FAKULTA MECHATRONIKY, INFORMATIKY A MEZIOBOROVÝCH STUDIÍ <u>TUL</u>

BIODIVERSITY IN WETLAND+ SYSTEM: A PASSIVE SOLUTION FOR HCH DUMP EFFLUENTS

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ABSTRAKT

Even though use of the hexachlorocyclohexane isomers (HCH) has been prohibited for decades, their presence in the environment is still reported worldwide. Wetland+[®] is a registered trademark of the remedial treatment technology consisting an aerobic sedimentary tank, a permeable reactive barrier (PRB), a biosorption system and an aerobic wetland. The average removal efficiency of the system was 96.8 % for chlorobenzenes (ClB) and 81.7 % for HCH, during the first twelve months of operation. This study evaluates biodegradation and diatom biodiversity as a measure of water quality in the system. The presence of the genes encoding enzymes involved in the degradation of the HCH compounds indicates that removal of HCH and ClB occured not only by chemical way but also through combined aerobic and anaerobic biodegradation. Significantly higher number of diatom species was detected in the Wetland+[®] effluent recipient - the Ostrovský Creek after one-year operation of the water quality .

ÚVOD



Hexachlorocyclohexane (HCH) isomers (α , β , γ , δ , ϵ) and their transformation products, such as chlorobenzenes (ClB), cause serious and persistent environmental problems in many places around the Alternatively, we have world. designed a robust, low-maintenance and sustainable treatment system that is cheaper than conventional WWTPs and can be implemented in remote locations where access to infrastructure is limited. The main objective of this study is to report on the initial performance of a newly constructed treatment system before and during the first growing season.

VÝSLEDKY A DISKUZE

Table 1. Average removal efficiency of individual modules and of the whole Wetland+® treatment system in the period September 20021 – September 2022

Module	∑нсн	∑CIB
А	3.9%	34.3%
В	45.0%	94.7%
С	21.2%	53.1%
D	37.3%	74.0%
Whole	81.7%	96.8%
system		

In total, 5.9 kg of HCH and 26.0 kg of ClB were eliminated from the drainage water within the 12-month operation of the system. Individual HCH isomers were not removed evenly but followed the pattern: $\alpha = \gamma = \delta > \varepsilon = \beta$. As a result, while δ -HCH dominated at the inflow to the system covering more than 60% of the total HCH concentration, ε -HCH prevailed at the outflow from the system.

Relative abundance of *Dehalococcoides* spp. and genes indicating total bacterial biomass (16S rDNA), dehydrochlorinase (*linA*), haloalkane dehalogenase (*linB*, *linB-RT*), and reductive dechlorinase (*linD*) in individual modules of Wetland+®. The color scale indicates the relative quantity of a given marker: red (+++) highest, orange (++) high, yellow (+) intermediate quantity, (+-) low quantity, ND = not detected or below the LOQ.

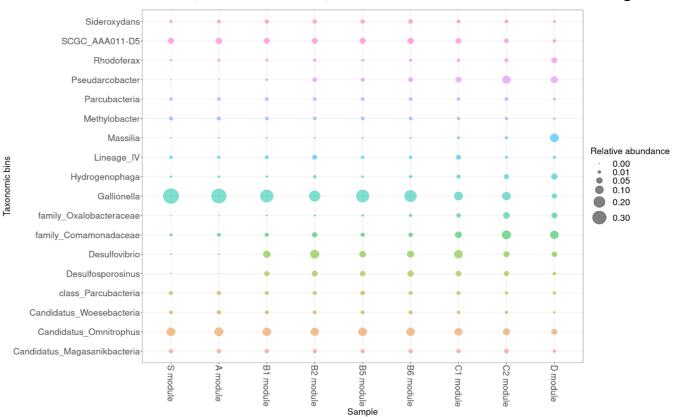
	Total bacterial biomass	Dehalorespiring bacteria	Lindane-degrading bacteria				
	16S rDNA	Dehalococcoides spp.	dehydrochlorinase	haloalkane dehalogenase	haloalkane dehalogenase	reductive dechlorinase	[mg/ 1]
	U16SRT	DHC	linA	linB	linB-RT	linD	
S (inlet)	++	+++	+	+	+	+	6.69
А	+	+++	+	+	+	+	9.68
В	++	+++	+++	+	++	++	9.71
С	++	++	+++	++	++	++	11.0 2
D	+++	+	+++	+++	+++	+++	12.8 7

The functional genes linA, linB, linB-RT, and linD encoding enzymes involved in aerobic HCH biodegradation were detected in all samples (Table 3). The highest level of the U16SRT gene, representing the total microbial biomass, was detected in the outlet from the aerobic wetland (module D). The relative abundance of the linA, linB-RT, and linD genes was the highest in the outlet. The quantity of the lin genes in the samples from modules S, A and B was similarly low, but in samples from module C

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This study took place in Hájko, Czech Republic (50°17'31.5" N 12°53'35.2" E). The location of Hájek is located in western Bohemia near Karlovy Vary. Since 1960, approx. 3-5 thousand have been deposited on the dump of the Hájek quarry. tons of residual ballast isomers HCH and ClB. The Wetland+® system was installed in September 2021. It consists of the following parts connected in series: aeration and sedimentation module (A), permeable reactive barrier (B), biosorption module (C) and aerobic wetland system (D). Water samples were taken at the outlet of individual Wetland+® modules and analyzed for physical-chemical parameters, molecular-biological parameters, diatom analysis and chemical analysis. Chemical analysis included the use of GC-MS arrays to determine the concentration of HCH isomers and their by-products, while molecular biology analysis included DNA extraction and qPCR reactions to assess microbial abundance and diversity. Finally, the efficiency of the Wetland+® treatment system for HCH and ClB removal was calculated based on the inflow and outflow concentrations per season.

were lin genes elevated. The highest relative abundance of Dehalococcoides spp. (DHC), was found in the inflow water (profile S) and in modules A and B. In direction of water flow (modules C and D), the abundance of DHC was decreasing.



The microbial profile demonstrates the high abundance bacteria belonging to genus *Galionella* and *Canditatus Omnitrophus* but with decreasing trend to the end of the wetland system. The D module were rich on bacteria of genera *Massilia, Rhodoferax, Hydrogenophaga, Flavobacterium, Caulobacter* and *Bacteriovorax* comparing to another modules. The abundance bacteria of the sulfate-reducing genera *Desulfovibrio* and *Desulfosporosinus* was found is become higher elevated in all modules profiles except for the inlet water S and A module. The abundance of the family Oxalobacteraceae, family Comamonadaceae and *Pseudarcobacter* increased in modules C1, C2 and D comparing to other.

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