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Elimination of bacterial contamination from domestic sewage using vertical flow constructed wetland

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ABSTRACT

Constructed wetlands (CW) plants are successfully used for treating various types of wastewater. Their most popular use is for the treatment of domestic sewage. The main aim of this research is to assess the change of microbiological parameters (total coliform, faecal coliform, total number of mesophilic and cryophilic bacteria, number of enterococci) during domestic sewage treatment in two CW beds with different fillings. The concentration of organic matter BOD_5, COD), total Kjeldahl nitrogen TKN, ammonia nitrogen NH_4–N, and total phosphorus were also analyzed. The research installation was based on two vertical subsurface flow constructed wetlands (VS-SF CW) – the first one filled with a Certyd aggregate and the second one with mineral material. Both beds were planted with reeds and worked in parallel with the same hydraulic load 0.1 $m^3/m^2 \cdot d$ (m/d). The research was conducted during vegetative and non-vegetative seasons. Both beds showed a high effectiveness of reduction of most microbiological parameters, amounting to over 95%. The removal efficiency of the total coliform (TC) and faecal coliform (FC) parameters for the Certyd bed during the vegetative season was 99.9%, 98.3% and 97.51%, respectively in the non-vegetative season. In contrast, for the mineral-filled bed, the TC and FC removal efficiency during the vegetative season were, respectively 78.1% and 74.3%, respectively, 65.7% and 58.9% in the non-vegetative season. A difference in efficiency was observed depending on the season. High removal efficiencies of organic matter (measured by BOD_5, COD values) and nutrient compounds was observed during the study. The conducted research proved a high efficiency of constructed wetlands in removing microbiological and chemical parameters. Overall, higher efficiency was observed in the bed filled with Certyd.

Keywords: Bacterial contamination; Constructed wetlands, Domestic sewage; Subsurface vertical flow system

1. Introduction

Due to pollution of water resources and water shortages, microbiological and biotechnological solutions are being sought to protect the environment. Conventional wastewater treatment plants (sludge activated system, trickling filter) are unable to provide adequate microbiological protection of bodies of water. Improper treatment or the discharge of

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Presented at the 3rd International Conference on Strategies toward Green Deal Implementation – Water, Raw Materials and Energy (ICGreenDeal2022), 5–7 December 2022, held online by the Mineral and Energy Economy Research Institute, Polish Academy of Sciences untreated wastewater can cause sanitary hazards. It is necessary to monitor the microbiological parameters of wastewater discharged to the receiver, for example, total coliform (TC) and faecal coliform (FC). To prevent disease transmission, effective wastewater treatment methods that meet, among others, microbiological quality guidelines, should be adopted [1–3].

Natural systems for removing various contaminants from wastewater, such as constructed wetlands (CW), are increasingly being used. They consist of four main components: wetland macrophytes, wetland substrate, water column and living organisms [4]. They are successfully used to treat various types of wastewater. They effectively remove organic and inorganic contaminants including: BOD_5, COD, potassium, sulfate, nitrogen, phosphorous, industrial chemicals, heavy metals and pharmaceuticals residues from domestic or municipal sewage.

Among the processes occurring in CW beds are microbial degradation, filtration, plant uptake, biological precipitation, chemical oxidation, adsorption, biostabilization and volatilization [5]. CW wastewater treatment is characterized by low energy requirements, does not require the use of chemicals and does not produce waste sludge. In addition, their efficiency, simple construction and low cost of construction and operation make these systems increasingly used by homeowners to treat domestic wastewater [6,7].

The selection of filtration media (grain composition and size) depending on the location (depth) in the bed is critical to effective treatment. Typically, well-sorted sand and gravel are recommended. An inappropriate fractional structure of the bed can cause hydraulic overloading and clogging [8]. In the study, classic mineral fill (gravel) and Certyd aggregate were used as fill. Certyd is a lightweight, porous ceramic material obtained by thermal treatment of ash [9].

Much attention has been paid to examining the effectiveness of systems in terms of physicochemical parameters, while there is less information and literature data on the removal of microbial contaminants. Hench et al. [10] proved that commonly used groups of indicator organisms (total coliforms, faecal coliforms, and enterococci) can be effectively removed with efficiencies ranging from 80% to 99%. Based on Jóźwiakowski's research [11], it was found that in vertical flow CW beds (97.70%–99.87%), the removal of TC and TC parameters is more efficient than in horizontal flow beds (68.26%–99.24%). The processes contributing to the removal of microbial contaminants are sedimentation, filtration, aggregation, oxidation, antibiosis, solar irradiation, competition and predation. It was also found that these systems were distinguished by a more effective removal of *Escherichia coli* (*E. coli*) bacteria compared to non-planted systems [10]. The scientific objective of the study was to determine and compare the efficiency of removing microbial contaminants depending on the season and the type of bed filling (gravel and Certyd). The novelty aspect is the practical investigation of the applicability of a new constructed wetland fill obtained from waste by sintering and the extension of the study to microbiological parameters due to the post-sintering methods of recovering water from wastewater.

2. Methodology

2.1. Research installation

In the study, a system based on two vertical flow CW beds (SS-VF CW) was used. Both beds were characterized by a depth of 0.80 m. The filtration media in each bed was composed of three layers of similar depths. Fig. 1 presents a detailed scheme of the research installation. Besides the two beds, the research installation also included a retention tank. Samples for testing were taken at three measurement points (I, II, III) [12,13].

The structure of the beds is shown in Figs. 2 and 3. Both beds were planted with reeds (*Phragmites australis*), which are most commonly used in constructed wetland systems.

Data obtained by Shahamat et al. [14] show that wetland plants such as reeds can be used as a cost-effective source for improving the quality of treated wastewater. The use of reeds in hydrophytic systems effectively removes various pollutants from wastewater in accordance with Effluent

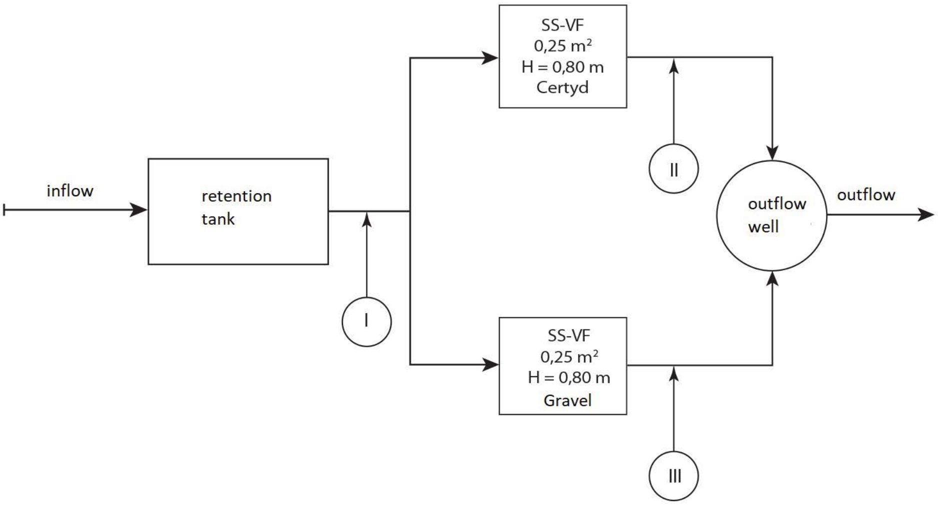

Fig. 1. Scheme of research installation.

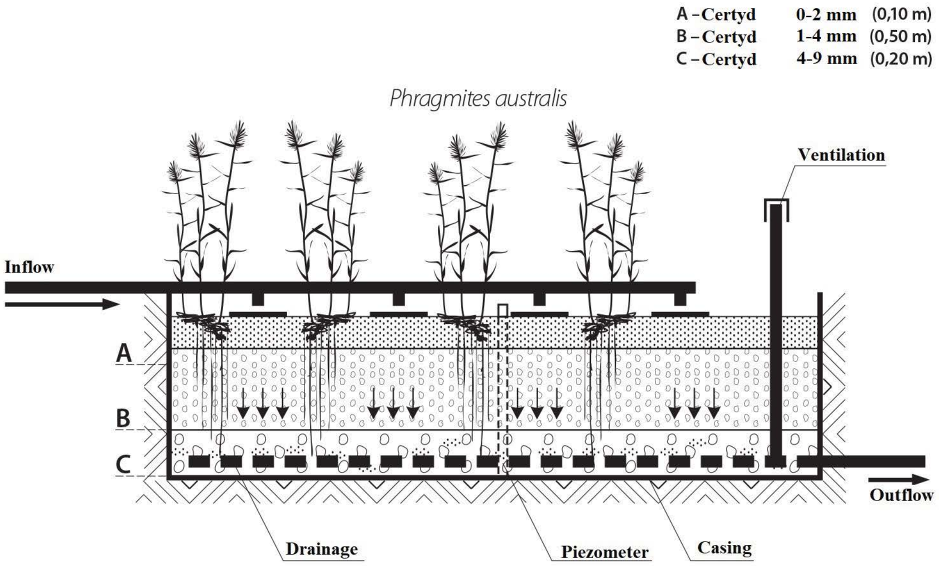

Fig. 2. Cross section of Bed A.

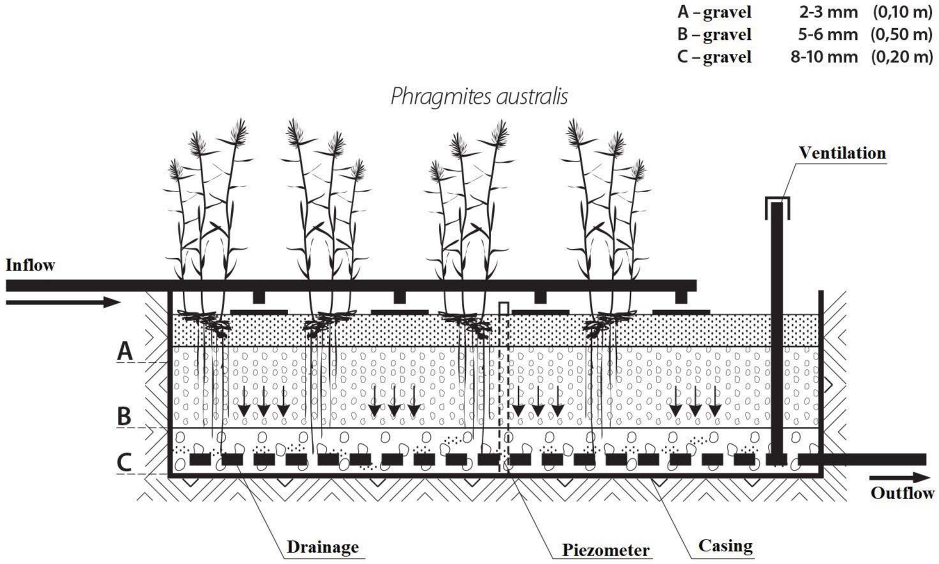

Fig. 3. Cross section of Bed B.

Guideline regulations and is an environmentally friendly and cost-effective method. Beds A and B were operated in parallel at a hydraulic load of 0.1 $m^3/m^2 \cdot d$ (m/d).

2.2. Collection of samples and analysis

The study evaluated microbiological and physicochemical parameters before and after the treatment of domestic wastewater in Beds A and B. The research was carried out in the period July–December 2021. 10 series of measurements were made (5 series in the vegetative period, 5 in the non-vegetative period). The study series covered a raw wastewater sample and two treated wastewater samples. The tests were performed in the Department of Environmental Engineering and Natural Sciences laboratory at Bialystok University of Technology. Wastewater testing was conducted in accordance with the requirements of the American Public Health Association (APHA) and Regulation of the Minister of Maritime and Inland Waterway Economy from 12th July 2019 [15,16]. Microbiological tests included: determination of the total number of mesophilic bacteria (Mesophiles) at a temperature of 37°C and cryophilic bacteria (Cryophiles) at 22°C, determination of the total coliform (TC) and faecal coliform (FC) index, and determination of the *Enterococcus* bacteria index. The total number of heterotrophic bacteria (mesophilic and cryophilic), was determined according to PN-EN ISO 6222:2004 [17]. Determination of the TC and FC index was performed following the fermentation-tube method. The number of enterococci was determined according to PN-EN ISO 7899-2:2004 by the membrane filtration method [18]. The content of organic matter (BOD_5, COD), suspended solids (SS), total nitrogen (TN) Kjeldahl nitrogen (TKN), ammonium nitrogen (NH_4–N) and total phosphorus (TP) were also analyzed. Spectrophotometer Spectroquant Pharo 100 was used BOD_5 was determined using OXI-TOP®.

2.3. Data refining

In order to compare the efficiency of wastewater treatment in the two beds, the average efficiency was calculated as the average difference between the inlet and outlet divided by the average value at the inlet.

$$\eta = \frac{\text{mean}\left(\text{value}_{\text{in}}\right) - \text{mean}\left(\text{value}_{\text{out}}\right)}{\text{mean}\left(\text{value}_{\text{in}}\right)} \quad (1)$$

where η – mean efficiency, mean ( ) – mean value, value_{in} – concentration/value on inflow, $\text{value}_{\text{out}}$ – concentration/value on outflow.

Due to different orders of magnitude involved, it is difficult to perform a direct comparison of all microorganisms removal across all studied groups, bed fillings and vegetative periods. In order to robustly present the obtained data, removal factors (*f*) were calculated as defined by the Eq. (2):

$$f = -\ln\left(\frac{\text{value}_{\text{out}}}{\text{value}_{\text{in}}}\right) \quad (2)$$

where f – removal factor, ln() – natural logarithm.

The higher the factor, the more microorganism are removed from sewage. Differences in factors represent number of e-folds between appropriate counts. Sets of the factors were plotted in the form of boxplots [19]. Each such boxplot consists of a box (marking quartile 1 and 3 along with median in the middle) and whiskers extending by distance d proportional to the interquartile range, but not further than minimum or maximum of the data. Observations outside whiskers are also plotted and can be interpreted as outliers.

$$d = \frac{\text{IRQ}}{\sqrt{n}} \quad (3)$$

where d – maximum extent of whiskers, IRQ – interquartile range, n – numbers of factors in given set.

3. Results and discussion

Calculations and graphs were prepared using Microsoft Excel spreadsheet and R statistical environment version 4.2.2 ("Innocent and Trusting") [20]. Table 1 shows microbiological and physico-chemical parameters of raw and treated wastewater using two types of beds in vegetative and in non-vegetative seasons. Figs. 4 and 5 show the comparison of removal efficiency between Beds A and B in vegetative and non-vegetative periods.

Based on the figures, it can be concluded that both beds showed better contaminant removal efficiency during the vegetative period. In the case of the Certyd-filled bed (Bed A), a high removal efficiency of 99.9% for total coliform and faecal bacteria was achieved during the vegetative period. The removal efficiency of faecal bacteria in CW beds varies with bed design, hydraulic residence time, temperature, and hydraulic and mass loading rate [21]. Removal efficiency of other microbial parameters in both periods was above 95% (except for Cryophiles in the non-vegetative period –92.02%). The removal efficiency of total coliforms and faecal bacteria in the bed filled with mineral aggregate (Bed B) during the vegetative period was 78.05% and 74.30%, respectively, and outside the vegetative period was 65.74% and 58.88%. In turn, the removal efficiency of the total number of mesophilic bacteria, psychrophilic bacteria and enterococci during the vegetative and non-vegetative periods ranged from 56% to 76%.

For both beds during the vegetative period, a high organic matter removal efficiency measured by BOD_5, COD and N–NH_4 values was obtained, amounting to: 94.27%, 86.69% and 86.13% (Bed A) and 91.28%, 83.28% and 84.92%, respectively (Bed B). In the non-vegetative period, the efficiency was slightly lower, at 87.84%, 78.78% and 80.83% for Bed A and 83.99%, 74.45% and 71.28% for Bed B, respectively. The removal efficiency of Kjeldahl nitrogen, total nitrogen, total phosphorus and total suspended solids for both beds during the vegetative period was in the range of 62%–87% and 52%–85% outside the vegetative period.

Comparing the results to the study conducted by García-Ávila [22], a higher efficiency was observed in own study. The removal efficiency of TC, FC, BOD_5, COD, ammonia nitrogen and phosphates was respectively: 96.02%, 93.74%, 75.39%, 64.78%, 70.70%, and 49.38%. In other studies, the average removal efficiency of BOD_5, COD, TN, N–NH_4 and TP was respectively: 82.12%, 79.79%, 51.46%, 74.06%, and 25.42% [12].

Based on their study, Sohair and Hellal [23] showed that the average removal efficiency of bacterial indicators TF and FC ranged from 94% to 99.9%. High removal of fecal coliform bacteria (~95%) was obtained by Ran et al. [24] using water lash (*Lemna gibba*) in this type of system. Sleytr et al. [25] proved that planted and unplanted SS-VF CW show high removal rates of faecal coliforms (*E. coli*, TC) and enterococci. There is no significant difference in microbial removal efficiency between VSSF-CVs with and without plants.

The efficiency of beds depends on microbial activity, hydraulic loading rate, hydraulic retention time, vegetation type and temperature [21]. Torrens et al. [26] proved that the presence of *Pharagmites austrialis* is of minor importance for the removal of faecal indicators in SS-VF CW beds. Bacterial indicators in these beds were better removed than viral indicators. In addition to the high efficiency of organic matter removal, nitrification and denitrification processes also occur in the beds. Compared to horizontal flow systems, denitrification is less effective. For that, vertical systems tend to have a higher removal efficiency of organic pollutants and nutrients [27].

Fig. 6 presents removal factors plotted against groups of microorganisms, types of beds and seasons. The values of the removal rates of the *Enterococcus* parameter are similar. The differences for Beds A and B were: 0.90 ± 0.01, 0.98 ± 0.23 (vegetative period) and 1.61 ± 0.01, 1.14 ± 0.23 (non-vegetative period), respectively. The removal coefficient of FC and TC parameter in the vegetative and non-vegetative periods were 7.7 ± 1.0, 5.0 ± 1.7 and 7.54 ± 0.46, 4.49 ± 0.74 (Bed A)

Table 1
Microbiological and physico-chemical parameters of raw and treated wastewater (beds A and B) during vegetative and non-vegetative periods

Vegetation	Raw wastewater	Treated wastewater	
Parameter		Bed A	Bed B
Enterococcus, CFU/mL	1.38E4 ± 6.57E3	4.90E2 ± 0.0	5.66E3 ± 2.72E3
FC, CFU/mL	3.16E6 ± 1.93E6	1.85E3 ± 1.77E3	8.12E6 ± 5.18E5
Mesophiles, CFU/mL	3.16E5 ± 3.88E4	7.70E3 ± 1.51E4	7.3E4 ± 1.72E4
Cryophiles, CFU/mL	2.8E5 ± 1.10E4	1.47E4 ± 2.87E4	8.96E4 ± 2.03E4
TC, CFU/mL	8.82E6 ± 7.79E6	6.12E3 ± 7.93E3	1.94E6 ± 1.57E6
BOD_5, mg/L	415.0 ± 13.4	23.4 ± 2.4	36.2 ± 3.5
COD, mg/L	738.0 ± 14.7	98.2 ± 7.5	123.4 ± 3.4
$N–NH_4$, mg/L	102.4 ± 7.0	14.2 ± 0.5	15.4 ± 0.3
TKN, mg/L	114.8 ± 4.4	16.6 ± 0.8	23.8 ± 0.7
TN, mg/L	115.6 ± 4.5	38.8 ± 2.5	41.0 ± 2.0
TP, mg/L	14.7 ± 1.4	5.6 ± 0.1	5.5 ± 0.4
SS, mg/L	72.0 ± 1.9	14.6 ± 1.4	9.4 ± 0.8
Non-vegetation	Raw wastewater	Treated wastewater	
Parameter		Bed A	Bed B
Enterococcus, CFU/mL	1.22E4 ± 5.23E3	4.00E2 ± 0.0	3.75E3 ± 1.78E3
FC, CFU/mL	2.14E6 ± 3.67E5	5.33E4 ± 8.86E4	8.80E5 ± 2.40E5
Mesophiles, CFU/mL	3.14E5 ± 1.35E5	7.72E3 ± 1.32E4	1.36E5 ± 1.72E5
Cryophiles, CFU/mL	5.56E5 ± 5.77E5	4.44E4 ± 8.28E4	1.42E5 ± 1.84E5
TC, CFU/mL	4.32E6 ± 1.51E6	7.26E4 ± 8.14E4	1.48E6 ± 2.99E5
BOD_5, mg/L	426.0 ± 18.5	51.8 ± 7.5	68.2 ± 10.3
COD, mg/L	762.4 ± 39.4	161.8 ± 6.7	194.8 ± 14.1
$N–NH_4$, mg/L	98.9 ± 5.3	19.0 ± 0.2	28.4 ± 2.4
TKN, mg/L	118.6 ± 10.4	35.1 ± 3.7	39.8 ± 4.1
TN, mg/L	119.7 ± 10.0	53.1 ± 3.4	52.3 ± 3.2
TP, mg/L	14.0 ± 2.6	6.7 ± 0.8	6.4 ± 0.6
SS, mg/L	72.0 ± 5.1	13.6 ± 1.5	11.2 ± 1.6

Note: Mean ± standard deviation.

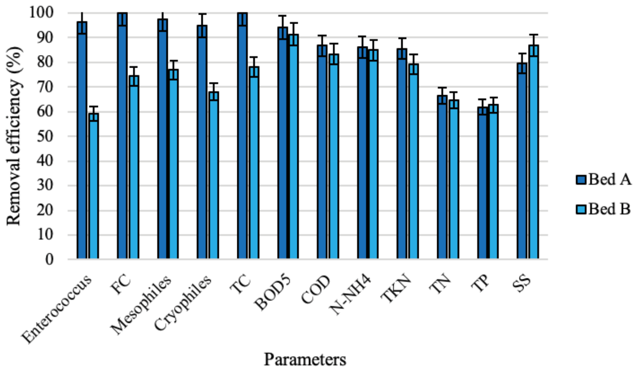

Fig. 4. Comparison of removal efficiency between Bed A and Bed B during the vegetative period.

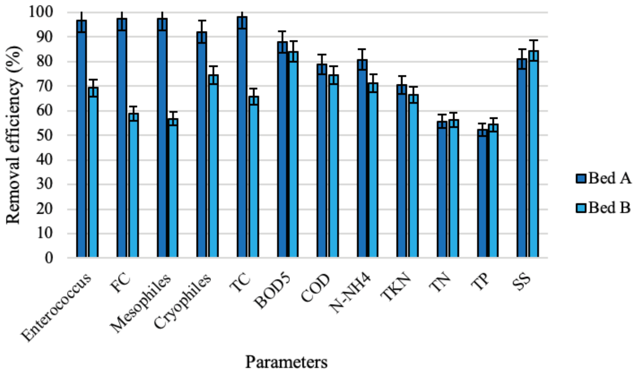

Fig. 5. Comparison of removal efficiency between Bed A and Bed B outside the vegetative period.

and 1.6 ± 1.4, 0.93 ± 0.36 and 1.8 ± 1.4, 1.02 ± 0.35 (Bed B), respectively. In turn, the removal rates of Mesophiles and Cryophiles during the vegetative and non-vegetative periods were 6.7 ± 2.7, 5.6 ± 2.0 and 5.6 ± 2.4, 4.5 ± 1.7 (Bed A) and 1.49 ± 0.27, 1.65 ± 0.89 and 1.16 ± 0.28, 1.56 ± 0.37 (Bed B), respectively.

In contrast to the mineral bed (Bed B), the Certyd bed (Bed A) behaves differently when it comes to vegetative

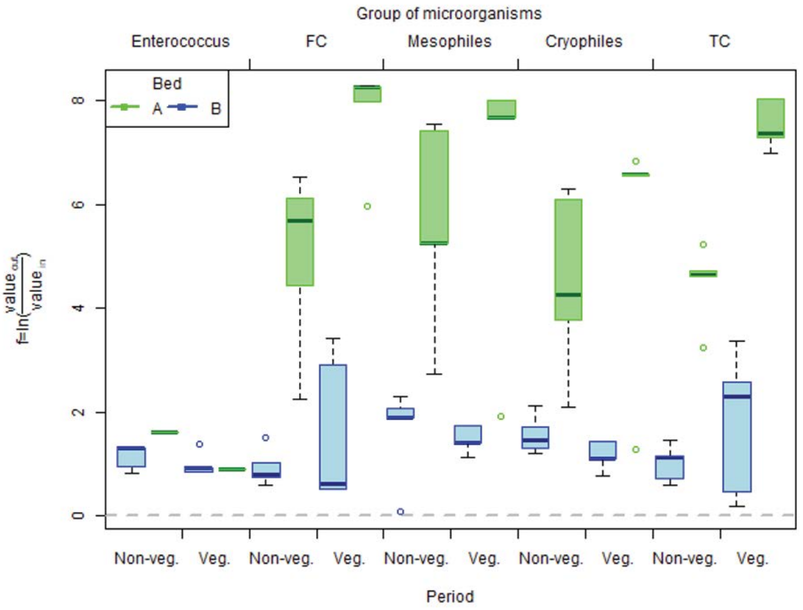

Fig. 6. Removal factors.

periods. In the examined groups of bacteria (with the exception of *Enterococcus*) in Bed A there is a visible difference. For example, for the FC parameter, the difference between the vegetative and non-vegetative periods is 2.7. This means that the non-vegetative inlet/outlet ratio is more than 14 times higher than the outlet/outlet ratio before vegetative. In the case of Bed B, the difference between periods is small, at 0.67 (almost 2 times the ratio). Regardless of the period, Bed A achieves better bacterial removal results than Bed B. The difference in removal ratios for Bed A is 4.1 (in the vegetative period) and 6.1 (in the non-vegetative period) higher (more than 58- and 445-fold higher ratios).

4. Conclusions

Vertical flow CW beds were found to effectively remove microbiological contaminants from domestic wastewater. The study shows that both beds showed high efficiency in reducing microbiological parameters (above 95%). The bed filled with Certyd had a higher efficiency compared to the bed filled with mineral aggregate. Removal efficiency of microbiological parameters: FC, Mesophiles, Cryophiles and TC during the growing season was more than 20% higher, and the parameter Enterococcus was 37% higher. Meanwhile, in the non-vegetative period, the difference in removal efficiency of microbiological parameters (Enterococcus, FC, Mesophiles, Cryophiles and TC) was: 27%, 39%, 41%, 18% and 33%, respectively. Removal factor analysis reveals that except for *Enterococcus* group, there is clear division between the two beds. While Bed A has a better value of removal factors in the vegetative period, differences between factors in Bed B are far less pronounced.

As in the case of microbial contaminants, better efficiency of organic matter and nitrogen removal was found in the Certyd-filled bed. The use of Certyd as a CW bed fill can help reduce environmental and landscape degradation associated with mineral aggregate mine operations.

The obtained research results can be helpful in the implementation of CW systems for the treatment of domestic wastewater and the secondary use of treated wastewater, for example, irrigation or other purposes. The use of the constructed wetland method for effective removal of microbial and physical–chemical pollutants may be limited to small wastewater treatment plants. On the other hand, these systems can effectively improve the quality of municipal wastewater by their application after typical biological treatment using activated sludge or a trickling filter.

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