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Humic derivatives as promising hormone-like materials

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The aim of this research is to prepare novel bio-inoculants derived from coal humic substances (HS) using bio-solubilization technique. This approach can be considered to some extent as model for supply plants with available nutrients throw the mineralisation of organic matter in soils by bacteria and fungi. Screening for the stable and active microorganisms' strains possessing ability to degrade humic substances was performed. The following subjects were examined using different isolation methods: natural microbial population from city soil, wood rot of Ulmis Pamila and biohumus of vermiculture of Eisenia foetida. Approaches for monitoring the humics-solubilizing fungi growth under liquid surface conditions in the presence of HS, proper conditions of bio-solubilization technique were elaborated. Coal humic acids (HA) from oxidized brown coal (Kyrgyz deposits) were isolated and added to a Czapek nutrient broth which was used either in full strength or without nitrogen source. The individual flasks were inoculated with natural microbial populations of corresponding cultivated soil, biohumus and wood rot samples for 12 months. Evaluation of phyto-hormonal activity of the produced HS and their derivatives in respect to higher plants with auxine and gibberellic tests was performed.

To characterize structure of the biopreparations obtained, an experimental approach was undertaken that implies application of different complementary techniques for the structural analysis of biopreparations. As those were used: elemental and functional analysis, FTIR and 1H, 13C NMR spectroscopy and size-exclusion chromatography.

According to the elemental composition of HS recovered from microbial cultures, a decrease in carbon and a significant increase of nitrogen in HS reisolated from the full strength broth inoculated with wood-decay microorganisms has been found. If biohumus microorganisms were used as inoculum, only minor changes were detected in the elemental composition of HS. A significant increase of H/C and O/C was also found in the HS. It can be attributed to formation new aliphatic and O-containing structures and decrease aromatic ones. Accumulation of fulvic acids was recorded in 6 months incubation. In 9 months, natural microbial populations from soil, biohumus, and wood rot had reduced the absorbance of HS media by 79, 75, and 62%, respectively.

A relative reduction of the molecular weight was noticed after 3 months incubation, and accumulation of new low molecular weight fraction after 6 months incubation was recorded after chromatography on Toyopearl HW-50S. Reductions in amount were due to a random degradation of substances in all molecular size classes. A formation the high molecular weight fraction has been found, that can be caused by cross-linking of structural constituents of molecules due to radicals forming after biodestruction or by their interaction with metabolites.

Data obtained by spectroscopic methods (UV/vis/FTIR) and element analysis indicated a decrease in particle size and a loss in aromaticity and aliphatic carbon in HS reisolated from microbial cultures. Simultaneously an increase in the N content of HS was observed, which probably from some constituents of microbial biomass such as proteins and aminosugars. The microbial degradation of HS strongly depended on the composition of the HS, the species selection of the microorganisms, and to a lesser extent on the culture conditions.

A hormone-like activity has been showed by HS preparations which were characterized with low molecular weights (∼5-15 kD). Each of these preparations was endowed with a single specific (auxin-like or gibberellin-like) activity. Biosolubilized HS with low molecular weight were displayed two kinds of activity.

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