DYNAMICS OF PCB-CONTAMINATED SOIL AND SILT TOXICITY CHANGE IN THE COURSE OF BIOREMEDIATION *IN SITU*

Vladimir V. Kapranov[1], Gennady A. Zharikov[1], Valentina P. Dyadishcheva[1], Alexey V. Konoplev[2].

1- SFES Research Center for Toxicology and Hygienic Regulation of Biopreparations, Bld.102A Lenin Str., Serpukhov, Moscow region, 142253, Russia, Tel.: +7(4967) 399738, Fax: +7(4967) 399738, E-mail: microb@rambler.ru; toxic@online.stack.net

2- SPA "Typhoon" 82, Lenin Ave., Obninsk, Kaluga region, 249038, Russia, Tel.: +7(48439) 44204, Fax: +7(48439) 44204, E-mail: konoplev@typhoon.obninsk.ru;

Introduction

Integral toxicity[1] – systemic toxicity of complex mixture of substances - is perhaps the most important indicator of soil contamination by xenobiotics. This parameter actually determines the bioavailability of toxic substance for living organisms. Aqueous extract from soil contaminated by polychlorinated biphenyls (PCB) causes loss of more than 50% test models *(Dafnia magna)*, which indicates toxicity of the soil.

Upon getting into soil, PCB is strongly absorbed and remains there in readily available, potentially available, or bound forms[2,3]. Bioavailable forms of PCB, i.e. readily and potentially available ones are distributed in liquid phase of soil. Integral toxicity of PCB-contaminated soil is usually about 100%.

Materials and Methods

In our field experiment we were determining the change in integral toxicity of PCB-contaminated soil during the course of remediation. Microbial bioremediation was used, since other types of remediation are either less efficient or not environmentally friendly.

PCB-contaminated soils used in experiments were soddy-podzolic. The soil was taken from territory near former electrical engineering plant (specialized in condenser manufacturing), which had been contaminating nearby territories with PCB for about 30 years. The main route of contamination was via industrial and storm sewage systems, followed by contaminated wastewater flow into Borovlyanka river and then into Oka river, where PCB was accumulated in bottom silts. During flood seasons PCB was migrating to inundable lands.

To study soil integral toxicity during bioremediation we used a technique of acute toxic effect assessment in aqueous extract obtained from PCB-contaminated soil or silt; the aqueous extract was tested without diluting. *Daphnia magna* were used as test models.

Aqueous extract was preliminarily prepared for analysis on daphnia. Soil sample (0.5 kg) was mixed, any large particles were stirred, the sample was placed into a flat-bottom flask, added with 1 L of tap water (the water precipitated for 2–3 days or distilled water was used), plugged, and mixed on a magnetic stirrer. The obtained suspension was precipitated during 2 hours; then the supernatant fluid was poured into a vessel, and filtered through a thin layer of cotton wool. After all these procedures the obtained soil extract was ready for testing. The same water as was used in extract preparation served as control.

100 ml of control water and test extract were poured into vessels. The test was performed in three replications.

Short-term bioassay (up to 96 hours) allows determining acute toxic effect of soil/silt aqueous extract for daphnia based on their survivability. The average mean of number of test-models survived during certain period of time in test aqueous extract and in control serves as survivability index. Loss of more than 50% of daphnia during 96 hours in test water as compared to control serves as toxicity criterion.

10 one-day aged daphnia were placed into each vessel and exposed under optimal conditions during 96 hours. In short-term bioassay daphnia were not fed. Survivability was evaluated after 1, 6, 24, 48, 72, and 96 hours. If during any of the time periods daphnia loss was 50 % or more the bioassay of the variant would be stopped.

As a remediation method the previously developed technology of *in situ* bioremediation of PCB-contaminated soils using microorganisms-degraders *Alcaligenus latus* and *Hansenula californica* was used.[4,5]

The cultivated microorganisms-PCB degraders were introduced into soil or silt sites contaminated by PCB. Prior to introduction the sites were dug to the depth of 20 cm. The amount of microorganisms introduced was 1 L of microbial suspension per 1 m^2 of soil or silt. Soil moisture was maintained at the level of 60%. New portions of microorganisms-PCB degraders were introduced every two weeks.

Experiment duration was 8 weeks, testing was performed every two weeks.

Results and Discussion
According to the results of field experiments (Table 1) after 14-28 days no toxic effect was observed both in silt and soil sites processed with microorganisms-degraders. Control sites were constantly toxic.

The given results are possible only if microorganisms-degraders have degraded PCB congeners available for water extraction from soil and silt. Due to this fact the amount of PCB extracted into water was insufficient to cause daphnia death.

A conclusion can be made from the results of field experiments, that microorganisms-degraders degraded the most bioavailable for them part of PCB on test sites during 28 days at maximum. As a result soil integral toxicity on these sites during bioremediation reduced to safe levels.

Therefore, introduction of strains-degraders leads to decrease in soil integral toxicity.

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5. Strain *Hansenulla californica* Yeast Used for the Decomposition of Polychlorinated Biphenyls// DOE Docket Number S–95385

Table 1: Soil integral toxicity during field experiment (% of daphnia loss)

Experiment variant	Amount of daphnia survived, units		% of daphnia loss as compared to control	Evaluation of integral toxicity
	in control	in test		
Initial soil (without microorganisms)				
Water (control)	30		0	not toxic
Soil + *Alcaligenes latus*		8	73	toxic
Silt + *Alcaligenes latus*		3	90	toxic
Soil + *Hansenula californica*		7	77	toxic
Silt + *Hansenula californica*		0	100	toxic
Soil (control)		8	73	toxic
Silt (control)		0	100	toxic
Soil after 14 days of bioremediation				
Water (control)	30		0	not toxic
Soil + *Alcaligenes latus*		21	30	not toxic
Silt + *Alcaligenes latus*		0	100	toxic
Soil + *Hansenula californica*		30	0	not toxic
Silt + *Hansenula californica*		8	77	toxic
Soil (control)		0	100	toxic
Silt (control)		0	100	toxic
Soil after 28 days of bioremediation				
Water (control)	30		0	not toxic
Soil + *Alcaligenes latus*		27	10	not toxic
Silt + *Alcaligenes latus*		25	17	not toxic
Soil + *Hansenula californica*		30	0	not toxic
Silt + *Hansenula californica*		20	33	not toxic
Soil (control)		7	73	toxic
Silt (control)		3	90	toxic
Soil after 42 days of bioremediation				
Water (control)	30		0	not toxic
Soil + *Alcaligenes latus*		30	0	not toxic
Silt + *Alcaligenes latus*		21	30	not toxic
Soil + *Hansenula californica*		27	10	not toxic
Silt + *Hansenula californica*		28	7	not toxic
Soil (control)		1	97	toxic
Silt (control)		0	100	toxic