MONITORING PROGRAMME FOR

DANUBE BARBEL (BARBUS BALCANICUS)

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Range

The range of *Barbus balcanicus* includes the Danube, Nistru/Dniester, Odra, Vistula and Vardar river basins. This species is present in several neighbouring countries of Croatia, namely Slovenia, Hungary, Serbia, Bosnia, Herzegovina and Montenegro. *B. balcanicus* can also be found in Italy and Macedonia.

Distribution in Croatia

Until now, distribution data for *B balcanicus* have not been systematically collected. The little knowledge of this species in Croatia is based on the last few decades of studies in Sava, Drava, Kupa and their tributary watersheds (Figures 1 and 2). The species is common or very common in some of the Danube basin watercourses.

There has been no national long-term specific monitoring on distribution or population ecological status for *B. balcanicus*, as not all areas were studied in this respect for the proposal of Natura 2000 sites.

B. balcanicus also occurs in border area of the Alpine Biogeographical Region, in the Kupa River and in the Continental Biogeographical Region.

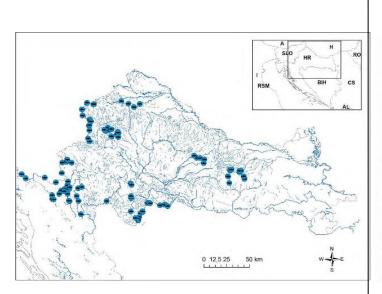


Figure 1: *B. Balcanicus* presence identified in the last years on the Continental part of the Croatian territory (prepared by SINP).

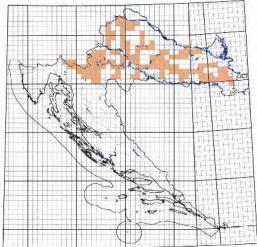


Figure 2: Distribution of *B. Balcanicus* (Mrakovčić *et al.*, 2010).





Habitat

B. balcanicus is a reophilic, lithophilic and benthopelagic freshwater fish species that lives mainly in mountainous and hilly rivers up to 500m above sea level and also some lowland streams. *B. balcanicus* prefers clear and fast flowing rivers with hard substrata, where the water temperature does not exceed 25°C. *B. balcanicus* is a short-lived species.

Phenology and population biology

B. balcanicus reaches sexual maturity in the second or third year of life. Reproduction usually occurs in the spring, but can be extended into the summer if conditions are suitable (from May to July). At the time of spawning, *B. balcanicus* gather in shoals and migrate upstream in search of favourable habitats with a gravel and stone substrate. Young fish fed mainly on benthic aquatic invertebrates (tendipedes, ephemeropterans, trichopterans, gamarids and oligochetes) and vegetation debris. Adult *B. balcanicus* also feed on fish fry and alevines.

Pressures and threats

B. balcanicus is threatened by the decrease in suitable habitats for spawning, schooling, feeding, sheltering, etc. Habitat quality has declined due to pollution, habitat modifications, degradations, destructions, disappearance (channelling, watercourses regulation, remodelling, etc.), flow regulation and water abstraction. The introduction of non-indigenous species has also been shown to have a negative impact on *B. balcanicus*. The number of locations and subpopulations of *B. balcanicus* currently displays significant flucuations.

Conservation measures

Specific recommended conservation measures for Natura 2000 sites for *B. balcanicus* are as follows: maintain or improve water quality by reducing or prohibiting pollution, build waste water treatment plants, protect natural reophylic habitats with high velocity and suitable substrate for *B. balcanicus*, prevent remodeling and slowing of river habitat (including construction of new dams), restrict regulations of banks and riverbed, prohibit extraction of sand and gravel in the riverbed, prevent entry of foreign and invasive species (namely g. *Neogobius*), achieve international cooperation in order to preserve parts of the boundary watercourses, implement revitalization and restoration of habitats and restore the watercourse to its original condition, allow natural flooding of certain areas around the river and build fish passages. Authors of the Croatian Red Book also propose to create special ichthyologic reserves, fixing limits for water flow regulation in the area of occurence, prohibition of non-native species and the construction of sewage treatment plants.

Specific measures are needed where the local situation requires action for preserving and improving the favorable ecological balance of the natural waters inhabited by *B. balcanicus*. In general, this is achieved by creating water and sediments flow conditions as close to the natural regime as possible, prohibition of alien/invasive species entry and reproduction, construction of appropriate devices for water recycling, avoiding lotic fragmentations due to different categories of buildings in the river bed, etc.

Annexes of the Habitats Directive

B. balcanicus is listed in Annexes II and IV of the Habitats Directive and also in the Annex III of the Berne Convention.

Red List

B. balcanicus is considered as a vulnarable (VU) species in Croatia. It is also a strictly protected species by law.





MONITORING PROGRAMME FOR THE CONTINENTAL BIOGEOGRAPHICAL REGION

Whilst *B. balcanicus* occurs in the border area of Alpine Biogeographical Region, in the Kupa River and in the Continental Biogeographical Region, this population is directly connected with Continental Biogeographical Region. Therefore, a monitoring programme and conservation status assessment is proposed only for the Continental Biogeographical Region.

The monitoring programme for the conservation status assessment is based on two parts. Firstly, a 6-year long surveillance (2013-2018) to establish the distribution of *B. balcanicus*. This will be achieved by field mapping and will fill gaps in knowledge about its distribution in the region. Secondly, the status of population will be monitored on plots from 2019 and the data gathered will help finalise the assessment. This approach supports the data needed for reporting and future management planning.

The monitoring programme will share the data on habitat quality with the system on evaluation of the ecological status according to the Water Framework Directive.

A similar approach should also be used for the other fish species of Community Interest, mainly Romanogobio kessleri, Romanogobio uranoscopus, Cobitis elongata, Sabanejewia balcanica and Cottus gobio.

Field workers should respect the national regulations regarding fishing and ichthyologic surveys, in particular electrofishing and avoiding hazardous substances. Field studies will only be carried out when necessary legal permits have been obtained from the Ministry responsible for nature protection and from the Ministry responsible for fisheries.

Field mapping

Objectives

The information on distribution of *B. balcanicus* is currently insufficient and this crucial component would be completed in the first monitoring period (2013-2018). The monitoring on plots should be subsequently designed based on the completed dataset on localities occupied by this species.

Field work instructions

The section of the river or stream is recorded into a map provided by SINP. The length of the section is not decisive. The mapping is carried out for 45 minutes (30 minutes if two anodes are used) on a section (including only work with electrofishing device in the water) in places where the electrofishing is possible. The time is the most useful unit of effort for this purpose, the speed of research should be oriented to catch the highest number of *B. balcanicus* individuals as possible.

The field survey must be carried out in the period from 15th April up to 15th November, excluding when maximum daily temperatures exceed 30°C (when there is increased risk of mortality due to low oxygen content in the water). The current flow cannot be higher than average flow. The electrofishing cannot be realized during in the rain because of safety.

The standard electrofishing method will be applied with the electric device set at the local water parameters. A backpacks electrofishing device could be used only in shallow water (depth not higher than 75 cm at more than 90% of the section surface). The field crew consists of a minimum of three persons (if backpacks electrofishing device is used), ideally five persons (for generators placed at the banks or in boats). As a minimum, the crew leader and person operating an electrofishing device must be specialists in ichthyology. The number of people moving in the water should be as few as possible (1-3 persons) to minimise damage on the animals and plants present.

The survey is oriented to all identifiable fish specimens going through the section. Only suitable parts for the occurrence of *B. balcanicus* are monitored. The river section should be bounded by stop nets or another temporary migration barrier. *B. balcanicus* is a species that can escape detection during the survey, therefore, it is appropriate to place the landing nets about 0.5 meters behind the anode, and use a recommended landing net mesh size of 4 mm.





All the fish will be held in containers with sufficient oxygenated water. Each individual is identified, measured to an accuracy of 5 mm and released as soon as possible back to the river section. The standard length (SL) is prescribed, whereby individuals are measured to the posterior end of body (of the last vertebra, not scales).

Sampling design

The mapping is based on surveillance in areas near the "sections of occurrence" (see subchapter Evaluation – Range). The ca. 10km sites for mapping are selected from the terminal parts of the selected "sections of occurrence" on same streams or streams with confluence between the buffer and conditions suitable for the species (using Strahler ordering, streams of 2nd and higher order). This way the mapping continues during all 6 years of the first period.

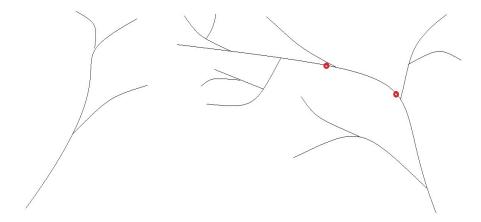


Figure 3a. Model example of species findings on partial river system with streams of 2nd and higher order draw

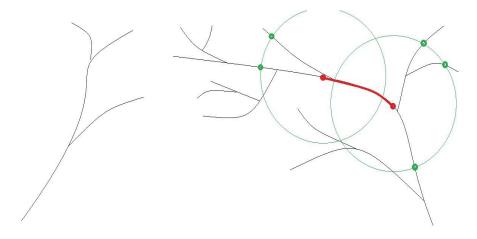


Figure 3b. Buffers and **5** sites chosen for mapping in the 1st year (green).



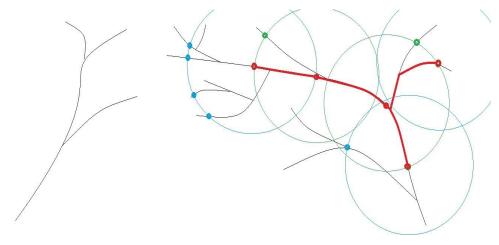


Figure 3c. Buffers and **5** sites chosen for mapping in the 2nd year (blue, all actually known occurrences in red).

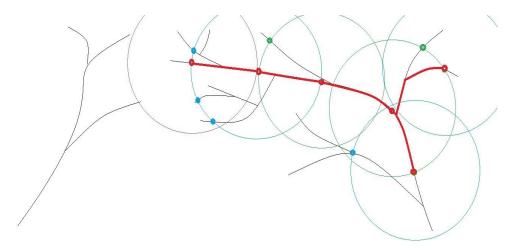


Figure 3d. Buffers and **5** sites chosen for mapping in the 2nd year (blue, all actually known occurrences in red).

The number of sites for mapping is limited to 30 sites per year. Specification of sites for specific years should be planned together with monitoring programmes for other fish species to be cost-effective.

This mapping is proposed only for the first 6-years period. It could be repeated in the future in the case of major changes in distribution (negative trend at level of > ca. 20% of the range in following periods). This repeated mapping should be oriented mainly to borders of the range and to the most endangered habitats only in the area (river basin) affected by the change.

During the period 2014-2015, the Natura 2000 Integration Project (NIP) inventory of freshwater ichthyofauna will be carried out in the areas where there are gaps in data, and could be connected with the mapping of *B. balcanicus*.

<u>Data forms</u>

The data form for mapping (and research on localities) is used (see part I of the data forms). Data on the character of the habitat, affecting pressures and conditions of the survey are recorded directly in the field in the attached data forms 1 and 2 (in white boxes). Information is then added to the electronic data form. Data on *B. balcanicus* are recorded only on paper in the field, which is subsequently analysed in the office and processed in electronic form in the structure of data form 3. Form 3 is also used for other target species (following other monitoring programmes). For species other than *B. balcanicus*, the numerical representation in the sample is completed in data form 4. The map is added to the data form 5 and representative photos to the data form 6.





Monitoring on plots

Objectives

The assessment for trends in population should be prepared according to the monitoring results. Monitoring will start in 2019 to ensure good reference values for the second monitoring period (after 2024).

Field work instructions

The field survey must be carried out in the period from 15th April up to 15th November, excluding when maximum daily temperatures exceed 30°C (where there is increased risk of mortality due to low oxygen content in the water). The current flow cannot be higher than average flow. The electrofishing cannot be realized during in the rain because of safety.

The standard electrofishing method will be applied with the electric device set at the local water parameters. A backpacks electrofishing device could be used only in shallow water (depth not higher than 75 cm at more than 90% of the section surface). The field crew consists of a minimum of three persons (if backpacks electrofishing device is used), ideally five persons (for generators placed at the banks or in boats). As a minimum, the crew leader and person operating an electrofishing device must be specialists in ichthyology.

The plot is represented by a 100 m long section of the river or stream. If the depth in the river is insufficient to permit a continuous survey and/or more than 80% of the width of the stream is less than 10 m, only a 3 m wide section along both shorelines is monitored.

The first sample will not finish before the end of 100 m long section is reached. After the first sample, each fish is measured to an accuracy of 5 mm and placed into containers with sufficient oxygenated water. The standard length (SL) is prescribed, whereby individuals are measured to the posterior end of body (of the last vertebra, not scales).

After one hour from the beginning of the first sample, the second sample can start. The full length of the plot is monitored again during the second sample. If more than 50% of *B. balcanicus* individuals are found in the second sample in comparison with the first sample, then a third sample is required for calculation of real species abundance.

The survey is oriented to all identifiable fish specimens going through the section. Profiles should be bounded by stop nets or similar temporary migration barriers.

The exact location of the plot is recorded into the map in order for comparisons in subsequent periods to be drawn.

Sampling design

The monitoring on plots follows the mapping. It is designed based on completed dataset on localities occupied by the species. Placement of monitoring plots on mapped sites is preferred as well as overlapping with monitoring plots for other fish species.

Monitoring will start in 2019. The frequency is once every 3 years for the 24 plots (i.e. 8 per year) and once year for the 8 control plots. Placement of control plots inside Natura 2000 sites is preferred. The total number of sites is 32 (16 per year).

The plots have to be placed on "sections of occurrence" completed after the mapping. The selection is based on the system of classification. Two main parameters are chosen as most representatives – Strahler order of the watercourse ($2^{nd}+3^{rd} \times 4^{th}$ and higher) and river basin (Drava, Sava and Kupa).

The proportion of the length of "sections of occurrence" between classes (combinations of these two parameters) is taken as determining the number of monitoring sites in these classes (to the ratio of 3 random plots: 1 control plot). The approximate position of sites is chosen at random by computer, and is specified on the map with regard to habitat and possibilities of field survey, and finally exactly determined in the field (in the first instance).

If there is no positive finding during first two surveys, another site is selected in the same class.





Length of "sections of occurrence" in classes:

	2 nd and 3 rd order	4 th and higher orders
Drava		
Sava		
Kupa		

	2 nd and 3 rd	4 th and higher
	order	orders
Drava		
Sava		
Kupa		

Data forms

The data form for monitoring is used (see part II of the data forms). Data on the character of the habitat, affecting pressures and conditions of the survey are recorded directly in the field in the attached data forms 1 and 2 (in white boxes). Information is then added to the electronic data form. Data on *B. balcanicus* are recorded only on paper in the field, which is subsequently analysed in the office and processed in electronic form in the structure of data form 3. Form 3 is also used for other target species (following other monitoring programmes). For species other than *B. balcanicus*, the numerical representation in the sample is completed in data form 4.

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Unsystematic data gathering

Objectives

There are available sources of information on actual occurrences of *B. balcanicus* which can be easily collected and interpreted for the purpose of determination of the species distribution.

Basic data sources are represented by:

- surveillance of other fish species organized directly by SINP
- sharing the data with Croatian Waters (mainly the data from monitoring of ecological status according to WFD)
- all ichthyologic surveys in the Continental biogeographical region (mainly done by universities and expert NGOs)

Field work instructions

No special field work is needed. All Croatian ichthyologist teams will be contacted and asked to provide information on the species occurrence discovered during different surveys in the field. Only presence data are required.

Data forms

The data form for unsystematic data gathering is used (see part III of the data forms). This form can be completed in the office since it only utilises data from other data sources. If there is any information on abundance or population structure, it should be noted into the field for "comments".





EVALUATION OF THE CONSERVATION STATUS COMPONENTS

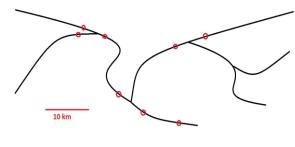
Range

The distribution of the species is connected with specific parts of rivers with high water velocity. The following approach will be used for preparation of the range and distribution map.

Range is evaluated based on distribution data during last 12 years or more if there is no actual research with negative findings (taking into account mainly results of mapping but also of unsystematic data gathering).

In the first step, all findings from the last 12 years with a distance less than 10 km are connected to "sections of occurrence" (also before starting the first monitoring period for use of selection of sites for mapping).

A)



B)

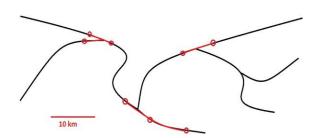


Figure 4: Model example of species findings (A) and findings distant less than 10 km connected to "sections of occurrence" (B).

In the second step, these "sections of occurrence" are prolonged 5 km upstream and 10 km downstream on occupied rivers, the result is "sections of distribution" representing the distribution of *B. balcanicus* depicted in the reporting.

In the third step, "sections of distribution" are prolonged 5 km upstream and 10 km downstream on occupied rivers and all quadrants 10x10 km overlapping obtained sections represent the range.

The favourable reference range will be specified according to principles described in official Guidelines (ETC/BD, 2011) by SINP with expert assistance of working group for fish monitoring.

Expansion or increasing of the range could be taken into account only in places where findings show *B. balcanicus* was not previously present. Other information would be interpreted as changes resulting from improvement in knowledge.

Potential loss in range could be indicated by repeated negative findings in some areas.

Population

For *B. balcanicus*, the number of individuals can be used as a population unit. This is calculated as the length of "sections of distribution" multiplied by the abundance coefficient from the monitoring plots.





This coefficient is calculated as an average number of individuals on unit of the length of the watercourse. For the first report in 2019, only the km of the river system occupied by the species calculated as the length of "sections of distribution" can be used as a population unit.

The evaluation of trends is based on the calculation of the indexes and the estimation of the total population size. The indexes are calculated in the following way: the results from one monitoring plot are averaged for one period from 2014-2019 (two numbers for standard plots, six for control plots). The results from the first period (average from 2019-2024) represent the reference baseline (100%), and it is proposed to take the value from the second reporting period as the favourable reference population, in the first report (2019) the reference value will not be evaluated. The results are representative on biogeographical level, and could be interpreted for categories of classification (except for specific river basins) and in the long-term, also on local level for specific plots.

The changes are evaluated as values of index on the Continental biogeographical level and the assessment of the component "population" follows the principles of the evaluation matrix for the conservation status assessment (for both – indexes and length of "sections of distribution", always the stricter rule is taken into account). Also if *B. balcanicus* disappears from more than 20% of plots inhabited in the previous period, the status of the component "population" must be evaluated as bad. If the species disappear from more than 10% plots inhabited in the previous period, the status of the component "population" would not be evaluated as favourable.

The evaluation of the population structure of *B. balcanicus* could be assessed only on the data about length structure. Only the data showing clear (negative) deviations in natural reproduction in some years should be taken into account in the conservation status assessment process. Analyses on reproduction, mortality and age structure are not recommended.

Habitat for the species

The evaluation of the habitat is completely taking over the assessment from the last report according to the Water Framework Directive.

The habitat quality is assessed as <u>favourable</u> if the ecological status of the length of inhabited watercourses is:

- from > 80% in high or good status
- from > 60% in high or good status AND from < 10% in poor or bad status

The habitat quality is assessed as <u>bad</u> if the ecological status of the length of inhabited watercourses is:

- from < 30% in high or good status
- from > 25% in poor or bad status

All other combinations of the ecological status assessment are taken as describing the inadequate status of the habitat for the species.

The numerical limits mentioned above should be calibrated during the first conservation assessment process in 2019 depending on the ecological status assessment for Croatian waters.

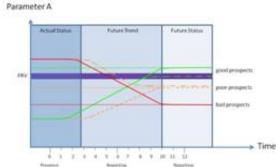
The habitat quality recorded in the field during mapping or monitoring make the evaluation of representativeness of chosen sites and plots possible.

Future prospects

This conservation status component should be evaluated by expert judgement according to methodology proposed by ETC/BD. The future trends and status will be estimated for range, population and habitat for the species following these principles:







Actual status of parameter	Future trend	Future status	Prospects (numbers refer to notes below)		
At/above FRV	+ (increasing)	> (above FRV)	/) Good		
At/above FRV	= (stable)	=/> (on/above FRV)	Good		
At FRV	- (decreasing)	<< (under FRV)</td <td colspan="2">Poor (1) Bad (1)</td> <td>Bad (1)</td>	Poor (1) Bad (1)		Bad (1)
Above FRV	- (decreasing)	>/=/ <<br (above/on/under FRV)	Good (2)	Poor (2)	Bad (2)
Below FRV	+ (increasing)	>/=/< (above/on/under FRV)	Good (3)	Poor (3)	Bad (3)
Below FRV	= (stable)	< (under FRV)	Poor (1)		Bad (1)
Below FRV	- (decreasing)	< (under FRV)	Poor (1)		Bad (1)
Unknown	+ (increasing)/ - (decreasing)/ = (stable)/ X (unknown)	X (unknown)	unknown		
under FRV X (unknown) on/above FRV		X (unknown)	unknown		

Parameter	Future Trend	Future Status	Prospects
Range			
Population			
Habitat			
Future Prospects			

Figure 5: Assessment of the future prospects of a parameter based on its future trend and predicted future status (A), evaluation matrix (B) and assessment table (C) for future prospects (ETC/BD, 2011)

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