The influence of wastewater on microbial contamination of the Vltava below Prague

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ABSTRACT

In terms of public health protection, the most important indicator in surface water monitoring is microbial fecal contamination. Despite the introduction of the best available technologies, their biggest source is treated and untreated municipal wastewater. Around 90 % of the Czech population use their local sewerage system, which is linked to a WWTP, treated, and discharged into recipient waters. Monitoring of microbial contamination of the Vltava below Prague CWWTP showed a level of fecal pollution in the 10 km section below the wastewater inflow in periods with different flow rates. Smaller tributaries of the Vltava, which bring treated wastewater from local WWTPs to the Vltava, were monitored as additional sources. From April 2022 to March 2023, the amount of Escherichia coli, enterococci, thermotolerant coliform bacteria, and Clostridium perfringens were monitored at ten sampling sites. The monitoring results showed relatively significant microbial pollution of the Vltava from Prague CWWTP discharge and, at the same time, the river's substantial self-cleaning ability in the following section. This creates good potential for the river's future utilization in the monitored area, with the exception of the section directly affected by the inflow of treated wastewater from Prague CWWTP. This study could be used to raise public awareness in order to minimize the health risk caused by the river's inappropriate utilization (possible presence of pathogenic microorganisms, including carriers of antimicrobial resistance).

INTRODUCTION

The Vltava is an important watercourse which, along its entire length, is the recipient of the inflow of a large volume of wastewater. Thanks to its high water bearing and cascades of water reservoirs, it can eliminate the pollution brought into it relatively effectively. The largest load on the Vltava comes from the capital city of Prague, not only from the Central Waste Water Treatment Plant (CWWTP), but also other smaller treatment plants. Municipal wastewater is one of the main sources of contamination of surface streams with microbial contamination, despite the obligation to use the best available wastewater treatment technologies at WWTPs. Some microorganisms significant for hygiene are not completely eliminated by treatment processes, and thus subsequently adversely affect the quality of water in watercourses, where they can represent an acute (viable microorganisms) or passive (e.g., spread of antibiotic resistance) health risk and a reduction in water usability, such as for recreation and irrigation. Despite this fact, the mechanisms for controlling microbial pollution of wastewater from WWTPs are not currently determined by legislation. In addition, the risk has increased with the current climate period, with the occurrence of extreme phenomena (drought and torrential rainfall). Both of these phenomena are critical for maintaining good microbial guality of surface waters. In periods of low flow rates, due to the low dilution of wastewater from WWTPs and other municipal sources, there is an increased concentration of microbial contamination in streams. In cities, torrential rain poses the risk of increased loading of streams by flood-ways, which bring untreated wastewater that the WWTPs are not able to manage. These facts have significantly contributed to the increased microbial load of watercourses in the long term [1, 2, 14]. Water quality in the Vltava is regularly monitored by the watercourse manager within the state surface water quality monitoring network for the ISVS (Informační systém veřejné správy, Public administration information system) – surface water quality records of the CHMI (Czech Hydrometeorological Institute). However, the sparse network of sampling sites (Vltava above Prague-Vrané nad Vltavou; Vltava below Prague-Zelčín) and the limited scope of monitoring cannot capture the risks associated with microbial contamination of the watercourse.

In our study, the Vltava in Prague and below the city was chosen for the purpose of a closer understanding of microbial contamination and its natural elimination in a large watercourse. Fecal pollution of the water in a recipient watercourse was assessed by determining standard indicator groups of microorganisms *Escherichia coli (E. coli)*, thermotolerant (fecal) coliform bacteria, intestinal enterococci, and *Clostridium perfringens*, whose spores are highly resistant to external environmental factors and survive in wastewater and surface water for a long time [3, 4].

The aim of the study was to characterize microbial pollution in the longitudinal profile of the Vltava below Prague and to determine the degree of increase in health risk and decrease in the usability of water burdened by fecal pollution due to the inflow of wastewater from the WWTP in the monitored section, and to assess the development trend in comparison with historical data [5].



Fig. 1. Vltava in Podbaba below the wastewater inflow from the current and new water line of CWWTP Prague



Fig. 2. Inflow of the Klecanský stream into the Vltava in Klecánky



Fig. 3. Vltava in Řež (the last sampling site of the monitored section)

METHODOLOGY

A 10 km section of the Vltava (from Troja to the Vltava-Řež sampling site) was monitored to determine the influence of treated wastewater discharge from the WWTP on water quality in the researched section of the river's longitudinal profile (*Fig. 1–3*). The descriptions and locations of the sampling sites are shown in *Tab. 1* and *Fig. 4*. Sampling sites on the Vltava were selected for monitoring where it is possible to assess the impact on its water quality by the inflow of treated wastewater from the Prague CWWTP and smaller WWTPs on the tributaries. The characteristics of these WWTPs are shown in *Tab. 2*. For technical reasons, the sampling sites capturing pollution input from the Prague-Bohnice, Nebušice, and Roztoky WWTPs were not included in the monitoring. Other tributaries in the monitored area have no WWTP inflow.

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Tab. 1. Description and location of sampling sites

Site No.	Name of sampling site	Description of sampling sites
1	Vltava-Troja	control sampling site above CWWTP Prague; sampling carried out from Troja footbridge
2	Vltava-Podbaba	sampling below both outlets of the CWWTP Prague from the left bank at the end of Císařský ostrov; the water below the outlets is not sufficiently mixed
3	Vltava-Sedlec	sampling on the left bank of the Vltava in Sedlec near a small beach used for recreation about 2 km below the CWWTP
4	Vltava-Roztoky	sampling near the outlet of the Roztoky WWTP to the Vltava, about 5 km below the CWWTP; is not suffi- ciently representative due to the irregular WW discharge
5	Vltava-Klecany	sampling from the centre of the watercourse about 6 km below the CWWTP; carried out from the ferry
6	Vltava-Řež	sampling from the centre of the watercourse about 10 km below the CWWTP; carried out from the footbridge
7	Drahanský stream	right-hand tributary of the Vltava, outlet of the Prague- -Čimice WWTP; sampling carried out about 1 km from the outlet to the Vltava
8	Přemyšlenský stream	right-hand tributary of the Vltava, outlet of the Zdiby WWTP; sampling carried out before the outlet to the Vltava
9	Klecanský stream	right-hand tributary of the Vltava, outlet of the Klecany WWTP; sampling carried out before the outlet to the Vltava
10	Podmoráňský stream	left-hand tributary of the Vltava, outlet of the Velké Přílepy WWTP; sampling carried out before the outlet to the Vltava
11	Únětický stream	left-hand tributary of the Vltava, outlet of the Horoměřice and Tuchoměřice WWTPs; sampling carried out from the road bridge about 150 m before the outlet of the Vltava



Fig. 4. Sampling site locations

WWTP name	Category according to PE	WW recipient	Number of people connected to the WWTP (2021)	Amount of treated WW in total [thousand m³/year]
CWWTP Praha EWL	over 100,000	Vltava	491,633	44,989
CWWTP Praha NWL	over 100,000	Vltava	706,012	64,601
WWTP Roztoky	2,000-10,000	Vltava	7,869	773
WWTP Dolní Chabry	2,000-10,000	Drahanský stream	4,632	264
WWTP Zdiby	2,000-10,000	Přemyšlenský stream	3,000	166
WWTP Klecany	2,000-10,000	Klecanský stream	3,117	343
WWTP Velké Přílepy	2,000-10,000	Podmoráňský stream	2,935	190
WWTP Horoměřice	2,000-10,000	Horoměřický stream (Únětický stream)	3,450	274
WWTP Tuchoměřice	up to 2,000	Únětický stream	1,816	149

Tab. 2. Basic WWTP characteristics

The amount of *E. coli*, intestinal enterococci, thermotolerant (fecal) coliform bacteria, and *Clostridia perfringens* were determined by standard culture methods. Determination of thermotolerant (fecal) coliform bacteria and *E. coli* was carried out according to ČSN 75 7835 standard, intestinal enterococci according to ČSN EN ISO 7899-2 standard, and *Clostridia perfringens* according to Decree 252/2004 Coll., Annex 6. For culture determination, methods according to the following procedures were used [6–8]. Plain samples were taken from April 2022 to March 2023 with a monthly frequency at the above-mentioned sampling sites.

The results were classified into quality categories according to ČSN 75 7221 [8] and compared with EQS limits (environmental quality standards) for surface waters according to Government Regulation No. 401/2015 Coll. [10]. For information, the results were compared with the limits for bathing waters according to Decree No. 238/2011 Coll. [11].

ČSN 75 7221 [8] uses the characteristic value C90 and, based on it, classifies flowing surface water into five classes according to quality (*Tab. 3*). The standard

allows the so-called purposeful classification according to its own range of indicators, when the resulting class is determined according to the most unfavourable classification. In our case, the classification was used for indicators of thermotolerant (fecal) coliform bacteria and intestinal enterococci.

Government Regulation No. 401/2015 Coll., on indicators and values of permissible pollution of surface waters and wastewater, requirements for permits for discharge of wastewater into surface waters and into sewers, and on sensitive areas from 2016 [10] specifies EQS values for microbiological indicators as P90 for *E. coli*, intestinal enterococci, and thermotolerant (fecal) coliform bacteria (*Tab. 3*).

Decree No. 238/2011 Coll., on the establishment of hygiene requirements for swimming pools, saunas, and hygiene limits of sand in sandpits of outdoor playgrounds from 2011 [11] differentiates between intestinal enterococci and *E. coli* indicators based on P90 and P95 into three quality levels (*Tab. 3*).

Tab. 3. Classification of microbial parameters

	Thermotolerant (fecal) coliform bacteria	Intestinal enterococci	Escherichia coli
		[CFU/100 ml]	
		ČSN 75 7221 (C90)	
Class 1 = unpolluted water	< 2,000	< 600	-
Class 2 = slightly polluted water	< 10,000	< 1,300	-
Class 3 = polluted water	< 20,000	< 2,500	-
Class 4 = heavily polluted water	< 40,000	< 4,600	-
Class 5 = very heavily polluted water	≥ 40,000	≥ 4,600	-
	Governme	nt Regulation No. 401/2015 Sb.	
EQS (P90)	4,000	2,000	2,500
	De	ecree No. 238/2011 Sb.	
excellent quality	-	200 (P95)	500 (P95)
good quality	-	400 (P95)	1,000 (P95)
acceptable quality	-	330 (P90)	900 (P90)
unsatisfactory quality			

Notes: C90 = characteristic value (value with a probability of not exceeding 90 %); P90 and P95 (95% and 90% percentile); EQS = environmental quality standard

RESULTS

The results of microbial pollution monitoring from April 2022 to March 2023 are shown in the graphs in *Fig.* 6–15.

The assessment according to ČSN 75 7221 (*Fig. 5*) showed the best quality at the Vltava-Troja control sampling site, which was classified as slightly polluted water (quality class 2). The outlet of treated wastewater from the CWWTP changed the water quality in Vltava-Podbaba to quality class 5 (very heavily polluted water). Thanks to the intensive dilution of microbial pollution and the active watercourse self-cleaning, the water in the other monitored locations of the longitudinal profile of the Vltava already corresponded to quality class 3 (polluted water). The discharge from heavily polluted tributaries (i.e., Drahanský, Klecanský, Podmoráňský, and Únětický streams) did not contribute to the significant deterioration of water microbial quality. Of the monitored streams, Přemyšlenský stream showed the best quality, on which the apparently well-functioning WWTP from the village of Zdiby-Přemyšlení is located.

The water quality at the monitored sampling sites was compared with EQS for microbiological indicators, expressing the surface water status (*Tab. 3*). The results for all monitored indicators are presented in *Figs. 6–13* and in *Tab. 4*. Both the graphs and the table show significant differences between the individual sampling sites, not only in the Vltava but also in its tributaries below Prague. Indicators of thermotolerant (fecal) coliform bacteria and *E. coli* were exceeded at 60 % of the sampling sites in the Vltava and in 80 % of the tributaries; the indicator of intestinal enterococci was exceeded at only 20 % of the sampling sites in the Vltava, but in 80 % of the tributaries. The control sampling sites on the Vltava above the CWWTP and

the Vltava-Klecany met the requirements of Government Regulation No. 401/2015 Coll. for all three indicators. Despite the inflow of pollution from many sources during its flow through Prague (including the significantly polluted Botič and Rokytka streams), the results showed that the Vltava in Troja has unexpectedly good water quality. According to the original assumptions, a significant increase in the number of microbial indicators occurs in the next section, where wastewater treated on two water lines of the Prague CWWTP flow into it. This is despite the fact that the CWWTP is equipped with modern treatment technology that meets the requirements for the best available wastewater treatment technologies and is actively taking other steps to improve the quality of discharged water, such as UV disinfection of treated wastewater (which was put into trial operation in October 2021). As the graphs in *Fig. 6–14* show, intensive dilution and self-cleaning occur in the Vltava, thanks to which there are already significantly fewer microbiological indicators in the section from Sedlec, namely about 2 km below the CWWTP.

Surprising differences were found in the microbial quality of the water of the Vltava tributaries below Prague into which wastewater from local WWTP is fed (*Fig. 6–14, Tab. 4*). Critical pollution was brought by the Klecanský and Podmoráňský streams, to which the Klecany and Velké Přílepy WWTPs are connected, i.e., around 6,000 connected inhabitants. In contrast, Přemyšlenský stream (Zdiby WWTP; 3,000 connected inhabitants) showed the lowest pollution throughout the monitoring period. Thanks to the relatively low water bearing of the polluted tributaries, the influence on the Vltava below their inflow is not significant (*Fig. 14*). According to ČSN 75 7143, Vltava water is "conditionally suitable" for irrigation at most sampling sites, i.e., the numbers of thermotolerant (fecal) coliform bacteria and enterococci range between 1,000 and 10,000 CFU/100 ml [15].

Sampling site	Thermotolerant (fecal) coliform bacteria		Intestinal enterococci		Escherichia coli	
	P90/ average [CFU /100 ml]	min./max. [CFU/100 ml]	P90/ average [CFU/100 ml]	min./max. [CFU/100 ml]	P90/ average [CFU/100 ml]	min./max. [CFU/100 ml]
Vltava-Troja	2,870/1,252	60/5,400	1,300/567	48/4,800	1,940/992	50/5,400
Vltava-Podbaba	27,800 /17,167	800/79,000	9,610 /5,258	80/2,800	19,800 /10,758	200/36,000
Vltava-Sedlec	8,800 /4,145	600/20,000	1,800/1,045	130/5,200	4,800 /2,655	400/13,000
Vltava-Klecany	3,790/2,575	130/11,000	1,640/691	90/2,100	2,490/1,668	110/6,000
Vltava-Řež	7,820 /2,837	200/12,000	1,990/763	90/2,400	4,540 /2,006	110/9,000
Drahanský stream	12,000 /19,245	500/180,000	3,700 /4,363	190/34,000	12,000 /19,245	500/180,000
Únětický stream	9,410 /5,981	110/32,000	6,020 /2,177	200/7,400	8,440 /4,501	990/22,000
Přemyšlenský stream	2,090/1,146	70/5,800	1,990/1,478	20/9,600	1,730/907	70/3,800
Klecanský stream	117,000 /59,492	600/520,000	76,200 /19,108	600/120,000	93,600 /42,808	500/360,000
Podmoráňský stream	37,900 /23,625	2,000/39,000	20,800 /12,033	800/52,000	27,700 /16,142	1,700/30,000

Tab. 4. Classification of microbiological parameters

Note: P90 values exceeding EQS Government Regulation No. 401/2015 Coll. are marked in bold

The results were further evaluated according to Decree No. 238/2011 Coll., which assesses water quality after the end of the bathing seasons (ideally 4) based on its determination according to the indicators listed in *Tab. 3*. Water is classified as "unsuitable" for bathing if the values of any of the indicators are higher than the values for "acceptable" quality. According to the above assessment, in the observed period, none of the Vltava's sampling sites achieved a quality acceptable for bathing. However, at the Vltava-Troja sampling site, in the period suitable for recreation (i.e., from May to September), the numbers of CFU in both indicators were found to be 100 % corresponding to the limits for "acceptable"

quality. The sampling site is located near Troja slalom channel, where canoeists often come into contact with water. In the next section of the watercourse, below the inflow of the CWWTP, values below the limit for unacceptable quality appeared only sporadically. The situation was better with intestinal enterococci. At the Vltava-Řež sampling site, about 10 km from the CWWTP, in the period suitable for recreation (May to September), the numbers of CFU in both indicators were found to be already 70 % corresponding to the limits for "acceptable" quality. Some sampling was carried out after heavy rain, which negatively affected the water quality (*Fig. 15*).



Fig. 5. Sampling site pollution rates for thermotolerant (fecal) coliform bacteria and intestinal enterococci according to Czech technical norm ČSN 75 7221













Clostridium perfringens — Vltava



Fig. 6–13. Microbial water quality of monitored sites of the Vltava and its tributaries





Fig. 14. Seasonal progress of selected microbial indicators in the Vltava; columns with values exceeding the bathing water limits are represented in grey – dark grey for *E. coli*, light grey for enterococci



Fig. 15. Sum of selected microbial indicators during the monitoring in comparison with Vltava flow rate

DISCUSSION

The aim of this study was to monitor the current state of water quality in the Vltava from Troja to the footbridge in Řež, along with several important tributaries with an inflow from local WWTPs. Water quality in this part of the Vltava is not systematically monitored and suggests very strong pollution, both from the many sources below Prague and from the discharge of treated wastewater from the Prague CWWTP on Císařský ostrov. Annual monitoring of this area was aimed at identifying limitations in the usability of the Vltava water for its further use (e.g., recreation and irrigation). The results showed that the Vltava has huge capacity to absorb pollution and eliminate it. However, the use of water for recreation is completely limited in a relatively short section below the CWWTP inflow. After a few kilometres, however, the watercourse regenerates from the point of view of microbial contamination, probably due to strong dilution, sedimentation, and active self-cleaning processes. Although the study was not focused on monitoring changes due to climatic episodes, it was found that both extreme conditions (drought with low flows and heavy torrential rains) have a negative effect on water quality. Low flows will limit the possibility of diluting the incoming pollution, which was already visible at the control Troja sampling site; on the other hand, torrential rain worsens water quality in the Vltava and its tributaries due to the effect of surface pollution flushes and the inflow of flood-ways directly into the recipient watercourses.

The microbial quality of the water in the Vltava below Prague has not been systematically monitored in recent years or the data has not been published. The sampling sites monitored as part of the surface water monitoring programme are relatively less dense in the section of the Vltava below Prague due to the larger size of the water bodies. The Vltava is regularly monitored at the representative sampling sites of Vrané nad Vltavou and Zelčín. In Prague, water quality is monitored in detail at the Vltava-Podolí sampling site, and basic physico-chemical and microbiological indicators are monitored at the Prague-Troja sampling site. Pražské vodárny a kanalizace, a. s. also monitors the Prague-Podolí sampling site.

The first, completely unique bacteriological study of the Vltava was carried out in 1931 by Kredba and Dvořák [5, 12]. In this study, a one-time monitoring of the presence of microorganisms was performed. Sampling was done in a short time interval (3 days) in April to be as little affected by climatic and other factors as possible. Samples were taken from both banks in the marked section, which started above Prague at the Zbraslav bridge and ended below Prague under the weir in Roztoky. Sampling sites were chosen so that each source of pollution could be captured. In accordance with the methods of the time, "*bact. coli* numbers in 1 l" (modified Ficker-Partiš method on Endo agar) and "number of germs in 1 ml" were determined. We assume that the method of determining *bact. coli* could be comparable to today's method of determining coliform bacteria on Endo agar [4]. A comparison of historical and current data is shown in *Tab. 5.*

Another source of older data was the study by Baudišová [13, 14] in 1997, which dealt with the water microbial quality in the Labe and the lower Vltava. The study was focused on the quality of the water before the Vltava meets the Labe, and coliform bacteria and thermotolerant (fecal) coliform bacteria were monitored in it. The values found at the Podolí, Troja, Roztoky and Zelčín sampling sites are shown in *Tab. 5.* A comparison of the values of coliform

Tab. 5. Comparison of present fecal pollution situation with historical data

n the Labe and the lower Vltava. water before the Vltava meets olerant (fecal) coliform bacteria Podolí, Troja, Roztoky and Zelčín arison of the values of coliform

1931 (Kredba)	1996 (Baudišová)		2022–2023	
<i>Bact. coli</i> (left/right bank) – one-time sampling	Coliform bacteria (annual average)	Thermotolerant (fecal) coliform bacteria (annual average)	Thermotolerant (fecal) coliform bacteria (annual average)	
		[CFU/1 000 ml]		
-	77,000	22,000	-	
-	57,000	23,000	12,500	
30,000/64,000	-	-	-	
94,000/1,200,000	-	-	171,670 (left bank)	
550,000/630,000	2,384,000	922,000	2,000*	
400,000/450,000	-	-	25,750 (left bank)	
-	1,309,000	576,000	-	
	1931 (Kredba) Bact. coli (left/right bank) – one-time sampling - - 30,000/64,000 94,000/1,200,000 550,000/630,000 400,000/450,000	1931 (Kredba) 1996 (Baudišová) Bact. coli (left/right bank) – one-time sampling Coliform bacteria (annual average) - 77,000 - 57,000 30,000/64,000 - 94,000/1,200,000 - 550,000/630,000 2,384,000 400,000/450,000 - - 1,309,000	1931 (Kredba) 1996 (Baudišová) Bact. coli (left/right bank) – one-time sampling Coliform bacteria (annual average) Thermotolerant (fecal) coliform bacteria (annual average) Image: CFU/1 000 ml] [CFU/1 000 ml] Image: CFU/1 000 ml] 22,000 Image: CFU/1 000 ml] 23,000 Image: CFU/1 000 ml] 1 Image: CFU/1 000 ml] 1	

* One-time sampling on 25th April 2022



Fig. 16. Amount of bact. coli in the Vltava in 1931

CONCLUSIONS

The updating of historical data describing microbial contamination of water in the Vltava below Prague showed a favourable trend in development, which is a consequence of improving technological processes of wastewater treatment.

It was found that fecal contamination at the Vltava-Troja control sampling site is unexpectedly low, despite the fact that it receives pollution from many sources in Prague. In the period when it could be used for recreation, the water showed "acceptable" quality according to Decree No. 238/2011 Coll.

Water quality at the other sampling sites was strongly affected by discharged purified wastewater from both CWWTP water lines, and in the area near the sampling site, its further usability was strongly reduced, especially for recreation.

bacteria in 1 l of water from the three periods indicated a significant decrease

However, the condition quickly improved in the next section of the Vltava thanks to dilution, sedimentation, and cleaning processes; the water quality at more distant sampling sites already showed "acceptable quality" during some samplings during the period of possible summer recreation.

Although the study was not focused on monitoring changes due to climatic episodes, it was found that both extreme conditions (drought with low flows and heavy torrential rains) have a negative effect on water quality. Low flows will limit the possibility of diluting the incoming pollution, which was already visible at the Troja control sampling site; on the other hand, torrential rain worsens water quality in the Vltava and its tributaries due to the effect of surface pollution flushes and the inflow of flood-ways directly into the recipient watercourses.

The study showed relatively good potential for the possible use of the Vltava in Prague and below Prague, with the exception of the section strongly affected by the inflow of treated wastewater from the Prague CWWTP. In general, it would be advisable to focus on increasing awareness with a help of this and similar studies, so that water can be used more efficiently and, conversely, so that it does not represent a significant health risk when used inappropriately (possible presence of pathogenic microorganisms, including antibiotic resistance carriers).

With regard to the growing requirements for the protection of the health of the human population and improvement of water quality in streams to the level of the standards of European directives, it is necessary to focus more attention on the monitoring microbiological indicators of water quality than is ensured by the current routine monitoring system.

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