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Anatomical particularities of the dentition in some fish species from the *Salmonidae* family

Radu Constantinescu, Vioara Mireşan, Viorica Coşier, Gabriel Friş, Daniel I. Cocan

Department of Fundamental Science – Biotechnologies, Faculty of Animal Science and Biotechnologies, University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, Cluj, Romania. Corresponding author: D. I. Cocan, cocandaniel@yahoo.com

Abstract. In this study anatomical particularity as morphology, positioning and orientation of teeth of four species of salmonids (*Oncorhynchus mykiss*, *Salmo trutta fario*, *Salvelinus fontinalis and Salvelinus alpinus*) were analyzed. In the upper jaw section, teeth are arranged in two rows, separated by a groove gum. In the midline of the buccal cavity's roof, dentition was found only in brown trout and rainbow trout. For the *Salvelinus* genus species, dentition is missing in that section. From the morphological point of view, brook trout presented the most developed dentition, regardless of anatomic segment. Regarding the length of teeth in this species (*Salvelinus fontinalis*) we obtained the following results: EUJ=1.816±0.06; MUJ=1.504±0.07; LJ=2.116±0.15; T=2.582±0.14. Regarding the diameter of the base of the teeth for the same species we obtained the following results: EUJ=1.05±0.14; MUJ=0.536±0.07; LJ=0.694±0.04; T=1.054±0.12. According to previous studies, we consider that this is the result of adaptations and predatory behavior of species in natural waters, which is a dominant species among salmonids.
Keywords: dentition, *Salmonidae* family, morphology, anatomical particularities

Rezumat. În acest studiu am analizat particularităţile anatomice, caracterele morfologice, dispoziţia şi orientarea dinţilor la patru specii de salmonide (*Oncorhynchus mykiss*, *Salmo trutta fario*, *Salvelinus fontinalis* şi *Salvelinus alpinus*). La nivelul fălcii superioare s-a constatat dispoziţia dinţilor pe două rânduri, separate de un şanţ gingival. La nivelul liniei mediane a plafonului bucal a fost constatată dentiţie doar la păstrăvul curcubeu şi la păstrăvul indigen. La speciile din genul *Salvelinus*, aceasta lipseşte. Din punct de vedere morfologic, păstrăvul fântânel a prezentat cea mai bine dezvoltată dentiţie, indiferent de segmentul anatomic. Referitor la lungimea dinţilor, la această specie am obţinut următoarele rezultatele: EUJ=1.816±0.06; MUJ=1.504±0.07; LJ=2.116±0.15; T=2.582±0.14. Referitor la diametrul bazei dinţilor pentru aceeaşi specie am obţinut următoarele rezultate: EUJ=1.05±0.14; MUJ=0.536±0.07; LJ=0.694±0.04; T=1.054±0.12. În concordanţă cu studiile efectuate anterior, considerăm că acesta este rezultatul adaptărilor şi a comportamentului agresiv al speciei, în apele naturale, aceasta fiind o specie dominantă între salmonide.
Cuvinte cheie: dentiţie, familia *Salmonidae*, morfologie, particularităţi anatomice

Introduction. On the evolutionary scale, different fish species show morphological and behavioral adaptations caused by geographical factors, environment, populational and access to food sources. Basically, their evolution is similar to that of mammals, of course, having specific features of the aquatic environment. Food has always been a basic requirement of living organisms, and depending on its nature and availability, in the case of fish were noticed morphological and behaviour adaptations.

Referring to the first segment of the digestive apparatus of fish -the oral cavity, the specialized literature presents various adaptations, specific to the native environment and feeding behavior (Schwenk & Rubega 2005; Gamal et al 2012). Starting from the position, size and orifice opening of the mouth, in case of predatory fish, one of the specific adaptations is the buccal dentition. This should be a clear distinction between oral cavity dentition and pharyngeal dentition, the latter being specific to fish with an herbivore or omnivore behavior.

Even among predatory fish, in terms of morphology, dentition presents various forms, from canines shaped teeth (*Esociformes*, *Perciformes*, *Anguilliformes*)

(Sadeghinezhad et al 2014), villiform dentition, brush-like arranged (*Siluriformes*) (Chattopadhyay et al 2014), incisor teeth specialized for cutting (*Tetraodontiformes*) (Santini & Tyler 2002), or even molar-shaped teeth (Hulsey et al 2006), used for food triturating (*Characiformes*). Structurally, fish teeth are composed of dentine and are covered with enamel, and in case of large predators such as sharks, the teeth have a central pulp cavity, highly vascularized and innervated (Enax et al 2012; Enax et al 2014). For most predatory fish, the teeth do not show the pulp cavity. They are needle-like formations with permanent replacement (polyphyodont teeth). The reasons for permanent replacement of teeth are multiple, and this mechanism is still being studied. It is possible that the teeth will be replaced only after loosing the previous teeth, but it is not a general rule. For some fish species (*Mullus barbatus*, *Mullus surmuletus*) dentition is monophyodont and is lost without being replaced immediately, after the juvenile stage (Aguirre 1997), due to changes in nutritional spectrum. On the other hand, the shape and dimensions of the teeth is influenced by several factors (Linde et al 2004). The changes occur rapidly, from one generation to another. This explains why, just one generation away, farmed specimens of *Micropterus floridanus* (Lergemouth Bass), have a poorly developed and numerically reduced teeth, compared to wild specimens from which they originated (Selvaraj 2010). They also present an involution, regarding pharyngeal teeth. Similarly, the size of the food consumed and its pressure affects the size of the teeth. This happens in the case of salmon that lives in the spring regions (poor habitats with less food), compared to specimens that live in large streams (Johnson et al 2006). Still in the case of salmon, it was demonstrated that they do not lose their proeminent teeth from the breeding season, which are resorbed and covered by the oral mucosa (Witten et al 2005).

Fish belonging to the Salmonidae family, are considered to be predatory, due to there feeding behavior in their natural habitat. They usually feed on animal origin food, due to its enzymatic equipment (Dabrowski & Glogowski 1977; Reimer 1986; Nya & Austin 2011), which improves the digestion processes. Moreover, in addition to these aspects of the digestion physiology, there are anatomical features that make these species, to have a predatory behavior (Sánchez-Hernández & Cobo 2015). Among these, can be mentioned the hydrodynamic shape of the body, adapted to swimming at high speed (Ojanguren & Braña 2003; Rouleau et al 2010), caudal positioning of dorsal, ventral and anal fins and well-developed caudal peduncle muscles (Standen & Lauder, 2007; Standen 2008). From the point of view of capturing live feed, salmonids are characterized by a relatively large opening of the oral cavity and by a highly complex dentition.

In Romania, salmonid rearing is on an upward trend and one of the measures taken in this regard is species diversification. Until recently, species reared in Romanian trout farms were rainbow trout (*Oncorhynchus mykiss*), brown trout (*Salmo trutta fario*) and brook trout (*Salvelinus fontinalis*). Recently it was introduced the arctic charr (*Salvelinus alpinus*). The four species of salmonids belong from the taxonomical point of view to three genres: *Oncorhynchus* (rainbow trout), *Salmo* (brown trout) and *Salvelinus* (brook trout and arctic charr). Phylogenetically, these species went through different routes (Crespi & Fulton 2004; Shedko et al 2013), thus presenting different anatomical and morphological features, depending on the adaptations made to environmental conditions and food availability (Jensen et al 2004; McMahon 2007). Not least, the feeding behavior influenced the digestive apparatus morphology, including dentition. Although, the specimens studied came from trout farms, their ancestral morphological features were not lost. For this reason, we decided to do a numerical and morphological analysis of oral dentition for the four species of trout.

Material and Method. The aim of this study was to highlight the differences between the four species studied, available on the teeth in the buccal cavity, their number, size and shape. To perform these studies, ten specimens were slaughtered (females) of each species, with weights ranging between 240 and 290 g, and similar age (1.5 years). The heads were detached from the bodies, and dried at 60^{o}C for four hours. By drying soft tissue, teeth become more visible. Depending on the species, determinations were made on the number and arrangement of teeth on the upper and lower jaw, tongue and the roof of the mouth. For counting the teeth, they were stained with methylene blue and silicone rubber molds were made. Statistical analysis of the data was made using GraphPad Prism 6 software. The digital imaging was processed by Adobe Photoshop.

Results and Discussion. By analyzing the images (Figure 1), we can observe a relatively uniform distribution (but random) of the teeth in all four studied salmonid species. Their presence is evident both in the upper jaw and the lower jaw and tongue. The distances between the teeth, regardless of anatomic segment analyzed, do not seem to follow a general rule, in the sense that some are closer to each other compared to others. What is definitely certain, is that starting from the commissures, the teeth show increasing size, as we approach the oral region of the mouth opening. Regardless of the studied species, the orientation of teeth on the sides is inward (medial), and the orientation of the teeth from oral region of the buccal orifice is aboral. Dentition of the tongue is also orientated aboral.

Figure 1. Profile picture of heads of the four species of salmonids (a - *Oncorhynchus mykiss*; b - *Salmo trutta fario*; c - *Salvelinus fontinalis*; d - *Salvelinus alpinus*). Dentition is well highlighted after thermostating.

In all four species of salmonids studied, is observed in the upper jaw, a disposition of two rows of teeth, which are separated by a gingival groove (Figure 2). Latero-medial and oral-aboral orientation of the teeth on the upper jaw clearly reflects the predatory behavior of the studied species. Such a disposition of teeth (prehensile orientation) has a key role in retaining food, generally composed of juveniles of other species, and fish that do not attain large size at maturity. Also, it is found that in all these species, the disposition of teeth is along the length of the upper jaw, from its apical pole and to the commissures of the two jaws. Comparing the teeth of studied species, *Oncorhynchus* and *Salmo* genus present dentition in the midline of the roof of the mouth, while for the species belonging to the *Salvelinus* genus is missing.

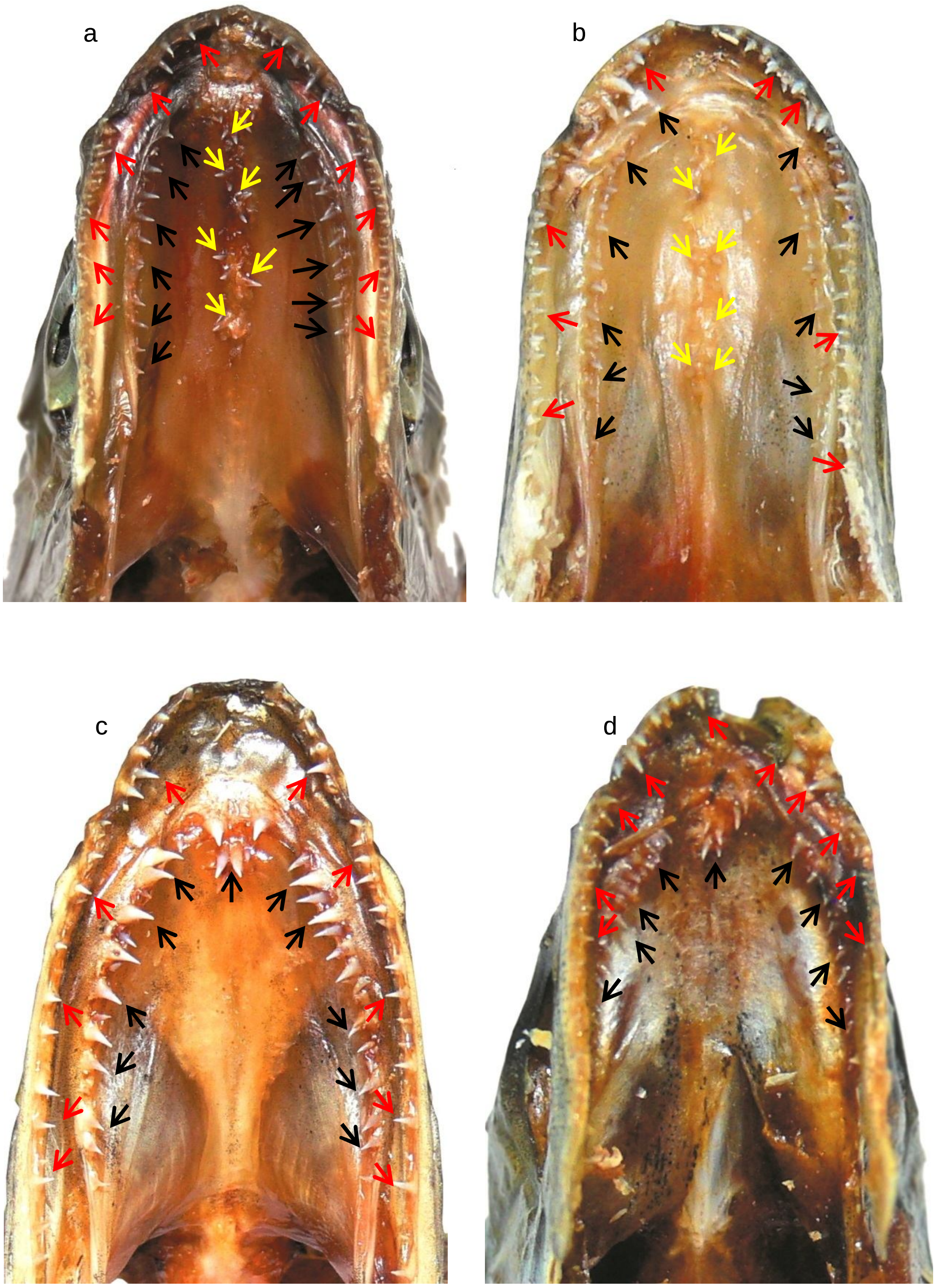

Figure 2. Upper jaw dentition from four salmonid species (a - *Oncorhynchus mykiss*; b - *Salmo trutta fario*; c - *Salvelinus fontinalis*; d - *Salvelinus alpinus*; red arrows - teeth on lateral line; black arrows - teeth on the inner line; yellow arrows - teeth of roof of the mouth).

The orientation of rainbow trout and brown trout teeth from the roof of the mouth is medio-lateral and oro-aboral. The actual origin of the teeth from the midline of the roof is common. They have a successive orientation to left and right, this giving the impression of disposition on two rows of teeth.

Even though for the species of the *Salvelinus* genus, the dentition of the roof of the mouth is missing, there is still the presence of a variable number of teeth (4 to 7) on the apical pole of the bone structure of the roof, less aboral to the cartilaginous papilla of the upper jaw.

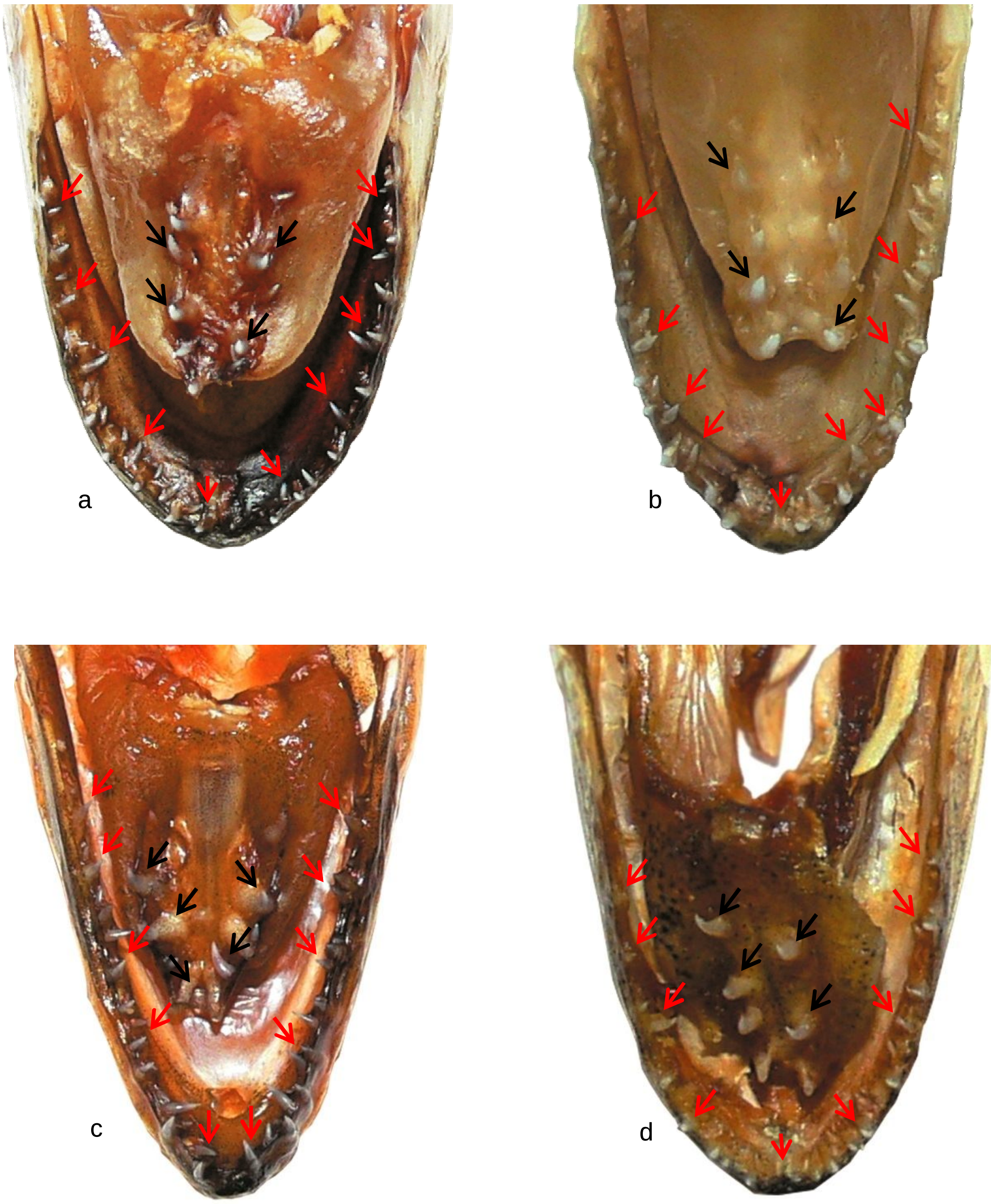

Figure 3. Lower jaw dentition of the four salmonid species (a - *Oncorhynchus mykiss*; b - *Salmo trutta fario*; c - *Salvelinus fontinalis*; d - *Salvelinus alpinus*; red arrows - teeth of lower jaw; black arrows - teeth of the tongue).

As can be seen in Figure 3, the lower jaw shows a single row of teeth on both sides. The teeth appear to be interposed. Between the two larger teeth there is one or two smaller teeth. On the upper jaw, teeth are presented on all full length of the external line. On the lower jaw, regardless of the species studied, teeth near the two commissures (forming the junction between the upper and lower jaws) are missing.

The teeth of the tongue are present in number of 7 to 9, depending on the species. On all the four species, the teeth are arranged on two lines. The lines of teeth in the case of brook trout, tend to merge at the apical pole of the tongue, meanwhile for the other three species, the lines have a parallel trend.

Both in case of the lower jaw and tongue dentition, teeth orientation is also prehensile, which is oro-aboral and latero-medial.

Regarding the mean values for tooth length of the four species studied, as can be seen in Table 1, on the external line of the upper jaw (EUJ), the highest value was recorded for brook trout (TL = 1.816±0.06) and the lowest value was recorded for arctic charr (TL = 1.122±0.06). The same situation was observed for the teeth situated on the medial line of upper jaw (MUJ), the highest value being recorded on brook trout (TL = 1.504±0.07) and the lowest value for arctic charr (TL = 1.035±0.09). Similar observations have been observed on the teeth from the lower jaw (LJ) and tongue (T). Thus, in the lower jaw, brook trout showed an average tooth length of 0.15±2.116 mm, while the arctic charr showed an average tooth length of 1.05±0.12 mm. The average values for tooth length of the tongue were: brook trout - TL = 2.582±0.14 mm vs. arctic charr - TL = 1.469±0.12 mm. Since the species belonging to Salvelinus genus (brook trout and arctic charr) have no teeth at the midline of the roof of the mouth, the mean length value at this level were: brown trout - TL = 1.14±0.08 mm vs. rainbow trout - TL = 1.068±0.08 mm.

Regarding tooth length, the highest value was recorded in brook trout. The same trend in found for the tooth base diameter. The lowest values for tooth base diameter were recorded for the brown trout. On the lateral line of the upper jaw (EUJ), brook trout showed the highest value of TBD = 1.05±0.14, while the lowest value was recorded from brown trout (TBD = 0.395±0.09). On the medial line of upper jaw (MUJ) the highest mean value was obtained for brook trout (TBD = 0.536±0.07), and the lowest mean value was obtained for brown trout (TBD = 0.315±0.03). The same situation was observed in the case of the lower jaw teeth: brook trout (TBD = 0.694±0.04) vs. brown trout (TBD = 0.411±0.08). For the teeth on the tongue the highest mean value regarding the base diameter was recorded for brook trout (TBD = 1.054±0.12), and the lowest mean value for arctic charr (TBD = 0.573±0.03). Because species from *Salvelinus* genus have no teeth at the midline of the roof of the mouth, the base diameter of teeth showed the highest mean value for rainbow trout (TBD = 0.535±0.09) compared to brown trout (TBD = 0.432±0.09).

In Table 2 are presented the mean differences and their statistical significance, regarding the length of teeth in the four salmonid species studied. Comparing the length of teeth of rainbow trout and brown trout, depending on their anatomical area (EUJ, MUJ, LJ, MUL), we obtained statistically insignificant mean differences. Regarding tooth length from the tongue (T), we obtained highly significant differences (MD = -0.616; $p < 0.001$) in favor of brown trout. Comparing rainbow trout and brook trout teeth length, highly significant differences are observed regardless of their anatomical area (EUJ - MD = -0.511, $p < 0.001$; MUJ - MD = -0.394, $p < 0.001$; LJ - MD = -0.802, $p < 0.001$; T - MD = -0.872, $p < 0.001$). Antagonistic to the situation above (when the mean of the differences were negative and in favor of brook trout), when comparing the length of the teeth of rainbow trout and arctic charr, mean differences are positive and in favor of rainbow trout as follows: EUJ - MD = 0.183, $p < 0.001$; MUJ - MD = 0.075, NS; LJ - MD = 0.264, $p < 0.001$; T - MD = 0.187, NS. Comparing the mean values of teeth length for brown trout and brook trout, we obtained negative differences, in favor of brook trout as follows: EUJ - MD = -0.469, $p < 0.001$; MUJ - MD = -0.448, $p < 0.001$; LJ - MD = -0.706, $p < 0.001$; T - MD = -0.256, $p < 0.05$.

Table 1

Mean values and variability indices regarding the length and base diameter of the teeth in the four species studied, according to their position in the buccal cavity

Variables		*Oncorhynchus mykiss*		*Salmo trutta fario*		*Salvelinus fontinalis*		*Salvelinus alpinus*	
		TL (mm)	*TBD (mm)*	*TL (mm)*	*TBD (mm)*	*TL (mm)*	*TBD (mm)*	*TL (mm)*	*TBD (mm)*
EUJ	X±sx	1.305±0.08	0.436±0.09	1.347±0.10	0.395±0.09	1.816±0.06	1.05±0.14	1.122±0.06	0.42±0.07
	SE	0.025	0.027	0.032	0.03	0.021	0.043	0.02	0.021
	V%	6.03	19.85	7.57	24.61	3.76	13.05	5.73	15.91
MUJ	X±sx	1.11±0.10	0.334±0.04	1.056±0.08	0.315±0.03	1.504±0.07	0.536±0.07	1.035±0.09	0.33±0.02
	SE	0.033	0.013	0.025	0.011	0.023	0.024	0.029	0.007
	V%	9.51	12.87	7.57	11.32	4.94	14.05	8.92	6.55
LJ	X±sx	1.314±0.09	0.482±0.16	1.41±0.11	0.411±0.08	2.116±0.15	0.694±0.04	1.05±0.12	0.416±0.09
	SE	0.027	0.05	0.035	0.024	0.047	0.013	0.039	0.028
	V%	6.63	32.8	7.99	18.72	7.01	5.93	11.7	21.51
MUL	X±sx	1.068±0.08	0.535±0.09	1.14±0.08	0.432±0.09	-	-	-	-
	SE	0.024	0.03	0.026	0.028	-	-	-	-
	V%	7.28	18.06	7.17	20.78	-	-	-	-
T	X±sx	1.656±0.08	0.596±0.05	2.272±0.34	0.993±0.22	2.582±0.14	1.054±0.12	1.469±0.12	0.573±0.03
	SE	0.031	0.017	0.106	0.068	0.045	0.037	0.038	0.009
	V%	5.92	9.03	14.78	21.74	5.61	11.22	8.32	5.46

TL - teeth length; TBD - teeth base diameter; EUJ - external line of upper jaw; MUJ - medial line of upper jaw; LJ - lower jaw; MUL - median upper line; T – tongue.

Comparing brown trout mean length of teeth to arctic charr mean length of teeth, we obtained positive differences, favoring brown trout: EUJ - MD = 0.225, $p < 0.001$; MUJ - MD = 0.021, NS; LJ - MD = 0.36, $p < 0.001$; T - MD = 0.803, $p < 0.001$. Finally, comparing the mean lengths of teeth in the two species belonging to the *Salvelinus* genus, the differences are positive and in favor of brook trout, as follows: EUJ - MD = 0.694, $p < 0.001$; MUJ - MD = 0.469, $p < 0.001$; LJ - MD = 1.066, $p < 0.001$; T - MD = 1.059, $p < 0.001$.

Table 2

Mean differences and their statistical significance, regarding the length of teeth in the four species studied, depending on their position in the buccal cavity (Tukey's multiple comparisons test)

Variables	*EUJ*		*MUJ*		*LJ*		*MUL*		*T*	
	MD	*SS*	*MD*	*SS*	*MD*	*SS*	*MD*	*SS*	*MD*	*SS*
Om vs. Stf	-0.042	ns	0.054	ns	-0.096	ns	-0.072	ns	-0.616	oooo
Om vs. Sf	-0.511	oooo	-0.394	oooo	-0.802	oooo	-	-	-0.872	oooo
Om vs. Sa	0.183	****	0.075	ns	0.264	***	-	-	0.187	ns
Stf vs. Sf	-0.469	oooo	-0.448	oooo	-0.706	oooo	-	-	-0.256	o
Stf vs. Sa	0.225	****	0.021	ns	0.36	****	-	-	0.803	****
Sf vs. Sa	0.694	****	0.469	****	1.066	****	-	-	1.059	****

MD - mean difference; SS - statistical significance; Om - rainbow trout; Stf - brown trout; Sf - brook trout; Sa - arctic charr; EUJ - external line of upper jaw; MUJ - medial line of upper jaw; LJ - lower jaw; MUL - median upper line; T - tongue.

Applying the same test of multiple comparisons, but this time regarding the base diameter of teeth of the four species (Table 3), it shows clearly that brook trout dentition is much better developed than in rainbow trout, brown trout and arctic charr. Regardless of the species with which it is compared, the mean differences are in favor of brook trout. Thus, comparing the mean values of teeth base diameter of rainbow trout and brook trout, we obtained the following differences: EUJ - MD = -0.614, $p < 0.001$; MUJ - MD = -0.202, $p < 0.001$; LJ - MD = -0.212, $p < 0.001$; T - MD = -0.458. For the same character comparison, between brown trout and brook trout, mean differences and statistical significance were: EUJ - MD = -0.655, $p < 0.001$; MUJ - MD = -0.221, $p < 0.001$; LJ - MD = -0.283, $p < 0.001$; T - MD = -0.061, NS.

Table 3

Mean differences and their statistical significance, regarding the base diameter of teeth in the four species studied, depending on their position in the buccal cavity (Tukey's multiple comparisons test)

Variables	*EUJ*		*MUJ*		*LJ*		*MUL*		*T*	
	MD	*SS*	*MD*	*SS*	*MD*	*SS*	*MD*	*SS*	*MD*	*SS*
Om vs. Stf	0.041	ns	0.019	ns	0.071	ns	-0.103	o	-0.397	oooo
Om vs. Sf	-0.614	oooo	-0.202	oooo	-0.212	ooo	-	-	-0.458	oooo
Om vs. Sa	0.016	ns	0.004	ns	0.066	ns	-	-	0.023	ns
Stf vs. Sf	-0.655	oooo	-0.221	oooo	-0.283	oooo	-	-	-0.061	ns
Stf vs. Sa	-0.025	ns	-0.015	ns	-0.005	ns	-	-	0.42	****
Sf vs. Sa	0.63	****	0.206	****	0.278	****	-	-	0.481	****

MD - mean difference; SS - statistical significance; Om - rainbow trout; Stf - brown trout; Sf - brook trout; Sa - arctic charr; EUJ - external line of upper jaw; MUJ - medial line of upper jaw; LJ - lower jaw; MUL - median upper line; T - tongue.

Thus, at the tongue level, the difference between base diameters of teeth was insignificant. Brown trout presents a well developed dentition on the tongue. In the comparison between brook trout and arctic charr, differences were highly significant in favor of brook trout.

Conclusions. Morphological characters, disposition and teeth orientation of the four salmonid species studied clearly reflects their predatory behavior. The fact that the specimens taken on our study were reared in trout farms, surely contributed to different results comparing to wildlife specimens. On phylogenetic basis, were transmitted specific morphological characters to the specimens from natural habitats, in spite of changing the nutritional spectrum and feeding behavior.

Statistical analysis revealed the fact that brook trout has a better developed dentition compared to the other three species, regarding teeth length and also the base of teeth diameter. According to previous studies, we consider that this is the result of adaptations and aggressive behavior of the species, in natural habitats this being a dominant species.

Certainly, fundamental studies in this field must be deepened, regarding the number of teeth, dentition formula, structure of teeth and their role. Such studies, don't have a practical applicability in trout farms, but they can sustain intra- and interspecific ecological relations, between components of aquatic biocenosis.

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Authors:
Radu Constantinescu, University of Agricultural Sciences and Veterinary Medicine, Faculty of Animal Science and Biotechnologies, Department of Fundamental Science – Biotechnologies, Romania, Cluj, Cluj-Napoca, 400372, Calea Mănăştur Street 3-5, e-mail: constantinescu.raduusamv@yahoo.com
Vioara Mireşan, University of Agricultural Sciences and Veterinary Medicine, Faculty of Animal Science and Biotechnologies, Department of Fundamental Science – Biotechnologies, Romania, Cluj, Cluj-Napoca, 400372, Calea Mănăştur Street 3-5, e-mail: vmiresan@yahoo.com
Viorica Coşier, University of Agricultural Sciences and Veterinary Medicine, Faculty of Animal Science and Biotechnologies, Department of Fundamental Science – Biotechnologies, Romania, Cluj, Cluj-Napoca, 400372, Calea Mănăştur Street 3-5, e-mail: viorica.cosier@gmail.com
Gabriel Friş, University of Agricultural Sciences and Veterinary Medicine, Faculty of Animal Science and Biotechnologies, Department of Fundamental Science – Biotechnologies, Romania, Cluj, Cluj-Napoca, 400372, Calea Mănăştur Street 3-5, e-mail: gabi_frish@yahoo.com
Daniel Ioan Cocan, University of Agricultural Sciences and Veterinary Medicine, Faculty of Animal Science and Biotechnologies, Department of Fundamental Science – Biotechnologies, Romania, Cluj, Cluj-Napoca, 400372, Calea Mănăştur Street 3-5, e-mail: cocandaniel@yahoo.com

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