Can waters from woodland areas be of poor quality? The problem of sanitary contamination of Białowieża Primeval Forest watercourses

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Abstract. Studies on the microbiological quality of selected watercourses in the Białowieski Primeval Forest were assumed (Narewka, Orłówka, Hwoźna, Braszcza, Łutownia, Przedzielnia). In samples of water taken in the years 2010–2015, the numbers of psychrophilic, coliform and *Escherichia coli* were determined. High organic contamination of all of the studied watercourses, typical of woodland and wetland areas, was confirmed. Increased microbiological contamination was indicated only for the Orłówka and Braszcza rivers, which suggests an inflow of contaminants from areas inhabited by wild animals and water-logged areas. The poor sanitary quality of waters from the Hwoźna River is most likely connected with the cross-border inflow of contaminants.

Key words: sanitary contaminants, Escherichia coli, freshwater quality, woodland.

1. Introduction

The quality of waters influences the biological variability and functioning of ecosystems. This fact is indisputable and, as of now, has been widely discussed. The excess of chemical contamination leads to changes in the intensity of ecological processes, which may sometimes lead to the significant degradation of the ecosystem. Is this similar in the case of contamination of a biological nature? Usually, attention to this group of contaminants is not given until determining the functional properties of waters. The issue pertains to, above all, microbiological contamination. Those considered most important from a health-oriented and legal point of view are described as of anthropogenic origin (Bielecka et al., 2006; Skorbiłowicz et al., 2008). Thus, waters in areas inhabited by people are usually characterized by the poorest microbiological quality. Forested areas, on the other hand, are considered clean, and their

waters as not containing harmful substances. It turns out, however, that contaminants influencing the periodic degradation of water quality are identified even in areas within forests (Miniuk, 1998; Pierzgalski et al., 2010; Baille & Neary, 2015; Schelker, 2013).

Among the biological factors, *Escherichia coli* are of key importance when assessing the quality of waters. It is a typical indicator of contamination, additionally helpful when identifying its source. The natural habitats of these bacteria are human and animal bodies, where their population reaches as many as a few million cells in 1g of faeces (Weaver et al., 2005). However, their presence is also registered in the natural water environment. Indicating the presence of *Escherichia coli* in surface ecosystems in the amount of approximately 10 cells in 1ml of water signifies the inflow of faecal contaminants, usually from domestic sewage (Frak & Jankiewicz, 2013). It is, therefore, obvious that *E. coli* will be present in waters from areas inhabited by people, especially in the neighbourhood of sewage treatment plants or places where livestock is raised. Is it, thus, possible for *Escherichia coli* to be aboundant in water ecosystems in woodland areas, especially those under environmental protection?

The aim of the study was to determine the level of sanitary contamination of watercourses in woodland areas, along with indicating their potential sources. The research were carried out in the area of Białowieża Primeval Forest, covering areas of varied levels of anthropogenic pressure. The potential threat to the quality of waters from forest areas as well as the influence of anthropogenic activity on the quality of water in the analysed watercourses were studied. The obtained results may be useful useful in planning the protection of valuable water and water-dependant ecosystems.

2. Study area

The study area situated in the area of the Białowieża Primeval Forest, located in the north-eastern region of Poland (so-called Podlasie Centralne). The area is of a typical forest character: it covers areas of species and habitat protection (National Park as well as core area of UNESCO Biosphere Reserve), including those under strict protection (not subjected to anthropogenic activity – reserves). Also present are numerous aquatic and wetland ecosystems, dependent on the amounts and quality of incoming waters (Pierzgalski et al., 2010; Bajkiewicz-Grabowska & Karczewska, 2012; Malzan et al., 2014). In the area of the Białowieża Primeval Forest, outside of the area of the National Park, human settlements (number of inhabitants: from 112 people (Budy village) to 2670 (Białowieża town)) which may influence the state of the forest water ecosystems are found (Fig. 1).

3. Material and methods

Water samples for studies were taken in the years 2010–2015, in July and September. The following watercourses in the area of the Białowieża Primeval Forest were covered by the study (Fig. 1):

- In the area of NP: Narewka River (Points No. 1, 2, 10), Orłówka (No. 4a and 4b), Hwoźna (No. 5, 6), Przedzielna (No. 7) and Braszcza (No. 8);
- (2) In areas of strict protection: Orłówka (No. 4a and 4b), Hwoźna (No. 5 and 6);
- (3) Areas outside of PN: Łutownia (No. 3) catchment with 92% forest cover, Narewka (No. 2, buffer zone of NP – catchment in the study area with a forest cover of 98%).

The samples were collected in accordance with ISO 5667, and next subject to laboratory analysis. The level of bacteriological contamination was assessed using standard culture methods. The numbers of psychrophilic bacteria were determined according to PN-EN ISO 6222:2002 (results given in CFU per 1 ml of water). The amounts

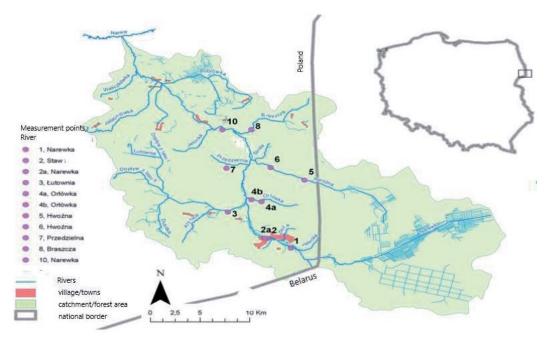


Figure 1. Study area – measurement points (water sampling points)

of total Coliform as well as *Escherichia coli* were determined according to ISO 9308, in respective temperatures of 37 and 44°C (results given in MPN per 1 cm³ water).

4. Results and Discussion

The assumed studies revealed strong microbiological contamination of all analysed watercourses. It was shown that the waters contain numerous psychrophilic bacteria (Fig. 2): the highest numbers were determined for the Braszcza River (sample No. 8) (up to 8000 CFU/1cm³), while the lowest for Narewka (No. 2 - from 1200 to 2500 CFU/1cm³). The most bacteria in the rover usually originated from outside of the ecosystem (so-called allochtonic bacteria) and multiply in favourable environmental conditions (e.g. temperature, energy source). The population of psychrophilic bacteria is dependent on the amount of organic substances present in water, found in the form of agglomerates (suspensions) or dissolved in water (so-called: DOC): along with an increase in the concentration of organic matter, the population of bacteria increases (Adame et al., 2012; Frak & Jankiewicz, 2013). This increase, however, is dependent on the season of the year and temperature of the water: the highest number counts of bacteria are confirmed in the warm months. The shown populations of psychrophilic bacteria therefore suggest high contamination with organic substances, most likely soil-borne. In forest soils, organic matter is derived mainly from plant droppings (remains of forest undergrowth, leaves, branches, etc.). Its amount is determined to be as much as 138 Mg on the surface of 1 ha (it can contain over 20% of organic carbon) (Bajkiewicz-Grabowska & Karczewska, 2012; Schelker, 2013; Baille & Neary, 2015). The inflow of organic matter is therefore

possible, especially in the period of rainfalls and along with surface runoff. As given by e.g. Miniuk (1998), Bielecka (2006), Pierzgalski (Pierzgalski et al., 2010) and Janek (2016), the watercourses of Białowieża Primeval Forest contain high amounts of organic carbon, which leads to the classification of their quality as inadequate. Over 20 mg C/dm³ (DOC), and sometimes even as much as 65 mg C/ dm³, were confirmed in half of the analysed water samples (Skorbiłowicz et al., 2008; Janek, 2016). The high numbers of psychrophilic bacteria in the waters of Białowieża Forest therefore confirm their high organic contamination. The highest values were shown for Braszcza and Orłówka rivers, which are small watercourses flowing through wetlands. A significant source of organic carbon in surface waters are hydrogenic soils, the share of which in the area of the Białowieża Primeval Forest is estimated to be around 15-20% (Bajkiewicz-Grabowska & Karczewska, 2012; Malzahn et al., 2014). The impact of wetland areas on watercourses of the Forest is confirmed by the shown domination of organic nitrogen (Skorbiłowicz et al., 2008; Janek, 2016), as well as ammoniacal nitrogen (Miniuk, 1998; Bielecka et al., 2006) among the contaminants. Increased numbers of psychrophilic bacteria were also stated confirmed periodically for Łutownia, which confirms its average organic contamination (35-50 mg DOC/dm³, according to Janek, 2016).

The presence of *Escherichia coli* was also noted in the analysed watercourses (Fig. 3). In the case of the Narewka (pts. No. 1 and 2), Przedzielna, Łutownia and Braszcza rivers, low populations were usually confirmed, up to approx. 5 MPN/1cm³ water, indicating average faecal contamination and not a reason for concern. These are populations usually found in the summer period in surface waters, not limiting their utility properties (Frąk & Jankiewicz, 2013).

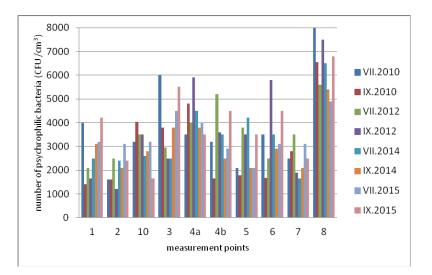


Figure 2. Number of psychrophilic bacteria in measurement points

However, in the case of Orłówka, Braszca, Narewka (Pt. No. 10) and Hwoźna rivers, higher populations (i.e. over 7 MPN/cm³) were registered more often, confirming an increased inflow of contaminants of a faecal origin. The indication of high amounts of *Escherichia coli* was surprising especially for the rivers Orłówka and Hwoźna, due to the specifics of their catchment: both rivers are found in an area under strict protection, where the presence of people is strongly limited (Miniuk, 1998; Bielecka et al., 2008). Increased faecal contamination of the Orłówka River (7 NPL/cm³) is, therefore, connected with runoff from the area of numerous woodland animal sanctuaries (strict nature reserves), as well as the accumulation of these contaminants in water-logged areas (similarly to the case of the Braszcza River) (Pierzgalski et al., 2010).

The high bacteriological contamination of the Hwoźna River is probably connected with the inflow of contaminants of unknown origin from outside the borders of Poland (Pierzgalski et al., 2010). In the area of Belarus, the Hwoźna River flows through grasslands, which may be fertilized organically - high sanitary contamination, in this case, is confirmed only in the spring and summer period (Frak, own studies). The Voivodeship Inspectorate of Environmental Protection in Białystok carried out, in the early spring of 2014 (WIOS, 2015), random studies of the Belarussian part of the waters of Hwoźna River: at the time, low biodegradable contamination (as BOD 2.8 mg O₂/dm³) and total phosphorus (0.42 mg P/dm³) was confirmed. This highlights instead of signifies contamination of waters of the Hwoźna river flowing into the study area. The cross-border influence on the quality of waters in the Hwoźna river is also mentioned by Bajkiewicz-Grabowska and Karczewska (Bajkiewicz-Grabowska & Karczewska, 2012).

The periodical abundant presence of *Eschericia coli* (up to 24MPN/cm³) in waters of the Narewka (Point No. 10) and Łutownia river suggests the possibility of their contamination by the inflow of domestic sewage. This is highly probable due to the close proximity of commercial and industrial areas (Gruszki, Budy, and Teremiski villages). Moreover, studies of waters of the Narewka river, along its entire length, showed regular contamination (Miniuk, 1998; Bielecka et al., 2006; Skorbiłowicz et al., 2008), including the sanitary one (Frąk, own studies), in the area outside of woodland areas (i.e. below the town of Narewka).

Moreover, in order to specify the influence of the catchment on the quality of waters in the Białowieża Primeval Forest, the total number of coliforms was tested (Fig. 4). This group of bacteria includes not only bacteria of faecal origin, but also species commonly found in the natural environment, e.g. Klebsiella aerogenes (formerly Aeromonas aerogenes). Indicating their presence in the analysed samples at a temperature of 37°C, we can obtain falsified results: the marked population may also include bacteria of a soil origin, the occurrence of which does not signify faecal contamination. Which source of contamination is dominant (surface runoff or the inflow of sewage) can therefore be determined by observing the difference in the obtained results (total coliform and faecal coliform) (Frąk & Jankiewicz, 2013). In our study, a constant high count of Total coliform could be unanimously confirmed for the waters of Orłówka, Hwoźna and Braszcza rivers (over 24 MPN/cm³). In the waters of Orłówka and Braszcza rivers, on the other hand, the number of Escherichia coli was determined at 7 MPN/cm3 - thus it is likely that the microbiological contamination is caused by the inflow of soil particles (Adame et al., 2012). The conclu-

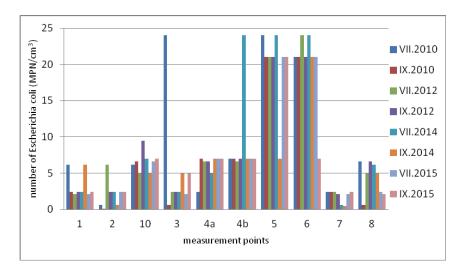


Figure 3. Number of Escherichia coli in measurement points

sion coincides with that suggested above (see: the number of psychrophilic bacteria), regarding the high contamination of soil origin in waters of the investigated rivers. Both indicators, therefore, unanimously indicate the strong influence of the forest catchment on the quality of waters in woodland area.

In the case of the Hwoźna River, on the other hand, a high number of total coliform and faecal coliform (24MPN/cm³) was constantly confirmed (Frąk, own studies). The results indicate that the low quality of river water is, therefore, caused not only by the inflow of contaminants of an anthropogenic nature from the outside of Poland (as mentioned above: high COD; WIOŚ, 2015). The inflow of substances from woodland areas also has a significant influence on the quality of waters in the river (Bielecka et al., 2006).

5. Conslusions

The presence of allochtonic microbiological contaminants was indicated in the watercourses of the Białowieża Forest: bacteria of a natural origin, as well as (periodically) anthropogenic origin were confirmed. Depending on the present climatic and hydrological conditions, and the quality of the entire catchment area, the contamination of the waters can be very high (Adame et al., 2012; Schelker, 2013; Baillie & Neary, 2015). Thus, the sanitary threat of water ecosystems can be significant – their insufficient/low quality may, therefore, influence the health condition of animals using them. However, the observed biological diversity in the Białowieża Primeval Forest (Bielecka et al., 2006; Bajkiewicz-Grabowska & Karczewska, 2012; Kujawa et al., 2016), is not directly threatened by a high number

of bacteria in the water. The analysis of the obtained results and available scientific findings indicates the accurateness of the above conclusions also for waters of other woodland areas. Therefore, it seems as though the problem (Pierzgalski et al., 2010; Schelker, 2013; Alonso & Hardtle, 2015; Baillie & Neary, 2015; Janek, 2016) of insufficient water quality of waters in woodland areas ought to be expanded to include studies of their microbiological contamination. This will make it possible to specify the degree of risk posed to forest ecosystems, including environmentally valuable areas.

What is more, high organic contamination of watercourses in Białowieża Forest, typical of watercourses flowing through woodland and wetland areas, was found. The assumption regarding the cross-border threat to waters in the Białowieża Forest (Hwoźna River) was also confirmed. Particular attention ought to therefore be drawn to international collaboration in the protection of water resources. WIOŚ also suggests the cross-border influence on the quality of waters in the Narewka River – however the studies presented in the present work do not support these assumptions (in the scope of sanitary contamination).

The obtained results suggest the need for the constant monitoring of watercourses in the area of the Białowieża Primeval Forest. Since the year 1990, the level of contamination of waters has been continuously decreasing (Pierzgalski et al., 2010; Janek, 2016), as influenced by the implementation of proper sewage management in neighbouring areas. However, the sanitary condition of watercourses is degraded by runoff from areas which are home to large populations of wild animals. Determining the genetic affiliation of isolated strains of *Escherichia coli* may aid the specific determination of the sources of origin of the contamination.

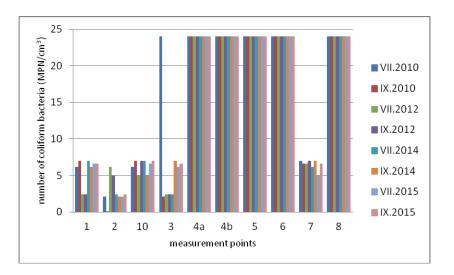


Figure 4. Number of coliform bacteria in measurement points

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