THE SEASONAL QUANTITATIVE DISTRIBUTION OF COLIFORM GERMS IN THE ARIEŞ RIVER (ROMANIA) WATER AFFECTED BY POLLUTION

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Abstract. Faecal pollution of river water can lead to health problems because of the presence of infectious microorganisms. These may be derived from human sewage or animal sources. Water safety or quality is best described by a combination of sanitary inspection and microbial water quality assessment. This approach provides data on possible sources of pollution in Arieş river water, as well as numerical information on the actual level of faecal pollution according with the analzyed bacteriological parameters.

Keywords: river, water, faecal pollution, hygienic bacteria

INTRODUCTION

Rivers represent easily pollutable environments, as various types of waste water – resulted from household activity, industry, agriculture, pesticides, inorganic fertilizers, all of may contain infectious agents – that are discharged into them [1, 25]. A however small, either quantitative or qualitative modification in a microbial population makes possible both the ecological state of an aquatic medium be determined and the identification of the possible source of pollution be achieved. The etiologic agents of the majority of water-transmitted illnesses are microorganism eliminated from the digestive tract [22].

Detailed knowledge of the extent of faecal pollution in aquatic environment is crucial for watershed management activities in order to maintain safe waters for recreational and economic purposes [20]. Techniques which enable rapid and sensitive detection of faecal pollution in environmental freshwaters are thus prerequisite for efficient water quality monitoring.

The sanitary quality of water is appreciated by the presence or absence of pathogen microorganism or of those which indicate the possibility of their presence. Given this, the following bacteriological parameters were analyzed in order to establish the nature of Arieş river's faecal pollution: probable number of total coliforms (PNTC), probable number of faecal coliforms (PNFC) and probable number of faecal enterococi (PNFE).

At sediment level, bacteriological parameters are more constant and more stable, being less influenced by alterations in the environment. Thus, in time, they may be considered to reflect the evolution of water quality. From this point of view, these indicators can be used as an appreciation criterion as well as an instrument for the prognosis of water quality evolution. Moreover, they may function as a decisive factor in what the measures for the rehabilitation of aquatic ecosystems are concerned [3, 16].

Despite the traditional use of fecal coliforms as fecal contamination indicator, numerous recent studies suggest that fecal coliforms are more susceptible to a variety of geographical, physico-chemical factors and/or stresses [12, 17, 23, 27, 29].

The aim of our study is to determine the degree to which hygienic bacteria – indicators of faecal pollution – are present in the waters of Arieş river. To this aim, three groups of indicator bacteria were studied: total coliforms (TC), faecal coliforms (FC) and faecal enterococi (FE).

Pollution sources of the Arieş River are various, starting with mining activity, house holding activity, as well as the anthropic, uncontrolled factors etc. The predominant heavy metal sources in Aries River are the acide mine waters drained from tailings and from deposed sediments rich in metals which in certain conditions are easily mobilized.

Heavy metal salts represent a very serious form of pollution for surface waters due to their toxicity and stability. They can induce disorders of the biological balance, with negative consequences over the various uses of water. The degree of pollution is dependent upon the nature of mine waste, the hydrological link between mines and local rivers, and the local physico-chemical environment. Thus, according with in the water of Aries river were detected increased quantities of trace metals (Cu, Zn and Pb) [2, 3, 11, 26]. The values of heavy metal salts` concentration are higher in the upstream sampling points as compared with those registered in the downstream of the river. It can be affirmed that these elevated values are a consequence of the pollutants` accumulation as a result minig activities.

MATERIALS AND METHODS

The water samples necessary for this study were taken seasonally (in January, April, July and October) from 10 sampling points upstream and downstream of the main towns the river passes through (Abrud, Baia de Arieş, Sălciua, Turda and Luncani). The main polluting sources in the area were also taken into consideration when establishing the sampling points. The water samples were drawn from the river bed at 1 m distance from the shore, being stored in glass containers previously sterilized at 180° C, for 60 minutes, according with SR ISO 5667-6/97 [31].

The term "total coliform" designates a group of bacteria belonging to the *Enterobacteriaceae* family, which is characterised by the ability to ferment lactose at 35-37°C, with gas and acid production for 24-48 hours [21]. Their presence in natural environments indicates a recently faecal contamination of a river's waters [6, 14, 20, 23]. The total coliform bacteria number determination is performed through the **MTM** (multiple tubes method) according to STAS 3001-91 [32] on a liquid medium (the presumptive test) and on a **GEAM solid medium** (the confirmation test) [5, 7].

The term “faecal coliforms” is attributed to the bacteria belonging to the *Enterobacteriaceae* family, Gram negative, unsporulated, oxidase-negative, able to develop in the presence of biliar salts and in that of other surface agents, as well as able to ferment the lactose at 44 °C, with acid and gas production [10].

Coliforms bacteria are present in large amounts in the excrements of man and warm-blooded animals, which makes the detection of these germs possible even after considerable dilution. Widespread in nature, they are not considerated to have a direct epidemiological importance in the case of water examination. The methods for detecting coliform bacteria of certain fecal origin have shown that this group of enterobacteriaceae is a better indicator of faecal pollution with intestinal content from warm-blooded animals than the total coliforms method, when considered for the investigation of river pollution [10, 24].

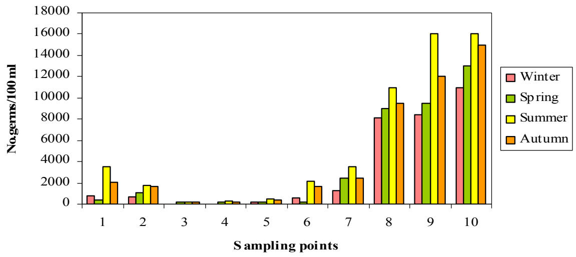
The probable number of faecal coliform and faecal enterococi bacteria is detected through the MTM method [5, 7] and according with STAS 3001-91 [32]. For the detection of faecal coliform (FC) (termotolerants) germ, we cultivated them in the **Mac Conkey medium** (beef extract, lactose, bile), while for the fecal enterococi (FE) we used a **natrium azide medium**, azida I (the prezumtive test) and azida II (the confirmation test) (STAS 3001-91) [5, 7, 32]. The modification of the medium’s colour towards yellow and the presence of sediment on the bottom of the test tube indicate a positive reaction, thus the presence of these bacteria in the tested water samples being asserted.

RESULTS

The bacteriologycal analysis of Arieş river’s water has demonstrated the presence of the total coliforms bacteria with maximum density all along the studied period and in all the sampling points.

The presence of bacteria belonging to the total coliforms group was detected in every sampling point and also in all the seasons. The numerical variation of the total coliform bacteria (TC) is represented in Figure 1.

The numerical fluctuation of the TC germs can be observed, on the one hand, in each of the sampling points, but more acutely from one sampling point to another. The number of TC germs grows in the downstream sections, achieving maximum values at the sampling point downstream of Luncani.



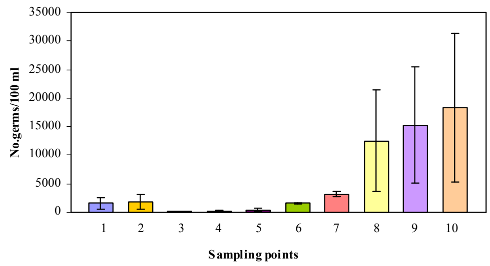


Figure 1. The numeric distribution and annual average values (right) of total coliform density (left) in Arieş river water over 2008 (1 – Abrud upstream; 2 – Abrud downstream; 3 – Baia de Arieş upstream; 4 – Baia de Arieş downstream; 5 – Sălciua upstream; 6 – Sălciua downstream; 7 – Turda upstream; 8 – Turda downstream; 9 – Luncani upstream; 10 – Luncani downstream).

The TC bacteria count registered the lowest values upstream and downstream of Baia de Arieş, as well as upstream of Sălciua in each of the analyzed seasons. The minimum value was registered upstream and downstream of Baia de Arieş in winter, while the maximum value was registered upstream and downstream of Luncani in summer.

Watching the seasonal variations one can be observe that the highest values for this group of faecal contamination indicators were registered in summer, when water temperature grows, causing a subsequent increase in the microorganisms’ development. The values obtained in autumn are the lowest, being, however, relatively close to those obtained in summer, while those corresponding to winter and spring are low. The high values obtained in summer may also be due to uncontrolled tourism and intensive grazing near the water. Similar seasonal variations with higher numerical densities registered in warm seasons were registered by other authors, in estuaries [15] and in Crişul Alb river [9].

The numerical distribution and annual average values of faecal coliform germs densities in the water of Arieş river during the four seasons analyzed in 2008 is graphically represented in Figure 2.

According to the results acquired by water sample analysis, the most probable number of FC bacteria in Arieş river is lower than that of the TC bacteria.

Seasonal variations of the FC germs number were recorded, reaching maximum values during summer (2200 germs/100 ml) downstream of Luncani and minimum ones (<20 germs/100ml) upstream of Baia de Arieş. The count of FC is high at the two extremeties, in the upper and lower sections of the river course, respectively. Upstream of Abrud, the maximum value was registered in summer, being lower (330 germs/100ml) than in the next sampling point – Abrud downstream, in which the maximum value was reached in autumn (720 germs/100ml). The increased number of FC germs in the downstream sampling points may

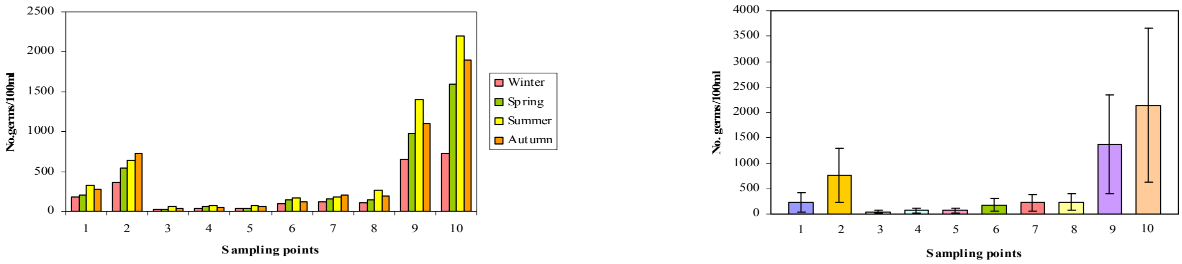


Figure 2. The numeric distribution (left) and annual average values (right) of faecal coliform density in Arieş river water over 2008 (1 – Abrud upstream; 2 – Abrud downstream; 3 – Baia de Arieş upstream; 4 – Baia de Arieş downstream; 5 – Sălciua upstream; 6 – Sălciua downstream; 7 – Turda upstream; 8 – Turda downstream; 9 – Luncani upstream; 10 – Luncani downstream).

have also been determined by inadequately treated human sewage, industrial discharges, the influence of anthropogenic factors and urbanization.

In their study [4] of the anthropic influence on some types of aquatic ecosystems (streams, beck, lakes and its sediments) and on the bacteria considered indicators of faecal pollution, show that the presence of FC bacteria is the consequence of some strong anthropic influences, respectively when water of household origin is discharged into these aquatic habitats, it has a contributory effect on water pollution.

In the P9 and P10 sampling points the high values of FC germs may be due to waters containing high level of domestic faecal pollution, caused, on the one hand, by the deficitary activity of the sewage station serving the Turda and Câmpia Turzii areas, and, on the other, by the growing number of industrial pollutants along the river course, as well as by the uncontrolled river shore grazing in Luncani.

The differences registered between the 10 sampling points are evident in this case, too, the biggest number of registered FC being still P10 in every season.

Watching the seasonal evolution, one can observe that the number of faecal coliform germs has an ascending seasonal evolution from winter to spring, reaches its maximum value in summer, a descendent evolution following in autumn, when water temperature decreases, causing a reduction in the number of coliform germs.

The faecal enterococi (FE) are only present in a few of the sampling points, while in others their presence could not be detected at all (Abrud downstream and Baia de Arieş upstream in winter, spring and autumn; Sălciua upstream in winter, summer and autumn). FE represent the less numerous group of the hygienic bacteria studied. Their numbers present us with only slight variations.

The distribution of the FE germs number is graphically represented in Figure 3.

Figure 3. The numeric distribution (left) and annual average values (right) of faecal enterococi density in Arieş river water over 2008 (1 – Abrud upstream; 2 – Abrud downstream; 3 – Baia de Arieş upstream; 4 – Baia de Arieş downstream; 5 – Sălciua upstream; 6 – Sălciua downstream; 7 – Turda upstream; 8 – Turda downstream; 9 – Luncani upstream; 10 – Luncani downstream).

Analyzing the numeric distribution of this bacteria group, the presence of FE germs in every sampling point on the course of Arieş river can be observed. The FE group registered the highest numbers in the downstream sampling points. For each season the maximum level was registered downstream of Sălciua. The high values which, especially in summer, were registered upstream of Abrud may be due to the washing of sheep that takes place in this part of the river, or even to the sampling point being located nearby some dung dischargements. The minimum value of this group of water faecal pollution indicators was registered in summer downstream of Abrud (18 germs/100ml), while the maximum (320 germs/100ml) – aupstream of Sălciua in summer as well.

In P4, the higher number of FE germs can be explained by the fact that the sampling point was located in an area where elbows were being formed on the river course as a result of rubble excavation, which favourized increased water stagnation as compared to the P3 and P5 sampling points.

When analysing the seasonal evolution we can observe the total absence of FE germs in the P2 and P3 sampling points during winter and spring and in the P4 sampling point during winter, summer and autumn, all indications of only minor faecal pollution of river waters. Seasonal fluctuations were registered in the other sampling points, their maximum values during summer, with lower, but parallel values in autumn, and minimum ones in winter and slightly higher in spring.

The seasonal analysis shows that there is an ascending seasonal evolution in the presumable number of faecal enterococi from winter to spring and summer when, due to higher air and water temperatures, the development of faecal pollution indicator bacteria is stimulated. In autumn, even if the FE number is reduced compared to summer, its level is, nevertheless, still high.

The reduced number of the studied hygienic bacteria in the upstream sampling points (P2, P3, P4) on the Aries river may be also explained as a consequence of the heavily polluted waters in these area with Cu, Pb and Zn [2, 3, 11, 26] which inhibitory effect over the bacterial growth and development.

The quantitative variation of the hygienic bacteria in Arieş river in 2008 is represented in Figure 4. One may observe that the total coliform (TC) germs are present in the highest number (77%) in each of the sampling points and in each of the studied seasons. The TC are followed by the FC (15%), the lowest percentage of all studied faecal pollution indicators being represented by the FE (8%).

As concerns the 2008 seasonal distribution of hygienic bacteria in the river Arieş, the fact that there are great fluctuations in the bacteria number (Fig. 5) was demonstrated, the minimum values being registered during the cold season, while the maximum ones during the warm season. By comparing our data with that from speciality literature, we may affirm the existence of similarities between them [8, 13, 19, 23, 29]. From the analysis of recorded data, the presence of all bacteriological parameters of faecal pollution could be ascertained for each of the studied seasons. Important differences were observed from one season to another and between the sampling points during the same season. Some of the studied hygienic bacteria was undetectable in some of the seasons, generally during the cold ones, while being present in low numbers during the warm ones.

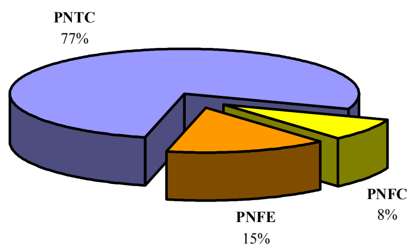


Figure 4. The quantitative variation (%) of hygienic bacteria in Arieş river water in 2008.

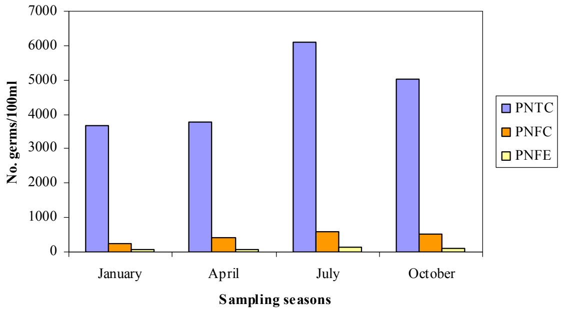


Figure 5. The seasonal distribution of hygienic bacteria medium density in Arieş river water in 2008.

Observing the seasonal distribution, the presence in a high number of TC germs in each season can be recorded, with maximum values in July and minimum values in January.

In the case of FC and FE a similar situation can be observed, with maximum values in July and minimum values in the cold season. The FC number is smaller than that of TC, but at the same time bigger than that of FE, which represent the lowest values among the studied hygienic bacteria and for each of the studied seasons.

DISCUSSIONS

In all the water samples that were analyzed, the presence of faecal pollution indicators (total coliforms, faecal coliforms and faecal enterococi) was detected. All studied groups showed seasonal fluctuations and numerical fluctuations depending on the sampling points. The values of this indicators increase from winter to autumn, the highest values being usually characteristic of summer, when higher water temperatures cause an increase in the microorganisms' development and activity. The lowest number of germs was registered during winter and spring, as the water's temperature was low when the samples were taken. The hygienic bacteriological indicator values are higher in the case of the sampling points that are situated downstream due to the anthropic influence and the effect of urbanization. These values are lower in the P3, P4, P5 sampling points, even undetectable in the case of FE, indicating in some seasons, the existence of less faecal contaminated water.

Among the hygienic bacteriological indicators that were studied, the best represented from a numeric point of view was the TC germs, while the least represented were the FE germs.

Comparing our results with those from the literature [9, 13, 18, 28, 30] we may affirm that, regarding the number of faecal coliform germs, Arieş River water is highly contaminated with human wastes especially in downstream of the river where it is strongly relieved the effect of urbanization. The increased numbers of these bacteria in Arieş river waters represent a serious concern taking into consideration the fact that these germs are considered as opportunist pathogens which may induce serious health affection for the human population. Our results demonstrate once again the necessity of a high depuration of faecal and residual sewage waters that are overflowed in the Arieş River, fact that was evidenced by the presence of pathogenic germs, especially by the high number of detected coliform germs.

According with our studies, we may affirm that the faecal coliform germs in the water of Aries river are strongly influenced by the modification of the chemical composition of the water as a result of the various sources of pollution along of the river course (mining activity in the upstream of the river and a highly contaminated water with human wastes especially in downstream of the river where it is strongly relieved the effect of urbanization).

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