STUDIES CONCERNING SEASON VARIATIONS OF THE FAECALIGENIC POLLUTION IN THE CRIŞUL ALB RIVER

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Summary: The water tests were made in every season at the Crişul Alb River during 3 years of study. The tests were analyzed in the lab in order to determine 3 faecal gene pollution indicators: total coliform bacteria, faecal coliforms and faecal enterococcus. The tests were made in 10 drawing points, located upstream and downstream in the following cities: Brad, Gurahonţ, Buteni, Ineu, Chişineu Criş.

Based on the recorded values, the hygienic and sanitary quality estate of the water has been possible. The main faecal gene pollution sources were found and identified. The nature of this faecal gene pollution was also found and identified based on a report between the faecal coliforms and the faecal enterococcus.

The study made on the Crişul Alb River assured during those 3 years a monitoring of the sanitary quality from the water and the establishment of eventual pollution sources. I hope that the obtained data will catch the attention of the local authorities, economic agents and citizens on the aspects related to the water pollution from the Crişul Alb River. I also hope that this will help them to take measures in order to limit the pollution sources and to use the water with more efficiency.

Keywords: water pollution, faecal colifroms bacteria, monitoring water quality, Crişul Alb River

INTRODUCTION

The bigger and bigger requirements for water resources from all over the world ask for a more efficient quantitative and qualitative use of water. It has been thought about the renewal and the regain of the used water sources. Water pollution is one of the greatest problems of the mankind and the water purification is very important for maintaining a biological balance on the Earth. The life of plants, animals and the entire biological activity of the human kind depend on this balance (Millea, 2001).

The running waters and lakes have water supplies for lots of cities and communities. Unfortunately, these water supplies are submitted more frequently to the actions of pollution sources with domestic and industrial residues. The law no. 107/25.09.2006 concerning the waters from Romania, define the water pollution like this: the water pollution is any kind of physical, biological, chemical or bacteriological pollution of the water, beyond an established admitted limit, even surpassing the normal limit of the radioactivity, directly or indirectly made by human activities.

The water cannot be used in the same way as it was before the pollution. Therefore, the water is considered impure when the usual thermal or biological conditions are changed and enriched with bacteria and other microorganism. The sanitary quality of the water can be appreciated by the presence or absence of the pathogenic microorganism and other microorganism showing their possible presence.

The water containing pathogenic agents jeopardize the life and health of other creatures. The most frequent pathogens met at the level of the surface waters are coming from the intestinal tractus or the urine of infected persons which can accidentally get into the domestic and industrial water sources used for drinking.

The generic term of coliform germs represent a category of bacteria member of the *Enterobacteriaceae* family and includes the following types: *Escherichia, Citrobacter, Klebsiella, Enterobacter, Salmonella* and *Shigella*. From all these, the *E. coli* is considered the most specific indicator for the detection and determination of the fecal water pollution. Therefore the existence of a sanitary risk is determined by the viral and bacterial pathogenic germs (Nestor & Lazăr, 1989).

The usage of the standard methods is suggested for evaluating the number of bacteria from the aquatic environment submitted to lab tests. Modern methods of detecting the fecal water can be used also. Fiksal et al. workers from 1994 suggest the use of enzymologic methods which are faster than the classic microbiological ones for locating the fecal pollution; Pyle et al. workers from 1999 suggest a faster and more efficient method which combine the imunomagnetic separation and the liquid laser citometric; Yu & Bruo in 1996 suggest a fast method (of approximately an hour) which consists of combining the imunomagnetic separation with the electrochemioluminiscent detection of the bacteria; Hagedorn et al. workers from 1999 improve a modern method which detect the fecal pollution sources with the help of a resistance test concerning the antibiotics of the fecal streptococcus.

No matter the used methods for detecting the fecal water pollution it is highly recommended for the methods to be efficient and to take immediate measures in order to limit the extension of this kind of pollution with a negative effect on the life and health of the living organism.

The importance of the monitoring activity from the water quality, comes from the permanent phase of the water resource quality. Based on this data, an efficient strategy of the quality resource is adopted.

The study emphasizes data referred to monitoring the faecal gene pollution along the Crişul Alb River, from the spring to the towns` exit. The data is obtained in the gathering company of the water tests in 2003, 2004, 2005, in the 4 seasons of the year. The determination of the fecal enterococcus and the total

and faecal coliforms are taken into consideration. Establishing the nature of the fecal gene, human and animal pollution source is also considerable.

MATERIALS AND METHODS

The water tests submitted to lab examination were taken from the Crişul Alb River during 3 years of study (in this period, the monitoring activity of the waters` quality has been made in the hydrographic basin) in the 4 seasons of each year, winter, spring, summer and autumn.

The stations were placed near the major localities crossed by the river: Brad, Gurahonţ, Buteni, Ineu, Chişineu-Criş in order to follow the effects of the human pressure on the microorganism from the water.

In order to determine the hygienicall and bacteriological parameters, water samples were submitted to analyzation, the following culture mediums were used: sodium laurilsulphat (simple or double concentrated) and Geam-Levine (total coliform bacteria), MacConkey medium (faecal coliform), simple or double concentrated medium of sodium azide and azide with purple cresol medium (faecal enterococci) (Cuşa, 1996).

The presence of coliform bacteria (total coliform) can be revealed also through the presumption test (presumptive test). First of all we put samples of water in several test tubes with medium and after that the confirmation test is done on a solid medium at a temperature of 37° C and a period of incubation of 24 hours. Starting from the number of tubes with positive reaction we calculate the probable number of coliform bacteria (total coliform). The coliform bacteria (total coliform) are confirmed if characteristic colonies have developed: flat colonies of dark blue-violet with metallic gloss or convex, opaque, mucous with central metallic gloss or pink colored with the centre blue-violet (STAS 3001/1991).

Based on the positive results of the presumption test for the coliform bacteria further on we can establish the presence of the coliform thermotolerant bacteria (faecal coliform). This is done with the help of liquid selective mediums at an incubation temperature of 44° C. After this time depending on the number of positive test tubes the probable number of thermotolerant coliform bacteria (faecal coliform) can be calculated. In order to confirm the thermotolerant coliform bacteria from the same test-tubes considered positive in the presumption test a few droplets are taken with a with Pasteur pipette and passed in test-tubes which contain either Bromocresol purple Lactose Bouillon or Brilliant Green Bile Lactose Bouillon.

The samples are incubated at 44° C for 24 hours. If the color of the medium turns into yellow and simultaneously a gas is released due to the fermentation processes of the lactose is clear that the thermotolerant coliform bacteria are present (faecal coliform).

The streptococci are evidenced also through the presumption test. In this case are used also test tubes with an enriched liquid medium. This medium with the bacteria are incubated at 37° C. The positive reaction is revealed through confirmation test on selective liquid medium incubated for 48 hours at 44° C. Starting from the number of confirmed positive tubes the probable number of streptococci is calculated. After the confirmation of streptococci in liquid medium, one or two drops from each test-tube considered positive in the presumption test are passed with a Pasteur pipette into a test-tube with Bromocresol purple Sodium azide Bouillon. We incubate this at 44.5º C for 24 hours. The turning of the color in yellow with apparition of sediment on the bottom of the test-tube demonstrates the presence of the streptococci in the water (Drăgan-Bularda, 2000).

REZULTS AND DISCUSSIONS

During the year 2003 the approximate number of total and faecal coliforms are relatively low in winter and higher in summer. The minimal value for total coliforms is 60 germs/100 ml. The test is made in winter at the drawing point level Gurahonţ upstream. For 110 germs fecal coliforms/ 100 ml, the tests are made at the drawing point level Brad upstream in the same season. The values of the total and faecal coli form bacteria during the year of 2003 are usually lower than the values from the years to come.

The maximum value of 2600 germs/100 ml tests for total coliforms are made in summer in the drawing point Buteni downstream. For faecal coliforms of 910 germs/100 ml, the tests are made in summer at the drawing point level Gurahonţ downstream. The high number of faecal coliform bacteria can give us information about the presence of a recent faecal pollution in the water mass.

The results confirm our expectations concerning the existence in the area of zootehnical microfarms, near the drawing point, which have insufficient limit systems of the faecal gene pollution. The recorded values can be found in table number 1. On the other hand, the approximate number of faecal enterococcus is very low and at the level of some drawing points and in certain seasons this faecal enterococcus doesn`t even exist. The values recorded during the year of 2003 are lower compared to the values from the following years.

The maximum value of 100 germs/100 ml tests were made at the drawing point Ineu downstream, in summer. Values close to maximum are confirmed in this season, too. In the water tests taken during the year 2004 total and fecal coliforms were found with minimal values in winter at the drawing point level Brad upstream: 220 germs/100 ml for total coliforms and 110 germs/100 ml for faecal coliforms (table 2).

The maximum value of 6600 germs/100 ml for total coliforms is recorded in autumn at the drawing point level Ineu downstream; 3800 germs/100 ml for faecal coliforms in summer at the drawing point level Chişineu Criş downstream. The faecal enterococcus is known by its absence or low number in winter when the water temperature is low compared to the most propitious temperature for their development. The maximum value of 2850 germs/100 ml is recorded at the drawing point level Ineu upstream, in summer.

The high number of faecal enterococcus certifies a recent and massive pollution in the water mass. The bacteriological study made during the year 2005 in the

four seasons at the level of the water tests, established the following individual values for those 3 sanitary and hygienic bacteria groups. The values can be seen in table no.3

The total coliform bacteria has numerical variations during 2005 between 380 germs/100 ml in winter at the drawing point level Buteni downstream and 6410 germs/100 ml in autumn at the drawing point Chişineu Criş downstream. The minimal value from the cold season is due to the low temperature compared to the most propitious temperature for their development. This can also happened because of the accidental overflow of polluted substances, coming from the leather processing companies in Sebiş, located in just a few km from the drawing point.

The faecal coliforms have numerical variations, too: between 200 germs/100 ml in the cold season, at the drawing point Brad upstream and 2700 germs/100 ml at the drawing point Chişineu Criş downstream, in autumn. The approximate number of faecal enterococcus can be zero in winter at almost every drawing point and the maximum value of 2000 germs/100 ml at the drawing point level Gurahonţ downstream in autumn. The high value from autumn can be determined by the growth of organic materia from the water mass, which favors the development of microorganism. Drawing point level, hygienic and sanitary bacteria, winter, spring, summer and autumn.

Analyzing the recorded values for all those 3 groups of hygienic and sanitary bacteria, during 3 years of study we got to the conclusion that the lower value for total coliform bacteria was recorded in 2003 and the highest value in 2004. The fecal coliform bacteria had the lowest value in 2003 and 2004. The highest value was recorded in 2004. The faecal enterococcus was absent at the level of certain drawing points. During those 3 years the highest value was recorded in 2004.

Table 1. The numerical distribution of the bacteria groups made in the Crişul Alb River during the year of 2003

Sampling sites	Bacteria groups	Winter	Spring	Summer	Autumn
Brad upstream	NTC	170	180	1700	1600
	NFC	110	140	300	250
	NFE	0	7	11	5
Brad downstream	NTC	210	290	1900	1850
	NFC	130	170	360	320
	NFE	0	17	30	40
Gurahonţ upstream	NTC	60	790	2200	2100
	NFC	220	240	800	700
	NFE	0	5	15	10
Gurahonţ downstream	NTC	110	880	2350	2500
	NFC	280	340	910	820
	NFE	0	40	90	70
Buteni upstream	NTC	220	810	1100	900
	NFC	210	250	400	300
	NFE	0	15	50	30
Buteni downstream	NTC	310	830	2600	2550
	NFC	170	410	560	380
	NFE	0	0	90	60
Ineu upstream	NTC	180	740	1800	1900
	NFC	220	350	500	490
	NFE	0	0	20	10
Ineu downstream	NTC	220	850	1900	2150
	NFC	210	320	590	520
	NFE	0	47	100	95
Chişineu Criş upstream	NTC	210	770	2300	1800
	NFC	170	280	550	500
	NFE	0	0	20	10
Chişineu Criş downstream	NTC	275	1100	2500	1950
	NFC	220	420	690	640
	NFE	0	56	95	87

NTC – the number of total coliforms (no./100ml), NFC - the number of faecal coliforms (no./100ml), NFE - the number of faecal enterococcus (no./100ml)

Comparing data from the 3 hygienic and sanitary indicators from the Crişul Alb River with data from specialized literature, the conclusion is that we have similar values at the Someş River (Buzan şi Drăgan-Bularda, 2005). The lower values are similar with the ones from the Mureş River (Ştef şi colab., 2005), Bahlu River (Ailiesei şi colab., 2001) and Trebeş (Măzăreanu şi colab., 2001). Related to all the 3 hygienic and sanitary bacteria groups, their high value at the drawing point level can give us information about the presence of a possible pollution source.

A percentage value had total coliform bacteria, the faecal coliforms were next and finally the faecal enterococccus. These were present or absent in a very low number compared to the other 3 studied bacteria groups. In 2003 the percentage was smaller, approximately equal with zero. From the studied data presented in the tables from above, we consider that the lowest values for the studied hygienic and sanitary indicators were recorded, as it should normally be, during the cold season - in winter.

Table 2. The numerical distribution of the bacteria groups made in the Crişul Alb River during the year of 2004

Shampoings sites	Bacteria groups	Winter	Spring	Summer	Autumn
Brad upstream	NTC	220	330	480	460
	NFC	110	180	330	320
	NFE	0	55	91	81
Brad downstream	NTC	400	520	700	680
	NFC	310	470	550	490
	NFE	51	70	190	150
Gurahonț	NTC	1500	1900	2700	2100
upstream	NFC	630	720	900	810
	NFE	30	90	170	120
Gurahonț downstream	NTC	1100	1500	3100	2600
	NFC	450	510	1050	920
	NFE	50	75	1130	1080
Buteni upstream	NTC	1850	2250	3100	2700
	NFC	410	650	1300	950
	NFE	0	81	105	82
Buteni downstream	NTC	560	900	1300	1100
	NFC	390	860	950	910
	NFE	10	30	87	63
Ineu upstream	NTC	2800	3600	4800	4600
	NFC	900	1850	2700	2100
	NFE	20	110	2850	900
Ineu downstream	NTC	3600	4700	5400	6600
	NFC	1700	2100	3700	3300
	NFE	98	180	380	210
Chişineu Criş upstream	NTC	2200	3100	4600	4200
	NFC	870	1600	2100	1800
	NFE	35	120	250	170
Chişineu Criş downstream	NTC	2000	4400	6100	5700
	NFC	1300	2200	3800	3500
	NFE	0	170	420	200

Tabel 3. The numerical distribution of the bacteria groups made in the Crişul Alb River during the year of 2005

Sampling sites	Bacteria groups	Winter	Spring	Summer	Autumn
Brad upstream	NTC	640	800	1000	1100
	NFC	200	400	900	1200
	NFE	0	0	0	0
Brad downstream	NTC	300	1200	1600	1700
	NFC	300	700	900	610
	NFE	0	0	20	55
Gurahonț upstream	NTC	680	1200	1700	2000
	NFC	200	500	1100	900
	NFE	18	36	91	114
Gurahonț downstream	NTC	830	1300	2300	2600
	NFC	500	1100	1700	1350
	NFE	20	97	1890	2000
Buteni	NTC	562	1150	1300	1400
upstream	NFC	350	900	1200	1100
	NFE	0	0	20	36
Buteni downstream	NTC	380	1000	1200	1300
	NFC	200	900	700	340
	NFE	11	36	72	100
Ineu upstream	NTC	490	910	3830	4760
	NFC	200	562	1200	1150
	NFE	0	55	105	114
Ineu downstream	NTC	562	1150	3900	5220
	NFC	348	775	2240	2300
	NFE	0	91	110	153
Chişineu Criş upstream	NTC	540	1300	2980	4900
	NFC	224	744	2200	2600
	NFE	0	84	97	120
Chişineu Criş downstream	NTC	925	1700	3800	6410
	NFC	326	820	2400	2700
	NFE	0	95	120	140

NTC – the number of total coliforms (no./100ml), NFC - the number of faecal coliforms (no./100ml), NFE - the number of faecal enterococcus (no./100ml)

A slight growth of this number was recorded in spring. The higher values were recorded during the hot season – in summer. In autumn the obtained values for the hygienic and sanitary indicators are close to the values from the last season. The reason might be the vegetal and animal organic materia in the water mass, brought by the rainfall in autumn and the flowing process of vegetal and organic waste from the river's shore.

The development of microorganism communities is stimulated. The low temperature in winter negatively influences the development of the microorganism communities. The increased temperature and the organic materia determines a higher density of the bacterial community from the water mass. Higher values for the hygienic and sanitary indicators at the drawing point level from downstream compared to the ones from upstream, can be the reason of the flowing process from the riverbed: domestic and industrial waste water from human communities.

Therefore we can affirm that the studied hygienic and sanitary bacteria has seasonal variations and it depends on the drawing point of the tests. The establishment of the faecal pollution at the level of the aquatic environment can be made by taking into consideration the report between the faecal coliform (FC) and the faecal enterococcus (FE). When the report`s value is bigger than 4, a human source pollution is evidenced. When the report`s value is between 2 and 4, the pollution is mixed, but the human source pollution is predominant.

When the report`s value is between 0,7 and 1 the pollution is also mixed, but this time the animal source pollution is predominant. The report`s value smaller than 0,7 indicate a faecal gene pollution containing animal dejects (Barbato şi colab., 1990; Cuşa şi Astratinei, 1996). During the year of 2003 the report`s value of the number of faecal coliforms/the number of faecal enterococcus have in every other drawing point higher values than 4, meaning a faecal gene pollution made by human dejects.

The fecal pollution in 2004 is pre eminently made by human dejects. But, there are drawing points like Brad upstream, Buteni upstream and Chişineu Criş downstream where the faecal enterococcus is absent during the cold season. The report`s value of the number of faecal coliforms/ the number of faecal enterococcus between 2 and 4 indicates a mixed faecal gene pollution (from animals and humans), but mostly human.

The drawing points which have this value are located upstream, where zootehnical microfarms can be found. The report`s value of 0,92 at the drawing point level Gurahonţ downstream got to be noticed. It means the presence in the area of a faecal gene pollution made by animal deject. For the year of 2005, in the cold season the faecal enterococcus is absent in almost every drawing point. There are 2 exceptions: Gurahonţ upstream and Buteni downstream.

At the drawing point level Gurahonţ downstream in autumn a report`s value of the number of faecal coliforms/the number of fecal enterococcus is lower than 0,7 meaning a faecal gene pollution made by animal dejects. The zootehnical microfarms from the area are the season of this value. Again in the winter season, but this time at the drawing point Buteni downstream, the report`s value is 3,4 meaning a mixed pollution made by humans.

CONCLUSIONS

The total and faecal coliform bacteria were detected in every studied water test, having minimal values in winter and high values in summer and autumn. The reason is the increased water temperature, the organic materia and even the spare time activities in the river`s water from this season.

The faecal enterococcus was not determined in every water test. They usually don`t appear in winter, because of the limited organic materia and the low water temperature compared to the temperature proper for their development. The approximate number of faecal enterococcus records an explosive growth in summer and autumn. This confirms the result obtained from the total coli forms and the faecal enterococcus and the growth of pollution sources in the hot seasons.

The values recorded for the 4 hygienic and sanitary bacteriological indicators are lower upstream and bigger downstream. The reason might be the waste water from the water cleaning stations, which seemingly have insufficient cleaning systems for the domestic and fecal gene deject.

The report between the approximate number of faecal coliforms and the approximate number of faecal enterococcus allow us to affirm that in the Crişul Alb River the faecal gene pollution is made by animals or humans. Sometimes it is mixed, mostly human or animal, in different seasons and depending on the drawing point tests.

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