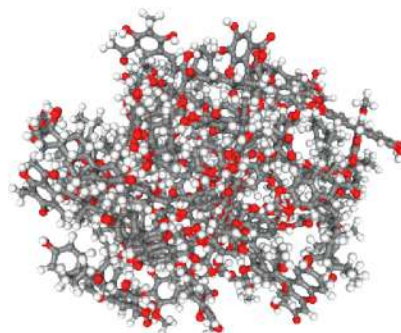


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**HUMIC
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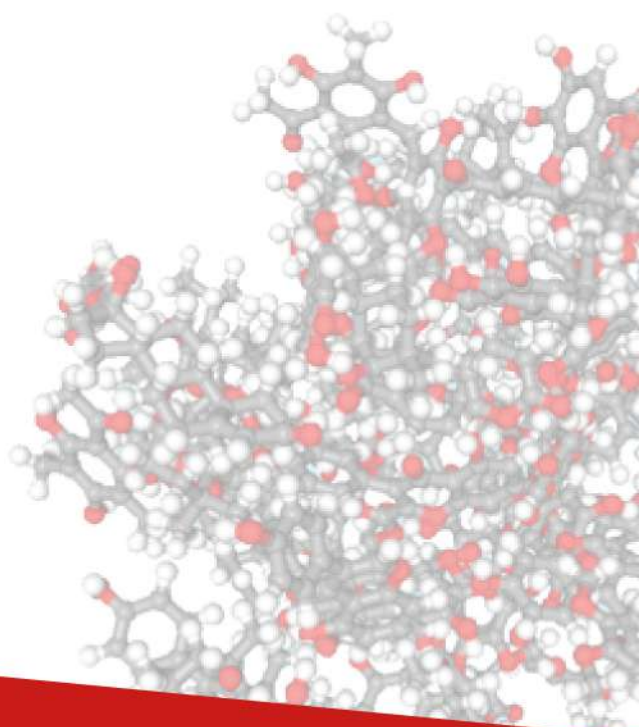
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**ГУМИНОВЫЕ ВЕЩЕСТВА И
ТЕХНОЛОГИИ ВЫЖИВАНИЯ**

Seventh International Conference of the CIS IHSS
on Humic Innovative Technologies

**HUMIC SUBSTANCES AND
TECHNOLOGIES FOR RESILIENCE**

**Книга тезисов
Abstract book**



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Humic substances as complex systems

Humus and carbon sequestration technologies in soil

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Keywords: carbon sequestration, soil

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The supramolecular understanding of humus suggests that the heterogeneous relatively small humic molecules surviving the progressive degradation of biomolecules released in soil by the cell lysis are progressively enriched with apolar components. There is thus a thermodynamic drive to associate the humic molecules into apparently large molecular sizes by relatively weak interactions such as hydrophobic interactions and H-bonds. This view implies that organic carbon in soil can be stabilized by protection into the hydrophobic domains of humic substances or mature compost. Evidence of such hydrophobic protection mechanism and its effective capacity to sequester labile organic carbon in soils are shown here. The supramolecular nature of humus also implies that stronger covalent bonds can be established between phenolic components of humus by catalyzed oxidative reactions. Synthetic metal porphyrins added to soils as either water-soluble compounds or immobilized on clay minerals are capable to efficiently catalyze the oligomerization of soil humus under either the addition of an oxidant, such as hydrogen peroxide, or solar irradiation. The in-situ photopolymerization of soil humus was proved in field trials to effectively sequester organic carbon in soil and have the potential capacity, if extended to the global agricultural scale, to offset the anthropogenic emissions of greenhouse gases in the planet atmosphere.

New mesoporous polymer materials: synthesis, structure, and properties'

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Keywords: polymer, nanocomposites

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This work addresses the preparation of mesoporous polymeric materials via eco-friendly strategy of environmental crazing which allows sustainable modification and functionalization of conventional polymers (polyethylenes, polypropylene, PTFE, UHMWPE, etc.). This approach involves plastic deformation of polymers and self-induced development of fibrillar-porous structure with nanoscale pore dimensions below 20 nm (Fig. 1). Structural parameters (porosity and pore dimensions) of as-prepared mesoporous materials can be controlled by the conditions of deformation, nature of polymers and crazing-promoting fluids. "Green" scenario of this process is concerned with the use ecologically safe crazing-promoting fluids as biphasic oil-in-water emulsions with high water content (> 97%). The inner structure of the mesoporous materials is presented by regular fibrils bridging the neighboring crystalline lamellae (Fig. 1). Analysis of the AFM images reveals the development of double surface relief provided by crystalline lamellae and fibrils. This surface modification makes it possible to control hydrophilic lipophilic balance of both hydrophobic and hydrophilic polymers. Despite high porosity, the mesoporous materials preserve high mechanical properties. The resultant materials can be used as membranes, sorbents, breathable materials, scaffolds, etc. Moreover, these materials can serve as host matrixes for the preparation of diverse nanocomposite materials for a broad range of practical applications.

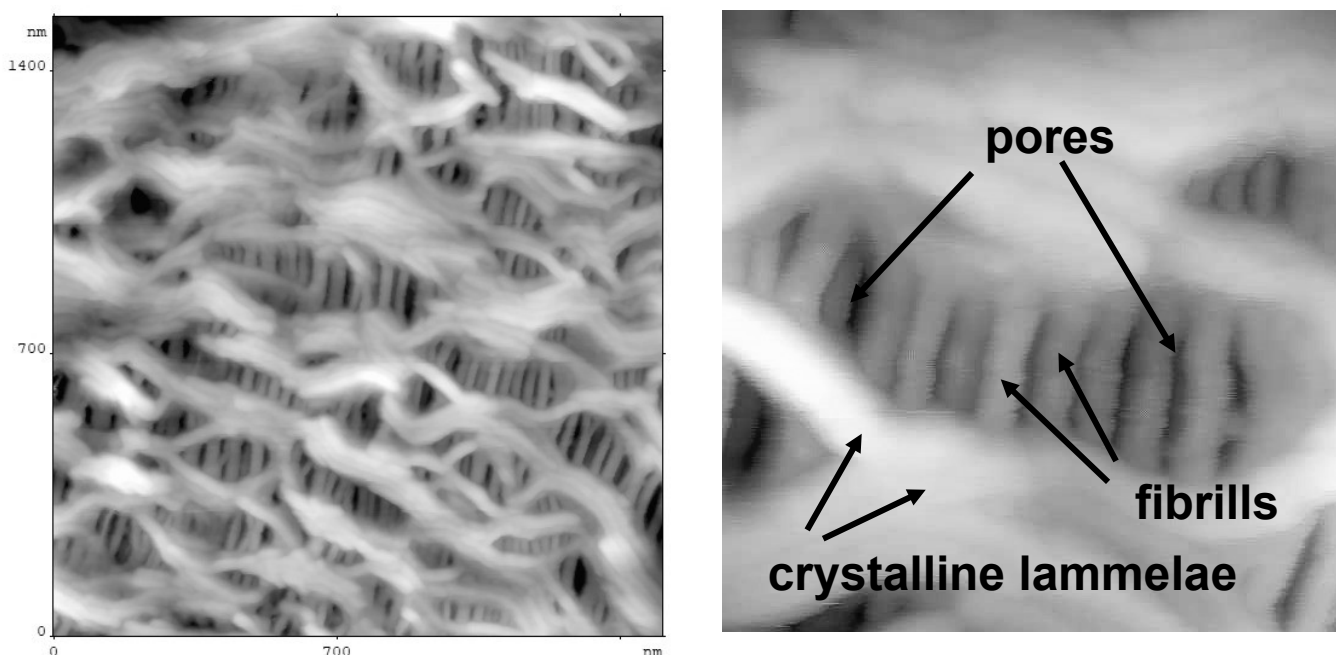


Figure 1. AFM images of the HDPE sample stretched by 200% in the O/W emulsion.

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Humic acids isolated from postpyrogenic soils of forest-steppe region: elemental and molecular composition by ^{13}C -NMR spectroscopy

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Keywords: wildfire, postpyrogenic succession, soil organic matter, ^{13}C -NMR spectroscopy, crown fire, surface fire

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Organic matter plays an important role in global carbon cycle of planet. Forest fires are one of the most significant types of disturbances on a global scale, affecting biogeochemical cycles and global warming. Pyrogenic components of soil organic matter are practically not taken into account when assessing the implications of global climate changes on SOM sequestration rate and its stability. Therefore, there are global changes in humosphere under unpredictable fires effect, and fingerprints of this key factor on the molecular structure of SOM is underestimated.

In this regard, both the amount and structural stability of SOM were investigated for postpyrogenic environments of forest-steppe. Mature unaffected by fire Psamment Entisols were studied in comparison with same soil strongly affected by surface and crown fires in 2010 in Tolyatti city. This article discusses postpyrogenic succession in case of comparing results of field works in 2010 with 2020.

The elemental composition of humic acids as well ^{13}C NMR spectra were investigated for studied humic acids. The obtained data clearly show that humic acids in soils of postfire and control plots form rather heterogeneous groups. The main difference in the degree of heterogeneity of HAs groups is the ratio of hydrogen, oxygen and carbon. There is a simultaneous decrease in the H/C and O/C atomic ratios at high temperatures, which indicates a slight loss of oxygen-containing functional groups (moreover, the loss is greater during crown fire), while the aromaticity degree of molecules increases. Changes also occur in the HAs molecules 10 years after the fires, that are manifested mainly in an increase in the content of oxygen-containing groups, which is accompanied by the oxidation of the HA molecule with the same increase in the aromaticity degree of the HA molecule.

Analysis of the ^{13}C -NMR spectra of humic acid from the studied soils made it possible to identify the ranges of chemical shifts belonging to carbon atoms of various functional groups and molecular fragments of HA. Results confirm many studies in this field that the aliphatic part is the main share in the humic acids' composition of non-affected by forest fire soils, while as a result of fires, the aromaticity degree of HAs molecules significantly increases, which is a typical zonal feature of soil organic matter in the forest-steppe zone. Moreover, for the first time ever integrated indicators of HAs molecular composition were presented for postpyrogenic soils. An analysis of integral indicators of the humic substances molecular composition showed that HAs isolated from samples taken in 2020 are generally more mature and resistant to oxidation (including microbial) compared to 2010. This indicates that periodically repeating low-intensity fires in forest-steppe ecosystems can contribute to the accumulation of stable forms of pyrogenic carbon in soils. Therefore, data obtained showed that there are significant changes in structural organization of organic matter as a result of forest fires.

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This work is dedicated to the 300th anniversary of Saint Petersburg State University.

Elemental composition of lignin preparations

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Keywords: lignin, elemental composition, soils, plant, humic acids, Transbaikalia

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The humus of the chernozems of Transbaikalia is formed by herbaceous vegetation, which, under the influence of cryoarid conditions, develops certain forms of preservation both in morphology and in the restructuring of the internal state of functional adaptation. In a short summer period, the combination of strong solar radiation with powerful ultraviolet radiation and sharp changes in night temperatures leads to internal self-regulation, to the accumulation of persistent lignins in phytocenoses, adapted both to a deficit of heat and moisture, and sometimes to their excess. Hence, a comparative analysis of the elemental composition of lignin preparations isolated from soils, vegetation, and humic acids (HA) was carried out. The elemental composition of lignin preparations is not constant, which is explained by morphological variability depending on the type of plant, its place of growth, the growing season, and the state of the environment.

The objects of study were dispersed-calcareous chernozems (Calcaric Chernozems) of the Tugnui hollow of the Selenga middle mountains and quasi-gley chernozems (Turbic Chernozem Molliglossic) of the Eravna hollow in the south of the Vitim Plateau of Western Transbaikalia. Lignin of vegetation, soils, and HAs were isolated by treating the samples with a dioxane:water = 9:1 mixture in the presence of 0.7% HCl at boiling temperature for 2 h. Before lignin isolation, the soil samples were extracted with cold water and an alcohol–benzol mixture. The elemental composition of lignins was determined on a CHNOS Vario MICRO Cube elemental analyzer (Elementar Analysen systeme, Germany).

Under the conditions of the permafrost forest-steppe, the elemental composition of lignin in vegetation, soils, and humic preparations undergoes significant changes compared to those in the steppes. In lignin of steppe chernozems, carbon is higher than in preparations isolated from permafrost analogs, which indicates that the dry climate of the steppes and uneven precipitation contribute to a better accumulation of carbon, which is a constitutional element of organic compounds. The content of hydrogen in soil lignin and humic acids from permafrost quasi-gley chernozems is higher compared to dispersed carbonate chernozems due to different natural conditions. The difference in the elemental composition of lignin macromolecules is a high amount of oxygen - 50.99 - 58.15% in quasi-gley chernozem, at O/C = 1.1-1.6. In chernozems of dispersed-carbonate oxygen content is 0.9-1.7 times less. The elemental composition of lignin in the studied soils shows that the studied samples are characterized by a high content of oxygen-containing functional groups.

The ratio H/C=0.06-0.09 confirms the aromatic character of the lignin samples isolated from all the studied samples, which indicates a high contribution of carbon to the formation of the lignin macromolecule.

It was found that regardless of the geographical location of the soils (steppe or permafrost forest-steppe), the content of carbon, hydrogen and oxygen in lignin in all samples decreases in the series: vegetation - soil - humic acids. In the lignin of dispersed carbonate chernozems, the condensation reaction occurs more actively, as evidenced by its high carbonation.

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Elemental composition of zonal and intrazonal soils in Transbaikalia

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Keywords: elemental composition, humic acids, dispersed carbonate chernozems, alluvial meadow soils

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The elemental composition of humic acids (HA) is one of the most important and stable identification characteristics of HA, which allows obtaining information about the general principles of molecular structure and the level of their condensation.

Dispersed carbonate chernozems are zonal soils. They are formed in the central part of the Tugnui hollow under a grass-forb community on loess-like loams. Alluvial meadow soils are intrazonal soils. They are located mainly in the central part of the river floodplain Selenga and develop under normal atmospheric-ground moisture under meadow plant associations.

Preparations of humic acids (HA) were isolated from the humus horizons of the studied soils (layer 0-20 cm) by extraction with 0.1 N. NaOH according to the method of D.S. Orlova - L.A. Grishina. The elemental composition of HA was studied on a CHNS/O Perkin Elmer 2400 Series II automatic elemental analyzer.

The studied soil types have distinctive features in the elemental composition of humic acids. A high carbon content was found in the HAs of alluvial meadow soils (C content = 43.9 at. %), which indicates a significant proportion of aromatic fragments of the nucleus in their macromolecules. In the HAs of dispersed carbonate chernozems, the condensation reaction occurs less actively (C content = 38.7 at.%). It would seem that the change in the conditions of soil formation in the direction of a gradual increase in temperature and a decrease in humidity is associated with an increase in the condensation of HA. But on the chernozem, the direction of the process is influenced not only by changes in climatic conditions, but also by chemical and biological factors: the reaction of the environment, the composition and amount of organic residues. In the HA preparations of dispersed carbonate chernozems, the hydrogen content was 33 at.%, which is closer to that of the chernozems of the ER. In alluvial meadow soils, the indicator is slightly lower - 32.4 at.%.

The oxygen content in the studied HAs is 25.2 at.% in dispersed carbonate chernozems. In the HA of alluvial meadow soils, the indicator is lower - 21.2. Due to the fact that the oxygen-containing functional groups of organic compounds determine the hydrophilic properties of molecules, they are concentrated in the peripheral part of the HS. Thus, the proportion of aliphatic structures in the HAs of dispersed carbonate chernozems is higher than in alluvial meadow soils.

The degree of benzoid in alluvial meadow soil (37%) is 2.2 times higher than in chernozems. In the studied HAs, the proportion of nitrogen atoms is 2.5%. The value of the atomic ratio H:C decreased with increasing degree of humification, i.e. the more HA is formed in the soil, the more they are condensed. In both studied samples, H:C is less than unity; in the alluvial meadow soil, the ratio is narrower and amounts to 0.73.

The data on the elemental composition of alluvial meadow humic acids indicate a high conservation of humus and, consequently, soil resistance to various influences. The elemental composition of humic acids in chernozems indicates that HAs with a low carbon content and an increased oxygen content are formed in cold areas. In dispersed calcareous chernozems, HAs are weakly condensed compared to those from alluvial meadow soils, which are more enriched in cyclic fragments.

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Thermal and microbiological stability of humic acid adsorbed on various clay minerals

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Keywords: fractionation, basal respiration, TGA-DSC, organo-mineral complexes

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Nowadays thermogravimetry (TG) and differential scanning calorimetry (DSC) are frequently employed techniques for assessing the stability of soil organic matter (SOM). However, the relationship between the biological and thermal stability of SOM is still not clear and existing results are often contradictive and ambiguous.

The main purpose of this research is to study the thermal characteristics of humic acid sorbed on bentonite, kaolinite and muscovite and their relationship with the resistance of sorbed humic acid to microbiological degradation.

Humic acid (HA) isolated from leonardite was adsorbed at pH = 4 on clay fractions of bentonite, kaolinite, and muscovite. After drying the samples, the experiment was repeated twice more. Thermal characteristics of organo-mineral complexes were measured using TGA-DSC and in order to assess basal respiration of samples gas chromatography was applied.

The amount of sorbed HA per unit area decreased in the order kaolinite > bentonite > muscovite. This finding would seem to show that kaolinite is the most effective sorbent due to hydrophobic interactions between HA molecules and uncharged silanol surface of kaolinite. The C/N ratio of HA significantly decreased after its sorption, but in different extent for different minerals, which may indicate HA fractionation by mineral-specific mechanisms.

After the sorption experiment, all organo-mineral complexes exhibit two exothermic effects Exo1 (300-350°C) and Exo2 (350-450°C), originated from the thermal destruction of HA sorbed on the surface of minerals. Exo 1 and Exo2 significantly shifted towards higher temperatures after HA sorption which is a possible indication of HA thermal stability increase due to interaction with clay minerals. The thermal stability of sorbed HA decreased in the order bentonite > muscovite > kaolinite and increased in the order 1 cycle < 2 cycle < 3 cycle of sorption.

For kaolinite and muscovite, the value of basal respiration appeared to be significantly higher compared to the corresponding pure minerals, which can be explained by the destruction of sorbed organic matter by microorganisms during incubation. Interestingly, a different pattern was revealed for the organo-mineral complex of bentonite: the intensity of basal respiration remains minimal throughout the entire incubation experiment. We are assuming that the reason for such finding is inaccessibility of organic matter for destruction by microorganisms. According to the increase in the intensity of basal respiration, the minerals are arranged in the order bentonite < kaolinite < muscovite.

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Extraction in alkali without nitrogen significantly alters physico-chemical properties of humic acids from compost

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Keywords: humic acids, alkaline extraction, oxidation, functional group content

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Humic acids (HA) are formed during biotransformation of organic residues in the diversity of the environments and represent polydisperse and heterogeneous compounds enriched in polar functional groups. Alkaline extraction with subsequent acidification is traditional way to obtain HA preparations from soils and sediments. However, alkaline treatment is criticized for the possibility of creating artifacts, such as oxidation of phenolic constituents, hydrolysis of ester groups, etc. International Humic Substances Society has recommended to perform alkaline extraction under the atmosphere of inert gas (e.g. nitrogen). However, extraction under N₂ is rarely used in humus chemistry and is ignored when HA are obtained for commercial purposes. Compost is one of the sources of commercial HA. It represents relatively fresh organic material if compare with peat, coal and soil humus. The aim of this work was to study the physico-chemical properties of HA from compost extracted by 0.1 M NaOH in absence or presence of oxygen.

Compost prepared from leaves and grass with the time of composting of about 10 years was used as a source of the HA. Compost material was air dried and passed through 2 mm sieve. Extraction of the HA was performed by three consecutive treatments with 0.1 M NaOH (1 hour each) under the N₂ atmosphere (HA-N₂) or without it (HA-O₂). The pH of each extract was adjusted to 7.0, extracts were pooled and purified by membrane filtration (0.22 μm). HA were then obtained by acidification and air-dried.

The infrared spectra of the preparations were identical as well as their molecular weight distributions on Sephadex-G-75 gel. However HA-O₂ was characterized by higher O:C and C:N ratios (0.6 and 13.6 respectively vs 0.5 and 12.5 in HA-N₂), higher optical density at 465 nm ($E^{0.001\%}$ 0.04 vs 0.03 in HA-N₂), higher E4/E6 ratio (5.5 vs 4.7 in HA-N₂), lower contribution of CH_n fragments and higher contribution of CH_n-O ones (17.3 and 21.2% respectively vs 22.9% and 16.7% in HA-N₂). The content of OC-O(N) (acetale) fragments was also higher in HA-O₂. The content of functional groups titrated in the regions of pH 5-8 and 8-10 were 4 and 2 times higher in the HA-O₂ suggesting an increase in the content of COOH-groups and in the content of OH-groups in this preparation.

Thus, we have shown that alkaline extraction of HA from relatively fresh humic material (compost) significantly alters physico-chemical properties of HA if no N₂ is applied. Most significant changes occur in the functional groups content and in the content of aliphatic O-containing fragments. We hypothesize that changes occur in the functional properties of the HA as well. Alkaline treatment under O₂ may enhance HA complexing and antioxidant capabilities and can be used as targeted modification of the HA preparations. We are testing this hypothesis in our current research.

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Synthesis of magnetic sorbents on the base of peat for purification of polluted waters from heavy metals

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Keywords: heavy metals, sorption, magnetic particles, peat

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Heavy metals are widespread pollutants of aquatic environment. The main sources are metallurgic plants, mining industry, coal mining industry. It is essential to have efficient tools for preventing migration of heavy metals upon their entering into natural water bodies. Sorbents are among the most widely used materials used for this purpose. In this work, we propose to prepare "green" magnetic sorbent based on peat and embedded into it ferromagnetic particles.

For preparing this sorbent we treated natural peat with mixture of iron(II,III) salts to yield magnetic particles of magnetite. The magnetite allows to remove the sorbent after application by applying an external magnetic field. The phase composition of the iron oxides within the modified sorbents was determined with a use of XRD spectroscopy

We conducted a series of model experiments on the sorption of Cu^{2+} and Ni^{2+} ions upon their simultaneous presence in mixture. We ran both kinetic and sorption experiments.

For this purpose, we prepared solutions of copper and nickel with different concentrations. Then, the known weights of the prepared sorbents were added, the tubes were topped with screw tops and placed onto the overhead shaker. For kinetic studies the samples were taken at the time exposure of 10, 20, 40, 60 and 120 min. Initial and final concentrations of Cu and Ni were measured using ICP AES. The difference between the concentrations was used for calculation of the sorption efficiency of the native and modified peat toward Cu and Ni.

The efficiency of sorption of the non-modified peat sample with regard to Cu^{2+} was 35 mg/g. At the same time, the efficiency of magnetite modified peat sample increased and accounted for 60 mg/g. For Ni^{2+} ions the modification of peat with magnetite did not play a substantial role: the sorption capacity of non-modified peat was 25 mg/g, whereas for the modified peat it accounted for 23 mg/g. It should be noted that the pure magnetite (Fe_3O_4) did not show sorption affinity with regard to nickel and copper, which was also found out during the experiments.

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Possibilities of using different approaches to assessing the resistance of organic matter of fallow soils to mineralization

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Keywords: fallow soils, labile organic matter, resistance to oxidation

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Fallow lands, the area of which in Russia can be up to 40 million hectares, are considered both a very significant potential sink and a source (with a change in the land use system) of CO₂ in the soil-plant-atmosphere system. The return of fallow lands to agricultural circulation can lead to an abrupt emission of CO₂ into the Earth's atmosphere due to the mineralization of a significant part of the organic matter (OM) accumulated under fallow vegetation over the past decades. Forecasting the scale of CO₂ emissions from fallow soils when changing the land use system is an actual task, the solution of which should consider both the quantitative parameters of the accumulation of pollutants under fallows and their potential susceptibility to mineralization.

The purpose of the work is to study the effectiveness of various approaches to the assessment of stable and labile components of the pool of OM of fallow soils, in the aspect of assessing the susceptibility to microbial destruction. As the object of the study, a fallow site aged 15-20 years was used, on which 3 sites with low, medium and high reserves of OM were allocated. At each of the sites, 2 pits were laid, for layer-by-layer (after 5 cm) sampling from the old arable horizon. In the samples, the total content of OM and soluble OM in a mixture of Na₄P₂O₇+NaOH was determined. Also, the amount of OM oxidized 0.167 mol/l K₂Cr₂O₇ in 5 N H₂SO₄ solution (easily oxidized fraction of OM according to the Khan method [1]) was determined.

A significant differentiation of the old arable horizon was revealed both in terms of the total content of OM and in terms of the content of alkali-soluble and easily oxidized OM. At the same time, when calculating the carbon content of the alkali-soluble and easily oxidized parts of the OM as a percentage of the total organic carbon, the patterns of differentiation of the old arable horizon are different. The proportion of alkali-soluble carbon in the total pool of organic carbon in the old arable horizon increases, from 44.3% in the 0-5 cm layer to 53.0% in its lower part (deeper than 15 cm). According to the content of easily oxidized carbon, there is a decrease in its proportion in the total pool of organic carbon, from 56.0% in the 0-5 cm layer to 43.6% in its lower part. The difference in the patterns of differentiation of the old arable horizon in terms of the content of labile OM is associated with the fact that a part of detritus, insoluble in alkali, also enters the composition of easily oxidized OM.

It can be concluded that the use of the stepwise oxidation method to assess the potential susceptibility of OM of fallow soils to mineralization is a more informative indicator, since along with taking into account the labile part of OM, it allows objectively taking into account its detritus component, potentially susceptible to oxidative degradation.

Possibilities of using different approaches to assessing the resistance of organic matter of fallow soils to mineralization.

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Humic acids. Age is not important

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Keywords: humic acids, peat, bog, Western Siberia

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A representative ombrotrophic peat bog was chosen as the object of study. Peat deposit 220 cm deep. The peat deposition dates back to 5200 calibrated years [1]. Peat samples were taken every 10 cm to the entire depth of the peat deposit. The deposit is homogeneous, composed of high-moor sphagnum peat to a depth of 190 cm. In the bottom layer (190-220 cm) there is grass-sphagnum and grass peat.

There is a low ash content ash in the studied subject (from 1.00 to 1.80% with an average index 1.40 ± 0.22). Increase in decomposition up to 60 cm $\leq 5\%$, up to 140 cm - 10%. The ash content increases to the bottom layer and demonstrates maximum up to 50%. Humic acids were isolated by alkaline extraction [2] and characterized by a number of analyzes (elemental composition, UV, IR spectroscopy).

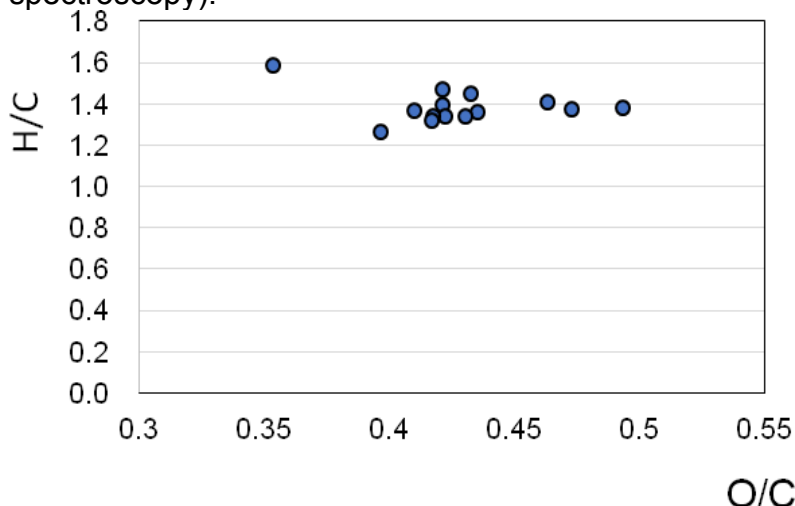


Figure 1 - Van Crevelen distribution of humic acids in an ombrotrophic peat bog.

A rather high C/N ratio (from 17.1 to 23.8) of humic acids demonstrates reduce in the degree of oxidation during peat decomposition processes. The dependence of O/C on H/C, recorded in the Van Crevelen diagram, is in the region above 1.2 of the H/C indicators. This range corresponds to weak oxidation of humic acid in peats with low nutritional maturity. The close arrangement of points on the diagram indicates the homogeneous composition of humic acids. Despite the fact that the process of peat genesis proceeded for several thousand years, the composition of humic acids changed insignificantly. This circumstance can be explained by the natural conditions of peat accumulation in the region.

Humic acid is a valuable raw material for large-scale scientific and industrial research.

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Flame-retardant hybrid organo-inorganic nanocomposite polymeric materials based on high-density polyethylene

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Keywords: nanocomposite, high-density polyethylene

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The development of flame-retardant hybrid nanocomposite materials presents the topical scientific direction of modern materials science which is oriented towards the search for sustainable low-cost and eco friendly approaches. This work offers a new method for the preparation of flame retardant polymeric materials based on polyolefins containing ecologically safe aluminum hydroxide as the fire-suppressing agent via the strategy of environmental crazing. This approach involves two stages: incorporation of precursors (as aluminum salts) into mesoporous matrixes and subsequent *in situ* hydrolysis in the basic medium to aluminum hydroxide (AH) within the confined space of mesopores. The optimal conditions for *in situ* hydrolysis were studied. TEM observations show that AH is uniformly distributed within the host matrix as spherical or needle-like particles with dimensions below 15 nm. The resultant nanocomposite materials with low content of AH (below 30%) shows high flame retardancy and are characterized by high mechanical properties which are by 15020% lower than those of the pristine polymer.

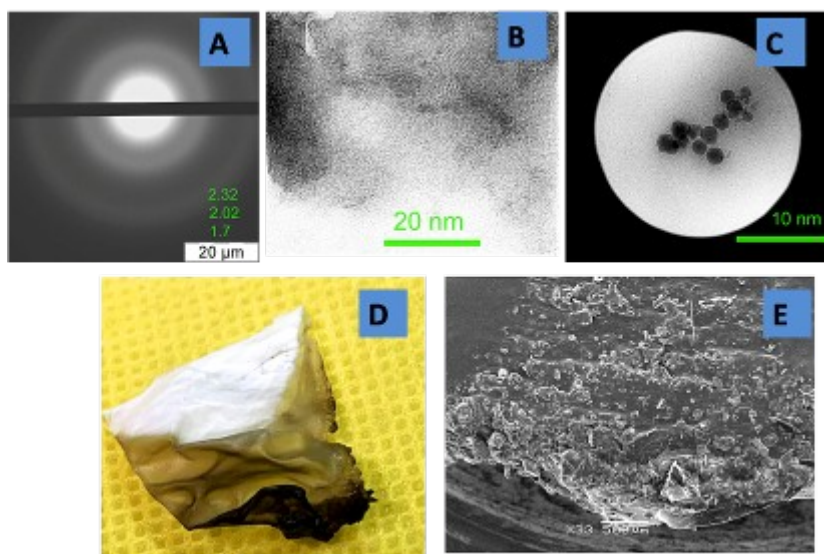


Figure 1. (A) Electron diffractogram of AH in mesoporous HDPE matrix, (B, C) TEM images of AH/HDPE, (D) carbonization, and € SEM images of the carbonized sample (coke).

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Enzymatic activity and humic acids of soils in the Selenga river delta

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Keywords: enzymatic activity, humic acids, humus, alluvial meadow and meadow-swamp soils

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Over the past decades, the soil cover of the region under study has been subjected to degradation influences. Against this background, the need to study the enzymatic activities in the soil and their relationship with the factors of soil formation is increasing. Unfortunately, insufficient data on the state of enzymatic activity and humus state have been obtained in the described region. In a comparative aspect, the enzyme activity of the near-terrestrial and near-river floodplains of a number of islands in the Selenga delta has been studied. Available literature data on the humus composition and biological activity refer to virgin soils.

According to the natural zoning, our study area belongs to the delta meadow-swamp and forest-steppe region. The Selenginsky deltaic area is unique within the Baikal region. It is characterized by peculiar processes and phenomena, the combination of which leads to the formation of unique landscape conditions and intrazonal ecosystems. Most of this area is a riverine plain of the Selenga River, indented by numerous channels and oxbow lakes. An important role of the modern delta of this river - the main tributary of the unique Lake Baikal as a natural natural filter. Lake Baikal itself was inscribed on the UNESCO World Heritage List in 1996.

The soil cover and ecosystems of the territory under study develop under the conditions of the continental climate of Eastern Siberia, somewhat transformed by the influence of Lake Baikal, especially during the spring-summer period. In addition, the vegetation cover of biotopes is influenced by uncontrolled pasture use, which reflected on the humus state of soils. Hence, the relevance of the topic of the research under consideration is predetermined by the importance of these natural objects.

This work deals with the search for more sensitive methods that will help to determine the perceptions of the enzymatic activity in the creation of soil fertility. The aim of the research was to study the chemical structure of humic acids (HAs), to identify the features of biological activity, and to determine the humus state of the riverside plain adjacent to the Lake Baikal water area.

The study of soils (gray forest, alluvial meadow and meadow-bog) showed that humus content increases from floodplain-meadow, meadow-bog to gray forest. In the same direction, the enzymatic activity also increases, which is characterized by the highest correlation coefficient [1]. Thus, the nuclear part is equally developed in the HA molecules of alluvial meadow and meadow-swamp soils, indicating greater condensation and their maturity, which allows us to predict the resistance of humus substances to stresses.

Acknowledgements; the work was carried out on the topic of the state order № 121030100228-4.

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Characteristics of soil organic matter in the Kola Subarctic

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Keywords: elemental composition of humic acids, infrared spectra, optical properties, stable isotopes of humus carbon

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Global climate warming primarily affects the processes of increasing carbon emissions in the ecosystems of the Arctic and Subarctic. Humic substances in the ecosystems of the Kola Subarctic have been studied in illuvial-humus-ferruginous Podburs (Turbic, Spodic & Folic Cryosols) under the lichen-shrub tundra. Soil pits were laid in the trough valleys of the Khibiny Mountains: Kukisvumchorr, Poachvumchorr, Takhtarvumchorr.

Soil organic matter was studied using the methods of analysis of the group and fractional composition of humus, analysis of the elemental composition and infrared spectrometry of humic acid preparations, analysis of stable isotopes of humus carbon ($\delta^{13}\text{C}$).

The content of organic carbon in the organic horizons of the studied soils is naturally high and can reach 15–20 %. This is due to the slow rate of mineralization of organic matter. On the other hand, the soils of the mountain-tundra belt, due to the well-developed process of alpha-humus illuviation, are characterized by a deep humus content of the mineral profile. The content of humus in mineral horizons can reach 5–6 %.

The nature of the distribution of humus along the soil profile is regressive-accumulative. The type of humus in the surface horizons is fulvate-humate and humate-fulvate, and in the mineral horizons it is fulvate. Very low and low values of the extinction coefficient, low values of humification indicate low aromaticity of humic acids and correspond to a short period of biological activity.

The ratio of the processes of humification and mineralization of organic matter in the conditions of the mountain tundra causes the convergence of the elemental composition of humic and fulvic acids. They are characterized by a low carbon content, a high degree of oxidation (O/C 0.7–1), a significant substitution of aromatic rings and the predominance of aliphatic chains in the construction of the molecule (H/C 1.3–2.2).

In accordance with the short period of biological activity, the enrichment of humus with nitrogen in both organic and mineral horizons is low and very low: the C/N ratio varies from 8 to 36. The C/N ratio in humic acid preparations ranges from 12.9 to 16.1.

The carbon isotopic composition of organic matter ($\delta^{13}\text{C}$) shows a range of values from -32.5 ‰ to -23.9 ‰, diagnosing the predominance of plants of the C3 type of photosynthesis and the high contrast of natural conditions: extremely cold and humid periods and cold and dry episodes.

The infrared spectra of humic acids reveal a significant proportion of oxygen-containing groups of various origins, methyl and methylene groups of paraffin chains and peptide groups characteristic of humic acids with a developed peripheral part.

Thus, the humus horizons of the soils of the lichen-dwarf shrub tundra are distinguished by the coarse nature of mor-type humus, its humate-fulvate composition, low extinction coefficients, a significant content of the Pg-fraction, low enrichment of humus with nitrogen, and a developed peripheral part of humic acid molecules with a small proportion of benzenoid structures. Similar properties of organic matter molecules reflect the conditions of increased moisture in the mountain tundra with a short period of biological activity.

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Content of free radicals in humic substances from taiga and tundra soils

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Keywords: humic acids, fulvic acids, paramagnetic activity, soil hydromorphism

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Numerous experiments studying soil organic matter have proved an immense role of free radicals (FRs) in their semiquinoid form in biochemical processes. The organic radicals, with their stability associated with the delocalization of an unpaired electron along the system of conjugated bonds of the polynuclear structure of humic substance (HSs) molecules, serve to determine the ability to be naturally paramagnetic for HSs. The analyzed results of researchers' work showed that the FR concentration in the HSs structure is strongly affected both by various zonal bioclimatic factors, and human activity.

The working hypothesis of our studies was that changes in the composition of incoming plant residues and hydrothermal conditions have an effect on soil humification rate and the content of free radicals (FRs) in the structure of humic (HAs) and fulvic acids (FAs). The purpose of this study is to examine the paramagnetic properties of HA and FA samples obtained from taiga and tundra soils – virgin and arable ones.

The research embraced six key soils in taiga (virgin Eutric Albic Histic Retisol, and two Eutric Albic Retisols – virgin and arable ones) and tundra zones (virgin Histic Cryosol, and two Gleyic Stagnosols – virgin and arable ones).

The evaluation of the paramagnetic activity of HS samples showed that the FR concentration in HA samples is several times higher than the one in FAs. It is explained by a higher content of aromatic and other polyconjugated structures in HA molecules, on which unpaired electrons can delocalize. PCA analysis of HSs shows a statistical relationship between $c(\text{FRs})$ and the proportion of aromatic fragments, which indicates the active participation of these structural units in the radical polymerization and complexation reactions.

The value of the g-factor for FA samples is averagely higher than the one for HAs. This indicates a greater shift of the electron density of the unpaired electron in the FA structure towards the oxygen atom due to its spin-orbit interaction with oxygen-containing functional groups, which demonstrate a higher content in FAs compared to HAs. A statistically significant relationship was established between the value of the g-factor and the content of O-CH₃ groups in FAs, which can be associated with lignin fragments.

An increase in hydromorphism in Retisols leads to a higher concentration of FRs in the molecular structure of HAs. It may be explained by a differing nature of plant residues entering and participating in the process of humus formation.

A higher moisture content in tundra soils leads to a lower content of free radicals in the structure of HSs. The revealed variations in the changing paramagnetic activity of HSs in virgin taiga and tundra soils following an increase in hydromorphism can be explained by the different nature of plant residues incoming and participating in the humus formation process.

In contrast to HAs, no clear regularity is observed for FAs in their paramagnetic activity when analyzing the profiles of the studied soils. It may be associated with a good solubility of FAs and their subsequent mixing in the course of downward migration across the profile.

The agricultural use of taiga and tundra soils reduces the paramagnetic activity of HSs by over 20% compared to their virgin counterparts, which is due to a higher rate of transformation of plant precursors and prohumus substances in the arable horizon.

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Two-dimensional correlation spectroscopy as a method to construct absorption spectra of low quantities of humates

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Keywords: humate, low quantities of humates, two-dimensional correlation spectroscopy, Beer-Lambert–Bouguer law

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Two-dimensional correlation spectroscopy (2D-COS) is a chemometric technique used in many fields of spectroscopy and applied to many types of objects. Polymers and bioorganic compounds are among the most studied ones. The mathematical apparatus of 2D-COS has been developing rapidly in recent years and can be used to improve the analytical characteristics of many widely used methods of analysis [1]. Still, its use for quantification is rather scarce.

UV-visible absorption spectroscopy is a conventional analysis method for humic substances (HS). However, natural waters contain very low quantities of HS, and even the application of cells with optical path lengths of 10 cm in UV-vis absorption spectroscopy does not always provide good results. Therefore, obtaining HS absorption spectra in UV and visible regions with high sensitivity is an essential task [2].

This study is devoted to the use of 2D-COS to reconstruct absorption spectra of low amounts of HS using the concentration as an external perturbation in correlation maps. For this purpose, an equation relating the parameters of the homospectral synchronous correlation matrix with the parameters of the Beer-Lambert law, and molar absorptivity ε was derived,

where n is the number of solutions in a test series, Δc is the concentration step for a given series of solutions used to build the correlation matrix, I_λ is the auto-peak intensity on the synchronous correlation map, and l is an optical path length. The equation was tested both on model systems, dyes (phenol red and bromothymol blue), and aqueous solutions of humates as a more complex object.

It was shown that the application of 2D-COS gives a significantly higher signal-to-noise ratio in assessing molar absorptivities for submicromolar concentrations compared to averaging the molar absorptivities from a set of spectra. Similar results are obtained for HS, which may allow increasing the information content of the spectral measurements for analysis without changing the hardware of the method.

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Spectroscopic analysis of chernozem natural organic matter with preliminary separation using polycarbonate track-etched membranes

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Keywords: natural organic matter, fractionation, ultrafiltration, track-etched membranes, fluorescence spectrometry, infrared spectrometry, inductively-coupled atomic emission spectrometry

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Natural organic matter (NOM) is a highly complex mixture containing thousands of different organic molecules. Due to its origin, NOM always presents in the form of particles of various sizes, thus membrane filtration is a promising fractionation method to simplify this complex system. It allows for studying particles ranging in size from 10 Da to several hundred micrometres [1]. Nowadays, there is a growing interest in using this separation method for organic matter, predominantly dissolved organic matter (DOM) of water.

This study used sequential filtration through polycarbonate track-etched membranes with pore sizes from 0.01 to 10 µm to fractionate water-extractable organic matter (WEOM, DOM obtained by extracting a soil mass/volume with an aqueous solution) from typical chernozem obtained at room temperature water extraction. The filter cut-off to distinguish particulate organic matter (POM) and DOM is somewhat inconsistent, but 0.2 to 0.6 µm pore size is usually considered the boundary between them [2]. Therefore, membrane filters of defined pore size used in this work cover not only DOM but also POM. The main focus of the study is on the fractions obtained by washout from the respective membrane filters.

In this study, we demonstrated the possibility to analyze WEOM narrow fractions with direct sputtering ICP-AES without decomposition. It was established that the trends in the elements' concentration change upon passing from large to smaller particles are reproduced regardless of the membrane material. To perform the functional-group analysis the FTIR-ATR spectra of the fractions were obtained. For this, a method of drying a drop on the surface of the ATR crystal was proposed, which makes it possible to use a very small amount of the fractions. This approach can be used for the characterization of narrow fractions with a particle size greater than 1 µm for this type of track-etched membranes. By FTIR and ICP-AES, it can be seen that all WEOM fractions contain clay minerals with variable compositions. To analyze fluorescent organic components, excitation-emission matrices were obtained, and specific regions were integrated. All the fractions contain humic-, tyrosine-, and tryptophan-like substances with variable compositions. The proposed concept is a logical extension of sieve cascade fractionation that involves particles with a size of less than 10 µm and expands knowledge of its chemical composition and its association with mineral phases.

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Estimation of carbon stocks and stabilization rates of organic matter in soils of the "Ladoga" carbon polygon

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Keywords: SOM quality; HAs; ¹³C CP/MAS spectroscopy

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Soil organic matter (SOM) is a major component of the pedosphere. Due to the climate change, SOM may be subject to active biodegradation processes, land transformation and become a major source of greenhouse gases in the atmosphere. However, the overall effect of climate change on SOM content varies by region and soil type. Climate change can have catastrophic consequences for SOM, soils, and landscapes in general. Nowadays, different natural zones of our planet are subject to landscape degradation. Sustainable soil management aimed at increasing the content of SOM can increase the process of carbon sequestration, thereby contributing to the mitigation of climate change. Establishment of a network of carbon polygons for monitoring climatic processes as well as emission and deposition of carbon compounds in various ecosystems is one of the priority tasks in the field of climate and biosphere conservation as a whole. The most important area of work on the carbon polygon is the problem of assessing the reservoir and trends of stabilization of organic matter in different natural zones and ecosystems. By stabilization we mean the process by which SOM is formed resistant to biotic and abiotic influences and leads to an increase in the time of its turnover in the environment. The opposite of this process is destabilization, during which SOM components become available for use by microorganisms. This work presents the results of the study of the soil sections of the "Ladoga" carbon polygon. The Ladoga carbon polygon will be located in Voeykovo village, the south of Vsevolozhsk district of Leningrad region of Russia. The area is represented by fluvio-glacial eskers relief form, in front of eskers uplands the Holocene lowland of Ladoga lake is located which is merged with current Ladoga waters edge. In the location the Folic Podzol (Arenic) and Fibric Histosol were considered. These soils are widely represented in the southern taiga subzone and can be used as benchmark monitoring soil sections. As a result of the analysis of chemical parameters it was revealed that Fibric Histosol and Folic Podzol (Arenic) are characterized by an acidic reaction of pH, pronounced podsolization and peat accumulation. In Fibric Histosol stores a significant amount of SOM up to 42.95 kgC/m². In the analyzing of the molecular composition of humic acids (HAs) extracted from the studied soils, it was found that HAs from Fibric Histosol were characterized by a relatively high content of aromatic structural fragments (51.28-56.83 %), while in Folic Podzol (Arenic) aliphatic structural fragments prevailed (56.23-63.08 %). The increase of aromatic structural fragments in the HAs composition indicates the stabilization of the SOM and an increase its turnover time in the environment. The creation of a monitoring network will make it possible to identify the main trends of SOM accumulation, to determine the conditions under which accumulation occurs, and to calculate its contribution to climate change on the planet.

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Fractal clusters is a form of existence of humic substances in soils

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Keywords: humic substances, structure of humic substances, fractal clusters

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Studying humic acid solutions, Osterberg showed using the small-angle neutron scattering method that primary humic acid particles with a size of 2-10 nm form fractal clusters with a size of 100-200 nm in solutions (1). Fedotov found that in soils, as well as in solutions of humic acids, there are also primary particles of humic substances of 2-10 nm in size, forming fractal clusters of 100-200 nm in size (2), which in turn, together with mineral particles, form soil gels.

Early studies have shown that individual primary particles of humic acid exist in solution only at concentrations below 10 mg/l (3). At high concentrations, they combine to form fractal clusters. Therefore, the only form of humic substances existence in soils should be precisely fractal clusters.

The purpose of the work: to verify the assumption of the existence of humic substances of soils in the form of fractal clusters.

The studies were carried out using a JEOL-6060A scanning electron microscope with a tungsten cathode at an accelerating voltage of 2-5 kW.

For the study, the following were used: humic acids and fulvic acids isolated from sod-podzolic soil; humates isolated from brown coal and peat; soil samples taken from the upper horizons different soils; dust, collected in the Kursk region, Moscow and Rostov region.

The conducted experiments show that F-clusters are found in all studied samples of soils, humates, fulvic and humic acids, have characteristic sizes and shapes (Figure 1).

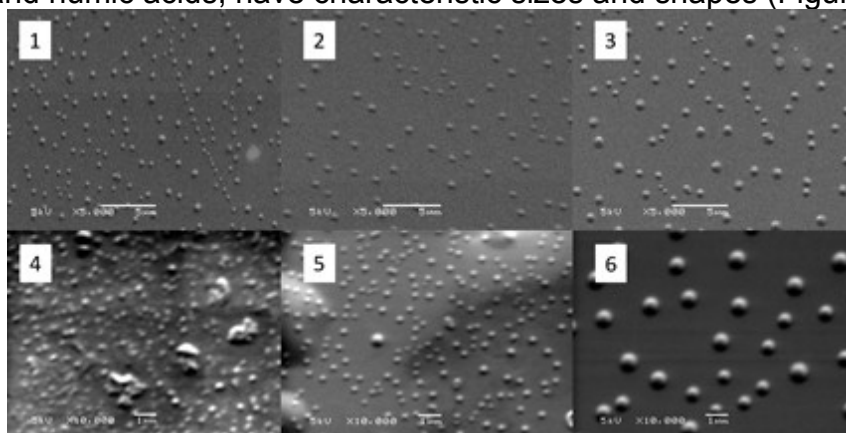


Figure 1. Fractal clusters isolated from soils: 1-sod-podzolic, 2-gray forest, 3-chernozem, 4-red earth, 5-brown earth, 6-mountain meadow.

Fractal clusters are the form of existence of humic substances in soils. Thus, they must perform certain functions in soils, being responsible for many of their properties. The study of fractal cluster organization of soils and its relationship with soil properties is a promising and underestimated direction in the study of soils.

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Chemical composition of the different fractions humic substances of sapropel in the Pskov region

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Keywords: sapropel, humic acids, fulvic acids, organic substances

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Using LC-MS analysis, the features of chemical composition of the main liquid fractions (humic acids - HA and fulvic acids - FA), included in the humic substances of sapropel in the Pskov region, were studied in detail.

Table 1. Content of carboxylic acids in different liquid fractions of sapropel, $\mu\text{g l}^{-1}$

Component	HA (22-24%)	FA (10-12%)
Fumaric acid	0.22	0.00
Acetic acid	14.70	31.40
Oxalic acid	0.84	0.00
Wine acid	0.30	0.00
Lemon acid	1.12	0.00
Succinic acid	0.22	0.00
Gallic acid	0.01	0.00
Apple acid	1.29	0.00
Adipic acid	0.06	0.00
Methionine	0.27	0.60
Valine	16.28	3.14
Histidine	4.15	0.50
Glycine	0.42	1.00
Glutamine	10.39	4.82
Serene	7.27	18.54
Leucine	9.67	0.52
iso-Leucine	7.00	0.10
Asparagine	4.27	0.27
Alanine	0.39	1.00
Tryptophan	0.14	0.01
Threonine	7.00	0.44
Lysine	17.00	0.16
Arginine	15.00	10.00
Sirin	4.00	0.00
Tyrosine	0.08	0.17
Threonine	0.29	0.07
Proline	0.32	0.10
Cysteine	0.09	0.01
Phenylalanine	0.21	0.41
Ferulic acid	16.10	29.00
Para-coumaric	13.77	35.73
Syringic acid	15.58	19.42
Vanillin acid	18.10	60.15
4-hydroxyphenylacetic	0.00	10.87
Σ	186.55	228.43

Photoactive hybrid organo-inorganic nanocomposite polymeric materials containing quantum dots

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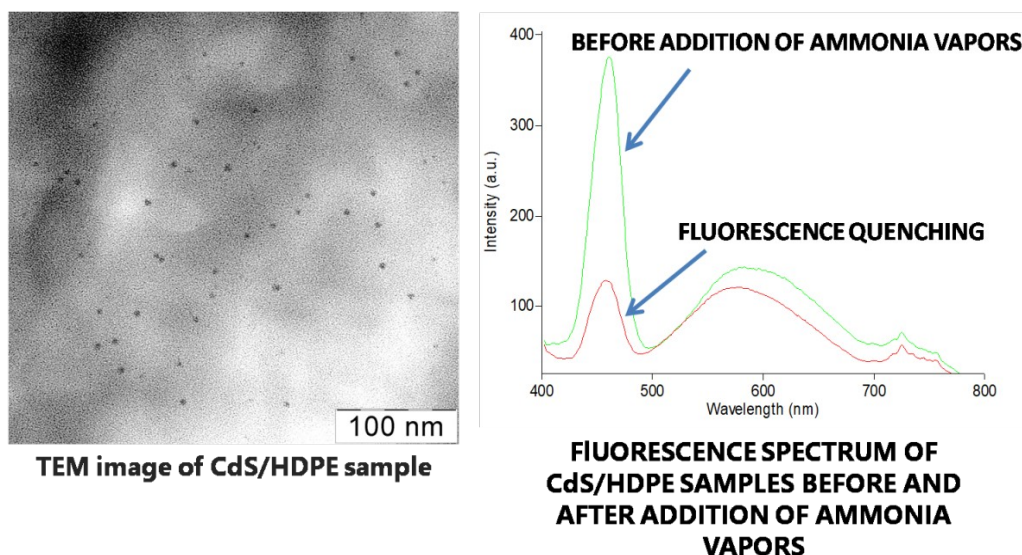
Keywords: nanocomposite, quantum dots

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Ecological problems present the most pressing issues of XXI century and demand the development of innovative high-performance sustainable sensors for safety control and ecological monitoring. The basic requirements to the sensing materials are concerned with their efficient and quick response towards the action of analytes and convenient practical use.

This work addresses the new approach for the preparation of high-performance optochemical sensors based on mesoporous polymeric matrices and inorganic quantum dots. The mesoporous HDPE and PTFE matrices were fabricated using the strategy of environmental crazing. The mesoporous polymeric matrixes were loaded with photoactive additives using the protocols of forced and passive wet impregnation. Quantum dots based on CdS were shown to be uniformly distributed within the polymer matrix as nanoparticles with mean dimensions of 3 nm without any aggregation. The resultant optochemical materials show high fluorescence intensity as presented by two well-resolved peaks and experience fluorescence quenching when subjected to the action of ammonia vapors. Upon removal of ammonia vapors, the fluorescence peaks are fully restored, and this behavior is reproduced in many cycles.

Hence, the proposed approach allows preparation of new optochemical nanocomposite sensors with quick and intensive response towards analytes which can be used for the safety control.



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Study of the humification of wood processing industry waste

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Keywords: bark and wood waste, humic acids, ¹³C NMR, IR spectroscopy

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Significant amounts of bark and wood waste (BWW) have been accumulated in the Republic of Komi as a result of the activities of the wood processing industry, which have a negative impact on the environment. Existing technologies allow converting BWW and their decomposition products into a bioresource, for example, humic preparations. On the other hand, BWW storage facilities are unique objects where it is possible to analyze the process of transformation of natural biopolymers under conditions close to natural ones. The study site is located in the vicinity of Syktyvkar and is a BWW storage site with an area of 8-11 ha and a depth of up to 27 m. BWW are represented by weakly decomposed fragments of wood chips and sawdust in the upper part of the profile to a depth of 3 m, the degree of decomposition of which is higher in the underlying layers up to the base of the massif. It has been established that the formation of humic acids (HA) occurs already at the initial stage of BWW decomposition in the two upper layers, which is confirmed by the HA content of 0.8 and 1.3%, respectively. The data of the elemental composition and molecular absorption spectra show the primary stage of the formation of HAs from wood raw materials, in which there is an increase in ring conjugation (increase in the degree of condensation) diagnosed by an intense decrease in the atomic ratio $x(H) : x(C)$ (0.90) and an increase in the extinction coefficient $E_{4650.001\%}$ (0.024) for the HAs from the layer (1.5-3.0 m) compared to the sample of the upper part of the profile (0-1.5 m) with indicators of 0.97 and 0.015, respectively. Chemical analysis of alkaline extracts of BWW samples for the entire thickness of the deposit using IR spectroscopy (IRS), UV-Vis, ¹³C NMR spectroscopy and liquid gel chromatography revealed the main mechanisms of formation of the structure of high-molecular organic compounds from precursor molecules and further directions for the transformation of molecular ensembles of HAs. An analysis of the results obtained shows that at the initial stage of BWW humification, high-molecular structures of HA are formed as a result of the interaction of molecules of natural biopolymers, such as lignin, (hemi)cellulose and lipids, etc. The number average molecular weights (M_n) of HAs decrease significantly down the profile. This is mainly due to the depolymerization and transformation of macrostructures of lignin fragments in the composition of HAs, which is confirmed by a decrease in the proportion of methoxyl groups according to ¹³C NMR data and the intensity of signals from syringyl- and guaicylpropionic structures in the IR spectra. Lignin fragments are transformed into polycondensed structures, which is confirmed by an increase in the degree of HA aromaticity in the central part of the profile with the highest degree of decomposition of the initial substrate. In the lower part of the profile, the decomposition of organic material is hard due to significant watering. This process is diagnosed by an increase in the proportion of paraffin fragments according to ¹³C NMR and M_n data.

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Impact of soil tillage on humic substances transformation according to ^{13}C -NMR study

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Keywords: humic substances, elemental analysis, soil tillage, ^{13}C NMR spectroscopy

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Soil cultivation negatively affects soil organic matter (SOM) quantity and quality, stimulates oxidative processes, removal of nutrients, and soil compaction by heavy machinery. Humic substances (HS) are the most essential component of SOM, constituting up to 60–90 %. This study was aimed to assess the changes in composition and molecular structure of HS under soil tillage. Two types of soils within Transbaikal territory were investigated: Eutric Cambisol Cryic and Haplic Cambisol Arenic. The cultivated soil variants have been the croplands over 40 years. HS were isolated from the soil followed recommendations of the International Humic Substances Society. Then humic materials were characterized by elemental analysis and ^{13}C NMR spectroscopy.

Humic preparations obtained are mainly differ in C content. The lowest C concentration was observed for native Haplic Cambisol HS, and it was significantly increased for arable soil variant. Both humic materials of Eutric Cambisol differed slightly in C amount. A regular decrease in N content was noted under soil cultivation. The atomic H/C ratio for HS under the study varied from 0.93 to 1.06. Higher value was observed for Haplic Cambisol, that reflects an increased share of aliphatic components into HS structure. Reduction in the H/C atomic ratio for the cultivated variant of this soil testifies aromatic carbon content enhancing. As a rule, soil cultivation intensifies microbial activity, destruction of alkyl constituents, N consuming, and HS polarity increase.

The ^{13}C NMR spectra were identified as alkyl C (5-50 ppm), O-alkyl C, divided into methoxyl C (50-60 ppm), carbohydrate C (60-95 ppm), aromatic C, included aryl C (110-145 ppm) and phenolic C (145-160 ppm), carboxyl C and ester groups (160-190 ppm), C atoms of quinone and ketone groups (190-220 ppm). According to the ^{13}C NMR spectra the proportion of aromatic carbon in HS was prevailed across all soil variants followed by alkyl C and O-alkyl C, and carboxyl C. When comparing the native soils, HS of Eutric Cambisol more reach in aromatic C. Aromatic carbon and carboxyl group contents were observed to elevate for cultivated soils HS. These changes are mostly pronounced in Haplic Cambisol. This data were consistent with elemental analysis. Our study has demonstrated that soil cultivation intensifies SOM labile components destruction and humic substances transformation. Organic matter of Eutric Cambisol Cryic are more resistant to soil tillage.

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Two-dimensional correlation spectroscopy for characterizing mineral and organic-matter bands in soil fractions

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Keywords: two-dimensional correlation spectroscopy, soil organic matter, chernozem, sod-podzolic soils, fractionation, infrared spectrometry

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Two-dimensional correlation spectroscopy (2D-COS) techniques improve traditional spectroscopic methods by transforming 1D spectra into 2D correlation maps and identify correlations between individual bands. 2D-COS simplifies the spectra of samples with many overlapping bands and increases the spectral resolution. This is essential for relatively weak soil organic matter (SOM) infrared bands compared to mineral-matrix bands. Thus, a method of IR measurements using 2D-COS for soil-fraction spectra is proposed for more accurate estimation and identification of bands, in particular for SOM.

2D-COS spectra of soil fractions of sod-podzolic, chestnut, and chernozem soils obtained using different fractionation and filtration options and IR modalities (attenuated total reflection, diffuse-reflectance, and photoacoustic FTIR) were made. Samples for contrasting formation conditions and different land use (native steppe, shelterbelt, fallow, and arable land for chernozems) and samples of sections of genetic horizons were used.

For sod-podzolic and chernozem soils, synchronous 2D-COS maps of the size fractions for DRIFT show a positive correlation in 4000–3600 (hydrogen bonds), 1700–1150, and 1100–200 cm^{-1} (quartz). Asynchronous 2D-COS maps show differences at 3690, 3660, 3620, 2930, 2830, 1643, 1562, 1444, 1251, 1158, 1070, 797, and 505 cm^{-1} . The first three bands belong hydrogen bonds and indicate an increase in the proportion of silicate and quartz not bound to SOM or bound to adsorbed water in small fractions, which is consistent with increased intensity for adsorbed-water band at 1643 cm^{-1} . The bands at 1070, 797, and 505 cm^{-1} belong to crystalline quartz, and their proportion increases with fraction size. The most important is the manifestation of peaks of 1562, 1444, 1251, and 1158 cm^{-1} , which have no significant contribution from inorganic components and are due to carboxylate groups, aromatic compounds, and Amide III band; the intensity of these correlation bands is significant for the discrimination of size fractions.

2D-COS reveals bands of functional groups of the surface of soil particles, organic components, in particular a larger number of aliphatic component bands of SOM, as well as adsorbed forms of water, including the long-wave region, where the dominance of the mineral components does not facilitate the use of either SOM extraction or destructive treatments (annealing or chemolysis). 2D-COS with granulometric methods for separating soil fractions showed that the range of 20 μm – 1 mm is informative and can be used for a detailed soil analysis.

The proposed approach for sod-podzolic and chernozem soils provides an extra data set for identifying SOM components with high reliability. The proposed 2D-COS approach can be extended to a broader range of soil fractions and separation methods (e.g., membrane separation of fine fractions). As a result, the methodology of 2D-COS IR experiments would be expanded to submicrometer fractions and extracts of SOM.

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On-line and off-line chromatographic separation with ultrahigh-resolution mass spectrometry of humic substances: comparison of FT-ICR-MS, Orbitrap and TOF

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Keywords: humic substances, fractionation, high-resolution mass-spectrometry, high-performance liquid chromatography, Orbitrap, TOF, FT-ICR-MS

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Humic substances (HS) are an extremely complex mixture of organic compounds. Qualitative analysis of such a mixture is a difficult task even when using ultrahigh-resolution mass spectrometry, such as Fourier-transform ion-cyclotron resonance mass-spectrometry (FT-ICR-MS), due to a number of physical limitations, such as ionization suppression effects in the source, space charge effects, charge capacity, etc. In addition, despite the great progress in instrumentation, FT-ICR-MS spectrometers remain quite rare and expensive instruments. Therefore, expanding the possibilities of HS analysis by such mass-spectroscopy methods as time-of-flight (TOF) and Orbitrap is a relevant task. However, the resolution and accuracy of determining the mass in these methods may not always be sufficient for HS. The main way to overcome this difficulty is to use on-line or off-line fractionation using high-performance liquid chromatography (HPLC). Although some findings have already been published on the use of chromatography with these methods [1–2], more research is required, both methodological studies and data gathering on various samples.

In this work, we have demonstrated the capabilities and limitations of Q-TOF and LTQ-Orbitrap in combination with reversed-phase HPLC on a number of humic and fulvic acid samples in terms of determining gross formulas in comparison with FT-ICR-MS. Fractionation provides a significant increase in the number of attributed gross formulas and enhances the reliability. On the other hand, the study revealed a number of problems, such as the lack of reference materials that can be used to compare different methods, as well as the need to build the criteria for correctness and precision of the data.

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Patterns of Pb(II) adsorption by the main genetic horizons of peaty-podzolic-gleyic soil

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Keywords: lead adsorption, soil organic matter, clay minerals, non-silicate iron minerals

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Clay minerals, non-silicate iron compounds, and soil organic matter are the main components capable of efficient Pb(II) sorption. However, the mechanisms of metal uptake by individual soil components are insufficiently studied, which complicates the prediction of the behavior of heavy metals in soils and landscapes.

The aim of this work was to reveal the patterns of Pb(II) uptake by individual soil components: organic matter, non-silicate iron compounds, and clay minerals in mineral horizons of peaty-podzolic-gleyic soil (PPG).

Sorption experiments were carried out on the EL_{ih}, EL, EL_{cn}, IIBD PPG soil horizons using lead nitrate in the concentration range from 0.0001 to 1.0 mmol/l. Sorption experiments were carried out before and after the removal of non-silicate iron compounds and organic matter from the soil. Sorption experiments were carried out at pH values corresponding to the pH of the aqueous suspension of each horizon, and the same ionic strength.

The removal of organic matter from the soil was carried out with a 10% solution of H₂O₂ while heating in a water bath, the removal of non-silicate iron compounds was carried out according to the Mehra-Jackson method. When both components were removed from the soil, organic matter was removed first and then non-silicate iron compounds were removed.

In the variant of the sorption experiment with untreated soil, lead was sorbed to the least extent by the EL horizon, which is explained by its depletion in organic matter and iron minerals. The EL_{cn} and IIBD horizons absorb lead to the greatest extent, due to the enrichment of these horizons in non-silicate iron compounds and clay minerals.

It has been established that after soil treatment with H₂O₂ and Mehra's and Jackson's reagent, the amount of absorbed lead increases, which can be explained by an increase in the surface area due to the removal of aggregating agents and the unblocking of sorption centers previously occupied by other components.

It is difficult to assess the role of organic matter in lead sorption by the EL_{ih} and EL horizons, because after its removal, the amount of sorbed lead increased. This may be due to close interaction of soil organic matter with the mineral components, and when the organic matter is removed from the soil, the absorption of Pb(II) involves the sorption centers of iron minerals and clay minerals. Non-silicate iron compounds play an important role in lead sorption by soil in all horizons, since after its removal, the amount of absorbed lead dropped significantly. Clay minerals make the most significant contribution to lead sorption in the EL_{cn} and IIBD horizons. The role of clay minerals in Pb(II) sorption in the EL_{ih} horizon is reduced, possibly as a result of the formation of a film of organic matter on the mineral surface.

Based on the data on the change in pH values in all sorption experiments, it can be concluded that at a Pb(II) concentration of less than 0.01 mmol/l, absorption proceeds according to the ion-exchange mechanism. If concentration is more than 0.01 mmol/l, there are 2 absorption mechanisms possible: ionic exchange and formation of intrasphere surface complexes on side cleavages of clay minerals, surfaces of non-silicate iron compounds, and deprotonated functional groups of organic matter.

This work was carried out within the framework of State Assignment No. 121040800154-8 "The role of organo-mineral interactions in the carbon cycle and environmental sustainability of soils and adjacent environments".

Obtaining and purification of fulvic acids from oxidized coal

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Keywords: oxidized carbon, fulvic acids, purification, adsorbent

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The paper presents the results of obtaining fulvic acids (FA) from oxidized coals of Kazakhstan and the study of their purification.

Oxidized brown coal from the Maikuben deposit with the following characteristics was used as a feedstock (wt. %): A^d - 25,8; W - 9,3; V^d - 46,6; S^d - 0,71. The content of humic substances is 56% [1].

Acid fulvic acids were obtained on the basis of potassium humate by acidifying them with a 5% solution of nitric acid; as a result of the reaction, humic acids precipitated in the form of amorphous brown precipitates. All organic substances remaining in the acidic solution after HA isolation were considered fulvic acids. FA was further purified by the Forsyth method [2], where activated carbons "Shoptykol" and "Shubarkol" (Kazakhstan) were used as an adsorbent to separate substances in the acid filtrate. The adsorption separation method makes it possible to obtain homogeneous preparations that are also not contaminated with precipitate ions.

The method for isolating fulvic acids according to Forsyth consists of the following stages: preparation of activated carbon, adsorption, purification, isolation, purification and concentration of fulvic acids, dialysis, drying. We carried out the stages before purification and concentration of fulvic acids.

The values of acidity, electrical conductivity, and ionic composition of the initial and obtained FA solutions after purification were studied. The ionic composition of FA solutions was studied on a two-channel capillary ion chromatograph Dionex ICS-5000. The results of the study are presented in table 1.

Table 1. Value of acidity (pH), electrical conductivity and ionic composition of the sample (T=25°C)

Name	pH	Conductivity, mS/cm	Cations, mg/l				Anions, mg/l			
			Na ⁺	NH ₄ ⁺	K ⁺	Mg ²⁺	Ca ²⁺	Cl ⁻	NO ₃ ⁻	SO ₄ ²⁻
Source FA	1.19	197.73	63.68	25.59	1873.57	9.40	57.76	10.75	51597.91	85.88
FA after cleaning with the adsorbent "Shoptykol"	1.36	145.52	80.48	49.27	1374.21	7.56	60.49	161.01	34106.45	70.48
FA after cleaning with the adsorbent "Shubarkol"	1.27	130.00	164.33	82.04	1197.41	7.46	49.42	281.05	31995.94	57.41

The data obtained showed that after the 4th stage of FA purification, the conductivity of the solutions decreases, which indicates a decrease in the K⁺, Mg²⁺, Ca²⁺, NO₃⁻, SO₄²⁻ ions in the solution after purification. An increase in the content of sodium and chlorine ions may be due to the fact that HCl and NaOH solutions were used when washing the adsorbent. Work will continue.

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Alkaline extraction and secondary synthesis in humic research

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Keywords: humic matter, alkaline extraction, oxidation, heterogeneous reactions

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Humic substances are polydisperse and heterogeneous compounds formed during decay and transformation of organic residues in the diversity of the environments (IHSS 2022). Their structure, composition and formation mechanisms remain poorly understood. The lack of common definition, the use of alkali as extractant solution and the paradigm of secondary synthesis as a key formation pathway has led to the deny of HS as a distinct category of natural compounds. We are going to discuss the notion of HS, to show some benefits of alkaline extraction and provide experimental evidence for oxidative coupling reactions under environmentally-relevant conditions.

Humic substances have long time been defined as specific products of humification distinct from the molecules of known chemical classes. HS are also defined operationally as fractions obtained during alkaline extraction (humic acids, fulvic acids, humin). Such dual definition is somewhat contradictory and causes problems with non-specific compounds in humic extracts (so-called non-HS) and the problem of identification of HS as chemically unique compounds in already formed humus. The origin-oriented understanding of HS as total of compounds comprising humus (synonymously to substances of humus, humus substances) can be a possible solution to this problem. It allows to link the notion of HS with the extraction method and is in line with current definition of HS by IHSS (www.humic-substances.org).

Alkaline extraction is traditional point of debate in humic research. We have studied the effect of alkaline treatment on the properties of humic acids extracted from chernozem, soddy-podzolic soil and compost in the absence and presence of oxygen. No differences in physico-chemical properties were observed in chernozem preparation, slight oxidation occurred in soddy-podzolic soil HA, while significant increase in functional group content and free radical content was observed in compost HA during extraction without N₂. Interestingly, compost HA-O₂ preparation was less resistant to degradation by laccase and showed higher physiological activity in priming tests with radish seedlings compared with HA-N₂. Thus, alkaline treatment of relatively fresh humic matter may really create artifacts but these may be beneficial for HA preparations used in commercial purposes.

Finally, it is questioned whether oxidative coupling reactions may occur in natural environments. We have studied synthesis of humic molecules in heterogeneous dynamic flow-through system, containing clay mineral, immobilized laccase as a biocatalyst and a mixture of phenolic monomers as humic precursors. We have shown that at low precursor concentrations typical of soil environment FA-like products are formed. Competitive sorption of the substrate and reaction products on the mineral phase as well as oxidation potential of phenolic monomers largely determine the composition of humic products in the solid and liquid phases.

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Synthesis of phenol-modified humic ligands with enhanced reducing properties and their use for preparation of silver nanoparticles

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Keywords: humic ligands, phenolic formaldehyde condensation, Fenton reagent, silver nanoparticles, antibacterial compositions

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The potential of AgNPs for therapeutic and biotechnological applications has gained much attention. Its major disadvantage is high toxicity. In this work, we proposed to modify humic substances (HS) with phenols for improving reducing power of natural HS. It is important for in situ synthesis of AgNPs assisted with HS. We expect synergistic antibacterial activities of both silver nanoparticles and phenoli-enriched humic ligands.

In this study, we used coal humate (CHP-Na) for chemical modification. Two synthetic pathways of incorporation of polyphenolic fragments into HS structures were used: the oxidative copolymerization of phenols initiated by Fenton reagent and phenolic formaldehyde condensation. The incorporated phenolic compounds were as follows: naphthoquinone, 1,4-hydroquinone, catechol, 2-methyl-hydroquinone, and 2,4-hydroxy-naphthoquinone. Structure of the obtained humic derivatives was characterized using ^{13}C NMR spectroscopy. The typical spectra are shown in Figure 1.

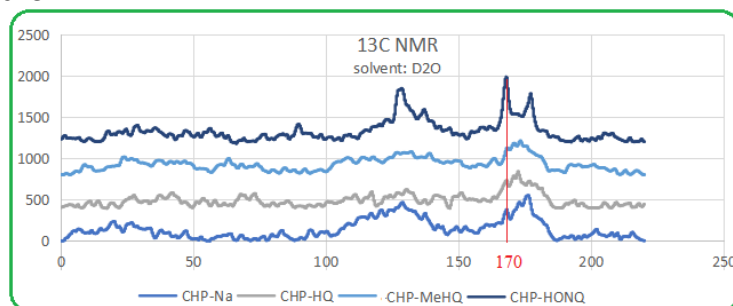


Figure 1. ^{13}C NMR spectrum of HA and the copolymers of hydroquinone, 2-methylhydroquinone, and hydroxynaphthoquinone.

The catechol derivatives showed narrow peaks at 170 ppm. This might indicate the presence of some oxidation products in the mixture. The spectra of other derivatives were very featureless - similar to the initial HA. The obtained ligands were used for synthesis of AgNPs. Kinetics of silver nanoparticles formation have been studied by potentiometric and spectrophotometric methods. The formation of AgNPs was confirmed by UV-vis spectroscopy and TEM (Figure 2).

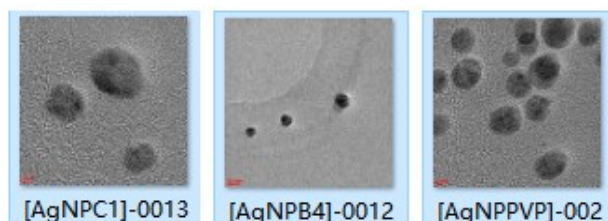


Figure 2. TEM revealed spherical AgNPs with the different diameters (8 nm – 25 nm.)

The antibacterial studies of the synthesized compositions are under way.

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Comparative characteristics of hepatoprotective activity of humic peloid preparations

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Keywords: humic acids, hymatomelanic acids, intoxication PCB, protective action

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Despite the constant increased attention to pollution problems, its severity does not decrease. The industry has often used substances that are naturally non-decomposing and so dangerous that its production and use are prohibited by the 2001 Stockholm Convention, but they are nevertheless present in the environment. The eco-toxicants of high priority include polychlorinated biphenyls (PCB), which tend to cumulate in humans.

The aim of this work was research of humic substances of peloid of protective action in model of acute intoxication caused by PCB.

The experiment was made on white outbred rats. Acute intoxication was simulated by single introduction via the probe of PCB mix to the stomach in a dose of 0,1 LD₅₀. To control group of animals (n=6) 0.9% chloride sodium solution was entered. As group of comparison were animals (n=6) to which after PCB intoxication daily entered 0.9% chloride sodium solution. With the medical purpose to animals (n=24) daily subcutaneously entered solutions of peloid drugs on the basis of the humic acids (HA), the hymatomelanic acids (HMA), fulvic acids (FA) and also initial humic substances extract (HFA) emitted from the low-mineralized silt sulfide muds (pH=7.4) by the patented techniques. The experiment was carried out within 10 days.

The therapeutic effectiveness of humic peloid drugs was evaluated by activity of enzymes of aspartate aminotransferase (AST) and alanine aminotransferase (ALT), change of bilirubin content, cholesterol and albumin on the 3rd and 10th day of the experiment in blood serum.

On the 3rd day of the experiment in comparison group under the influence of PCB there was a significant increase in activity of AST and ALT for 27% and for 19.7% respectively in comparison with healthy animals, the content of bilirubin increased by 10 times, cholesterol - by 1.5 times, at the same time the content of albumine decreased by 17%.

The use of peloid drugs has been resulted in smoothing of toxic action of PCB in various degree of intensity: on the 3rd day under the influence of all drugs the AST and ALT level has been sharply decreased; bilirubin content under the influence of HA and HMA decreased in comparison with comparison group more than twice – from 50 mkmol/L to 25 and 21 μmol/L respectively; action of FA and HFA decreased bilirubin slightly – till 31 and 44 μmol/L. Level of cholesterol decreased to physiological norm under the influence of all drugs, and the content of albumin was normalized only under the influence of HMA. On the 10th day all biochemical indicators of animals were normalized under the influence of humic substances.

Protective effect of all studied peloid drugs is established, at the same time the most effective was HMA on extent of influence on activity of enzymes, protein and lipidic metabolism.

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Humic substances in processes and technologies for reducing anthropogenic impact

Investigation of the effect of humus on some properties of urban soils

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Keywords: humic substances, chitosan, soil magnetism, X-ray diffraction, urban soils

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Soil is one of the most important rapidly changing components of the urban ecosystem, it needs a comprehensive study and monitoring measurements by available methods. The purpose of the work is to analyze the strength stability for the subsequent systematization of anthropogenically altered soils, as well as stabilization and management of the structural stability of the soils of the metropolis. The choice of a method for monitoring measurements of soil condition is one of the tasks of this work.

One of the important properties of the soil structure is the ability to resist mechanical loads under the action of humidity. The peculiarity of urbanized soils is the change in depth properties at shallow depths (up to 20-30 cm). The paper presents the results of a layer-by-layer (in increments of 3-7 cm) comparative analysis of the waterproofness of the soils of two sites differing in anthropogenic load and the presence of humus. In addition, together with soil samples in their natural state, experiments were conducted with model soil on the effect of humic acid (humate) and chitosan on the strength characteristics of ground sand in the process of increasing the amount of moisture in the sample.

The granulometric composition, bulk density, hygroscopic humidity, humus content, mineral composition, magnetic properties, and the content of a number of metals, including lead, cadmium, iron, cobalt, and nickel, were also determined for the selected objects.

Soil magnetism is the leading factor in the relationship of soil parameters: the content of iron in the soil and the total amount of metals in the soil profile, the content and distribution of humus in soil layers [1]. The strength was estimated by the penetrometric method according to the load required to submerge the cone (angle at the top 40°) to a certain depth (0.5 cm). To determine the magnetic properties of soils, the author's device "Magnetometer WAVE-2" (registration No. 2021666155/69, 2021, with the developed software) was used [2]. Particle sizes were estimated using the laser size analyzer Microtrac S3500 (Microtrac Inc., USA). The mineralogical composition of the samples was determined by X-ray diffraction using a universal X-ray diffractometer HZG-4A from Carl-Zeiss Jena (Germany).

The influence of humus on the values of maximum strength and the range of humidity at which the soil sample remains stable is shown. More humus-rich horizons (upper ones) show increased strength and a wider range of humidity.

Treatment of the sample with a chitosan solution leads to its hardening. The methods used (magnetic induction and penetrometric measurements) can be used as methods for monitoring measurements of the state of the soil cover.

The work was carried out within the framework of the state task "Colloidal Chemistry" (CITIS No. 121031300084-1), "Ecology" (CITIS No. 122040600057-3) and No. 075-01477-22-00 topics 0439-2022-0013.

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The activity of copper cations to form complexes with natural organic impurities

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Keywords: activity, complexes, copper, spectrophotometric method, natural organic impurities

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It is known that copper ions form strong complex compounds with amino acids, peptides, carboxylic acids, as well as with other organic ligands. The activity of copper cations to form complexes with natural organic impurities was evaluated by the spectrophotometric method in the interaction of copper ions with potassium iodide. Experimental results were published in conference proceedings [1,2].

It is established that:

- the interaction of copper (II) with glutamic acid solutions from 50 to 100 mg/l (TOC) prepared in distilled water reduces the oxidizing properties of Cu(II) by 9% - 39%, respectively. However, if solutions of glutamic acid with the above concentrations are prepared in natural water, the oxidizing ability of copper decreases by 53-88% [1];

- trilon B in concentrations from 2 to 5 mg/l per TOC unit reduces the oxidizing properties of copper(II) by 9.5 - 52%, respectively. The concentration of organic impurities in bottled drinking water ranged from 0.76 to 6.48 mg/l (according to TOC), and their activity to complexation with copper (II) ions was comparable to trilon B. Water treatment, for example, with red light, had a significant effect on the activity of organic impurities [2].

The above properties are important to take into account when analyzing the antimicrobial properties of copper (II), which, although long known, are being actively studied in connection with the Covid-19 pandemic.

The complexing properties of copper were evaluated by spectrophotometric method ($\lambda=285\text{nm}$). The relative error of determination, according to the results of parallel measurements, was 5-7%. The decrease in the amount of iodine formed in the presence of surface water of the Volchikhinsky reservoir with organic impurities in an aqueous solution of 2.8 mg / l (TOC) was 81% compared to the blank experiment, and in the presence of the same water, but after chlorination – 37.5%.

The assessment of the complexing ability of natural organic impurities showed that copper (II) ions bind into strong complex compounds with organic impurities of natural origin.

Thus, the properties of natural organic impurities, as well as special organic additives in drinking waters that are used to prepare disinfectant solutions containing copper (II) compounds can be evaluated using the previously proposed methodology [3].

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Improving the environmental monitoring system

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Keywords: environment, aluminum, humic acids, organic impurities

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Improving the system of environmental monitoring is impossible without the integration of scientific knowledge about the properties of humic and fulvic acids and administrative decisions in the field of environmental policy.

Surface waters in large cities are sources of drinking water supply, but can be contaminated with heavy metal compounds, which are not adequately determined by existing methods, since they are contained in low concentrations and in the form of strong complex compounds with fulvic acids. For example, the addition of mercury ions to surface water at a concentration exceeding twice the threshold limit value does not make it possible to determine the mercury content in the water system using the existing analysis method.

In traditional water purification technologies, aluminum compounds are used as coagulants: aluminum sulfate and aluminum chlorohydrate (aluminum oxychloride). The efficiency of removing organic compounds in the form of humic and fulvic acids depends on seasonal changes in organic impurities, the dose for primary chlorination, the dose of the coagulant and is 50-60% (by permanganate index), but 30-40% by the content of total organic carbon (TOC). Determination of the aluminum content in the source and purified water is possible by different methods. It has been proven that the results of analysis with aluminon (traditional method according to GOST 18165-89) are significantly affected by the properties of organic impurities contained in the source or purified water. How to evaluate properties and what methodology to take as a basis? Currently there are no substantiated solutions, although there are instrumental methods of analysis. It has been proven (HIT-2019, Karpukhina E.A.) that the best method for determining metals in the presence of humic substances is inductively coupled plasma atomic emission spectroscopy (ICP-AES). It is recommended to use the ICP-AES method for environmental monitoring, since more than 70 elements, including aluminum, can be determined simultaneously.

It is recommended to carry out chlorination with a TOC content of less than 5 mg/l to exclude the possibility of the formation of carcinogenic organochlorine compounds in the regulatory documentation (SanPiN 2.1.3685-21). However, in the publications on water treatment, there is no information not only about the properties of natural organic impurities and the possible total content of organochlorine compounds after chlorination, but also about the content of organic impurities in terms of TOC. In addition, methodological problems remain:

- how to evaluate the properties of organic impurities formed in the source water after primary and secondary chlorination in relation to biometals (magnesium, calcium, iron);

- how to replace primary chlorination, which not only contributes to the destruction of humic and fulvic acids and increases the efficiency of their removal from water, but also maintains the condition of the equipment in sanitary standards. As an alternative to replace gaseous chlorine or sodium hypochlorite, some companies offer the use of polyhexamethylene guanidine (PHMG), a biocide that is banned in South Korea due to its particular health hazard.

The lack of an integrated approach to solving the problems of assessing the composition and properties of natural organic impurities, humic and fulvic acids, does not allow justifying the types of technological and administrative decisions. This leads to incorrect policy in the field of drinking water supply, as well as in the field of ecology in general, and harms the health of the population.

Humic substances in methanogenesis

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Keywords: humic acids, fulvic acids, methanogenesis, microbial consortia

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Scientific studies have been carried out that allow us to approach a deeper understanding of the role of humic substances (humic acids, fulvic acids) in the processes of anaerobic fermentation, called methanogenesis, in the implementation of which, as a rule, natural evolutionarily formed heterogeneous microbial consortia take part. The inhibitory effect of humic acids on various hydrolytic enzymes of those microorganisms that catalyze the primary stage of depolymerization of organic substances that are part of the raw materials for the methanogenic process has been shown [1].

It is shown that it is possible to regulate the intensity of methanogenesis catalyzed by consortia of different composition by introducing humic substances (humic acids, fulvic acids, sapropel) into the reaction medium [2] with the control of the effect on biocatalysis by determining the intracellular concentration of ATP in cells of microbial consortia [3, 4].

Various combinations of approaches to the suppression of methanogenesis and the formation of methane with a simultaneous decrease in the part of methane in its composition with carbon dioxide were investigated [1]. Redox derivatives of humic acids introduced into reactions of methanogenesis results in suppression of the process. The use of same redox compounds concurrently with potassium persulfate (possessing oxidizing and electron acceptor properties), hexahistidine-containing organophosphorus hydrolase with high lactonase activity, decreasing the resistance of microbial methanogenic community, and natural polypeptide antimicrobial agent bacitracin produced by *Bacillus* cells into the media with anaerobic consortia were investigated in regulations of methanogenic suppressions. The possibility of notable and quick as well as slow and soft regulation of methanogenic processes was demonstrated in a newly developed and patented method of methanogenesis suppression [5].

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Behavior of plutonium in the mineral-natural organic matter system

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Keywords: Plutonium, natural organic matter, goethite, smectite, sorption

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Since the beginning of the atomic era, humanity has faced the persistent issue of preventing environmental pollution with radionuclides. Therefore, one of the most urgent and modern aims is to study the migration behavior of radionuclides in contaminated environmental objects. In Russia, storage pools for liquid radioactive waste (LRW) are operated. In accordance to the specifics of these objects, special geochemical conditions are created in them, which determine the possible ways of migration of radionuclides from them. At the same time, it is important to ensure the safety of such facilities for the entire period of their operation, as well as to consider measures for the further rehabilitation of these facilities and the area around them.

Plutonium is one of the most toxic man-made radionuclides, and it has extremely complex chemical properties. In the environment the main reactions that determine the behavior of plutonium are sorption, complexation and redox reactions. The purpose of this work was to study the behavior in the Pu(V)-goethite-natural organic matter and Pu(V)-illite/smectite-natural organic matter systems, including under the operating conditions of LRW storage pools.

In the work, a synthesized and characterized sample of goethite (α -FeOOH) and standard samples of humic (potassium humate (Powhumus, Humintech) and fulvic acids (Fulvagra, Humintech) were used as materials. Illite-Smectite (70/30 ordered) sample (ISCz-1) that was characterized by Clay Minerals Society was used. In the course of the work, the effect of time and pH on the sorption of Pu(V) in individual and ternary systems. Experiments were carried out in the pH range from 2.0 to 11.5 in a solution of 0.05 M NaNO₃.

To study the colloidal properties of the systems, the ζ -potential was measured in the pH range from 2.0 to 11.5. Because of this experiment, it was found that the colloidal system potassium humate - goethite is stable at pH values from 4 and above, the potassium humate system is stable at pH above 9, the goethite system is stable at pH below 3 and above 8. To control the dissolution of natural organic substances, spectrophotometry was used. For fulvic acid, no dependence of solubility on pH was revealed, however for potassium humate an increase in dissolution is observed with increasing pH.

Dependences of Pu(V) sorption in binary and ternary systems containing humic and fulvic acids and colloidal mineral particles were obtained. A change in the sorption value with time was established, which indicates the occurrence of redox reactions in the system. In individual systems Pu(V) – goethite and Pu(V) – smectite equilibrium is reached in a long time (more than 2 months). In presence of NOM redox reactions are much faster and equilibrium is reached in shorter time. Presence of natural organic substances strongly affects the sorption of Pu(V) and the aggregative stability of the colloidal system. In presence of NOM in ternary systems sorption of Pu(V) increases at low pH (lower than 4), but it decreases at high pH comparing to binary systems.

The results of sequential leaching showed that the forms of plutonium in the ternary system are similar to the forms in the LRW storage.

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Evaluation of the effectiveness of the use of humic-bentonite washing agents for cleaning oil-contaminated soils

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Keywords: washing agents, humic-bentonite suspension, encapsulation

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The use of highly dispersed solid particles as stabilizers of oil dispersions is very relevant from the point of view of developing environmentally safe and cost-effective ways to minimize the impact of oil spills based on natural self-purification processes. The use of such technologies reduces the risk of secondary pollution and is especially relevant for water protection and protected areas that are most sensitive to purification from petroleum hydrocarbons. In this regard, an approach to the restoration of oil-contaminated environmental objects was proposed, based on the use of humic-bentonite suspension on a water basis with additives of nonionic surfactant of the 4th hazard class. Such agents will contribute to the dispersion of the oil pollution film, followed by its encapsulation in the composition of ultrafine suspensions.

For the purpose of comparative evaluation of the effectiveness of water-based washing agents of bentonite clay for the purification of oil-contaminated soils, the following were studied: bentonite-humic reagent-surfactant (B-H-S); bentonite-surfactant (B-S); bentonite suspension (B); nonionic surfactant (S). Artificial pollution was carried out for model substrates – fine-grained river sand (0.4-1 mm fraction) and low-grained river pebbles (2-5 mm fraction) with two types of crude oil: low-viscosity, with a density of 0.816 g/cm³ and high-viscosity, with a density of 0.896 g/cm³. River pebbles were additionally polluted with diesel fuel (D)

The results showed the greatest efficiency of washing the pebble substrate contaminated with D with the studied agents. The degree of washing varied from 48.7 to 84.1%, depending on the type of agent. The maximum effect was observed with the use of organomineral agent B-H-S. For a pebble substrate contaminated with low-viscosity and high-viscosity oil, the washing efficiency was slightly lower: from 28.6 to 73.1% and from 33.3 to 71.2%, respectively, depending on the nature of the agent and was also maximal when using B-H-S. There was no significant difference in the degree of flushing from the type of oil. For sandy substrates: the agents proved to be more effective for flushing low-viscosity oil compared to high-viscosity.

An analysis of the distribution of oil in the sedimentary and supramental phases of the organomineral suspension formed after washing showed that most of the petroleum hydrocarbons pass into the solid phase of the washing agent. At the same time, most of the petroleum hydrocarbons, regardless of the type of contamination and the nature of the substrate, were contained in the solid phase of agent B-H-S. According to the content of petroleum hydrocarbons in the solid phases of the washing waters formed after washing, it can be arranged in the following row B-H-S > B-S > S > B. This fact confirms the assumption about the effective encapsulation of petroleum hydrocarbons in the composition of the mineral phase of the suspension.

Dynamics of removal of rare earth and heavy metals with water-soluble organic matter from soils when phosphogypsum is applied (in a laboratory experiment).

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Keywords: organic matter (OM), lysimetric waters, amphiphilic properties, phosphogypsum, rare earth elements (REE), heavy metals (HM)

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Phosphogypsum (PG) is a waste from the production of phosphate fertilizers that accumulates in greater quantities. It gets into the soil both as a result of dusting of dumps, and as an ameliorant, that is deliberately introduced into the soil. The high content of rare earth elements (REE) is one of the potential dangers of FG, entering the soil. It is necessary to evaluate the dynamics of the transformation of REE coming from FG into the soil. Soil organic matter (OM) can play an important role in this regard, since the significance of OM in the fixation of REE was previously shown.

For the experiment, plastic vegetation vessels (columns) were filled with soil sifted through a sieve with a diameter of 2 mm for podzolic soil and 3 mm for chernozem. A layer of gravel washed with nitric acid was placed at the bottom of the vessel for drainage, then a layer of inert non-woven material and a soil sample: podzolic or chernozem. The vessels were spilled with distilled water until completely wetted, and then phosphogypsum was added to the surface (2 g per vessel). The experiments were performed in duplicate. Lysimetric water was discharged into glass vessels. The duration was 18 weeks, lysimetric waters were sampled once a week.

Results.

In lysimetric waters molecular weight (Sephadex G-50 gel) and amphiphilic distributions (OCTYL-SEPHAROSE CL-4B gel) were determined with a BioRad chromatograph. The total content of REE and HM in lysimetric waters, hydrophilic and hydrophobic subfractions after their separation, was determined by the ICP-MS method (Bruker ICP-MS 820).

In the experiment with podzolic soil, most of the organic matter with the maximum degree of hydrophilicity was taken out from the 4th to the 9th week of the experiment; the introduction of two different doses of phosphogypsum has practically no effect on the overall pattern of OM removal by weeks. In addition, the composition of the removed OM changed somewhat over time. According to amphiphilic distributions, REE and HM are mainly removed at the expense of the hydrophilic fraction of OM. The maximum amount of REE and HM is taken out in the period from 3 to 9 weeks. In the experiment with chernozem compared with podzolic soil REE and HM are sorbed more efficiently. It should also be noted that in the experiment with chernozem, several other patterns of removal of both OM and REE and HM are observed.

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Selective sorption materials based on mesoporous polymers for oil recovery and water treatment

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Keywords: mesoporous polymers, oil recovery

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Development of innovative materials with high sorption capacity and selectivity presents the ongoing challenge for academic community from both scientific and practical viewpoints, and special focus is placed on the solution of demanding ecological problems. For example, oil spillage, environmental pollution, emission of toxic organic wastes, and water contamination dictate the need for the persistent search of new approaches and high-performance materials for pollution control and safety.

In this work, a new eco-friendly approach for the development of high-performance polymeric materials with high sorption capacity and selectivity is proposed. Mesoporous polymeric materials (high-density polyethylene, ultrahigh-molecular-weight polyethylene, polytetrafluoroethylene, etc.) are prepared using the strategy of environmental crazing in the ecologically safe mode in the presence of the biphasic oil-in-water emulsions with high water content (above 95%). The resultant materials are characterized by high porosity (above 40%), nanoscale pore dimensions (below 15 nm), specific pore area $23.2 \pm 0.1 \text{ m}^2/\text{g}$. The development of specific fibrillar-porous structure is responsible for new highly unique properties of the polymer materials, including superhydrophobicity and selectivity. Mesoporous materials based on hydrophobic polymers (HDPE, PTFE, PP, UHMWPE, etc.) show highly selective oil sorption (hydrocarbons, plant oil, petrol) from water environment, and sorption capacity is 2 g/g. The advanced mesoporous materials can be easily recovered and repeatedly used, and the entrapped oil can be safely collected. Moreover, these materials can be successfully used for the recovery of high-cost natural essential oils.

This approach offers new benefits for the controlled development of high-performance polymeric materials with high sorption capacity and selectivity for diverse practical applications.

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Direct and indirect detoxification effects of humic substances

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Keywords: humic substances, detoxification, bioassay, luminescent bacteria, adaptive response

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Humic substances (HS) are ubiquitous natural products of decomposition of dead organic matter. For a long time, HS have been generally regarded to be inert in ecosystems. Thus, ecotoxicological aspects of HS have mainly been discussed in connection with their ability to bind heavy metals and various organic xenobiotics, with consequent modulation of their bioavailability and toxicity. These types of effects on organisms are called 'indirect effects'. In reality, the detoxifying effects of HS in water ecosystems are quite complex: HS not only neutralize free pollutants (indirect bioeffects), but also stimulate the protective response of organisms (direct bioeffects).

Prospects and potential problems of bioluminescent bacteria-based assay to monitor toxicity of solutions in the presence of HS are discussed. The main criterion for the application of the bioluminescent unicellular bioassay is versatility and ease of use.

The detoxification efficiency of HS in different pollutant solutions was evaluated, and the detoxification mechanisms are discussed. Particular attention is paid to the direct and complex direct + indirect effects of HS. The work focuses on the protective function of HS in solutions of radionuclides and salts of stable metals, organic oxidizer solutions, with special consideration of antioxidant properties of HS. Evidences for the direct HS effects on the bacteria were presented, such as an increase of rates of NADH-dependent enzymatic processes in cells, synthesis of mucous layers fixed by HS in the toxic solutions at cell membranes. Hence, the direct bioeffects of HS in toxic solutions might form a basis for the stimulation of the protective bacterial response.

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The effect of humic acid and chitosan on coagulation of iron(III) hydroxide by NaCl

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Keywords: iron hydroxide, humic acid, chitosan, river-sea zone

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Boundary zones play an important role in ocean ecosystems, as zones of considerable biogeochemical activity [1]. The river-sea transition zone is of particular importance due to the increased anthropogenic impact on river ecosystems. Transformation of the forms of riverine matter (dissolved, colloidal and suspended) in this zone significantly affects the state of marine ecosystems. The colloidal component of river runoff practically has not been studied, in contrast to the dissolved and suspended forms.

This work is part of our research on the transport of riverine matter (clays, carbonates) to the sea and is aimed at studying the influence of electrolytes and dissolved organic matter (DOM) on the stability of colloidal iron [2].

The variety of factors affecting the processes in the river-sea zone required the study of the transfer process in laboratory conditions. Iron(III) hydroxide sol - $(\text{Fe}(\text{OH})_3)$ (particle size 10-30 nm) in NaCl solutions was chosen as a model system. Humic acid (HA) separated from coal (leonardite) (Powerhumus, Humintech Ltd.) was chosen as a model of transformed DOM of river water. Biopolymer chitosan (Ch), 83 kDa, 82% deacetylation degree, chloride form (Bioprogress Co) was a model of fresh DOM (flocculant) released in situ in the river-sea area. The sol was prepared by FeCl_3 hydrolysis under boiling. The size of $\text{Fe}(\text{OH})_3$ particles was determined with a Zetatrac analyzer (Microtrac Inc., USA).

The aggregative stability of the sol was shown to remain constant until the salt concentration of 18‰, after which the particle size increased sharply from 10 to 500 nm. The presence of HA does not significantly affect the behavior of the sol. The addition of a Ch solution (50 mg/l) to the sol leads to a loss of its stability at 3‰ NaCl, while the lower concentrations of Ch (10 mg/l) do not affect the behavior of the sol. That is consistent with our previous studies in which the introduction of freshwater bacteria in the sol caused an increase in its particle size at 5‰ NaCl, which can be explained by the flocculation of the sol by active substances, released during the viral lysis of bacteria [2].

The sol behavior changes significantly if a Ch solution was added to $\text{Fe}(\text{OH})_3$ pre-modified with HA, the sol stability in this case was maintained up to 18‰ NaCl. HA is likely to create a protective layer on the iron hydrosol particles, which reduces the flocculation activity of Ch. It was determined that about 50% of the total iron carried by rivers is deposited in the river-sea mixing zone. According to our data, HA can facilitate the transport of colloidal iron to the outer part of the mixing zone even in the presence of flocculants (fresh DOM), capable of depositing it in the inner part of the mixing zone.

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Reduction of radiation effects of tritium and americium on luminescent bacteria by humic substances

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Keywords: low-dose; radionuclides; radiotoxicity; luminous bacteria; humic substances

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It is known that microorganisms of soils and aqueous media are sensitive to the presence of humic substance (HS), products of natural decomposition of organic matter, which play a role of natural attenuators of environmental toxicity. We study the effects of alpha- and beta- emitting radionuclides (americium-241 and tritium, respectively) in the presence of HS under the conditions of low-dose exposures (<0.1 Gy). Luminous marine bacterium *Photobacterium phosphoreum* was applied as a model unicellular water microorganism to monitor toxicity and activation ability of the radionuclide solutions. To imitate the marine environment for bacterial cells and to balance osmotic processes, the 3% NaCl solutions were used.

The bioluminescence response of the marine bacteria to americium-241 and tritium corresponded to the "hormesis" model: it included stages of bioluminescence inhibition and activation, as well as the absence of the effect. HS were shown to decrease the inhibition and activation effects of the radionuclides on the bacterial luminescence. The changes were shown to be related with the accumulation of americium-241 in the bacterial cells [1]. The correlations between the bioluminescence intensity and the content of Reactive Oxygen Species (ROS) were found in the radioactive bacterial suspensions in the presence of tritiated water [2].

The results demonstrate an important role of HS in natural processes in the regions of low radioactive contaminations: HS can mitigate radiotoxic effects and adaptive response of microorganisms to low-dose radioactive exposure of alpha- and beta- types. The involvement of ROS to these processes was demonstrated.

The results can provide a basis for predicting a response of living organisms to radiation at large territories infected with low-intensity radiation after accidents, discharges of nuclear plants, or underground mining of natural resources.

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Transformations of humic acids and redistribution of rare earth elements during mechanochemical treatment of metal-bearing lignite

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Keywords: brown coal, rare earth elements, humic acids, mechanochemistry

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Active use of rare earth elements (REE) by mankind has set the task of finding alternative sources, and coal and products of its processing are among the most promising. Previously, we showed the possibility of mechanochemical concentration of REE in the humic acid fraction (HA) in brown coal (BC) [1]. The aim of this work is to study the changes in HA that occur during mechanochemical treatment and the effect of these changes on the binding of REE by HA. BC mined from the Azeiskoe deposit (Russia) with the total content of REEs of $(9.7 \pm 0.4) \cdot 10^2$ g per ton was used in this study. Table 1 shows concentrations of REE in HA determined by ICP-MS.

Table 1. REE concentrations in HA of the pristine BC, BC subjected to mechanochemical activation with $\text{Na}_2\text{CO}_3 \cdot 1.5\text{H}_2\text{O}_2$ (MCA), and mechanochemically activated in the absence of reagents (MA) BC

	REE concentration, g/t		
	HA of Pristine coal	HA of MCA coal	HA of MA coal
45 Sc	13 ± 0	15 ± 0	14.5 ± 0.7
89 Y	51 ± 0	135 ± 7	175 ± 7
139 La	35 ± 1	90.5 ± 3.5	105 ± 7
140 Ce	145 ± 7	330 ± 0	360 ± 0
141 Pr	25.5 ± 0.7	50.5 ± 0.7	53 ± 1
146 Nd	110 ± 0	220 ± 0	230 ± 0
147 Sm	25 ± 1	44 ± 1	47 ± 1
153 Eu	5.75 ± 0.07	10 ± 0	10.5 ± 0.7
157 Gd	20 ± 1	38.5 ± 0.7	43.5 ± 0.7
159 Tb	2.7 ± 0.1	5.6 ± 0	5.9 ± 0
163 Dy	14.5 ± 0.7	29 ± 1	34 ± 0
165 Ho	2.55 ± 0.07	5.75 ± 0.21	6.55 ± 0.21
166 Er	6.65 ± 0.21	14.5 ± 0.7	16 ± 0
169 Tm	0.86 ± 0.04	1.8 ± 0	2.1 ± 0.1
172 Yb	5.2 ± 0.3	10 ± 0	12.5 ± 0.7
175 Lu	0.77 ± 0.04	1.45 ± 0.07	1.9 ± 0.1
∑ REE	463	1002	1117

The report will present CHN-analysis, infrared spectroscopy, and potentiometric titration data for coal and HA subjected to MCA under different conditions. The difference of mechanochemical oxidation of HA in coal and previously isolated ones will be shown.

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The humic acids impact on the photodegradation process of some phenolic derivatives under UV irradiation in aqueous solution

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Keywords: humic acids, peat, photodegradation, excilamps, phenolic compounds

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Modification of humic acids (HAs) properties is possible under the influence of, for instance, mechanochemical or photochemical actions or other chemical methods. HAs can absorb light and transfer light energy to other components of aqueous solutions, in some cases strongly influencing the photolysis of pollutants.

Phenols are one of the most common pollutants in industrial wastewater, chemical, petrochemical, paint, textile, pesticide plants, etc. They serve as intermediate links in the industrial synthesis of products, such as a variety of adhesives and antiseptics [1].

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 group of studied HAs was isolated from an undisturbed high-moor peat with different humidity in the Mezensky District. The thickness of the peat deposit was 10-60 cm and 60-80 cm. The concentration of the studied solution of humic acid was 0.05 g/l. The standard commercial preparation of humic acids Aldrich from brown coal was also studied. 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, which indicates the presence of several fluorophore groups in the composition of the objects under study. The effect of UV radiation from excilamp (KrCl, 222 nm) on the spectroscopic properties of humic acids has been investigated. *p*-Cyanophenol and 2,6-Bis(hydroxymethyl)-4-methylphenol (Aldrich Chemical Co) were selected as a phenolic compounds. The presence in the water solution of phenolic compound the humic acids has a noticeable effect on the processes of photochemical degradation under the action of UV radiation. The fluorescence intensity of the mixture drops sharply after irradiation, to a much greater extent than in substituted phenols. This allows us to conclude that the addition of Aldrich humic acid significantly increases the photodegradation of the compound under study. The addition of peat humic acid leads to much weaker changes in the absorption and fluorescence spectra of the mixtures, though in the same direction as the addition of the Aldrich humic acid. HAs were found to have an enhancing effect on the photodegradation of substituted phenols, which could be due to various processes. First of all, humic substances are important photosensitizers in soil and water environments, due to their ability to produce reactive species under irradiation. This effect may be also caused by energy transfer and by formation of π -complexes. 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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Assessment of efficiency of several surfactants in combination with humic substances for cleaning oil polluted sand

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Keywords: humic substances, oil pollution, surface washing agents, surfactants

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Despite many environmental treaties and international agreements on climate change, oil is still one of the most needed resources and a key-element in international trade. Although oil usage and oil transportation are severely regulated, the accidents take place. Oil spillages may disrupt entire ecosystems leaving dead animals and plants behind as well as causing damage to the human health. There are several approaches for removal of oil films from the surfaces [1]. For aquatic environments, one of them is to use surfactants to disperse oil and make it thus more biodegradable. For contaminated soil surface another broad class of remedial agents is used under the category "surface washing agents".

The goal of this work was to test combinations of different surfactants with humic substances as the components for SWA compositions. Humic substances are natural surface active agents. We hypothesized that their joint use with the synthetic detergents could increase efficiency and improve safety profile of the SWA compositions.

For testing efficiency of different SWA compositions we used approach developed by the Exxon company, which is called "beach washing" test. We tested a number of different surfactants. A number of them were kindly provided by the NIIPAV LLC (Volgodonsk, Russia), nominally: Betapav 1830, Betapav AP45, Katapav 1618. Two surfactants were provided by "Zavod sintanolov LLC"/Norchem Group (Nizhny Novgorod, Russia): Bianol, Stearox 6, ethoxylated rapeseed oil. In addition, we used Span80 and Tween80 nonionogenic surfactants.

Effectiveness tests were conducted with different oil products such as crude oil, diesel fuel and natural gas condensate. These tests shown that cleaning effectiveness for several washing agent mixtures may reach up to 30% for crude oil polluted sand, up to 40% for diesel fuel and 60-70% for natural gas condensate.

Experiments regarding different modes of washing and their respective effectiveness (single and double washing) of oil polluted sand were also carried out.

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Complexes of polyacrylic acid with amino organosilanes as dust suppressants

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Keywords: dust, pm2.5, pm10, sorption, humics

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Air pollution by dust is one of the acute modern problems of megacities. Large particles ($\leq 10 \mu\text{m}$ in diameter) and small particles ($\leq 2.5 \mu\text{m}$ in diameter), better known as PM 10 and PM 2.5 (particulate matter), respectively, can be airborne and, if inhaled, can cause serious respiratory problems for many years. Modification of acrylic acid with amino organosilanes leads to the possibility of their immobilization on various mineral particles, which can be used to bind dust and prevent wind erosion. Previously, our laboratory developed methods and approaches for the synthesis of highly adhesive derivatives of humic substances - silsesquixane-humic complexes (SSHC). It is shown that they can be effective for dust suppression, but they have the disadvantage of having a strong black-brown color. Which, despite their harmlessness, can cause inconvenience when used in an urban environment, therefore, complexes with polyacrylic acid - SSPA were tried, which are transparent, but form similar complexes.

To synthesis SSPA 20 ml of a 10 g/l solution of polyacrylic acid (Aldrich, Mv 450 kDa) were prepared. Then the aliquot of APTES (3-aminopropyltriethoxysilane) was added (50, 100, 200 mg for SSPA-25, -50, -100, respectively). To prepare dust models the dried soil was sieved with 100 μm mesh. For the experiment, 10 grams of model dust was placed in Petri dishes, after which 5 ml of derivatives or distilled water as control were added to dust and dried in the electric oven at 50 °C. To study of binding particulate matter by SSPA lab-scale air-blowing tests were used. In this test after treatment of model dust with SSPA and water as control, the samples were placed in a plastic container equipped with a compressed air source and a particle matter sensor. The determined concentration of particulate matter after exposure of compressed air on the samples is shown bellow

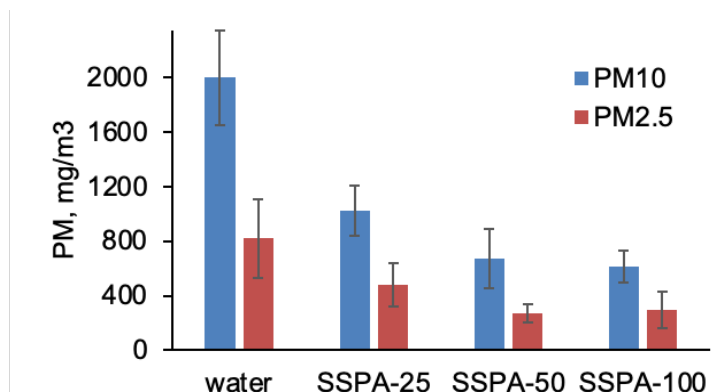


Figure 1. Determined concentration of particulate matter after exposure of compressed air to dust samples.

As can be seen from the above results, treatment with SSPA leads to a decrease the content of particulate matter in the air. Treated samples are more resistant to wind erosion. It can also be seen that this effect increases with an increase of organosilane content in the complex. This can be explained by the binding of small particles in the dust by silanol groups in the complexes. Thus, the complexes can be considered as promising dust suppressants also as SSHS complexes.

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Cluster analysis of different water quality indicators

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Keywords: water quality indicators, water treatment, chlorinated byproducts, cluster analysis

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In order to find ways to manage water treatment technology, cluster analysis of different quality indicators of raw water of the Ufa river (prior to treatment) and potable water was conducted. The indicators analyzed were: turbidity, chromaticity, permanganate index, dissolved organic carbon (DOC) of raw water; specific ultraviolet absorbance at 254 nm (SUVA) of raw and potable water; and concentrations of byproducts of water chlorination, such as trihalomethanes (THM), haloacetic acids (HAA) and halogenated semi-volatile organic compounds (SVOC) [1, 2]. The sample size was 200 measurements carried out in five years. The Euclidean distance was used as a linkage distance measure.

According to the data obtained, the producing of volatile chlorinated byproducts (THM and HAA) in potable water was linked to the DOC concentration, while the formation of halogenated SVOC was related to such a qualitative characteristic of DOC as its aromaticity shown through the SUVA parameter (Figure 1). The dose of a chlorinating agent depends on chlorine demand and therefore is strictly related to the raw water chromaticity, turbidity and SUVA.

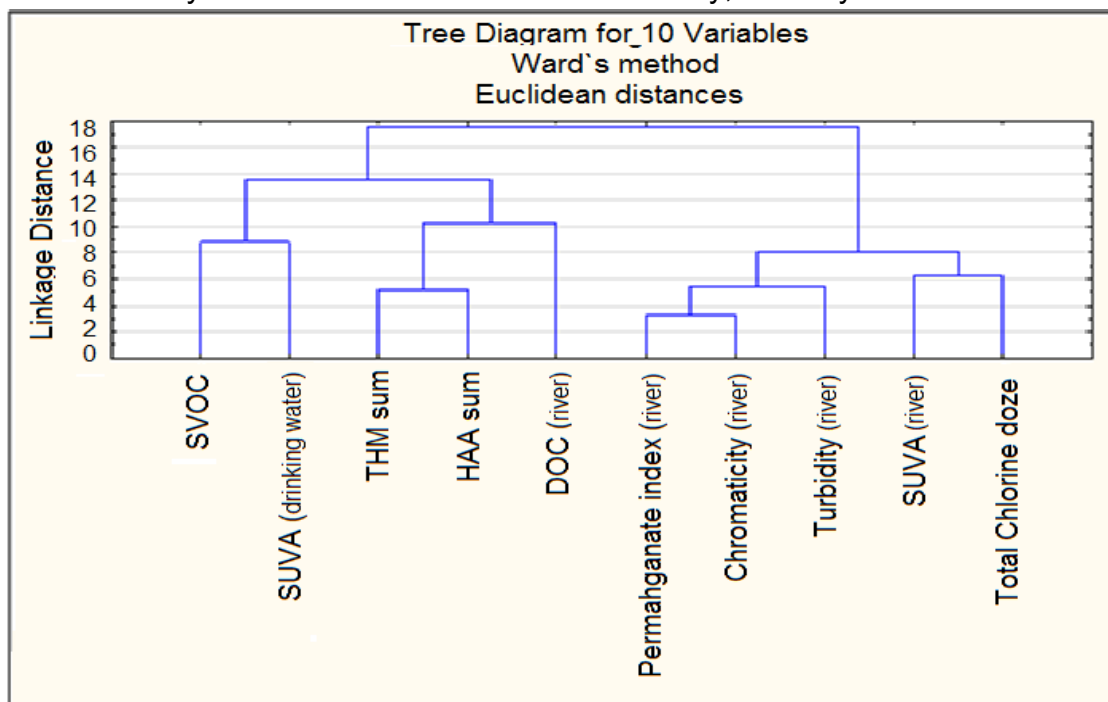


Figure 1. Results of cluster analysis of raw and potable water quality indicators.

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Studies on CO₂ absorption using humic substances

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Keywords: humic substances, carbon dioxide, absorption, capacity

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The purpose of this work was to study the effectiveness of humic acid salts in capturing CO₂. An aqueous solution of potassium and ammonium humate in different concentrations was used as absorbents. Salts of humic acids were obtained at the Institute of Coal Chemistry and Technology.

The content of carbon dioxide at the inlet was fixed and amounted to about 13.6% vol. The CO₂ absorption process was carried out at constant temperatures (25°C and 40°C respectively). The volume of the absorbing liquid was also known and used in the calculations. The CO₂ concentration in the outgoing gas was measured continuously during the experiment using a GA-40 Tplus IR gas analyzer. The process was carried out until the CO₂ concentration at the outlet was close to the concentration at the inlet. The absorption capacity was calculated from the gas balance (it was based on the measured CO₂ content and known gas flow rates). In addition, the absorption capacity was checked using a Scheibler (it is based on volumetric analysis, it is a kind of gas burette). The pH of the solution was also checked before and after saturation with CO₂.

The results of the tests are summarized in table 1. For comparison, a solution of monoethanolamine (MEA) and water were compared.

Table 1. Results of CO₂ uptake in the studied

Test №	Solvent	Amount of solution, g	Inlet gas concentration, %	Process temperature, °C	Testing time, min	Absorbed CO ₂ , g/kg	Absorbed CO ₂ (liquid analysis)	pH, before/after saturation
1	1% PH	500	13.5	25	6.63	1.14	-	-
2	1% AH	500	13.6	25	10.05	1.35	-	-
3	Dis. water	500	13.54	25	5.63	0.64	-	-
4	0.1% PH	500	13.42	25	6.15	0.79	-	-
5	30% PH	500	13.5	25	18.03	2.77	-	11.0/6.8
6	30% AH	500	13.6	25	29.28	8.12	6.68	9.854/7.415
7	50% PH	300	13.53	25	15.95	3.75	2.57	11.09/7.074
8	50% AH	300	13.65	25	28.88	10.33	7.63	9.726/7.426
9	10% PH	500	13.62	25	10.73	1.31	-	10.96/6.828
10	10% PH	500	13.5	40	10.82	1.67	-	10.99/6.820
11	50% PH	300	13.48	40	14.32	4.19	2.58	11.43/7.685
12	30% MEA	522.5	13.38	25	187.10	127.03	100.8	13.3/8.700
13	30% MEA	512	13.42	40	154.80	123.53	96.41	13.3/8.784
14	50% PH	300	13.57	75	11.67	5.13	3.89	11.087/8.38

AH – ammonium humate, PH – potassium humate, MEA - ethanolamine

According to the obtained data, it can be seen that the AH solvent has a higher absorption capacity than the PH solvent. An increase in salt concentration causes an increase in absorption capacity. This effect is especially noticeable for ammonium humate. The maximum absorption capacity was 10.33 g CO₂/kg using 50% HA (T=25°C). The best absorption capacity of

potassium humate was 4.19 g/kg when using 50% HA (T=40°C). Diluted solvents (1% HA and 1% HA) have the same absorption capacity as distilled water and are therefore ineffective for absorbing CO₂.

Despite the significantly lower absorbing capacity compared to MEA (about 10 times), the use of humic salts to capture CO₂ can be justified. Namely, the advantage is that the solution of humic salts after absorption can be used as a fertilizer. In addition, it is harmless and non-toxic. Therefore, it can probably be considered a green solvent. Further research is needed to determine the effect of temperature and to learn more about the chemistry between humic salts and CO₂.

Acknowledgements. The work was carried out within the framework of project No. IRN AP14871353 on the topic "Research of the scientific foundations and development of technology for obtaining organomineral-fertilizer by absorbing greenhouse gases with natural humic substances", funded by the Science Committee of the Ministry of Education and Science of the Republic of Kazakhstan.

Humic substances as biologically active compounds in biomedical technologies

Studies on latent and soluble polyphenol oxidase from *Moringa oleifera* Lam. leaves

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Keywords: *Moringa oleifera*, polyphenol oxidase, novel peptide, amino acid sequence, kinetics analysis

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Polyphenol oxidase (PPO) from *Moringa oleifera* leaves was investigated to unravel its unique properties that can be exploited for possible biotechnological application(s).

Soluble and latent PPO were extracted and purified to homogeneity using a combination of three-phase partitioning and gel filtration chromatography. The purity and molecular weight of the purified protein were estimated by SDS-PAGE and further by nano liquid chromatography-electrospray ionization tandem mass spectrometry (Nano-LC-ESI-MS/MS) and amino acid composition determination. Physicochemical and kinetic properties of the purified *Moringa oleifera* PPO (*mo*PPO) were investigated following established protocols.

Two soluble isoforms and a latent form of PPO were purified from the leaves of *Moringa oleifera*. All purified proteins were monomeric with molecular weights ranging from 10-65 kDa. PPO with the lowest molecular weight was further confirmed to be 7.6 kDa, 10.5 ± 0.06 kDa 7.2 kDa, and 8.61 ± 1.69 kDa as estimated by Nano-LC-ESI-MS/MS, SDS-PAGE, amino acid composition determination, and SEC respectively. The soluble proteins have pH optima at pH 6.5 and slightly basic pH 8.0, and that of the latent PPO was found to be 5.5 and 8.0 respectively. The optimal temperature for the purified enzymes was between 15 °C – 25°C and their catalytic efficiency was between $0.4 - 1.9 \times 10^5 \text{ M}^{-1}\text{s}^{-1}$.

The study concluded that *M. oleifera* leaves contain at least three forms of PPO, a novel low molecular weight peptide enzyme, a soluble form and a latent form with high k_{cat}/K_m similar to those obtainable from other sources which could be of interest in biotechnological applications.

Acknowledgements. We sincerely appreciate the Tertiary Education Trust Fund (TETFUND) and the Senate of Obafemi Awolowo University for providing financial support for this investigation.

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Assessment of the complexing ability of humic substances in natural substances and fertilizers in relation to metal ions

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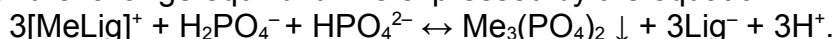
Keywords: stability constant, humic acids, complex compounds, iron (III), copper (II), magnesium, lead

<https://doi.org/10.36291/HIT.2022.051>

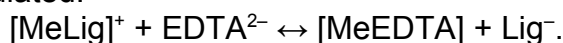
The value of the stability constant (Kst) characterizing the strength of complex compounds can be determined by various methods: chromatographic, extraction, electrochemical (capillary electrophoresis method, potentiometric titration method), kinetic (absorption spectroscopy method, electron paramagnetic resonance method, turbidimetric method, fluorometric method). However, despite the undeniable advantages of each of the listed methods, none of the methods is suitable for determining the stability constants of humic substances (HS), while complex formation is the main property of humic acids (HA) due to the presence of functional groups of acids: carboxylic, phenolic, carbonyl, which bind metal ions as in natural waters, and in fertilizers.

The activity of HA to form complex compounds with iron (III), copper (II), magnesium and lead ions was compared with the activity of metal ions in model solutions containing salicylic, sulfosalicylic, phthalic acids and EDTA. It was found that the complexing ability of HA with iron (III) is 4 times higher than with salicylic acid, and with phthalic acid – 2 times and can be comparable with EDTA. In relation to lead (II) ions, the activity for the formation of complex compounds in natural impurities can be 2 times higher than that of sulfosalicylic acid (HIT 2014).

To quantify HA to the formation of complex compounds with metal ions: magnesium, calcium, zinc, a method was used in which, as a model reaction, the reaction of formation of a coarsely dispersed system (CDS) containing a phosphate buffer. When a substance forming complex compounds with metal ions is introduced into the reaction system, simultaneously with the CDS formation reaction, the complexation reaction occurs. The reaction of the combined heterogeneous-ligand-exchange equilibrium is expressed by the equation:



This method allows you to calculate the activity of the studied ligand in comparison with the activity of EDTA (standard with a complexing activity of 1.00). To compare the experimental values of the activity of the organic ligand with the known value of the instability constant of its complex, the equilibrium constants (K_{eq}) of the reactions of the corresponding complexes of metal ions with EDTA are calculated:



According to this method, the complexing properties of HS in fertilizers were evaluated (HIT 2017). In addition, experimental data on the complexing properties of drugs – fluoroquinolones were obtained using the same method: the complexing activity of levofloxacin and moxifloxacin exceeds the activity of citrate ions and glycine, but does not exceed the activity of EDTA.

The activity of complex compounds of natural organic substances with metal ions (calcium, magnesium, zinc) before and after the introduction of sodium hypochlorite was also evaluated by the above method. It was found that after chlorination, the complexing ability increases by 2 times and is comparable to EDTA.

Thus, this technique confirms the fundamental possibility of using it to assess the complexing activity of organic ligands (including HA) in relation to some metal ions.

Antioxidant activity of iron-containing humic compounds with antianemic activity

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Keywords: humic substances, iron(III) hydroxocomplexes, iron deficiency anemia, antioxidants

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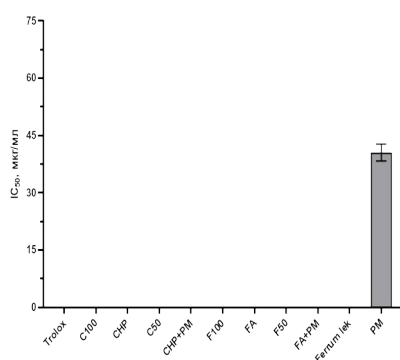
Globally iron deficiency anemia (IDA) affects 2% of the male population and up to 10% of the female population. The main treatment approach for iron deficiency anemia is iron replacement therapy, but there is a risk of oxidative stress development. Thus, the development of new iron containing drugs with antioxidant properties is very relevant. As a new methodological approach in the IDA treatment drugs synthesis a selective addition of the humic substances (HS) fractions carrying functional groups with close acidity was used. The targeted selection of the humic ligands with specific acid, iron-binding, and redox properties capable of both binding ionic iron(III) and stabilizing iron(III) hydroxocomplexes was done.

Experimental samples of iron-containing humic compounds were synthesized in the Natural Humic Systems Laboratory of the Chemical Faculty of Lomonosov MSU, there were 10 samples: CHP – original HS sample (the coal humic acids "Powhumus"), C100 (Fe(III) hydroxide complex with CHP), CHP+PM (CHP with polymaltose, 1:1), C50 (Fe(III) hydroxide complex with CHP and polymaltose, 1:1), FA - original HS sample (the peat fulvic acids "Fulvagra"), F100 (Fe(III) hydroxide complex with FA), FA+PM (FA with polymaltose, 1:1), F50 (Fe(III) hydroxide complex with FA and polymaltose, 1:1), PM - polymaltose and Ferrum Lek® - a reference control.

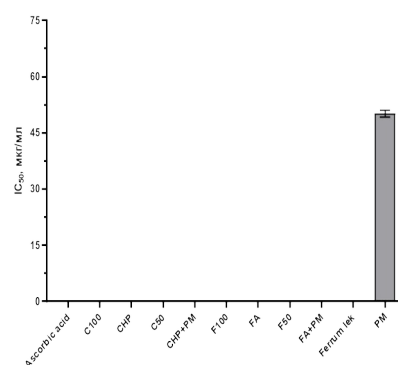
The study of antioxidant properties with used ABTS-test and NST-test (with O₂^{-•} -in non-enzymatic generation) of the Ferrum Lek®, HS and iron-containing humic compounds was done.

It was shown that an introduction of iron into the humic matrix significantly reduces their antioxidant activity in comparison with the original HS according to ABTS-test and NST-test. The preparation Ferrum Lek® showed lower antioxidant properties in comparison with the experimental samples of iron-containing humic compounds.

a)



b)



Values of IC₅₀ index for Ferrum lek and the experimental samples of iron-containing humic compounds and positive control preparations (Trolox, Ascorbic acid) in experiments: a) ABTS-test (ABTS^{•+}) and b) NST-test (with O₂^{-•} - in non-enzymatic generation).

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Cytotoxic properties of humic substances-containing wound healing ointments

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Keywords: humic substances, cytotoxicity, ointments, wound healing properties

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The cytotoxic properties are important indicators for compounds promising in the development of wound healing agents for local applications like ointments.

In the experimental design following objects were included: basic ointment base - Vaseline; commercially available Lincomycin 2% ointment; a complex ointment Lincomycin 2% + Protargol 2%; complex ointments Lincomycin + CHP-Ag (silver-containing complex based on humic matrix) with different concentrations of active components (Table 1). Cytotoxicity was assessed *in vitro* in 3T3-L1 and HepG2 cell cultures. The experimental protocol complied with ISO 10993-5:2001 Biological evaluation of medical devices - Part 5: Tests for *in vitro* cytotoxicity. The method of extracts was used. The level of cells viability was assessed by the method using Neutral Red Vital Dye, followed by spectrophotometric detection at OD 540 nm on a Tecan Infinite 200Pro plate reader.

All experimental data were subjected to statistical analysis. The value at which the viability of a cell line after incubation with ointment extracts for 24 hours was decreased by 50 percent (IC_{50}) was calculated. All calculations were performed using Prism GraphPad 8.0 software (USA).

The results of cytotoxicity assessment are presented in Table 1.

Table 1 - Values of ointment extract concentrations at which cell viability was reduced by 50 percent (IC_{50})

The ointment's composition	IC_{50} 3T3-L1, extract concentration (%)	IC_{50} HepG2, extract concentration (%)
Vaseline	n.d.	n.d.
Lincomycin 2%	58.24	143.52
Lincomycin 2% + Protargol 2%	38.46	103.84
Lincomycin 2% + CHP-Ag 2%	55.29	81.36
Lincomycin 0.2% + CHP-Ag 2%	n.d.	n.d.
Lincomycin 2% + CHP-Ag 0.5%	50.82	102.39

Note: n.d. - the decrease in cell culture viability after incubation with the test object for 24 h was practically undetectable.

As a result of the experiments, it was found that all the studied samples have greater cytotoxic properties against the normal fibroblast cell line (3T3-L1) than against the hepatocellular carcinoma cell line (HepG2). The manifestation of cytotoxic properties is more associated with the presence of an antibacterial agent (Lincomycin) in the composition of the ointment. The lowest toxicity was shown for the complex ointment with minimal antibiotic content (Lincomycin 0.2% + CHP-Ag 2%) - a decrease in cell viability was not actually registered. A similar result was shown for the ointment base - Vaseline. Changing the content of the humate-containing active ingredient (CHP-Ag) actually had no effect on the change in the overall cytotoxicity of the complex ointments.

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Clinical studies of humic acids

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Keywords: humic acid, humaton preparation, antioxidant, malondialdehyde

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According to the studies on the clinical properties of humic substances conducted by the Institute of Chemistry and Chemical Technology, Mongolian academy of Sciences and Garaany Uujim Bulgan company, in cooperation with the Institute of Veterinary Medicine and other organizations and researchers, humic acids isolated from Mongolian lignite /leonardite, and feed additives and its preparation "Humaton" has been found to support the growth and development of animals, stimulate the immune system, and have antioxidant effects.

For example, the humic acid preparation given to animals reduces the number of inactive 0 lymphocytes in the blood of animals by 26%, increases the number of T and B cells by 26-38%, activates the immune system, its antioxidant activity is similar to that of albumin protein, and it reduces the content of malondialdehyde produced in the peroxidation process by 4 times. According to the research results of Dr. Y. Ganbold, Institute of Veterinary Medicine, the phagocytic activity of the blood of calves given Humaton increased by 3.8%, the number of phagocytes by 22.3%, and the phagocytic integral index by 25.0%.

The average daily increase in live weight of calves given Humaton was 12.6% and 7.5% in heifers, while the increase in live weight during the 60-day period of two – year old lambs and goats was 20-25%. Our tests carried out in cooperation with the Agricultural Research Station of the Highland Region, Ikhtamir and Chuluut Soum in Arkhangai province against the disease of seagulls and scurvy gave positive results with more than 95%, the tests against gastroenteritis in young and large animals - 92%, respectively,

Farmers who use the product proudly talk about "not only the fatness of the animals and the milk yield of the cows have increased, but they have not stopped milking even in the winter and spring seasons, their tired young animals have been able to enter the year fully, and their horses have galloped and won in Suma, provincial and regional festivals." Humaton preparation is a product with Mongolian standards and registered in the register of animal and animal feed additives. Thus, it is clear that the use of humic acid preparation is of great importance in increasing the efficiency of animal husbandry production and improving the health of livestock.

Effect of humic products on the antagonistic properties of micromycetes

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Keywords: humic products, soil fungi, pathogen, antagonistic properties, *Trichoderma*

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Antagonist fungi of the genus *Trichoderma* can act as natural suppressors of phytopathogenic soil fungi [1, 2]. The suppression of phytopathogenic activity is influenced by abiotic (amount of organic matter - OM) and biotic factors. We studied antagonistic properties of *T. harzianum* against the phytopathogens *Alternaria alternata* and *Fusarium oxysporum*. Antagonistic activity was studied by the co-culture assay [1] on Czapek-agar medium with 3 g/L sucrose and with various OM content. The radius of the pathogen colony was measured both facing the antagonist colony and in the perpendicular direction. Various OM content was created by introducing humic products (HPs) from lignosulfonate (HP1) and coal (HP2) at 0.02 and 0.1 g/L.

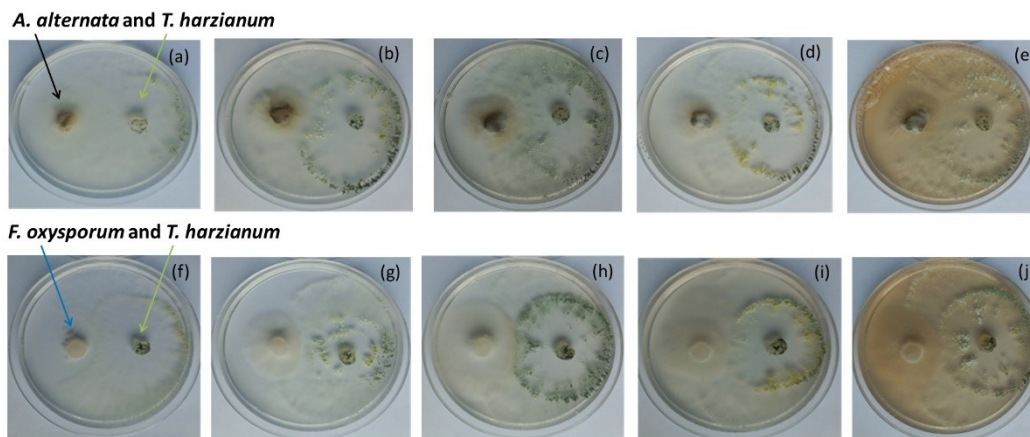


Figure 1. Effect of humic products on interaction of micromycete colonies:

A. alternata - *T. harzianum*: (a) control; (b) 0.02 HP1, (c) 0.1 HP1, (d) 0.02 HP2, (e) 0.1 HP2;

F. oxysporum - *T. harzianum*: (f) control; (g) 0.02 HP1, (h) 0.1 HP1, (i) 0.02 HP2, (j) 0.1 HP2.

HPs significantly activated the growth of *F. oxysporum* and *T. harzianum*, but not *A. alternata*. When HPs were added *Trichoderma* can act as a more active antagonist in relation to the slowly growing *A. alternata* (Fig. 1b-e). As we have shown earlier for *A. alternata* - *T. viride* [2]. In the pair *F. oxysporum* - *T. harzianum*, HPs did not give a competitive advantage to any of the species. Thus, an increase in the content of OM does not always determine the antagonistic activity of micromycetes in relation to pathogens. In some cases, the biological potential of fungal species is of decisive importance.

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Survival of lactic acid bacteria in silanol-humate gels

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Keywords: silanol-humate gels, humates, bacterial survival, lactic acid bacteria, biopreparations

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The problem of preserving the viability of lactic acid bacteria (LAB) in dairy products and various biological products is one of the key problems in the food and pharmaceutical industry. Therefore, the search for new ways to increase the viability of bacteria during storage and increase the shelf life of products is relevant. It was previously shown that when included in silanol-humate gels (SHG), the titer of viable cells of hydrocarbon-oxidizing bacteria during long-term storage (2 years) increases significantly (by 2 orders of magnitude) [1]. The gel was developed at the Department of Medical Chemistry and Fine Organic Synthesis of the Chemical Faculty of Moscow State University [2]. Gels are able to release cells from their matrix when dissolved in water. We suppose that the reason for increasing the viability of bacteria is their polymer environment and the presence of energy sources (ethanol, acetate). In this work we investigated the effect of immobilization of lactic acid bacteria *Lactobacillus paracasei* AK508 and *Enterococcus faecium* M3185 cells in SHG based on Powhumus humates on their survival during long-term storage. When preparing the gel, five different organic acids were used as titrants, which, according to our hypothesis, can be used by bacteria as an energy substrate.

It was shown that when *E. faecium* cells were immobilized in a silanol-humate gel with malic, lactic and citric acids and then stored for a month, the titer of viable cells was six, three and three times higher, respectively, than in the control (1.1% of the initial level). When storing *L. paracasei* cells in gels with ascorbic acid for a month, the titer of viable cells remained at the level of 32% (32 times more than in the control), with the addition of malic acid – at the level of 16% (16 times more than in the control), lactic and citric – at the level of 4.5-5% (in 4.5-5 times more than in the control). Such protective effect of SHG was observed during 4 month. High technological capacities of cultures in SHG were kept also.

Ascorbic acid can be fermented by LAB to form lactate and formate (from which acetate is subsequently formed), and malic acid is fermented to lactate. Thus, ascorbic and malic acids can be a source of energy for maintaining the vital activity of the LAB [3,4].

Thus, the data obtained show the possibility of using SHG for long-term storage of various microorganisms and the importance of an energy source for maintaining bacteria in a viable state.

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Research of zinc-containing humic compounds wound-healing properties and zinc assay content in biomaterial after their topical application

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Keywords: humic substances, zinc, vulnerary properties, topical application

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Zinc is present in all organs, tissues, fluids and secretions of the body, mainly in the skin, hair and bone tissue. More than 95% of all zinc in the body is contained in cells, it is involved in almost all stages of cell growth. Zinc is most often prescribed for medical purposes, such as for the treatment of skin ulcers, dermatitis, acne, and in antiviral therapy.

The goal of this work was to research the zinc-containing humic compounds wound-healing properties with an improved safety profile on an aseptic wound model by rats and to assay content zinc accumulation processes in animal hair.

Experimental samples of zinc-containing humic compounds (Zn-HS) were synthesized in the Natural Humic Systems Laboratory of the Chemical Faculty of Lomonosov MSU, there were 6 samples: Zn-CHP – Zn and the coal humic acids (HA) "Powhumus", Zn-FA – Zn and the peat fulvic acids (FA) "Fulvagra", Zn-CHS - Zn and the Sakhalin coal HAs, Zn-Peat1 - Zn and the angustifolium peat HAs, Zn-Peat2 - Zn and the sphagnum peat HAs, and zinc sulfate (ZnSO₄) - a reference control.

The wound-healing properties of zinc-containing humic compounds were research on an aseptic wound model by rats. The 20 mm diameter round-shaped wound was done in the area below the shoulder blades of rats by an excision method of the skin and a subcutaneous tissue layer. Experimental samples and ZnSO₄ were injected into wounds at 1.67 mg/ml Zn concentration. The general animal's condition was assessed daily; the planimetric analysis method of wounds was used on the 3rd, 5th, 7th, 9th, 11th, 13th, 15th, 17th, 19th and 21st days. The analysis of the zinc assay content in biomaterial was performed on a quadrupole mass spectrometer with inductively coupled plasma by Agilent 7500cx (Agilent Technologies, USA).

A statistically significant decrease in wound size during treatment with experimental samples Zn-CHS, Zn-Peat1 and Zn-Peat2 was established on the 5th day of the experiment, and with samples Zn-CHP and Zn-FA - on the 7th day, in comparison with the group of animals treated with ZnSO₄. Thus, the course application of zinc-containing humic compounds accelerates the healing of aseptic wounds, in comparison with a reference control.

The zinc content in the animal hair from rats after external application of experimental samples of zinc-containing humic compounds (Zn-HS) is comparable with the physiological values (160.2±3.8 mcg/ml). Thus, the animal hair from rats treated with samples of ZnSO₄, Zn-CHP and Zn-FA contained 120.0±11.9; 81.2±21.5 and 111.7±5.7 mcg of elemental zinc in 1 g of the dry animal hair, respectively. Higher values of zinc contain in animal hair samples were established for Zn-CHS, Zn-Peat1 and Zn-Peat2 samples, namely, 274.9±16.6; 268.3±23.6 and 259.4±8.7 mcg of elemental zinc per 1 g of the dry animal hair, respectively. Thus, higher values of zinc contain in the animal hair from rats after using the experimental samples Zn-CHS, Zn-Peat1 and Zn-Peat2 can indicate their cumulation property in the body tissues.

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Antibacterial materials based on levofloxacin and mesoporous polymer matrices

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Keywords: antibacterial materials, mesoporous polymers, levofloxacin, crazing

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Challenging demands of XXI century pose the urgent necessity for the development of innovative antibacterial eco-friendly and low-cost materials with high performance, prolong lifetime, and good mechanical characteristics. This work addresses the preparation and characterization of new antibacterial polymeric nanocomposite materials containing antibacterial broad-spectrum medicine levofloxacin (LF), which is the third generation fluoroquinolone. Host mesoporous matrixes based on high-density polyethylene (HDPE) and poly(ethylene terephthalate) (PET) were prepared by the strategy of environmental crazing in the "green" mode using the biphasic oil-in-water emulsions as crazing-promoting environments. Introduction of LF into the mesoporous host matrixes was performed by force and passive wet impregnation. Within porous matrixes, LF is shown to be uniformly distributed throughout the whole volume in the nanoscale state as nanoparticles with dimensions within ~2–20 nm. The content of LF in the host matrixes can be controlled by the concentration of LF feed solution and porosity of the host matrix. In the LF/HDPE under study, the content of LF in HDPE/LF nanocomposites is $\omega = 0.8 \pm 0.2\%$. The method of equilibrium dialysis shows that the release of LF from the polymer matrix (PBS, pH 7.4, 37°C) proceeds with a low release rate: within the first 40 min, up to 15% of LF is released, and within 7 days, less than 20%. The rapid release of the drug at the initial stage is provided by dissociation of drug molecules from the surface of mesoporous HDPE films. Hence, these materials show the prolong drug release. To assess the antibacterial performance of the LF/HDPE materials, bacterial strains were seeded on a solid nutrient medium and incubated at 37°C for 24 h. The inhibition zone (S_{inh}) was measured. The antibacterial performance (A) is evaluated as the ratio of S_{inh} to the content of LF in the composite material. For *Lactobacillus plantarum* 8P-A3, $A = 78 \pm 5 \text{ m}^2/\text{g}$, and for *Escherichia coli* ATCC 25922 $A = 420 \pm 20 \text{ m}^2/\text{g}$. To study the prolong antibacterial action of the proposed materials, the cell suspension, nutrient medium, and LF/HDPE sample were incubated at 37°C upon stirring. The tests show that, within 5 days, no bacterial growth was detected.

Hence, innovative and high-performance polymeric materials for biomedical purposes with high antibacterial properties and prolonged drug release were prepared, and this approach offers new benefits for the development of a broad scale of antibacterial materials for wound healing and protection.

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The drug plants extracts and humic substances based functional nutrition product development to improve the quality of men's health "GentlemenHum"

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Keywords: humic substances, drug plants, functional nutrition, men's health

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Every year in Russia and the CIS countries the number of men suffering from diseases of the genitourinary system increases. Chronic stress, harmful working conditions, deficiencies of micro- and macronutrients in food, unhealthy habits - all this leads to serious problems: prostate adenoma, prostatitis, impotence, infertility, obesity, etc. In this regard, the development of functional nutrition products for the prevention of men's diseases is an urgent area of preventive medicine.

The goal of this work was to development the drug plants extracts and humic substances (HS) based functional nutrition product to improve the quality of men's health and investigate its safety parameters.

The product contains extracts from drug plants and the low-mire peat HS. *Hippophae rhamnoides* L. fruit extract contains many vitamins, has regenerative, wound healing, antiulcerogenic, general strengthening effect. Extract from *Viburnum opulus* L. fruits has cardiotoxic, hypotensive effect, lowers blood cholesterol concentration. *Levisticum officinale* Koch. roots extract has a relaxing and antifatulent effect, increases libido. *Hedysarum neglectum* Ledeb. roots extract has androgenic and anti-inflammatory effects. *Rhaponticum carthamoides* Willd. roots and rootstock extract has anabolic and psychostimulant action, reduces alcohol cravings. The peat HS are effective immunomodulators, have antitumor and antiallergic effects, also have pronounced antioxidant and antiradical properties [1, 2]. The use of these plant extracts and HS together contributes to the amplification of major biological effects for the prevent diseases of the men's genitourinary system and eliminates the occurrence of side effects. The quantitative content of the main active componentry, chemical and microbiological safety was investigated according to the Pharmacopoeia XIV of Russia.

The product "GentlemenHum" numeric indicators was established: pH 7.68, weight fraction of dry substances 4.02%, one portion of the product contains polyphenolic compounds in terms of gallic acid 98 mg, tannins in terms of tannin 326 mg, organic acids in terms of malic acid 52 mg, carotenoids in terms of beta-carotene 6 mg, HS 20 mg, ecdystene and ecdysterone 12 mg. The content of radionuclides complied with the standard: Cs-137 not more than 400 Bq/kg; Sr-90 - 200 Bq/kg. The content of heavy metals and arsenic complied with the norm: Pb not more than 6.0 mg/kg; Cd - 1.0 mg/kg; Hg - 0.1 mg/kg; As - 0.5 mg/kg. The microbiological purity corresponded to the established category 3B of microbiological purity by parameters the aerobic microorganisms total number, yeasts and molds, absence of *Escherichia coli*, absence of the genus *Salmonella* bacteria, absence of *Staphylococcus aureus*, enterobacteriaceae resistant to bile.

Thus, the chemical and microbiological safety parameters correspond to those established by the Pharmacopoeia XIV and the Technical Regulations of the Customs Union TR CU 021/2011 "On food safety", in accordance with the safety requirements adopted in the Russia.

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Experience in the use of humic ointment for the treatment of burns

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Keywords: ointment, potassium humate, thermal burn, cure

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Mechanistic studies done by my research group on brown coal derived potassium humate indicates that potassium humate inhibits the activation of complement, the expression of adhesion molecules and the release of cytokines associated with inflammation [1].

Kazakhstan potassium humate was used to produce Gumaz ointment, the main advantages of which are the absence of antibiotics, preservatives, hormones. Indications for the use of ointment are conjunctivitis, keratoconjunctivitis of non-infectious, infectious and invasive etiology, as well as wounds, burns, dermatitis [2].

The ointment was used to treat a thermal burn of the 2nd degree of the proximal phalanx of the index finger of the left hand in a volunteer (female, 56 I.). The drug was applied to the wound area, capturing healthy tissue up to 0.5 cm (Figure 1).



Figure 1. Thermal burn of the proximal phalanx of the index finger of the left hand and the stages of its healing in the treatment with humic ointment

During the treatment, it was revealed that Gumaz ointment facilitates the course of the wound process, has an antimicrobial effect, reduces soreness and local irritation immediately after application at the burn site, stimulates the growth, development, reproduction of cells, which accelerates regeneration. The antimicrobial effect was manifested in the absence of reproduction of pyogenic bacteria, despite the absence of a bandage and the patient's work with raw fish and vegetables. On day 9, the process was resolved by the rejection of a soft crust without the formation of scars. Active granulation of cells, absence of edema, soreness were noted. The bottom of the wound had a pink color; erosion, bleeding foci were absent.

The increased content of biologically active substances that make up the veterinary drug in an accessible chelated form; simplicity and convenience of use, complete safety of the drug used, ensure high rates of therapeutic activity of the ointment in the treatment of thermal burns.

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Development and quality control of humic ointment

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Keywords: eye ointment, potassium humate, antimicrobial properties, conjunctivitis

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Various preparations are known for the treatment of lesions of the mucous membranes and skin of animals of various etiologies [1], for example, "Tetracycline eye ointment 1%"; ointment "Misophen"; gel for external use "Ophthalmo-gel"; a drug for the treatment and prevention of infectious keratoconjunctivitis of cattle containing an aqueous solution of Sulphur, Echinacea purpurea, Hepar sulphur, Belladonna albus, Apis mellifera and tissue nosode; a drug for the treatment of animals with mucosal and skin lesions, including a solution of cefazolin, novocaine and dexamethasone and others. The disadvantages of known drugs include the presence of antibiotics or chemical components in their composition, because of which restrictions are imposed on animal products according to veterinary and sanitary rules.

The uniqueness and effectiveness of the veterinary drug ointment "Gumaz", intended for the treatment of animals with lesions of the mucous membranes and skin of various etiologies, is due to the use of local potassium humate – a natural source of mineral and biologically active substances. Potassium humate obtained from brown coals of Kazakhstan contains a high concentration of the active substance (the mass fraction of humic acids in terms of dry matter is 56%), a complex of minerals is presented as additional components: sodium, magnesium, potassium, calcium, iron, copper, zinc, selenium and a number of other trace elements. Also, domestic potassium humate has a pronounced antimicrobial effect against bacteria and micromycetes [2].

The quality control of the ointment showed that the eye ointment "Gumaz" in appearance is a preparation with a weak nonspecific odor of a homogeneous jelly-like consistency of brown color, due to the presence of humic substances, and the formation of a black-brown liquid with prolonged storage of up to 10%. The drug is sterile packaged in syringes or jars. The hydrogen index (pH) of the aqueous extract of the drug is $6.5-8.5 \pm 0.5$ units. Eye ointment is non-toxic and harmless, does not cause negative effects on the body of laboratory mice. The use of the veterinary drug "Gumaz" ointment in experimental rabbits causes healing and disappearance of signs of the disease by 5-6 days. Treatment of calves with "Gumaz" ointment helps to reduce the recovery time for various acute forms of conjunctivitis. Animals with acute catarrhal keratoconjunctivitis recover on the second day; with chronic – on 3-4 days; with infectious purulent keratoconjunctivitis – on 7-8 days in 80% of animals; with fibrinous keratoconjunctivitis in 20% of animals – recovery is noted on 25-28 days from the start of treatment. Recovery in the treatment of skin lesions of various etiologies with the veterinary drug "Gumaz" ointment occurs on average 3-4 days earlier, wounds on the skin heal by primary tension.

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The effects of silver and zinc-containing humic pharmaceutical compositions on the functional state of peritoneal macrophages

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Keywords: humic substances, silver, zinc, macrophages, nitric oxide, arginase, endotoxin

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Humic substances (HS) are promising biologically active substances with antibacterial, wound-healing and interferogenic activity. The new pharmaceutical metal-organic compounds created on their basis will make it possible to produce innovative antibacterial and wound-healing medicines. The goal of this work was to study the effect on the peritoneal macrophages functional state of the original humic substances samples (HS), the samples silver nanoparticles ultradispersed in humic substances (HS-AgNPs) and zinc complexes humic compounds (HS-Zn).

Experimental HS-AgNPs and HS-Zn samples were synthesized in the Natural Humic Systems Laboratory of the Chemical Faculty of Lomonosov MSU, there were 21 samples: the coal humic acids (HA) "Powhumus" (CHP, CHP-AgNPs, CHP-Zn), the peat fulvic acids (FA) "Fulvagra" (FA, FA-AgNPs, FA-Zn), the Sakhalin coal HAs (CHS, CHS-AgNPs, CHS-Zn), the high-moor peats HAs: the angustifolium peat HAs (Peat1, Peat1-AgNPs, Peat1-Zn) and the sphagnum peat HAs (Peat2, Peat2-AgNPs, Peat2-Zn), the Irkutsk coal HAs (CHI, CHI-AgNPs), the "Genesis" coal HAs (CHE, CHE-AgNPs), the unfractionated peat preparation (PHF-T3, PHF-T3-AgNPs). Also, the colloidal silver preparation Protargol® and zinc sulfate (ZnSO₄) as reference controls were used.

The study of immunomodulatory properties (activity balance of NO-synthase / arginase) of the HS, HS-AgNPs and HS-Zn samples in the cell culture of peritoneal macrophages, including relative to LPS-stimulated control was done. Conventional mice of the C57BL/6 line were used. The nitric oxide (NO) production analysis was done by the nitrites content in the supernatants with used Grace reagent (Sigma-Aldrich, USA) by colorimetric method. The arginase activity analysis was done by the urea concentration in the cell lysate with used the Urea-450 test system (Bio-LA-Test, Czech Republic) by colorimetric method. Also, the admixture of endotoxin with used polymyxin B ("InvivoGen", USA) and cell proliferation were evaluated by colorimetric method (Titertek Multiskan® MCC, "Labsystems").

Some HS samples (FA, CHS, CHE, Peat1, Peat2) contributed to the polarization of peritoneal macrophages according to the classical type (M1) by increasing the activity of NO-synthase and inhibition of arginase. However, the Peat1 and Peat2 samples stimulating effect can be associated with an admixture of endotoxin in these samples. The CHP sample intensified its NO-stimulating properties against the background of a stable arginase state. At the same time, all these samples did not influence a toxic effect on cells. Experimental HS-AgNPs samples (CHP-AgNP, PHF-AgNP, CHS-AgNP and CHE-AgNP) activated alternative properties of peritoneal macrophages (M2), they also inhibited the proliferation of the tested cells, except for the CHE-AgNP sample. The others HS-AgNPs samples and Protargol® did not influence on the NO-synthase and arginase activity. Samples FA-Zn and Peat1-Zn showed the highest stimulating effect in the activation of NO-synthase and arginase, but their stimulating effect was associated with an admixture of endotoxin. Among non-pyrogenic samples, only CHS-Zn sample had the greatest activity, and only in relation to arginase

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Study of the slow release of Zn²⁺ and Ag⁺ from gels loaded with ZnO and Ag nanoparticles

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Keywords: ZnO, Ag, slow release, gels, humics

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Zinc oxide is known for its wound healing properties. It is used in various gels and ointments for wound dressing, and an important characteristic of such substances is the release of zinc in ionic form. Previously, we have developed a method for the synthesis of zinc oxide nanoparticles in humic media and methods for loading it into gels, with the addition of silver nanoparticles that perform an antibacterial function. Gels containing 2% Zn, 0.5% Ag, and 2% carboxymethylcellulose (CMC) as a gelling agent were the most promising in terms of physicochemical properties and content of metals.

The kinetics of the release of the ionic forms of Zn²⁺ and Ag⁺ was estimated from the fraction of the equilibrium concentration of ions in the permeate, estimated after dialysis of the gels through a membrane with an average pore size of 13500 kDa. As solutions for dialysis were used: to control the influence of competing ligands: Muller-Hinton broth (MH) and 0.2M Tris buffer pH=7.4; to control the effect of pH solutions with pH = 6, 7 and 8; to control the effect of ionic strength: distilled water, 0.09% and 0.9% aqueous sodium nitrate solution. The figure below shows an example of determining the release of metal ions from a gel against MH

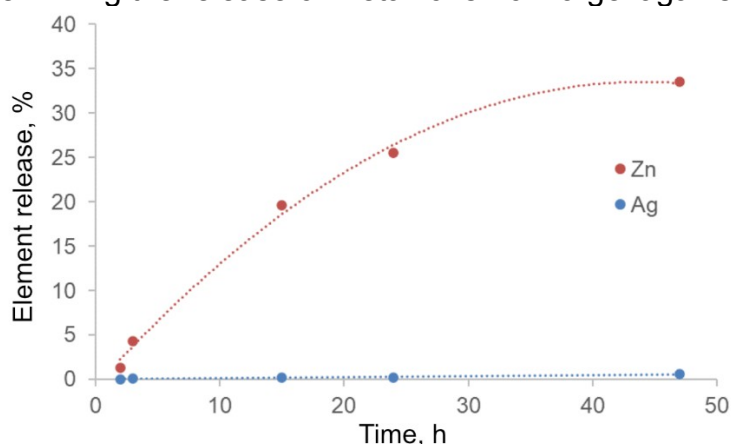


Figure 1. Release of ionic zinc and silver from the gel against MH.

The results of the evaluation of the release of Ag and Zn ions from the gel showed a pronounced dependence of the rate and concentration of released ions depending on the nature of the dialysis solution and the composition of the gel. Experimental data indicate the preferential release of zinc ions from the gel into the dialysis solution containing competing ligands. For silver, the highest yield was found for the Tris buffer, however, the rate of silver release from the gel and the total concentration of silver that passed into the dialysis solution turned out to be significantly (in some cases, an order of magnitude) lower than for Zn²⁺. In general, the maximum yield of zinc ions was found for CMC gel in MH broth - 34% after 48 hours, and for silver the maximum yield is typical for Tris buffer also from CMC - 3.1%. Thus, these gels can be considered as promising substances for wound healing.

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Sol-gel synthesis of zinc oxide nanoparticles with controlled dimensions in the environment of humic substances

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Keywords: nanoparticles, zinc oxide, humic substances

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Zinc oxide nanoparticles (ZnO-NP) demonstrate a range of biomedical applications. The sol-gel method is the most applicable synthesis for the nanoparticle obtaining. The purpose of this work was to obtain ZnO nanoparticles with controlled sizes in a medium of humic substances (HS) for use as the wound healing compositions. Addition of the humic substances (HS) coating to the ZnO-NPs may improve the colloidal stability of the nanoparticles by creating a charged surface layer, which increases electrostatic repulsive forces and steric repulsion between nanoparticles [1]. In addition, HS are natural compounds, which demonstrated no toxicity over the entire range of natural concentrations. The HS salts are soluble in water.

To achieve this goal, sodium humate from coal (CHP) was used as HS. Zinc acetate was used as a zinc precursor. The synthesis conditions were chosen as described in [1]. Synthesis was carried out in the range of HS concentrations from 0.1 to 10 g/l. The concentration of zinc acetate was 0.025 M. The synthesis was carried out by mixing the reagents at pH 12 and subsequent stirring at $T = 20^{\circ}\text{C}$ for 2 h. The result of reaction was the precipitation of an insoluble product. The precipitate was centrifuged, washed with water, and dried in a vacuum oven at $T = 60^{\circ}\text{C}$. The resulting product was characterized by XRD, UV-vis spectroscopy, and TEM. TEM images are shown in Fig. 1.

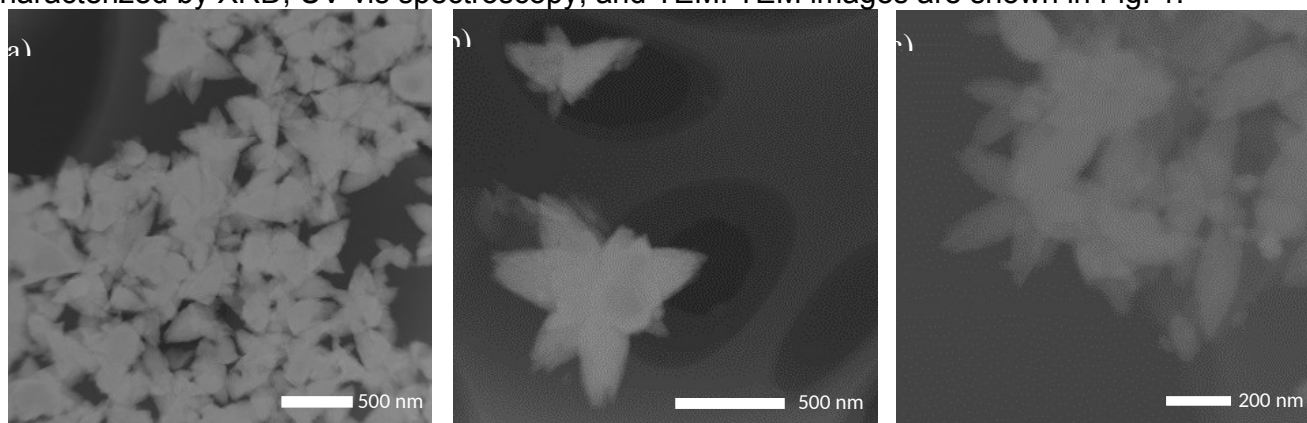


Figure 1. TEM images of ZnO nanoparticles obtained in GW solutions with concentrations (g/L): 0.1 (a); 1.0 (b); 10 (c).

According to TEM data, the size of ZnO particles was in the range of 400–500 nm. The aggregation of nanoparticles might occur as a result of the centrifugation and the absence of the HS macroligands. In accordance with the XRD data, an increase of the HS concentration contributed to the formation of smaller nanoparticles. Experiments without washing from humic ligands are planned.

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Nootropic activity of humic substances

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Keywords: humic substances, exploratory behavior, open field (animal test)

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Nootropics are over-the-counter medications that can specifically affect cognitive and mnemastic functions both in healthy individuals and in patients with organic brain lesions due to their neurometabolic, neurotransmitter, and vasotropic effects. Moreover, they have a high level of safety and are very popular among the adult population of the world (they are often called "drugs of the XXI century"). Therefore, the nootropic activity presence at the natural biologically active substances is one of their competitive advantages. The goal of this work was to research the nootropic activity of humic substances (HS) aqueous solutions (by NovoLand company, Russia).

The nootropic activity of experimental HS was tested on 70 female and male CD-1 effluent mice. Animals were divided into 7 groups (n=10) to study conditioned reflex in the "open field" test and the scopolamine cholinergic dysfunction (conditioned passive avoidance reflex). The comparison drug was "Piracetam".

A course of administration (ten times) of HS at a dose of 500 mg/kg promoted the development of a conditioned reflex in intact animals (in the "open field" test) (Figure 1). A single administration of 500 mg/kg of HS against the background of cholinergic dysfunction increased the time the animal spent on the platform, but did not increase the number of animals with the developed CRPA (Table 1).

Figure 1. Effect of the experimental HS on the orienting and exploratory behavior of mice in the open field test

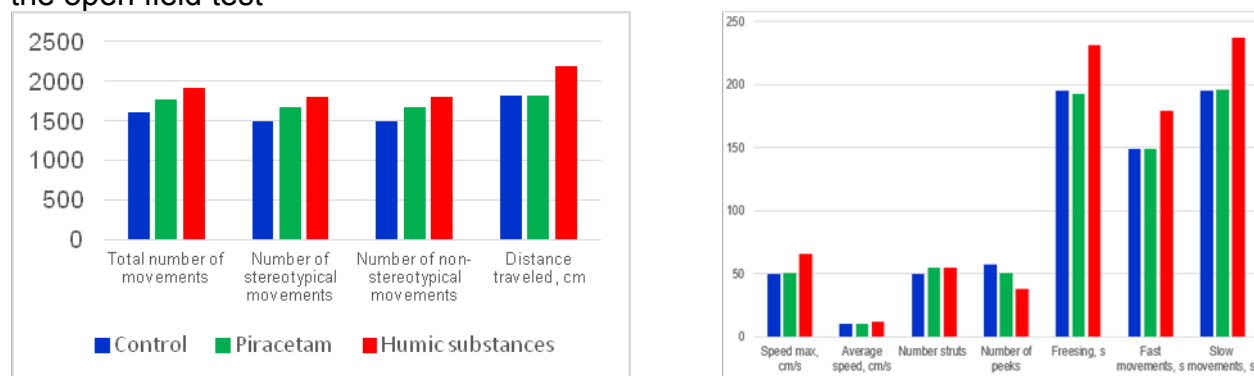


Table 1. Effect of the experimental HS on the scopolamine cholinergic dysfunction (conditioned passive avoidance reflex)

Groups	Indicator	
	the time the animal spent on the platform, sec (X±SE)	number of animals with the developed CRPA, %
Physiological solution	40.5±5.8 [#]	40
Physiological solution + Scopolamine, 1 mg/kg	13.2±5.8 [*]	10
Piracetam, 300 mg/kg + Scopolamine, 1 mg/kg	51.5±4.9 [#]	70 [#]
HS, 500 mg/kg + Scopolamine, 1 mg/kg	37±6.55 [#]	30

* differences with the group Physiological solution statistically significant, p<0.05; # differences with the group Physiological solution + Scopolamine, 1 mg/kg statistically significant, p<0.05

Antimicrobial activity of humic-based bionanomaterials containing silver against opportunistic pathogens

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Keywords: humic substances, antibiotics, bacteria, antibiotic resistance

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The antibacterial agents use led to the emergence and spread of resistant microorganisms, in some cases resistant to several classes of antibiotics. In this regard, the antimicrobial chemotherapy improvement, aimed at increasing the effectiveness of antibacterial agents and reducing the number of multiresistant strains of pathogens of infectious diseases, remains a highly relevant task.

The goal of this work was to research the antibacterial activity of humic-based bionanomaterials containing silver (HS-AgNPs) and their modification by naphthoquinone (HS-2OHNQ-Ag) and hydroquinone (HS-HQ-Ag), also study their effects on museum strains of *Escherichia coli* and *Klebsiella pneumoniae* and multidrug-resistant clinical strains of *Klebsiella pneumoniae* and Methicillin-resistant *Staphylococcus aureus* (MRSA).

Experimental samples were synthesized in the Natural Humic Systems Laboratory of the Chemical Faculty of Lomonosov MSU, there were 8 samples: the coal humic acid (HA) "Powhumus" (CHP-AgNPs), the peat fulvic acid "Fulvagra" (FA-AgNPs), the Sakhalin coal HA (CHS-AgNPs), the angustifolium (Peat1-AgNPs) and the sphagnum (Peat2-AgNPs) peat HAs, the Irkutsk coal HA (CHI-AgNPs), the "Genesis" coal HS (CHE-AgNPs), the unfractionated peat preparation (PHF-T3-AgNPs). All modification samples were synthesized on the CHP sample.

Minimum inhibitory concentrations (MIC) were determined for experimental substances with established antibacterial activity in combination with lincomycin. The study was done on the 96-well flat-bottomed tissue culture plate. Mineral nutrient medium M9 and Mueller-Hinton broth were used as nutrient medium. Results were evaluated after 24 h of incubation of the plates at a temperature of 37°C using spectrophotometry (605 nm).

As a result of the study of the effect of the studied substance on the development of periodical cultures of museum strains of *E. coli* and *K. Pneumoniae* in the mineral nutrient medium M9, it was found that CHS-AgNP, FA-AgNP, CHP-AgNP, Peat1-AgNPs, Peat2-AgNPs at concentrations of 0,5-8 mg / having pronounced antimicrobial activity, which manifests in the complete absence of bacterial growth in the wells. The antimicrobial activity of the studied substances against clinical multi-resistant strains of *K. pneumonia* was demonstrated by CHS-AgNP, FA-AgNP, CHP-AgNP, Peat1-AgNPs, Peat2-AgNPs at concentrations of 3-16 mg/l. The modification samples were studied on bacteria in the Mueller-Hinton nutrient broth and they showed the most stable results against opportunistic microorganisms. So, CHP-Ag, CHP-HQ-Ag and CHP-2OHNQ-Ag samples showed pronounced antibacterial activity at concentrations of 600-1200 mg/l to museum strains, which manifested itself in the absence of a change in the density of the contents of the well after 24 hours. The most pronounced activity was demonstrated by two substances - CHP -Ag and CHP - HQ Ag. These substances were tested on the possible potentiation of the action of lincomycin against the museum strain of MRSA. The CHP-Ag and CHP-HQ Ag samples were added to lincomycin at concentrations of 60 mg/l and 40 mg/l, respectively, as result the MIC of lincomycin was 0.1 mg/l, while without the addition of HS, it was more than 200 mg/l.

Thus, the humic-based bionanomaterials containing silver have an antibacterial effect and can potentiate the action of known antibacterial substances.

This work was supported by the financial Russian Science Foundation (Grant No. 20-65-47052).

The study of wound-healing and antibacterial properties of bionanomaterials based on humic substances and silver nanoparticles *in vivo*

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Keywords: humic substances, silver nanoparticles, antibacterial, wound-healing

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The problem of treatment of chronic wounds due to the high incidence of ulcers and pressure ulcers is of great concern in society. Particularly acute is the problem of treating such ulcers infected with antibiotic-resistant strains. For example, Ag / Zn nanocomplexes are described covalently grafted to an organic matrix, and their ability to suppress the growth of both gram+ and gram- microorganisms has been shown. The dressings with various compositions of silver has their antibacterial effect and is using to treat acute aseptic and infected wounds. Humic substances (HS) have a wide spectrum of biological activity, therefore with the creation of bionanomaterials based on HS as a matrix containing silver and zinc nanoparticles is of great interest today for the treatment of bacterial infections and for wound healing.

The goal of this work was to research antimicrobial and wound-healing properties the bionanomaterials based on humic substances and silver nanoparticles (HS-AgNPs) on a model of a septic wound.

Experimental HS-AgNPs samples were synthesized in the Natural Humic Systems Laboratory of the Chemical Faculty of Lomonosov MSU, there were 8 samples: the coal humic acids (HA) "Powhumus" (CHP-AgNPs), the peat fulvic acids (FA) "Fulvagra" (FA-AgNPs), the Sakhalin coal HAs (CHS-AgNPs), the high-moor peats HAs: the angustifolium peat HA (Peat1-AgNPs) and the sphagnum peat HA (Peat2-AgNPs), the Irkutsk coal HA (CHI-AgNPs), the "Genesis" coal HS (CHE-AgNPs), the unfractionated peat preparation (PHF-T3-AgNPs). Also, the colloidal silver preparation Protargol® as reference control was used.

The septic-inflammatory disease model of soft tissues by implanting a gauze sponge infected with a clinical strain of methicillinresistant *Staphylococcus aureus* (MRSA) into the wound space was done.

As a result, the application of CHS-AgNPs, FA-AgNPs, PHF-T3-AgNPs, CHE-AgNPs, Peat1-AgNPs, Peat2-AgNPs did not lead to a change in the proportion of the initial area of the wound surface on the 3rd and 5th days ($p>0.05$). Starting from the 7th application day of FA-AgNPs, PHF-T3-AgNPs and Protargol, also starting from the 9th application day of all the others studied substances there were a statistically significant decrease in the proportion of the initial area of the wound surface ($p<0.05$). A similar tendency to decrease the proportion of colony-forming units from their initial number was also noted for all experimental samples.

The histological evaluation of wounds after 21 days of application of the studied substances showed that at this time of observation, a pronounced phase of repair was noted in all the studied groups of skin samples of the simulated infectious wound process. However, the morphological picture of regeneration is heterogeneous. The best results of a wound defect were determined in samples taken from animals after applying the reference control Protargol and CHP-AgNPs sample. Thus, the antimicrobial activity of the CHP-AgNPs sample is higher than that of the reference drug Sialor (Protargol®), and the wound healing activity is comparable to that of the reference drug Sialor (Protargol®).

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Trans-generational carcinogenesis induced *in vivo* and its mitigation by lignin-derived composition with ammonium molybdate (BP-C2)

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Keywords: bpc2, radioprotection, lignin, polyphenols, genoprotective activity, antimutagenic activity

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Chemo- and radio-therapy are used to treat malignant neoplasms. These cause immediate and late side effects such as secondary neoplasms, disturbance of cardiovascular and central nervous systems, impairment of fertility [1]. Besides, some also exert genotoxic/mutagenic potential [2]. Side effects influence treatment strategies and, in patients with reproductive potential and good survival prognosis, genotoxic/mutagenic side effects are viewed as significant risk factors. According to Childhood Cancer Survivor Study, 30-years cumulative incidence of second neoplasms in childhood cancer survivors is 20.5% [3, 4].

To propose a strategy for ameliorating these deleterious effects we have assessed the antigenotoxic/antimutagenic activity of a novel lignin derived polyphenolic composition with ammonium molybdate (BP-C2) in a transgenerational study in BalbC mice.

Completed *in vivo* studies demonstrate adequate safety of BP-C2 (Safety Index >40). BP-C2 efficiently mitigates multiple acute radiation syndromes (gastrointestinal, hematopoietic, cutaneous). The minimum efficient doses (MED) of BP-C2 showing radiomitigative effects in murine animal models range from 35mg/kg (mouse, topical) to 81mg/kg (mouse, oral) or 93.7mg/kg (rat, oral). The MED of BP-C2 protecting bone marrow, blood and liver cells of Balb/c mice from cyclophosphamide and dioxidine is 60mg/kg [5].

Recently we have evaluated effects of oral BP-C2 on the development of lung tumors induced with 1,2-Dimethylhydrazine in the offspring of irradiated parents (Balb/C mouse). In this study BP-C2 significantly reduced the incidence and size of lung tumors in the offspring of parents that received BP-C2 following exposure to gamma radiation (Cs-137).

Experiments in Balb/C mice were supported with funds from RSF 20-15-00330,

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Determination of conversion degree of silver ions into nanoparticles in the presence of humic substances

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Keywords: humic substances, silver nanoparticles, conversion

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Silver nanoparticles (NP) are widely used as antiseptic agents as an alternative to antibiotics. They can be used as antibacterial agents, including antibiotic-resistant strains of microorganisms. There are many ways to synthesize silver NP, but the use of natural-like technologies for their synthesis is an urgent task since the synthesized preparations can be used for application to human tissues. It is known that humic substances can reduce silver with the formation of nanoparticles and, thus, they can be used as a basis for the synthesis of such antibacterial agents. In our previous work we have determined the optimal conditions for obtaining silver NP [1]. Continuing this topic, we determined one of the most important characteristics of this process - the degree of conversion of Ag⁺ into AgNP.

In this work, the content of silver in various forms was determined using ICP-AES (ICP-AES 5100, Agilent Technologies, USA). The separation of silver forms was carried out by two different methods - ultrafiltration and ultracentrifugation.

For ultrafiltration, we used Microcon YM-100, 100 kDa filters (Merck, Germany), and a miniSpin centrifuge with F-45-12-11 rotor (Eppendorf, Germany). Ultrafiltration was performed for 12 min, 13400 rpm (~12100g). Using this method, we found that after passing through the membrane in the solution remains 0,012mg/l ± 3% of the initial 2,64 mg/l ± 3%. Thus, the degree of conversion of Ag⁺ to AgNP was 99 ± 3 %

Ultracentrifugation was performed using an L8-80M ultracentrifuge (Beckman Coulter Inc., USA). A 45Ti rotor was used, at 25°C, 40000 rpm, 185677g, for 30, 60, 90, and 120 minutes. We found that complete separation of nanoparticles from silver ions is achieved in 90 minutes (Figure 1).

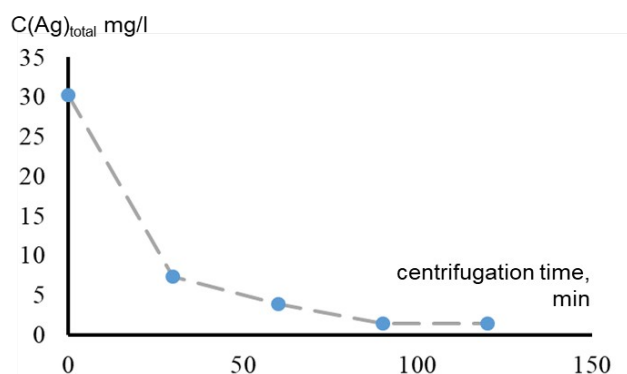


Figure 1. Total silver content versus centrifugation time.

Using this method, the degree of conversion of silver ions into nanoparticles was determined to be 95,4 ± 0,6%.

Acknowledgements. This work was supported by the Russian Science Foundation (project 20-63-47070).

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Synthesis of aromatic derivatives containing 1-hydroxy-1,1-bis(phosphonic) group

Vasiliev M.V., Zhirkova A.M., Zaitsev K.V., Prishchenko A.A., Ushakova K.A., Konstantinov A.I., Perminova I.V.

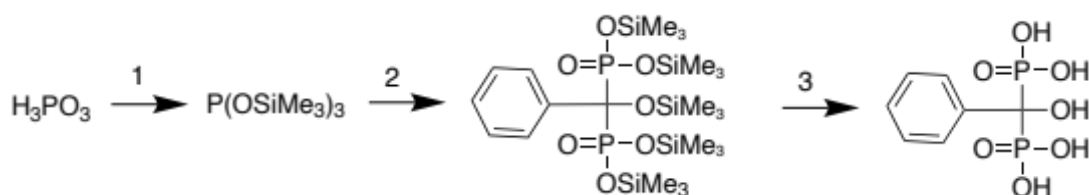
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Keywords: benzoic acid, bisphosphonates, tris(trimethylsilyl)phosphite

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The wide interest in the study of derivatives of 1-hydroxy-1,1-diphosphonic acids is due their useful biological and chemical properties. Chelating properties of the 1-hydroxy-1,1-bis(phosphonic) group are used for the treatment of metal poisoning [1]. Recent work on coordination ability has shown their high efficiency for binding ions of metals or metalloids [2]. The introduction of a 1-hydroxy-1,1-bis(phosphonic) group requires the elaboration of a practical and effective method suitable for working with multifunctional substrates such as humic substances.

At the beginning of our synthetic research on selective and efficient modification of humic substances, we studied model reaction, based on interaction of tris(trimethylsilyl)phosphite with carboxylic acid chlorides. We synthesized [1-hydroxy-1-phenyl methane-1,1-diyl]diphosphonic acid according to the scheme:



The following precursors were used: phosphoric acid, hexamethyldisilazane, benzoyl chloride, methanol. Isolation of the final product was carried out by precipitation with 90% ethyl alcohol. The yields of the target compound, $\text{PhC}(\text{OH})[\text{P}(\text{O})(\text{OH})_2]_2$, are high (>70 %).

The reaction products were characterized with a use of ^1H , ^{13}C , and ^{31}P NMR spectroscopy.

We could come up with reaction conditions for incorporation of 1-hydroxy-1,1-diphosphonic acidic group in model substrates and performed successful synthesis of [1-hydroxy-1-phenyl methane-1,1 diyl]diphosphonic acid. In the future, this method will be used to modify humic substances in order to increase their chelating properties.

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Searching for bioactive drug lead compounds from natural humic substances

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Keywords: drug lead compound, natural compound isolation, humic substances, fractionation

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The use of plant materials to treat human diseases has a long history, and ancient natural medicines are still in use. Some of them are used directly as medicinal plants, and the others are active ingredients discovered as lead compounds for drug development. The precursors of natural humic substances (HS) are lignins, tannins, lipids, peptides, carbohydrates, etc. This is why natural HS display a wide range of biological activities. Our previous studies [1] found that the ethanolic extract of coal-derived humic acid had a significant inhibitory effect on the class A β -lactamase TEM-1. This finding indicates that HS may contain lead compounds of β -lactamase inhibitors, and the inhibitory activity of β -lactamases may be related to the molecular composition of HS.

In order to reduce the complexity of HS system and to better understand their molecular compositions and biological activity, we adopt the method of biological activity-guided fractionation to extract and separate natural HS. In this study, we isolated HS using solid phase extraction (SPE) technique. Given the highest activity of phenolic components, we ran HS sorption at pH 8 in spite of the recommended pH 2. We passed HS solution through the Bondesil PPL column, eluted the sorbed components with methanol, and obtained the pH8-fraction. The molecular composition was analyzed using ultra-high resolution mass spectrometry (FTICR MS). The biological activity was detected using chromogenic analogue of β -lactam antibiotic cephalotine (CENTA) as a substrate for TEM-1 β -lactamase. The mass spectrum and inhibition curves are shown in Figure 1.

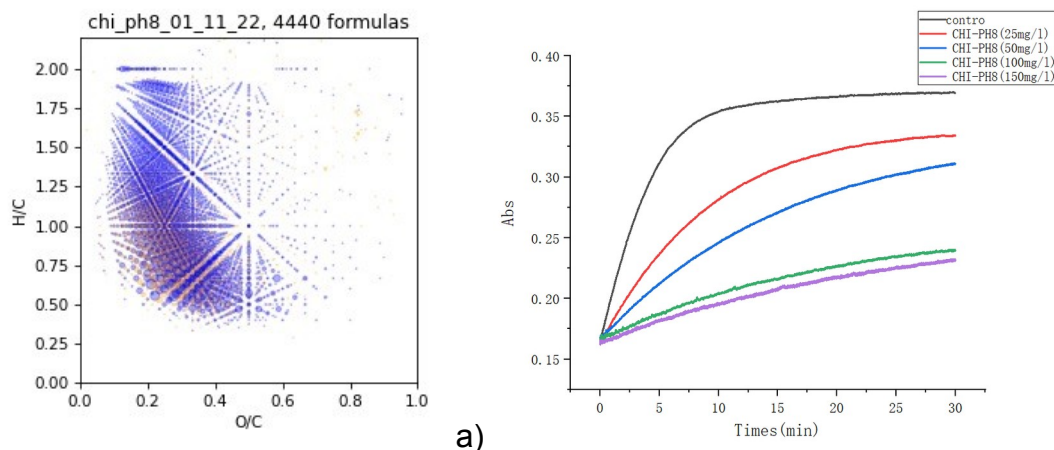


Figure 1. FTICR MS spectrum of CHI-pH8 fraction (a) and kinetic curves of its inhibitory activity towards TEM-1 β -lactamase.

The obtained results show that the pH8 fraction was enriched with least oxidized phenolic structures and it displayed substantial inhibitory effect on β -lactamase.

Acknowledgements. The research was supported by the Russian Science Foundation, project # 21-73-20202. Center of Collective Use of the Zelinsky Institute of Organic Chemistry of RAS is appreciated (15 T Solarix mass spectrometer, Bruker).

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Comparison of synthetic pathways of iron (III) complexes with macroligands of fulvic acids for correction of iron deficiency anemia

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Keywords: humic substances, iron (III), iron deficient anemia, bioavailability, cytotoxicity, Caco2

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There are more than 2 billion people in the world who suffer from anemia. Its most prevalent type is iron deficiency anemia (IDA). Despite the variety of drugs produced, the incidence rate does not decrease. The search for new drugs for iron-replacement therapy is in high demand. We have proposed two different pathways for synthesis of bioavailable forms of iron (III) in the form of a nanodispersed ferrihydrite stabilized by fulvic acid. We also assessed cytotoxicity and compared the bioavailability of iron in this form.

For the purpose of this study, we used novel humic macroligands of fulvic acid nature - the commercially available peat fulvic acids (tradename Fulvagra, Humintech Ltd). The first synthetic pathway was a one-pot synthesis. The ferrihydrite phase was generated in the humic ligand solution by rapid hydrolysis of FeCl₃ in the alkaline medium followed by precipitation of the product (F100). The second pathway was a two-stage synthesis. At the first stage, iron (III) hydroxide was obtained by hydrolysis of the FeCl₃ solution with a saturated NaHCO₃ solution, followed by precipitation. At the second stage of the synthesis, the precipitated iron(III) hydroxide was dissolved in a heated ligand solution (FAFe). The amount of Fe was measured by the ICP AES method. As a result, it was 20% and 15.5 % of the mass for the preparations F100 and FAFe, respectively.

The obtained samples were characterized with a use of XRD spectroscopy and TEM. Diffraction patterns confirmed that all samples consisted of amorphous iron-containing phase in the form of ferrihydrite. The TEM data revealed the presence of discrete nanoparticles with sizes in the range of 2-5 nm.

The cytotoxicity was assessed on epithelial cells isolated from the colon tissues (Caco2) with a use the neutral red test (MTT test). To assess the bioavailability of iron in the composition of F100, FAFe and water (control), the Caco2 cell line was used. The concentration of iron in cells was measured with a use of the ferrozine method. The results are shown in Figure 1.

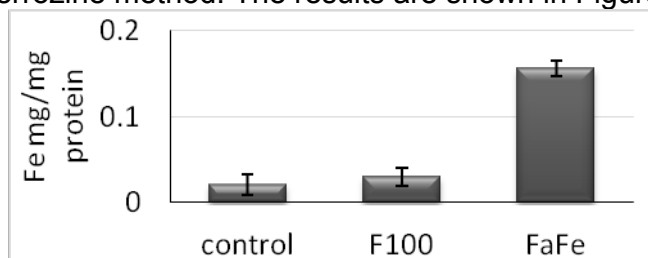


Figure 1. The amount of iron accumulated by the cells from the samples F100 and FAFe over 24 hours exposure.

Iron was present in the form of nanodispersed ferrihydrite. The samples did not display cytotoxicity: the IC₅₀ values were not achieved. The bioavailability studies have shown better bioavailability of iron in the composition with fulvic acid synthesized by the two-stage synthesis. The obtained data point out to the prospects for further study of this synthesis method in order to obtain a new active pharmaceutical substance (APS).

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The effects of humics-based Ag-nanomaterials on the pharmacological activity of antibiotic "Lincomycin" on the pyoinflammatory process model *in vivo*

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Keywords: humic substances, silver nanoparticles, bionanomaterials, antibacterial activity, pharmacological properties

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The resistance of microorganisms to antibiotics is one of the most important problems of world medicine and is the cause of the appearance of superbugs and millions of deaths, since some infections have become absolutely incurable. Antibiotic resistance occurs mainly due to the improper use of antibiotic drugs. Exposure on resistant strains of bacteria becomes a very difficult clinical task and requires the use of alternative drugs or the use of high doses of antibiotics, which leads to toxic effects for humans and further mutation of microorganisms. One of these alternative ways to combat antibiotic resistance is a combination of different bionanomaterials and antibiotics. The goal of this work was to research antimicrobial and wound-healing properties the composition of humics-based bionanomaterials containing silver and the antibiotic Lincomycin® on a model of a septic wound.

Experimental sample was synthesized in the Natural Humic Systems Laboratory of the Chemical Faculty of Lomonosov MSU on the coal humic acids (HA) "Powhumus" (CHP sample) by polyelectrolyte assembly and template synthesis of silver nanoparticles (CHP-AgNPs). The reference controls were the colloidal silver preparation Protargol® and the the antibiotic ointment Lincomycin®. The septic-inflammatory disease model of soft tissues by implanting a gauze sponge infected with a clinical strain of methicillinresistant *Staphylococcus aureus* (MRSA) into the wound space was done. The healing process of a purulent septic wound in rats was evaluated by the following indicators: 1) wound closure, % of the initial area and 2) contamination of the wound surface, CFU, % of the initial state. There were several experimental groups: group 1 – treatment of pyoinflammatory process with antibiotic ointment Lincomycin® 2%; group 2 – treatment with the ointment composition by Lincomycin® 2% and Protargol® 2%; group 3 - treatment with the ointment composition by Lincomycin® 2% and CHP-AgNPs 2%; group 4 - treatment with the ointment composition by Lincomycin® 0.2% and CHP-AgNPs 2%; group 5 - treatment with the ointment composition by Lincomycin® 2% and CHP-AgNPs 0.5%. Before the experiment the sensitivity of the used bacterial culture to Lincomycin® was evaluated by the EUCAST disco-diffusion method according to the principles of the "International Collaborative Study of Antimicrobial Susceptibility Testing, 1972" using Muller-Hinton agar. It was established that methicillin resistant *Staphylococcus aureus* (MRSA) clinical strain is absolutely insensitive to the antibiotic Lincomycin®. In the treatment of purulent-inflammatory process, it was noted that the use of antibacterial ointment Lincomycin® and the ointment composition by Lincomycin® and Protargol® had no therapeutic activity. The highest activity was shown by the sample in the 4th experimental group (the ointment composition by Lincomycin® 0.2% and CHP-AgNPs 2%) in all indicators of the healing process, on average 1.6-2.1 times compared with groups 3 and 5. It is important to note that there was a 10-fold reduction in the dose of the antibiotic against the background of the highest efficiency.

This work was supported by the financial Russian Science Foundation (Grant No. 20-65-47052).

The effectiveness and safety clinical evaluation of the humic substances based product "FitoX" use in the undergone a new coronavirus infection (Covid-19) patients' rehabilitation

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Keywords: humic substances, pharmacological activity, coronavirus infection

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There is a lot of information in the scientific literature about the high biological activity of humic substances and based on it, food products are created intended for the prevention of various diseases. The food product "FitoX" (by NovoLand company, Russia) is a soft drink concentrate, contains a complex of humic and fulvic acids, as well as extracts of medicinal plants of *Polygonum aviculare* L. and *Thymus serpyllum* L. The "FitoX" product is a natural product for protecting and preventing the effects of therapy with antibiotics, antiparasitic drugs, including to acute respiratory viral infections, is effective in combating chronic fatigue and various types of stress, helps the body to recover effectively after diseases and injuries, as well as severe physical and emotional stress. The open randomized single-center comparative prospective study (clinical evaluation) of the effectiveness and safety of the "FitoX" food product was conducted. The selection criteria of patients were: men and women over the age of 18, a anamnesis should be a verified and documented diagnosis of a new coronavirus infection covid-19 (positive test result for the virus (U07.1 according to ICD-10)), patients should have a score of 2-3 points on the rehabilitation routing scale, those in need of medical rehabilitation. The duration of the study was 21 days after the first intake of the product, the duration of the course of use is 14 days, the duration of the observation period for patients is 7 days after the end of the course of treatment. The experimental group of patients took the "FitoX" product every day at a dose of 1 mg per 1 kg of weight in terms of humic substances. The control group of patients were treated with standard methods used in the rehabilitation of patients who had suffered a new coronavirus infection (Covid-19).

Criteria for evaluating effectiveness: functional disorders based on an assessment of the severity of dyspnea during exercise on the Borg scale; affective disorders based on an assessment on the Hospital Anxiety and Depression Scale (HADS); respiratory disorders based on an assessment of the severity of dyspnea on the MRC scale (dyspnea). Safety assessment criteria: the frequency of occurrence and development of adverse events recorded according to spontaneous reports of patients, as well as according to physical examination, according to clinically significant changes in vital signs (blood pressure, heart rate, respiratory rate, body temperature) relative to the baseline; clinically significant deviations in laboratory tests and instrumental methods surveys from reference values; pathological changes in the electrocardiogram relative to the initial level.

It can be noted that based on the indicators of clinical, instrumental and laboratory examinations, no adverse events, serious adverse reactions and deaths were detected in this clinical study. Thus, the data obtained were consistent with the results of a preclinical study on the established safety profile of the active substance (humic substances) of the "FitoX" product under study and convincingly confirm the high level of safety of its use. The "FitoX" product has proven in clinical testing the previously stated effectiveness as a natural preventive and protective agent. A particularly important property was that the use of the product contributed to a faster recovery of respiratory functions, assessed on the MRC dyspnea severity scale. In addition, a statistically significant trend towards more effective recovery of the psychoemotional state of patients associated with the disease was demonstrated.

Humic substances in Arctic ecosystems

Protective properties of humic substances in Arctic lakes: geochemical and technogenic influences

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Keywords: Arctic lakes, humic substances of water, speciation

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Currently, increased attention is being paid to the study of the Arctic region, given its role in maintaining the ecological balance on the planet. The increased multifactorial pollution of natural waters is of concern due to the high concentration of industrial and technical sources on the Kola Peninsula. One of the main sources of negative impact on the water bodies of the Kola Peninsula is the Monchegorsk site of Kola MMC JSC, one of the largest copper-nickel ores processing enterprises.

Organic humic substances are high-molecular compounds of variable composition and supramolecular structure, formed as a result of complex biochemical processes. These mechanisms depend on specific landscape characteristics, climatic factors and anthropogenic impact. Natural macromolecular compounds are acids of various activities and are characterized by significant complexing properties with respect to metal ions in natural waters, which determine their protective role in the Biosphere.

We have studied the physicochemical characteristics of humic substances isolated from the waters of small lakes in the Arctic regions of the Kola Peninsula and Western Siberia (zeta potential, size, MM, IR spectra), the balance of allochthonous and autochthonous organic substances and their complex formation with several metal ions. In addition, the influence of natural factors (marine influence, alkaline, acidic rocks) and technogenic (metallurgical enterprises and nuclear power plants) on the formation of the chemical composition of waters and the qualitative features of natural organic substances was assessed.

Multivariate statistical methods of discriminate and canonical analysis revealed significant differences in the content and metals specification lakes water with different anthropogenic loads (Fig. 1). The differences were provided by the following parameters (F not less than 10): organic properties (molecular mass, zeta-potential), suspended and different colloid forms (above 0.45; from 0.45 to 0.05 μm), and Fe, Al, and Ni labile or non-labile speciation.

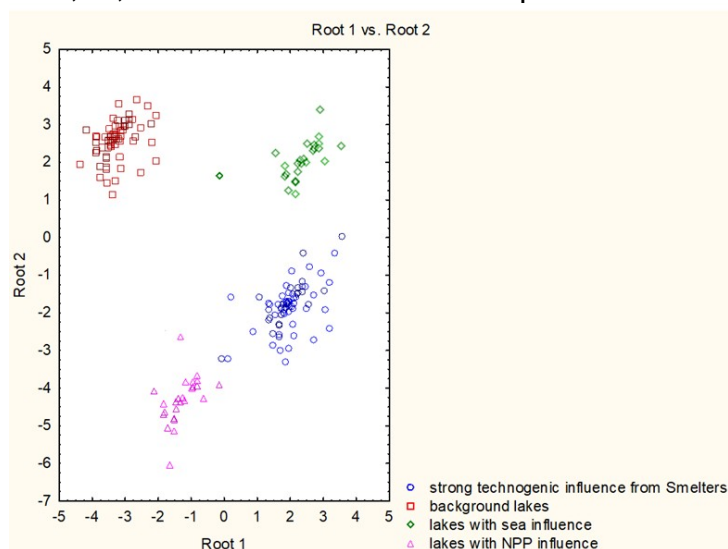


Fig. 1 Canonical visualization of Discriminant Multivariate Analysis for 4 Lake Types.

Molecular signatures of methane seeps unfold in the composition of dissolved organic matter of the Laptev and East-Siberian Seas

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Keywords: dissolved organic matter, methane, molecular composition

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The Arctic Shelf is a great unique geographical region, including diverse areas with considerable influences of mighty river mainstreams, permafrost and ice wedges. Global warming has caused substantial contribution of a large pool of dissolved organic matter (DOM) and abyssal methane CH₄ from the permafrost thawing into the Arctic Ocean [1]. As a result, surface waters can be systematically oversaturated with CH₄ due to vertical and lateral transport from bottom and littoral sediments.

DOM is a major pool of reduced organic carbon, characterized by varied origin and unique molecular composition within aquatic systems [2]. Determining the chemical constituents of DOM by the complex of analytical methods such as Fourier Transform Ion Cyclotron Resonance Mass Spectrometry (FT ICR MS), Nuclear Magnetic Resonance (NMR) and Elemental Analysis (CHNS) is supposed to be the ultimate measure for probing the source material, transport and evolution of methane in the Arctic region. The goal of the work was to investigate methane fingerprint on the base of molecular study of DOM.

Solid-phase extraction (SPE) procedure was conducted aboard research vessel Ac. Mstislav Keldysh in October 2020. The SPE was performed for 20 marine water samples from the Laptev and East Siberian Sea. The release of high concentrations of methane was observed for the nine samples from the whole set. Marine water (20-30 L each) was filtrated through GF/F filters, acidified and discharged through Mega Bond Elut PPL cartridges (5 g, 60 mL tube). The cartridges were unloaded in the lab using methanol. Recovery of the cartridges ranged from 80 to 95% for the whole set of samples. The isolates were studied by FT ICR MS (15 T Solarix mass spectrometer located at the Zelinsky Institute of Organic Chemistry of RAS), NMR spectroscopy and elemental analysis. The principal component analysis (PCA) was performed for the sample set on the base of the obtained FT ICR MS and NMR data. The molecular structures of organic matter isolates under methane conditions revealed an abundance of nitrogen, the aliphatic structures and low content of oxygen, whereas the molecular signature of the samples without methane impact were characterized as more saturated with oxygen and aromatic structures.

Acknowledgements. This research was financially supported by the Russian Science Foundation grant no 21-77-30001 (experiment on DOM isolation) and 21-73-20202 (FT ICR MS measurements). Center of Collective Use of Zelinsky Institute is appreciated. This research was conducted in the framework of the scientific educational school of the Lomonosov Moscow State University "Future of the planet and global environmental change".

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Adsorption study of copper and nickel onto organic matter of peat in relationship with the anthropogenic pollution of the Russian Arctic

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Keywords: adsorption, copper, nickel, peat, organic matter, Russian Arctic

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Arctic and sub-Arctic zones cover vast areas of the Russian Federation – more than 3 million km². Whilst only about 1% of its total population permanently live in this area. Nevertheless, due to the fact that a significant part of the minerals, fossil fuels, and bioresources are located in the Arctic part of the Russian Federation, it contributes about 12% of Russia's GDP. Active mining and industrial activities negatively affect the fragile ecosystem of the Arctic. Non-ferrous metallurgy plants – one of the major industrial facilities in the Russian Arctic, located on Kola and Taymyr peninsulas emit about 400 t of Ni and more than 500 t of Cu each year into the environment [1]. Humic substances (HS) are natural sorbents that reduce the mobility and toxicity of metals in the environment. Peat is the major source of HS in the Arctic as marshes occupy significant land areas in the region. Because of this, peat accumulates atmospheric and water pollution, specifically metals.

Research on the adsorption of metal ions on HS of peat is an important task. Obtaining such data is an important step toward the development of approaches to predicting the sorption and migration dynamics of metal complexes with organic matter in the Arctic. It is important to note that nowadays most of the adsorption studies devoted to the sorption of metals on peat and other natural organic matter (NOM) are mostly focused on toxic elements such as Pb, Cd, and Hg. However, in the case of the Russian Arctic where non-ferrous metallurgy plants emit toxic amounts of Ni and Cu into the environment focus of adsorption studies needs to be shifted to these elements.

In our study, we characterized the kinetics of Cu and Ni adsorption on peat. The concentration of elements varied from 50 to 2500 mg·L⁻¹, the mass of peat was taken as 30 mg, 0.03 M pH 5.5 acetate buffer was used for pH and ionic strength maintaining. We found out that experimental data comply with the pseudo-second-order kinetic model with R²=0.999 and R²=0.998 for Cu and Ni, respectively. The obtained data fitted well to the Langmuir model [2]: R²=0.970 (Cu) and R²= 0.979 (Ni), the sorption capacity of peat with regard to Cu²⁺ and Ni²⁺ accounted for 63 and 43 mg/g, respectively. Based on the obtained data, we can conclude that the maximum sorption capacity of peat is higher for copper ions as compared to nickel ions. We also found that the values of Freundlich constants were higher for Cu: 10.1 against 7.3 for Ni, which might be indicative of formation of more stable complexes of Cu with peat NOM as compared to Ni.

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Humic acids isolated from various types of podzols in Arctic ecosystems – ¹³C CP/MAS NMR spectroscopy

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Keywords: nuclear magnetic resonance, soils, agrosoils, YANAO

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The soil cover of Arctic ecosystems is highly differentiated in terms of soil types. Here one can see various typical background soils of the region, soils subjected to the effects of urbanization and even agrogenic and post-agrogenic soils. Our study focused on various podzols localized around the town of Nadym (Yamalo-Nenets Autonomous Okrug, Russia, 65.5333 72.5166). Four key objects of study were selected: 1 – typical (mature) podzol (Albic Podzol (Turbic)); 2 – urbanized podzol (Albic Podzol (Anthrotoxic)); 3 – agrogenic podzol (Plaggic Podzol (Turbic)); 4 – post-agrogenic podzol (Plaggic Podzol (Turbic)). Humic acids (HA) were isolated from the topsoil horizons AO, AOu, Ap, by the standard IHSS method. Major differences in the chemical composition of HA were found, with urbanized podzols containing a greater proportion of aromatic compounds (110-160 p.p.m.), and agrogenic and natural more alkyl compounds (0-46 p.p.m.). Alkyls content decreases in the series pristine>agrogenic>post-agrogenic>urbanized (Figure 1).

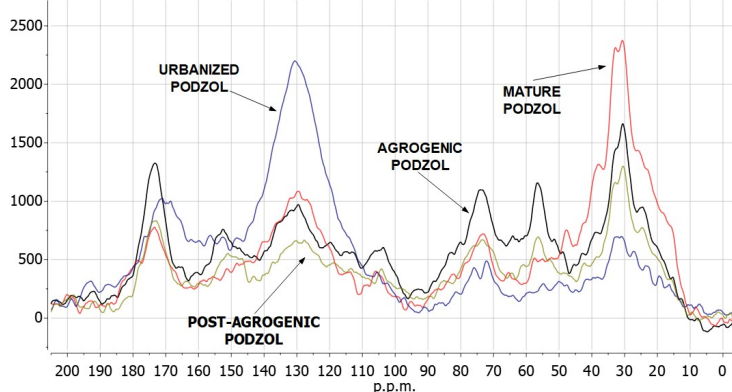


Figure 1. ¹³C NMR spectroscopy of the humic acid isolated from the topsoil horizons. X-axis – chemical shifts (p.p.m.); Y-axis – intensity of the signal.

Calculation of the quality indicators of humus indicates that in pristine soils humic acids are more stable, compared to agro- and urban ecosystems (Table 1).

Table 1. The content of the structural fragments in the humic acid

Soil	Chemical shifts from ¹³ C, %						AR	AL	AR/ AL	Al h,r+ Ar h,r	C,H- Al/O, N
	0- 46	46- 60	60- 110	110- 160	160- 180	180- 200					
Mature	41.1	6.6	13.8	23.6	9.1	5.8	32.8	67.2	0.49	55	2.0
Agrogenic	34.2	9.0	14.9	20.6	14.0	7.3	34.6	65.4	0.53	49	1.4
Post-agro	34.8	7.7	14.5	23.1	13.2	6.7	36.3	63.7	0.57	49	1.6
Urbanized	18.5	3.7	9.7	46.0	14.5	7.6	60.6	39.4	1.54	28	1.4

AR (aromatic fraction) – the sum of the AR structural fragments; AL (aliphatic fraction) – the sum of the AL structural fragments; AL h,r + AR h,r – degree of hydrophobicity (%); C,H-AL/O,N-AL – degree of humification

Increased anthropogenic pressure on the ecosystem leads to a decrease in the degree of hydrophobicity and humification of humic acids.

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Study of the molecular composition of dissolved organic matter in Arctic Sea water

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Keywords: dissolved organic matter, Arctic Shelf, molecular composition, optical measurement

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The Arctic Shelf is the most sensitive region to the global warming. Therefore, climate changes of the region are supposed to be the indicator of global warming [1]. Rising average air temperatures, melting glaciers, and progressive degradation of permafrost ultimately lead to an increase of the greenhouse gas emission [2]. The emission of methane in the Arctic region is of high importance. Thus, the investigation of carbon cycle in the Arctic Shelf is the urgent scientific task.

The goal of this work is a comprehensive interpretation of the molecular, structural and elemental composition of dissolved organic matter (DOM) of permafrost in the aquatic system of the Eastern Arctic and the determination of metabolic relationships between the molecular composition of DOM extracts, humic substances of sediments and methane release.

The research and sampling were carried out aboard the vessel Ac. Mstislav Keldysh in the Kara Sea region in October 2021. The marine water was sampled and filtrated through GF/F filters to investigate the optical characteristics of the DOM. The former was isolated by solid phase extraction (SPE) procedure using Bond Elut PPL cartridges. The extraction of cartridges was conducted in the lab using methanol. To ensure greater stability of the isolates, the lyophilization stage was performed for the methanol DOM extracts.

The whole set of extracted samples are planned to study by measuring of total organic carbon concentrations and elemental analysis. The structural and molecular composition of isolates will be investigated by Fourier Transform Ion Cyclotron Resonance Mass Spectrometry (FT ICR MS) and Nuclear Magnetic Resonance (NMR) spectroscopy. The optical parameters of the isolates and marine waters are supposed to be investigated by fluorescence and UV-vis spectroscopy.

The next step of the study is to investigate the correlation between the molecular composition of DOM and to released amount of methane. The main result of the work will be the model of permafrost degradation.

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Pool of amino acids in the mountain soils, Crimea

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Modern laboratory methods provide the ability to study soil organic matter at the molecular level, where individual detectable compounds can act as biomarkers (such as amino sugars, lignin phenols, n-alkanes, etc.) for a more accurate reconstruction of paleoecological conditions. The purpose of this study is to consider the pool of amino acids in the soils of the Crimean Mountains, the genesis of which is poorly understood, and to assess the possibility of using these compounds as such markers.

We have studied the profiles of Phaeozem and Cambisol located on the southern slope of the Ai-Petri Yayla - a mountain plateau in the central part of the Main ridge of the Crimean Mountains. The analysis was performed by HPLC in water (free amino acids) and acid (bounded Amino acids) extracts.

The distribution of some amino acids, especially phenylalanine (in mountain-meadow chernozem-like soil $r = 0.90$ and in burozem $r = 0.81$), closely correlates with the profile distribution of roots. This is due to the fact that the main sources of low molecular weight aromatic acids in the soil are the products of lignin degradation [2]. This fact is especially relevant for mountain soils characterized by an increased accumulation of root detritus. The content of (free) phenylalanine is higher under the woody vegetation of the beech forest. In the Phaeozem the presence of (free) dicarboxylic amino acids (glutamic, aspartic) is more significant, due to the high excretion of these compounds by herbaceous vegetation.

A connection was also found between the content of humus and (free) proline ($r = 0.6$), which accumulates in plants under stressful conditions [1]. Analysis of the amino acids pool using the principal component method revealed a high dynamics of soil formation processes in Cambisoli, due to the displacement of the upper boundary of the altitudinal belt of beech forests as a result of climate change.

Among the bounded amino acids, alanine and histidine are found in the greatest amount in the Phaeozem. According to Vasilievich, the ratio of the content of heterocyclic (more stable) and oxy-amino acids (most rapidly assimilated by microorganisms) allows us to judge the magnitude of biological activity [3]. With an equally low content of oxy-amino acids in both profiles and a significant predominance of histidine in Phaeozem, it can be concluded that there is a higher biological activity in it.

Thus, the content of individual amino acids allows us to reconstruct the types of plant communities, evaluate the paleoecological environmental conditions, as well as the biological activity of soils.

Acknowledgements. The work was done with the support of theme №122011800459-3.

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The use of fulvic acids for the purification of heavy metals in the aquatic environment

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Keywords: fulvic acid, heavy metals, purification

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Fulvic acids are an important class of soil organic matter. They arise in soils as a result of plant decay and are soluble at all pH values [1]. Fulvic acid is ubiquitous in the aquatic environment and has many functional groups that allow it to form complexes with metal ions.

Sorbents with fulvic acid remove heavy metals from the body, since fulvic acid has a unique ability to "bind" with heavy metals, forming complexes with free metals or attached to metalloproteins, which contributes to the removal of these metals from the body. Complexes of metals with fulvic acid are characterized by sufficiently high strength. In particular, the logarithms of the stability constants of fulvates are: Be - 9.4; Sr - 9.8; Cu - 12.1; Ni - 8.4; Zn- 10.1; Cd - 9.2. Heavy metals differ significantly in the strength of their bonds with organic matter. Its strength depends on the composition of the latter, the chemical properties of the element, as well as on the acid-base characteristics of the medium.

Much less studied is the question of the influence of cations of various metals on the adsorption of fulvic acids by minerals. At the same time, this problem is of great interest from the point of view of the processes occurring in soils and other media enriched with humic substances. Soil organic acids and their compounds with metal cations have different chemical affinities for the surface of clay minerals, and the mechanism of their interaction with the surface may also change [2].

To conduct research on the purification of heavy metals using fulvic acids as an adsorbent, a model solution of heavy metals was prepared from state standard samples. In a conical flask (250 ml), a weighed portion of the sorbent (0.5 g) was mixed with a model solution (50 ml, pH=3.26) and stirred in a shaker at room temperature (25°C) at a stirring speed of 200 rpm for 3 h. After the process, the adsorbent with heavy metals was removed from the solution by filtration. The solution filtered from the adsorbent (pH=5) was analyzed for the content of heavy metals on a Ta-lab voltammetric analyzer. The results of the elemental analysis presented in Table 1.

Table 1. Results of water purification from heavy metals

Heavy metals, mg/l	C ₀ (Me), mg/l	Fulvic acids	
		C _{res} (Me), mg/l	Degree of purification, %
Cadmium	0.10	0,0036	96,40
Lead	0.10	0,0042	95,80
Copper	0.10	0,0062	93,80

The results show that the fulvic acid preparation used in this study has a high affinity for metal ions such as Cd, Pb and Cu. This is explained by the fact that when interacting with heavy metals, fulvic acids form a chelate complex due to carboxyl groups [3], since at pH below 6, strongly acidic carboxyl functional groups, as well as carboxyl groups of medium strength, will be completely dissociated. In this regard, fulvic acid can be considered as a sorbent for water purification from heavy metals.

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Humic substances in the soil – plant system: new humic agrochemicals and agrotechnologies

Protective effect of humic acids. Pesticides of the pyrethroid class

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Keywords: humic acids, pesticides, pyrethroid, protective effect

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Soil is a biologically active part of the lithosphere and the first recipient of any contamination arising from various human activities. Currently, farmers are widely using intensive field cultivation technologies. Abundant fertilization, the use of pesticide and a failure to adhere to crop rotation causes a degradation of the soil. Negative changes of the soil biota and its biochemical processes take place as a result of a biological misbalance. This becomes a limiting factor for achieving the potential fertility of plants. The main component of soil involved in the process of sorption is organic matter (OM), the structure of which should be distinguished in three fractions: humic acids (HA), fulvic acids (FA) and humin (H), which differ in solubility in water, acid and alkali, thereby characterized by varied properties. The purpose of this study is to evaluate the possibility of using HS in the processes of environmental detoxification, using the example of pesticides of the pyrethroid class.

Materials and methods. Five peatlands have been selected for the study in the floodplain meadows of the Yenisei River. Extraction of humic acids (HA1, HA2, HA3, HA4, and HA5) was performed using a procedure recommended by the International Humic Substances Society. One of the most common pyrethroids at the moment is cypermethrin and its isomers, which have been used in research. HA have been assessed on the basis of their elemental composition. Spectroscopic techniques have been used to study the HA structure and their interaction with pesticides including: UV-visible spectroscopy, FT-IR spectroscopy and LC-MS/MS.

Results. For cypermethrin and its isomers, sorption was well described by a Freundlich isotherm where $1/n$ values indicated that the sorption mechanism could be mainly explained by physical reactions in all samples of HA. The complex mechanisms of interactions between pesticides and HA based on van der Waals forces, ligand exchange and charge transfer complexes formation were demonstrated. It was also proved that the nature of interactions is strictly dependent on the structure of the pesticide molecule.

The conclusion of our study may contribute to the choice of plant protection products that, in addition to their effectiveness, are as little ballast for the environment as possible.

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The concept of evolutionary humification

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Keywords: soil organic matter, humic substances, free radicals

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Soil humic substances (HS) are the largest reservoir of carbon and nitrogen in the biosphere and perform a wide range of environmental services. The process of humification due to the polychemism of soil organic matter has not yet been fully studied.

If we consider this process from an ecological point of view, then humification is the result of the joint interaction of plants and soil, which has developed over millions of years of biosphere evolution. Due to their hydrophobicity, humic acids are protected from leaching from soil and are able to accumulate in the upper, root-inhabited part of the soil profile. Due to their relatively high biochemical stability, they accumulate long-term reserves of nitrogen and other nutrients necessary for the sustainable development of plants.

If we consider humification from a biochemical point of view, then it seems to use the theory of random transitions from the apparatus of supramolecular chemistry. In a generalized form, three stages of humification can be distinguished as a process of formation of a spatial lattice of self-organizing systems. In the soil, it begins with the stage of small-scale template fluctuations of monomers with the formation of the simplest associates from molecules with a minimum degree of complementary binding of the decomposition products of organic residues, their hydrophilic part adsorbed on the surface of mineral particles. Then the stage of medium-scale fluctuations develops. There is a "self-organization" of the system through the formation of longer-lived cluster ensembles of pro-HS in the form of short chains - microlayers and microframeworks. This stage ends with the complementary formation of two- or three-dimensional associates in the form of supramolecular ensembles by free radical polymerization.

These two stages take place under optimal hydrothermal conditions for microbiological activity, when monomers of organic compounds of various nature are present in the soil in sufficient quantities. The pro-HS formed under these conditions are highly hydrated and have a maximum conformational volume.

The next stage is large-scale fluctuations - autocatalysis or "self-assembly" of the system: complementary three-dimensional condensation or free radical polymerization of supramolecular ensembles of HS into global macromolecular structures of HS. It occurs when hydrothermal conditions change, causing dehydration and a decrease in the conformational volume of (decrease by about 10 times) as a result of moisture deficiency or other reasons. A decrease in the volume of branched macromolecules leads to the convergence of individual branches and the emergence of covalent bonds. As a result, HSs of a stochastic nature are formed in the soil based on ring structures with the participation of benzene and pyrene nuclei with the formation of a stable "two-layer" amphiphilic HS film on the surface of solid particles.

Further evolution of HS in the soil proceeds towards a decrease in the concentration of free radicals, an increase in biothermodynamic stability and the content of condensed polyconjugated aromatic structures. These processes are reflected in the conceptual scheme of evolutionary humification developed by us.

When a state of equilibrium is reached (when the processes of transformation of organic carbon compounds in the soil are balanced), the process of oxidative free radical polymerization is gradually replaced by a process of fragmentary renewal of HS.

Development of new humic fertilizers

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Keywords: humic fertilizers, microelements

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A non-waste technology for the production of humic fertilizers of various composition and purpose from lowland peat has been developed and patented. Macronutrients have been introduced into the composition of agrochemical grades. The new fertilizers were successfully tested on various crops and received state registration under the general trade name Tyumen agrochemical.

The purpose of further research is the selection of trace elements to enhance the effect of the humic preparation.

Doses of microfertilizers were determined based on the analysis of literary sources. Testing was carried out on seeds of spring wheat. The seeds were germinated in Petri dishes for 7 days on solutions of a mixture of a humic preparation with microfertilizers. Two biotests were carried out, the control in the experiments was the humic preparation Rostok 0,1%. The concentration of the working solution of the humic preparation is 0,001%.

In the first experiment, boric acid H_3BO_3 , copper sulfate $CuSO_4 \cdot 5H_2O$, potassium iodide KI, ammonium molybdate $(NH_4)_2MoO_4$ were dissolved separately in a humic preparation.

According to the number of roots of spring wheat seedlings, there were no significant differences between humic fertilizer and with the introduction of microelements into its composition. Exceeding the control was observed only when boric acid was added – by 0,1 pc.

The maximum length of the root during seed germination in the solution of the Rostock preparation was 10,8 cm. When microelements were introduced into the composition of the humic preparation, a decrease in root length was observed, but the difference with the control was not significant. The minimum decrease in this indicator was determined when potassium iodide was added – by 0,7 cm, the maximum decrease when copper sulfate was dissolved in a humic preparation – by 2,5 cm. The pattern along the length of a spring wheat sprout is similar to the length of the root.

The mass of seedlings significantly exceeded the Rostock preparation only when boric acid was added to it – by 5%. Other microfertilizers reduced the weight of seedlings: potassium iodide – by 15%, copper sulfate – by 36%, ammonium molybdate – by 45%. The difference with control is significant in the last two cases.

In the second experiment, the effect of a mixture of microfertilizers was studied: H_3BO_3 , $MnSO_4$, $ZnSO_4$, $CuSO_4 \cdot 5H_2O$. First, microfertilizers were dissolved in distilled water, then they were added to the Rostock preparation (1 and 10 ml/l), because sulfates of manganese and zinc do not dissolve in the alkaline environment of the drug.

The germination of seeds during germination on a solution of the Rostok preparation was 90%, on a mixture of microfertilizers – 77%, with the addition of microfertilizers to a humic preparation – 80%. All biometric indicators of test-culture plants during seed germination on a solution of micronutrient fertilizers and when they were introduced into a humic preparation significantly decreased compared to a humic preparation without additives: the number of roots – by 3-10%, root length – by 12-31%, sprout length – by 8-23%, the mass of seedlings – by 23-47%.

Thus, the addition of microfertilizers to the humic preparation reduced its effect on the test culture.

Humic-based polyelectrolytes: effect on Cd and Pb mobility in contaminated soils

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Keywords: soil conditioner, cationic polymer, anionic biopolymer, interpolyelectrolyte complex, heavy metal

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Interpolyelectrolyte complexes (IPECs), the products of interaction between two oppositely charged polyelectrolytes (PEs), have been suggested for soil structure stabilization. An ability of IPECs to improve soil hydrophysical properties, prevent soil erosion and act as soil conditioners have been studied in our previous works [1]. The efficacy of IPECs was dependent both on chemical composition of IPEC and soil properties. Using humic substances as the polyanionic ingredient of IPEC is a prospective approach to develop environmental-friendly and effective formulations.

In this study we used two IPECs: positively and negatively charged. Both were prepared from the commercial polymers with the excess either of polycation or polyanion: cationic poly(diallyldimethylammoniumchloride) (PDDA) and anionic biopolymer lignohumate (LH), a humic-based plant growth stimulator. The objective of the study was to assess the effects of the three PEs (LH, IPEC+ and IPEC-) on mobilization/immobilization of Cd and Pb in soil spiked with these metals, as well as on soil structural composition and plant growth in a model experiment.

Experiments were conducted in a small-plot trial using artificial soil consisting from sand (48%), clay (48%) and peat (4%). Soils were first treated with 2% PE-solutions (1.5L per plot) and then spiked with CdCl₂ and Pb(CH₃COO)₂ in rates equal to 3 tentative allowable concentrations and *Festuca pratensis* was sown. After 2 months plant biomass, soil aggregate composition and content of Cd and Pb (mobile species in ammonium acetate buffer, pH 4.8) were determined.

Both IPECs and LH decreased the content of mobile species of cadmium and lead in soil comparably to the spiked control. The highest detoxifying effect was observed for the negatively charged IPEC, with a 34-35% decrease of Cd and Pb mobile species. Positively charged IPEC and LH also demonstrated detoxication ability: content of cadmium and lead decreased by 21% and 24-25%, respectively.

In phytotests all polymers did not show a significant effect on plant growth parameters.

Composition of soil structural aggregates was not significantly affected by all the PEs and the share of agronomically valuable aggregates (0.25-5 mm) varied within 84-89%.

The results of the work show that negatively charged IPEC and LH have the potential to decrease content of mobile HM-species and thus to mitigate their toxicity for plants, but did not provide a significant effect on soil structure for the used soil. Positively charged IPEC destroy large soil aggregates and can improve soil structure in a case of cloddy soils. In general, humic-based IPECs can be regarded as a tool to create productive artificial soils.

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Impact of growth regulator of humate and fulvate type on productive factors of potatoes, onions and tomatoes while using in the system of dribble irrigation

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Keywords: lignohumate of brand AM, growth regulator, potatoes, onions, system of dribble irrigation

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Under the conditions of intense plant science resource saving technologies come out on top. In particular, the system of dribble irrigation implies water saving and pesticidal load upon agrocenosis. The use of Lignohumate in the technology of dribble irrigation can improve profitability of production without using additional resource spend. For scientific support of practice in the application of Lignohumate in the system of dribble irrigation tests were carried out on the basis of the Federal State Funded Research Institution "Pre-Caspian Agrarian Federal Research Center" (Astrakhan region), part of the results are shown in table No. 1.

Table 1 Impact of preparation Lignohumate of brand AM on yielding capacity of potatoes, onions and tomatoes in 2021

No.	Variant	Crop	Yield t/ha	Increment of the background	
				t/ha	%
1-K	Background*	Potatoes	71.8	–	–
2-K	Background + Leaf treatment		80.3	8.5	11.8
3-K	Background + Dribble irrigation		83.0	11.2	15.6
4-K	Background + Leaf treatment + dribble irrigation		88.1	16.3	22.7
1-T	Background	Tomatoes	141.0	–	–
2-T	Background + Leaf treatment		150.5	9.5	6.4
3-T	Background + Dribble irrigation		165.0	24.0	17.0
4-T	Background + Leaf treatment + dribble irrigation		172.0	31.0	21.9
1-L	Background	Onions	59.7	–	–
2-L	Background + Leaf treatment		73.0	13.3	22.4
3-L	Background + Dribble irrigation		72.9	13.2	22.2
4-L	Background + Leaf treatment + dribble irrigation		75.3	15.6	26.1

* Background – basic cultivation technology

Inclusion of Lignohumate into background cultivation technology in the farmstead makes it possible to get considerable increase of yielding capacity without using additional facilities which is confirmed by the results received in the course of the tests:

In variant 4-K it was possible to get the increase of yielding capacity of 16.3 t/ha in regard to the background yielding capacity;

In variant 4-T the increase of yielding capacity makes 21.9%;

In variant 4-L the increase of yielding capacity was 15.6 t/ha.

Lignohumate increased commercial yielding capacity in all three variants, as well as positively affected quality factors of the yield; thus, we can observe the increase of starch and vitamin C quantity in potatoes and the decrease of nitrates accumulation. In all variants of synergetic effect of Lignohumate was observed in case of combination of leaf application via

dribble irrigation. The results which were received in the course of the tests found scientific evidence of the positive practice of Lignohumate application in the systems of dribble irrigation.

On the global scale implementing Lignohumate into the basic technology of the farmland is possible except the increase of production profitability, obtain the decrease of anthropogenic load on geocenosis, saving water resources, as well as decreasing chemical load upon the soil.

The application effectiveness of activated peat hydrosol based biopreparation for barley cultivation

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Keywords: humic biopreparations, barley

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The paper describes the production testing to determine the application effectiveness of the activated peat hydrosol based biopreparation within the cultivating of barley of the cultivar "Prairie" in the conditions of Orlovsky district of Rostov region.

The biopreparation is a peat hydrosol that was activated by ultrasonic cavitation process at a high static pressure without the use of any chemical reagents [1]. The dispersed phase of the sol consists of nanoparticles tending to agglomerate and produce amorphous structures with micrometer sizes.

The main biologically active components of the biopreparation are humic acids capable to stimulate the growth and development of plants.

The biopreparation being researched has been used for pre-sowing seed and foliar processing of sowing in phase of stem elongation combined with herbicides, macro- and microelement fertilizers (Table 1).

Table 1. Test scheme

	Pre-sowing seed processing	Foliar processing
Control	–	–
Test site No 1	5% solution	–
Test site No 2	5% solution	0.5% solution
Test site No 3	5% solution	1.0% solution

When the grain reached the kernel milk phase we estimated the intermediate productivity by mowing method with the further monitoring of barley crops' morphometric indicators.

Within the production testing the foliar treatment with 0.5% biopreparation working solution tended to significantly raise the raw crop biomass by 20.1% together with less significant increase in grain yield by 4.9% , while foliar treatment with 1% solution provided the significant increase in raw biomass by 64.8% coupled with an grain yield increase by 11.4% relatively to control site. As for the single foliar treatment in the phase of tillering - stem elongation we have not registered any sensible change in the quality of barley grain within all variations of the experiment parameters.

It should be remarked that almost all climatic conditions were favorable for the formation and the maturation of the barley grain. However, during the period of wax ripeness barley grain encountered the unfavorable conditions: high temperatures, combined with low air humidity and soil drought during this period, limited the possibility of realizing the productivity laid down in earlier periods of productivity development.

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Priming with leonardite humate affects the early growth performance of common wheat in excessively wet and dry conditions

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Keywords: *Triticum aestivum* L., drought, water excess, carotenoids, chlorophyll, relative water content

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Wheat is the most abundant crop in the world: it is the first rain-fed crop after maize and the second irrigated crop after rice (Portmann et al., 2010). Unfavorable environmental conditions affect crop productivity and quality. In recent times, the global rainfall pattern has also changed, leading to an expansion of drought-stressed zones as well as water excess extreme events, where excessively wet and dry conditions cause severe stress to plants. Wheat is both sensitive to water deficiency and surplus. Seed priming is defined as a pre-sowing treatment that partially hydrates seeds without allowing emergence, following by drying back to the original moisture level. The primed seeds have been reported to demonstrate enhanced germination and increased stress tolerance under various adverse environmental conditions. Humic substances are biostimulants that are natural constituents of different environments which are considered to relate to mitigating activity against biotic and abiotic stresses. Despite a substantial amount of data indicating the prospects of using both seed priming and HS application to improve crop performance, ready-to-use technologies are still lacking. The aim of the study was to assess the effects of seed priming with leonardite humate (LH) on the early growth performance of common wheat in excessively wet and dry conditions.

To reach this aim, common wheat (*Triticum aestivum* L., cv. L-1) seed priming with LH (2 g/l) was conducted, followed by a laboratory pot trial. Soil moisture was kept at 70%, 50% and 30% of soil water holding capacity (SWHC) imitating excessively wet, optimal, and excessively dry conditions, respectively. The experiments were established on Albic Retisol, which was silt loam soil with a pH value of 6.9 (1:5, H₂O) and humus content 1.8%. To estimate effect of priming with LH, germination rate were monitored during the wheat growing. At 21 day after sowing (DAS), plants were harvested followed by plants' length and fresh weight measurement, determination of pigments' (carotenoids, chlorophylls *a* and *b*) content and assessment of water deficiency in plant leaves.

At DAS 3 the emergence rate index of primed seeds was 3.3 or 6.0 times higher than the control ones under water deficiency or surplus, respectively. However, effect of priming on final germination was observed only under water surplus, where this parameter was 3 times greater as compared to control. Though no statistically significant ($p < 0.05$) effect of priming on the plants' length or weight was found, wheat plants raised from the primed seeds under water surplus possessed higher content of carotenoids and chlorophylls, and plants raised from the primed seeds under water deficiency demonstrated lower water deficiency of leaves.

Our results point to the seed priming with humic substances as a promising pre-sowing treatment to increase stress tolerance of common wheat in excessively wet and dry conditions.

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The reasons of humic substances biological activity

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One of the effective and economically justified methods of influencing the production process of green vascular plants is their foliar treatment with solutions of humic substances (HS). Most often, HS are isolated from various natural objects: composts, brown coals, peats, soils, and others using aqueous alkaline or neutral salt solutions, as well as a combination of aqueous alkaline solutions and sodium pyrophosphate. There is an opinion [1] that isolation of HS by any aqueous alkaline solutions and their subsequent fractionation is not correct. In addition, as shown in the studies of O. Schreiner and E.C. Shorey [2], soil humic acids, which isolated by alkaline solutions and precipitated by mineral acids, include resin acids and their esters, fatty acid glycerides (fats), agosterol, phytosterol, paraffinic, lignoceric and agocereric acids, and soil fulvic acids, which non-precipitated by mineral acids, include dioxysteric acid, picoline carboxylic acid, pentosans, xanthine, hypoxanthine, cytosine, histidine, and arginine. From the point of view of chemists dealing with cellulose production technologies, dark-colored substances formed because of alkaline pulping of plant raw materials are black liquor [3].

In our opinion, HS isolated by different aqueous alkaline solutions from natural objects are black liquor too. The black liquor is a complex mixture consisting of (1) organic monomers and oligomers (products, which are inherited from initial material and/or formed resulting from the alkaline hydrolysis of natural organic polymers), (2) colloidal systems of natural organic polymers (lignin, polysaccharides, fats, waxes, resins, kerogen, etc.), (3) organo-mineral compounds, including colloidal systems, and (4) inorganic ions and a variety of compounds, including colloidal systems. It is the complex chemical composition of black liquor (or HS) that determines its biological activity. Since the components of this organic mixture contain a variety of functional groups, they form colloidal systems and are characterized by a large set of universal blocks of biological macromolecules. These substances, getting into plants, can: accelerate the circulation of nutrients, increase the permeability of cell membranes, relieve crop stress after pesticide application, optimize the ratio of organic and mineral anions in plants, Absorption, and assimilation by green vascular plants of structural fragments of biological macromolecules leads to their enrichment with energy as well as to the induction of gene expression.

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Characteristics of the group composition of peloids of various origins

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Keywords: humic acids, elemental composition

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The biological activity of low-mineralized peloids is determined by organic components that are currently insufficiently studied. It is customary to distinguish groups of non-specific and specific organic compounds in organic components. The predominant component in the composition of therapeutic muds are specific organic complexes - humic substances, which are represented by humic acids, pro-humic substances and humin components.

The aim of the work was to study the effect of mineralization on the composition of peloids of various origins.

In all analyzed objects, humic substances account for approximately 86.46%, and the group of non-specific organic substances is represented by a smaller amount and averages 13.54%. In the composition of specific organic substances, the maximum proportion in all samples falls on humic acids. The humin content is characterized by significant fluctuations, while the humic acid content is stable and specific for each mud deposit. The maximum amount of humic acids is represented in lake Sernoe and lake Molochka where their content reaches 95.61% and 92.98%, respectively, of specific organic substances. In the Teplovka and Solodovka deposits, the ratio of humic acids is slightly less and is approximately 72.30%.

In the therapeutic muds of the lake Teplovka and lake Malting are characterized by a high humin content of 25.85% and 29.46%, respectively, while lake Teplovka differs from other mud sources in shallow water, large redox potential, as well as high mineralization of mud solution. Lake Solodovka is close to the lake by hydrogeological characteristics. The humin content and humin content in it are also high compared to lakes Molochka and Sernoe. The content of humin in the lake Molochka and lake Sernoe is composed of 7.02% and 4.39%, respectively.

Based on the data obtained, it can be concluded that the content of organic substances that determine the biological activity of peloids is determined by the thermodynamic conditions of the formation of deposits and the presence of inorganic components in the mud solution. The different composition of the organic components of therapeutic mud implies a different level of their biological activity.

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Feed additive based on humic substances

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Keywords: potassium humate, feed additive, dairy calves

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There are many publications on the presence of a wide range of biological activity of humic substances. Humic acids inhibit the growth of pathogenic bacteria, mold, and reduce the level of mycotoxins. Humic substances are biologically active compounds, therefore they are actively used as components of various veterinary preparations, feed additives and sources of biologically active substances in food products. In animal husbandry, the use of preparations based on humic acids promotes weight gain, reduces the spread of infectious diseases and activates metabolic processes [1]. Analysis of the results of bioregulatory properties, antimicrobial activity, biological activity of drugs of domestic origin based on humic acids on laboratory and farm animals showed the safety and activity of veterinary drugs for the treatment of purulent-necrotic processes of hooves and claws, purulent and surgical wounds [2].

The analysis of the obtained feed additives based on potassium humate for elemental composition and physicochemical was carried out within the framework of the project IRN AR09260096 "Development of technology and organization of pilot production of modified organic biological products based on humic polyelectrolyte acids obtained from coals of Kazakhstan" (LLP "Institute of Chemistry of Coal and technology").

Production experiments were carried out on the basis of the dairy farm of Kamyshenka LLP in 2021-2022. within the framework of scientific research under the program BR10764965 "Development of technologies for keeping, feeding, growing and reproduction in dairy cattle breeding based on the use of adapted resource-energy-saving and digital technologies for various natural and climatic zones of Kazakhstan." To conduct a production experiment on calves, two groups (control and experimental) were formed, 20 heads each. The age of setting up for the experiment is 3 months, the average live weight is 82 kg, the duration of the experiment is 90 days. The animals were fed according to the scheme adopted in the farm, the composition of the diet included reverse, concentrated feed and green mass (clover, alfalfa, cereals, corn). The set of feeds and their quantity fully met the needs of calves of this age - young animals up to 6 months of age, green feeds were fed ad libitum. According to the main indicators, the rations were balanced in accordance with the recommended norms. Additionally, the animals of the experimental group in a mixture with skim milk received the studied feed additive in the amount of 15 ml per head. At the end of the research, indicators characterizing the growth of animals, feed costs, hematological indicators were calculated and an appropriate conclusion was made on the use of feed additives.

The results of the production experience showed that the feed additive had an effective effect on weight gain in calves. At the beginning of the experiment, the whole was 82 kg, at the end of 112 kg. The weight was 30 kg.

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Effect of modified organic fertilizers based on potassium humate on the growth of biological objects

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Keywords: potassium humate, organic fertilizer, enrichment

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Scientists of the "Institute of Coal Chemistry and Technology" LLP obtained a line of modified organic fertilizers based on potassium humate with the addition of Fe, NPK, Mo, Mg. Tests of the influence of a line of modified organic preparations on biological objects were carried out in the Botanical Garden of Nur-Sultan. The following bioobjects were selected: Blue spruce (*Picea pungens*), Dalecarli birch (*Betula pendula* 'Dalecarlica'), Rose (*Rosa*), Nedzwiecki apple tree obtained by micropropagation (*Malus niedzwetzkyana* Dieck), Pear (*Pyrus*) Watering the control groups with solutions of preparations (0.1) was carried out every 10 days as a root top dressing to enhance growth.

According to the results of the research, the best performance was observed in Spruce blue when using the fertilizer "Potassium Humate + Mg", used to take into account the needs of this biological object (Fig. 1).

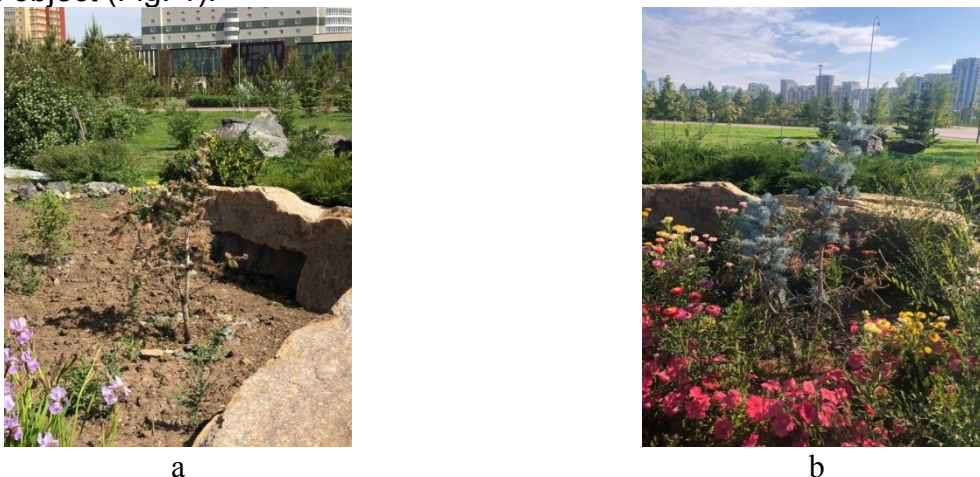


Figure 1. Spruce blue (*Picea pungens*) before (a) and after (b) the start of research.

During the period of the experiment, all of the above objects showed positive results: the splendor and leaf surface increased, the stems of the experimental groups acquired hardness in comparison with the control groups. The fruits of the pear tree ripened, and the appearance of large buds was observed in roses. Thus, these results indicate the effectiveness and prospects of using modified organic fertilizers based on potassium humate.

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Influence of structural-group composition and some trace elements on the biological activity of brown coals humic compounds

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Keywords: humic compounds, biological activity

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Humic acids (HA) and fulvic acids (FA) are raw materials for the production of highly effective preparations - long-acting plant growth stimulants. The ability of HA and FA to stimulate plant growth has been well confirmed by many studies conducted using a variety of plant species cultivated under various conditions [1-2]. The influence of HA and FA on the development of plants is reflected both in the growth of roots and the growth of seedlings, and in productivity.

Preparations based on HA and FA isolated from brown coals of the Tisulskoye deposit of the Kansk-Achinsk coal basin and the Tyulganskoye deposit of the South Ural basin were used in the work. Coal, HA, and FA samples were characterized by elemental and technical analyses, IR and ^{13}C NMR spectroscopy in the solid state.

Biological activity (BA) was assessed by phytotesting on wheat seeds of the Iren variety according to the phytoactivity index (FI), taking into account the seed germination energy (GE), root length (RL), and seedling height (SH) [2].

It has been shown that the biological activity of HA (in the form of soluble humates) and FA with respect to wheat seeds is directly proportional to the following structural parameters calculated from NMR spectroscopy data: the degree of aromaticity f_{ar} , the hydrophilic-hydrophobic parameter f_{hh} , and the parameter reflecting the ratio of aromatic and aliphatic fragments of the organic $f_{ar/al}$ masses. A study was made of the influence of the concentration of preparations on the growth parameters of plants. The most positive effect on wheat seeds is exerted by sodium humates with a concentration of 0.005% (IF 1.14 - 1.27). For FA isolated from the same coals, phytoactivity indices of 1.10–1.39 were obtained at the lowest concentration (0.00025%). The most pronounced stimulating effect of drugs is manifested on the length of the roots and the height of the seedlings.

It has been shown that the presence of plant micronutrients such as copper, zinc, cobalt and manganese increases the biological activity of sodium humate solution. The main significance of trace elements is to increase the activity of various enzymes that catalyze many biochemical processes. The concentration of sodium humate in the solution varied from 0.01 to 0.0005%, the concentration of microelements from 0.1 to 0.001%. The maximum biological activity is shown by preparations (0.005% by humate), the content of trace elements of which does not exceed 0.001%.

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Effect of the humic drug "EldORost" on the yield of potatoes and vegetables

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The use of environmentally safety products of natural origin is a global trend today. Currently, worldwide interest in humic fertilizers has sharply increased. This is due to the fact that more and more data is being accumulated on the positive impact of humic substances on plant growth and development, as well as on the quality of agricultural products and soil fertility. Humates, being physiologically active substances, regulate and intensify metabolic processes in plants and soil, and contribute to the active intake of nutrients in a form that is digestible for plants. "EldORost" is a ballastless preparation of humic nature of a new generation with a high degree of humification. The product contains easily soluble physiologically active salts of humic and fulvic acids (humates and fulvates), a complex of amino acids, macro- and microelements in a form accessible to plants. It is an environmentally friendly product, completely soluble in water, which is extremely convenient for drip irrigation system. It is designed for all types of crops in any soil and climate zones, acts as plant hormones: the optimal concentration of the drug is 0.0001%.

Laboratory and demonstration field tests conducted on potatoes and main vegetable crops (tomato, cucumbers, cabbage, carrots, onions, beets) showed high efficiency of the growth stimulator of the growth and development of plants "EldORost", the indicators of which exceeded the known reference drug. The preparation significantly improves the sowing qualities of seeds: increases germination and seed germination energy, intensively stimulates root formation in plants. Accelerates growth processes: increases the height of plants, the length of leaves, the number of leaves on the plant, the number of lateral stems, the number of tubers on the bush, the number of fruits on the plant, the mass of tubers from the bush, the total mass of plants, the diameter and length of the root crop. It increases the yield, the period of fruiting, reduces the term of crop ripening by 10-12 days, improves the consumer properties of the obtained agricultural products. "EldORost" has a positive effect on the productivity of potatoes and major vegetable crops, significantly increasing the productivity of their yield compared to the control variants of the field experiments, additional yield: cabbage - 31.3%, cucumbers - 30.2%, tomatoes - 34.7%, table beets - 28.7%, carrots - 33.3% and potatoes - 36.8%.

The preparation is used for pre-sowing seed treatment, foliar treatment during the vegetation period as an independent fertilizer and post-harvest soil treatment. Its application does not require changing the existing agricultural technologies - it is compatible with any plant protection agents and other fertilizers. It is used in the form of a working solution both independently and in tank mixtures with plant protection agents and soluble fertilizers within the planned treatments. It increases the efficiency of mineral fertilizers and pesticides, reducing their use by 20-40%. The results obtained experiments on potatoes and main vegetable crops showed high efficiency of the humic nature growth stimulator "EldORost". This confirms that the use of humic compounds - the most promising direction of agricultural technology. Their use is actively expanding, especially in connection with the desire to make agriculture as environmentally friendly, efficient and economical as possible.

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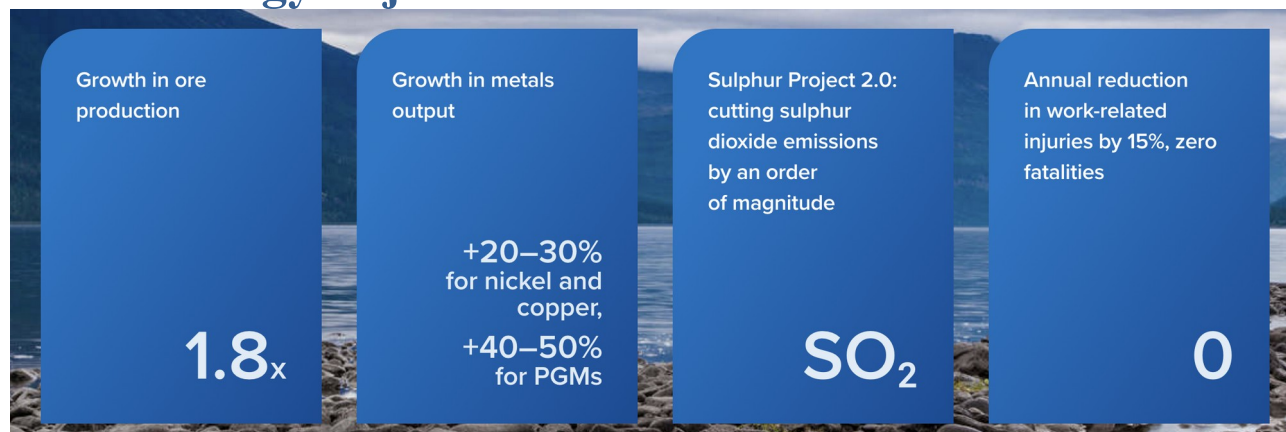
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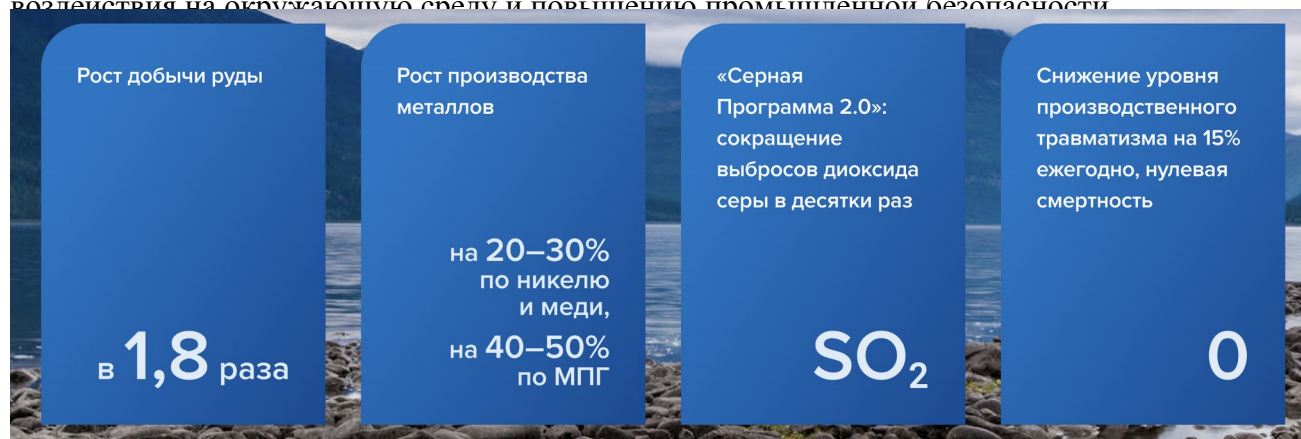
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Leonardite mining is carried out on the Sakhalin island, Russian Federation. All stages of extraction and selection of raw materials are carefully monitored by our specialists.



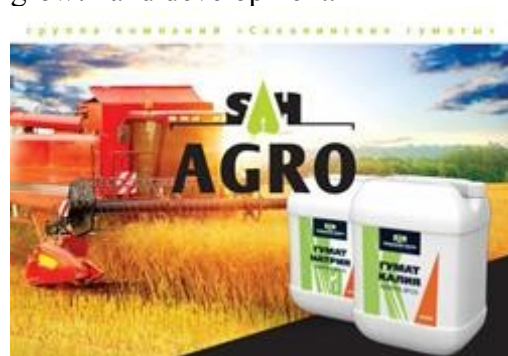
Sakhalin Humate GC products have a high content of humic substances and are available in various formulations (powder, aqueous solution, granules). Especially for agriculture, the SAKHALIN HUMATES Group of Companies developed Potassium Humate and Sodium Humate. These preparations are produced under the brand name AGRO and are positioned as a line of professional agrochemistry. Distinctive features of humates of this line are high degree of oxidation of hydrocarbon chains and increased content of low molecular weight humic acids. Humate "Sakhalinsky" under the AGRO trademark contain an optimum set of microelements in chelated form: silicon, iron, magnesium, sulfur, zinc, cobalt, copper, manganese, etc.

Contacts:

www.humate-sakhalin.ru 105082, Russian Federation, Moscow, ul. Bakuninskaya, d. 74-76, building 1, room 29/1328 e-mail: green_island@inbox.ru
Tel / fax: +7 (495) 648-90-55

It goes well with fungicides, herbicides, growth regulators, with urea and alkaline microelement solutions.

The preparations have successfully passed a full range of agrotechnical tests and are recommended for widespread implementation, both for reclamation and restoration of depleted and infected lands, and as an effective accelerator of plant growth and development.



The use of humates significantly increases uptake of mineral nutrition and productivity of plants.

The high effectiveness of their use is confirmed both with the facts and figures of scientific reports of the best agricultural chemists and soil scientists of Russia, as well as by the positive reviews of agricultural enterprises that already use Potassium Humate and Sodium Humate ® AGRO. Humate "Sakhalinsky" is fully suitable for organic farming.



NL International - международная торговая марка, представленная более чем в 12 странах мира с большой сетью дистрибьютеров и клиентской базой в России. На российском рынке компания работает с 2000 года. В ассортименте NL — более 450 наименований продукции, среди которых функциональное питание, БАД, уходовая и декоративная косметика, средства для ухода за домом.

Собственный производственный комплекс компании расположен на родине бренда в г. Новосибирске и сертифицирован по стандарту ISO 22000 — соответствие международной системе менеджмента в области безопасности пищевой продукции. Сертификацию предприятия проводила SGS – ведущая независимая мировая компания.

В 2022 г. компания получили сертификат GMP – Good Manufacturing Practice. С 2018 г. производство NL имеет сертификат «Халяль», который подтверждает, что компания соответствует исламским требованиям в вопросе производства продукции.

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We are the Regional Chapter of the Commonwealth of Independent States of the International Humic Substances Society (CIS IHSS). The chapter was founded in Zvenigorod, Moscow Region, Russia at the Open Meeting of the Russian IHSS - Chapter held on September 25, 2002. As such, CIS-IHSS is a successor of the Russian IHSS Chapter founded in 1994. The main goal of the CIS IHSS is consolidation of the efforts of the CIS scientists in the field of basic and applied humic research. At present, the CIS IHSS has 150 members and is the largest chapter of the IHSS. The membership includes scientists, students, engineers, practitioners, and business representatives from Belarus, Kazakhstan, Kyrgyzstan, and Russia. Since 2022 the CIS members from Ukraine formed a separate national chapter of IHSS - Ukraine Chapter. The scientists and engineers are both from research institutes and industrial firms. The scope of scientific interests includes structure, molecular properties, genesis of humic substances, interactions of humics with heavy metals and organic ecotoxins, application of humics for remediation technologies, biological activity of humics; technologies of manufacturing and agricultural applications of humics, and others. We are glad to be a part of the world-wide humic research community. We are open for cooperation and any kind of HUMIC activities.

For more information about the CIS IHSS visit our website at <http://www.humus.ru/ihss/>. Information about the IHSS you can find at the website: <http://www.humic-substances.org>

You are very welcome to join us!

Mariya V. Zykova
Regional Coordinator of the CIS IHSS

Kristina A. Bratishko
Secretary of the CIS
IHSS

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