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Occurrence of Pharmaceuticals in Wastewater and Their Interaction with Shallow Aquifers: A Case Study of Horní Beřkovice, Czech Republic

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Abstract: The application of innovative technologies in water management, such as wastewater reuse, requires a deeper understanding of emerging pollutants, including pharmaceuticals. This study presents a unique pilot site at Horní Beřkovice in Central Bohemia, where wastewater parameters are significantly influenced by the effluent from a local psychiatric hospital, and where the treated wastewater infiltrates into a shallow aquifer over a long period. The survey compared the quality parameters of local wastewater with those of the wastewater in four other catchments with no sources of concentrated pharmaceutical contamination. A total of 10 pharmaceuticals were detected while monitoring a common sewage system, but their number increased 3-fold at Horní Beřkovice. The water quality data revealed the effectiveness of the removal of pharmaceuticals from wastewater at the local sewage treatment plant and tracked the fate of substances that move from the treatment plant into the recharge ponds and then gradually into groundwater. The findings showed a significant decrease in all the monitored micropollutants that remained bound in sediments and in the unsaturated zone. Their passage into groundwater was highly reduced, and they virtually disappear after a few hundred meters in the saturated zone. The only exception is carbamazepine. This substance passes through the treatment technology and unsaturated zone. It systematically appears in the groundwater samples collected about 1 km from the infiltration site.

Keywords: micropollutants; pharmaceuticals; wastewater; groundwater; infiltration

1. Introduction

The issue of the so-called micropollutants in surface water and, subsequently, in groundwater receives considerable attention from the professional community. These substances are, in most cases, not listed in current legislation that specifies the requirements for drinking water, because until relatively recently analytical methods capable of detecting them with reasonable accuracy did not exist. Nevertheless, the development of laboratory methods and procedures and their feasible application leads to a wide range of new questions and challenges. An extremely varied range of substances is present in the hydrosphere. In most cases, their impact on the ecosystem and, consequently, on human health is unknown.

Pharmaceuticals (PPCP—pharmaceuticals and personal care products), which are usually present in water in very low concentrations ranging from ng/L to µg/L, are a crucial group of micropollutants. Their assorted composition encompasses a wide variety of medicines that leave the human body and pass into the sewer. The situation is compounded further by the formation of highly variable decay products of primary contaminants. Hormones, contraceptives, and even narcotics are found in the water [1] as well. Livestock and fish farms, where various products are commonly used to treat and prevent diseases or to accelerate weight gain in animals, are another source of pollution in addition to the healthcare sector. It turns out that these substances are widespread in almost all industrialized or developed countries [2]. The highest concentrations logically occur around large urban agglomerations and medical facilities such as nursing homes for seniors or hospices. The best understanding regarding the behavior of pharmaceuticals in surface water and their impact on the environment was documented by Costanzo et al. [3], Peake and Braund [4], Jobling et al. [5], and Deo and Halden [6]. Petrie et al. [7] and Vymazal et al. [8,9] dealt with the occurrence of pharmaceuticals in wastewater treatment plants and in constructed wetlands. Their results indicate that the removal efficiency of pharmaceuticals varies greatly among different wastewater treatment systems and among the substances studied.

Far less information is available on pharmaceuticals in groundwater. The assumption that these particular contaminants are effectively eliminated in the soil profile and by the sorption processes in the unsaturated zone was disproved by the first hydrogeological studies [10]. For example, concentrations of anthropogenic hormones, diclofenac, and carbamazepine, ranging between 1.4 and 6.5 ng/L [11], up to 46 ng/L [12], and up to 610 ng/L [13], respectively, were identified in the groundwater of aquifers fed by treated wastewater in urban agglomerations in the USA. Banzhaf et al. [14] studied surface water and groundwater interaction and classified selected pharmaceuticals as mobile and sorbing/degradable. According to some studies, the natural attenuation of pharmaceuticals is greatly affected by temperature, redox potential, mineralogy, and sorption ability [15]. Laboratory experiments indicated that sorption is the main attenuation process for pharmaceuticals rather than degradation [16]. Swartz et al. [17] found a relationship between lower pharmaceutical concentrations and higher oxygen concentration in the rock environment, and they also provided evidence of less intensive attenuation of drugs under reducing conditions. According to Yamamoto [18], the sorption coefficients are generally higher for amines and lower for carboxylic acids and neutral pharmaceuticals. Organic carbon and iron oxides are the major sorbents in sediments. Roberts et al. [19] published their results of sorption modeling for selected pharmaceuticals and calculations of the sorption parameters. The results of some studies carried out in the USA [13] showed that organic pollutants such as caffeine, gemfibrozile, and many analgesics were removed from groundwater 6 months after the infiltration of wastewater. On the other hand, some drugs such as carbamazepine and primidone remained in the environment for up to 8 years.

The risk posed by pharmaceutical contamination of water is not clearly defined. For example, Kostich et al. [20] showed that the disruption of the endocrine system of fish occurred due to trace amounts of the contraceptive ethinylestradiol in a stream. The effect of diclofenac on histological changes in the kidneys and gills of rainbow trout [21] was observed in Germany. Some pharmaceuticals were found to be very stable in the environment and, therefore, dangerous because of bioaccumulation, and they may pose a risk to terrestrial animals. A typical example is diclofenac residues, which caused a disruption in the ecosystem in many places in India and Pakistan [22]. Another group of pharmaceuticals abundant in wastewater is antibiotics [3,23,24]. These substances may greatly affect the resistance of bacteria and, consequently, pose a human health risk. A study from China [25] detected several pharmaceuticals in tap water and attempted to assess the human health risk of selected substances based on the U.S. Environmental Protection Agency's (EPA's) Exposure Factors Handbook.

The occurrence of pharmaceuticals in drinking water sources were addressed by Furlong et al. [26]. Their study detected 118 different pharmaceuticals in drinking water samples from 25 treatment plants in the USA. Another study [27] found a positive correlation between pharmaceutical concentrations in groundwater sources and urban land use. The findings of Morteani [28] confirmed the presence of anthropogenic estrogen concentrations exceeding 2 ng/L in the Želivka Reservoir, which is the main source of drinking water for the Czech capital Prague. For this reason, among others, another team of researchers performed broadscale monitoring of selected pharmaceuticals (naproxen, ibuprofen, diclofenac, carbamazepine, and 17α-ethynylestradiol) in Czech water supply systems, which did not confirm any contamination of the drinking water [29]. The probable reason for this is that most of the raw surface water that serves as a source for drinking water came from protected water reservoirs situated on the upper reaches of rivers. However, some substances such as ibuprofen and carbamazepine in particular occur in Czech water supplies. This is due to the low efficiency of existing treatment technologies for carbamazepine and due to the high consumption of ibuprofen in the Czech Republic, which according to the State Institute for Drug Control was about 15.6 g/person/year in 2007 and more than 3 times higher when compared, for example, to Germany [30].

The purpose of this study is to document and evaluate the occurrence of pharmaceuticals at a specific pilot site located at Horní Beřkovice in the Czech Republic. The findings are based on data from a complex monitoring system, which records pharmaceutical concentrations at the source of the contamination, throughout the wastewater treatment process, and in the affected environment. The study also aims to compare the efficiency of wastewater treatment plants at different localities and with different technologies.

2. Materials and Methods

2.1. Characteristics of the Pilot Locality at Horní Beřkovice

The issue of the occurrence of pharmaceuticals in groundwater was studied for the first time in connection with the infiltration of treated wastewater into the ground in the village of Horní Beřkovice in Central Bohemia (Figure 1). The village currently has 938 permanent residents and there is no industrial production or other sources of pollution with the exception of local farming. A psychiatric hospital with 587 beds has been operating in the village since 1891. Operating at full capacity, the hospital produces about one-third of the local wastewater. Various pharmaceuticals that are systematically administered to the patients of the psychiatric hospital pass through the digestive tract and are excreted into the sewage. The municipal sewer system is connected to a wastewater treatment plant from where the treated water drains into recharge ponds after about 800 m. The current Czech legislation permits the infiltration of treated wastewater into the ground only in exceptional circumstances, specifically in cases where natural conditions do not allow wastewater to be discharged into surface streams. There are no streams in the wider surroundings of Horní Beřkovice, so that the village was granted a permit by the water authority to drain the treated water directly into the ground.

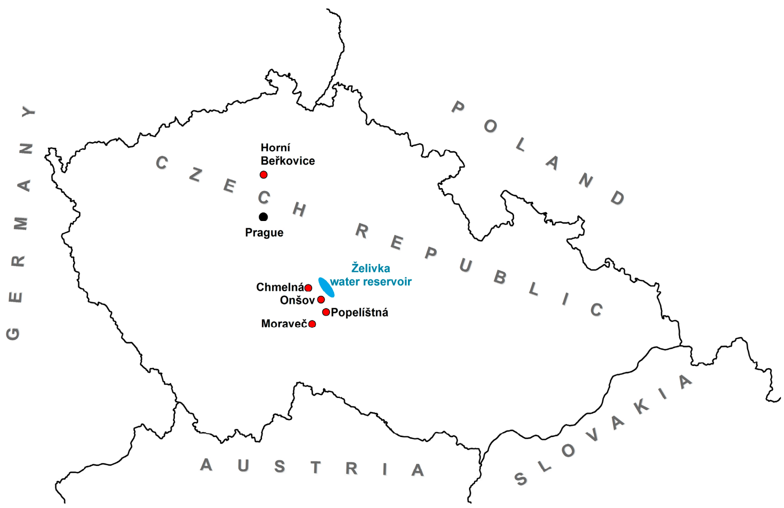

Figure 1. Location of pilot sites.

Systematic monitoring of the groundwater quality below the local sewage treatment plant was carried out at Horní Beřkovice in 2007–2009. At that time, however, it focused only on the basic chemistry of the water, concentrations of trace metals, and organic pollution. As late as 2012, the study was extended to monitor four pharmaceuticals [31] as well. The present study is an output of project AQUARIUS (see Acknowledgements), which was launched in 2015. Since then, the monitoring in Horní Beřkovice was expanded to 32 pharmaceuticals. For the sampling conducted in January 2016, the scope of the analysis was expanded to include an additional 12 pharmaceuticals and their intermediate products. Samples were collected at regular quarterly periods.

2.2. An Overview of the Natural Conditions at the Locality of Horní Beřkovice

The infiltration occurs on flat cultivated land in a region with an average annual temperature of +8.5 °C and annual rainfall of 500–550 mm. The immediate bedrock of the recharge ponds consists of a brown earth soil profile comprising very permeable Quaternary fluvial gravels and sands. Loess and loess loams a few meters thick occur on the slopes of a gentle morphological depression. The filtration coefficient of the Quaternary fluvial aquifer corresponds to 1.3×10^{-5} to 1.3×10^{-3} m/s, while the coefficient of transmissivity equals 2.7×10^{-4} to 5.2×10^{-3} m^2/s [32]. Sandy marls and calcareous siltstones of the Jizera Formation of Middle Turonian age occupy deeper parts, of which only the first few meters containing products of weathering are more permeable. More or less impervious Turonian sediments occur at greater depths. Therefore, the infiltrated water moves only in the shallow aquifer toward the east, toward the drainage basin formed by the Elbe River.

2.3. Monitoring System at the Horní Beřkovice Locality

The monitoring was intended to identify and document the migration path leading from the source of the pharmaceutical contamination, which is the psychiatric hospital at Horní Beřkovice, all the way to the potential consumers of the contaminated water, who are the residents of the village of Daminěves located along the path of groundwater flow (Figure 2, Table 1).

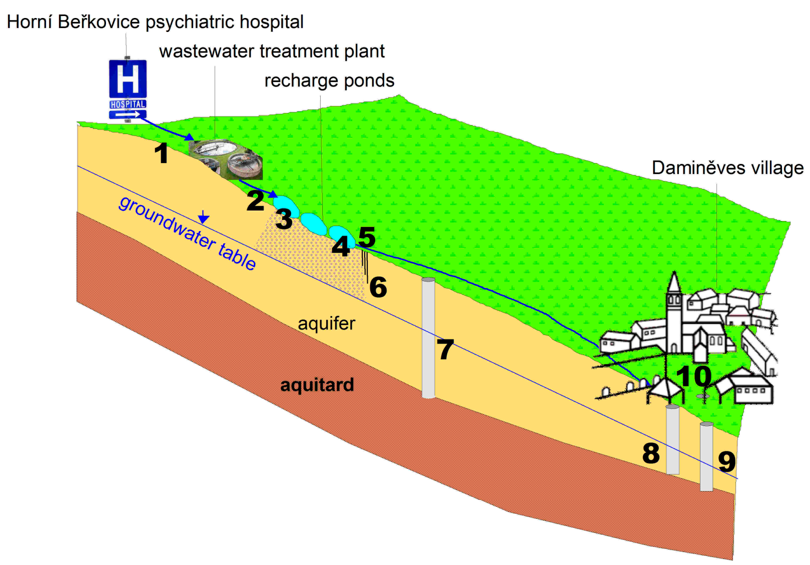

Figure 2. Horní Beřkovice monitoring scheme.

Table 1. Characteristics of the individual monitoring sites.

Sampling Site	Characteristics of the Monitoring Site
1	The inflow into the wastewater treatment plant—a mixture of pharmaceuticals in the wastewater from the village and in the wastewater from the psychiatric hospital
2	The outflow from the wastewater treatment plant (the difference between the results of the monitoring at points 1 and 2 indicates the efficiency of purification)
3	Sediments deposited in the first recharge pond
4	Sediments deposited in the third recharge pond
5	Outflow from the third recharge pond
6	Zonal sampling of soil from the unsaturated zone
7	Monitoring borehole below the third (last) recharge pond. This section provides information on the behavior of drugs after passing through the unsaturated zone and then through about 100 m in a saturated environment.
8–10	Wells in the village of Daminěves determine the concentrations of pharmaceuticals after passing through about 1 km of the aquifer. The pharmaceuticals that are detected in this area passed through various degradation processes, dilution, sorption, etc.

Wastewater and surface water samples were collected directly with amber glass vials, and sediments and soils in polyethylene boxes. The zonal samples of soils in the unsaturated zone were taken from a borehole core with a diameter of 50 mm. The AQUARIUS project included several other pilot catchments that focused on the monitoring and study of nutrients and related substances. Four of them—Chmelná, Onšov, Popelištná, and Moraveč—drain into the Želivka water reservoir (Figure 1). Wastewater from settlements located in the catchment areas are treated in local constructed wetlands. The sampling was carried out at quarterly intervals in 2014–2015, above and below the constructed wetlands, and the selection of the analyzed substances and sampling and analytical methods corresponded to those used at Horní Beřkovice.

The results obtained in these catchments within the scope of the AQUARIUS project were used as reference data, which define the background values or common pharmaceutical concentrations in the wastewater in the Czech Republic.

Furthermore, some analyses were compared with pharmaceutical concentrations in the influent and effluent of four mechanical–biological treatment plants operating in large Czech cities (data from 2009 to 2011). In agreement with the operators, the data in Table 2 are listed anonymously as wastewater treatment plant (WWTP) A–D.

2.4. Characteristics of the Wastewater Treatment Plants

As mentioned above, this study presents the findings from several localities with different types of wastewater treatment facilities. The facility in Horní Beřkovice (Figures 1 and 2) is a conventional mechanical–biological plant. The main aim of the mechanical stage is grit removal. The biological treatment comprises a denitrification stage, nitrification stage, and settling tanks. The required efficiency is achieved by low F/M loading (“food to mass” ratio) of the sludge and high sludge age. The process does not include chemical precipitation of phosphorus.

The wastewater treatment plants WWTP A–D (Table 2) that were compared are larger facilities constructed for tens to hundreds of thousands of inhabitants, but the process of wastewater treatment is basically the same as in Horní Beřkovice, with the exception of a unit for the chemical precipitation of phosphorus.

The last type of wastewater treatment plant presented is the constructed wetlands in the Želivka reservoir watershed: Chmelná, Onšov, Popelištná a Moraveč (Figure 1, Table 3). They are designed to treat sewage from municipalities with 60–200 inhabitants. Such facilities comprise a grit removal pit at the point of inflow and artificial wetlands with horizontal flow and permanent saturation of the filtration bed, where predominantly anaerobic treatment of the water occurs.

Table 2. Average value from four large sewage treatment plants (A–D) in Bohemia in 2009–2011.

Substance	WWTP A			WWTP B			WWTP C			WWTP D		
	Input	Output	Cleaning Efficiency	Input	Output	Cleaning Efficiency	Input	Output	Cleaning Efficiency	Input	Output	Cleaning Efficiency
	ng/L		%	ng/L		%	ng/L		%	ng/L		%
Ibuprofen	21,600	380	98	8700	930	89	7700	125	98	18,500	180	99
Diclofenac	400	910	−128	600	860	−43	420	500	−19	870	890	−2
Carbamazepine	604	924	−53	260	330	−27	350	420	−20	1230	1325	−8

Note: mutually comparable values are marked by same shades of grey.

Table 3. Average concentrations of individual pharmaceuticals at the inflow to and outflow from monitored constructed wetlands (CW) and wastewater treatment plants (WWTP) in 2014–2015.

Substance	Data from the Želivka Reservoir Catchments															Horní Beřkovice WWTP		
	Chmelná CW			Onšov CW			Popelištná CW			Moraveč CW			CW average					
	Input	Output	Cleaning Efficiency	Input	Output	Cleaning Efficiency	Input	Output	Cleaning Efficiency	Input	Output	Cleaning Efficiency	Input	Output	Cleaning Efficiency	Input	Output	Cleaning Efficiency
	ng/L		%	ng/L		%	ng/L		%	ng/L		%	ng/L		%	ng/L		%
Furosemide	13,000	3700	72	71,500	11,000	85	1700	2160	0	16,900	3500	79	25,775	5090	59	1745	144	92
Paracetamol	12,100	34	100	45,500	9800	78	239	305	0	8450	11,670	0	16,572	5452	45	39,225	25	100
Caffeine	7800	<10	100	17,000	5250	69	10,750	2000	81	26,500	10,750	59	15,513	4500	77	185,000	38	100
Saccharin	11,500	4900	57	11,350	5200	54	940	480	49	6350	11,000	0	7535	5395	40	43,250	60	100
Ibuprofen	9900	5250	47	2550	1650	35	6550	5200	21	2950	12,950	0	5488	6263	26	37,250	42	100
Hydrochlorothiazide	5050	3200	37	400	1600	0	2000	2300	0	19,500	3900	80	6738	2750	29	1512	2550	0
Metoprolol	1720	176	90	341	164	52	664	487	27	1618	1745	0	1086	643	42	1980	29	99
Diclofenac	720	495	31	700	325	54	770	765	1	750	695	7	735	570	23	1237	500	60
Atenolol	350	55	84	1700	1020	40	455	240	47	<10	<10		626	329	43	643	<10	100
Warfarin	45	24	47	37	15	59	21	11	48	60	42	30	41	23	46	27	16	41
Gabapentin	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	14,050	1177	92
Carbamazepine	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	2850	2725	4
Clarithromycin	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	2035	84	96
Naproxen	<50	<50	-	<50	<50	-	<50	<50	-	<50	<50	-	<50	<50	-	1054	<50	100
Sulfamethoxazole	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	621	633	0
Ketoprofen	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	393	<10	100
Triclocarban	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	303	<10	100
Trimetoprim	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	250	3	99
Erythromycin	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	152	6	96
Iopromide	<50	<50	-	<50	<50	-	<50	<50	-	<50	<50	-	<50	<50	-	95	<50	100
Sulfamethazine	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	53	7	87
Sulfapyridine	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	57	534	0
Penicillin G	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	15	<10	100
Sulfamerazine	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	<10	<10	-	12	<10	100
Chloramphenicol	<20	<20	-	<20	<20	-	<20	<20	-	<20	<20	-	<20	<20	-	9	<20	100
Sulfanilamide	<50	<50	-	<50	<50	-	<50	<50	-	<50	<50	-	<50	<50	-	0	40	0

Note: mutually comparable values are marked by same shades of grey.

2.5. Analytical Methods

The analyses of the collected samples of wastewater, surface and groundwater, and mud and soil were carried out according to valid procedures and EPA method 1694 in the Vltava catchment laboratory.

Samples were collected in 60 mL amber glass vials (filled only halfway). The samples were stored in a freezer (in an inclined position). They were defrosted at a maximum temperature of 30 °C on the day of analysis. It was necessary to conduct the analysis immediately after defrosting.

Two methods were developed for the analysis of PPCPs—Method A (ESI+ mode) and Method B (ESI− mode). The samples of water were centrifuged in headspace vials for 5 min at about 3500 rpm. Subsequently, 1.50 g of each sample were weighed in a 2 mL vial on an analytical balance. Then, 1.5 μL of formic acid (Method A) or 1.5 μL of acetic acid (Method B) were added to each sample. An isotope dilution was performed in the next step. Deuterated internal standards of d10-carbamazepine, d6-sulfamethoxazole, d3-iopromide, and 13C2-erythromycin (Method A), or d3-ibuprofen, d4-diclofenac, d3-naproxen, d5-chloramphenicol, and d3-iopamidol (Method B) were used.

The extraction procedure for the sediment samples consisted of two steps. During the first step, 0.25 g of each sample was weighed in a vial and 1 mL of acetonitrile was added. The vial was then placed in an ultrasound pot for the extraction. Each sample was centrifuged after the extraction, and the extract was then separated in another vial. During the second step of the extraction, each sample was mixed with 1 mL of formic acid and sonicated and centrifuged as in the first step. The extracts from both steps were then poured together for further processing. Before chromatography, the extracts of the solid samples were filtered with a glass syringe through a 0.45 μm filter.

PPCPs were separated and detected by LC–MS/MS methods based on direct injection of the sample into a chromatograph. A 1200 ultra-high-performance liquid chromatograph (UHPLC) coupled with an Agilent 6410 Triple Quad Mass Spectrometer (MS/MS) of Agilent Technologies, Inc. (Santa Clara, CA, USA) were used in ESI+ or ESI− modes.

Method A (ESI+)—the separation was carried out on a Zorbax Eclipse XDB-C18 analytical column (100 mm × 4.6 mm, 3.5 μm particle size). The mobile phase consisted of methanol and water with 0.1% formic acid and 5 mM ammonium formate as the mobile phase additives. The flow rate was 0.25 $mL \cdot min^{-1}$. The injection volume was 0.50 mL.

Method B (ESI−)—the separation was carried out on a Zorbax Eclipse XDB-C18 analytical column (150 mm × 4.6 mm, 3.5 μm particle size). The mobile phase consisted of methanol and water with 0.05% acetic acid as the mobile phase additive. The flow rate was 0.25 $mL \cdot min^{-1}$. The injection volume was 1 mL.

The samples for nonsteroidal anti-inflammatory drugs were first acidified with acetic acid, filtered through a 0.45 μm filter paper and mixed with an internal standard solution. The internal standard solution was prepared from deuterated solids (98% purity) and water from a UHQ system. The solid-phase extraction (SPE) was performed on a high-performance liquid chromatography column.

The detection limit for each analyte is shown in Table 4.

Each series of samples were verified by calibration control and by maintaining a clean environment, equipment, and agents. The performance of the analytical system was ensured by blind and spiked samples. The chemicals used for the preparation of calibration solutions had a certified purity of 99%. They were prepared from neat analytes or from solutions with certified concentration.

Each fifth sample in a series was processed by the method of standard addition, which was used to control the effect of the matrix of the sample and to reset the actual recovery ratio of a specific analyte. The measuring instruments were under regular control, and measuring vessels were metrologically tested.

The chemicals used were supplied from renowned manufacturers in the EU and USA: Dr. Ehrenstorfer GmbH (Augsburg, Germany), LGC Ltd. (Teddington, Middlesex, UK), Honeywell International Inc. (Morris Plains, NJ, USA), HPC Standards GmbH (Cunnersdorf, Germany), Absolute

Standards Inc. (Hamden, CT, USA), CIL Inc. (Tewksbury, MA, USA), Analytika spol s.r.o. (Prague, Czech Republic).

Table 4. Detection limits for analyzed pharmaceuticals.

Substance	Detection Limit (ng/L)	Substance	Detection Limit (ng/L)
Carbamazepine	10	Chloramphenicol	20
Erythromycin	10	Bezafibrate	10
Sulfamethoxazole	10	Warfarin	10
Iopromide	50	Saccharin	50
Ibuprofen	20	Gabapentin	10
Diclofenac	20	Tramadol	10
Iopamidol	50	Paracetamol	10
Atenolol	10	Sulfanilamide	50
Caffeine	100	Clarithromycin	10
Ketoprofen	10	Roxithromycin	10
Metoprolol	10	Carbamazepine-10,11-epoxide	10
Penicillin G	10	Carbamazepine 10,11-dihydro-10-hydroxy	10
Sulfamerazine	10	Carbamazepine 10,11-dihydroxy	10
Sulfamethazine	10	Oxcarbazepine	10
Sulfapyridine	10	Ibuprofen-2-hydroxy	30
Trimetoprim	10	Ibuprofen-carboxy	20
Furosemide	50	Diclofenac-4′-hydroxy	20
Gemfibrozil	10	Naproxen-O-desmethyl	20
Hydrochlorothiazide	50	Venlafaxine	10
Naproxen	50	Sertraline	10
Triclocarban	10	Ranitidine	10
Triclosan	20	Iohexol	50

3. Results and Discussion

3.1. Comparison of the Data from Horní Beřkovice to the Common Values in the Czech Republic

The monitored and compared catchments of the Želivka Reservoir are very similar to that of Horní Beřkovice as far as land use and areal extent are concerned. The only difference is the absence of concentrated sources of pharmaceutical contamination such as hospitals or other medical facilities. The only sources of contamination in these catchments are represented by the wastewater from local villages, which contain only commonly used drugs.

In general, the comparison of the measured values and data clearly suggests that the pilot locality at Horní Beřkovice is a unique site where, in some cases, much higher concentrations of pharmaceuticals (even in comparison with the mechanical–biological treatment plants in large cities) were detected. Moreover, the uniqueness of this locality is supported by a much broader number of detected substances.

Table 3 shows that, of all the drugs monitored, only 10 pharmaceuticals were detected in the tributaries of the Želivka Reservoir. Their concentrations in wastewater prior to treatment in the constructed wetlands ranges from a few micrograms up to tens of micrograms per liter, and they include only commonly used drugs. The highest concentrations were observed in the case of furosemide, a diuretic used for heart failure. Other commonly used drugs that were detected are paracetamol, ibuprofen, caffeine, and saccharine. The lower part of Table 3, with levels mostly below 1 μg/L, includes the pain killer diclofenac; medications for high blood pressure such as hydrochlorothiazide, atenolol, and metoprolol; and warfarin, which is used to prevent thrombosis.

The average concentrations of individual pharmaceuticals in the monitored catchments usually have similar or comparable values, but this is not a general rule. This is reflected, for example, in the case of paracetamol, the concentrations of which differ in the catchments of Onšov and Popelištná by a few orders of magnitude. This is not the result, for example, of the greater number of inhabitants in the Onšov catchment area, because the opposite applies in the case of ibuprofen. On the other hand, the concentrations of this substance in the Popelištná catchment were found to be twice as high as in the

Onšov catchment. It seems that the concentrations of pharmaceuticals are highly variable depending, for instance, on the social and age composition of the local population. The unreliability of existing sampling methods may be another reason for the spatial and temporal variability. As discussed by Petrie et al. [7], the grab sampling method provides us with a sample for a specific point in time and may be significantly influenced by fluctuations in flow.

All substances analyzed and monitored in the tributaries of the Želivka Reservoir were also recorded in Horní Beřkovice. Some of them show comparable concentrations (atenolol, metoprolol, or warfarin). In most cases, however, the pharmaceutical concentrations in Horní Beřkovice exhibit values that are higher in the case of caffeine, twice as high in the case of paracetamol, and nearly 7 times higher in the case of ibuprofen. By contrast, the substances that are usually not used in the psychiatric hospital do not correspond to the concentrations which are common in the Želivka catchment (e.g., furosemide).

Nevertheless, Horní Beřkovice differs fundamentally because of the fact that a much larger number of substances were detected that would otherwise not occur in water. Striking examples are the very high concentrations of gabapentin and carbamazepine that are used to treat schizophrenia and epilepsy. An average concentration of 2850 ng/L was recorded in the wastewater from Horní Beřkovice in the case of carbamazepine, but this substance has not been detected at all in the Želivka catchment. For comparison purposes, various studies document concentrations of 1680 ng/L in the wastewater in Kälby in Sweden [33], 84–1300 ng/L in Minnesota in the USA [34], 637 ng/L in Australia [35], or 152–4596 ng/L in the UK [36]. Other substances that occur in high concentrations in Horní Beřkovice are clarithromycin (an antibiotic) and naproxen (a nonsteroidal anti-inflammatory drug). All other drugs detected in Horní Beřkovice occur occasionally in concentrations that usually do not exceed hundreds of ng/L.

The efficiency of the wastewater treatment plant in Horní Beřkovice is comparable to that of other large mechanical–biological wastewater treatment plants. Table 2 provides the results for three abundant pharmaceutical substances from four facilities. The input values in the inflow at Horní Beřkovice are very similar to those of the other sites. Diclofenac and carbamazepine are poorly degradable, while ibuprofen is removed very efficiently at all of the facilities studied. Compared to the efficiency of ibuprofen removal in the constructed wetlands (Table 3), the efficiency of the mechanical–biological wastewater treatment plants is much higher. The likely reason for this fact is that the wastewater in the outdoor tanks of the mechanical–biological wastewater treatment plants was exposed to sunlight and intensive aeration during the treatment process. As reported in literature [8], aerobic biodegradation and photodegradation are important processes of ibuprofen removal from wastewater, but they are very limited in constructed wetlands.

The last analyses of the samples from Horní Beřkovice were extended to a larger range of substances. The predictive ability of single results from each sampling point is obviously lower than the 2-year monitoring series. However, the first documented occurrence of pharmaceutical metabolites (Table 5) is of particular interest. Ibuprofen, diclofenac, and naproxen metabolites were efficiently removed in the wastewater treatment plants, while the carbamazepine metabolite carbamazepine-10,11-epoxide apparently passes through the treatment process unchanged.

Table 5. Single measure of metabolites and additional pharmaceuticals at the Horní Beřkovice site.

Metabolite	Horní Beřkovice Data		
	Input	Output	Cleaning Efficiency
	ng/L		%
Carbamazepine-10,11-epoxide	320	460	0
Carbamazepine 10,11-dihydro-10-hydroxy	<10	<10	-
Carbamazepine 10,11-dihydroxy	<10	<10	-
Oxcarbazepine	<10	<10	-
Ibuprofen-2-hydroxy	<30	<30	-

Table 5. *Cont.*

Metabolite	Horní Beřkovice Data		
	Input	Output	Cleaning Efficiency
	ng/L		%
Ibuprofen-carboxy	31,000	<20	100
Diclofenac-4′-hydroxy	140,000	<20	100
Naproxen-*O*-desmethyl	400	<20	100
Venlafaxine	210	<10	100
Sertraline	3100	1600	48
Ranitidine	580	220	62
Iohexol	430	19	96

Note: mutually comparable values are marked by same shades of grey.

3.2. Removal Efficiency of Pharmaceuticals during Wastewater Treatment

Table 3 shows that the efficiency in removing pharmaceuticals from wastewater varies considerably. Studies in the UK [7] documented the wastewater treatment plant efficiency for the most abundant pharmaceuticals, ranging from low (<50%) to high (>80%). Such variability is the result of the different physicochemical properties of substances and their susceptibility to biological attack. Kasprzyk-Hordern et al. [36] reported a removal efficiency of activated sludge wastewater treatment of over 85%, based on the average values for 55 studied substances. The wastewater treatment plant at Horní Beřkovice removed 95%–100% of the following drugs: paracetamol, caffeine, saccharin, ibuprofen, atenolol, metoprolol, naproxen, clarithromycin, ketoprofen, triclocarban, trimethoprim, erythromycin, iopromide, penicillin G, sulfamerazine, and chloramphenicol (determined as the primary substance).

Just the opposite applies to the next group of pharmaceuticals, which pass through the sewage treatment processes at the wastewater treatment plant in Horní Beřkovice virtually unchanged, or their concentration may even increase. They include the high blood pressure medication hydrochlorothiazide, the antibiotics sulfamethoxazole, sulfapyridine, sulfanilamide, and the antiepileptic carbamazepine, including its metabolite carbamazepine-10,11-epoxide. According to a study from Australia [24], the average removal efficiency of antibiotics in conventional (activated sludge) and advanced (microfiltration/reverse osmosis) wastewater treatment plants was 92%. Also, compared to our findings, the efficiency of 75% in the case of sulfamethoxazole in particular is surprisingly high.

However, data from a single facility should not be generalized. For example, when comparing the removal efficiency of monitored substances at different localities that are located along the tributaries to the Želivka Reservoir (Table 3) and that use similar treatment technologies (mechanical treatment, biological treatment in constructed wetlands), the obtained results often lead to quite different conclusions. This fact is best evidenced by the virtually zero removal efficiency of paracetamol at the localities of Popelištná and Moraveč, while the other constructed wetlands from the area are able to remove this substance with an efficiency approaching 100%. Compared to the monitored constructed wetlands, the treatment technology used at the wastewater treatment plant in Horní Beřkovice is much more effective in removing ibuprofen, saccharin, metoprolol, and atenolol.

3.3. The Behavior of Pharmaceuticals after Passing through Wastewater Treatment Plants

According to the abovementioned data, it is obvious that various concentrations of pharmaceuticals are released into the local environment. The following chapter describes the results of the monitoring of the drugs and characterizes their behavior at Horní Beřkovice. Table 3 summarizes individual substances and their average concentrations that the wastewater treatment plants discharge to three infiltration ponds. Their infiltration is limited by the composition and thickness of sediments that accumulated at the bottom of the ponds. However, when the volume of treated wastewater effluent from the sewage treatment plant is compared to the outflow from the last infiltration ponds in the winter with limited evaporation, it is evident that the ponds still maintain their infiltration function. The discharge of the wastewater treatment plant typically reaches 3–5 L/s, while the outflow from the last infiltration pond is only sporadic, usually after intensive rainfall.

Extremely high levels of some drugs were detected in the sediments of the infiltration ponds (Table 6). Considerably higher concentrations were determined in the first pond and lower concentrations in the third pond. In most cases (carbamazepine, sulfapyridine, diclofenac, triclocarban, triclosan, or hydrochlorothiazide), the concentrations in the sediments of the first pond are several times higher than those in the influent entering the wastewater treatment plant. This indicates that these drugs are sorbed onto the deposited sediments and may act as a secondary source of contamination. The occurrence of high caffeine concentrations in the sediments is enigmatic, because this substance is removed with a very high degree of efficiency in the wastewater treatment plant.

Table 6. Detected pharmaceutical concentrations in the mud of the infiltration ponds.

Sampling Site	**Carbamazepine**	**Ibuprofen**	**Diclofenac**	**Caffeine**	**Metoprolol**	**Sulfapyridine**	**Hydrochlorothiazide**	**Triclocarban**	**Triclosan**	**Tramadol**	**Clarithromycin**
	ng/kg										
1st infiltration pond	76,000	27,000	15,000	27,000	5850	2350	36,000	46,000	107,000	75,000	13,300
3rd infiltration pond	10,000	<10	<10	4700	8100	1500	<10	<10	<10	1500	<10

Soil samples from the unsaturated zone just behind the third infiltration pond were taken at intervals of 50 cm to a depth of 3 m (Table 7). Nine pharmaceuticals were detected in the soil. Four of them (hydrochlorothiazide, gabapentin, clarithromycin, and roxithromycin) were determined only at a depth of 1 m. By contrast, carbamazepine and caffeine appeared systematically in all samples down to a depth of 3 m. An interesting finding appears to be that quite significant roxithromycin concentrations occur at a depth of 1 m.

Table 7. Detected pharmaceutical concentrations in the unsaturated zone.

Depth (m)	**Carbamazepine**	**Caffeine**	**Metoprolol**	**Sulfapyridine**	**Hydrochlorothiazide**	**Gabapentin**	**Tramadol**	**Roxithromycin**	**Clarithromycin**
	ng/kg								
0.5	15,000	<10	8600	<10	<10	<10	<10	<10	<10
1.0	110,000	6700	47,000	5000	11,000	11,000	21,000	5700	5200
1.5	12,000	<10	10,000	2200	<10	<10	<10	<10	<10
2.0	16,000	5400	7900	2700	<10	<10	1700	<10	<10
2.5	12,000	<10	<10	<10	<10	<10	<10	<10	<10
3.0	6600	2300	<10	<10	<10	<10	<10	<10	<10

Attenuation processes and especially dilution in the aquifer apparently have a great influence on the reduction of pharmaceutical concentrations in groundwater. Analyses of water from the monitoring borehole drilled 100 m behind the last infiltration pond show that the vast majority of drugs is below the detection limit. The only exception worth noting is carbamazepine and its metabolite carbamazepine-10,11-epoxide, whose concentration does not change and more or less remains at the level observed in the wastewater entering the sewage treatment plant. The concentrations of other drugs that appear in the groundwater of the monitoring borehole—such as sulfamethoxazole, hydrochlorothiazide, gabapentin, tramadol, and sulfanilamide—are low.

The monitoring facilities in the village of Daminěves, located about 1 km from Horní Beřkovice, occasionally detected only slightly increased concentrations of caffeine and saccharin, which may be of local origin and which are most likely unrelated to the contamination of the treated wastewater at Horní Beřkovice. It is highly probable that carbamazepine is the only contaminant that comes from Horní Beřkovice. Carbamazepine concentrations reaching up to 890 ng/L systematically appeared in two of the three wells monitored at Daminěves. The metabolite carbamazepine-10,11-epoxide was found in all three wells in the village of Daminěves as well. Figure 3 illustrates the evolution of the average concentrations of carbamazepine in 2014–2015 and its variation along the entire profile from the contamination source to the village of Daminěves.

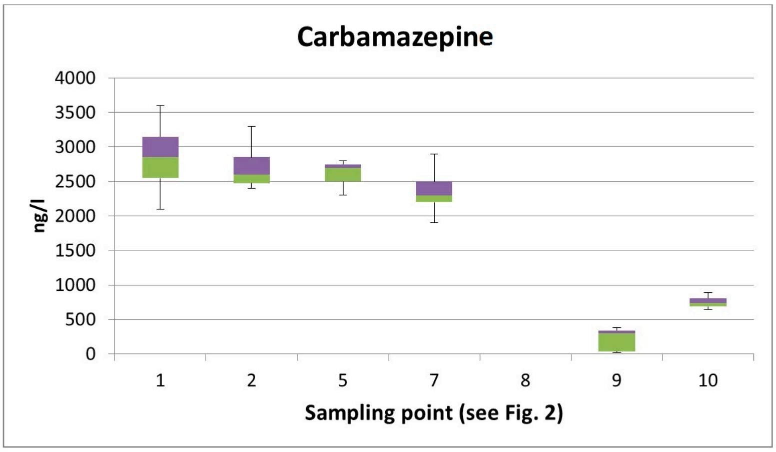

Figure 3. Box plots of carbamazepine concentrations monitored in 2014–2015 in the effluent from the wastewater treatment plant in Horní Beřkovice downstream to the village of Daminěves.

The persistence of carbamazepine in groundwater has been documented in literature as well. Drewes et al. [13] noted that the carbamazepine concentration in samples from boreholes downgradient from wastewater infiltrations ponds showed no significant reduction after travel times of approximately 2 years. Our study may confirm this finding. Based on approximate values of hydraulic parameters, we can estimate the travel time from the infiltration ponds in Horní Beřkovice to the wells in Daminěves (a distance of 900 m) to several years. The mean reduction in carbamazepine concentration from 2200 ng/L in the infiltration ponds to 753 ng/L in the groundwater in Daminěves may be interpreted as the result of dilution.

4. Conclusions

The findings showed that the current purification technologies are ineffective for a variety of pharmaceuticals. At Horní Beřkovice, these pharmaceuticals include mainly hydrochlorothiazide, gabapentin, carbamazepine, carbamazepine-10,11-epoxide, sulfamethoxazole, sulfapyridine, and sulfanilamide. However, despite using similar treatment technologies, the removal efficiency of some substances differs at various localities. The reason for this phenomenon may be dissimilar technological parameters of the wastewater treatment plants (residence time, wastewater composition, sludge age, etc.).

In the case of infiltration of treated wastewater through recharge ponds, large volumes of pharmaceuticals are sorbed onto the mud at the bottom of the ponds. However, this material may become a secondary source of contamination at an increased flow rate. The unsaturated zone and soil play an important role in the removal of pharmaceuticals during their infiltration.

The attenuation processes are effective, and all the observed pharmaceuticals fell below the detection limit after having passed through about 1 km of a relatively highly permeable sandy-marly aquifer. The only persistent problem is carbamazepine and its metabolite carbamazepine-10,11-epoxide. At the present time, the existing wastewater treatment procedures are not able to remove these substances. Concentrations of carbamazepine continue to reach values of up to 890 ng/L at the village of Daminěves even though large volumes of this contaminant are deposited in mud and sorbed in the unsaturated zone at Horní Beřkovice.

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