MONITORING PROGRAMME FOR

BITTERLING (*RHODEUS AMARUS*)

by Jan Dušek

with participation from Marko Ćaleta and Zoran Marčić

using parts of the text and list of references prepared by Doru Bănăduc

Range

The range of *Rhodeus amarus* includes Eurasia with a disjunctive distribution. In Europe, *R. amarus* inhabits waters from the East of France, from the Alps and Dinarics mountains to the Ural and Caucasus mountains, including the Balkans area.

Distribution in Croatia

R. amarus is considered to be well distributed, common and present in high abundances in the suitable habitats in the continental biogeographical region (Drava, Sava, Ilova, Kupa, Sutla and Una and their alluvium and tributaries watersheds) (Figures 1 and 2). It often disappears or is not found in samples in some areas and reappears in nearby areas. In other biogeographic regions, it is an introduced invasive species. Consequently, it was considered that a monitoring program was needed for the Continental Biogeographical Region only.

Nationally, there has been no permanent or long-term specific monitoring on distribution or population status of *R. amarus*. In spite of the fact that no exhaustive data about this fish species distribution in Croatian national territory is available, (a relatively common situation in some other European countries also) the present known data (Figure 2) represent reliable data for the proposal of a short medium-term monitoring elements proposal for Croatia. Nevertheless, the knowledge regarding *R. Amarus* in Croatia is considered to be sufficient when taking the status of the species into consideration.
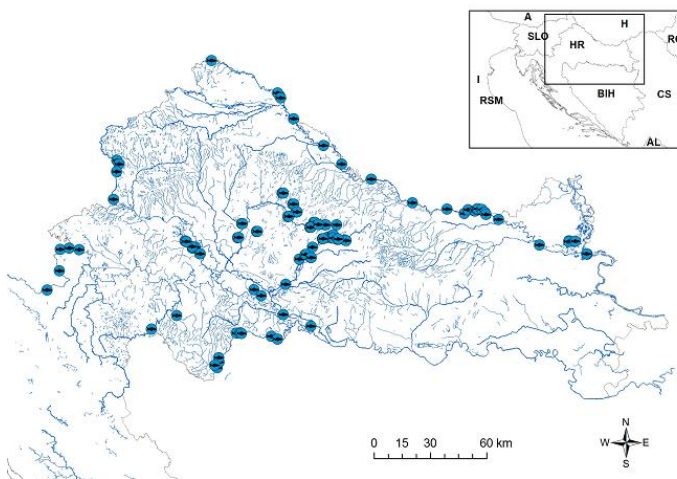

Figure 1: *R. amarus* presence identified in the last years on Continental part of the Croatian territory (prepared by SINP).
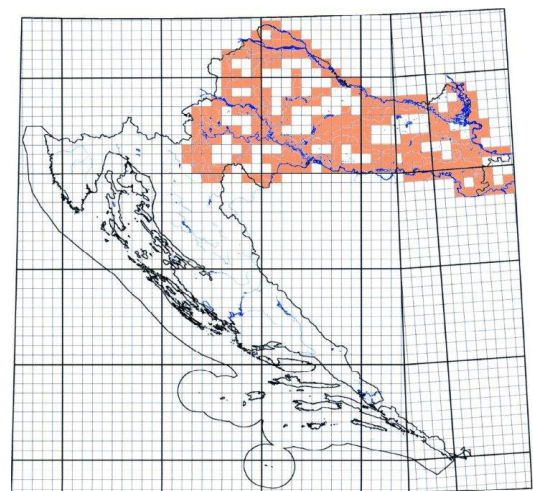
Figure 2: Distribution of *R. amarus* (Mrakovčić *et al*., 2010).

Habitat

R. amarus is a benthopelagic fish species, living in temperate areas, in fresh standing or slow flowing waters, with aquatic vegetation and a sand-silt bottom (canals, slow-flowing rivers, backwaters, oxbows). These habitats belong to group A of the National Habitat Classification, habitats with the following codes: A.1.1., A.2.3.2., A.2.4, A.3., A.4.1.3. They also belong to two habitat types from the Habitats Directive with the following codes: 3140 and 3150.

Phenology and population biology

R. amarus fed mainly on unicellular filamentous algae and vegetation debris. The maximum reported age is 5 years and sexual maturity is reached after one year. The length range at first maturity is between 3-6 cm. Reproduction occurs between the end of April and August. Reproduction is triggered by an increase in water temperature, with a threshold for spawning of between 10 and 15°C. *R. amarus* is a batch-spawning species, with each female producing several batches of eggs over a long reproductive season. Several ovipositions are completed within one day, divided from other such spawning bouts by a period of several days with no spawning activity. The roes are laid in the gill cavities of the molluscs' *Unio* and *Anodonta*. During the reproductive period, the males defend territories around the freshwater unionid mussels in which females lay one to six eggs during each oviposition event. Absolute female fecundity depends on female body size and typically reaches 80-300 eggs per season. The eggs hatch within 24-36 h, followed by a 3-4 week larval stage which also occurs in the mussel cavity. Larvae leave the mussel cavity when they reach a length of around 10 mm and enter shallow, vegetated regions that serve as nursery areas until late autumn. It is a species well understood in respect of its biology and partially of its ecology in the studied Croatian areas of interest.

Pressures and threats

R. amarus is threatened directly by pollution and also indirectly by pollution effects on freshwater mussels. The aquatic and semi aquatic (riverine habitat) habitat degradation and river remodelling, regulation and flooding control, indirect contamination, can also negatively influence *R. amarus*, both directly or/and indirectly. At present, there are significant fluctuations in numbers of *R. amarus* in its locations and subpopulations.

Conservation measures

Specific recommended conservation measures for Natura 2000 sites for *R. amarus* are as follows: preserve stable populations of freshwater mussels, prevent degradation of the structure of substrata (mainly prohibit the extraction of sand from the riverbed) and water pollution and raise the water quality.

Conservation measures should be done in general where the local situation requires action for preserving and improving the favourable ecological balance of the natural waters inhabited by this species, creating water and sediments flow conditions as close to the natural regime as possible, construction of waste water treatment plants, avoiding lotic fragmentations due to different constructions in the river bed, etc.

Annexes of the Habitats Directive

R. amarus is listed in: Annex II of the European Union Directive on the conservation of natural habitats and of wild fauna and flora (92/43/EEC), and also in the Annex III of the Bern Convention.

Red List

R. amarus is not listed in the Red List.

MONITORING PROGRAMME FOR THE
CONTINENTAL BIOGEOGRAPHICAL REGION

The most important information for the evaluation of the conservation status of *R. amarus* regards the distribution of the species. However, a comprehensive mapping programme would not be an efficient spending of research effort. Due to the good current status of this species in Croatia (well spread, common, abundant and non-endangered), research is to be focused on the localities (alluvial areas) using systematic data collection accompanied by unsystematic data gathering.

The monitoring programme is also based on sharing the data on habitat quality with the system on evaluation of the ecological status according to the Water Framework Directive.

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

The mapping of *R. amarus* is **not recommended** in the Croatian territory for the period 2013-2018. This is due to the fact that it is common and without significant changes in its distribution until now and no significant changes were registered in relation with characteristic habitats. Only in the case of major changes in distribution (negative trend at level of > 10% of the range in following periods) is it recommended to plan detailed mapping. 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 *R. amarus.*

Monitoring on plots

The monitoring on plots representatively spread in Continental biogeographical region is also **not recommended** for the period 2013-2018. Only in the case of major population size loses (loses of occurrence >25% of sites surveyed during the research on localities between two following periods OR negative trend at level of > 10% of the range in following periods) it is proposed to choose ca. 30 plots for monitoring. The exact number depends on variability of parameters for classification. Objectives of the monitoring should be oriented to representative plots chosen according to the following principle of classification. The plots would be representatives for type of the habitat (rivers, canals, oxbow lakes, ponds etc.) in river basins of Sava, Kupa and Drava (+ Danube).

Research on localities

Objectives

This level of surveillance will ensure sufficient data about the status of the species in representative areas and enable SINP to evaluate potential deviations which can signify negative trends in population as well as in distribution of *R. amarus*. If these negative trends are established, the field mapping and the monitoring on plots have to start in the next period.

This surveillance is not too time-consuming and is adequate with regard to actual status of the species in Croatia. It will also give useful information for planning and setting of conservation measures in alluvial areas.

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 exceeding 30°C (when there is increased risk of mortality due to low

oxygen content in the water) or not less than 15°C (as *R. amarus* is not active and is oriented to deep water where is difficult to sample). The electrofishing cannot be realized during extremely high flow rates, at excessively increased turbidity of water and 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 3 m wide sections along the shoreline is monitored, the electrofishing from boat instead of wading is needed if the depth is continuously higher than 1 m near the bank. The section may not be bounded by stop nets or other temporary migration barrier. *R. amarus* is among the species that can easily escape detection during the survey, it is therefore appropriate to place the landing nets about 0.5 meters behind the anode, and using the recommended landing net mesh size of 4 mm.

The electrofishing (including only work with electrofishing device in the water) per site is carried out for 60 minutes (45 minutes if two anodes are used) or until the catch of 250 *R. amarus* individuals. The speed of research should be oriented to catch the highest number of *R. amarus* individuals as possible. The exact location of transect will be recorded into the map in order for comparisons in subsequent periods to be drawn.

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

For each proposed site, the number of individuals and population structure (proportion in length classes) will be determined.

<u>Selection of localities</u>

The selection of monitored areas will be based on the expert knowledge of the Croatian ichthyologists. Classification is not possible or meaningful for *R. amarus,* as the information about parameters (width and depth) is not available for localities with the presence of the species. Six areas would be chosen in river basin of:

- Danube+ lower part of Drava (proposed is the area in Kopački Rit)
- middle part of Drava
- middle part of Sava (proposed is the area in Lonjsko or Mokro polje)
- Ilova
- Odra
- Kupa (proposed is the area in Pokupski bazen)

An alluvial area is a locality with sites represented by main river and canals, oxbow lakes, ponds etc. (if present in locality). Five sites have to be chosen in each locality (altogether 30 sites). At least one of the selected sites would have been verifiably occupied by *R. amarus* in the last 6 years. The selection of sites should take into account the opportunity of surveillance and evaluation of possibilities of connection (migrations) between different parts of area. The selected sites should also have aquatic vegetation and sand-silt bottom, a habitat characteristic needed for *R. amarus*.

Each monitored site would be monitored once every six years, the sites without occurrence of *R.amarus* will not be monitored in the subsequent period, however alternative sites will be chosen.

The crew should complete the field survey in two days for each locality. The field work effort can be distributed in this relatively long period of time, therefore only two days of the field work will be conducted for the monitoring of *R. amarus* per year.

Figure 3: Model example of selection of sites in an alluvial area - area of the confluence of Drava and its tributary near to Petrijevci

<u>Data forms</u>

The data form for research in localities (and mapping) 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 *R. amarus* 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 *R. amarus*, 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.

Unsystematic data gathering

<u>Objectives</u>

There are available sources of information on the actual occurrences of *R. amarus* 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 or expert NGOs)

<u>Field work instructions</u>

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.

<u>Data forms</u>

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

Distribution of the species is continuous in all main streams where *R. amarus* occurs. For this reason, 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 no actual research with negative findings (taking into account mainly results of unsystematic data gathering) – all quadrants 10x10 km between findings on the same river should be connected if gaps are not larger than 30 km (length of the river between localities, not by air). Also, all following parts of rivers are taken as part of the range – 10 km upstream from the upper locality and 20 km downstream from lower locality. Distribution maps are prepared using the method with half distances – gaps to 15 km, 5 km upstream and 10 km downstream.
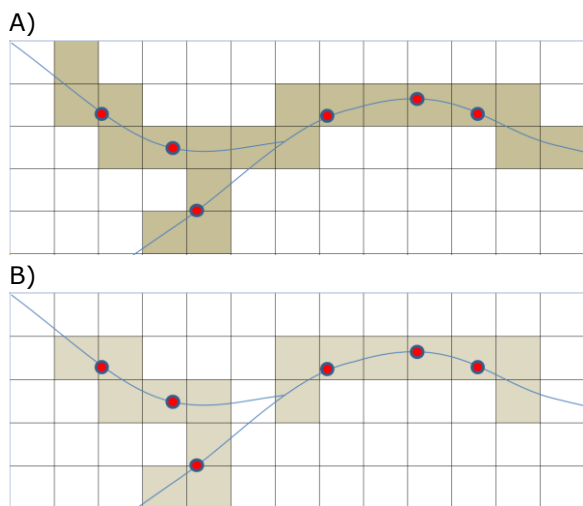
A)

B)

Figure 4: Model example for range (A) and distribution (B) map using quadrants 10x10 km (findings by red points).

The reference range is proposed as the same as the value in the first report for the period 2013-2018. Expansion or increasing of the range could be taken into account only in places where findings show *R. amarus* 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. It can only be validated by systematic mapping.

Population

The most useful population unit is number of quadrants 10x10 km occupied by the species. The possibility of estimation of the population size using the number of individuals is unfortunately inconceivable.

The calculation of this number represents a simple GIS analysis described above for the preparation of the distribution map.

Analyses on reproduction, mortality and age structure is not recommended for this short-lived species (max. 5 years). The description of the population structure from monitored sites during research on localities gives only the information about local status and cannot be analyzed at biogeographical level. Monitoring on plots will start if trends in population structure would be important, even it is difficult to determine from the data any clear (negative) deviations in natural reproduction in some years.

The favourable reference population is proposed as the same as the value in the first report for the period 2013-2018. The trend is interpreted in % of increase/decrease of the inhabited quadrants 10x10 km. If the species disappear from more than 10% of sites inhabited in the previous period, the status of the component "population" would not be evaluated as favourable. If *R. amarus* disappears from more than 25% sites inhabited in the previous period, the status of the component "population" must be evaluated as bad.

A)   B)   C)

5 10 15 20 25 30 35 40 45 50 55 60 65 70 75 80   5 10 15 20 25 30 35 40 45 50 55 60 65 70 75 80   5 10 15 20 25 30 35 40 45 50 55 60 65 70 75 80
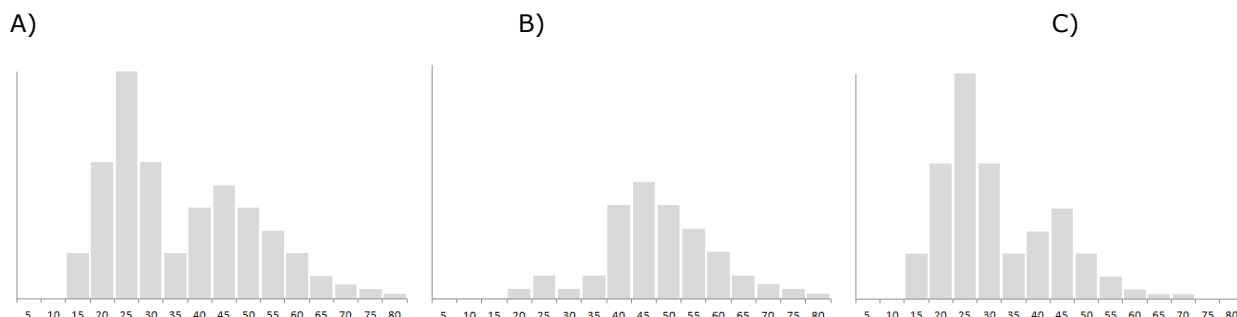
Figure 5: Standard length (SL): A) model of "ideal" recorded population structure; B) frequent situation without detecting youngest category (0+); C) situation deviating from normal with lack of adults

Habitat for the species

The surface depends directly on chosen population unit (length of inhabited rivers and streams). For this reason the component "habitat" is assessed based on quality.

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

The habitat quality is assessed as favourable if the ecological status of the surface of inhabited areas is:
- from > 70% in high or good status
- from > 50% 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 surface of inhabited areas is:
- from < 25% in high or good status
- from > 30% 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.

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:

A)
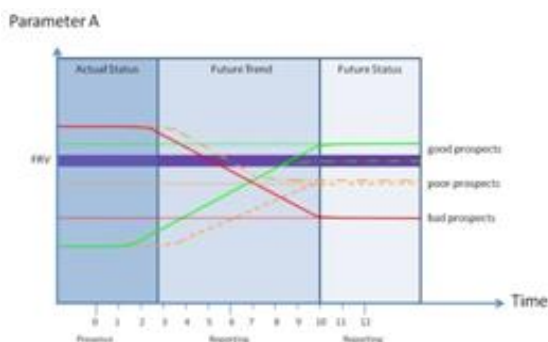
B)

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)	Poor (1)	Bad (1)	
Above FRV	- (decreasing)	>/=/</<< (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 on/above FRV	X (unknown)	X (unknown)	unknown		

C)

Parameter	Future Trend	Future Status	Prospects
Range			
Population			
Habitat			
Future Prospects			

Figure 6: 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)

References

Baptist M. J. et al., 2006 – Flood detention, nature development and water quality along the lowland river Sava, Croatia, Springer.

Bănăduc D. and Bănăduc A., 2002 – A Biotic Integrity Index adaptation for Carpathian River assessment, Acta oecologica IX, 1-2, 77-95.

Bănărescu P. M. and Bănăduc D.: Habitats Directive (92/43EEC) fish species (Osteichthyes) on the Romanian territory; Acta Ichtiologica Romanica II, 2007.

Bonacci O., Oskorus D., The influence of three Croatian hydroelectric power plants operation on the river Drava hydrological and sediment regime.

Bošnir J. et al., 2007 – Organochlorine pesticides in freshwater fish from the Zagreb area. Arhivza Higijenu Radai Toksikologiju vol. 58, no. 2, 187-193, ISSN 0004-1254.

Ćaleta, M., Mustafić, P., Mrakovčić, Milorad. & Marčić, Z. (2009): Studija inventarizacije ihtiofaune donjeg toka rijeke Une. PMF Zagreb.

Dragun Z. et al., 2011 – Water quality of medium size watercourse under baseflow conditions: the case study of river Sutla in Croatia, AMBIO 40: 391-407.

Dumbović, Vlatka, Posavec Vukelić, V., Duplić, A., Katušić, L., Jelić, D., Boršić, I., Partl, A. (2009): Studija inventarizacije flore i faune rijeke Une i priobalnog pojasa . Sisak : Sisačko-moslavačka županija.

Dušek, J., 2007: Metodika terénního sběru dat o populacích vranky obecné (*Cottus gobio*) v rámci sledování stavu z hlediska ochrany. Agentura ochrany přírody a krajiny ČR, Praha, 11 pp. + Annexes.

Evans, D., Arvela, M., 2011: Assessment and reporting under Article 17 of the Habitats Directive.

Explanatory Notes & Guidelines for the period 2007-2012. ETC/BD, Paris, 123 pp.

Fausch K. D. et al., 1984 – Regional application on an index of biotic integritz based on stream fish assemblages, Trans. Of the Am. Fish. Soc. 113, 39.55.

Fausch K. D. and Schrader L. H., 1987 – Use of index of biotic integrity to avaluate the effects of habitat, flow, and water quality on fish assemblages in three Colorado Front Range streams – Colorado Division and the Cities of Forth Collins, Loveland, Greelez, Longmont and Windsor, department of Fisheries and Wildlife Biology, Colorado State Universitz, Fort Colins, Colorado.

Frančišković-Bilinski S., Bilinski H. and Širac S., – Organic pollutants in stream sediments of Kupa River drainage basin, Fresenius Environmental Bulletin, vol. 14, no. 4, 2005, 282-290.

Gvozdic V., Brana J., Puntaric D., Vidosavljevicl D. Roland D., 2011 – Changes in the lower Drava River water quality parameters over 24 years, Arh Hig Rada Toksikol, 62: 325-333.

Karr J. R. 1981 – Assessment of biotic integrity using fish assemblages, Fisheries, vol. 6, 21-27.

Karr J. R. and Dudley D. R., 1981 – Ecological perspective on water quality goals, Environ. Manage., 5, 55-68.

Karr et al., 1986 – Assessing Biological Integritz in Running Waters A Method and Its Rationale, Illinois Natural Historz Survey, Special Publication 5 September 1986, 1-20.

Kottelat, M. and J. Freyhof, 2007. Handbook of European freshwater fishes. Publications Kottelat, Cornol, Switzerland. 646 p.

Konečná, Reichard, M, 2011: Seasonal dynamics in population characteristics of European bitterling *Rhodeus amarus* in a small lowland river. Journal of fish biology, 78 (1): 227-239.

Habeković D., Mrakovčić M. I Bogdan M., 1986: Ichtiofauna dijela rejeke Drave nakon izgradnje sustava HE Čakovec. Ribarstvo Jugoslavje, 4: 57-61.

Milorad Mrakovčić, Andreja Brigić, Ivana Buj, Marko Ćaleta, Perica Mustafić, Davor Zanella: Red Book of Freshwater Fish of Croatia/Cravena Knjiga Slatkovodnih Riba Hrvatske, 2006.

Mrakovčić M., Kerovec M., Mišetić S., Schneider D., Tomaskovic N. i Šurmanović D., 1996: Ichthyofauna of the Drava River (Croatia). Internationale Arbeitsgemeinschaft Donauforshung, 1: 345-348.

Mrakovčić, M., Mustafić, P., Ćaleta, M., Zanella, D., Buj, I. & Marčić, Z. (2008): Ihtiološka raznolikost rijeke Mure. PMF Zagreb.

Mrakovčić, M., Ćaleta, M., Mustafić, P., Marčić, Z. & Zanella, D. (2010): Značajke ihtiofaune rijeke Sutle. PMF Zagreb.

Mrakovčić, M., Ćaleta, M., Mustafić, P., Marčić, Z., Zanella, D. & Buj, I. (2010): Rhodeus amarus iz Slatkovodne ribe. Izvješće za potrebe izrade prijedloga potencijalnih NATURA 2000 područja. PMF Zagreb.

Muus, B.J. and P. Dahlström, 1968. Süßwasserfische. BLV Verlagsgesellschaft, München. 224 p.

Picer M., Perkov S. and Picer S., 1995 – Contamination of Bela Krajna, Slovenia with polychlorinated byphenyls. 1. Levels of some high molecular chlorinated hydrocarbons in the water and fish of the Kupa River in Croatia, Water, Air and Soil Pollution, 82(3-4), 559-581.

Popović I., 2008 – Implementation of the Water Framework Directive and Urban Waste Water Treatment Directive in the Republic of Croatia – Investments, operation-maintenance, adaptation.

Povz M. and Sket B, 1990 – Naše slatkovodne ribe. Založba Mladinska knjiga.

Skoberne, P. (ed.), 2009: Recommended conservation measures for Natura 2000 in Croatia. Report of the project EuropeAid/123526/D/SER/HR Institutional building and implementation of NATURA 2000 in Croatia, 125 pp.

Teskeredžić E. Z., Teskeredžić M., Tomec B., Kurtović B., Raspor D., Kapetanović D., Dragun I., Vardić

D., Valić Z., Strizzak B., Španović Z., Šoštarić V., Roman Z., 2009 – Programme for the monitoring of the freshwater fishery status in the year 2009 – Group D – Fishing area Sava, River Sutla.

Vostradovsky, J., 1973. Freshwater fishes. The Hamlyn Publishing Group Limited, London. 252 pp.