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Morphological Differentiation of Salmonines (Subfamily Salmoninae) with Emphasis on Trout *Salmo* spp. Stocks in Serbia and Adjacent Regions

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Abstract: The multivariate analysis of the external morphology of 22 continuous characters reveals that huchen *Hucho hucho* and Arctic charr *Salvelinus alpinus* are different from all trout stocks, whereas the Lake Ohrid belvica *Salmothymus ohridanus* is close to the brown trout complex. The position of rainbow trout *Oncorhynchus mykiss* implies greater similarity in trout-salmon morphology in relation to that of huchen and charr. The phenetic relationships derived on external morphology suggest similarity of the west-Danubian brown trout stock of the Black Sea basin to the brown and marble trout stocks from the Adriatic Sea basin, difference of brown trout stock from the Mlava River and similarity of brown trout stock from the Velika Morava River drainage in comparison to that of the Aegean Sea basin. Those results are initial hints for the conservation of brown trout stocks of Serbia.

Key Words: phenetics, evolutionary history, river drainages, brown trout stocks

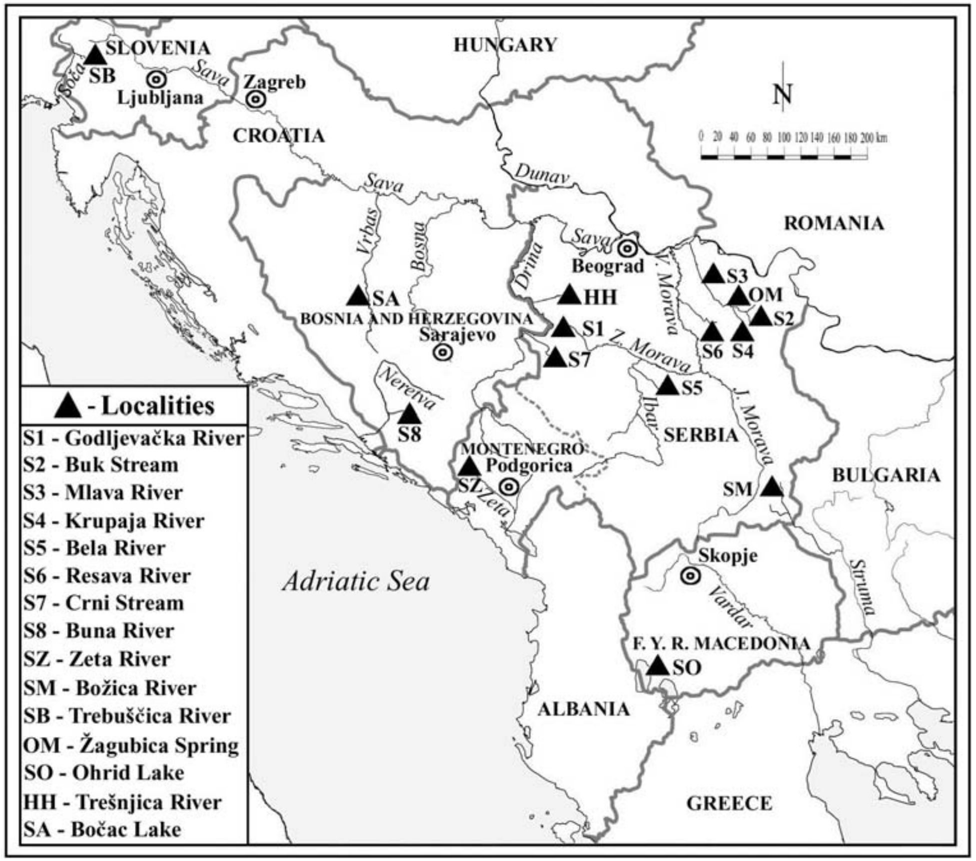

Fig. 1. Sampling area and localities in the West Balkans.

provided from fish farms (Table 1). In addition to the brown trout, samples of several salmonid taxa, e.g., Lake Ohrid belvica *Salmothymus ohridanus* (Steindachner, 1892), rainbow trout *Oncorhynchus mykkis* (Walbaum, 1792), huchen *Hucho hucho* (L., 1758) and charr *Salvelinus alpinus* (L., 1758) were included into the analysis as outgroups. The samples comprised 176 individuals totally, the majority of them was sexually immature and hence the sex was not taken into account as the source of variability.

Analysis of external morphology was accomplished on 22 continuous characters (Fig. 2). Those characters were chosen following Holčík (1989) and Bookstein *et al.* (1985, slightly modified). All measures for bilateral characters were taken from the right side of the fish, except when this was impossible due to loss or damage of the character in concern. The measuring of continuous characters was accomplished using the digital caliper up to the nearest 0.01 mm.

In order to avoid the taxonomical assignments in advance, each sample was considered an OUT (Operational Taxonomic Unit) of Sneath, Sokal (1973). Each character was regressed vs. standard length of fish, in order to transform the sample to the same scale size (Sokal, Rohlf 1995), i.e., to exclude the size-effect on variability as much as possible and residuals were used for the analysis. The morphometric data set

Table 1. Samples of salmonid fish, with their locality names, taxonomical assignments, sample sizes (N), geographical determination of localities from which samples originate, notification on particular specificities and abbreviations used in the paper.

Locality	Taxon	N	Area, drainage, sea basin	Note	Abbr.
Buna River	*Salmo farioides*	12	Herzegovina, Neretva River, Adriatic Sea	Trešnjica fish farm	S7
Trebuščica River	*Salmo marmoratus*	18	Slovenia, Soća River, Adriatic Sea	Tolmin fish farm	SB
Lake Ohrid	*Salmothymus ohridanus*	6	FYR Macedonia, Crni Drim River, Adriatic Sea		SO
Drina River	*Hucho hucho*	11	Western Serbia, Sava River, Black Sea	Trešnjica fish farm	HH
Mlava River	*Oncorhynchus mykkis*	14	Eastern Serbia, Danube River, Black Sea	Žagubica fish farm	OM
Lake Bočac	*Salvelinus alpinus*	10	Bosnia, Sava River, Black Sea	Introduced - stocked	SA
Zeta River	*Salmo taleri*	10	Montenegro, Skadar Lake, Adriatic Sea	Upstream of subterranean pass	SZ
Božica River	*Salmo macedonicus*	25	Southern Serbia, Struma River, Aegean Sea		SM
Resava River	*Salmo trutta*	9	Eastern Serbia, Velika Morava River, Black Sea		S6
Godljevača River	*Salmo trutta*	13	Western Serbia, West Morava River, Black Sea		S1
Buk Stream	*Salmo trutta*	17	Eastern Serbia, Mlava River, Black Sea	Upstream of subterranean pass	S2
Mlava River	*Salmo trutta*	9	Eastern Serbia, Danube River, Black Sea		S3
Krupaja River	*Salmo trutta*	5	Eastern Serbia, Mlava River, Black Sea		S4
Bela River	*Salmo trutta*	7	Central Serbia, West Morava River, Black Sea		S5
Crni Stream	*Salmo taleri*	10	Western Serbia, Drina River, Black Sea		S8

of residuals was analyzed by MANOVA (Sneath, Sokal 1973), which tested the significance of difference in morphological variability between OTUs, by Canonical Discriminant Analysis, which defined multivariate morphological difference between OTUs (Manly 1986), and by UPGMA Cluster Analysis (Sneath, Sokal 1973) on Mahalanobis D^2 distances between OTUs (Sokal, Rohlf 1995), which grouped the most similar OTUs. The analysis was performed using the package STATISTICA v. 5.1. for Windows 95 (StatSoft Inc.). The discriminating power of the most discriminative morphometric characters transformed into the percentages of either head length Lcap (e.g., Lmand), or standard length (e.g., Lcap, tA1) of fish was *a posteriori* tested univariately for the statistical significance afterwards, using ANOVA and Tukey HSD *post hoc* test for unequal sample sizes, i.e., Spjotvoll, Stoline (1973) test.

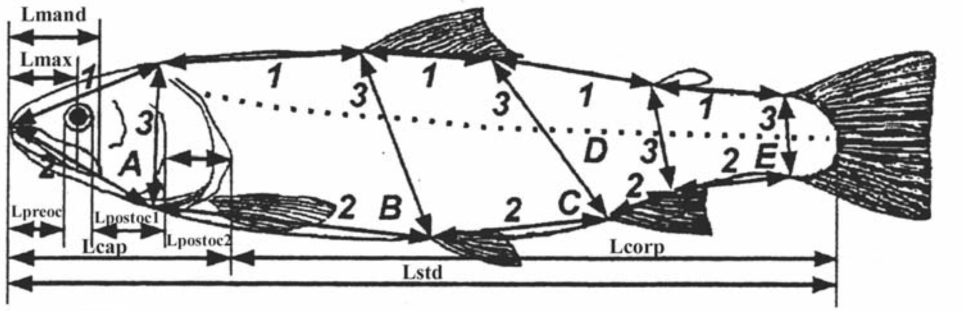

Fig. 2. Character set (LCAP - head length; LCORP - body length; LPREOC - preocular length; LPOSTOC1 - first postocular length from the rear edge of eye cave to the front edge of preopercular; LPOSTOC2 - second postocular length from the front edge of preopercular to the rear edge of opercular; LMAX - upper jaw length from the tip to its most rear spot at upper jaw; LMAND - lower jaw length from the tip of lower jaw to its most rear spot.; tA1 - distance from the tip of snout to the rear end of skull; tA2 - distance from the tip of snout to the isthmus base; tA3 - distance between the rear end of skull and isthmus base; tB1 - distance between the rear end of skull and the beginning of the dorsal fin base; tB2 - distance between the isthmus base and pelvic din base; tB3 - distance between the beginning of the dorsal fin base and pelvic fin base; tC1 - length of the dorsal fin base; tC2 - distance between the base of the pelvic fin and beginning of the anal fin base; tC3 - distance between the end of the dorsal fin base and beginning of the anal fin base; tD1 - distance between the end of the dorsal fin base and beginning of the adipose fin base; tD2 - distance of the anal fin base; tD3 - distance between the beginning of the adipose fin base and end of the anal fin base; tE1 - distance from the end of the adipose fin base and the dorsal end of the caudal fin base; tE2 - distance between the end of the anal fin base and the ventral end of the caudal fin base; tE3 - distance between the dorsal and the anal ends of the caudal fin base).

On testing the consistency of OTUs, the second Canonical Discriminant Analysis on distinct groups (i.e., stocks, or probable taxa) of brown trouts (with *Salmothymus ohridanus* included) as OTUs was accomplished, in order to test the group consistencies, and to gain information on characters discriminating them.

Results

The MANOVA testing of OTUs revealed significant general between-group variability (Rao's $R_{286, 1576} = 12.515$, $p < 0.01$). The majority of characters tested by ANOVA were significantly variable at the level of $p < 0.001$, being with values of $F_{13, 158}$ between 4.956 and 39.773. Only for LMAX the significance of between-group variability was less than 99.9% ($F_{13, 158} = 1.974$, $p < 0.05$).

The Canonical Discriminant Analysis results corroborated that difference in variability of particular OTUs by a total of 97.09% of correct OTU classification (Appendix 1). Only the samples from Godljevačka River (S1, 76.92% of correct classification) and from the Bela River (S5) contained one individual each, both of them were classified to the Buk Stream (S2, 88.23% of correct classification). The first eight Canonical Roots explained 97% of the total discrimination (Table 2) with the character set (LCAP, LCORP, LPREOC, LMAND, tA1, tA2, tA3, tB2, tC1, tD2 and tE3) which repeatedly appeared discriminative. The lower jaw length (LMAND) had the greatest discriminative power (54% of the total one), whereas character tC1 had the smallest (3%). All Mahalanobis D^2 distances between OTUs (Appendix 2) were significant at $p < 0.001$,

Table 2. Standardized Canonical Discriminant Analysis Coefficients (Roots) for morphometric characters of OTUs (characters with strong discriminative power bolded).

Character	Root1	Root2	Root3	Root4	Root5	Root6	Root 7	Root8
LCAP	**0.97**	-0.35	0.13	-0.11	0.25	-0.03	**0.63**	0.05
LCORP	0.06	0.27	0.42	**-0.55**	0.01	**0.51**	-0.41	-0.08
LPREOC	0.47	0.29	**0.52**	0.35	0.12	-0.29	-0.82	0.44
Lpostoc1	0.11	-0.29	0.08	0.03	-0.27	0.13	-0.37	-0.40
Lostoc2	0.05	0.12	-0.03	-0.45	-0.05	-0.18	0.07	-0.14
Lmax	-0.02	0.09	-0.12	-0.01	0.15	0.07	0.09	-0.24
LMAND	**-0.84**	-0.15	**0.67**	**-0.55**	-0.03	0.05	0.30	0.08
tA1	0.08	-0.50	-0.48	**0.63**	0.03	**0.66**	-0.30	-0.04
tA2	-0.81	-0.40	-0.10	0.09	-0.21	**-0.65**	-0.24	0.10
tA3	-0.11	**0.57**	**-0.60**	-0.18	0.25	0.34	0.29	-0.01
tB1	-0.35	-0.29	-0.44	-0.18	-0.03	-0.48	0.13	-0.19
tB2	0.40	-0.39	-0.04	-0.20	**-0.52**	-0.01	0.24	**0.56**
tB3	0.35	0.30	-0.17	-0.25	-0.02	-0.26	-0.44	0.29
tC1	-0.11	0.13	0.20	-0.37	0.10	-0.04	-0.06	**-0.67**
tC2	-0.25	0.20	-0.32	0.17	-0.06	-0.04	0.28	-0.16
tC3	-0.14	-0.15	0.18	0.07	0.33	0.16	-0.27	-0.11
tD1	0.15	0.18	0.18	0.44	0.24	-0.26	0.36	0.33
tD2	0.21	0.25	0.07	**0.63**	**-0.72**	-0.12	**0.55**	-0.03
tD3	-0.19	0.06	-0.12	0.07	0.06	-0.27	-0.10	-0.02
tE1	-0.22	0.30	-0.34	0.32	-0.48	-0.31	-0.40	-0.11
tE2	-0.07	0.16	-0.03	-0.01	0.32	0.22	0.28	-0.11
tE3	-0.34	0.25	0.23	-0.22	-0.37	**0.51**	0.19	**0.61**
Eigenvalue	16.55	12.45	4.51	3.47	3.00	2.04	1.59	1.33
% of discrimination	36	26	10	8	6	4	4	3
Cumulative %	36	62	72	80	86	90	94	97

except S2 - S4, S2 - SM and S5 - SO, being of $p < 0.01$ significance. The UPGMA Cluster Analysis (Fig. 3) clearly separated two non-salmonine outgroup taxa, huchen and charr, from the salmonine ones, since the head length (LCAP) and lower jaw length (LMAND) were the most discriminative characters on the Canonical Root 1. However, the topology of OTUs within the salmonine branch had unexpected configuration. Thus, the Lake Ohrid belvica clustered together with the brown trout from the Mlava and Krupaja rivers (Danube River drainage), who created the distinct brown trout branch, whereas the rainbow trout clustered with the marble trout and brown trout from the western part of the Balkans (Montenegro, Herzegovina and Drina River drainage in West Serbia). The distinct branch of OTUs contained the brown trout samples from streams in the Morava River (Velika - Great and Zapadna - Western) system together with the OTU from the Aegean Sea basin (Struma River drainage). In addition, the group, which consists of Lake Ohrid belvica and brown trout OTUs from Mlava and Krupaja rivers, was distinct from the group of other brown trout OTUs, in which the

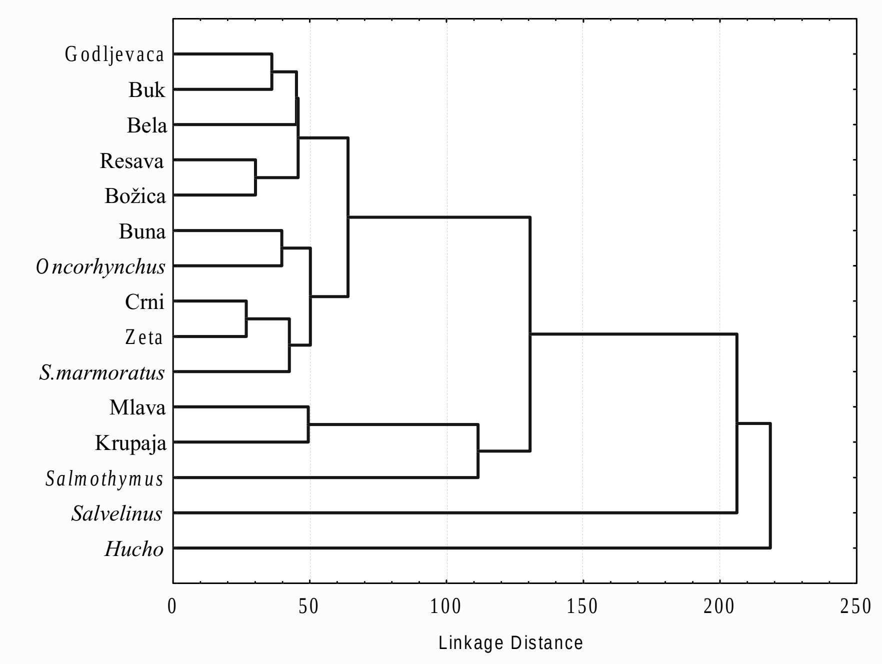

Fig. 3. UPGMA phenogram of salmonid OTUs derived from Mahalanobis distances between them.

western brown trout was distinguishable from the one of the Morava River drainage. Canonical Root 2, with the tA3 as discriminative character, separated marbled trout from the majority of western brown trout samples. Canonical Root 3 with LPREOC, LMAND and tA3, that appeared most discriminating characters, amalgamated the position of OTUs from the previous two Canonical Roots by separating rainbow trout, belvica and marble trout from the large central group of brown trout OTUs and small distinct group of brown trout OTUs from Godljevaua and Krupaja Rivers and Buk Stream. The most discriminative combination of canonical roots appeared to be that of Roots 1 (explained by LCAP and LMAND) and 3 (explained by LPREOC, LMAND and tA3) (Fig. 4). ANOVA testing of OTUs for discriminating characters did not reveal the discrimination concordant with that from Canonical Discriminant Analysis, since the differences between OTUs for characters were inconsistent (Appendix 3), implying an occurrence of a lot of homoplasies.

The second Canonical Discriminant Analysis of the pooled Danube River drainage brown trout stocks into the Danube River drainage western (upper Zeta River, Crni Stream), eastern (Buk Stream, Godljevaua, Resava and Bela Rivers) and Danubian (Mlava and Krupaja rivers), as well as the valid taxa from the Adriatic and Aegean seas' basins revealed the *a priori* classification correctness of 96.32% (Appendix 4), with one western, one Adriatic *S. farioides* and three Aegean *S. macedonicus* incorrectly classified. Certain new characters on head (tA1, tA2), body (tC1) and tail (tD2, tE3) appeared discriminative (Fig. 5). The phenogram (Fig. 6) confirmed the similarity in external morphology between eastern brown trout and *S. macedonicus*. In addition, the western brown trout from the Danube River drainage (*S.* cf. *taleri*) and *S. farioides* appeared more similar in body form than each of them was to the *S. marmoratus*. The

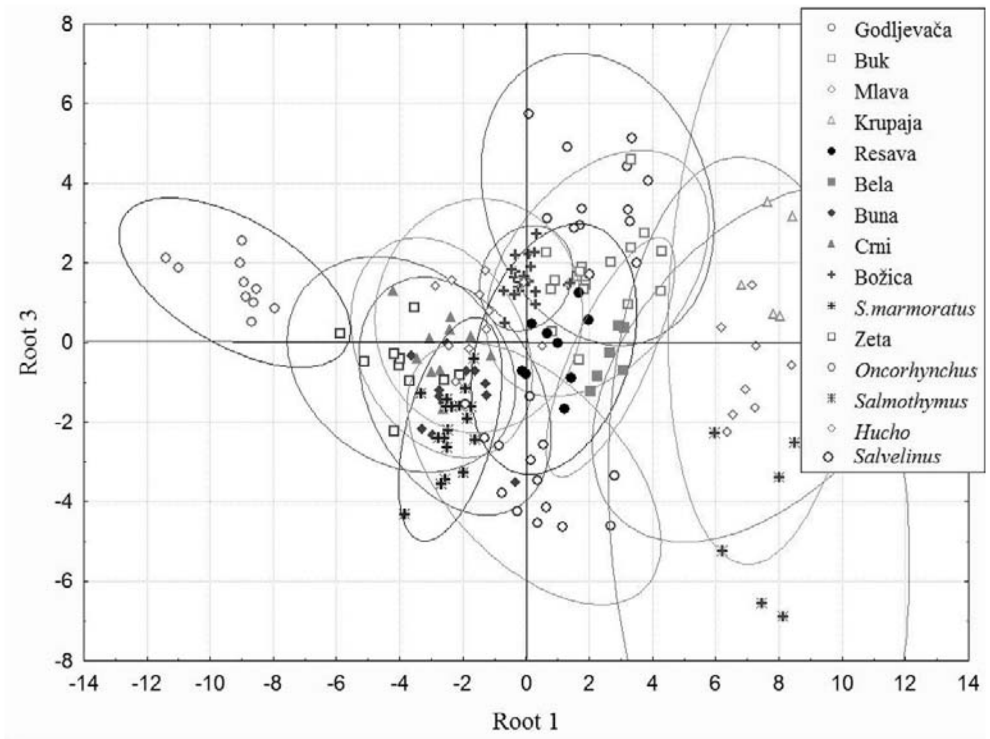

Fig. 4. Position of salmonid OTUs in the space defined by their Canonical Discriminant Scores on Canonical Roots 1 and 3.

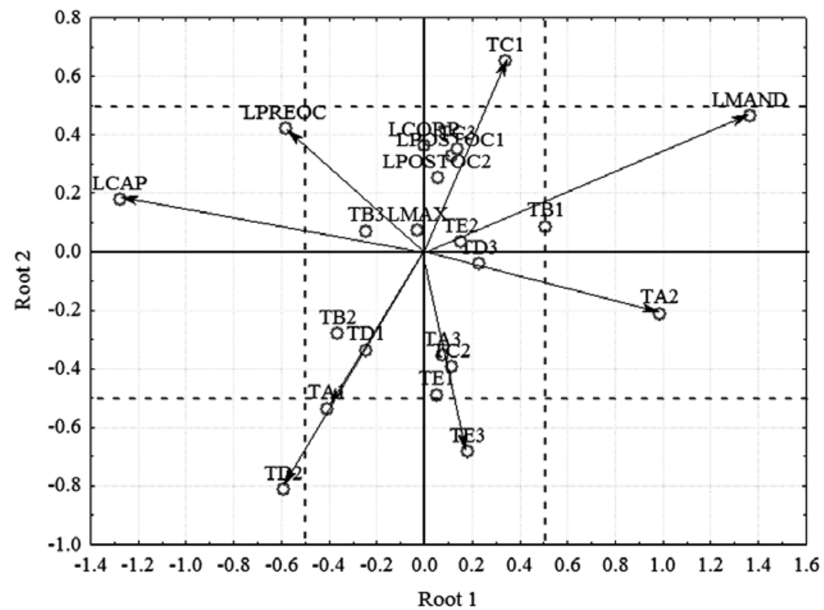

Fig. 5. Canonical loadings of groups of pooled *Salmo* spp.and *Salmothymus* OTUs on Canonical Roots 1 and 2.

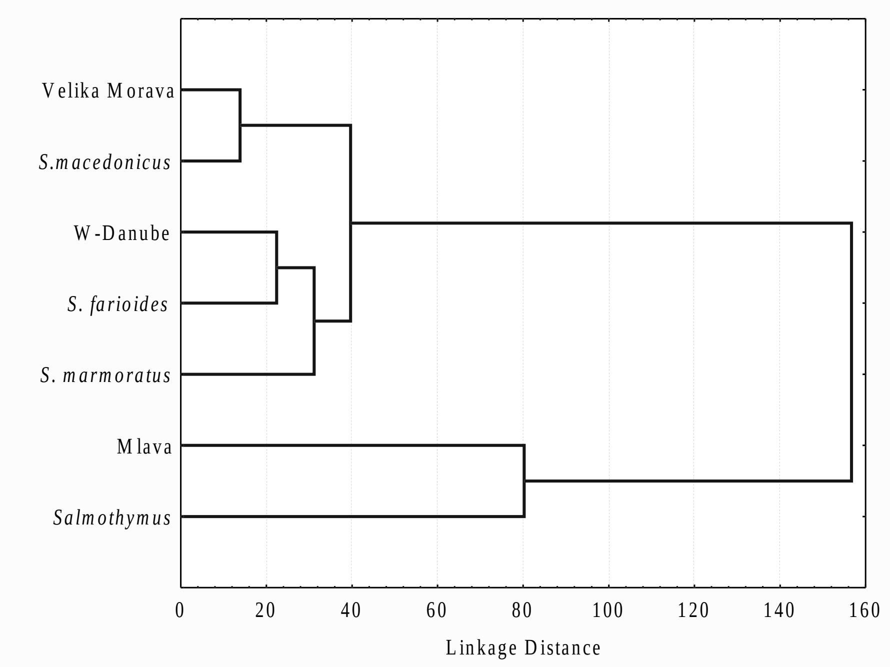

Fig. 6. UPGMA phenogram of groups of pooled *Salmo* spp. and *Salmothymus* OTUs derived from Mahalanobis distances between them.

only deviation from the expected configuration was the Danubian brown trout from the Mlava River drainage, who appeared more similar to the *Salmothymus ohridanus* than to other brown trout taxa.

The phenetic, i.e., morphological relationships between OTUs (Figs 3 and 6) suggest the clear division between the western and eastern brown trout stocks in the West Balkans. Regardless of belonging to the Adriatic Sea basin (e.g. Buna River, upper Zeta River brown trout OTUs and marble trout), or to the Black Sea basin (Crni Stream brown trout, Drina River drainage), western brown trout stocks clustered together. The eastern stocks that belong to the Velika Morava River drainage did the same. Even the isolated Buk Stream sample, which is not in the Velika Morava River drainage (it is in the Mlava River drainage), was the most similar to OTUs from that drainage. Another out-drainage East Balkans sample, the Božica River brown trout from the Struma River drainage, Aegean Sea Basin, clustered with the brown trout OTUs from the Velika Morava river drainage. The brown trout stocks from Mlava and Krupaja (the tributary of Mlava) rivers in the Danube River drainage clustered separately from other brown trout stocks together with *Salmothymus*. The huchen and charr were clearly distinct from all trout stocks in external morphology.

Discussion

The position of OTUs in phenogram (Fig. 3) is informative for the generic relationships of huchen and charr, who clearly separated both from trout (*Salmo*, *Salmothymus* and *Oncorhynchus*) stocks and from each other. This is in contrast to the cladogram of Osinov, Lebedev (2000) derived from 36 enzyme loci, where taimen *Hucho hucho* *taimen* is the salmonid closest to brown trout and to the Atlantic salmon *Salmo salar*. The position of huchen and charr derived from phenetic analysis on external morphological characters is concordant with phylogenetic relationships found on 119 morphological characters by Stearley, Smith (1993). However, the kind of ancestral position of Lake Ohrid belvica *Acantholingua* (*Salmothymus ohridanus* here) close to the huchen and char in their cladogram does not agree with the closer relationship to trout taxa resolved both here (Figs 3 and 6) and in Wilson (1974), Kendall, Behnke (1984), Dorofeyeva (1989) and Oakley, Phillips (1999). Moreover, Snoj *et al.* (2002) found on the mtDNA control region, cytochrome b gene and LDH-C* gene that *Salmothymus* clade holds the intermediary position between the Atlantic salmon and all five lineages of brown trout, thus supporting the statement on the inclusion of *Salmothymus* into the genus *Salmo*.

The dispersal and recent distribution of brown trout stocks in the south-western part of the Balkans seems to be more complex than would be expected on their locking in particular sea basins. The similarity of brown trout OTUs from the upper Zeta River and Crni Stream in Lmand and tA3 (Appendix 3) supports the presumption of Karaman (1938) that after the rising of Dinarid Alp and formation of subterranean break in the Zeta River (or its predecessor lake) continuum, the subsequent (probably glacial) connection of the upper Zeta River with the Piva - Tara - Drina rivers system took place, followed by introgression of brown trout from the ancient Black Sea basin. Karaman (1938) designated brown trout stocks from the West Balkan part of the Black Sea basin (Slovenia, Lika, north-western and central Bosnia and northern Montenegro) *Salmo taleri*, differentiating it from the Adriatic Sea brown trout *S. farioides* by the "slimmer body, smaller mouth opening, scarcer dentation, smaller scales, lower dorsal fin and bigger number of pyloric caeca". The brown trout OTUs from the upper Zeta River and Crni Stream had lower tA3 - rear head height and longer Lmand - lower jaw length than *S. farioides* from the Buna River. In contrast, by shorter Lcap, longer Lmand and higher tA3, *S. taleri* was different from the brown trout stocks of Velika Morava and the Danube rivers' drainages. Until the future confirmation or rejecting by additional morphological and molecular analyses, as well as by phylogeographic processing of brown trout stocks in the Black Sea basin in the Balkans, the assignment of *S. taleri* in conservational sense should be adopted in fishery practice (e.g., in fish-stocking).

The similarity between the brown trout stocks from the Velika Morava River drainge in Lmand and tA3 suggests that this stock differs enough from those of the western part of the Black Sea basin in the Balkans. The close relationship of the Buk Stream with OTUs in the Velika Morava drainage implies that this isolated stock geographically closest to the Mlava River was once connected to the ancient common eastern stock of brown trout. The close morphological relationship of stocks from Božica (*S. macedonicus*) and Resava rivers might suggest their ancient connection, which is to be cleared by both molecular and phylogenetic analyses. The grouping of the brown trout stocks from the Velika Morava (Black Sea basin) and Božica (Aegean Sea basin) rivers in the distinct cluster different from the western brown trout stocks resembles the grouping of brown trout populations in Greece (Apostolidis *et al.*, 1996), where western and north-eastern groups of brown trout stocks were detected, with stocks from the eastern, Ponto-Aegean zone being related to the Danubian, i.e., the Black Sea stocks. By data from Grubić *et al.*, (1982), who designated brown trout from the Resava River (Velika Morava River drainage) *Salmo trutta* m. *fario*, it is not possible to conclude on the status of this stock in relation to the western one (*S. taleri*) from the Black Sea basin. ŠORIĆ (1990) reported that brown trout from the Resava River differs from the Adriatic *S. farioides* by the greater number of shorter and thinner pyloric appendices, slightly larger number of scales above and below the lateral line, smaller depth of head, bigger eye, lower anal fin and smaller pelvic fins. In conservational sense, it seems that stocking with brown trout from hatcheries on streams and rivers in the Velika Morava River drainage should be accomplished exclusively by material from that drainage.

The distinct phenogram branch consisting of brown trout stocks from Mlava and Krupaja rivers suggests their distinctness from other stocks of the Black Sea basin in the region. However, the drawing of a final conclusion from this result is very difficult. Mlava and Krupaja rivers are the only two in Serbia that are typical chalk streams, with the brown trout stocks completely isolated from neighbour trout-holding rivers. Further investigation on brown trout from unstocked streams flowing directly into the Danube River reservoir between the two Djerdap dams shall give more elements for concluding distinctness of the Danubian brown trout stock. Since then, the stocking material that originates exclusively from the Mlava River drainage should be provided, as occurred hitherto once in 2001 in small extent.

In spite of the similarity found between brown trout stock from the Velika Morava River system and that from the Božica River (Struma River drainage), the crossed fish-stocking of those waters has to be avoided until the phyletic and taxonomic status of both stocks is examined in details. This is especially important, since the Božica River stock is strongly defficient and needs urgent conservational measures (MARIĆ *et al.* 2004). In addition, the brown trout stock of the Jerma River, the tributary of Nišava River, Velika Morava River system appeared to hold the same brown trout occurring in the Božica River (unpublished), which reportedly was never stocked by fisheries managers from Serbia, implying that Aegean brown trout was stocked into the Jerma River (Fig. 7) by fisheries managers in adjacent Bulgaria. The brown trout *S. macedonicus* from the Božica River drainage (Fig. 7) is easily distinguishable from that of the Velika Morava River (Fig. 7) system. Brown trout from the Božica river features numerous very large strongly ocellated spots that are dark-red (above the lateral line almost violet) in colour, as well as four dark vertical bars in backgound, whereas those from the Vlasina River feature less numerous small black and slightly ocellated bright red dots and more than 5 (up to 9) less prominent vertical bars in colour. Preliminary molecular data (unpublished) strongly warns for much more care in fish stocking practice, when brown trout stocks of the south-western part of Serbia, both in the Aegean Sea basin and in the Danube River drainage are in concern, since the molecular (i.e., haplotype) diversity has ancient character there and is in a strong need for conservation.

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Fig. 7. Brown trout samples from the Ljubata River, a tributary of the Božica River (middle), Vlasina River, a tributary of the Južna Morava River (bottom) and Jerma River, Južna Morava River drainage (top).

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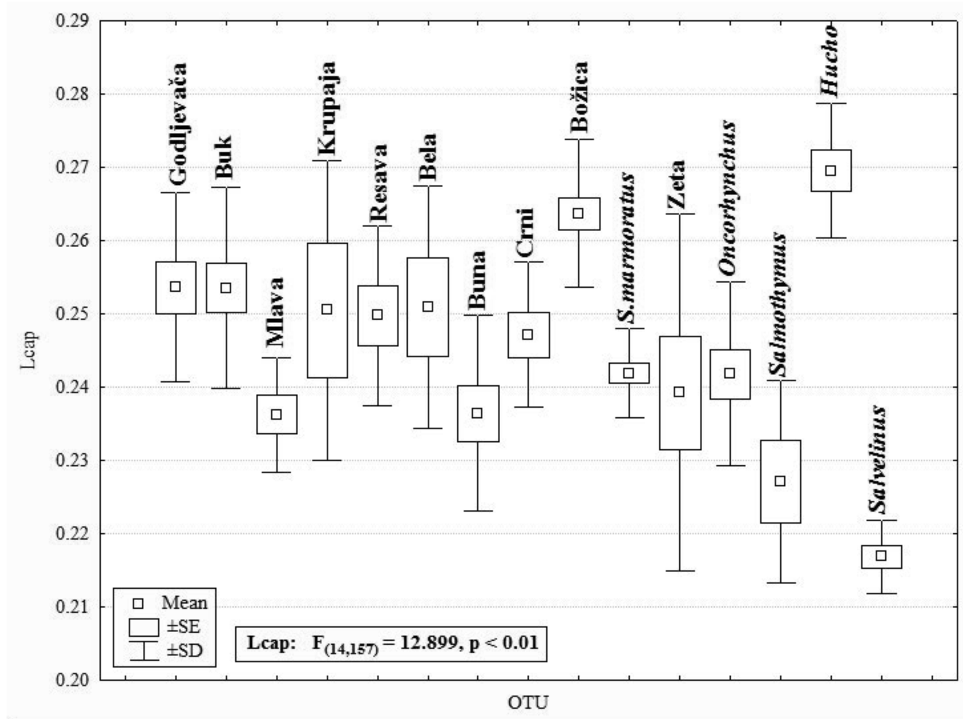
Appendix 1. Percentage of correct classification (%) of OTUs in Canonical Discriminant Analysis (all brown trout OTUs from the Danube River drainage denoted *S. trutta*).

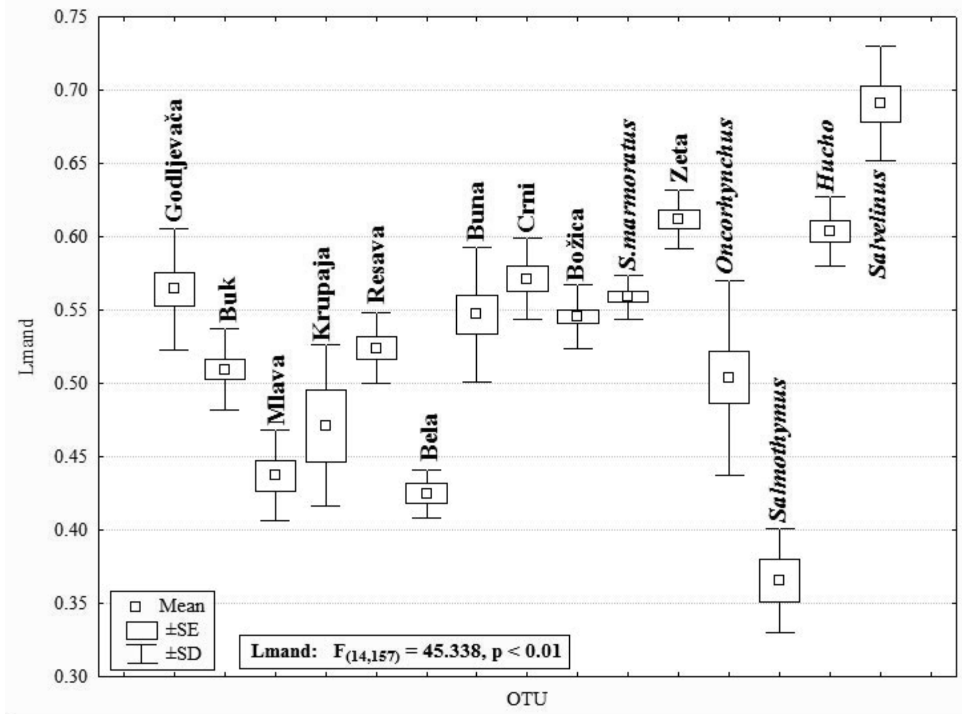
OTU	%	S1	S2	S3	S4	S6	S5	S8	S7	SM	SB	OM	SO	HH	SA	SZ
S. trutta Godljevača R. (S1)	76.92	10								3						
S. trutta Buk Stream (S2)	88.24	1	15				1									
S. trutta Mlava R. (S3)	100.00			9												
S. trutta Krupaja R. (S4)	100.00				5											
S. trutta Resava R. (S6)	100.00					9										
S. trutta Bela R. (S5)	100.00						6									
S. farioides Buna R. (S8)	100.00							12								
S. trutta Crni Stream (S7)	100.00								10							
S. macedonica Božica R. (SM)	100.00									22						
S. marmoratus (SB)	100.00										18					
Oncorhynchus (OM)	100.00											14				
Salmothymus ohridanus (SO)	100.00												6			
Hucho hucho (HH)	100.00													11		
Salvelinus alpinus (SA)	100.00														10	
S. trutta Zeta (SZ)	100.00															10
Total	97.09	11	15	9	5	9	7	12	10	25	18	14	6	11	10	10

Appendix 2. Mahalanobis D^2 distances between OTUs (above the matrix diagonal) and their F values (below the matrix diagonal) with OTU abbreviations, localities and sample sizes (in bracket); plain numbers denote distances with $0.01 < p < 0.001$, and bold ones those with $p < 0.001$.

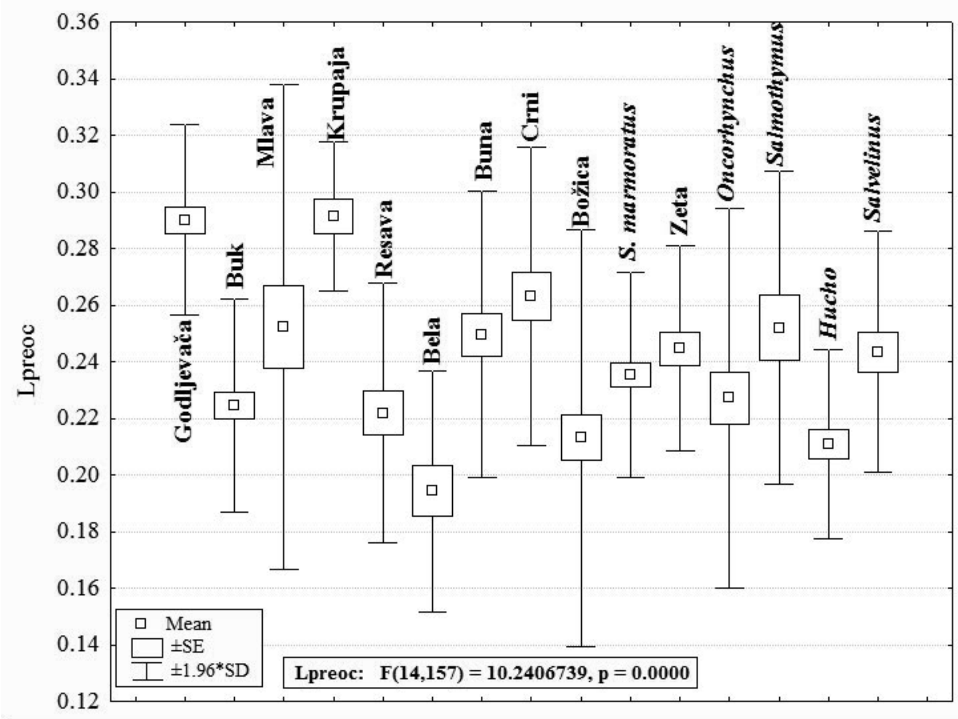
	S1 (13)	S2 (17)	S3 (9)	S4 (5)	S6 (9)	S5 (7)	SF (12)	S8 (10)	SM (25)	SB (18)	OM (14)	SO (6)	HH (11)	SA (10)	SZ (10)
S. trutta Godljevača (S1)	0	36.076	75.846	98.924	68.813	37.766	72.013	66.297	31.322	90.864	78.040	150.959	191.454	200.787	103.252
S. trutta Buk (S2)	**9.740**	0	54.309	60.409	50.808	52.509	50.210	51.621	31.680	81.821	44.524	141.932	214.853	181.080	80.208
S. trutta Mlava (S3)	**14.335**	**11.405**	0	49.399	76.846	47.797	125.315	132.406	74.621	130.722	93.033	80.212	238.618	353.083	178.760
S. trutta Krupaja (S4)	**11.685**	7.611	**5.187**	0	117.683	117.317	149.319	177.267	102.305	176.933	139.192	142.862	313.851	384.002	185.922
S. trutta Resava (S6)	**13.006**	**10.670**	**12.103**	**12.357**	0	53.347	52.514	51.595	30.168	69.280	74.130	122.931	232.915	160.791	74.068
S. trutta Bela (S5)	**5.248**	**7.876**	**5.791**	**10.265**	**6.463**	0	70.072	57.511	38.294	58.134	63.810	114.893	156.576	234.031	96.257
S. trutta Buna (SF)	**16.273**	**12.887**	**22.853**	**17.246**	**9.577**	**9.484**	0	36.341	32.583	41.293	39.700	150.468	201.531	87.868	43.614
S. trutta Crni (S8)	**13.425**	**11.708**	**22.081**	**19.329**	**8.604**	**7.279**	**7.083**	0	31.278	40.910	46.399	189.360	139.107	112.257	26.781
S. macedonica (SM)	**9.418**	11.328	**17.021**	**13.535**	**6.881**	**6.089**	**9.261**	**7.759**	0	40.824	58.484	142.577	139.499	139.005	50.401
S. marmoratus (SB)	**25.168**	**26.554**	**28.000**	**22.559**	**14.840**	**8.844**	**10.859**	**9.479**	**15.101**	0	60.236	212.482	137.202	160.164	44.089
Oncorhznchus (OM)	**19.174**	**12.574**	**18.141**	**16.764**	**14.455**	**9.073**	**9.314**	**9.716**	**18.490**	**17.472**	0	134.104	203.237	169.278	72.966
Salmothymus (SO)	**20.979**	**21.290**	9.718	**12.500**	**14.893**	11.310	**20.366**	**23.966**	**22.672**	**32.325**	**19.068**	0	345.035	374.519	242.414
Hucho (HH)	**41.119**	**52.060**	**41.758**	**35.308**	**40.760**	**20.550**	**41.565**	**25.945**	**37.209**	**34.014**	**45.231**	**45.285**	0	367.820	175.814
Salvelinus (SA)	**40.659**	**41.069**	**58.881**	**41.871**	**26.814**	**29.619**	**17.126**	**19.890**	**34.482**	**37.111**	**35.447**	**47.400**	**68.603**	0	123.587
S. trutta Zeta (SZ)	**20.908**	**18.191**	**29.811**	**20.272**	**12.352**	**12.182**	**8.501**	**4.745**	**12.503**	**10.216**	**15.279**	**30.680**	**32.791**	**21.898**	0

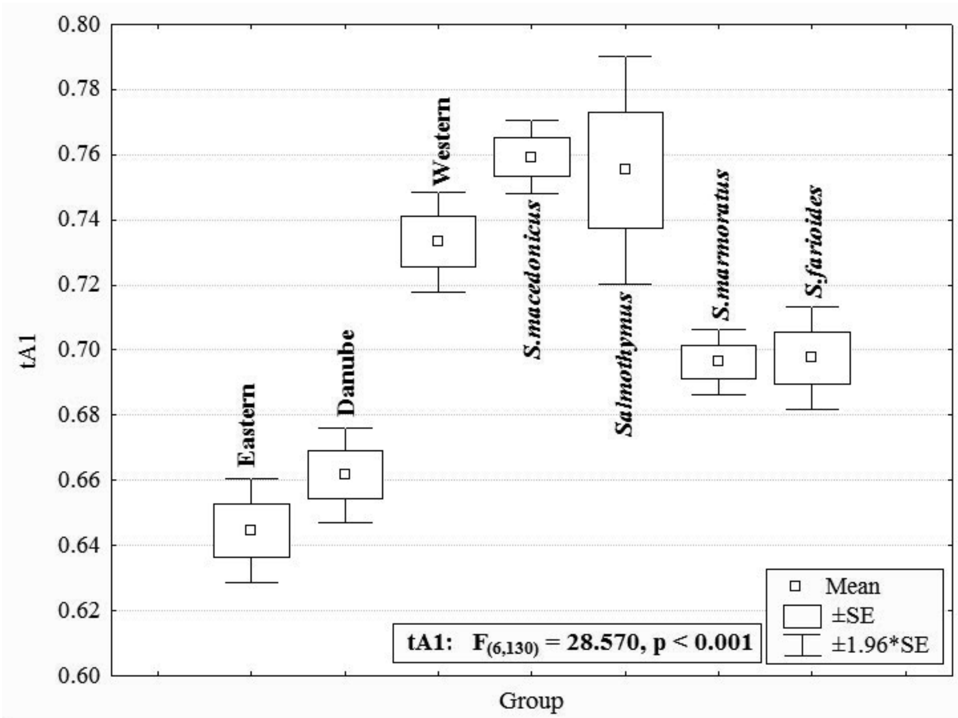
Appendix 3. Box - Whisker plots for discriminating characters with the ANOVA testing results.



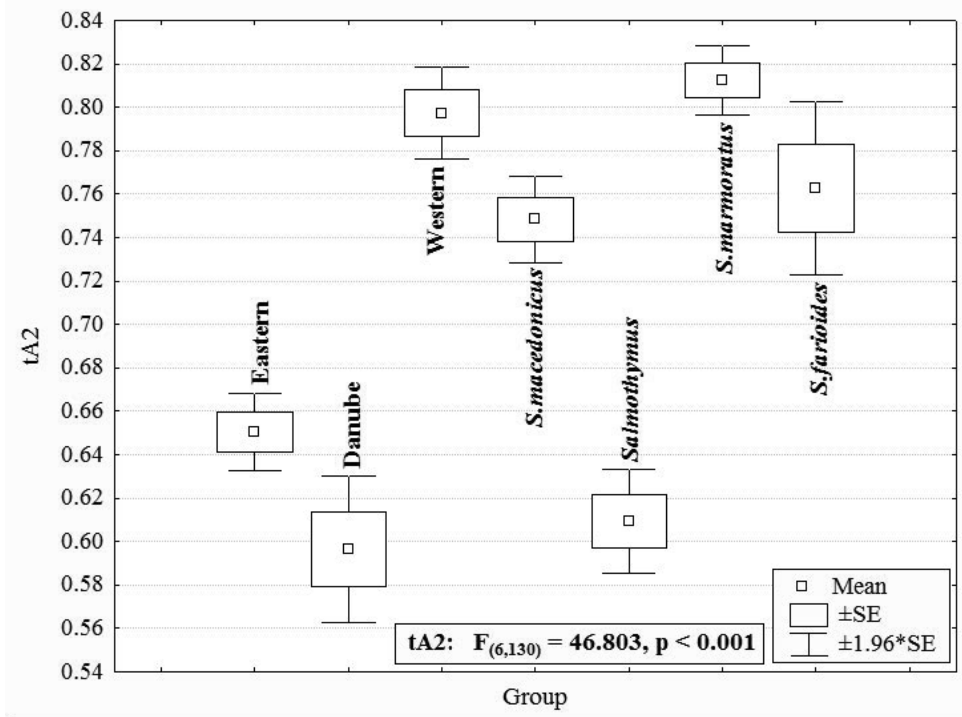


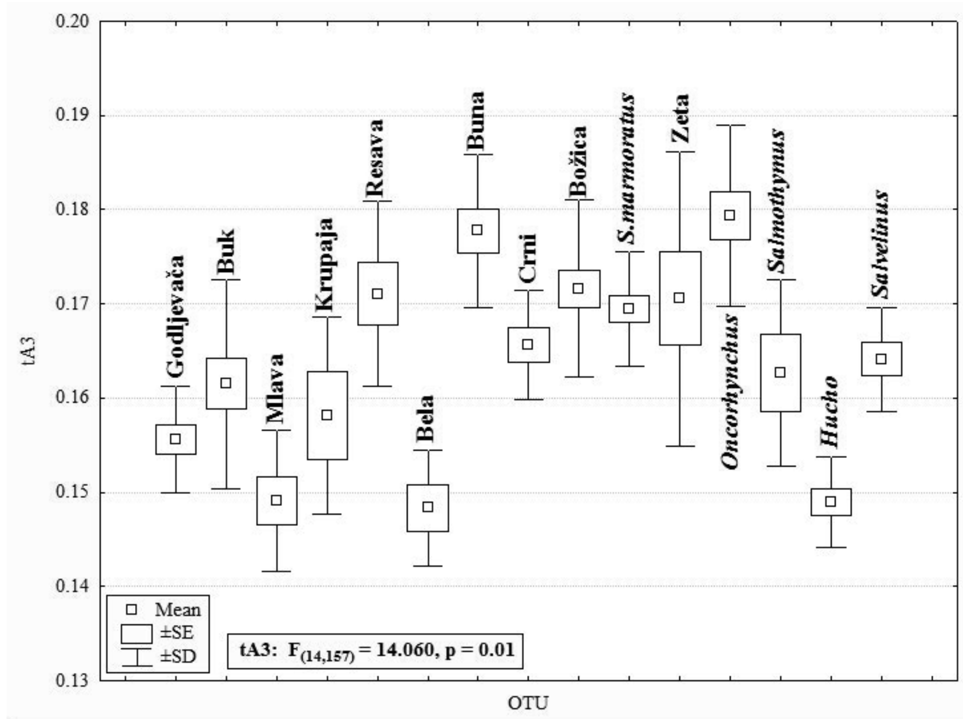
Appendix 3. Continued



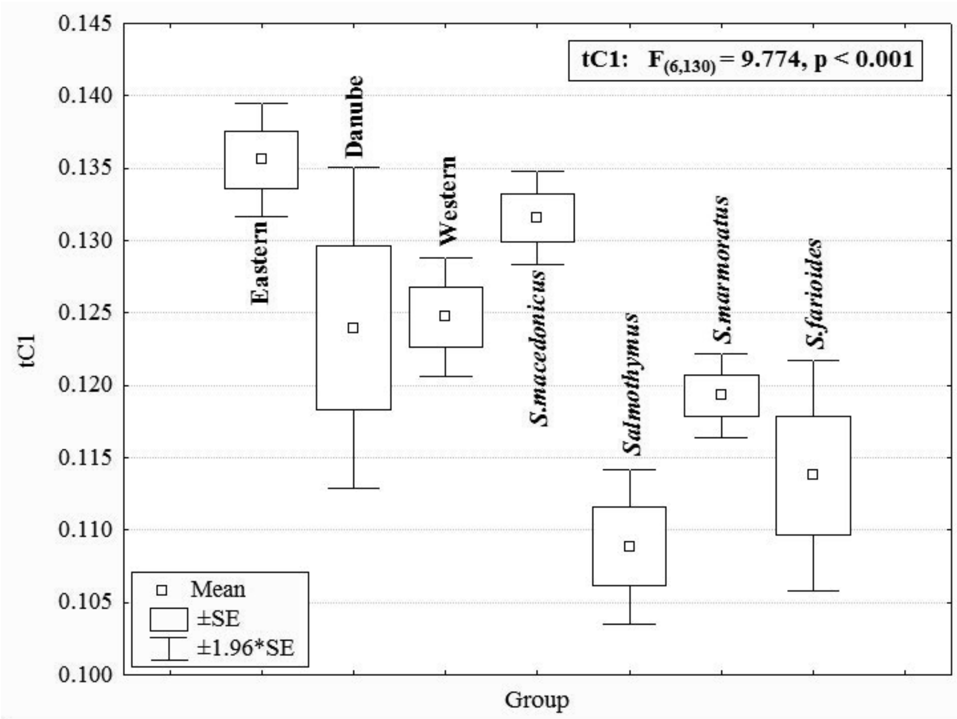


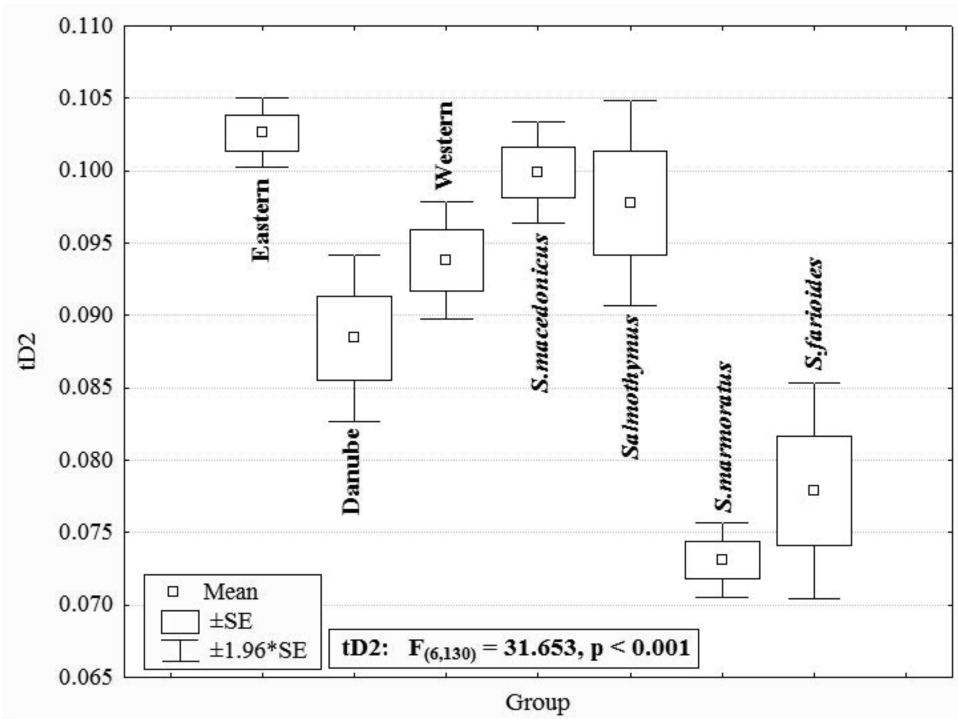
Appendix 3. Continued



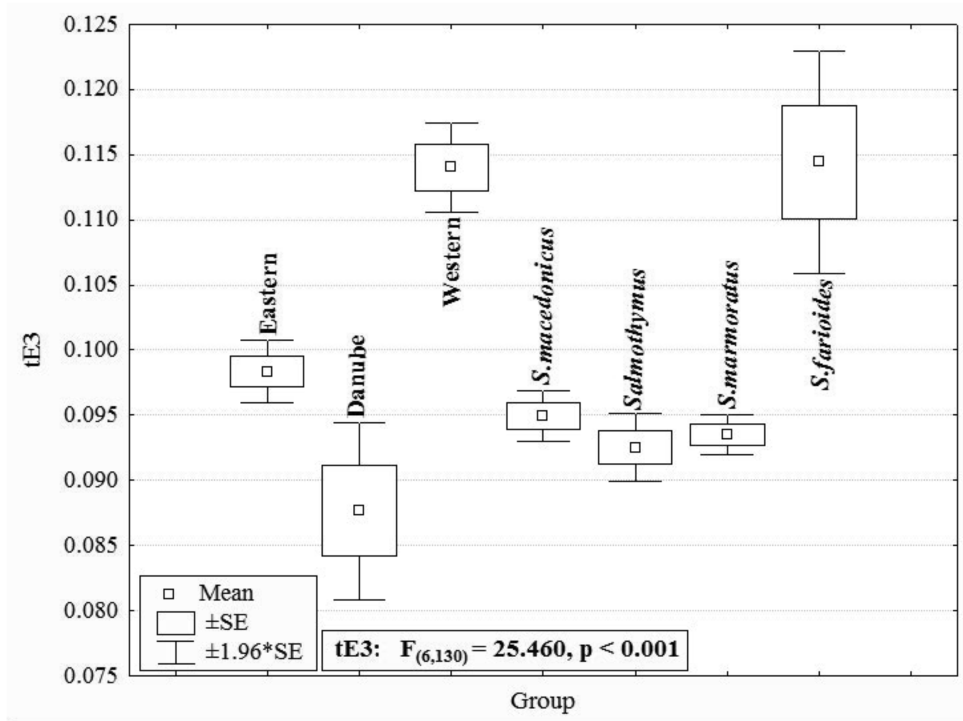


Appendix 3. Continued





Appendix 3. Continued



Appendix 4. Percentage of correct classification (%) of brown trout groups in Canonical Discriminant Analysis.

Brown trout groups	**%**	**E**	**W**	**D**	**SM**	**SB**	**SO**	**SF**
Eastern (E)	93.33	42			3			
Western (W)	95.45		21					1
Danube (D)	100.00			14				
S. macedonicus (SM)	100.00				22			
S. marmoratus (SB)	94.44		1			17		
Salmothymus (SO)	100.00						6	
S. farioides (SF)	100.00							9
Total	96.32	42	22	14	25	17	6	10

Морфологични различия между видовете от подсемейство Пъстървови (Salmoninae) и главно от род Пъстърви (*Salmo* spp.) в Сърбия и прилежащите региони

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(Резюме)

Мултивариантният анализ на 22 външни морфологични белега при Пъстървовите показа, че дунавската пъстърва (*Hucho hucho*) и арктическият сивен (*Salvelinus alpinus*) се различават от всички останали популации, докато белвицата (*Salmothymus ohridanus*) от Охридското езеро принадлежи към комплекса на речната пъстърва. Положението на дъговата пъстърва (*Oncorhynchus mykiss*) показва по-голяма близост с морфологията на видовете от род Salmo, отколкото с тази на дунавската пъстърва и сивена. На базата на външната морфология бе установено, че популациите на речната пъстърва от реките на Западен Дунав (Черноморския басейн) са фенотично близки с тези на речната и мраморната пъстърва от басейна на Адриатическо море и фенотично различни от тези на речната пъстърва в р. Млава. Популациите на речната пъстърва от басейна на р. Велика Морава показват фенотична близост с тези от Егейския басейн. Тези резултати са първите показатели за необходимостта от опазване на популациите на речната пъстърва в Сърбия.