No harm to living beings: SibFU scientists to test contaminated soils on ferments

Scientists of Siberian Federal University and the SB RAS Institute of Biophysics have developed the concept of comprehensive testing of agricultural soils contaminated with various chemicals, including heavy metals, pesticides and fertilizer components.

The proposed test systems are based on enzymes, not living organisms. With the help of new test systems, the conclusion about toxic substances in the soil can be made by suppressing the enzymatic reactions responsible for the various functions of a living organism: respiration, glow, etc., but the result is more accurate than when we use classical methods of biological analysis. Significant research results are published in Proceedings of the Academy of Sciences.

The anthropogenic pressure on the environment is increasing year by year, so is the need to control the environmental safety of water, soil, crops which agro-industry is growing in different countries. The industry massively uses biotests to detect contaminants and take prompt measures to eliminate them. For example, they check the pollution of water or soil with the help of simple organisms — bacteria, algae or Daphnia crustaceans. If their well-being changes, this indicates the presence of hazardous substances in the samples. The accuracy of such biotests is low as living organisms are variable and heterogeneous even within the same population, and the results of the analysis can vary significantly due to the conditions of preparation of the test isolate of the organisms, to put it simpler, the results are affected by how the living testers were grown and stored before the experiment.

'Our research team has developed integrated test systems designed for environmental monitoring of soils. These are enzyme biotests, which are more sensitive and accurate than commonly used standard biotests. They provide reliable results and are less dependent on random factors than, for example, daphnia tests – by the way, no harm was done to even a single living creature as a result of our work,' said **Elizaveta Kolosova**, junior researcher at the Laboratory of Bioluminescent Biotechnology of Siberian Federal University.

For their experiments, the scientists selected chains of enzymes that ensure the functioning of certain functions of a living organism - one enzyme is responsible for respiration, the other is involved in digestion, etc. As a result of studies of more than 10 such systems, the researchers chose three enzyme systems with the maximum sensitivity to different classes of toxic substances found in soils. These are butyrylcholin esterase, a bienzyme chain of NAD(P)H: FMN-oxidoreductase + luciferase, and a three-enzyme system of lactate dehydrogenase + NAD(P)H: FMN-oxidoreductase + luciferase.

'The important thing here is that when we use all three systems in a complex to test soil samples, we get an exhaustive answer, let's say, the most complete list of noxiousnesses which these samples contain,' **Ms. Kolosova** continues.

The scientists chose the main agricultural pollutants as model toxicants: organophosphorus and organochloride pesticides, titanium dioxide nanoparticles (it is used, in particular, as a plant growth stimulator) and copper chloride contained in some pesticides.

> *'We tested the sensitivity of each enzyme to toxicants first. An enzyme is essentially a catalyst. It starts and intensifies chemical reactions. If the enzymatic reaction does not occur, it is quenched in the presence of some toxicant — this is the pollution-detected! signal. So we selected enzymatic systems that revealed the presence of contaminants were selected. Next, we evaluated the sensitivity of the enzyme systems to pollutants in our experiments with soil extracts,'* commented **Oleg Sutormin**, co-author of the study, assistant to the Department of Biophysics.

According to the researcher, the team chose five standard soil samples for their experiments: sand, light loam, medium loam, heavy loam and chernozem (fertile black soil rich in humus and with a lighter lime-rich layer beneath). They used an aqueous extract from the soil without adding toxic substances as the reference soil to exclude the influence of the components of uncontaminated soil on the test results.

'Ultimately, the maximum objective of this study is to understand how toxicants found in the soil affect the health of a person who eats vegetables and cereals grown in the land of Krasnoyarsk Territory and, in the future, other regions of Russia. After all, all living organisms, not only insects or animals but also humans, have the enzymes used in the new test systems. It's impossible to experiment on humans for obvious reasons, but we managed to select precisely the set of enzymes which, on the one hand, detects well all kinds of contaminants of such a complex multi-component object which soil is, and on the other hand, most of these enzymes dwell naturally in each of us. Now this area seems very promising since it was possible to prove the ability of enzyme tests to replace living organisms during biotesting of complex natural environments,' said **Valentina Kratasyuk**, Doctor of Biological Sciences, professor, head of the Department of Biophysics, Siberian Federal University.

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