ASSESSMENT OF THE ECOLOGICAL STATUS OF VARIOUS LOTIC ECOSYSTEMS FROM THE H.B. JIU USING BIOTIC COMMUNITIES ACCORDING TO THE WFD REQUIREMENTS

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Abstract. The paper presents aspects regarding assessment of ecological status of some aquatic lotic ecosystems and characterization of biotic typology by inventorying the biotic communities according to the E.U. Framework Directive in the field of water policy and Romanian Law 310/2004. The following biotic communities were inventoried in the chosen 8 river sections: phytoplankton, phytobenthos, macrophytes, macroinvertebrates, fish fauna. Specific parameters were calculated, then bioindicator values for each case and subsequent ecological status were assessed. Anthropic impact was generally of low importance or not visible in upstream/reference sections. A more important impact was observed on the mainstream of Jiu river, amplified by human settlements (Tg.Jiu, Craiova) and by mining facilities from sub-mountain areas.

Key words: Jiu river, biomonitoring, aquatic biotic communities, ecological status assessment

Rezumat. Evaluarea stării ecologice a diferitelor sisteme ecologice acvatice lotice din bazinul hidrografic Jiu pe baza comunităților biotice conform Directivei Cadru a apei. Lucrarea prezintă aspecte privind evaluarea stării ecologice a unor ecosisteme acvatice lotice și caracterizarea tipologiilor biotice prin inventarierea comunităților biotice în conformitate cu Directiva Cadru a Uniunii Europene din domeniul politicii apei și cu legea 310/2004. Următoarele comunități biotice au fost inventariate în cele 8 secțiuni de râu studiate: fitoplancton, fitobentos, macrofite, macronevertebrate și fauna piscicola. S-au calculat parametri specifici, apoi au fost evaluate valoarea bioindicatoare pentru fiecare caz și starea ecologică respectivă. Un impact mai important a fost observat pe cursul principal al Jiului, amplificat de așezarile umane (Tg. Jiu, Craiova) și de exploatarea minieră din zonele submontane.

Cuvinte cheie: râul Jiu, biomonitoring, comunități biotice acvatice, evaluarea stării ecologice

INTRODUCTION

The ecological status evaluation of the aquatic ecosystems has to be done based on several biological elements: phytoplankton and fish fauna from water column, phytobentos, macrophytes and macroinvertebrates from substrate, according to WFD and Law 310/2004 requirements. These elements and their parameters have to be used for biotic typology definition, reference conditions and ecological status limits, as well. Studying the above mentioned biological elements allows these 5 ecological status to be defined: high, good, moderate, poor, bad.

Therefore, for complete characterization of the lakes and rivers in Romania it is absolutely neccesary to use data about aquatic flora and fauna. Methodologicaly speaking, based on case studies, biological elements are studied according to WFD and Law 310/2004 requirements. The main biotic communities are catalogued and the WFD recommended parameters are mentioned. The determinations are made down to the lowest level possible: species, genus, family, in order to permit the best and complete characterization of the rivers and lakes types and their differentiation.

The aspects taken into account for lotic aquatic ecosystems are: compozition and abundance of the macroinvertebrates fauna, compozition and abundance (biovolume) of the aquatic macrophytes, compozition, density and abundance of the phytobenthos, compozition, abundance and age structure for fish fauna, taxonomic diversity and biomass of the phytoplankton.

The following objectives were: the inventory of biotic communities specific to different lotic aquatic ecosystems, based on tipologic categories and ecoregions, with case study within catchment area and ecoregion (Jiu); bringing the information up to date about representative biotic elements belonging to aquatic ecological systems, according to WFD and Law 310/2004 requirements; the evaluation of actual ecological status of different aquatic ecological systems based on biological elements and charaterization of biotic typology.

MATERIAL AND METHOD

In order to reach the targeted objectives, together with MMGA (MMDD) and ANAR reprezentatives it has been agreed to investigate aquatic ecosystems belonging to Jiu river catchment area. The following sections from 4 rivers have been investigated: : Jiul de Est – Cimpa, Jiu – Balteni, Jiu – Malu Mare, Jiu – Zaval; Gilort – Bengesti; Motru – upstream Valea Mare reservoir, downstream Valea Mare reservoir; Balasan – upstream Motatei. These sections have been selected based on actual monitoring network and accesibility.

The 8 investigated sections belong to several types: RO 01a, RO 02a, RO 03a, RO 15a, RO 17a and RO 18^a. All these sections have silicious substrate.

The next steps have followed after identifying the representative sections: field evaluation, sampling (quantitative and qualitative sampling using Romanian and European methodologies) followed by laboratory analysis, data processing and ecological status evaluation according to WFD requirements. Phytoplankton, phytobenthos, macroinvertebrates, macrophytes have been sampled and fish fauna was investigated by Bacau University specialists as well.

A new element is represented by macrophytic vegetation inventory using Kohler method in its modified form made by prof. Georg Janauer (Viena University), a modern method with minimum impact on this biotic community, the samples being qualitative ones, for confirmation of species identification, the qualitative aspects (biovolume in corelation with biomass) being estimated by standardized precedures.

The method is based on aquatic macrophitic vegetation (characeae, aquatic briofite, ferns, spermatofite – hydrofite and helofite as well, and in special cases filamentous algae) within continous inventory units with an individual length, each one being a unit with ecologic uniformity. For all presented species in each inventory unit the EMP is estimated (Plant Mass Estimate, "*Pflanzenmenge*", Kohler et al., 1971 – mass plant estimation) using a 5 level scale: 1 = rare; 2 = occazional; 3 = freevent; 4 = abundant; 5 = very abundant, prevailed. EMP takes into account the vertical development of the plants groups, not only the horizontal one. In general, EMP is equivalent with 3-D abundance, plants quantities and bio-volume. AS numerical interpretation, the used scale respect a mathematical function a 3rd power (Melzer et al. 1986; Janauer et al. 1993; Schneider 2000).

The results have been presented within standardised form, developed by prof. Janauer based on Kohler method. The automatic calculation of parameters was available using electronic access starting in 2002 (web address www.midcc.at). The recorded data has been processed and the numerical derivation of EMP has been obtained, with graphic inter-comparison representation. The distribution diagram shows each species quantity/biovolume (EMP) within each inventory unit.

The data regarding EMP field colected is the base for numerical calculation of MRP (*Relative Plant Mass*), abundance index/average mass (*Mean Abundance/Mass Index*) and Distribution Ratio (Steinberg et al. 1995, Janauer at al. 1993; Kohler&Janauer 1995; Pall&Janauer 1995) and standard diagrams allow the comparison between quantitative relations on macrophytes within lotic and lentic ecosystems and avoiding the errors.

Romanian and European key works have been used for the identification. Lists with identified species and parameters for each sample have been made. The taxonomic identification have been performed in ICIM.

RESULTS AND DISCUSSIONS

Phytoplankton

Regarding the phytoplankton, hydrological conditions and anthropic impacts were the main factors that had a strong influence on the specific composition of this community and specific quantitative indicators.

A limited specific composition was observed with 28 taxa belonging to Bacillariophyta and Chlorophyta. The diatoms were prevailed (80%).

The algal density showed values between 10.000 cel/l (Jiu river– Zaval, Motru river – upstream Valea Mare) and 59.000 cel/l (Gilort river – Bengești), the diatoms being the most abundant group (until 100%).

The phytoplaktonic biomass has varied between 0,02 Motru river – downstream Valea Mare and 0,11 Gilort river – Bengeşti, with predominance of diatoms.

Both, oligo- and oligo-betamezosaprobic organisms (clean water specific: *Ceratoneis arcus, Coconeis placentula, Diatoma hiemale, Gomphonema constrictum, Actinastrum hantzschii, Ulotrix tenuissima*), and beta and beta-alfamezosaprobic organisms (polluted water specific: *Nitzschia sigmoidea, Pinnularia viridis, Synedra ulna*) were observed in different investigated stations.

Phytobenthos

The phytobenthos is considered a representative community for watercourses. The phytobenthos analysis is consisted of specific composition, density, abundance. 70 taxa belonging to the following groups have been inventoried: Cyanophyta, Pyrrophyta, Bacillariophyta, Euglenophyta, Chlorophyta. The most taxa belong to diatoms – 53, then cloroficee – 11, euglenoficee – 4. Some phytobenthos taxa have been observed within phytoplanktonic communities as well.

In some water bodies the diatoms were the prevailed algae – 100% (Jiu river – Balteni, Motru river – downstream Valea Mare). The common taxa were: *Ceratoneis arcus, Coconeis placentula, Diatoma hiemale, Mastogloia Braunii, Melosira granulata, Nitzschia linearis, Nitzschia sigmoidea, Chlorella vulgaris, Genicularia spirotaenia.*

The numerical abundance shows the dominance of the diatoms -75,5%, and then cloroficee -15,7%, euglenoficee -6%, cianobacteria and piroficee -1,42%.

The densities of the benthonic algal communities have varied between 11.000 ex/m² (Jiu river – Cimpa) and 53.000 ex/m² (Gilort river – Bengeşti). The abundances again show the dominance of diatoms.

Based on bioindicators and using the Pantle-Buck method, the saprobic index was calculated with values between 1,10 upstream Valea Mare and 1,71 Balasan river – Motatei (tabel 1).

 Table 1. Values of saprobic index (s) calculated considering phytobenthos from watercourses belonging to Jiu River catchment area (september 2006)

No.	Sampling station	Saprobic Index	Saprobic zone	Ecological status
1	Gilort - Bengesti	1,20	oligosaprob	high
2	Jiu - Cimpa	1,14	oligosaprob	high
3	Jiu - Balteni	1,56	o-β - mezosaprob	high
4	Jiu - Malu Mare	1,65	β - mezosaprob	good
5	Jiu - Zaval	1,40	oligosaprob	high
6	Motru - up. Valea Mare	1,10	oligosaprob	high
7	Motru - do. Valea Mare	1,50	o-β - mezosaprob	high
8	Balasan - up. Motatei	1,71	β - mezosaprob	good

Tabelul 1. Valorile indicelui saprob (s) calculat pe baza fitobentosului din râuri aparținând b.h. Jiu (septembrie 2006)

A light increasing of contamination was observed alos upstream and downstream along Jiu river. The Valea Mare dam and reservoir influenced the development of phytobenthos communities, downstream the composition and the density of benthonic algae were decreased.

Macroinvertebrates

110 taxa from different groups have been cattalogued: Oligochaeta, Gasteropoda, Bivalvia, Hirudinea, Isopoda, Amphipoda, Ephemeroptera, Plecoptera, Trichoptera, Odonata, Coleoptera, Diptera. The distribution was the following: true flies -39 taxa, mayflies -21 taxa, caddis flies -20 taxa.

The number of taxa was low. Between 6 (Jiu – Malu Mare) and 42 (Jiu de Est – Cimpa) taxa per station were calculated depending on abiotic characteristics and the value of anthropic impact. A dramatical decrease of taxa number was observed along Motru river due to hydromorfological changes and other disturbance factors, from 26 (upstream Valea Mare reservoir) to 9 (downstream Valea Mare reservoir) taxa. Along the Jiu river a decrease of taxa number was observed from upstream (Cimpa) to downstream (Malu Mare) due to anthropic impact. At Zaval, an increase of taxa number was established, most likely due to the Danube water influence.

The proportions of different groups were different. On Gilort river the mayflies represented 38% from the total number of taxa, while the true flies and caddis flies were 19% each. Along Motru river, upstream reservoir, the mayflies represented 38%, followed by caddis flies (28%) and true flies (24%).

A certain number of taxa exist in more than one investigated stations. For instance, *Acentrella sinaica, Eukiefferiella gracei, Baetis rhodani, Limnodrilus udekemianus* and *Baetis* sp.

The most determinations were achieved at the species and genus level. The ratio of determinations varied between 89-100% (Balasan river – upstream Motatei). The number of the groups varied from 3 (Jiu river – Malu Mare) to 7 (Balasan river – upstream Motatei). The presence of many invertebrates groups and their balanced development shows the potential of reference station.

Quantitatively, the densities have varied in large limits. For instance, along the Jiu river the densities decreased from upstream to downstream, except Zaval station. At upstream Motatei on Balasan river the densities showed the dominance of mayflies (44%) and true flies (39%).

The most identified forms were bioindicators for oligosaprobic, oligo-betamezosaprobic and betamezosaprobic zones, taking into account the specific of investigated stations as reference or the best available sections for different types. Upstream Valea Mare reservoir on Motru river, 12 oligosaprob taxa and 2 oligo-betamezosaprob taxa were identified from a total number of 25 taxa (56%). These taxa are considered sensitive. At Bengesti station, on Gilort river, 5 oligosaprob taxa and 1 oligo-betamezosaprob taxa represented 50% from all taxa in the sample. On Jiu river, at Malu Mare, sensitives taxa have not observed.

In order to point out the presence of sensitive taxa, the ecological status of rivers and the intensity of anthropic impact EPT (efemeroptera-plecoptera-trichoptera) index was used. The value of this index increases with water quality. The EPT index is given by the number of 3 mentioned groups divided to the total number of taxa and shows the richness of sensitive taxa (table 2).

Table 2. Variation of EPT index from watercourses belonging to Jiu River catche	ment area (september 2006)
Tabel 2 . Variatia indicelui EPT în cursuri de apă apartinând bh Jiu (su	eptembrie 2006)

Investigated station	Total	EPT Index				
	no.of	(absolute value)			(%)	
	taxa	Total	Е	Р	Т	
Gilort - Bengesti	16	11	6	2	3	69
Motru - up. Valea Mare	25	19	8	4	7	76
Motru - do. Valea Mare	9	6	3	0	3	57
Balasan - up. Motatei	26	6	4	0	2	23
Jiu - Cimpa	42	22	6	7	9	52
Jiu - Balteni	12	6	4	0	2	50
Jiu - Malu Mare	6	1	1	0	0	17
Jiu - Zaval	16	4	3	0	1	25

The EPT index values were higher on Gilort river (69%) and Motru – up. reservoir (76%). Due to human impacts the values of EPT index decresed along the Jiu river.

Taking into account the position of investigated stations within mountain and hill areas, the dominance of scrapers, shredders and predators feeding groups was observed. At the downstream stations the number of detritivourous and filtrators increases. The presence of scrapers and shredders deepend on subtrate bioderma. The analysis on feeding groups for all taxa in a sample shows for instance at Cimpa the balance among scrapers (13-31%), predators (12-29%) and shredders (11-26%).

Fish fauna

37 fish species have been identified in all investigated water bodies. Some species have been observed only in mountain rivers, very well oxygenated ones, belonging to oligosaprobic zone. For instance, the trout was observed on Jiu de Est river, Motru river – upstream and downstream reservoir.

All data regarding numerical and gravimetrical fish stock have been recorded. The ecological status of species: indigenous or introduced, feeding regime, ecological preferences were recorded as well. From 37 observed species only 2 are acclimatized: *Pseudorasbora parva* and *Lepomis gibbosus*.

The biggest number of taxa were observed at Zaval (20) on Jiu river, and the smallest number on Motru river – downstream reservoir and on the Jiu de Est river at Cimpa (2). Regarding the number of captured specimens the most representative fish assemblages were on Jiu river at Zaval (276) and at Balteni (516). The densities at 100 m2 varied, taking into account the station and the water body specifics, from 2,5 at Motru - downstream reservoir to 48,75 at Balasan, upstream Motatei.

On Gilort river, at Bengesti, most of the 167 observed fish specimens belonged to the species *Barbus petenyi* (99), *Phoxinus phoxinus* (32) and *Orthrias barbatulus* (20). The dominant species were *Romanogobio kessleri* and *Carassius gibelio* on Jiu river at Malu Mare; *Proterorhinus marmoratus* and *Gobio obtusirostris* on Balasan river; *Rhodeus amarus, Squalius cephalus* and *Alburnoides bipunctatus* on Jiu river, Balteni; *Phoxinus phoxinus* and *Salmo trutta fario* on Motru river, upstream reservoir; *Barbus petenyi*, on Motru river, downstream reservoir Valea Mare; *Salmo trutta fario*, on Jiu de Est river at Cimpa; *Carassius gibelio, Idus idus* and *Alburnus alburnus* on Jiu river, at Zaval.

The repartition of species is characteristic to existing habitats, the number of species depending on their size and existing impact. The number of species gradually increased starting from the springs of the rivers and with the size of habitats. Moreover, the salmonicol species present in some sections are indicators of natural or almost natural conditions, with minimal human influences. The salmonids allow for identifying the reference conditions along the rivers and the types of the rivers.

The age structure of fish population shows the potential of that population, increasing or decreasing trend respectively. *Macrophytes*

This biological community was superficially considered in the past, so the main objective of inventorying the macrophyte communities from Jiu catchment was obtaining reliable, actual information by using a standardized methodology, information to be used in conjunction with the one obtained for other biotic communities for assessment of current ecological status of the studied aquatic ecosystems according to WFD.

Quality parameters for macrophyte communities are taxonomic composition and abundancy (E.U. 2000).

For each section (either with our without macrophytes) habitat parameters were recorded. Various sets of habitat parameters were considered, to allow for relating growth forms with abiotic conditions.

A total of 31 different macrophyte taxa were recorded in all 4 inventorying sections from Jiu river - Cimpa, Bâlteni, Malu Mare şi Zăval.

For the other three rivers, results were as follows :

- Motru river (upstream and downstream Valea Mare reservoir): 7 and 15 species;
- Gilort (Bengeşti) river: 15 species;
- Balasan (upstream Moțăței): 21 species.

For each river section a *concise species list* was composed, mentioning species name and author, standard abbreviation and growth form. From the DCA or other monitoring schemes perspective only a concise species list fulfills the monitoring requirements. For each species Plant Mass Estimates were recorded for each of the survey units where the plants were seen.

Overall, the recorded 42 taxa were distributed by growth forms as follows :

- acro-pleustophytes (plants floating on the water surface): 1 species;

- submersed pleustophytes (plants neither floating on the water surface, nor being really attached to the sediment): 1 species;

- submersed anchored (plants are fixed in the sediment or attached to the bottom: 4 species;

- amphiphytes (plants grow in the same survey unit on the bank, like a helophyte, and at the same time in the water body, like a submersed anchored plant): 6 species;

- helophytes (all bank vegetation still clearly associated with the water body): 30 species.

In every case helophytes were dominant plants, amphiphytes being relatively under-represented. Submersed anchored macrophytes were recorded in most sections, but in small amounts, represented only by a species of chlorophyta (*Cladophora glomerata*), one bryophyta (*Fontinalis antipyretica*) and 2 species of spermatophyta (*Cardamine amara* and *Ceratophyllum demersum*).

It is known that reference sections of rivers from mountain areas are generally unfavourable for macrophyte growth. Water flow is generally high and the sediment type (gravel, stones, rocks, sand) is not suitable for proper anchoring, so only a limited number of aquatic macrophytes can be found in these habitats, helophytes being always dominant. Yet generally helophytes are not very suitable as quality indicators, hence it is difficult or impossible to assess the ecological status of those sections based only on macrophyte community.

In our survey such situations were found in several sections from Jiu river (Cimpa, Malu Mare) and Gilort (Bengesti). Anyway, inventorying the macrophyte communities allowed for some conclusion to be drawn and it proved useful for present comparisons and it will be useful for future ones.

The two sections from Motru river were interesting because they were chosen upstream and downstream Valea Marea reservoir, the upstream section being a reference one. Macrophytes were poorly represented in this section (only 7 species of helophytes, with low biovolumes), while in the downstream section they showed a marked difference: more species and higher biovolumes, together with occurrence of few indicator species allowed for concluding that even if both sections have good ecological status, there was a subtle nutrient increase from upstream to downstream sections, confirmed also by analysis of other biological communities.

On Balasan river, the last river inventoried in Jiu catchment, upstream Moțăței section was chosen. The macrophyte communities were quite well represented, being recorded 4 species of hydrophytes, 2 amphyphyte and 15 helophyte species. Low water flow was associated with general occurence of *Lemna minor*, the only acro-pleustophyte found. The assemblage of indicator species (*Sparganium erectum, Ceratophyllum demersum* together with *Lemna minor*) in significant biovolumes marked a higher nutrient load of the water, probably caused by nearby crop fields and the pollution from fertilizers. A moderate ecological status was assigned to this section.

CONCLUSIONS

Adding up all the data obtained by studying certain biotic communities from different water bodies along the rivers allows for drawing a wide range of conclusions.

The phytoplankton had low taxonomic diversity. The diatoms were the dominant algae.

The phytobenthos communities were diversified with 70 identified taxa from different groups: Cyanophyta, Pyrophyta, Bacillariophyta, Euglenophyta, Chlorophyta. Most taxa were from diatoms.

110 macroinvertebrates taxa were inventoried in all studied rivers with organisms belonging to different taxonomic groups: Oligochaeta, Gasteropoda, Bivalvia, Hirudinea, Isopoda, Amphipoda, Ephemeroptera, Plecoptera, Trichoptera, Odonata, Coleoptera, Diptera. The number of taxa was generally low per station. The majority of identifications have been achieved at the species and genus level. Most identified forms were bioindicators and sensitive species. The values of EPT index showed the presence of natural or almost natural conditions on Gilort river – Bengesti, on Motru river – upstream reservoir, and a dramatic anthropic impact on Jiu river at Malu Mare (17%).

The saprobic index values based on phytobenthos and macroinvertebrates bioindicators revealed the high and good ecological status of investigated stations. Based on saprobic index, in general, the surveyed stations stayed within oligosaprobic, oligo-beta-mezosaprobic or beta-mezosaprobic zones. The lowest values were observed on Motru river – upstream Valea Mare reservoir and Jiu river at Cimpa, stations with little anthropic impact. On Balasan river, upstream Motatei, because of the characteristics of the station the ecological status was good. Along the Jiu river a decrease of the ecological status and water quality was observed from upstream to downstream.

37 fish species have been identified in the investigated water bodies. The biggest number of taxa was observed at Zaval (20) on Jiu river, and the smallest number on Motru river – downstream reservoir and on Jiu de Est river at Cimpa (2). The presented taxa depended on specific characteristics of the investigated water bodies. The trout was observed only on upstream stretches of the Jiu and Motru rivers, for instance. The species repartition is characteristic to existing habitats, the number of species depends on their size and human impacts. Regarding the number and spatial distribution of fish species collected in 2006 as compared with the previous situation (before 1960), a decrease of species number from springs to mouth of the rivers due to anthropic impact was noticed. The exception is at Zaval, where the Danube has influenced the fish fauna of the Jiu river.

Overall, it can be stated that significant anthropic impact was detected for the main channel of Jiu river, starting from Petrosani, impact amplified by important human settlements (Tg.Jiu, Craiova) and by mining facilities from pre-Carpathian area. Main tributaries (Motru, Gilort, Amaradia) and rivers from mountain and sub-mountain area are much less affected, native fish fauna being present in greater ratio. For a proper, precise ecological diagnosis there is a need for other surveys for the next two years, coupled with increased frequency of sampling points and sampling campaigns.

In lotic systems occurrence and distribution of macrophytes is influenced by to several significant parameters: water flow regime, river bottom type, availability of light, physico-chemical conditions of aquatic environment. All these criteria have impact upon general patterns of distribution of individual species and macrophyte assemblages. From the six groups of macrophyte species considered (Chlorophyta, Bryophyta, Spermatophyta – submersed anchored, Spermatophyta – plants floating on the water surface, Spermatophyta – amphiphytes, Spermatophyta – helophytes), three groups can be useful as water flow indicators, i.e. Bryophyta, Spermatophyta – submersed anchored, Spermatophyta – submersed pleustophytes.

As previously mentioned, there are some sections where abiotic parameters are not suitable for any macrophyte growth, not only for indicator species. Anyway, even if macrophytes are completely absent or only species without indicator value appear, there is impossible to drawn any conclusion regarding water quality and ecological

status if no other biological community, better represented, is assessed. It can be noticed that for the river sections where macrophytes were present and indicator species appeared in significant amounts, the results were similar to those obtained by studying other biological communities, such as phytobenthos. It was even possible to determine relatively small differences between water quality from one section to another, on the same river (as for sections from upstream and downstream Valea Mare reservoir, Motru river).

The investigated water bodies and rivers belonged to different types: RO01a – Jiu de Est river (Cimpa), RO02a – Motru river (upstream and downstream Valea Mare reservoir), RO03a – Gilort river (Bengesti), RO15a – Balasan river (upstream Motatei), RO17a – Jiu river (Malu Mare) and RO18a – Jiu river (Balteni and Zaval).

Sometimes it is difficult to confirm abiotic typology using biota, due to disturbance factors. For instance, on Motru river, upstream and downstream Valea Mare reservoir, there are not too many similarities between the two water bodies, even though they belong to the same type. The changes arise because of the dam and reservoir impacts which modified the hydrologic characteristics with consequences on biota.

On RO01a, RO02a and RO03a types or upper stretches of the rivers, respectively, stoneflies, together with mayflies and caddies flies have been observed. The stoneflies are missing in the middle and downstream stretches of the rivers due to human impacts. The trout characterizes only RO01 and RO02 types. Other species, like *Thymallus thymallus*, have not been captured, though they may be present in those water bodies and rivers.

Very detailed identification of aquatic flora and fauna, down to subspecies and variety level, are needed for biotic typology characterization in order to reveal the differences. The abiotic typology confirmation will take a long time, due to lack of enough specialists. A grouping of different abiotic types and also dampening their number is possible.

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