ECOLOGIA BALKANICA

2018, Vol. 10, Issue 2 December 2018 pp. 63-72

Intercalibration of Macroinvertebrate-Based Method for Status Assessment of Bulgarian Tributaries of the Danube River

Rabia Soufi[1], YankaVidinova[1],Violeta Tyufekchieva[1], Vesela Evtimova[1*], Desislava Stoianova[1], Maria Kerakova[1], Galia Georgieva[1], Stefan Stoichev[1], Ivailo Dedov[1], Georg Wolfram[2]

1 - Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, 2 Yurii Gagarin Str., 1113-Sofia, BULGARIA
2- DWS Hydro-Ökologie GmbH, Zentagasse 47, A-1050 Wien, AUSTRIA
[*]Corresponding author: vesela.evtimova@gmail.com

Abstract. Bulgaria has joined the official intercalibration (IC) in order to complete the state commitments for the classification of rivers based on Biological Quality Elements (BQE). The objective of this paper was to verify whether the present Bulgarian classification method for ecological status of large lowland (national type R7) and medium-sized lowland (national type R8) rivers was compliant with the normative definitions of the Water Framework Directive. Thus, a two-year study (2014 - 2015) on macroinvertebrate communities was conducted at 43 sites (18 from R7 and 25 from R8). Differences in qualitative and quantitative composition in the above-mentioned river types, as well as indicative potential of BQE in relation to different anthropogenic pressures were analysed. Based on alternative benchmark sites (identified using the criteria defined by the EU), the High/Good boundary and a reference value for the Biotic index were defined. Class boundaries were in line with the results of the completed intercalibration exercise. We found a significant association between the biotic index and all three groups of pressures (land use, chemistry and hydro-morphology) and these results could be used for assessment of the ecological status. The national assessment method (based on our data) was in good agreement with the accepted methods from other member states who share the same river types within the Eastern Continental Geographical Intercalibration Group (EC-GIG).

Key words: benthic macroinvertebrates, river types, large and medium-sized lowland rivers, ecological quality assessment, pressure, Bulgaria, Geographical Intercalibration Groups.

Introduction

The European Water Framework Directive (WFD) 2000/60/EC (EC, 2000) requires national classifications of ecological status to be harmonised through an intercalibration. In this intercalibration exercise, significant differences in status classification among member states are harmonised by comparing and, if necessary, adjusting the good status boundaries of the national assessment methods. The intercalibration is performed for rivers, lakes, coastal and transitional waters, focusing on selected types of water bodies (intercalibration types), anthropogenic pressures and Biological Quality Elements (BQE). Among these BQE are also benthic macroinvertebrates (Directive 2000/60/EC –

 Union of Scientists in Bulgaria – Plovdiv
University of Plovdiv Publishing House

EC, 2000). The development of ecological assessment and classification systems is considered as one of the most important and technically challenging parts of the implementation of the WFD.

The official intercalibration of invertebrate-based methods of ecological status assessment in the Eastern Continental (EC) rivers has been finalised within the EC-GIG (Geographical Intercalibration Group) intercalibration in 2011 (Opatrilova, 2011). The EC-GIG includes nine types, four of which are relevant for Bulgaria. Bulgaria has already joined the IC round but only the methods for IC types R-E1a (= national type R2) and R-E4 (R-E1b) (= R4) have been successfully intercalibrated at the first stage (European Union, 2013). The Bulgarian method for IC types R-E2 (= R8) and R-E3 (= R7) has to be approved and recognised as compatible with the methods of other EC-GIG countries and are treated in this publication (Table 1).

Various pressures have been addressed by the different methods in the finalised IC exercise. Most countries indicated as detected pressures general and hydro-morphological degradation and pollution by organic matter. The Bulgarian (BG) method addresses mainly catchment land use, pollution by organic matter and eutrophication, as well as habitat destruction. The BG method is, therefore, comparable to the methods which have already been successfully intercalibrated.

The objective of this paper is to present the results of the completed intercalibration exercise for the Bulgarian classification method of ecological status assessment of rivers belonging to the IC types R-E2 (= R8 national type) and R-E3 (= R7 national type) based on benthic invertebrates. This is in line and compliant with the WFD normative definitions and its class boundaries. Validation of the final results allowed the successful IC of the method for analysing these river types of surface water.

Material and Methods

Site selection

All visited sites belonged to Ecoregion 12 and are Bulgarian tributaries of the Danube River (Fig. 1). The number of sampled sites sharing the common types R-E2 (= R8) and R-E3 (= R7) were 25 and 18, respectively (Table 2).

Table 1. Main IC types in the EC GIG (2015) and corresponding national types.

Type	Common intercalibration type	Ecoregion	Catchment area [km²]	Altitude [m]	National type	Status IC for Bulgaria
R-E1a	Carpathians: small to medium, mid-altitude	10	10–1,000	500–800	R2	Finalised 1st stage
R-E4 (R-E1b)	Carpathians: small to medium, mid altitude	11, 12 (10)	10–1,000	200–500	R4	
R-E2	Plains: medium-sized, lowland	11,12	100–1,000	<200	R8	Finalised 2nd stage
R-E3	Plains: large, lowland	11,12	>1,000	<200	R7	
R-EX4	Large, mid-altitude	10, 11, 12	>1,000	200–500	–	Not applicable
R-EX5	Plain: small lowland	11, 12	10–100	<200	–	
R-EX6	Plain: small, mid-altitude	11, 12	10–100	200–500	–	
R-EX8	Balkan: small to medium-sized, calcareous, karst spring	5	10–1,000		–	

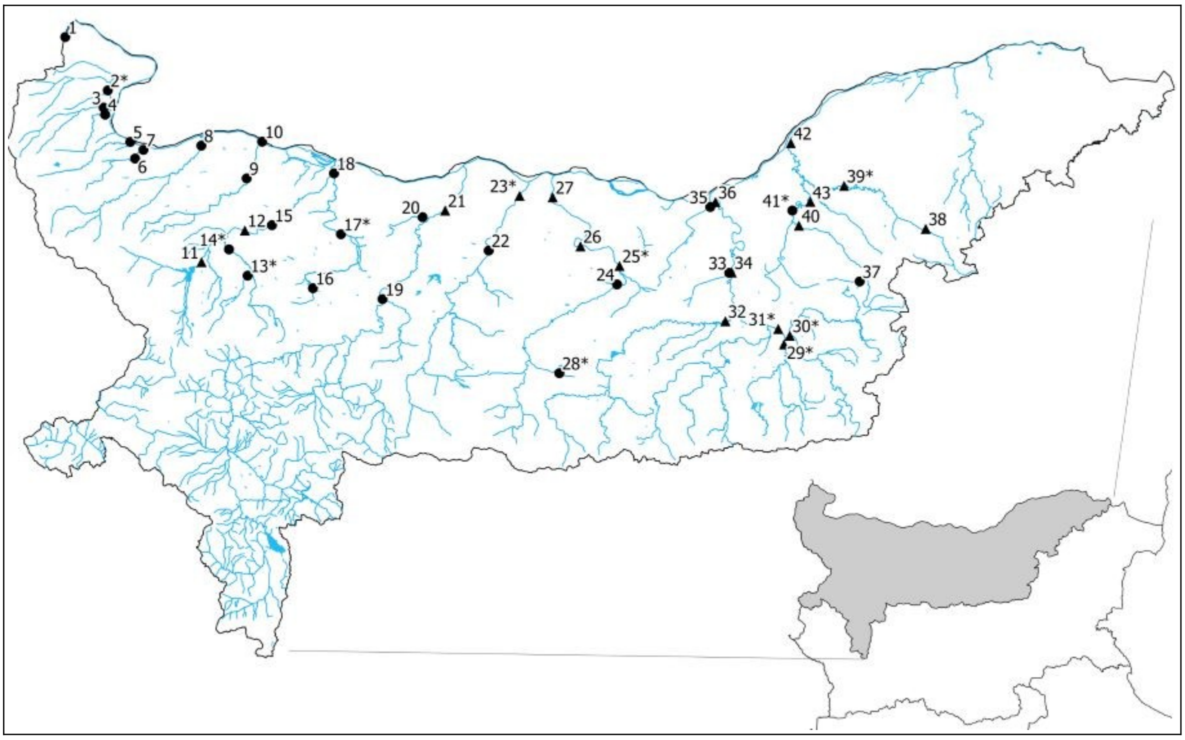

Fig. 1. Scheme of the sampled localities. Legend: R7- triangles, R8- circles.
With * are marked the alternative benchmark sites.

Table 2. List of the sampled sites, including their location and the years they were sampled. With * are marked the alternative benchmark sites.

N	River/Site	River Code	Latitude	Longitude	River Type	2014	2015
1	Timok, Bregovo	Tim_Bre	44.15879	22.62339	R8_RE2	x	x
2*	Topolovets, outflow	Top_out	43.98285	22.81737	R8_RE2	x	x
3	Voynishka, Tarnyane	Voy_Tar	43.92621	22.79877	R8_RE2	x	x
4	Vidbol, Dunavtsi	Vid_Dun	43.90302	22.80533	R8_RE2	x	x
5	Archar, outflow	Arc_out	43.81282	22.92031	R8_RE2	x	x
6	Skomlya, Septemvriitsi	Sko_Sep	43.75767	22.9422	R8_RE2	x	x
7	Skomlya, Dobri Dol	Sko_DD	43.78646	22.98025	R8_RE2	x	x
8	Lom, outflow	Lom_out	43.80043	23.24489	R8_RE2	x	x
9	Tsibrica, Vulchedrum	Tsi_Val	43.69198	23.45198	R8_RE2	x	x
10	Tsibrica, Dolni Tsibar	Tsi_DTs	43.81373	23.52269	R8_RE2	x	x
11	Ogosta, Montana	Ogo_Mon	43.41376	23.24561	R7_RE3		x
12	Ogosta, Kobilyak	Ogo_Kob	43.51896	23.44371	R7_RE3		x
13*	Botunya, Babino	Bot_Bab	43.36862	23.45628	R8_RE2	x	x
14*	Botunya, Ohrid	Bot_Ohr	43.45611	23.37115	R8_RE2	x	x
15	Ribine, outflow	Rib_out	43.53667	23.56741	R8_RE2	x	x
16	Skut, Peshtene	Sku_Pes	43.32685	23.75459	R8_RE2	x	x
17*	Skut, Turnava	Sku_Tur	43.5066	23.88259	R8_RE2	x	x
18	Skut, Miziya	Sku_Miz	43.70873	23.85094	R8_RE2	x	x
19	Zlatna Panega, outflow	ZP_out	43.29042	24.07267	R8_RE2	x	x
20	Gostilya, outflow	Gos_out	43.56333	24.25523	R8_RE2	x	x
21	Iskar, Orechovitsa	Isk_Ore	43.58523	24.35858	R7_RE3		x
22	Tuchenitsa, outflow	Tuc_out	43.45216	24.5576	R8_RE2	x	x
23*	Vit, Gulyantsi	Vit_Gul	43.6341	24.69868	R7_RE3		x

N	River/Site	River Code	Latitude	Longitude	River Type	2014	2015
24	Shavarna, outflow	Sha_out	43.33947	25.14396	R8_RE2	x	
25*	Osam, Levski	Osa_Lev	43.40111	25.1551	R7_RE3		x
26	Osam, Izgrev	Osa_Izg	43.46626	24.97677	R7_RE3		x
27	Osam, Cherkovitsa	Osa_Che	43.62909	24.84998	R7_RE3		x
28*	Krapets, outflow	Kra_out	43.04267	24.8802	R8_RE2	x	x
29*	Dzhulyunishka, Dzhulyunitsa	Dzh_Dzh	43.139	25.90652	R7_RE3		x
30*	Stara, Kesarevo	Sta_Kes	43.16817	25.9328	R7_RE3		x
31*	Stara, Bryagovitsa	Sta_Bry	43.19072	25.8803	R7_RE3		x
32	Rositsa, Polikraishte	Ros_Pol	43.21666	25.63865	R7_RE3		x
33	Eliiska, outflow	Eli_out	43.37786	25.65659	R8_RE2	x	x
34	Yantra, Karantsi	Yan_Kar	43.37904	25.66774	R7_RE3		x
35	Studena, outflow	Stu_out	43.59722	25.56976	R8_RE2	x	x
36	Yantra, Novgrad	Yan_Nov	43.61338	25.5933	R7_RE3		x
37	Popovski Lom, Popovo	PL_Pop	43.34932	26.25231	R8_RE2	x	x
38	Beli Lom, Razgrad	BL_Raz	43.52449	26.55396	R7_RE3		x
39*	Beli Lom, Pisanets	BL_Pis	43.66717	26.18122	R7_RE3		x
40	Cherni Lom, Ostritsa	ChL_Ost	43.53482	25.97467	R7_RE3		x
41*	Cherni Lom, Shirokovo	ChL_Shi	43.58523	25.94469	R8_RE2	x	x
42	Rusenski Lom, outflow	RL_out	43.80841	25.9377	R7_RE3		x
43	Cherni Lom, Cherven	ChL_Che	43.61258	26.02452	R7_RE3	x	

Sample collection and processing

Benthic macroinvertebrates were collected by wading in the river or along the riparian zone in deep rivers (ca. 100 m). The procedure followed a multi-habitat sampling strategy, where several subsamples from representative habitats were combined to one mixed sample, depending of their relative proportion in the river (Cheshmedjiev & Varadinova, 2013). The sampling followed EN 16150:2012 (Pro-rata multi-habitat sampling) using a hand net (mesh size 500 µm) with 10 units (~ 0.9 m^2) and was done once a year (2014-2015) in late summer (August–October). Benthic samples were preserved in 70 % alcohol. All macroinvertebrates were sorted and determined using a stereo microscope. For large samples, subsamples were taken. All taxa were identified to family level, where possible to species/genus level.

Additionally, hydro-morphological alterations, such as impoundment, hydropeaking, abstraction, dams within river segment, dams at the site, channelisation, riparian vegetation within the river segment, riparian vegetation at the site, habitat alteration within the river segment, habitat alteration at the site and dykes were assessed in situ using a scale from 0 (no) to 3 (high) for all variables except water abstraction, where the maximum value was 2 (moderate).

Further, physical and chemical parameters of the water were studied. Electric conductivity (EC) was measured *in situ* using portable Windaus Labortechnik Package. Water samples were collected for measuring biological oxygen demand (BOD_5), orthophosphate-P (SRP), nitrate-N (NO_3-N) and ammonium-N (NH_4-N). Chemical parameters were analysed in an accredited laboratory using the following standards: pH - ISO 10523; oxygen concentration (mg/l) and saturation (%) - EN 25814; electric conductivity (µS/cm) - EN 27888; BOD_5 (mg/l) - EN 1899-2; SRP (mg/l) - ISO6878; NO_3-N (mg/l) - ISO 7890-1; NH_4-N (mg/l) - ISO 7150-1.

Data analyses

Due to biogeographical and typological reasons, as well as differences in data acquisition, biological data of different countries or different water types cannot be compared without concern. For this reason, WFD (Directive 2000/60/EC - EC, 2000) requires the use of reference conditions within each GIG as a benchmark to standardise biological assessment metrics and assessment results have to be expressed as Ecological Quality Ratios (EQR). In this study, the setting of reference values was done based on alternative benchmark sites, since true reference sites were lacking. The criteria for alternative benchmark sites were according to the EC-GIG report (Opratilova, 2011).

Metric calculation

The Irish Biotic index (BI) was used for both river types according McGarrigle *et al.* (1992, McGarrigle & Lucey, 2009). The metric calculation is described in detail in Ordinance No 412 (2011) (Appendix II.2.). The BI was calculated from the relative proportions of the tolerance groups of macroinvertebrates. Depending on additional criteria, the calculated values might be downgraded (Yaneva & Cheshmedjiev, 1999). EQR is the ratio between the observed index value and the index value typical for reference sites (for R7/R8 rivers the maximum BI value is 5; Cheshmedjiev & Varadinova, 2013): EQR_{BI} = measured value / reference value.

The biological monitoring working party (BMWP) is a procedure for measuring water quality and is calculated as the sum of the indicator values of the presented families. We also calculated the average score per taxon (ASPT) by dividing BMWP by the number of scoring taxa in the sample (Armitage *et al.*, 1983; Hawkes, 1998).

Pressure-impact relationships

The impact of three types of pressures on macrozoobenthos was assessed. For two of the pressure types we used data obtained *in situ* from 2014-2015:

1. Hydro-chemistry (including EC, BOD_5, SRP, NO_3-N, NH_4-N);

2. Hydro-morphology (for details see Sampling collection).

The impact of the third type of pressure, land use, was assessed based on data collected for R-E2 and R-E3 sites between 2009 and 2015. The impact of land use was analysed using CORINE Land Cover (CLC), as well as the land-use index (LUI), which was derived from CLC and defined as: $LUI=4.CLC_{urban} + 2.CLC_{agr.\ intens.} + CLC_{agr.\ extens.}$.

The relationships between these pressures and BI were explored by fitting linear models in R software, version 3.4.2 (R Core Team, 2017).

Results and Discussion

Invertebrates

We recorded 259 aquatic macroinvertebrate taxa belonging to 23 systematic groups: 130 taxa were identified to species level, 72 - to generic, 52 – to family, two – to order, two – to class and one to phylum level. The highest taxon richness was found in the following groups: Gastropoda (41), Ephemeroptera (39), Oligochaeta (26) and Chironomidae (25). Approximately half of the groups were presented with less than five taxa. Macroinvertebrate communities were more diverse at the sites of R8 river type (238 taxa), as compared to R7 sites (125 taxa). Furthermore, 134 taxa were found only in small and medium-sized rivers, while in large rivers the typical inhabitants were just 21 taxa. This was most probably due to the more diverse microhabitats at R8 sites, on the one hand, and due to the fact that R7 sites were sampled only once (in 2015), on the other.

Biotic indices

The intercalibration exercise follows similar assessment concepts, *viz.* multimetric index based on structure and "species traits" metrics. Only the method of Bulgaria is based on the Irish Biotic index. This index was also used in the previous round of IC in Bulgaria, when R-E1a and R-E1b river types had been successfully intercalibrated. Hence, this method can be accepted and the IC exercise using the fit-in procedure (European Union, 2015) is considered as feasible in terms of assessment concepts. The values of the indices/metrics for the alternative benchmark sites are presented in Fig. 2.

According to the alternative benchmark criteria (Opatrilova, 2011), ASPT index should be in the range 5 - 6.4. According Armitage *et al.* (1983) and Zamora-Muñoz *et al.* (1995), ASPT ignores random factors, gives a more realistic picture of the conditions in a river and records smaller differences in water quality as compared to BMWP. Our results showed identical dynamics for both indices (Fig. 2a). Despite the lower ASPT values at three of the observed benchmark sites (CHL_Shi, Osa_Lev and Sta_Kes), according their BI-value (resp. EQR_BI), they belonged to the category of rivers with/in good ecological quality. Most probably the higher BOD_5 values there (Table 4) was responsible for the loss of sensitive taxa and consequently - lower BMWP/ASPT- values at these sites. Contrary, higher values of all indices were established at sites Bot_Ohr, Bot_Bab and Sta_Bry, where BOD_5 values were the lowest.

Reference values and class boundaries

The water quality assessed using BI (resp. EQR_IBI) of the selected benchmark sites varied from excellent to good (Fig. 2b, Table 3). Both IC types were treated together, due to the small data set (n = 16 for both types) and because the values of BI for the alternative benchmark sites did not differ significantly between the two types. The calculated value for BI was 3.84.

Neither any discontinuity in the relationship between pressure and the impact, nor a paired metric analysis (both approaches according to Guidance Document No. 14, EC (2011)) were helpful to set the boundaries for the status classes. Following Step 8 of the Boundary Setting Protocol (Guidance Document No. 14, EC (2011), page 65), the continuum of impact was divided into equal width classes, starting from the H/G boundary (reference value 4.7 х 0.8). As the lowest possible value of BI is 1, which has been found for instance in Beli Lom near Razgrad, the whole gradient of ecological status is covered. The maximum BI value found in the data set was 4.5 (e.g. Skomlya near Septemvriytsi Village).

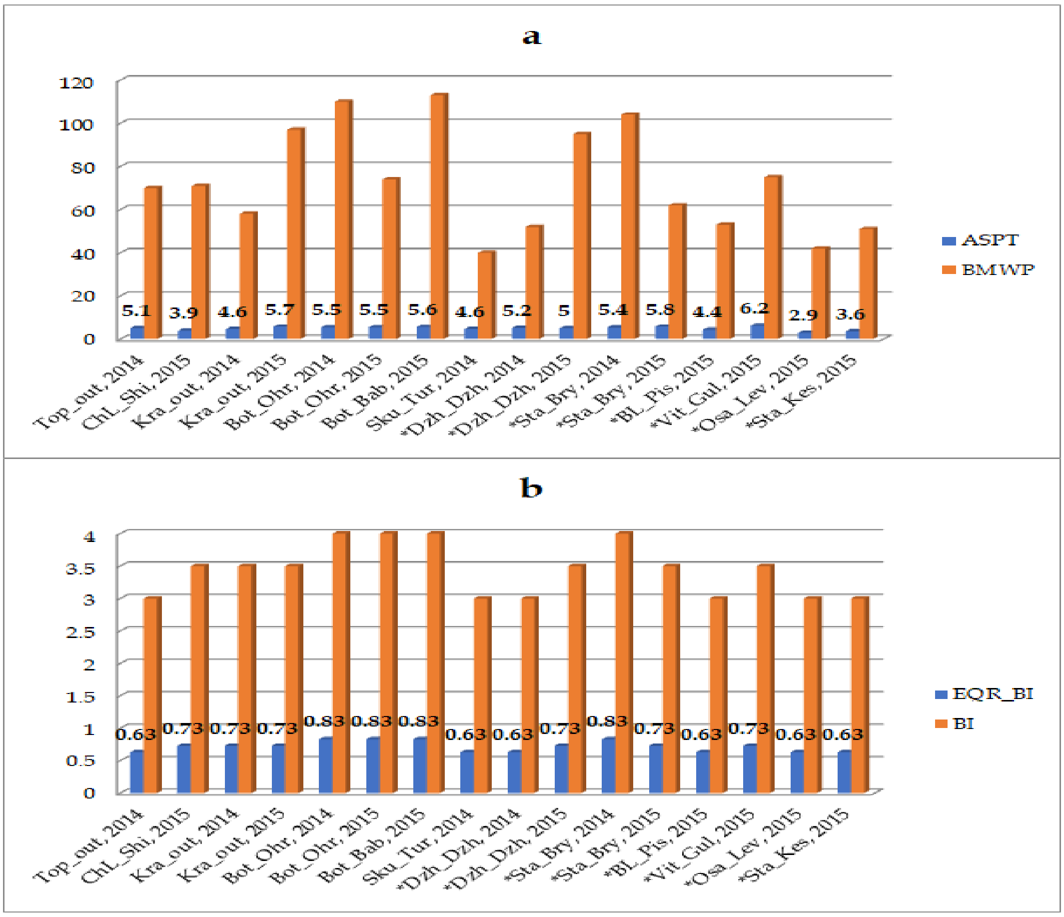

Fig. 2. Alternative benchmark sites and measured values for the ASPT, BMWP metrics **(a)** and IBI, EQR_IBI metrics **(b)** for IC river types R-E2 (= R8) and R-E3 (= R7) in Bulgaria.

The following algorithms were used to transform EQR values to nEQR values:

EQR_i	$nEQR_i$
≥ 1	1
$\geq EQR_{H/G}$	$(EQR_i - EQR_{H/G}) / (1 - EQR_{H/G}) * 0.2 + 0.8$
$EQR_{H/G} > EQR_i \geq EQR_{G/M}$	$(EQR_i - EQR_{G/M}) / (EQR_{H/G} - EQR_{G/M}) * 0.2 + 0.6$
$EQR_{G/M} > EQR_i \geq EQR_{M/P}$	$(EQR_i - EQR_{M/P}) / (EQR_{G/M} - EQR_{M/P}) * 0.2 + 0.4$
$EQR_{M/P} > EQR_i \geq EQR_{P/B}$	$(EQR_i - EQR_{P/B}) / (EQR_{M/P} - EQR_{P/B}) * 0.2 + 0.2$
$< EQR_{P/B}$	$(EQR_i - EQR_{min}) / (EQR_{P/B} - EQR_{min}) * 0.2$

By dividing the class boundaries by the reference value, the EQR for all class boundaries was calculated. The EQR was transformed to normalised EQR in order to define H/G as 0.8, G/M as 0.6, *etc*. Each class includes the lower class boundary (e.g., nEQR = 0.8 → high status).

The percentiles, which have often been used in defining the H/G boundary, cannot be used in the case of BI, because the index is ordinal and not metric (only values in 0.5-steps). As alternative, a logit regression was used to derive the H/G boundary, defined at 85%. The inverse EQR of the H/G boundary (3.84/0.8) gave the reference values for R-E2 and R-E3 (rounded to 1 digit): reference BI = 4.8. Using the fit-in procedure (European Union, 2015), the method for these rivers is considered as intercalibrated.

Pressure-impact relationship

According OPRATILOVA (2011), at least four of the seven parameters used for the screening of alternative benchmark sites have to fit within the given threshold (Table 4).

According to the latest revision of the water bodies in the Danube Region (Water basin Directorate "Danube Region", 2010), the sites Dzh_Dzh (Dzhulyunska/ Dzhulyunitsa bridge) and Sta_Kes (Stara/below Kesarovo) belong to national river type R4 rather than R7. However, since they are close to R7 in terms of their abiotic characteristics, they are included in the calculations for the IC river type R-E3 (Table 4).

For the hydro-morphological screening parameters, it was required that each site has the following parameter values equal to „no" or „low" status, while also allowing for at most three parameters corresponding to „medium" and just one to „high" status: impoundment, hydro-peaking, water abstraction, upstream dam influence, water temperature modification, channelisation, alteration of riparian vegetation, local habitat alteration, dykes, toxic risk, water acidification, navigation and recreational use.

By varying the weight of these variables, the following combinations performed best (i.e. resulted in the highest R^2 = highest proportion in the variance of BI explained by the pressure index):

Pressure Index I (for R-E2) = Abstract. + Dams_seg + Dams_Site + VegRip_seg + 4 x HabAlt_seg + 2 x HabAlt_site

Pressure Index II (for R-E3) = Channelisation + VegRip_seg + HabAlt_site + 4 x Dykes.

Pressure-response relationships for the Biotic index in R-E2 and R-E3 river types.

The impact of hydro-chemistry, hydro-morphology and land use on macrozoobenthos was assessed (Table 5).

Table 3. Reference values and class boundaries for the Biotic index (BI), as well as EQR and normalised EQR (nEQR) values in the IC river types R-E2 (=R8) and R-E3 (=R7).

	BI	EQR	nEQR
Reference values	4.80	1.00	1.00
High / Good Boundary	3.84	0.80	0.80
Good / Moderate Boundary	3.13	0.652	0.60
Moderate / Poor Boundary	2.42	0.504	0.40
Poor / Bad Boundary	1.71	0.356	0.20
Lower boundary of Bad status	1.00	0.208	0.00

Table 4. Alternative benchmark sites for IC river types R-E2 (= R8) and R-E3 (= R7) in Bulgaria. EC=electric conductivity, BOD_5=biological oxygen demand, SRP=orthophosphate-P, NO_3-N=nitrate-N, NH_4-N=ammonium-N, LUI=Land use index.

	SiteCode	EC µS/cm	SRP mg/l	NO_3^-N mg/l	NH_4^-N mg/l	LUI %
Alternative benchmark sites ranges		250-620	0.04-0.25	2.0-6.0	0.1-0.25	50-170
	Top_out, 2014	352	0.09	0.66	0.74	150
	ChL_Shi, 2015	793	0.186	2.06	0.04	142
	Kra_out, 2014	537	0.038	2.22	0.11	65
R-E2	Kra_out, 2015	519	0.03	0.401	0.04	65
(=R8)	Bot_Ohr, 2014	445	0.07	1.66	0.04	106
	Bot_Ohr, 2015	540	0.102	1.27	0.04	106
	Bot_Bab, 2015	321	0.03	0.66	0.04	76
	Sku_Tur, 2014	542	0.083	1.65	0.04	170
	Dzh_Dzh, 2014	520	0.03	1.52	0.04	63
	Dzh_Dzh, 2015	449	0.03	0.15	0.06	63
	Sta_Bry, 2014	505	0.03	1.28	0.084	78
R-E3	Sta_Bry, 2015	817	0.054	0.6	0.07	78
(=R7)	BL_Pis, 2015	849	0.179	4.64	0.094	149
	Vit_Gul, 2015	550	0.159	1.17	0.07	120
	Osa_Lev, 2015	585	0.051	0.66	0.29	117
	Sta_Kes, 2015	438	0.087	0.3	0.4	85

Table 5. Pressure-response relationships for the Biotic index in R-E2 and R-E3 river types.

River type	Pressure/Environmental factor	Response	n	R^2	p
R-E2 (= R8)	CLC urban	BI	26	0.49	<0.001
R-E3 (= R7)	CLC agriculture intensive	BI	22	0.32	0.006
R-E2 (= R8)	BOD_5	BI	43	0.06	n.s.
R-E2 (= R8)	PO_4-P (SRP)	BI	43	0.48	<0.001
R-E2 (= R8)	NO_3-N	BI	43	0.14	0.012
R-E2 (= R8)	NH_4-N	BI	43	0.31	<0.001
R-E2 (= R8)	Pressure Index Hydro-morphology	BI	20	0.43	0.002
R-E3 (= R7)	Pressure Index Hydro-morphology	BI	17	0.36	0.011

For all three groups of pressures (land use, chemistry, hydro-morphology), the significant regressions are reported (for chemistry only for type R-E2). In all of the regression models, BI increased with the decrease of the pressures' intensity. R-E2 (= R8) sites were more sensitive to hydro-morphological alterations. The relationship between $CLC_{natural}$ and BI showed the same tendency: when the conditions were close to the natural ones, the BI was higher. The Corine Land Cover (category urban) was negatively associated with values of BI in urban areas in both river types. Densely populated areas in the European lowlands suffer from direct human activities causing morphological alteration and habitat loss (CIS, 2006). However, the small and medium- sized rivers were more sensitive and vulnerable. Similar was the pressure-response relationship between the intensive agriculture and BI for R-E3 (= R7). Among the hydro-chemical parameters, BI was more sensitive to increasing of SRP and NH_4-N.

Therefore, the Biotic index clearly responds to anthropogenic impacts and can be used for the assessment of the ecological status, confirming the findings of HOLT & MILLER (2010).

Conclusions

We have enabled establishing a comprehensive dataset for the IC river types R-E2 (= national type R8) and R-E3 (= national type R7). For both river types, we found significant relationships between various pressures (land use, hydro-chemistry, hydro-morphology) and response (Biotic index). Alternative benchmark sites were identified, using the criteria defined in the EC GIG report (EC GIG, 2015). Based on the selected benchmark sites, the H/G boundary and a reference value for the BI were defined. The other class boundaries were set by using the equal class width approach.

Based on the data from 2014 and 2015, the national assessment method was compared with the final IC exercise of the EC GIG following the fit-in procedure of the European Union (2015). The analysis revealed a good agreement of the national method with the methods from other member states of the GIG. Since all criteria were met, no adjustment was required.

Following the criteria defined in the fit-in-procedure of the European Union (2015), the national assessment method of Bulgaria is considered as comparable with the existing methods. The method was officially approved by ECOSTAT group.

Acknowledgments

This study was funded in the frame of the project „Intercalibration of the methods for analysis of biological quality elements (BQE) for the types of surface waters on the territory of Bulgaria, corresponding to common European types in the Geographical intercalibration groups“, conducted by consortium DIKON-UBA.

References

ARMITAGE P., D. MOSS, J. WRIGHT, M. FURSE. 1983. The Performance of a New Biological Water Quality Score System Based on Macroinvertebrates Over a Wide Range of Unpolluted Running - Water Sites. – *Water Research*, 17: 333-347. [DOI]

CHESHMEDJIEV S., E. VARADINOVA. 2013. [Chapter 5. Demersal macroinvertabrates]. - In: Belkinova D. *et al.* (Eds.), [*Biological analysis and ecological status assessment of Bulgarian surface water ecosystems*]. University of Plovdiv "Paisii Hilendarski", Plovdiv, pp. 147-162. (In Bulgarian).

CIS. 2006. *WFD and hydromorphological pressures.* Good practice in managing the ecological impacts of hydropower schemes; flood protection works; and works designed to facilitate navigation under the Water Framework Directive. Water Framework Directive Technical report. European Commission, Brussels, 68 p.

EC. 2000. Directive 2000/60/EC of the European Parliament and of the Council of 23 October 2000 establishing a framework for Community action in the field of water policy. – *Official Journal of the European Communities* I.327/1-72. Available at: [eur-lex.europa.eu].

EC. 2011. *Guidance document on the intercalibration process 2008–2011. Guidance Document No. 14. Implementation strategy for the Water Framework Directive (2000/60/EC).* European Commission, Technical report-2011-045.

EC GIG. 2015. *Eastern-Continental Lake GIG – Phytoplankton*. Technical Report, 20th July 2015.

European Union. 2013. *Commission decision of 20 September 2013 establishing, pursuant to Directive 2000/60/EC of the European Parliament and of the Council, the values of the Member State monitoring system classifications as a result of the intercali-*

bration exercise and repealing Decision 2008/915/EC.

European Union. 2015. *Procedure to fit new or updated classification methods to the results of a completed intercalibration exercise. Guidance Document No. 30. Technical Report - 2015 – 085.*

Hawkes H.A. 1998. Origin and development of the biological monitoring working party score system. - *Water Research*, 32(3): 964–968. [DOI]

Holt E.A., S.W. Miller. 2010. Bioindicators: Using Organisms to Measure Environmental Impacts. - *Nature Education Knowledge*, 3(10): 8.

McGarrigle M., J. Lucey, K. C. Clabby. 1992. Biological assessment of river water quality in Ireland. - In: Newman, P.J., M.A. Piavaux, R.A. Sweeting (Eds.), *River Water Quality Ecological Assessment and Control*. Brussels (Commission of the European Community), pp. 371-385.

McGarrigle M., J. Lucey. 2009. Intercalibration of ecological status of rivers in Ireland for the purpose of the Water Framework Directive. - *Biology and Environment, Proceedings of the Royal Irish Academy*, 109: 237-246. [DOI]

Opatrilova L. (Ed.) 2011. *WFD Intercalibration Phase 2: Milestone 5 report – River/EC GIG/Benthic Invertebrates.* European Commission Directorate General, JRC, Institute of Environment and Sustainability.

R Core Team. 2017. *R: A language and environment for statistical computing.* Vienna, Austria: R Foundation for Statistical Computing. Available at: [r-project.org]

Yaneva I., S. Cheshmedjiev. 1999. Hydrobiological Monitoring. Surface Running Waters. Irish Biotic Index (Clabby, Bowman, 1979, Clabby, 1989). – In: Peev D., S. Gerasimov (Eds.), *National Program for Biomonitoring of Bulgaria*. Gea Libris Publisher, pp. 158-162. (In Bulgarian).

Ordinance No - 412 from 15. 06. 2011 of the Minister of environment and waters. Appendix II.2. Concerning methods on hydrobiological monitoring of surface waters. (in Bulgarian).

Water basin Directorate "Danube Region". 2010. Danube River Basin Management Plan, 2010-2015. Pleven, Bulgaria. Available at: [bd-dunav.org]. (In Bulgarian).

Zamora-Muñoz C., C. Sàinz-Cantero, A. Sànchez-Ortega, J. Alba-Tercedor. 1995. Are Biological Indices BMWP and ASPT and their Significance Regarding Water Quality Seasonally Dependent? Factors Explaining their Variations. – *Water Research*, 29 (1): 285-290. [DOI]

Received: 05.11.2018
Accepted: 20.12.2018