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INFLUENCE OF RIVER WATER CONTAMINATION ON DISTRIBUTION OF FISH SPECIES

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Резюме: *В резултат на това обширно изследване, главният извод, който е направен е, че най-разпространеният замърсител на реките в изучения транс-граничен регион между 4 европейски страни България, Сърбия, Македония и Гърция са нерегламентираните сметища за битови отпадъци и битовите и отходни канали, резултат от жизненият процес на местното население без да се спазват законите на националното и европейско законодателство. Едновременно с това местните власти прилагат лошо управление на отпадъците. Вторият голям източник на замърсяване са фермите и строителните дейности. Изплзването на изкуствени и естествени торове над допустимите норми и изхвърлянето на отходни води от животновъдството, както и сторителните дейности, голяма част от които битови замърсяват водата и почвите в района, който не е добре развит в индустриално отношение. Установи се, че рибните видове се влияят от замърсяването на водата. Като био-индикатор е използван вида Балканска пъстърва (Salmo trutta fario), която се влияе от концентрациите на цианурова киселина във водата, а такива съдържания се установиха в реките Бистрица и Новоселска, а водата на река Банщица е замърсена освен от битови отпадъци и отходни канали, също и от изливаща се в нея минерална вода. Проектът е финансиран от три програми: Проект "Екологичен мониторинг на река Драговищица" по Програми BG02 и BG03 по Финансовия механизъм на Европейското икономическо пространство с бенефициент Сдружение "Български икономически форум" и програмен оператор Министерство на околната среда и водите, Проект "Учи-БАН" на Българска академия на науките и Проект"Европейско състезание за млади учени EUCYS 2015" на Министерство на образованието и науката.*

General Information

Studied area is situated in South-West Bulgaria, near mountain Osogovo (Fig.1). It is located in the Central part of the Balkan Peninsula (South Europe) near town of Kyustendil with average altitude of 512 m. The climate of the town is transitional Continental to Mediterranean. Rainfalls are not very intensive with an annual rate of about 589 mm. They are almost equal distributed over the seasons. Snow falls from November to March and the average thickness of snow is 30 cm with duration about 15 days. However, the area has many water sources: rivers, springs, lakes, mineral and ground water [1].

The biggest river in the area is the Struma River. The Struma River springs from 2246 m above sea level, closely to the peak Cherni Vruh (2290 m) of the Vitosha Mountain, Bulgaria. The length of the river is 415 km from which 290 km are situated in Bulgaria and the rest 125 km are situated in Greece, where the river flows into the Mediterranean Sea (Fig.1). By this way it might appear as a cross-border pollutant. On the territory of Kyustendil Municipality, the river has four big feeders: Dragovishtitsa River, Bistritsa River, Novoselska River, and Banshtitsa River. Dragovishtitsa River which length is 70 km is a right tributary to the Struma River and flows through Southeastern Serbia (45 km) and Western Bulgaria (25 km). Bistritsa River has 51 km length. Banshtitsa River is 22 km long, and Novoselska River is 25 km long. All four feeders spring at the cross-border area between Bulgaria and neighbor countries Serbia and FYR Macedonia. Information about the topic is published only by our research team [1,2,3,4,5,6,7,8].

Fig. 1. Location of the studied area, * place of sampling

The most widespread fish species in all 5 of the studied rivers are: *Leuciscus cephalus, Barbus barbus,* but most important as an environmental bio-indicator is *Salmo trutta fario (Brown trout).* This specie is under special fishing regime by the Bulgarian Government. It is sensitive to some parameters of the environment such as dissolved oxygen, temperature and most important it is sensitive to presence of Cyanuric acid (CYS) in the water. Cyanuric acid is a product of decomposition of the domestic waste in the illegal landfills along the rivers, which are the main source of contamination observed in the studied area. It is considered that this fish specie disappears when the concentration of the Cyanuric acid becomes more than 8 mg/l. Other bio-indicator for this parameter is the fresh water shrimp *Branchiopoda,* which we also studied.

All territory is a part of the European Green Belt. Total 37 248 hectares from the territory of municipality Kyustendil are included in the protected area NATURE 2000.

Fish *Salmo trutta fario (Brown trout)* is under protection and under special fishing and breeding regime.

Methods and Materials

General aim of the project is to underline the importance of the environmental monitoring for achieving preservation of the natural resources for future generations, including water, soil, air, forests, fish, agricultural lands, and etc.

Specific aim of the project is to obtain information about the quality and possible contamination of the water, sediments and soils along the cross-border Struma River and some her feeders and influence of the water contamination on the distribution of the river fish species. Study is necessary, because the biggest river in the area and its 4 flows cross large cross-border area between 4 European countries. Area has about 100% agricultural and fishing mean for the livelihood. Water of the studied rivers is used mainly for irrigation, farming, drink water, fishing, and food industry.

Project is financed by three programs: Project “environmental Monitoring of Dragovishtitsa River” on Program BG02 и BG03 of the Financial Mechanism of European Economic Area with Program Operator Ministry of Environment and Water, Project “Student Institute of BAS” of Bulgarian Academy of Sciences, and Project “European Competition for young scientists-EUCYS 2015” of the Ministry of Education and Sciences of Republic of Bulgaria.

The study is performed along the length of the feeders and along Struma River at the places of the penetrating of her feeders: Dragovishtitsa River, Banshtitsa River, Bistritsa River, and Novoselska River. The research is done in order to gather perennial data about the condition of the Struma River and to prepare a database for comparison for future studies. The data will be used to obtain valuable information about the agricultural and fishing value of the river at the time of the study.

Methods are selected in accordance to the Bulgarian National System for monitoring the environment which supports database at national and regional level. National System for Environmental Monitoring performs constant reviews in many static and mobile stations. It is coordinated with the EU environmental regulations.

The present study includes measurements of basic physical and chemical parameters of the water of Dragovishtitsa River, Novoselska River, Banshtitsa River, Bistritsa River, and the Struma River. Samples were taken and measurements were made at intervals of about 300 meters along the rivers and at the point of their penetration into the Struma River. Points of measurement are situated

at the confluence places and before and after them also. Investigation of fish passages – bio-monitoring was done using radar (sonar) for fish passages. Fish species are determined by method of interview of the fishermen.

Radiation of the water and the common radiation background were measured with a Geiger counter "Radex" RD1503.

Measurements were performed also with an instrument "Hanna" HI9813-6, which measures the acidity of the water (pH), water temperature (t, °C), electrical conductivity (EC, µS), total dissolved solids (TDS, ppm).

Another device used for testing the water in the river is spectrophotometer (colorimeter) "Lovibond".With the help of this tool are identified: free, total and combined chlorine Cl, acidity pH, cyanuric acid CYS, total alkalinity $CaCO_3$, free, total and combined copper Cu and iron Fe.

It is also used binocular (stereo) microscope USB“CETI” (STAR-24ED) with a computer program “Globecam-D” and digital microscope USB 2.0 DigiScope with white and fluorescent (blue) light in order to study the micro-detritus in the sediment of the river.

Nitrate and nitrite content in the water are measured by using test strips with a range of 0-10-25-50-100-250-500 mg/l. The team will continue environmental measurements in the future to determine the seasonal variation of the parameters of the water, as well as taking steps and decisions in case of the event of established dangerous levels of pollution.

For bio-monitoring we used portable sonar (radar) for fish passages “Fish Finder” with monochromatic LCD screen, one-ray, frequency 200 kHz, maximum depth 100 m, picture of bottom relief.

The applied method of measurement is "on spot" ("in-situ"), on terrain, throughout direct sampling ("grab samples"), because the advantages of this method are high accuracy and correctness of the research. Digital (electronic) devices are used for the accomplishment of the study because of their ability to quickly, easily and accurately measure the parameters on terrain.

Results

The air temperature during the measurement varied from 14 °C to 18 °C. The Struma River before the confluence of it inflows is relatively clean. The acidity of the water is almost neutral with a pH=7.69. Water of Dragovishtitsa River has pH=7.40, with presence of Cu (0.16 mg/l) and Fe (0.05 mg/l). All other measured parameters are in normal rates. Because of the clean water in this river the fish species Brown trout (*Salmo trutta fario)* is widely spread. At the point of confluence in Struma River parameters do not change significantly. At the confluence of Banshtitsa River the acidity does not change significantly, but at the confluence of Bistritsa River the acidity becomes to strong alkaline pH = 8.26, which means that Bistritsa River is contaminated probably with washing chemicals from the households. Evidence for this contamination is the high foaming at the point of confluence of the two rivers. Canals for domestic waste water are observed. The electrical conductivity is relatively high EC = 3.25-4,20 µS , which means that the water is saturated with electrolytes. The mineral content is relatively high, probably because of the drying up of the rivers during summer period. This parameter is almost constant along the river and does not effect by the observed sources of contamination as canals and landfills. Water of Novoselska River has high content of Cyanuric Acid CYS (16 mg/l). (Fig. 2) (Table 1). There are no fish species observed. Fishermen also were not observed in the Banshtitsa, Bistritsa and Novoselska Rivers.

The amount of total dissolved solids TDS is within the normal range, with low concentrations of dissolved solids ranging from 50 to 169 (ppm). This shows low level of sodium carbonate, sodium bicarbonate and sodium chloride contamination. Only traces from nitrates and nitrites are established (according EU Nitrate Directive), probably nitrogen fertilizers are not used this season (autumn). There is presence of total, free and combined chlorine (Cl), but in low concentrations from 0 to 0.08 mg/l.

Particular attention should be paid to cyanuric acid CYS, which is in quantities of 0 to 16 mg/l. There is increased content before the feeders and in the confluence of the Bistritsa River and Banshtitsa River. This pollutant is established at place of the illegal landfills and after them along all studied rivers. Cyanuric acid CYS is a product of the chemical industry and is contained in detergents, disinfectants, adhesives and others. This water is not suitable for direct consummation and livestock watering, also the irrigation of agricultural areas may be a problem. The water has high total alkalinity (hardness $CaCO_3$). The content of calcium carbonate (Total Alkalinity) ranges from 111 to 250 mg/l and it is higher around the illegal landfills with building waste - lime, concrete, cement, ceramics.

The amount of free, combined and total copper Cu is not high, but systematic irrigation of agricultural areas may enrich the soil with copper and exceed the limit values for soils under Bulgarian State Gazette, No54/1997. According to this Gazette, for low acidic and neutral soils, Cu must be under 250-260 mg / kg, but for acidic soils – must be much lower. Free copper in the studied water

varies between 0 and 0.8 mg/l, total copper from 0.19 to 0.46 mg/l and combined copper is from 0 to 0.46 mg/l.

Free Iron (Fe) is in low concentrations from 0 to 0.05 mg/l, again it is higher before the inflows and in the places of confluence.

Radioactivity of the water, sediments and the common radiation background are normal and fluctuate around 0,16-0.20 μSv/h.

River sediments and alluvial soils are contaminated with various anthropogenic micro-detritus (microscopic fragments by human waste) as polyethylene (10%), plastics (5%), textile (10%), ceramics (7%), metals (4%), building materials (15%), glass (7%), coal, ash, rubber, paper, styropor (1% each), and others.

Discussion

Dragovishtitsa River is not contaminated. Water is relatively clean and because of this reason the fish species *Salmo trutta fario* is widely spread. There are also *Leuciscus cephalus, Barbus barbus* and some other fish species. Fresh water shrimp *Branchiopoda* is widely spread also, which is the main food for the fishes. The overall anthropogenic pollution of the river is low which results in a wider biodiversity.

Novoselska River is contaminated with Cyanuric acid, copper and high alkalinity around the illegal dumpsites. No fish species in the river. *Branchiopoda* disappears around the illegal landfills.

Banshtitsa River was contaminated with almost all studied parameters, but after our publication and letters to the Government it had been cleaned and now it is relatively pure with high hardness ($CaCO_3$) and salinity, which could be explained with the drought of the river in the end of the summer season. Biological species *Branchiopoda* disappears at places of illegal landfills at about 300 m after them. No fish species present in the river. The temperature of the water of Banshtitsa River is abnormally high because of the high amount of hot mineral water which inflows artificially into the river. It was localized near the town's swimming pool.

Bistritsa River is contaminated with washing chemicals from the villages and farms where it passes through and as a result has high alkalinity. Because of this reason foaming was observed at her confluence into the Struma River. No fish species observed into this river.

Struma River and its feeding inflows have increased conductivity (high salt/mineral content) which probably is a natural process because of the low level of rivers their salinity is increased. Electro-conductivity parameter is almost constant for each river in different measurement points. It does not help to be localized any source of contamination. The disinfectants and the detergents in Struma River have high concentrations, above 8 mg/l. Probably, because of this reason the fish species *Salmo trutta fario* is rare in this river.

Total alkalinity is high, which might be explained with the low level of rivers and the fact that it passes through carbonate rocks and dumping of construction waste at the illegal landfills along the rivers. Carbonate content increases at places of dumping of domestic and building wastes.

The water of Banshtitsa, Dragovishtitsa and Novoselska rivers contains copper and should be taken into consideration for systematic irrigation of agricultural lands and cattle-breeding. Cu presence is established around the illegal landfills, where local population dumps illegally domestic waste. One part of the Cu in the water has probably natural origin.

Table 1. Measured parameters of the water of Struma River and its feeding streams

Description	Ponit of Measurement	pH	EC, μS	TDS, ppm	t,°C air	t,°C water	NO3, mg/l	NO2, mg/l	free Cl, mg/l
Struma River before the streams	1	7.69	3.50	150	18	11	2.5	5	0.08
Bistritsa River	2	8.26	3.60	75	18	11	0	0	0
River Struma before flowing of the Bistritsa River	3	7.15	4.20	79	14	11	2.5	5	0.08

Banshtitsa River	4	7.34	3.34	169	15	12.5	0	0	0
Struma River after flowing of Banshtitsa River	5	7.79	3.25	80	16	11	0	0	0.39
Novoselska River	6	7.55	3.20	120	16	11	2.5	5	0.12
Dragovishtitsa River	7	7.40	3.48	92	18	12	0	0	0

Description	Ponit of Measu-rement	total Cl, mg/l	Combi-ned Cl, mg/l	Cianuric acid CYS mg/l	Total alkalinity CaCO3, mg/l	free Cu, mg/l	total Cu, mg/l	combined Cu, mg/l	Iron Fe, mg/l	radiation backgroun d µSv/h	radiaiton water, µSv/h
Struma River before the streams	1	0.07	0	7	250	0.10	0.37	0.27	0.04	0.20	0.28
Bistritsa River	2	0	0	12	198	0.80	0.27	0	0	0.16	0.20
River Struma before flowing of the Bistritsa River	3	0.15	0.07	0	250	0.27	0.28	0.01	0	0.20	0.22
Banshtitsa River	4	0	0	5	155	0.13	0.19	0.06	0.04	0.18	0.20
Struma River after flowing of Banshtitsa River	5	0	0	0	197	0.00	0.46	0.46	0	0.16	0.20
Novoselsk a River	6	0.10	0	16	160	0.18	0.32	0.14	0.12	0.20	0.20
Dragovishti tsa River	7	0	0	0	111	0	0.16	0.16	0.05	0.16	0.16

The river sediments contain copper minerals as virgin copper, chalcopyrite, malachite [9].

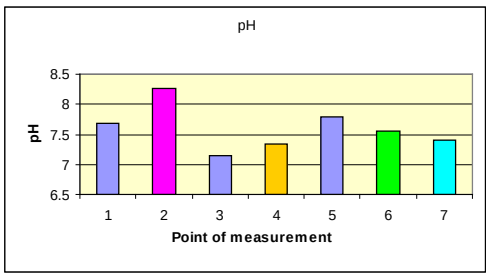
Anthropogenic micro-detritus is widely spread in the sediments and soils and measures for this contamination must be taken. Such as localization, more strict regulations of illegal dumpsites and cleaning of river beds and shores.

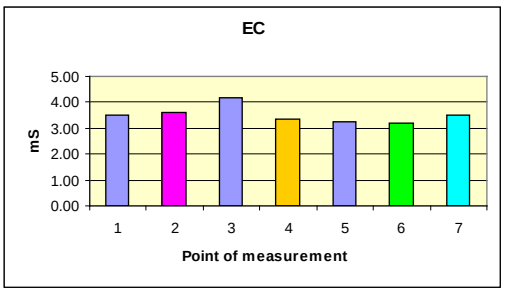
Quality of the water and soils along the studied rivers influence directly on the quality of the crops, livestock, produced food, and fish species. For example as a results of abuse of nitrogen fertilizers traces of nitrates and nitrites have been established in different vegetables tomatoes and

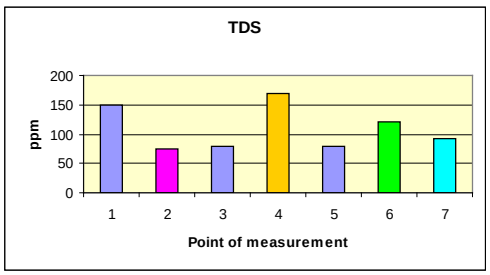
cucumbers of local producers. The bio-indicator fish specie *Salmo trutta fario* disappears in places with high concentration of Cyanuric acid.

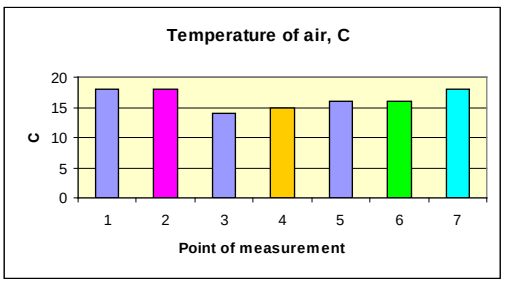
Through using of microscope with fluorescence (blue) light it was established polyethylene micro-detritus in soils, sediments, but also additional investigations established microplastics in nylon-packaged food as cheese, yellow-cheese, sausages, and in dog and birth excrements.

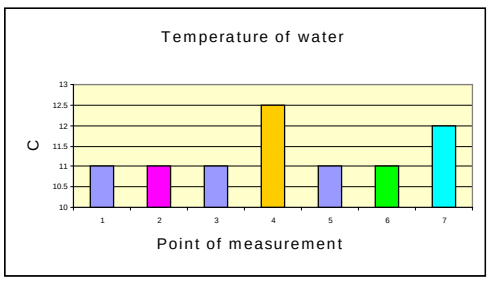
Polyethylene fragments may become a part of the food chain and might cause strangling of the animals, occlusion of the digestive tract, trauma of the internal organs, necrotic and inflammation of the intestines, intoxication of man and animals. Presence of the anthropogenic micro-detritus might become poor the soil and might change the soils structure. Pure plastics have low toxicity due to their insolubility in water and because they are biochemically inert, due to large molecular weight. Plastic products contain a variety of additives, some of which can be toxic. For example, plasticizers like adipates and phthalates are often added to brittle plastics like polyvinyl chloride in order to make them pliable enough for usage in food packaging [10].

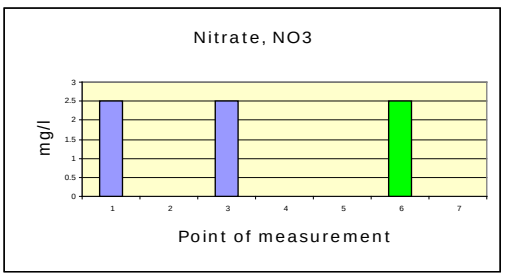


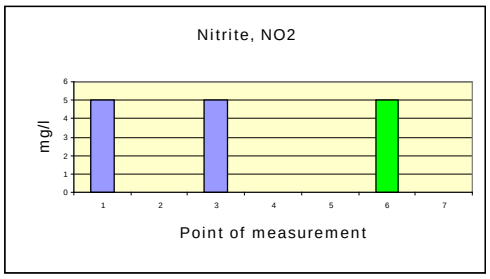


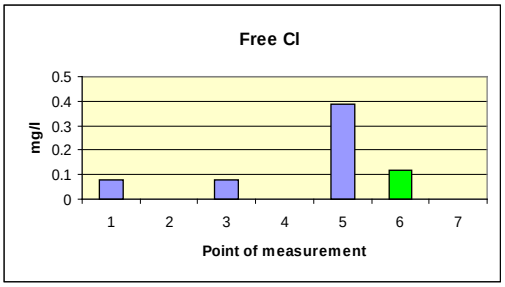


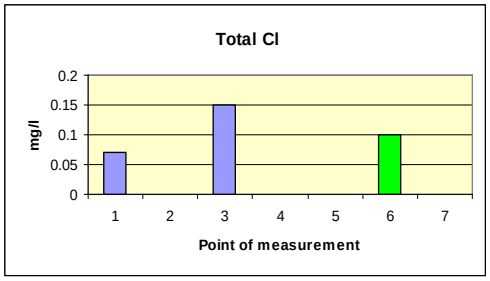


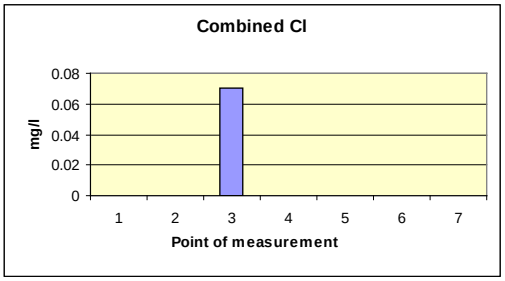












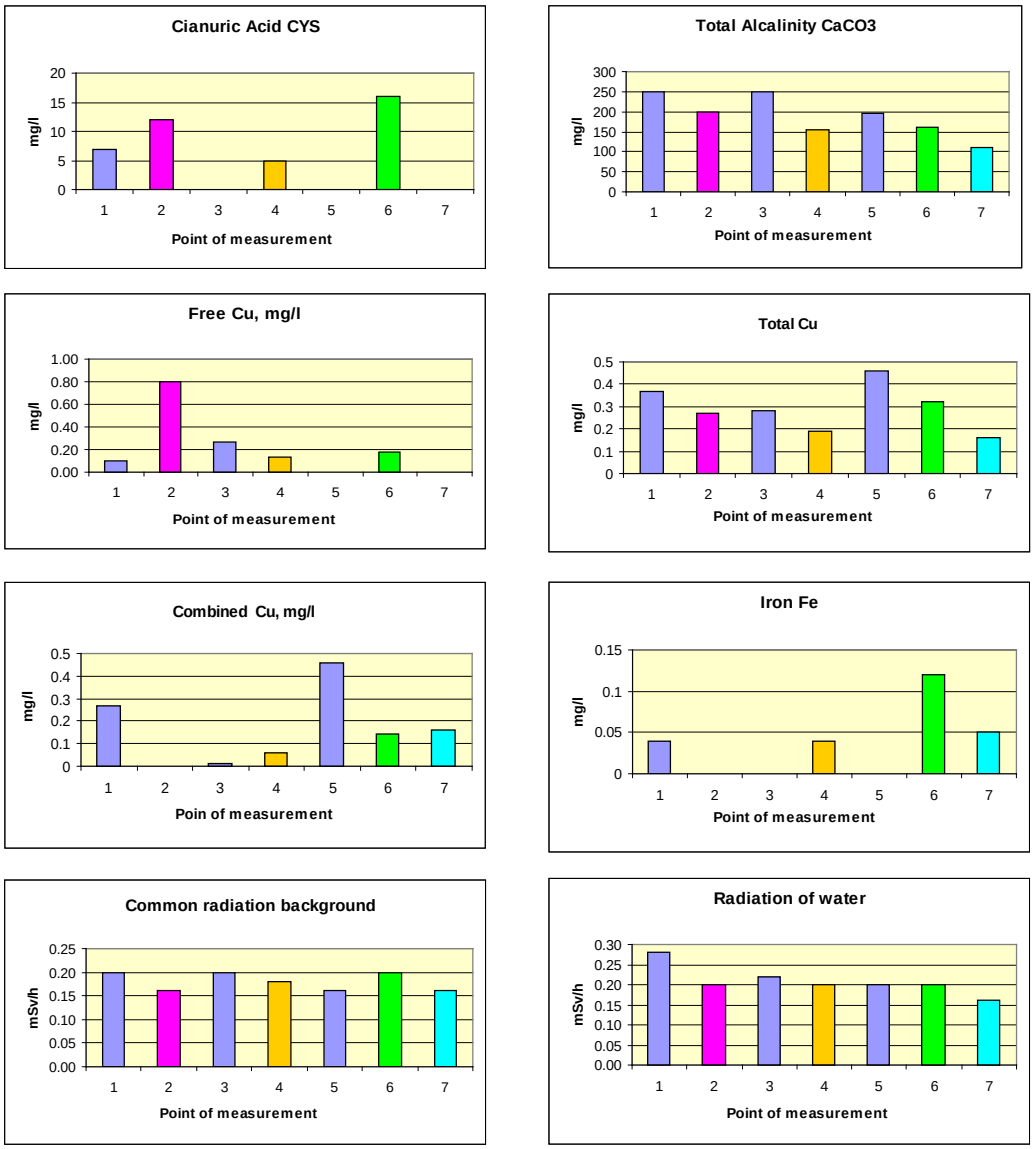


Fig. 2. Diagrams of the measured parameters of the water of Struma River and its feeders

Conclusions

As a result of this vast investigation, the main conclusion done is: the most widespread sources of contamination in the studied cross-border Euro-region between 4 European countries (Bulgaria, Serbia, FYR Macedonia, and Greece) are the illegal landfills, dumping waste and household waste water canals, result of the living process of the local population without respect to any National and EU legislation. Equal to this harm is the bad work of municipal and governmental authorities about waste management of the area. Second source of contamination is farming and construction activities. Using of fertilizers is widespread in the area during spring and summer period. Farming and construction waste dumped illegally in river beds also contaminates the water and soils in the area. Region is not industrial developed and industry is not the main pollutant of the area.

It is established that population of the fish species closely relates to contamination of the water. Contaminated rivers do not have any fish species. Feeding streams bring contamination into Struma River and decrease population of some fish species as Brown trout fish (*Salmo trutta fario).* This specie disappears in water, contaminated with Cyanuric acid CYS (Novoselska and Bistritsa Rivers) or in water contaminated with thermal mineral water (Banshtitsa River).

References:

1. Ivanchev, E. Kyustendil. "James Kraikov", 1996, 98 pp. (In Bulgarian)
2. Sotirov, A., Malwood, D., Pistalov, N., Vezenkova, R., Yerusalimova, M., Stanchev, L., Rasulski, T., Savova, S. Anthropogenic micro-detritus as a factor for co-temporary soil and sediment forming. Journal of Ecological Engineering and Environmental Protection, 2015, 3, 15-21.
3. Sotirov, A., Malwood, D., Pistalov, N., Vezenkova, R., Yerusalimova, M., Stanchev, L., Rasulski, T., Savova, S. Influence of some feeders on contamination of Struma River. Journal of Ecological Engineering and Environmental Protection, 2014, 3-4, 25-32.
4. Sotirov, A., Malwood, D., Pistalov, N., Vezenkova, R., Stanchev, L., Rasulski, T., Savova, S. Comparison of the data for environmental monitoring of River Novoselska and River Banshtitsa, region Kustendil, Bulgaria. Proceedings University Annual Scientific Conference, Veliko Turnovo, Bulgaria, July 2014.
5. Sotirov, A., Vezenkova, R., Yerusalimova, M., Savova, S., Stanchev, L., Rasulski, T. Quality of the water of Novoselska River, intended of the future reservoir Kyustendil. Second scientific conference of ecology (SACE), Plovdiv, Bulgaria, November 2013.
6. Sotirov, A., Yerusalimova, M., Vezenkova, R., Savova, S., Stanchev, L., Rasulski, T. Environmental monitoring of Banshtitsa River – part 2. Journal of Ecological Engineering and Environmental Protection, 2013, 3-4, 34-42. (in Bulgarian)
7. Sotirov, A., Vezenkova, R., Pistalov, N., Savova, S., Stanchev, L., Rasulski, T. Environmental monitoring of town Kyustendil. Journal of Ecological Engineering and Environmental Protection, 2013, 1, 19-28. (in Bulgarian)
8. Sotirov, A., Vezenkova, R., Savova, S., Stanchev, L., Rasulski, T. (2013). Environmental monitoring of Novoselska River, Kyustendil region. XII National conference of Chemistry for students, Sofia University “St. Kliment Ohridski”, Sofia, Bulgaria, May, 2013.
9. Vitov, O., Sotirov, A. Mineral content of the sands from rivers in Kyustendil region with ecological risks for the quality of the soils, water and building materials. Journal of Ecological Engineering and Environmental Protection, 2014, 3-4, 86-93. (in Bulgarian)
10. Wright, S.L., Thompson, R.C., Galloway, T.S. The physical impacts of microplastics on marine organism: A review. Journal of Environmental Pollution, Elsevier, 2013, 178, 483-492.