

BACTERIOLOGICAL INVESTIGATIONS FROM WATER AND SEDIMENT IN THE LONGITUDINAL SECTION OF THE RIVER TISZA

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Abstract. In 30th January, 2000 the dam of the clarifying plant of an Australian-Romanian mining-company named Aurul Corporation located on the border of Zazár settlement near to Nagybánya was broken through and more than 100,000 m³ cyanide-containing water polluted with metal-complex had got into the Szamos through Lápos-stream then finally into the Tisza. According to the calculations referring to the turnover of substances the whole amount of about 105-110 ton cyanide remained in aqueous phase and together with attached heavy metals it left Hungary at the exit of polluting wave. The Ministry of Environmental Protection has elaborated a program of investigation for surveying the environmental and natural damages of the Szamos and the Tisza caused by cyanide pollution originated from Romania. The organic part of this program was the investigation with laminar stream performed in the longitudinal section of the Tisza which happened between 19th and 30th September in 2000 at the whole Hungarian reach of the Tisza from Tiszabecs to Tiszasziget. Bacteriological examinations were performed from water and sediment. In the seventh month after marching down of pollution the hygienic bacteriological state of water of the Tisza developed according to the characteristics of the autumn state with low water. The effect of cyanide pollution on the aquatic communities of microbes has not been already detectable. However, the results of bacteriological investigations with the character of material-cycles can be considered as a basic research on the Hungarian reach of the river Tisza.

Keywords: cyanide pollution, microbial cycles of elements, sampling with laminar stream.

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Introduction

In 30th January, 2000 at 22 o' clock the dam of the clarifying plant of an Australian-Romanian mining-company named Aurul Corporation located on the border of Zazár settlement near to Nagybánya was broken through and more than 100,000 m³ cyanide-containing water polluted with metal-complex had got into the Szamos through Lápos-stream then finally into the Tisza. In the case of total dissolved cyanide-content we can tell about the concentration-relations developed during the time of

marching down of pollution that the maximum concentration was between 20 and 30 mg/l in the Szamos, between 10 and 15 mg/l at the reach of the Tisza located under the Szamos and then moving downstream this concentration gradually decreased. The maximal concentration in water leaving Hungary was 1.49 mg/l (which was still 150 times higher than the permissible value). Metal-complexes — well soluble in water — come with polluting wave were very stable. In consequence of this the whole amount of about 105-110 ton cyanide remained in aqueous phase according to the calculations referring to the

turnover of elements and together with attached heavy metals it left Hungary at the exit of polluting wave. (This fact was also verified by the results of samples taken at the reservoir of Vaskapu).

In the February of 2000 the Ministry of Environmental Protection elaborated a program of investigation (with the collaboration of concerned Environmental Protection Inspectorate, National Parks, the Water-quality Protection Institute of VITUKI Corporation and KGI) titled: "Surveying the environmental and natural damages of the Szamos and the Tisza caused by cyanide pollution originated from Romania". The implementation of the program consisted of three phases. In the first phase the tracking of pollution marched down and laboratory measurements had been done as well as the estimation of the extent of destruction appearing in the communities of living organisms had been started. In the second phase the surveying of the extent of damage caused by polluting material marched down had taken place as well as the project of the investigation performed in the longitudinal section of the Tisza had been created. According to this the expedition would be performed in the period of low water at the Hungarian reach of the Tisza form the national boundary to the national boundary. The work of the expedition was planned for the period of two weeks which would be repeated in every two or three years. For the third phase the Ministry has planned the investigation of the regeneration of aquatic communities of living organisms, the reduction of detected irreversible damages and the implementation of a rehabilitating program.

The organic part of this program was consequently the investigation with laminar stream performed in the longitudinal section of the Tisza which happened between 19th and 30th September in 2000.

The client was the Ministry of Environmental Protection, the executor was the Middle-Tisza Water Authority. Chemical and biological processing of samples were performed by the following institutes: Budapest Institute of the National Public Health and Medical Officer Service (Hungarian abbreviation: ÁNTSZ), Department of Public Health Biological Laboratory (Budapest); Ecological Department of Debrecen University (Debrecen); Fish-culture Research Institute (Szarvas); North Hungarian Environmental Inspectorate (Miskolc); Environmental Inspectorate of Upper-Tisza Region (Nyíregyháza), Environmental Inspectorate of Trans Tisza (Debrecen); Environmental Inspectorate of the Middle-Tisza Region (Szolnok), Middle-Tisza Water Authority; Environmental Inspectorate of Lower

Tisza Region (Szeged); VITUKI Corporation (Budapest); MTA ÖBKI — Hungarian Danube Research Station (Göd); Eszterházy Károly College (Eger); Biological Department of Szeged University (Szeged).

Review of literature

During last thirty years the Middle-Tisza Water Authority (MTWA) has performed investigations with laminar stream performed in the longitudinal section of the Tisza in many occasions. In six occasions (June of 1975, September of 1975, August of 1977, September of 1979, September of 2000, November of 2002) the sampling of the whole Hungarian reach of the Tisza has happened from which the majority was a sampling with laminar stream. During each expedition the sampling of water (and sediment in some cases) has happened at 30 to 50 sampling sites. On the occasion of single investigations performed in the longitudinal section of the river the majority of sampling sites was the same. In the seventy' years biological examinations also have been performed beside physical and chemical examinations of water and sediment. The aim was to determinate those parameters among microbiological ones which were examined in the hygienic routine. The authors published the results of measurements in the volumes of Tiscia (B. Tóth 1981, Estók 1980, 1981, Hegedűs and Zsikó 1981). In the autumn of 2000 the microbiological examinations had been extended also to the microbiological parameters which are important from the aspect of material-cycles of the sediment of the Tisza and its tributaries.

Necessity of material-flow investigations performed on the basis of bacterial activity

In the latter years Hungarian bacteriological research and the applied, routine bacteriological activity have found themselves face to face with fundamental methodological challenge and a challenge in the interpretation. A continuous change can be experienced in the terminology of environmental bacteriology. Today we do not distinguish so sharply the autochthonous and allochthonous microbiota discussed and examined separately earlier as e.g. in receiving water bodies the one part of the allochthonous microbiota originated from sewage waters can infiltrate into the functional microbe-community of a given surficial water.

According to Némédi (Gorzó *et al.* 1998), we ought to classify the elements of microbiota (taxa, taxon-groups) on the basis whether they have a

function in the place of detection, or this activity is occasional only, or they do not have any role at all in the material-cycles of a given medium. The same taxon can be found on either side so in this relation. The function is decisive not the classification into a species. Certain taxa have shown activity of material-cycles very widely noting also that bacteria showing obligate function can occur too. Thus the 'microbiota elements' of public health can already be well embedded in the general structure of microbial material-cycles.

Though the quantitative examination of bacteria receives lots of criticism (differences of in vivo/in vitro circumstances), yet the determination of actual number of microbes is a very important question of environmental microbiology. There is every hope that the extent of bacterium-density, which is necessary for producing a given function (activity), would be traceable by using more reliable methods.

However, today there are limits yet in the knowledge of qualitative composition (soil, river, lake) with whole level of species. There are such micro-organisms presence of which can be detected by electron microscope, but they can not be cultured in any medium (oligotrophic microbes). However, one part of microbes, which can be cultured, is undeterminable (according to the present-day state of taxonomy). However, we have to take into consideration the fact, too, that during the annual dynamics the composition of microbiota is also changing, thus it is understandable that the tracking of population dynamics in qualitative level can mean insoluble task many times. However, by spreading the use of molecular methods and by appropriate use of automatic identifying systems of microbes we can get closer to the answer of the question.

Materials and Methods

The investigation of the Tisza performed in the longitudinal section was implemented between 19th — 30th September, 2000. The expedition sampled the Tisza and its tributaries (the Szamos, the Lónyai-channel, the Bodrog, the Sajó, the Zagyva, the Hármas-Körös, the Maros) from Tiszabecs (744.2 r.km) to Tiszasziget (167.0 r.km) altogether at forty sampling sites from the board of an exploratory ship (Table 1).

The bacteriological processing of forty water-samples taken from the channel line was performed in the Regional Laboratory of MTWA and the laboratory of the Budapest Institute of ÁNTSZ, Department of Public Health Biological Laboratory. The sampling of sediment happened at forty sections, tree samples were taken per sections (right bank, left

bank and channel line). So altogether 120 sediment-samples were collected for physical, chemical, heavy-metal and macro-zoobenthos examinations. From this collection in the case of 19 sediment-samples the Budapest Institute of ÁNTSZ, Department of Public Health Biological Laboratory performed the microbiological examinations important from the aspect of material-cycles.

In this paper we report particularly on microbiological relations of the investigation performed in the longitudinal section of the Tisza in 2000.

Bacteriological examinations performed at forty water-samples taken from channel line: Aerobe total count at 22 and 37 °C, number of Coliforms MPN, number of Faecal Coliforms MPN, number of Faecal Streptococci CFU, number of Clostridia CFU at 46 °C, total number of bacterial CFU (Accridin Orange Direct Count method, Hobbie *et al.*, 1977).

Bacteriological examinations performed at seven water-samples taken from channel line: Determination of *Salmonella*-positivity, *Escherichia coli* and *Pseudomonas aeruginosa*.

Bacteriological examinations performed from 19 sediment-samples: Determination of number of Coliforms CFU, number of Faecal Coliforms CFU, number of Faecal Streptococci CFU, number of Clostridia CFU, *Salmonella*-positivity, *Pseudomonas aeruginosa*, aerobe and anaerobe total count at 22 and 37 °C, *Staphylococcus aureus*, number of colonies indicating proteolytic activity, Mould-fungus CFU, Yeast-fungus CFU, total count of Desulphurylating bacteria, total count of Cellulolytic bacteria, total count of Ammonifier bacteria, total count of Nitrifier bacteria, total count of Denitrifier bacteria.

Bacteriological qualification was performed according to the C group of the Table 2 of MSZ 12749:1993 standard and on the basis of the guide of the Budapest Institute of ÁNTSZ, Department of Public Health Biological Laboratory.

Results

Water-bacteriology

Number of saprophyte bacteria CFU indicates the extent of bacterium-biota which depends on the presence of quickly decomposable organic materials and decomposes those materials.

Viable microorganisms existing in water reproduce generally better at 22 °C, than at 37 °C in the mediums used in laboratories.

Microorganisms reproducing at 37 °C (which get into our waters with external pollution) are less viable in water. They are trend-like indicators of

Table1. CFU and Coliform counts from the longitudinal section of Tisza performed between 21st and 30th September, 2000.

| Time of sampling | Name of the water body | r.km | Total count at 22°C, CFU/ml | Total count at 37°C, CFU/ml | Number of Coliforms in 1 ml |
|------------------|------------------------|-------|-----------------------------|-----------------------------|-----------------------------|
| 20.09.2000 | TISZA | 744.2 | 1 300 | 740 | 17 |
| 20.09.2000 | TISZA | 687.0 | 640 | 380 | 4.9 |
| 20.09.2000 | Szamos | 1.0 | 28 000 | 13 000 | 350 |
| 20.09.2000 | TISZA | 685.0 | 18 000 | 13 000 | 350 |
| 20.09.2000 | TISZA | 683.0 | 12 000 | 7 200 | 160 |
| 21.09.2000 | TISZA | 627.8 | 5 600 | 4 400 | 14 |
| 21.09.2000 | TISZA | 616.5 | 5 200 | 4 000 | 13 |
| 21.09.2000 | TISZA | 591.9 | 6 800 | 6 400 | 3.3 |
| 22.09.2000 | TISZA | 568.7 | 6 600 | 6 300 | 3.3 |
| 22.09.2000 | TISZA | 559.9 | 2 700 | 2 500 | 1.3 |
| 22.09.2000 | Lónyai-csatorna | 1.0 | 180 000 | 110 000 | 2 400 |
| 22.09.2000 | TISZA | 557.9 | 3 100 | 1 600 | 11 |
| 22.09.2000 | TISZA | 555.9 | 2 300 | 1 100 | 13 |
| 23.09.2000 | TISZA | 544.7 | 600 | 350 | 2.6 |
| 23.09.2000 | Bodrog | 1.0 | 480 | 150 | 3.3 |
| 23.09.2000 | TISZA | 542.7 | 880 | 190 | 1.3 |
| 23.09.2000 | TISZA | 523.5 | 1 100 | 900 | 1.7 |
| 24.09.2000 | TISZA | 493.4 | 280 | 120 | 0.68 |
| 24.09.2000 | Sajó | 1.0 | 1 800 | 750 | 7.9 |
| 24.09.2000 | TISZA | 491.4 | 250 | 130 | 0.45 |
| 24.09.2000 | TISZA | 489.4 | 400 | 100 | 0.78 |
| 25.09.2000 | TISZA | 464.0 | 400 | 300 | 1.1 |
| 25.09.2000 | TISZA | 453.0 | 220 | 75 | 0.4 |
| 26.09.2000 | TISZA | 431.0 | 8 400 | 4 300 | 3.3 |
| 26.09.2000 | TISZA | 415.0 | 20 000 | 160 | 0.78 |
| 26.09.2000 | TISZA | 404.0 | 17 000 | 4 000 | 0.78 |
| 27.09.2000 | TISZA | 395.0 | 12 000 | 250 | 2.2 |
| 27.09.2000 | TISZA | 336.6 | 11 000 | 900 | 7.0 |
| 27.09.2000 | Zagyva | 1.0 | 6 500 | 2 000 | 160 |
| 27.09.2000 | TISZA | 334.6 | 11 000 | 300 | 7.9 |
| 28.09.2000 | TISZA | 330.0 | 2 400 | 2 200 | 7.0 |
| 28.09.2000 | TISZA | 266.4 | 10 000 | 3 600 | 170 |
| 29.09.2000 | TISZA | 244.6 | 6 000 | 1 500 | 140 |
| 29.09.2000 | Hármas- Körös | 1.0 | 1 800 | 450 | 28 |
| 29.09.2000 | TISZA | 242.6 | 16 000 | 1 500 | 54 |
| 29.09.2000 | TISZA | 215.0 | 2 000 | 1 400 | 110 |
| 30.09.2000 | TISZA | 178.0 | 680 | 440 | 35 |
| 30.09.2000 | Maros | 1.0 | 1 500 | 1 200 | 22 |
| 30.09.2000 | TISZA | 176.0 | 1 800 | 1 200 | 92 |
| 30.09.2000 | TISZA | 167.0 | 3 000 | 2 400 | 160 |

Table 2. Faecal coliform, *Streptococcus*, *Clostridium* and planctonic bacterium counts from the longitudinal section of the river Tisza performed in between 21st and 30th September, 2000.

| Time of sampling | Name of the water body | r.km | Faecal coliforms MPN/ml | Faecal streptococci CFU/ml | Clostridia CFU/ml | Planctonic bacteria million cell/ml |
|------------------|------------------------|-------|-------------------------|----------------------------|-------------------|-------------------------------------|
| 20.09.2000 | TISZA | 744.2 | 4.9 | 1.5 | 16 | 2.35 |
| 20.09.2000 | TISZA | 687.0 | 1.3 | 0.3 | 12 | 2.52 |
| 20.09.2000 | Szamos | 1.0 | 35.0 | 3.2 | 210 | 5.11 |
| 20.09.2000 | TISZA | 685.0 | 22.0 | 2.2 | 160 | 3.63 |
| 20.09.2000 | TISZA | 683.0 | 35.0 | 1.6 | 88 | 2.90 |
| 21.09.2000 | TISZA | 627.8 | 7.9 | 0.1 | 70 | 3.08 |
| 21.09.2000 | TISZA | 616.5 | 1.7 | 0.1 | 45 | 3.02 |
| 21.09.2000 | TISZA | 591.9 | 0.78 | 0.1 | 65 | 3.68 |
| 22.09.2000 | TISZA | 568.7 | 0.78 | 0.1 | 53 | 3.71 |
| 22.09.2000 | TISZA | 559.9 | 0.45 | 0.1 | 52 | 4.00 |
| 22.09.2000 | Lónyai-cs. | 1.0 | 920.0 | 60.0 | 1 200 | 10.04 |
| 22.09.2000 | TISZA | 557.9 | 1.1 | 0.2 | 96 | 5.34 |
| 22.09.2000 | TISZA | 555.9 | 4.9 | 0.0 | 90 | 4.18 |
| 23.09.2000 | TISZA | 544.7 | 0.2 | 0.0 | 60 | 4.29 |
| 23.09.2000 | Bodrog | 1.0 | 1.4 | 0.2 | 70 | 3.37 |
| 23.09.2000 | TISZA | 542.7 | 0.2 | 0.0 | 70 | 3.63 |
| 23.09.2000 | TISZA | 523.5 | 0.0 | 0.0 | 70 | 3.71 |
| 24.09.2000 | TISZA | 493.4 | 0.2 | 0.1 | 70 | 3.77 |
| 24.09.2000 | Sajó | 1.0 | 4.9 | 0.7 | 680 | 4.09 |
| 24.09.2000 | TISZA | 491.4 | 0.0 | 0.2 | 100 | 3.37 |
| 24.09.2000 | TISZA | 489.4 | 0.0 | 0.1 | 120 | 2.87 |
| 25.09.2000 | TISZA | 464.0 | 0.2 | 0.1 | 180 | 2.21 |
| 25.09.2000 | TISZA | 453.0 | 0.0 | 0.2 | 96 | 2.73 |
| 26.09.2000 | TISZA | 431.0 | 0.0 | 0.1 | 45 | 2.29 |
| 26.09.2000 | TISZA | 415.0 | 0.45 | 0.1 | 40 | 2.73 |
| 26.09.2000 | TISZA | 404.0 | 0.0 | 0.0 | 82 | 2.41 |
| 27.09.2000 | TISZA | 395.0 | 0.4 | 0.0 | 95 | 2.18 |
| 27.09.2000 | TISZA | 336.6 | 0.45 | 0.1 | 100 | 2.26 |
| 27.09.2000 | Zagyva | 1.0 | 7.0 | 7.7 | 960 | 2.90 |
| 27.09.2000 | TISZA | 334.6 | 0.45 | 0.1 | 37 | 2.29 |
| 28.09.2000 | TISZA | 330.0 | 4.9 | 0.7 | 110 | 2.32 |
| 28.09.2000 | TISZA | 266.4 | 160.0 | 0.7 | 120 | 2.32 |
| 29.09.2000 | TISZA | 244.6 | 92.0 | 0.9 | 180 | 2.41 |
| 29.09.2000 | Hármas- Körös | 1.0 | 24.0 | 0.4 | 130 | 2.73 |
| 29.09.2000 | TISZA | 242.6 | 43.0 | 0.5 | 100 | 2.93 |
| 29.09.2000 | TISZA | 215.0 | 92.0 | 1.0 | 77 | 2.47 |
| 30.09.2000 | TISZA | 178.0 | 17.0 | 0.1 | 65 | 2.50 |
| 30.09.2000 | Maros | 1.0 | 3.3 | 0.5 | 63 | 3.34 |
| 30.09.2000 | TISZA | 176.0 | 13.0 | 0.4 | 76 | 2.76 |
| 30.09.2000 | TISZA | 167.0 | 24.0 | 1.5 | 72 | 2.76 |

sewage waters with communal and agricultural origin, or rather they can indicate hygienic problem, too.

Aerobe total count at 22 and 37 °C

The aerobe total count was not significant (II. class) at the Upper-Tisza. The polluted (IV. class) Szamos with major water-discharge influenced the bacterium-biota of the Tisza to a large extent. The aerobe total count increased threefold in the Tisza downstream the mouth of the Szamos (685.0 r.km) compared to the value detected at section of 744.2 r.km (Table 2). Considering the aerobe total count at 22 and 37 °C the quality of water belonged to the III-IV. classes in this region.

Considering the number of aerobe colony forming units at 22 and 37 °C, river Tisza belonged to the II. class from the area of Gávavencsellő (559.9 r.km) to Tiszacsege (453.0 r.km) downstream the river.

From Tiszafüred (431.0 r.km) to Mindszent (215 r.km), the total count at 22 °C had a magnitude of ten thousand (III-IV. classes), the number of colony forming units at 37 °C had a magnitude of hundred (II-III. classes).

Among tributaries, Lónyai-channel was the most polluted (Table 2), the aerobe colony forming bacteria had been cultured in the magnitude of hundred thousand (V. class).

Facultative faecal-indicator bacteria having the type of *Clostridium* are sulphite-reducing anaerobe spored microorganisms which have short thick rod-like shape, form a capsule and their spores are oval-shaped. One part of them is the inhabitant of human alimentary canal, the other part lives in sediment or soil, for this very reason they are not really suitable for classifying categories in surface waters. In the case of sediment-disturbing the spores can get into water body again and being more resistant than vegetative forms they can be the indicators of former faecal pollution.

Clostridium-type bacteria were present in a great number at the whole reach of the Tisza (from 687 r.km to 167 r.km) in the water body. From water of the Lónyai-channel we have cultured these bacteria in larger amount with one order of magnitude — compared to the other tributaries.

The members of Enterobacteriaceae family are facultative faecal-indicator organisms. Their presence proves the load of organic material getting into water from outside.

The number of Coliform bacteria belonged to the II. class at river Tisza upstream the mouth of river Szamos. The high number of coliform bacteria of Szamos was measurable also at the Tisza after the

moth (Table 2). This reach of the river (687-683 r.km) can be categorized into the 'polluted' category. The river can be characterized by the I-II. categories of water-quality on the basis of the number of coliform bacteria up to the area of Tiszaug (266.4 r.km) with regard to this bacterium-group. The number of bacteria has increased with two orders of magnitude at Tiszaug (probably) due to the effect of sewage water of Szolnok and it does not decrease significantly up to the area of Tizzasziget (167 r.km). Among tributaries, river Szamos and the Lónyai-channel were the most polluted.

Obligate faecal-indicators

Those coliform bacteria, which ferment lactose also at 44 °C, are faecal coliform bacteria. Obligate faecal indicators originate from faeces solely, getting into water their time for surviving is short, they are not able to post-reproduce in water. They are reliable indicators of new faecal pollution.

The habitat of faecal Streptococci is the human and animal alimentary canal therefore their cultivation from water (together with faecal Coliforms) indicates new faecal pollution.

Faecal Streptococci are very resistant, they are able to survive in unfamiliar circumstances for a longer term.

According to this, if the number of faecal Streptococci is not accompanied — simultaneously — to the large number of faecal Coliforms then it indicates former already terminated pollution in most cases.

The obligate faecal indicators occurred in the greatest number at the reach of river Tisza (685.0-616.5 r.km) located downstream the mouth of river Szamos (Table 2). The quality of water belongs to the III-IV. classes in this water body. Faecal pollution was not significant from the area of Tuzsér to Szolnok (616.5-330.0 r.km), while the extent of pollution justified a classification into III-IV. classes from Tiszaug to Tizzasziget (266.4-167 r.km).

Among tributaries, faecal pollution of Lónyai-channel was significant.

Sometimes the total number of planktonic bacteria determined by direct microscopic counting is greater with more orders of magnitude than the number of bacteria determined by cultivating methods because there are microorganisms of which presence can be detected by microscope though, but they can not be cultivated in any kind of medium. By AODC technique the living (but it is not sure that continuing active metabolism) bacterium-cells can be counted.

Total number of bacteria (or the number of planktonic bacteria) has ranged between 2.18×106

cell/ml and 5.34×10^6 cell/ml at the examined reach of the Tisza. Among tributaries the number of planktonic bacteria of Lónyai-channel was the highest: 10.04×10^6 cell/ml (Table 2) which represented a significant biomass.

The Budapest Institute of ÁNTSZ, Department of Public Health Biological Laboratory has performed hygienic bacteriological examinations from water at seven sampling sites in the course of the investigation of the Tisza performed in the longitudinal section (at 687; 683; 489; 266.4; 215; 167 r.km and at the mouth of the Szamos). On the basis of these examinations *Escherichia coli* was detectable at each of seven sampling sites, in the largest number at the section of the Tisza (683 r.km) located downstream the mouth of the Szamos (21 000 colony/100 ml).

Salmonella-positivity was detectable in the Szamos, the presence of *Pseudomonas aeruginosa* was demonstrable only in the sections of river Tisza upstream (687.0 r.km) and downstream (683.0 r.km) the mouth of river Szamos.

Sediment-bacteriology

As the Hungarian Standards valid at the present do not determine a limit-value for the quantitative relations of bacterium-communities living in the sediment therefore the assessment has happened only on the basis of trend-lines fitted to the diagrams.

Examination of hygienic parameters

According to the bacteriological results of the Budapest Institute of ÁNTSZ, Department of Public Health Biological Laboratory the aerobe total count at 22 and 37 °C was present in the magnitude of ten thousand while the anaerobe total count (at the two same degrees of temperature) in the magnitude of thousand in the sediment of the section of river Tisza from 568.7 r.km to 493.4 r.km (Tiszabercel – Tiszagyulaháza).

The number of these bacteria has decreased significantly in the sediment-samples of the Middle-Tisza, while the aerobe and anaerobe total counts of sediment have increased threefold at the Lower-Tisza from the area of Tiszaug to Tizzasziget (266.4 - 167.0 r.km). Among tributaries, the aerobe and anaerobe total counts of the sediment of rivers Hármas-Körös and Szamos were high outstandingly.

Number of Clostridia exceeded one thousand in the sediment originated from the reach of Upper-Tisza (559.9 – 491.4 r.km) which, however, decreased significantly at the Middle- and Lower-Tisza. The number of colony (7500 colony/g wet sediment) in the sediment of the Lónyai-channel has indicated a former pollution with large probability.

The number of Coliform bacteria was surprisingly small in the sediment-samples of Tisza. Their number does not exceed the value of 100/g wet sediment in each case.

The sediment of the Lower-Tisza (266.4 – 167 r.km) proved to be the most polluted.

Among tributaries, river Sajó had the best quality considering the number of Coliform bacteria.

Faecal pollution of sediment was not significant at the examined reach of Tisza. The number of faecal Coliforms and the number of faecal Streptococci were larger in the area of Tiszaújváros at the Upper-Tisza (491.4 r.km), then they decreased at the Middle-Tisza. The number of faecal indicators had increased again in the area of Tiszaug (266.4 r.km).

Among tributaries, the sediment of Lónyai-channel and Hármas-Körös was very polluted by microorganisms originated from faeces.

Salmonella- and *Staphylococcus aureus*-positivity was not demonstrable in the 19 sediment-samples.

Pseudomonas aeruginosa-positivity was detected in four cases by the Budapest Institute of ÁNTSZ, Department of Public Health Biological Laboratory in the sections of Tisza located at 559.9; 336.6; 215.0 and 167.0 r.km.

Quantitative relations of microbiota elements participating in the microbial cycles of elements in river-sediments

Aquatic plants rich in cellulose die in water and sink down to the bottom, sewage waters getting into the river also can contain by-products with cellulose content (e.g. sewage water of a paper-mill) which are decomposed by the activity of cellulose-decomposing bacteria, ray-fungi and fungi.

The amount of Cellulose decomposers (Table 3) is significant in the sediment of the Upper-Tisza from the area of Gávavencsellő to Tiszaújváros (559.9 – 491.4 r.km). This amount has decreased significantly at the Middle-Tisza, then it has increased slightly downstream the mouth of the Zagyva after the entering of sewage water of the Paper-mill of Szolnok.

The largest amount of cellulose decomposers was demonstrable in the sediment of the Lónyai-channel and river Bodrog (Table 3).

Bacteria indicating proteolytic activity participate in the process of proteolysis occurring in the course of biodegradation. During proteolysis amino-acids are produced as final products of lytic proteins.

The total count indicating proteolytic activity has increased threefold in front of the mouth of Lónyai-channel (559.9 r.km) and has reached the magnitude of hundred thousand to the area of Tiszatardos

3. Table: Results of sediment-bacteriological examinations of the investigation of the Tisza performed in the longitudinal section of the river (in 1 g wet sediment).

| Name of the water body | r.km | 1. | 2. | 3. | 4. | 5. | 6. | 7. | 8. | 9. |
|------------------------|-------|-------|--------|------|-----|--------|------|--------|------|-----|
| TISZA | 744.2 | - | - | - | - | - | - | - | - | - |
| TISZA | 687.0 | 9.3 | 100000 | 9.3 | 4.3 | 24 | 10 | 3000 | 600 | 0.0 |
| Szamos | 1.0 | 24 | 100000 | 9.3 | 0.3 | 24 | 35 | 2500 | 0.0 | 0.0 |
| TISZA | 685.0 | - | - | - | - | - | - | - | - | - |
| TISZA | 683.0 | 12 | 100000 | 9.3 | 0.3 | 24 | 30 | 5000 | 100 | 0.0 |
| TISZA | 627.8 | - | - | - | - | - | - | - | - | - |
| TISZA | 616.5 | - | - | - | - | - | - | - | - | - |
| TISZA | 591.9 | - | - | - | - | - | - | - | - | - |
| TISZA | 568.7 | >1100 | 100000 | >110 | 24 | >110 | 2000 | 25000 | 800 | 700 |
| TISZA | 559.9 | >1100 | 100000 | 46 | 1.5 | 110 | 300 | 60000 | 600 | 800 |
| Lónyai-cs. | 1.0 | >1100 | 100000 | >110 | 9.3 | 110 | 7500 | 60000 | 100 | 0.0 |
| TISZA | 557.9 | - | - | - | - | - | - | - | - | - |
| TISZA | 555.9 | - | - | - | - | - | - | - | - | - |
| TISZA | 544.7 | >1100 | 100000 | 110 | 24 | >110 | 1500 | 200000 | 700 | 0.0 |
| Bodrog | 1.0 | >1100 | 100000 | >110 | 110 | >110 | 1125 | 200000 | 1500 | 0.0 |
| TISZA | 542.7 | - | - | - | - | - | - | - | - | - |
| TISZA | 523.5 | >1100 | 100000 | 110 | 46 | >110 | 1375 | 80000 | 800 | 0.0 |
| TISZA | 493.4 | 29 | 100000 | 110 | 110 | 1.5 | 750 | 10000 | 500 | 0.0 |
| Sajó | 1.0 | 46 | 100000 | 14 | 2.3 | 2.3 | 50 | 4000 | 0.0 | 0.0 |
| TISZA | 491.4 | - | - | - | - | - | - | - | - | - |
| TISZA | 489.4 | - | - | - | - | - | - | - | - | - |
| TISZA | 464.0 | >110 | 100000 | 9.3 | 4.3 | 2.8 | 37 | 4000 | 0.0 | 0.0 |
| TISZA | 453.0 | - | - | - | - | - | - | - | - | - |
| TISZA | 431.0 | - | - | - | - | - | - | - | - | - |
| TISZA | 415.0 | - | - | - | - | - | - | - | - | - |
| TISZA | 404.0 | >110 | 100000 | 9.3 | 4.3 | 15 | 150 | 9000 | 0.0 | 0.0 |
| TISZA | 395.0 | - | - | - | - | - | - | - | - | - |
| TISZA | 336.6 | 1100 | 100000 | 110 | 24 | >110 | 97 | 20000 | 0.0 | 0.0 |
| Zagyva | 1.0 | - | - | - | - | - | - | - | - | - |
| TISZA | 334.6 | - | - | - | - | - | - | - | - | - |
| TISZA | 330.0 | - | - | - | - | - | - | - | - | - |
| TISZA | 266.4 | >1100 | 100000 | 46 | 4.3 | 10000 | 50 | 10000 | 500 | 0.0 |
| TISZA | 244.6 | - | - | - | - | - | - | - | - | - |
| Hármas- Körös | 1.0 | >2400 | 100000 | 46 | 9.3 | 10000 | 450 | 100000 | 800 | 0.0 |
| TISZA | 242.6 | - | - | - | - | - | - | - | - | - |
| TISZA | 215.0 | >1100 | 100000 | 24 | 24 | 10000 | 122 | 100000 | 400 | 0.0 |
| TISZA | 178.0 | - | - | - | - | - | - | - | - | - |
| Maros | 1.0 | >4600 | 100000 | 46 | 24 | 100000 | 650 | 200000 | 500 | 0.0 |
| TISZA | 176.0 | - | - | - | - | - | - | - | - | - |
| TISZA | 167.0 | >2400 | 100000 | 21 | 4.3 | 10000 | 800 | 80000 | 300 | 0.0 |

Key to the signs used : 1.: Heterotrophic H₂S producers MPN/ml, 2.: Ammonifying bacteria MPN/ml, 3.: Cellulose-degrading bacteria MPN/ml, 4.: Nitrifying bacteria MPN/ml, 5.: Denitrifying bacteria MPN/ml, 6.: Clostridia CFU/ml, 7.: number of colony indicating proteolytic activity CFU, 8.: CFU of mould fungi, 9.: CFU of yeast fungi

(523.5 r.km). The number of these microbial elements has decreased at the Middle-Tisza (Table 3), and the total count indicating proteolytic activity has increased to the magnitude of hundred thousand only downstream the mouth of Hármas-Körös. We have managed to detect these colonies in the largest

number from the sediment of rivers Bodrog and Maros.

Chemolithotroph Nitrifying bacteria oxidize the large part of ammonia found in water through nitrite to nitrate and they assimilate carbon dioxide by the help of obtained energy. The most frequent nitrite-forming microorganisms are the members of the

genera *Nitrosomonas* and *Nitrosococcus*. Nitrate is produced as a result of the activity of the members of the genus *Nitrobacter*. Nitrate produced during nitrification has a great importance. Algae and higher plants assimilate nitrate in a large mass, and the denitrifying bacteria utilize nitrate as electron-acceptor.

The number of nitrifying bacteria was the largest in the sediment at the reach of the Upper-Tisza located between Tiszabercel and Tiszagyulaháza (568.7 – 493.4 r.km). Their amount has decreased at the Middle-Tisza, then their number has increased slightly (Table 3) in the area of Szolnok - Mindszent (336.6 – 215.0 r.km). Among tributaries, the number of nitrifying bacteria reached the magnitude of hundred in the sediment of river Bodrog.

Denitrifying organisms reduce nitrite produced in the first step of nitrate-reduction to elemental nitrogen. Organisms performing denitrification do not belong to a uniform group neither morphologic nor biochemical way. Denitrification can be performed by e.g. the members of the genus *Bacillus*, certain genera of the family Micrococcaceae, *Pseudomonas aeruginosa*, *Thiobacillus denitrificans* and so on. The small amount of dissolved oxygen, abundant nitrate and organic material refer to the presence of denitrifiers.

Denitrifying organisms can utilize the most diverse materials (e.g. the intermediate products of cellulose-decomposition) as electron donor.

The number of denitrifiers reached the multitude of hundred in the sediment-samples taken at the reach of the Upper-Tisza located between 568.7 and 523.5 r.km (Table 3), then their number has decreased significantly at the reach of the Middle-Tisza. Denitrifiers were present in a huge amount (in the magnitude of ten thousand) in the sediment-samples of the area located between Tiszaug and Tiszasziget (266.4 – 167.0 r.km). Among tributaries, mainly the sediment of rivers Maros and Hármas-Körös contained these microbiota elements in the largest number.

The total count of mould fungi was the largest in the sediment-samples taken at the reach of the Upper-Tisza located between Tiszabercel and Tiszagyulaháza (568.7 – 493.4 r.km). The amount of mould fungi was not considerable at the Middle-Tisza. The total count of mould fungi has increased to the magnitude of hundred at the reach of the Lower-Tisza located on the area of Tiszaug and Tiszasziget (266.4 – 167.0 r.km).

Among tributaries, we managed to isolate mould fungi in the largest number from the sediment of the Bodrog (Table 3).

Yeast fungi were demonstrable only in two sediment-samples of the reach of the Upper-Tisza (568.7; 559.9 r.km). We have not managed to culture these fungi in the sediment-samples of the Middle- and Lower-Tisza.

Table 3. contains the data referring to the amount of desulphurylating and ammonifying bacteria. However, as the results of measurements are only approximate, therefore they have not been suitable for drawing a diagram.

Discussion

From the results we can conclude that after marching down of pollution in the seventh month the hygienic bacteriological state of water of river Tisza developed according to the characteristic of the autumn state with low water. The effect of cyanide pollution on the aquatic communities of microbes has already not been demonstrable.

According to the hygienic water-bacteriological examinations, the Upper-Tisza located upstream the mouth of the Szamos belongs to the I-II. classes. Downstream the mouth of the Szamos (Gergelyugornya – Záhony), each examinations with regard to hygienic bacteriological group belong to the IV. class ('polluted').

At the following reach of the river (Záhony – Tiszafüred), water quality is of I-II classes. On the basis of the number of aerobe organic-material decomposers water belongs again to the III-IV. classes from Tiszafüred to Tiszaug. In the area of Tiszaug – Tiszasziget the quality of the Tisza is of III-IV. classes according to every bacterium-group. The section located downstream the mouth of river Szamos (Gergelyugornya – Vásárosnamény) and the section of Tiszaug – after receiving sewage water of Szolnok – are very polluted by faecal way.

Among tributaries of the Tisza, the Lónyai-channel and the Szamos were the most polluted from hygienic bacteriological point of view. Positive activity of *Salmonella* was detectable in the Szamos. *Pseudomonas aeruginosa* was only demonstrable from the sections of the Tisza located upstream (687.0 r.km) and downstream (683.0 r.km) the mouth of the Szamos.

The results of bacteriological examinations with the character of material-cycles, performed from sediment are considered as a basic-research at the Hungarian reach of river Tisza. Namely, in the course of the investigation of Tisza performed in its longitudinal section in 1979 only the hygienic bacteriological examination of sediment took place.

According to the results of bacteriological examination of sediment performed in 2000, the

bacterium-groups having importance from both hygienic aspect and the aspect of material-cycles were present in a huge number at the reach of the Upper-Tisza (Gávavencsellő – Tiszaújváros). The amount of the microbiota elements of sediment has decreased in the Middle-Tisza, while the hygienic bacteriological parameters were the most disadvantageous at the Lower-Tisza and the presence of such a bacterium-biota was detectable which had been working increased way from the aspect of material-cycles. According to the results of hygienic bacteriological examinations performed from the sediment of the tributaries, the sediment of the Lónyai-channel, Hármas-Körös and Maros proved to be the most polluted. The microbiota elements participating in the microbial cycles of elements were the most active in the sediments of rivers Hármas-Körös, Bodrog and Maros.

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References

- B. Tóth, M. (1981): Bacteriological study of the sediment in the Tisza and its tributaries. — *Tiscia* 16, 55-64.
- Estók, B. (1980): Sediment investigations carried out in the longitudinal section of the Tisza and in the mouth of its major tributaries for establishing the presence of faecalindicator bacteria. — *Tiscia* 15, 27-34.
- Estók, B. (1981): Faecal indicator bacteria of the sediment in the Tisza and at the mouth of its greatest tributaries. *Tiscia* 16, 75-82.
- Gorzó, Gy., Reskóné Nagy, M. and Kupainé Pálfi, K. (1998): Bacteria participating in cycles of elements. — In: Némedi, L. (ed.) *Environmental Bacteriology KGI*, Budapest, pp. 83-86.
- Hegedűs, M. and Zsikó, M. (1981): Changes in the *Clostridium* count of the sediment in the longitudinal section of the Tisza. — *Tiscia* 16, 65-71.
- Hobbie, J. E. , Daley, R. J. and Jasper, S. (1977): Use of nuclepore filters for counting bacteria by fluorescence microscopy. — *Applied and Environmental Microbiology* 33, 1225-1228.
- MSZ 12750-18.1974. Felszíni vizek vizsgálata. Nitrátion meghatározása. (Testing of surface water. Determination of nitrate ion.) Budapest.
- MSZ EN ISO 26461-2.1994. Vízminőség. A szulfidredukáló anaerobok (clostridiumok) spóráinak kimutatása és számlálása. 2. rész: membránszűrési módszer (Water quality. Detection and enumeration of the spores of sulfite-reducing anaerobes. Part 2: Method by membrane filtration. EN 26461-2:1993) Budapest
- MSZ EN ISO 6222. 2000. Vízminőség. Tenyésztető mikroorganizmusok számának meghatározása. Telepszám meghatározás agar táptalaj beoltásával. (Water quality. Enumeration of culturable micro-organisms. Colony count by inoculation in a nutrient agar culture medium. ISO 6222. 1999) Budapest.