Original article

DOI: 10.2478/agri-2025-0014

PCBs: *Ex situ* short-term vegetation screening of the immobilization effect of differentially activated soil-applied humic preparations

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Tóth, Š. (2025). PCBs: *Ex situ* short-term vegetation screening of the immobilization effect of differentially activated soil-applied humic preparations. *Agriculture (Poľnohospodárstvo), 71*(4), 161 – 175.

The container trial scope was to screen the immobilisation effect of humic acids on polychlorinated biphenyls (PCBs), verifying a set of soil-applied preparations of HUMAC® Agro (HA) – Enviro (HE) – Sorbent (HS). A secondary goal was to formulate main closures for subsequent semi-operational *in situ* experiments focused on the Slovakian infamous case of Chemko Strážske, a facility of the former PCB producer, where the surrounding environment is polluted. Two experiments were established on August 3, 2020, by treating the upper 15 cm layer of soil. A week later, the sown radish *Raphanus sativus* L. var. *sativus* cv. Vinara F1 was sown. The crop was harvested in four cumulative harvests according to maturity, with soil samples collected on September 30 at the end of the trial. In trial A, the substrate was contaminated by extremely highly polluted bottom sediment taken from the waste channel of the facility and separately treated with the humic preparation in the ameliorative dose of 3 t/ha. In radish tubers, the sum of PCBs (PCB 28, 52, 101, 138, 153, 180) decreased by 32.73, 39.41 and 29.4% in comparison to the contaminated control, while in the leaves this reduction was 7.0, 15.6, 53.8%, to HA – HE – HS. The content of PCBs released from the soil into the soil leachate decreased by 49.3, 48.8 and 32.2%, respectively. Trial B was designed to test the double-dose effect, where agricultural soil with aged contamination was treated with HE only. Relating to the untreated control, a higher effect of a simple dose was recorded in comparison to double one, the content of PCBs was reduced by 73.2 vs. 60.5% in tubers, and by 23.1% vs 22.2% in leaves, respectively.

Keywords: PCB, environmental pollution, decontamination, leonardite activated, bioconcentration factor

The priorities of developed society, to which the majority of European countries, belong and Slovakia as well, include the improvement of multifunctional agriculture. From an agro-ecological point of view, such agriculture not only produces but also participates in the protection of the environment and the maintenance of the natural character of the landscape (Javorská *et al.* 2007; Di Guardo *et al.* 2020; Terzaghi *et al.* 2021, 2019; Aviantara *et al.* 2023; Amutova *et al.* 2024). To achieve such broadly defined goals, it is also necessary to manage problems related to old ecological burdens that pollute the environment and especially agricultural land (Kays & Zhang 2004; Turhan 2007; Waterfall *et al.* 2009; DiGuardo *et al.* 2017). One such issue is the region of Zemplín, a historical county in Slovakia and Hungary, whose middle part is considered to be one of the most polluted areas with polychlorinated biphenyls (PCBs) in the whole of Europe. In the Middle Zemplín, PCBs are considered to be ubiquitous, which is documented especially for the broad area near the town of Strážske, just the site where PCBs were produced in large amounts in the past (Pavlíková *et al.* 2007; Pilváňová 2007; Papajova and Juris 2009; Lászlová *et al.* 2018; Horváthová *et al.* 2018; Fülöp *et al.* 2021).

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The solution to this environmental issue remains highly relevant even nearly four decades after the production of PCBs at the Chemko Strážske facility in Slovakia ceased on 14 January 1984, especially for residents of the Michalovce district, and partly for neighbouring districts as well (Danielovič *et al.* 2003; Turčeková *et al.* 2008; Danielovič *et al.* 2014). The average PCB levels in the Slovak population are among the highest compared to data from other countries, while the average levels in the bodies of residents of the Michalovce district were even four times higher than those in a control group from the Slovakian district of Stropkov (Pavlíková *et al.* 2007; Pilváňová 2007; Turčeková *et al.* 2008; Novotny *et al.* 2021; Horváthová *et al.* 2024). This also indicates significant environmental contamination. Since PCBs mainly enter the human body through the food chain, addressing this problem should particularly focus on primary agricultural production to protect public health (Tóth 2024).

Historically, PCBs were regarded as harmless substances mainly used in paint formulations, heat transfer fluids in heat exchangers, or insulating fluids in transformers and capacitors. The potential environmental hazard of PCBs was first highlighted by Jensen in 1966 (Jensen 1966), who associated changes in the reproductive capacity of birds in Sweden with PCB levels in tissues. Today, numerous experiments and studies confirm the harmfulness of PCBs, particularly their adverse effects on human health. Since PCBs belong to the group of persistent organic pollutants, considerable efforts are directed towards reducing their presence. Globally, various decontamination and immobilisation methods are being tested, with humic acids often showing promise (Pignatello & Xing 1996; Cocarta *et al.* 2016; Lászlóová *et al.* 2018; Sadaňoski *et al.* 2020; Amutova *et al.* 2024; Lü *et al.* 2024; Shi *et al.* 2024).

The primary aim of the research was to assess the immobilisation effect of variously activated soil-applied humic acids, the active component of HUMAC® Agro – Enviro – Sorbent formulations, on PCB indicative congeners during *ex situ* short-term vegetation screening. If results proved environmentally beneficial, the secondary aim was to develop key guidelines for potential solutions to address this environmental issue in future semi-operational pilot studies focused on the agronomic immobilisation of PCBs under *in situ* conditions.

MATERIAL AND METHODS

Implementation, Short Preview and the Activities Chronology

First, soil and sediment sampling were carried out from the contaminated area and control area, whereby (i) sampling of highly contaminated bottom sediment was performed in March, and (ii) arable soil samples were taken from the contaminated and control area in July 2020 (Figures 1, 2).

Second, two container trials (AB) were established on 3 August 2020 and lasted until 30 September 2020. Both trials aimed for *ex situ* vegetation screening, using a short-term model crop of sown radish *Raphanus sativus* L. var. *sativus,* a cultivar of Vinara F1. Five seeds were sown into each container, at a depth of 1.5 cm, with the containers having a volume of 12.133 dm³, a slightly conical shape, and a surface area of 615.44 cm². In trial A, the substrate was contaminated on 3 August, while the soil was not contaminated in trial B. The treatment and application of differentially activated soil-applied humic preparations of the HUMAC® series (based on leonardite, manufactured by HUMAC Ltd., Košice, Slovakia) to the substrate were performed on 3 August. The sowing of the model crop occurred a week later, on 10 August, and the crop was sampled on 14 August. The model crop was sampled on four consecutive dates (9, 16, 19, and 29 September) as individual tubers ripened to harvest maturity, while the soil was sampled on 30 September at the end of the experiment.

Third, the plant and soil samples were processed after collection, drying them at room temperature in a well-ventilated storage room until reaching a constant weight. Laboratory analysis of the samples was conducted in October 2020.

Trial A, the Contamination

The extremely highly contaminated bottom sediment was collected from the waste channel of the Chemko Strážske facility, the former producer of PCBs. This sediment was used to contaminate

soil 1, which is an arable soil obtained from an uncontaminated area, as the site is located 30 km in a straight line to the southwest of the facility.

Both of the matters were homogenised separately, the soil was sieved with a mesh size of 3 × 3 mm, and the sediment was sieved with a mesh size of 1 × 1 mm. Then, the moisture content of the matter was measured just before the contamination, dry matter (DM) content was 48.8% at the bottom sediment, and 67.1% at Substrate 1 (68.4% at Substrate 2 applied in trial B).

The contamination was carried out as follows: a dose of 50.0 g of bottom sediment per 3,000.0 g of soil 1 was applied, both in dry matter (DM). Then, both homogenised masses were mixed for 2 minutes, and the contaminated substrate was placed into a container as a 15 cm upper layer, with a 15 cm lower layer of uncontaminated soil 1 added first. Before adding the upper layer, the contaminated substrate was treated with a humic preparation at a dose of 3 t/ha, with separate treatments using HUMAC® Agro (T1), HUMAC® Enviro (T2), and HUMAC® Sorbent (T3). Two humic-free controls were performed, consisting of contaminated substrate (T4, soil 1 + bottom sediment) and uncontaminated soil (T5, soil 1). The experiment included two repetitions, with ten trial units randomly assigned by container number, which used two-digit numbers.

Trial B, Dose Doubling

As the bottom sediment and soil/substrate 1 were applied in trial A, trial B involved applying soil/substrate 2 without the bottom sediment. Soil 2 was sourced from arable land in a contaminated area, specifically from a site 500 metres from the Chemko Strážske facility. Since there was no need to contaminate Soil 2, it was more suitable to carry out treatments with varied doses of humic preparation. The selected doses of HUMIC Enviro were the simple dose of 3 t/ha (T6), the double dose of 6 t/ha (T7), along with an untreated control (T8, soil 2). The process of preparing, placing the substrate into a container, and randomising the trial units was similar to that of trial A, apart from the contamination process.

Laboratory Analyses and Soil Conditions

Plant and soil samples for laboratory analyses were collected in the manner described hereinbe-

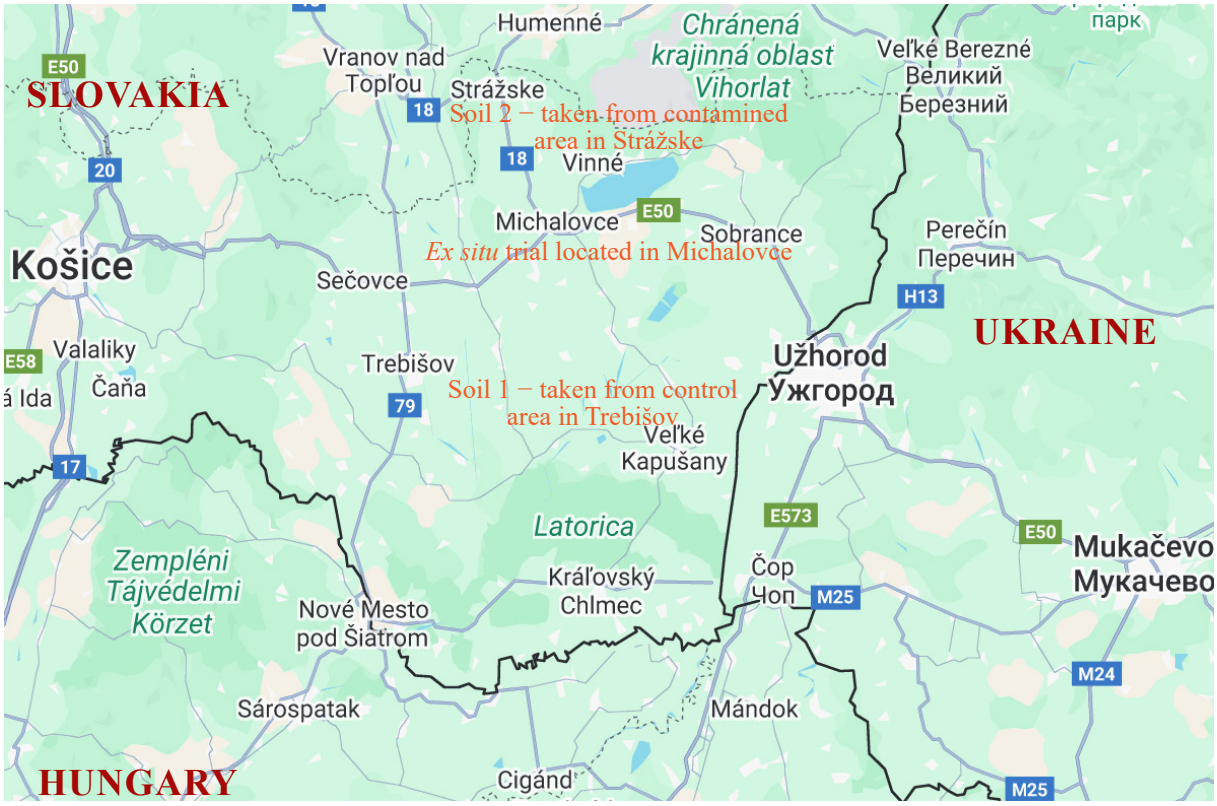


Figure 1. The map of the *ex situ* trial and the area of collection and contaminated soil.



Figure 2. Withdrawal of bottom sediment from the waste channel of the PCB producer's former facility in Strážske (A); conducting an *ex situ* container experiment in Michalovce (B); performing the experiment in Michalovce: the model crop *Raphanus sativus* L. var. *sativus,* cultivar Vinara F1 at full emergence (C), and an individual plant at harvest maturity (D); continuous processing of plant samples (EF); March (A), August (BCD), and September 2020 (EF).

fore. Before the experiment, the soil samples were analysed for key chemical properties (Table 1). The soil sampling, sample storage and processing were conducted according to Slovakian national law no. 151/2016 Z.z. (Codex), with laboratory analyses performed using the Mehlich 3 method (content of P, K, Ca, and Mg) (Mehlich 1984). Total nitrogen (Nt) was determined by the Kjeldahl method (Hraško *et al.* 1962). Soil organic carbon (C-ox) and humus content were indirectly measured by Tjurin's method (ISO 14235:1998). The soil pH in KCl (exchangeable soil reaction) was measured using a potentiometric method (ISO 10390:2005). For clay content and soil type classification, the standard Novak method was applied (Sobocká 2000).

The bottom sediment and soil collected from the contaminated area were analysed for initial PCB content (Table 2). At trial A, final soil samples were taken the day after the last crop harvest, from a depth of 0 – 0.15 m. During both trials (AB), all five plants per container were harvested on four occasions, when the tubers reached maturity. All samples were air-dried at room temperature in a well-ventilated storage room. PCB levels in the samples were measured using High-Resolution Gas Chromatography with an electron capture detector (HRGC, GC/ECD). Plant sample analyses were conducted at the National Reference Laboratory for PCBs (ISO/IEC 17025:2005 accredited Laboratory of Foreign Substances of the State Veterinary and Food Institute in Košice, ŠVPÚ, Slovak Republic), while soil samples were analysed at the Laboratory of Foreign Substances at the Research Institute of Agroecology in Michalovce (NPPC), Table 3.

The PCB substances were extracted from soil and bottom sediment samples using an extraction agent consisting of n-hexane and acetone (2:1), with the Soxhlet apparatus employed. For the purification of extracts and qualitative-quantitative estimations, the protocol of Obligatory Methods of Soil Analyses (Kobza *et al.* 1999) was followed. HRGC was characterised with the column HP-5 (5% diphenyl and 95% dimethyl polysiloxane), which was 50 m long, had an internal diameter of 0.2 mm, and a stationary phase width of 0.33 μm. It was equipped with an automatic sample injector, an electron capture detector, and a chromatographic station, providing sufficiently specific and selective identification and quantification of individual PCBs at trace levels (Němeček *et al.* 1994). For releasing PCB congeners from radish tubers and leaves, samples were extracted with diethyl ether, while purification and quantification procedures matched those used for soil and sediment samples. The concentrations of PCBs, specifically congeners nos. 28, 52, 101, 138, 153, and 180 (according to IUPAC classification), were measured using gas chromatographs: (i) Agilent GC 6890 Series for the plant samples (ŠVPÚ Košice) and (ii) HP 5890 Series II for the soil samples (NPPC Michalovce).

Table 1

The selected chemical parameters of soil/substrate, and classification of the soil type

Substrate	Nt [mg/kg]	P [mg/kg]	K [mg/kg]	Ca [mg/kg]	Mg [mg/kg]	pH/KCl	C-ox [%]	Humus [%]	Clay content [%]	Soil type
Soil 1	0.178	50.8	202.6	5,982.7	478.1	6.61	1.629	2.808	38	middle
Soil 2	0.191	68.5	297.8	3,244.7	480.7	6.32	1.850	3.188	32	middle

Note: Soil 1 – arable soil from the control area, Substrate 1 for trial A; Soil 2 – arable soil from the contaminated area, Substrate 2 for trial B.

Table 2

The content of PCBs indicative congeners in the Bottom sediment and Substrate 2 (in mg/kg)

Substrate / PCB congener	28	52	101	138	153	180	Σ PCB
Bottom sediment	2.246	4.340	2.799	2.978	2.963	2.610	17.936
Substrate 2	0.008	0.012	0.021	0.043	0.041	0.037	0.162

Table 3

Parameter, test method, and determination and quantification limits, according to PCBs indicative congeners and workplaces

Parameter / Lab	ŠVPÚ Košice			NPPC Michalovce		
Material	Plant samples			Soil samples		
PCB congener	Method	LOQ / LOD [µg/kg]	U (k=2)	Method	LOQ / LOD [mg/kg]	U (k=2)
PCB-28	GC/ECD	0.14 / 0.05	14%	GC/ECD	0.002 / 0.001	20%
PCB-52	GC/ECD	0.16 / 0.05	16%	GC/ECD	0.002 / 0.001	20%
PCB-101	GC/ECD	0.15 / 0.05	15%	GC/ECD	0.002 / 0.001	20%
PCB-138	GC/ECD	0.14 / 0.05	17%	GC/ECD	0.002 / 0.001	20%
PCB-153	GC/ECD	0.17 / 0.06	16%	GC/ECD	0.002 / 0.001	20%
PCB-180	GC/ECD	0.17 / 0.06	14%	GC/ECD	0.002 / 0.001	20%

Note: LOQ – limit of quantification; LOD – limit of detection; U (k=2) – expanded combined uncertainty with a coverage factor of 2 (for LOQ).

Weather Conditions

During the experiment, meteorological conditions were also monitored. Data on average daily temperature and precipitation for the short-term growing period of the model crop, sawn radish cv. Vinara F1, were obtained from a meteorological observation station located near (up to 1,500 m in a straight line) the *ex situ* experiment site in Michalovce. This station is part of the SHMÚ (Slovak Hydrometeorological Institute) network, ensuring high-quality records. The average daily temperature and total precipitation for August and September 2020 were 20.1°C and 81.6 mm, and 16.7°C and 54.8 mm, respectively.

Data Processing and Applied Statistics

All laboratory analyses, on the main chemical properties as well as on PCBs content, were done in classic twin laboratory analyses. The content of six indicative congeners of PCBs was detected analytically, and the sum of the six congeners (∑ 6PCB) was calculated mathematically:

$$\sum 6PCB = \text{content of PCBs } (28+52+101+138+153+180); \text{ in ppm or ppb}$$

It is notable also that the content of PCBs in soil samples is expressed in mg/kg (ppm, 10^{-6}), while the content in plant samples is in µg/kg (ppb, 10^{-9}). In the case of plant samples, the values of the content of PCB congeners under the quantification limit (LOQ) are considered zero, in the case of soil samples, the same is valid for the values under the limit of detection (LOD). A descriptive statistics procedure was performed to obtain the sum and average data as well, whereby simple counting of authentic data by the treatments was applied, and in the figures, the standard deviation was added. Two indexes were calculated as well:

(i) the effect of reduction, which is calculated according to the humic treatments by the trials (T1, T2, T3 relating to the untreated control of T4 concerning trial A; and T6, T7 relating to the untreated control of T8 concerning trial B):

$$\text{effect of reduction} = 100 - \frac{\sum 6PCB \text{ (treatment)}}{\sum 6PCB \text{ (control)}} \times 100; \text{ in \%}$$

(ii) and the bioconcentration factor, the ratio between the contaminant concentration in the plant (content in bulb and leaves together) and concentration in soil (end status concerning trial A, initial status concerning trial B), which is calculated according to the humic treatment:

$$\text{bioconcentration factor} = \frac{\sum 6PCB \text{ (in plant, ppm)}}{\sum 6PCB \text{ (in soil, ppm)}}; \text{ number}$$

RESULTS

The Trial A

The overall average content of indicative PCB

congeners in the model crop was 10.43 µg/kg, with radish tubers containing less than radish leaves −11.42 µg/kg vs. 10.05 µg/kg, respectively (Table 4). For individual congeners, the average content increased in the order PCB 28<PCB 180<<PCB 52 <PCB 101<PCB 138<PCB 153 (0.74<1.06<1.69<2.19<2.22<2.53 µg/kg, respectively), with significant differences in the first two compared to the others (Table 4, Figure 3). The similar order of the treatments was T5<<T3<T2<T1<<T4 (5.21<<8.89<10.90<12.05<<15.11 µg/kg, respectively), considering the average content in tubers and leaves, with the highest content in the contaminated control and the lowest in the uncontaminated control (Table 4, Figure 4). According to these results, each treatment reduced the content of the indicative

T a b l e 4

The content of indicative congeners of PCBs in the tubers and leaves of the sown radish in trial A (in µg/kg)

Treatment / PCB congener	R/L	PCB 28	PCB 52	PCB 101	PCB 138	PCB 153	PCB 180	Σ PCB
T1, HUMAC Agro	tubers	0.64	1.13	2.56	2.55	2.66	0.94	10.49
	leaves	0.97	2.96	2.26	2.93	2.96	1.53	13.61
T2, HUMAC Enviro	tubers	0.72	1.45	1.93	2.11	2.27	0.96	9.44
	leaves	0.91	2.58	2.40	2.43	2.76	1.28	12.36
T3, HUMAC Sorbent	tubers	0.71	1.68	2.37	2.54	2.67	1.03	11.00
	leaves	0.44	0.91	1.76	1.39	1.60	0.67	6.77
T4, control contaminated	tubers	0.75	2.80	3.45	3.07	4.18	1.33	15.58
	leaves	0.99	2.05	3.05	3.23	3.60	1.72	14.64
T5, control uncontaminated	tubers	0.45	<0.16	1.21	0.70	0.93	0.44	3.73
	leaves	0.81	1.38	0.87	1.24	1.64	0.74	6.68
Average	tubers	0.65	1.41	2.30	2.19	2.54	0.94	10.05
	leaves	0.82	1.98	2.07	2.24	2.51	1.19	11.42
	average	0.74	1.69	2.19	2.22	2.53	1.06	10.43

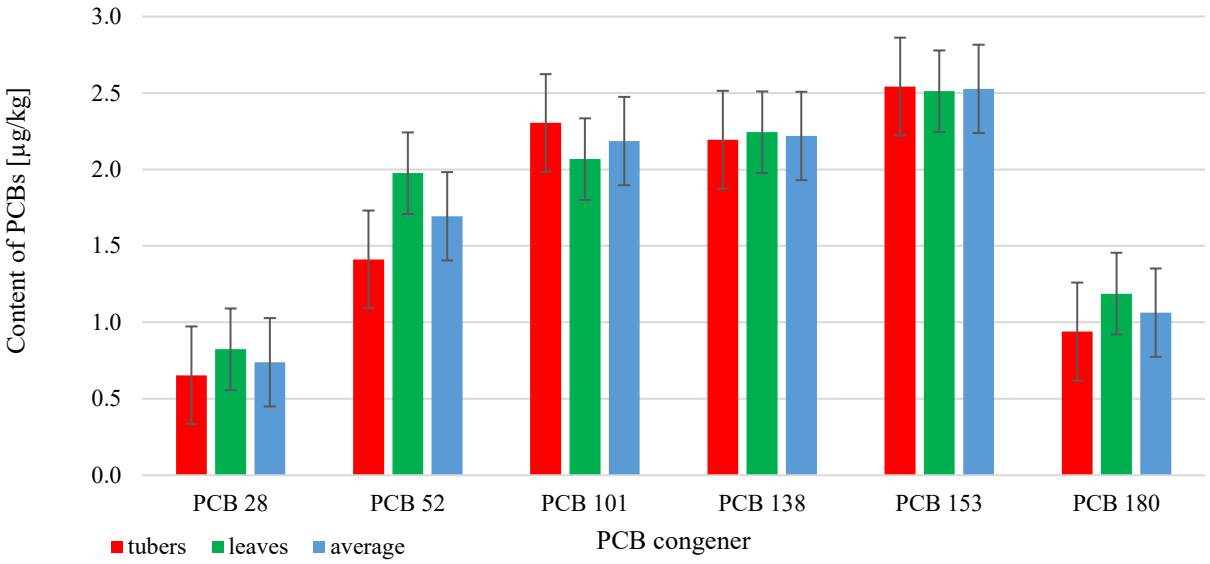


Figure 3. The content of PCBs in trial A by the indicative congeners, standard deviation added.

PCBs. Contrary to the general trend of higher PCB content in the leaves, treatment T3 showed a higher content in the tubers.

The bottom sediment used in trial A for substrate contamination was highly contaminated, as evidenced by the value 17.936 mg/kg (Table 2). The initial levels of most congeners ranged from 2.246 to 2.978 mg/kg, except for PCB 52, which had the highest content of 4.340 mg/kg. The soil-applied humic preparations from the HUMAC Agro (HA), Enviro (HE), and Sorbent (HS) series, used at their ameliorative dose of 3 t/ha, reduced the levels of indicative PCB congeners in the tubers and leaves of the model crop, and a decrease in PCBs was also detected in the soil at the final stage (Table 5). In radish tubers, the sum of PCBs decreased by 32.73%, 39.41%, and 29.40% compared to the contaminated control, while in the leaves, the reduction was 7.04%, 15.57% and 53.76%. In the soil leachate, reductions were 49.32%, 48.82% and 32.24%, respectively, for HA, HE, and HS. The bioconcentration factor declined in the order of HS < HE < HA, with values of 0.048 < 0.078 < 0.087, corresponding to T3 < T2 < T1, with untreated controls at 0.055 (T4) and 0.473 (T5).

The Trial B

In trial B, the overall average content of indicative PCB congeners in the model crop was 8.48 µg/kg, with the average content in radish tubers higher than in leaves, at 8.93 versus 8.03 µg/kg (Table 6). This was lower than in trial A, with a ratio that contrasts with it. The average content of PCB indicative congeners increased in the order of PCB 52 < PCB 28

T a b l e 5

The content of indicative congeners of PCBs in the soil, according to the treatments of trial A (in mg/kg)

Treatment / PCB congener	PCB 28	PCB 52	PCB 101	PCB 138	PCB 153	PCB 180	Σ PCB
T1, HUMAC Agro	0.046	0.055	0.041	0.048	0.046	0.042	0.278
T2, HUMAC Enviro	0.036	0.052	0.042	0.051	0.051	0.049	0.281
T3, HUMAC Sorbent	0.051	0.073	0.057	0.066	0.066	0.059	0.372
T4, control contaminated	0.070	0.097	0.086	0.105	0.101	0.090	0.549
T5, control uncontaminated	<0.001	<0.001	0.001	0.007	0.007	0.007	0.022

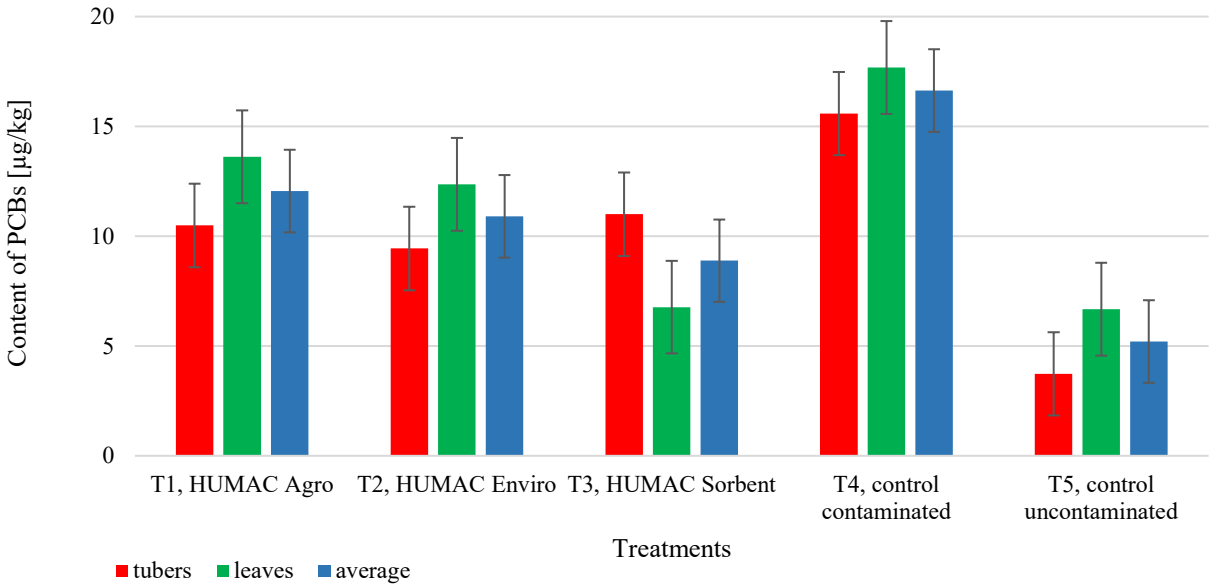


Figure 4. The content of PCBs in trial A according to the treatments, and standard deviation added.

T a b l e 6

The content of indicative congeners of PCBs in the tubers and leaves of the sown radish in trial B (in µg/kg)

Treatment / PCB congener	Plant part	PCB 28	PCB 52	PCB 101	PCB 138	PCB 153	PCB 180	Σ PCB
T6, HUMAC Enviro 3 t/ha	tubers	<0.14	<0.16	<0.15	1.54	1.75	0.77	4.06
	leaves	0.70	<0.16	0.86	1.87	2.42	1.02	6.87
T7, HUMAC Enviro 6 t/ha	tubers	<0.14	<0.16	1.68	2.16	2.74	1.00	7.58
	leaves	<0.14	<0.16	1.57	1.71	2.62	1.05	6.95
T8, untreated control	tubers	<0.14	<0.16	6.45	3.00	4.56	1.14	15.15
	leaves	<0.14	<0.16	2.12	2.94	3.57	1.64	10.27
Average	tubers	<0.14	<0.16	2.71	2.23	3.02	0.97	8.93
	leaves	0.23	<0.16	1.52	2.17	2.87	1.24	8.03
	average	0.12	<0.16	2.11	2.20	2.94	1.10	8.48

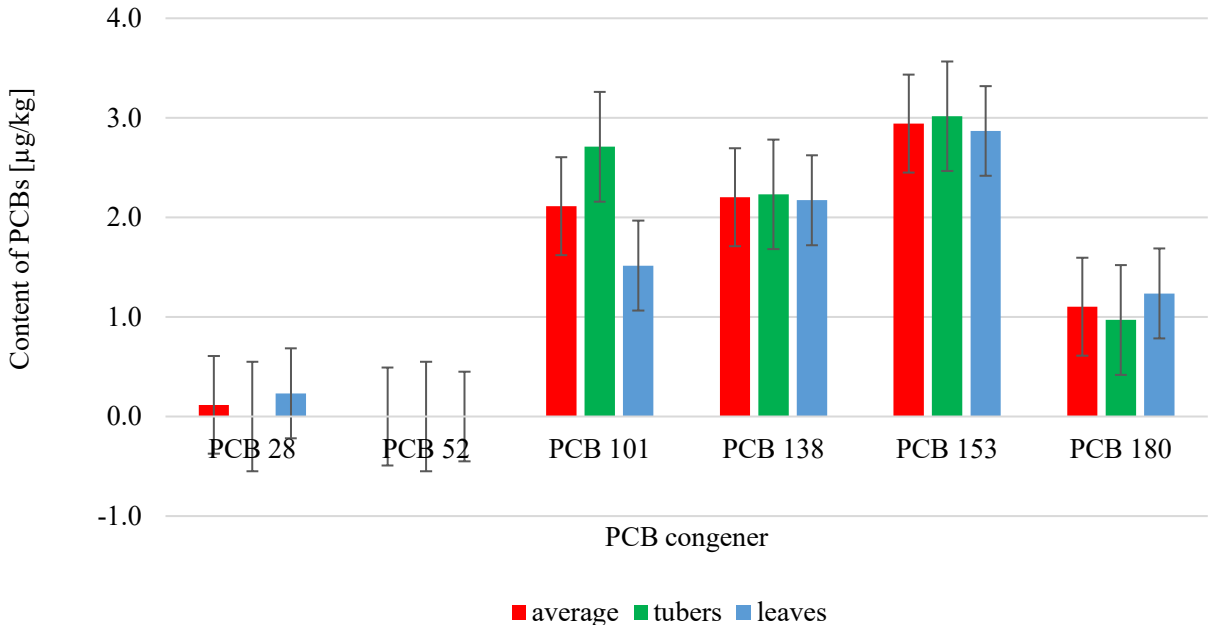


Figure 5. The content of PCBs in trial B by the indicative congeners, standard deviation added.

<<PCB 180<<PCB 101<PCB 138<PCB 153 (0.00<0.12<<1.10<<2.11<2.20<2.94 µg/kg, respectively), with significant differences between the first two and the third and subsequent congeners, as well as between the third and the others (Table 6, Figure 5). The treatments followed a similar order: T6< T7<<T8 (5.47<7.27<<12.71 µg/kg, respectively), considering the average content in tubers and leaves, with the highest levels found in the untreated control (Table 6, Figure 6). The lowest PCB content was observed in the treatment with the lower dose (3 t/ha) of HUMAC Enviro, compared to the treatment with double that dose.

The soil or substrate 2 used in experiment B was highly contaminated, as indicated by the aged contamination levels at T8 (Table 2). At Substrate 2, the sum of PCBs was detected at 0.162 mg/kg, while the contents of indicative congeners ranged from 0.008 to 0.043 mg/kg, with PCB 138 reaching the highest level of 0.043 mg/kg. Similar to trial A, the humic preparations of HUMAC Enviro applied to the soil in either a simple or double dose reduced the lev-

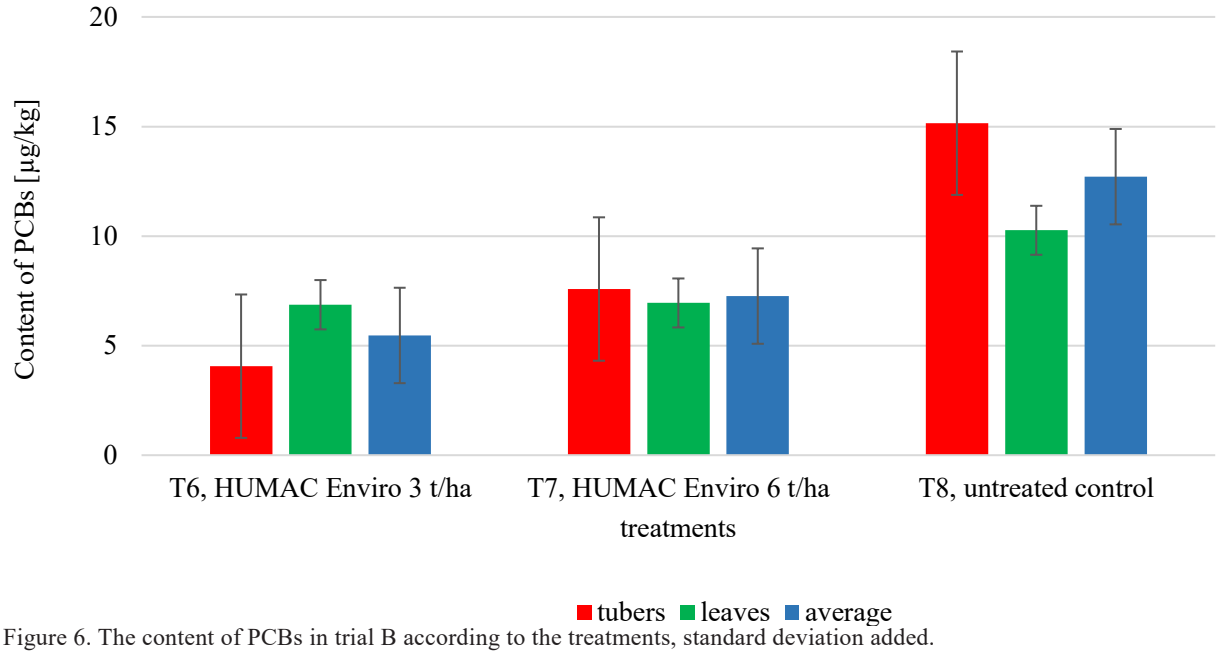


Figure 6. The content of PCBs in trial B according to the treatments, standard deviation added.

els of indicative PCB congeners in the tubers and leaves of the model crop. Compared to the untreated control, the simple dose had a greater effect than the double dose, decreasing PCB content by 73.20% versus 60.47% in tubers, and by 23.07% versus 22.17% in leaves, respectively. The bioconcentration factor decreased in the order of HE/simple dose < HE/double dose, with values of 0.067 and 0.090, respectively, for T6 and T7, while on the untreated control (T8), it was 0.157.

DISCUSSION

According to the Slovakian National Law "On the protection and use of agricultural land" and "On integrated prevention and control of environmental pollution" Act No. 220/2004 (Codex) and 245/2003 (Codex) respectively, and their subsequent amendments, the limit content of PCBs is set at 0.04 mg/kg for agricultural soil and 0.05 mg/kg in agricultural non-used soil, considering the sum of the six indicative congeners. In agricultural soil near the pollution source, the facility of the former PCB producer Chemko Strážske, related works document the local nature of the pollution, as most samples taken from the affected area show PCB levels below the specified limit. The trace content of PCBs in the soil complies with the mentioned law standard, making it possible to carry out agricultural activities.

According to the mentioned limit of 0.04 mg/kg, the content of 0.162 mg/kg recorded as the initial status concerning Substrate 2 exceeds the limit by 4.1 times. The content of 0.022 mg/kg, recorded as aged contamination and final status concerning Substrate 1, indicates that it was well below the limit. The value of 17.936 mg/kg recorded at the bottom sediment signifies that the sediment contained 359.7 times more PCBs than the limit of 0.05 mg/kg, indicating it was extremely highly contaminated. All these values are consistent with previous findings regarding the case of Strážske (Danielovič *et al.* 2003, 2014); however, the content of 0.022 mg/kg detected in soil from an uncontaminated area is unexpected and relatively high.

In a recent study based on passive air sampling across Europe, analysing spatial and temporal trends of persistent organic pollutants including PCBs, only 12 of the 32 sites had long-term PCB data, one of which was in Slovakia (White *et al.* 2023). Although that site is located relatively close to the pollution source, it is situated in a control region in the north-east direction, away from contamination. Significant attention has been directed towards this environmental issue worldwide, especially concerning soil in countries that historically produced

PCBs. Consequently, PCB monitoring was carried out in contaminated areas of the USA, Great Britain, Canada, Japan, and Sweden towards the end of the last century. Soil data indicate generally high levels of pollution, with recorded PCB contents of 0.567, 0.400, 0.200, 0.1000, and 0.332 mg/kg, respectively (Webber & Wang 1995; Kannan *et al.* 1997; Backe *et al.* 2004; Wilcke *et al.* 2006). Lower PCB levels were observed in Italy, specifically in the Piedmont region, where soil used for agriculture contained an average of 0.002 mg/kg, with values ranging from 0.0001 to 0.0160 mg/kg (Fabietti *et al.* 2010). However, Italian sources regarding PCB levels in bottom sediment of waste channels report higher concentrations, comparable to those mentioned in the paper (Sinche *et al.* 2018; Terzaghi *et al.* 2019). Slovakian data from the Strážske region, collected a decade ago, ranged from 0.005 to 0.009 mg/kg, indicating that PCBs persist in soil even 30 years after production ceased (Danielovič *et al.* 2014). These findings align with the data presented in the paper.

Similar to Slovakia, the need to address this issue ergonomically exists in many countries, even decades after production ceased (Webber & Wang 1995; Kannan *et al.* 1997; Backe *et al.* 2004; Fabietti *et al.* 2010; Cocarta *et al.* 2016; Kukharchyk & Kozyrenko 2018; Sinche *et al.* 2018; Sadaňoski *et al.* 2020; Ouyang *et al.* 2021; Lü *et al.* 2024; Shi *et al.* 2024; Subramani *et al.* 2024). Due to limited mobility, low water solubility, and high persistence of PCBs, soil is among the greatest reservoirs of these contaminants in the environment. Unfortunately, this causes accumulation, which now sits at the start of the food chain (Fernández-Sánchez *et al.* 2001; Amakura *et al.* 2005; Javorská *et al.* 2005, 2006; Mikes *et al.* 2009; Cervantes *et al.* 2011; Umlauf *et al.* 2011). It is thus strategically important to target this environmental burden directly in the soil, through decontamination or immobilisation (Bokvajova *et al.* 1994; Freidig *et al.* 1998; Pehkonen *et al.* 2010; Nybom *et al.* 2015; Guo *et al.* 2018; Holatko *et al.* 2022; Symanowicz & Toczko 2023). Such complex soil sanitation procedures have yet to be developed, although various elements that could be used are known, studied, or available. Most related research focuses on both soil-applied humic preparations and PCBs, usually in laboratory conditions, examining chemical aspects such as bonding and dynamics (Pignatello & Xing 1996; Eretsky 2000; Zhu *et al.* 2005; Birdwell *et al.* 2007; Trimble *et al.* 2008; Cervantes *et al.* 2013; Wei *et al.* 2015). These studies suggest that most PCBs sorption is spontaneous and exothermic, involving various types of chemical bonds, with humic acids showing high sorption affinity. However, from a practical agronomic perspective, these studies are somewhat limited, as open-field trials are influenced by numerous factors. Referring to the results of Wei *et al.* (2015), who observed a free energy of -6.01 kJ/mol at 15°C, it is likely that weather conditions − possibly during August and September − were favourable during the *ex situ* trial (see section 2.5 regarding weather), but they are unlikely to be the sole decisive factor. The existing literature mainly addresses the environmental fate and properties of PCBs, with little extensive *in situ* research on the immobilisation and decontamination effects of different activated soil-applied humic preparations (Di Guardo *et al.* 2017, 2020; Terzaghi *et al.* 2019, 2021). Similarly, this environmental aspect remains underexplored in agronomic, phytotechnical studies primarily focused on yield effects or technological parameters related to soil-applied humic preparations (Tóth *et al.* 2015; Wilcsewski *et al.* 2018; Holatko *et al.* 2022; Tóth & Dupľák 2023).

Among the research mentioned, a few more closely related studies also address the topic. Amakura *et al.* (2005) found that when evaluating the quality of baby food in Japan, a type of Japanese radish had the second highest rate of toxic equivalent quantity, which confirms the suitability of this rare model crop for short-term vegetation screening. According to a study by Mikes *et al.* (2009), where the uptake of PCBs into radish tubers and leaves was monitored at a heavily contaminated field site, uptake from the air was even more significant than from the soil. The shoot value of the bioconcentration factor was quite consistent, ranging between 0.01 and 0.22 during the *in situ* trial, compared with ranges of 0.048 to 0.090 under humic treatments and 0.055 to 0.473 at untreated controls within both *ex situ* trials (AB). In contrast to *in situ* conditions, *ex situ* screening can assume a lower intake from the air since the experiment was not conducted in a heavily contaminated area. The results of Sinche *et al.* (2018) showed that the resorption of PCBs was

slower with 6% amendments compared to 3%, most notably in humic acid treatments. However, the results related to a stronger effect of the simple dose compared to the double dose in trial B may align with the expected physiological impact of the humic preparations. Due to the stronger physiological effect of humic acids at their double dose, which probably leads to higher PCB intake with a more developed root system, there is likely a higher contaminant content in the model crop (Olaetxea *et al.* 2018). The findings from this short-term vegetation screening are environmentally favourable, and it is desirable to examine them in further detail.

CONCLUSIONS

The effect of variously activated soil-applied humic preparations from the HUMAC® series Agro – Enviro – Sorbent was examined for their ability to immobilise PCBs during short-term *ex situ* vegetation screening. The content of PCBs, following six indicative congeners, decreased in the sown radish tubers and leaves compared to the untreated control. At the same time, the amount of PCBs migrating from the soil into the soil leachate also diminished. However, a greater effect was observed with the lower dose of humic treatment compared to the higher dose, which is particularly important when considering the agro-ecological design of the subsequent *in situ* semi-operational experiment.

The environmentally advantageous nature of these initial results suggests the need to establish an agronomically designed pilot experiment. This should be conducted on arable soil at a polluted site near the source of pollution, with a clear goal of developing a phytotechnically manageable immobilisation process or even decontamination of PCBs. To achieve the most environmentally beneficial impact and ensure the high persistence of the immobilisation effect on PCBs, the *in situ* experiment must focus on several key factors. It is necessary to treat the entire topsoil layer and reach at least an equilibrium between the opposing effects of soil-applied humic preparations. Similarly, it will be essential to address how to eliminate or reduce the influence of numerous differing agro-ecological and technological factors. Ultimately, it is important to verify the durability of the environmentally positive impacts and to explore soil treatments based on different modes of action to maximise synergies.

Acknowledgement: This research was funded by a series of projects in the assignment of the Ministry of Agriculture and Rural Development of the Slovak Republic: (i) ÚOP no. 79/2020 „Verification of the immobilization effect of differentially activated humic acids on PCB substances in *ex situ* conditions, for subsequent option of *in situ* solution", (ii-iv) ÚOP nos. 77/2021 – 61/2022 – 61/2023 "Conduction- / Continuity- / Termination of *in situ* semi-operational experiment to design phyto-technical technology for immobilization / decontamination of PCB substances at agriculturally used land".

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Received: October 24, 2025
Accepted: March 17, 2026