

Analysis of the Impact of Stormwater Systems on Receiving Waters Based on the Analysis of Algal Community Structure

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ABSTRACT

One of the important ways to prevent permanent environmental pollution is to constantly monitor its quality, which can be performed in several ways. The present bioindication study analyzed the level of diversity and abundance of biofilm microorganism communities, which illustrate the state of the studied aquatic environment, enabling to determine its quality. The impact of stormwater discharge on the receiver at particular points was evaluated on the basis of the reaction of selected microorganisms or their groups to the substances appearing in the watercourse. The study of indicator organisms gives information about the waters of a given body of water without expensive hydrochemical tests and without causing a burden on the environment during the production as well as disposal of reagents that are consumed in many classical physicochemical analyses. On the basis of selected algal species, the Shannon index and McArthur index were calculated, and the effect of storm sewer discharge on the communities of indicator organisms was determined. The best visible impact of storm sewer discharge was seen on the basis of the entire study cycle in relation to the median of the McArthur index.

Keywords: surface water quality, bioindication, algae, community structure, stormwater systems.

INTRODUCTION

Rivers are reservoirs characterized by a constant flow of water and continuous changes occurring in the biocenosis and biotope [Starmach, 1976]. Rivers are also open ecosystems to which organic matter is supplied from the land, and not only from the site of photosynthesis production [Starmach, 1976, Mikulski, 1982]. Natural flowing waters are fed by groundwater, rainfall, surface runoff, or point discharges of wastewater of various types. The quality of stormwater is affected by the constant generation of pollutants and production of waste; this, in turn, is inherent in the functioning of human civilization [Januchta-Szostak, 2019]. Stormwater washes substances from the surface of the catchment area, which are delivered to the receiver through the storm or

combined sewer system. Surface runoff waters, which leach compounds formed, among other things, as a result of automobile transportation – oils, fuels, as well as fertilizers and other chemicals used in agriculture – also contain pollutants leached from the air, resulting from the activities of industrial plants [Bobrowski, 2002]. The content of substances in rainwater depends on traffic volume, land use or urban infrastructure, as well as the number and size of industrial plants. The amount of allochthonous substances transported with wastewater is increased along with anthropogenic pressure [Babko et al., 2016]. Water, due to its structure, is a carrier through which pollutants can move continuously and without constraints [Wiatkowski and Kózka, 2014].

Depending on the quality of the stormwater flowing into the receiving water body, its

parameters change, including the amount of dissolved oxygen [Spänhoff et al. 2007; Wakelin et al. 2008; Pliashchynk et al. 2018]. This causes irreversible changes in aquatic ecosystems [Gücker et al. 2006; Gorzel and Kornijów, 2007]. An important feature of rivers, which should be taken into account when compiling the results, is that the flow of matter takes place in only one direction, i.e. downstream [Chełmicki, 2001] Due to the effects that can be caused by inflows of polluted stormwater, i.e. containing increased amounts of total suspended solids, nitrogen, or heavy metals [Joshi and Balasubramanian, 2010], it is important to constantly monitor the quality of the environment.

Various methods are used for the study of water quality and the impact of point sources on receiving waters, ranging from standard physical and chemical methods, through various types of bioindication methods. In the future, these will allow implementing innovative methods of multidimensional data analysis which can be treated as a kind of soft-sensors, or elements of artificial senses [Tixier et al., 2023; Piłat-Rożek et al., 2023; Wang et al., 2023].

Bioindication constitutes a method of studying the impact of pollution on the environment, which involves observing the presence of representatives of selected species or communities of specific species of organisms and using them as bioindicators, i.e. biological indicators of quality. Changes in the abundance of organisms of the aquatic environment, allow assessment of water quality. This way of conducting research enables to determine the combined effect of all pollutants, considering their simultaneous occurrence in the environment, on living organisms. The use of a monitoring system using bioindication method, also enables to determine the dynamics and structure of a selected ecosystem responding to changing environmental conditions, caused, for example, by the impact of urban infrastructure systems. Thus, a comprehensive picture of changes in water quality is obtained over a longer period of time, by checking the impact of pollutants on the studied organisms and their communities [Starmach et al., 1976; Łagód, 2017]. Thus, bioindication is a method of biological environmental monitoring, and its purpose is to control as well as measure the changes that occur in the natural environment due to anthropogenic pressure [Dynowska, 2013]. Biological indicators of water quality, i.e. bioindicators, are all aquatic organisms (plant as well as animal), the presence of which gives information about the quality of

the studied ecosystem. Indicator organisms should meet the following criteria: long life cycle, narrow and specific range of requirements, wide occurrence, and characteristic appearance [Gorzel and Kornijów, 2004].

Observations of communities of organisms can be the basis for assessing the impact of pollutants entering with precipitation on receiving waters. Stability of indicators of selected groups of organisms is one way of biomonitoring and collecting information on the aquatic environment in a selected area. Organisms can be identified during microscopic observation of samples of biological material collected from selected points. During further analysis of the results, the organisms can be presented in the form of fractional shares, and then biocenotic indices can be calculated, including: Shannon index, MacArthur's index, and changes in these indices occurring during the study at a given point or at subsequent points of the site at the same time can be observed. The use of biocenotic indices based on the abundance of appropriately selected groups of organisms allows, in a relatively short period of time, to characterize biocenoses as well as their interrelationships and relationships within them. The Shannon diversity index takes into account the number of individuals as well as the proportion of given individuals in a group. It determines the probability that two individuals drawn from a sample will belong to different species [Łagód, 2017; Głowacki, 2013]. The calculated Shannon index is the basis for calculating the MacArthur index, which expresses the richness of taxa in the community for which the observed H value is equal to H_{\max} [Głowacki, 2013; Jost, 2006]. The employed indices are tailored to communities in which the abundance of individuals changes and reflect situations that arise in the natural environment, such as surface waters stressed by point source discharges of pollutants.

The aim of the study was to check the impact of storm sewers on the receiver – the Bystrzyca River, based on bioindication studies of biofilm samples using classical biocenotic indices.

MATERIALS AND METHODS

The material for study was taken from the Bystrzyca River, the largest river flowing through the city of Lublin. It is a tributary of the Wieprz River, and begins its course in Sulowo, where

its source is located. In its upper reaches it is fed only by small streams. Within the city of Lublin, it has 3 tributaries, namely: the Czechówka, Czerniejówka and Krężniczanka. The total length of the river is 74 km, of which 22.5 km flows through the city of Lublin. Lublin is also the only large urban area through which it flows [Misztal et al., 1996]. The wastewater flowing to the receiver comes from stormwater systems and treated domestic sewage, which is discharged from the “Hajdów” Wastewater Treatment Plant. The samples for the study were taken in the section of the river between Zemborzycki Lake and the wastewater treatment plant, which receives the stormwater collected from ul. Muzyczna catchment (Fig. 1) [Kalinowska et al., 2013].

Along the course of the Bystrzyca River, five measurement points were located at an equal distance from the bank. The first selected point was located at a distance of about 15 meters in front of the stormwater drain discharge. Point 2 was located directly in the stormwater drain discharge from the main conduit located in ul. Muzyczna. Point 3 was located downstream of the tributary. Point 4 was located at the same distance. Point 5 was located at a considerable distance, about 50 m, from Point 2. The sampling points were selected to include the locations of expected microorganism diversity, but also changes in water quality. Location of biological film sampling points is shown on map in Figure 1.

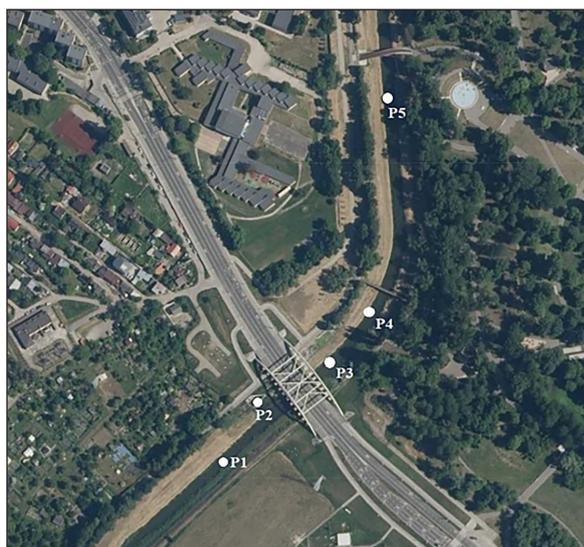


Figure 1. The locations of the biofilm sampling points were marked in white, the order of collection corresponds to the numbers. Own elaboration. Satellite photo obtained from geoportal.gov.pl

The study was carried out on the basis of samples of biofilm, formed on a solid substrate, (microscope slides) taken from the five aforementioned measuring points with specially designed devices, installed at the same depth below the water surface in the watercourse, taking care to ensure that the amount of above-water vegetation was similar. Efforts were made to ensure that conditions were constant and the same, at all points, in terms of insolation and depression below the water surface, as well as hydraulic conditions. Water temperature, air temperature and atmospheric conditions were recorded at the time of sampling. The slides, along with the biofilm covering them, were transported to the laboratory in clean containers filled with water drawn from the sampling point; then, they were placed under an optical microscope. Observations of biofilm-forming microorganisms were carried out under transmitted light and bright field of view, taking digital images for each field. The samples were examined in the laboratory of the Faculty of Environmental Engineering at Lublin University of Technology on the day they were collected, so that the time between sampling and observation under the microscope did not affect the quality of the biological membrane. During the study period from 2020-10-31 to 2021-08-13, sampling was carried out 20 times at five points. During the microscope observation, about 50 fields of view were taken at each point, which translated into a total of about 5500 digital images. As a result, 33917 individuals of selected species were observed.

In the digital images taken, representatives of 12 species of algae were sought: *Achnanthes lanceolata*, *Cyclotella comta*, *Microspora amoena*, *Nitzschia acicularis*, *Pediastrum duplex*, *Pinnularia microstauron*, *Rhoicosphenia curvata*, *Synedra acus*, *Synedra ulna*, *Scenedesmus quadricauda*, *Tabellaria flocculosa*, and *Ulothrix tenuissima*. Therefore, each observation (for each date and sampling point) had information on the abundance of representatives of each of the listed species, which accounted for 12 explanatory variables.

The number of organisms at each point, for selected days, was used to calculate biocenotic indices: the Shannon index and the MacArthur's index [Chomczyńska et al., 2009; Łagód et al., 2009; Montusiewicz et al., 2007].

The results of the study were compiled using indices, selected based on the literature:

- species richness:

$$S = \sum_{i=1}^{S^*} s_i \quad (1)$$

where: s_i – considered groups of organisms.

- Shannon index:

$$H = - \sum_{i=1}^{S^*} (\Pi_i \cdot \log_z \Pi_i) \quad (2)$$

where: S^* – the number of species;

Π_i – the fractional share of individuals counted in each species, which is the quotient of individuals in the i -th species (n_i), and the sum of individuals in all analyzed species (N);
 z – the base of logarithm.

- MacArthur’s index:

$$E = z^H \quad (3)$$

where: H – Shannon index;

z – the base of the logarithm used in Shannon index.

For abovementioned indices the binary logarithm was used i.e. $z = 2$.

Due to the fact that with the help of these indices it was possible to obtain relevant information from the original dataset, such a procedure can be treated as a non-classical approach to reducing the dimensionality of the dataset from 12 to 1 dimension.

The coefficient of variation is a measure of the variation in the distribution of the variable under study. Its estimator is calculated as:

$$\hat{c}_v = \frac{s}{\bar{x}} \quad (4)$$

where: s – the estimator of the standard deviation, while \bar{x} – the sample mean.

It was applied to compare the variation in the index values obtained at each of the biofilm measurement points.

The values of the above-mentioned indices for all measurement days during the course of the study, from all points of collection of biofilm, are shown in boxplots.

A boxplot chart shows the range of values of the described data, their skewness, as well as intergroup differences [Wilcox, 2009; Nuzzo, 2016]. In a boxplot chart, there is a frame covering the middle 50% of the values (interquartile range),

two vertical whiskers, and points that are outliers. The bottom edge of the box shows the value of the first quartile (Q_1), which is the value that separates the bottom 25% of data values from the top 75% when the data is sorted in ascending order. The horizontal line in the box is on the median value which shows the middle value in a row of ordered values. Whereas the top edge of the box shows the value of the third quartile (Q_3), which is separating the bottom 75% of values from the top 25% of the ascending sorted data [Dekking, 2005]. In the R programming language, the length of the whiskers departing from the frame is by default based on the length of the interquartile range. Namely, the lower whisker starts at a value equal to $\max\{\min(x), Q_1 - 1.5 \cdot (Q_3 - Q_1)\}$, while the upper whisker ends at a value equal to $\min\{\max(x), Q_1 + 1.5 \cdot (Q_3 - Q_1)\}$, where x is the variable under consideration. The data values that are outside the range of the box and whiskers are denoted by points above or below the whiskers with dots.

Boxplots were used in this work because they show multiple measures of the distributions of the values shown and allow visual comparison on a single graph of all measurement points while referring to information calculated from all available data.

All visualisations were produced with R packages ggplot2 [Wickham, 2009] and cowplot [Wilke, 2020].

RESULTS AND DISCUSSION

The biofilm fragments taken were analyzed, and the results are presented in graph (Fig. 2). The results for three days are shown, each from a different season (winter, spring, summer). The difference in the value of the species richness between the sampling points is clearly visible. Differences in the value of this index show a sudden decrease at Point 2 – located directly in the storm drain discharge – which indicates the influence of pollutants (heavy metal compounds, total suspended solids, residual petroleum substances) contained in the water supplied to the receiver. At subsequent points downstream of the discharge, there is a return of the indicators to their initial values as well as higher ones.

The Shannon index at selected points is in the range of 0.00 to 0.974 (Fig. 3). The MacArthur index ranges in values from 1.00 to 1.965. Then,

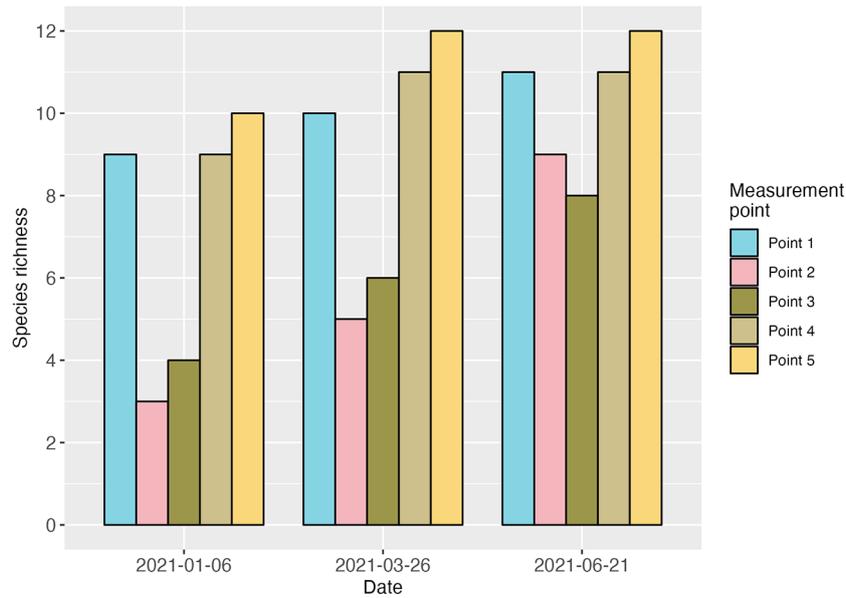


Figure 2. Species richness of organism communities on 2021-01-06, 2021-03-06 and 2021-06-21 in all measurement points

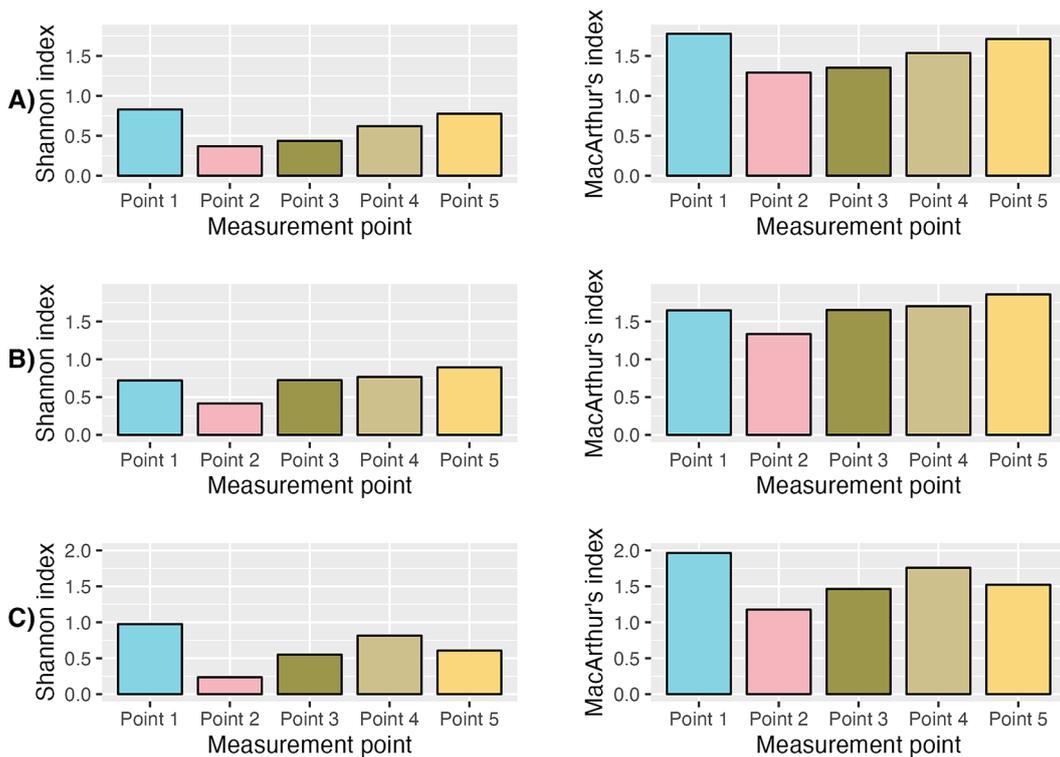


Figure 3. Biocenotic indices based on the abundance of microorganisms on (a) 2021-01-06, (b) 2021-03-06, (c) 2021-06-21

from Point 3 onward, the index values gradually return to the value observed at Point 1 or significantly exceed it. The return of the values of the Shannon and MacArthur indices to those found at the reference point, before the point pollution, and their exceedances, indicate the ability of the river to self-purify.

There are several works assessing water quality [Jaromin et al. 2012; Grzywna et al. 2014]. Similar results but relating to the saprobity of the river were observed in the work by R. Babko et al. [Babko et al., 2019] where it was found that water flowing into the river through the storm drain system, causes turbidity and increased suspended

solids in the receiving waters, and clearly affects the structure of algal communities found in the waters of the Bystrzyca River.

In the study of Babko et al. (2019), the value of saprobity changed with the points: the lowest was for the reference point (before the catchment) where it was 1.71, at the discharge point it increased to a value of 2.09, and then the value of this index decreased approaching the initial value at the reference point. Another work by R. Babko [Babko et al., 2020] presented differences in the value of the Shannon index between different points indicating changes in algal assemblages caused by inflow from storm sewers. In the work of Babko et al., studies were conducted on the same section of the Bystrzyca River, for the same group of distinguished indicator organisms. The difference with the presented study lies in the number of points subjected to analysis, which in the afore-mentioned works was 3 points, while in the presented study it was extended to 5 measurement points, according to the course of the river, in order to check the length of the range of impact of the storm sewer discharge. The assessment of the impact of the storm drain system in the present work was also based on additional indices: MacArthur's, species richness, and the coefficient of variation for these indices was calculated as well.

Grzywna studied water quality indicators, including the oxygen conditions of BOD₅ and COD in the catchment area of the Ochozanka River, the waters of which are polluted with wastewater of agricultural origin. His study shows that flowing

surface waters were characterized by COD values in the range of 38–189 mgO₂/l and BOD₅ values in the range of 10–45, and these values are similar to those presented by R. Babko and, as in the present work, it follows that waters collecting surface pollutants, discharged into a receiving body, affect its quality [Babko et al., 2019, Grzywna, 2014].

In this paper, of all the days of the survey, the results of those for which the trend was most visible are presented; at the other days the trend persisted, although it was less noticeable. This is most likely due to the fact that the value of indicators is affected by air temperature, water temperature, the amount of rainfall, while with large fluctuations in the parameters of atmospheric conditions, the trend may be less pronounced.

When analyzing the mean and median values that are shown on the boxplots (Figures 4–6) (mean values are at the height of star markers and the median values are on the black, thick lines on all boxplots) on all the days of experiment, it was found that the Shannon index reaches its lowest values at the point of discharge. This is also noticeable in the mean and median graphs showing species richness. In the boxplots for Shannon and MacArthur indices, it can also be seen that the interquartile range (the box, see description of boxplots in Materials and Methods) of Point 2 as a whole is below the range for Point 3. Thus, the return to the state of the higher values of these indices is noticeable in the distribution of these values, not just for their mean and median.

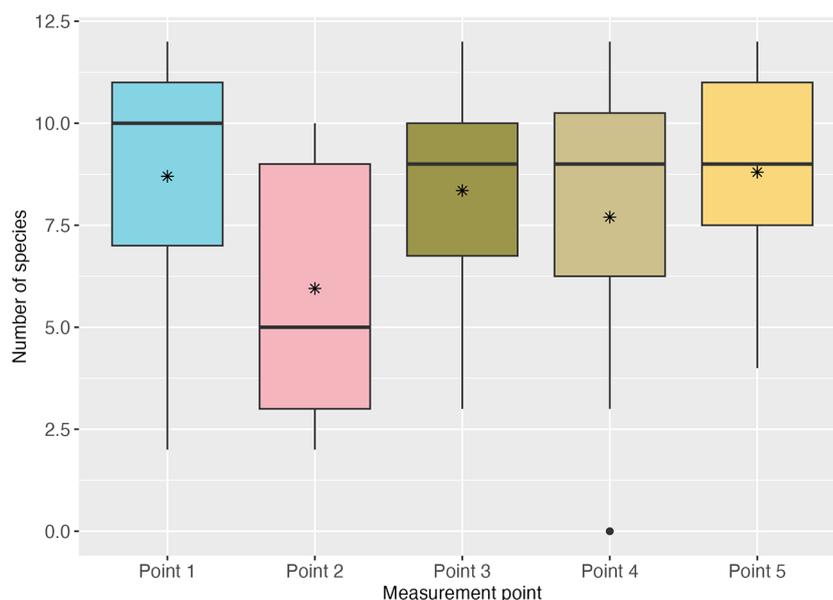


Figure 4. Boxplots of the number of species in measurement points: * – mean value, black line – median value

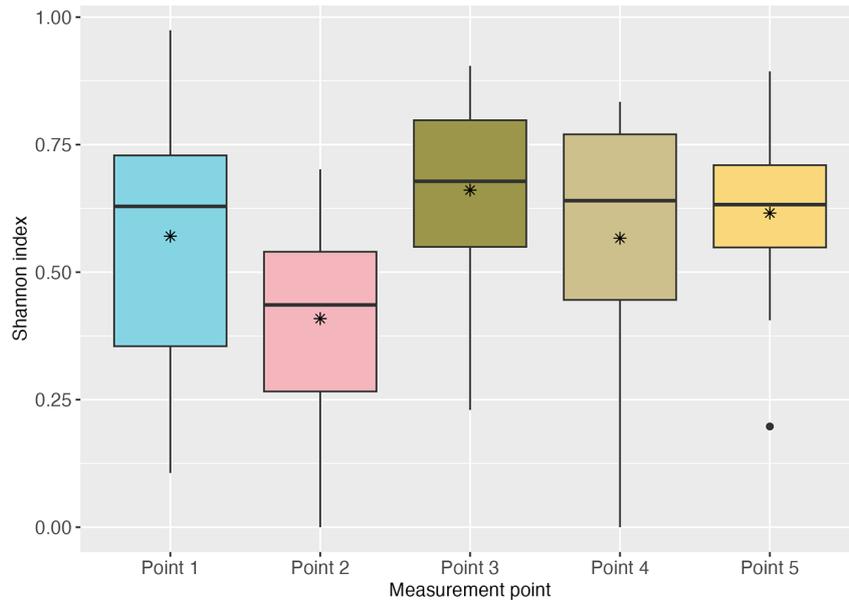


Figure 5. Boxplots of the Shannon index in measurement points: * – mean value, black line – median value

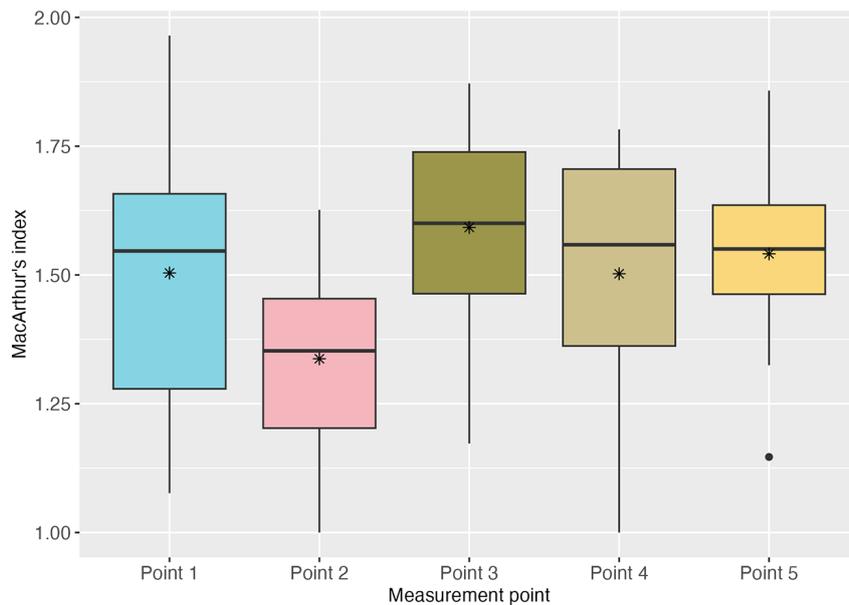


Figure 6. Boxplots of the MacArthur's index in measurement points: * – mean value, black line – median value

The MacArthur index confirms the differences between the points and, consequently, also the correctness of the previous graphs; this in turn indicates that the points for the study were selected correctly. In the graph of Figure 4, it can also be seen that there are no notable differences between the values of species richness from Points 3, 4 and 5, their boxplots being of similar length, mean and median at similar levels. The situation changes in Figure 5 and Figure 6, as the singular box and whiskers for Point 5 are much shorter than those of Points 3 and 4. Thus, the values of Shannon and MacArthur indices

have much less variation in the last point, except for the one outlier observation observed in the graph of their values. This is because species richness is an integer, while the indexes in question are based on logarithms of probabilities of occurrence for individual species. The matching shapes of the boxplots and bar charts for the Shannon and MacArthur indices are due to the fact that the MacArthur's index is an exponential transformation of the Shannon index value.

In addition, an analysis of the coefficient of variation calculated for each sampling point showed that in the case of species richness, the

highest coefficient with a value of 0.49 was achieved at Point 2. In the case of the Shannon index and MacArthur index, the highest coefficient of variation was achieved for Point 4, equal to 0.46 and 0.16. Point 3 is located next to the discharge of drainage water and stormwater drains from the Ludowy Park, which affects the water quality and microbial communities found in Point 4; this is related to the direction of flow and the way the river's water strata are mixed.

Many methods can be used to determine the structure of communities, and it is possible to determine the structure in several ways, based on any measurable parameter that will characterize the analyzed community. The parameters taken into account should be described by numerical values that can be related to the considered characteristic of the community. The use of indicators and biocenotic indices to analyze the structure of appropriately selected algal communities allows characterizing the biocenoses and the relationships occurring in them in a relatively short period of time. Measures such as the Shannon index, the MacArthur's index, do not use complex formulas, and are based on relatively simple assumptions. An important feature of the aforementioned measures is that they can be used in determining the quality of many environments, not just the aquatic environment.

CONCLUSIONS

Studies conducted on the basis of biofilm samples collected from five measurement points located in the Bystrzyca River showed that the most common and abundant species included: *Cyclotella Comta*, *Pinnularia Microstauron*, and *Nitzschia Acicularis*.

The impact of stormwater discharge is evident at all points, taking into account observations of the values of species richness, Shannon biodiversity, and the MacArthur's index. Analysis of all test days allows concluding that the most noticeable changes in biological material occur at Point 2 – the site of stormwater discharge. At successive sampling points, there was a gradual return of indicator values to the level at the reference point. The apparent reductions in indicator values at Point 2 are indicative of the impact of the storm drain system on the receiver, pointing to the need for constant monitoring of storm drain discharges.

Analyzing the values of species richness and Shannon or MacArthur index on the basis of box-plots leads to more in-depth observations than species richness alone, as changes in the variation of the values of the considered indexes at different sampling points can be observed on them.

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