice), vegetables (potatoes, carrots, cabbage, cucumbers, tomato), soybeans and fodder crops (Sudanese grass) were conducted in laboratory and field conditions. The new product (growth stimulator) "EldoRost" showed a higher stimulating efficacy in all cultures in comparison with the standard product.

The organic product "EldOrost" increases the crops of the studied plants by 10-50%, increases the root system, enhances the immunity and water-holding capacity of the soil, and accelerates the synthesis of chlorophyll and the maturation of the crop by 10-12 days.
Corrosion of non-ferrous metal in the presence of humic substances

Zimbovskaya M.M., Makunin A.V., <u>Pankratov D.A.</u> Lomonosov Moscow State University, Moscow, Russia, pankratov@radio.chem.msu.ru

Keywords: corrosion, non-ferrous metals

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Chemistry of corrosion processes is one of the most important scientific domain. The most thoroughly studied mechanisms are that of corrosion of engineering materials under the influence of reagents of anthropogenic origin. However, a significant part of metallic structures contacts with ground and soils, containing natural polyelectrolyte of a multifactorial action (acid-base, redox, complexing, etc.) – humic substances (HS). On the one hand, due to their high reactivity, HS can directly interact, for example, with metallic iron. On the other hand, it is known that archeologists often find metallic artifacts in very good condition in saturated HS waters (bogs, peat bogs, etc.). In addition, mixed corrosion inhibitors based on lignosulfonates, tannins and amino alcohols are suggested for the protection of steel reinforcement in reinforced concrete. It is obvious that the mechanisms of interaction of HS with metals are complex, and require a comprehensive study.

Earlier [see HIT-2017], we showed the role of HS in the corrosive destruction of ironcontaining engineering materials with metallic protective coatings of various nature. This article discusses corrosion under the action of HS of the following non-ferrous metals: aluminum, chrome, nickel, copper, zinc, tin, bronze, brass, nickel plated brass. As the corrosive medium were used: a) tridistilled water (pH 6.5); b) 3% NaCl solution (pH 8.5); c) 0.01% HS solution (obtained by alkaline extraction from "Natural Humic Acids", LifeForce, Saratov, Russia) (pH 7.6); and d) a solution containing 3% NaCl and 0.01% of the above HS (pH 7.7). Samples of materials were exposure in these solutions at a temperature of 45°C in the course of 48 hours. After the expiration, the properties of solutions for practically all samples have changed significantly. So, solutions containing HS was almost completely discolored (except for solutions that contacted with Cr, Ni and Al. For all samples, formation of brown sediments was observed (with the exception of the zinc which dense white sediment was observed). Scratches on the surface of some samples was covered with a dense layer of brown corrosion products. Visually, the highest corrosion intensity was observed for solutions with NaCl. The surfaces of the initial samples and the samples after treatment was studied by optical microscopy $(5^{x} - 20^{x})$, and SEM (with resolution to 200 nm). The compositions of the metal substrate and the coatings of the materials were determined by the energy-dispersion analysis method.

The nature of the changes on the metal samples surface and in the depth of the "scratch" was controlled by comparison with similar data for untreated materials. From the data of a microscopic study, it follows that on the surface of "active" metals (Al, Cu, Zn) corrosion products are formed unevenly - distinct regions with more and less dense coatings of corrosion products are clearly distinguished, clearly differing in their morphology and, possibly by chemical composition. On the surface of the samples of "passive" metals (Cr, Ni), thin layers of compounds are observed, but only under requirement of the presence of HS in the electrolyte. The most intense formation of corrosion products for all samples is observed in the area of "scratches" and natural defects, presented in the coating of the studied materials. Moreover, products of corrosion are formed in water or NaCl solution as loose morphology. In the case of solutions containing HS, including the presence of NaCl, a dense coating is formed above the active (unprotected coating) surface of the sample, possibly from sparingly soluble metal compounds and HS. Thus, using several samples of non-ferrous metals, it was shown by direct experiment that despite its high chemical activity to many metals, and in particular to iron, HS can serve as shielding corrosion inhibitors even in such aggressive media as NaCl solutions.

Сапропель – ценнейшее сырье для получения гуминовых препаратов

Митюков А.С. Институт озероведения РАН, ул. Севастьянова, д. 9, 196105 Санкт-Петербург, Россия, mitals@yandex.ru

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Научно-исследовательские работы Института озероведения РАН (ИНОЗ РАН) направлены на всестороннее изучение водных ресурсов континентальных водоемов и перспектив их практического применения, включая различные виды гуминовых веществ и сапропелей [1,2,4]. В настоящее время сапропели рассматривают как ценное органическое и органоминеральное сырье для различных отраслей страны и закономерно стали объектом детального изучения. Напомним, что сапропель – это продукт донных отложений пресноводных водоемов, образующийся в результате постмортальных превращений озерных гидробионтов и трансформации почвогрунтовых частиц при ограниченном доступе кислорода. Этот природный материал представляет собой желеобразную массу, которая постепенно уплотняется по мере увеличения глубины отложений и их возраста. Скорость седиментации и накопления сапропеля зависит от многих факторов - биотических (планктон и бентос) и абиотических (освещенность, градиент солености воды, концентрация растворенных и коллоидных веществ) [1]. Однако, несмотря на мощные и постоянно пополняющиеся запасы сапропеля в России, широкого применения этого природного сырья в экономике страны и, в частности, в сельском хозяйстве до сих пор очень ограничено. В этой связи целью настоящего исследования является эколого-биологическая оценка возможности и безопасности применения в сельскохозяйственном производстве ультрадисперсных гумато-сапропелевых суспензий.

В настоящее время сапропель континентальных водоёмов России становится востребованным сырьем для использования в лечебных целях, для производства сорбентов, органоминеральных удобрений и пищевых биологически активных добавок (БАДов). В ИНОЗ РАН проведено сравнительное исследование гранулометрических и биологических свойств гуминовых веществ, полученных из сапропеля типичным (щелочным способом экстрагирования) [2.3] и путем применения ультразвуковой кавитации. Показано, что применение гуминовых веществ сапропеля с повышенным содержанием наночастиц в качестве препаратов в животноводстве приводит к существенному экономическому эффекту. Использование гумато-сапропелевой суспензии в животноводстве позволяет увеличить среднесуточный прирост живой массы животных до 24% и выше по сравнению с Исследования, проведенные совместно контрольными группами. С Научноисследовательским институтом экспериментальной медицины РАН, показали, что суспензия сапропеля обладает адаптогенным свойством, повышает устойчивость клеток к неблагоприятным условиям, увеличивает функциональный резерв иммунных клеток животных. Препарат обладает стресспротективным и ранозаживляющим действием. Прирост урожайности пшеницы достигает 20% с 1 га при значительном снижении нормы высева семян за счет увеличения кущения, активации биохимических процессов , повышенного поглощения ультрафиолетового излучения и ускорения процессов фотосинтеза в листьях за счет нано частиц с высокоразвитой поверхностью и наличием химически активных функциональных групп в гумато-сапропелевой суспензии. Применение в растениеводстве в качестве удобрения и в животноводстве в виде кормовых добавок ультрадисперсной гумато-сапропелевой суспензии из озерного сапропеля позволит в дальнейшем заменить дорогостоящие импортные препараты и решить проблему замещения их импорта.

Литература

- 1. Штин С.М. // М.:Издательство МГГУ, 2005. 373 с.
- 2. Орлов Д.С., Триш Л.А. // М.:МГУ, 1981. 272 с.
- 3. Румянцев В.А., Крюков Л.Н. // Общество. Среда. Развитие. 2015. № 3. С. 170–174.
- 4. Перминова И.В. // Дисс. на соиск. уч. ст. докт. хим. наук. 2000. 359 с.

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