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Spring diet composition of Rainbow Trout, *Oncorhynchus mykiss* (Walbaum, 1792) in the Urederra River (Spain)

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This study describes the diet composition of 42 rainbow trout, *Oncorhynchus mykiss* (Walbaum, 1792) (90-480 mm TL), captured in May 1995 in the Urederra River (North of Spain). Rainbow trout of all sizes fed mainly on midge (adults, larvae and pupae), but there were some differences in the feeding habits of small and large fish. We found that as the trout grew larger terrestrial prey became the most important food item. Rainbow trout avoided Leuctridae and showed some preference for Ephemeroptera, Diptera larvae and Nemouridae larvae. Native fish predation and exploitation of the same trophic resources in the Urederra River, could negatively affect native fishes, such as European minnow, *Phoxinus phoxinus* L., 1758 or brown trout, *Salmo trutta* L., 1758.

Keywords : *Oncorhynchus mykiss,* rainbow trout, diet composition, prey selection, Urederra River, Spain.

Introduction

Rainbow trout, *Oncorhynchus mykiss* (Walbaum, 1792), occur naturally in the eastern Pacific Ocean, and in fresh water they exist mainly in the west of the Rocky Mountains from northwest Mexico to the Kuskokwim River, Alaska (Jonsson et al. 1993). The species was first introduced in Spain in the late 19th century (Elvira 1995) and it is still present in all Spanish basins (Doadrio 2001), especially in areas close to fish farms, from where specimens frequently escape (Carss 1990). The introduction of rainbow trout can alter native fish populations through predation, competition for food or territorial space, acting as carriers of different diseases and through destruction of earlier trout spawning beds (Landergren 1999).

Knowledge of the interaction between exotic and native species is necessary for adequate aquatic management. In this way, diet studies constitute an important tool (Neveu 1979), because trophic interaction between species (competition and predation) is an important mechanism in determining the distribution of aquatic communities (Lammens et al. 1992, Declerck et al. 2002).

The aim of this study is to describe the diet composition of rainbow trout in the Urederra River, as a first step in evaluating the interaction between rainbow trout and native fishes. The possible interaction will be discussed in comparison with previous works describing the feeding preferences of brown trout, *Salmo trutta* L., 1758 (Oscoz et al. 2000, Oscoz 2003) and European minnow, *Phoxinus phoxinus* (L., 1758) (Oscoz et al. 2001) in the nearby Larraun River.

Study area

Our study was carried out in the Urederra River (Navarra, North of Spain), a tributary of the Ega River (Ebro basin) (Fig. 1). The Urederra River has a catchment area of 319 km^2 and a total length of 20 km. Its altitude ranges from 750 m at the source to 450 m at its confluence with the Ega River, flowing mainly over a limestone watershed. Bedrock, boulders and cobbles dominate the river substrate, whereas the riparian vegetation consists mainly of alders *(Alnus glutinosa)* and willows (*Salix alba* and *S. purpurea*).

Whilst Brown trout (*Salmo trutta* L., 1758) and Eu-

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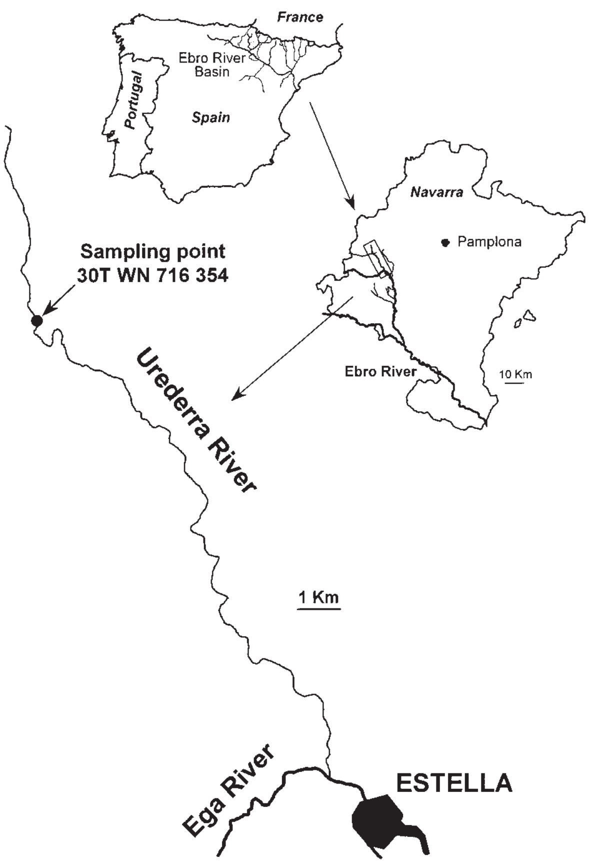

Fig. 1. Maps locating the Ebro River Basin and Navarra in the Iberian Peninsula (top left), the Urederra River in Navarra (right), and the sampling point location in the Urederra River.

ropean minnow (*Phoxinus phoxinus* (L., 1758)) occur with the rainbow trout in the studied river stretch (Oscoz et al. 1999), the rest of the river contains French nase (*Chondrostoma miegii* Steindachner, 1866), Graells barbel (*Barbus graellsii* Steindachner, 1866), Stone loach (*Barbatula barbatula* (L., 1758)), Gudgeon (*Gobio gobio* (L., 1758)) and eel (*Anguilla anguilla* (L., 1758)) (Campos et al. 1997).

There is an aquaculture farm in the upper stretch of the Urederra River where species frequently scape into the river. This farm is dedicated exclusively to cultivating rainbow trout up to a size of 60-80 g. Although the escaped rainbow trout have been reported to breed to maturity, there is no evidence of successful reproduction. The studied river stretch in base flow has an average depth of 34 cm and a mean water surface width of 9 m, and the river bed is dominated by rubble and cobble (64-256 mm size, Simonson et al. (1994)).

Material and methods

Rainbow trout were captured by electrofishing surveys carried out in a 100-m-length river stretch in May 1995. A detailed description of the river stretch characteristics and electrofishing surveys are in Campos et al. (1997). Rainbow trout were preserved in ice, but not frozen. In the laboratory, the stomachs were removed and preserved in a 4% formalin solution. The stomach content was identified under a magnifying microscope (x7-45). The stomach contents of 42 rainbow trout (90-480 mm total length (TL)) were analyzed. Fish age determination was done by scale annuli reading and validated with length-frequency histograms. Age classes 1 (<120 mm TL), 2 (120-170 mm TL) and >2 (>170 mm TL) were identified.

Four prey groups were identified : plant material, fishes, terrestrial invertebrates and aquatic invertebrates. When possible, aquatic invertebrates were identified to family level, terrestrial invertebrates to order level and fishes to species level. Because of the low occurrence and volume of plant material present in the stomachs the abundance of plant material was not quantified, and only the number of stomachs in which it appeared was noted.

In animal prey items, the frequency of occurrence of a given prey type is defined as the number of stomachs in which that prey occurs, expressed as a frequency of the total number of stomachs in which prey are present (Hynes 1950). The relative abundance of a prey (or contribution to the stomach contents) is defined as the percentage of total stomach contents in all predators comprised by that given prey. In mathematical terms, the percentage occurrence ($\%F_i$) and the percentage abundance ($\%A_i$) of prey type i can be described by the equations :

$$\%F_i = (N_i / N)\text{x}100 \qquad \%A_i = (\sum S_i / \sum S_t)\text{x}100$$

where, N_i is the number of predators with prey i in their stomach, N is the total number of predators with stomach contents, S_i the stomach content (number) composed by prey i, and S_t the total stomach content of all stomachs in the entire sample (Amundsen et al. 1996). The differences in diet composition between age classes were analyzed with a χ^2 test (significance $P<0.05$).

Trophic diversity was calculated according to Shannon's index ($H' = -\sum S_i \cdot \log_2 S_i$). Furthermore, in order to evaluate specialization in the diet of rainbow trout evenness index ($E = H'/H'_{max}$) was determined, considering that values close to zero mean a stenophagous diet and those closer to one a more euryphagous diet.

Feeding strategy diagrams were constructed following the Costello (1990) method with the modifications suggested by Amundsen et al. (1996). These diagrams are based on a two-dimensional representation, where each point represents the frequency of occurrence (%Fi) and the prey-specific abundance ($\%P_i = (\sum S_i/\sum S_{ti}) \cdot 100$), where P_i is the prey-specific abundance of prey *i*, and S_{ti} is the total stomach content in only those predators with prey *i* in their stomach.

Prey selection was also analyzed, comparing diet composition with benthic macroinvertebrates present in the river stretch. A benthos sample was collected using a Surber net with 0.1-mm mesh size in four replicates of 0.25 m^2. The sample was preserved at the capture site with 4% formalin solution. In the laboratory all the macroinvertebrates were counted and classified. Prey selection was quantified using Savage's index (Savage 1931) $W_i = A_i/D_i$, where A_i is the relative abundance of prey *i* in the stomach content, and D_i is the relative availability of this resource in the river. The values close to 1 in W_i mean no selection of prey *i*, and values lower and greater than 1 show avoidance (negative preference) and selection (positive preference) respectively. This index was chosen because it is more objective than other similar indices, and it is possible to verify its statistical significance with a χ^2 test (Manly et al. 1993).

Results

A total of 42 fishes were analyzed. Three rainbow trout had empty stomachs and were not analyzed further. In the other stomachs analyzed, 4203 preys were identified (Table 1). These belonged mainly to terrestrial Diptera and aquatic Chironomidae (larvae and pupae). One specimen (391 mm TL) fed only on fish (35 minnows) and another specimen (480 mm TL) fed only on snails (67 Lymnaeidae). Three age >2 individuals presented plant material in their stomach contents.

Relative abundance of prey items differed significantly between different age classes (χ^2= 563.3, 16 df, P<0.001). Older specimens increased the use of terrestrial invertebrates, until aquatic and terrestrial prey were equally consumed (Fig. 2). With reference to aquatic prey, the use of Ephemeroptera and Diptera larvae decreased as trout grew older, but the use of Chironomidae pupae increased (Table 1). The mean number of prey per stomach was higher in the older trout (Age 1 : 65.0 prey per stomach, Age 2 : 118.8, Age >2 : 141.3).

Trophic diversity and evenness indices decreased as trout grew larger (Table 1), suggesting less euryphagous feeding behavior. This feeding strategy change was confirmed by the feeding strategy plots (Fig. 3).

On the other hand, benthic prey selection was similar between different age classes (Table 2). All trout positively selected Baetidae, Heptageniidae, Nemouridae, Rhyacophilidae, Stratiomyidae and Nematoda, whereas Leuctridae was avoided. Furthermore, age 2 and age 1 trout positively selected Simuliidae, while age 2 and age >2 trout positively selected Chironomidae.

Discussion

The diet of rainbow trout in Urederra River includes benthic invertebrates, fishes and terrestrial invertebrates. A similar diet composition has been described in e.g., American lakes (Rabe 1967), Australian rivers (Pidgeon 1981), an Argentinean reservoir (Ferriz 1988) and Hawaiian streams (Kido et al. 1999). Different works have highlighted the opportunistic feeding behaviour of rainbow trout (Artigas et al. 1984, Gibson 1988, Ferriz 1994), identical to other salmonids (Vignes 1998), that modify their diet depending on prey availability. For this reason, the high consumption of Chironomidae pupae and terrestrial Diptera could be explained by the fact that the study period coincided with the highest emergence period of these prey, when usually they are very abundant and readily available.

Diet variation in fishes is related to prey availability, their accessibility and the risk of predation (Eggers 1982, Greenberg et al. 1997). As a result bigger prey that are easier to capture or with high energetic value are consumed more. Since terrestrial prey are easier to detect and have lower evasive ability (McLaughlin et al. 1994), during their emergence they become an important and valuable prey for visual predators such as rainbow trout. Nevertheless, terrestrial prey consumption is also related to other factors like topography, riparian vegetation or weather (Artigas et al. 1984, Vollestad & Andersen 1985, Cavalli et al. 1997).

The presence of European minnow in larger rainbow trout's (Age >2) stomachs confirms their predation on small native fishes. The apparently high consumption of Nematoda may not be real since some could be trout parasites (Molloy et al. 1995, Brotheridge et al. 1998, Byrne et al. 2002). Further studies would be necessary to determine whether their presence is due to real active consumption or parasitism.

The point distribution of the feeding strategy plots and the decrease of the trophic diversity and evenness indexes suggested less euryphagous feeding behaviour as trout grew larger. The positioning of the points mainly in the lower part of the feeding strategy plots show that the average contribution of the prey items to the stomach contents was low, indicating a generalized feeding strategy (Amundsen et al. 1996). However, some prey items contribute more to the stomach contents as trout grow larger, i.e. they became less generalist.

Diet variation as fishes grow is well documented in

Table 1. Diet composition of rainbow trout in the Urederra River (May 1995). The data are expressed as percentage of occurrence (%Fi) and relative abundance (%Ai). (L: Larvae; P: Pupae; I: Adult). Values of trophic diversity (H') and evenness index (E) are shown.

	Age 1		Age 2		Age >2		Total	
	$\%F_i$	$\%A_i$	$\%F_i$	$\%A_i$	$\%F_i$	$\%A_i$	$\%F_i$	$\%A_i$
AQUATIC INVERTEBRATES								
Hidracarina	7.69	0.12	-	-	16.67	0.12	7.69	0.07
Nematoda	61.54	15.98	92.86	7.64	58.33	5.96	71.79	8.64
Gammaridae	15.38	0.24	7.14	0.06	-	-	7.69	0.07
Isopoda	-	-	-	-	8.33	0.06	2.56	0.02
Lymnaeidae	-	-	-	-	8.33	3.95	2.56	1.59
Baetidae	84.62	18.11	00.00	7.52	75.00	3.36	87.18	7.97
Heptageniidae	69.23	4.85	57.14	1.62	50.00	2.60	58.97	2.66
Leuctridae	7.69	0.12	-	-	-	-	2.56	0.02
Nemouridae	84.62	2.37	57.14	1.56	41.67	0.47	61.54	1.28
Perlodidae	-	-	7.14	0.06	-	-	2.56	0.02
Hydroptilidae	15.38	0.36	7.14	0.06	8.33	0.06	10.26	0.12
Limnephilidae	-	-	-	-	33.33	0.24	10.26	0.10
Rhyacophilidae (L)	15.38	0.59	64.29	0.90	33.33	0.35	38.46	0.62
Rhyacophilidae (P)	-	-	7.14	0.06	8.33	0.06	5.13	0.05
Chironomidae (L)	84.62	13.25	85.71	3.49	66.67	1.95	79.49	4.83
Chironomidae (P)	69.23	11.72	00.00	20.87	75.00	27.49	82.05	21.70
Limoniidae	-	-	21.43	0.18	8.33	0.06	10.26	0.10
Simuliidae (L)	69.23	12.07	85.71	5.05	58.33	2.42	71.79	5.40
Simuliidae (P)	23.08	1.18	35.71	0.48	-	-	20.51	0.43
Stratiomyidae	15.38	0.71	28.57	0.66	16.67	0.24	20.51	0.50
Dytiscidae (I)	7.69	0.12	7.14	0.06	-	-	5.13	0.05
Haliplidae (I)	-	-	7.14	0.06	-	-	2.56	0.02
TOTAL AQUATIC INV.	00.00	81.78	00.00	50.33	91.67	49.38	97.44	56.27
FISHES								
Phoxinus phoxinus	-	-	-	-	8.33	2.06	2.56	0.83
TERRESTRIAL INVERTEBRATES								
Diptera	69.23	17.40	92.86	48.59	75.00	46.90	79.49	41.64
Coleoptera	15.38	0.47	42.86	0.66	50.00	0.65	35.90	0.62
Lepidoptera (L)	7.69	0.12	14.29	0.12	16.67	0.12	12.82	0.12
Molusca	-	-	-	-	8.33	0.06	2.56	0.02
Formicidae	7.69	0.12	14.29	0.24	33.33	0.35	17.95	0.26
Hemiptera	-	-	7.14	0.06	8.33	0.06	5.13	0.05
Non-identified Insecta	7.69	0.12	-	-	41.67	0.41	15.38	0.19
TOTAL TERRESTRIAL INV.	69.23	18.22	00.00	49.67	75.00	48.55	82.05	42.90
PLANT MATERIAL								
Plant material	-	-	-	-	25.00	-	7.69	-
Number of stomachs	13		14		12		39	
Total number of preys	845		1663		1695		4203	
H'	3.09		2.41		2.39		2.70	
E	0.72		0.54		0.52		0.55	

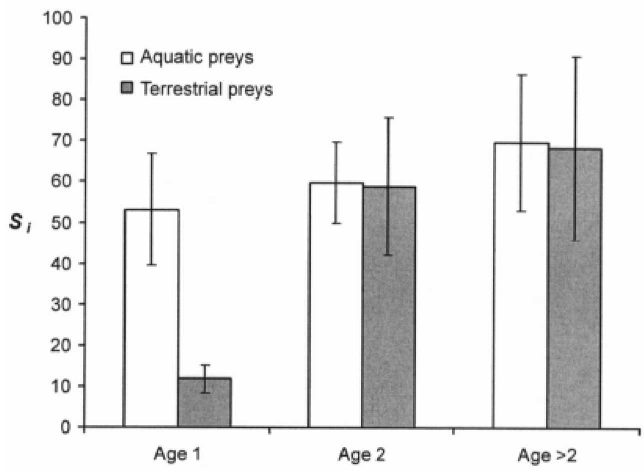

Fig. 2. The mean number of prey (Si) consumed by rainbow trout of different age classes in the Urederra River (May 1995). Error bars indicate standard error.

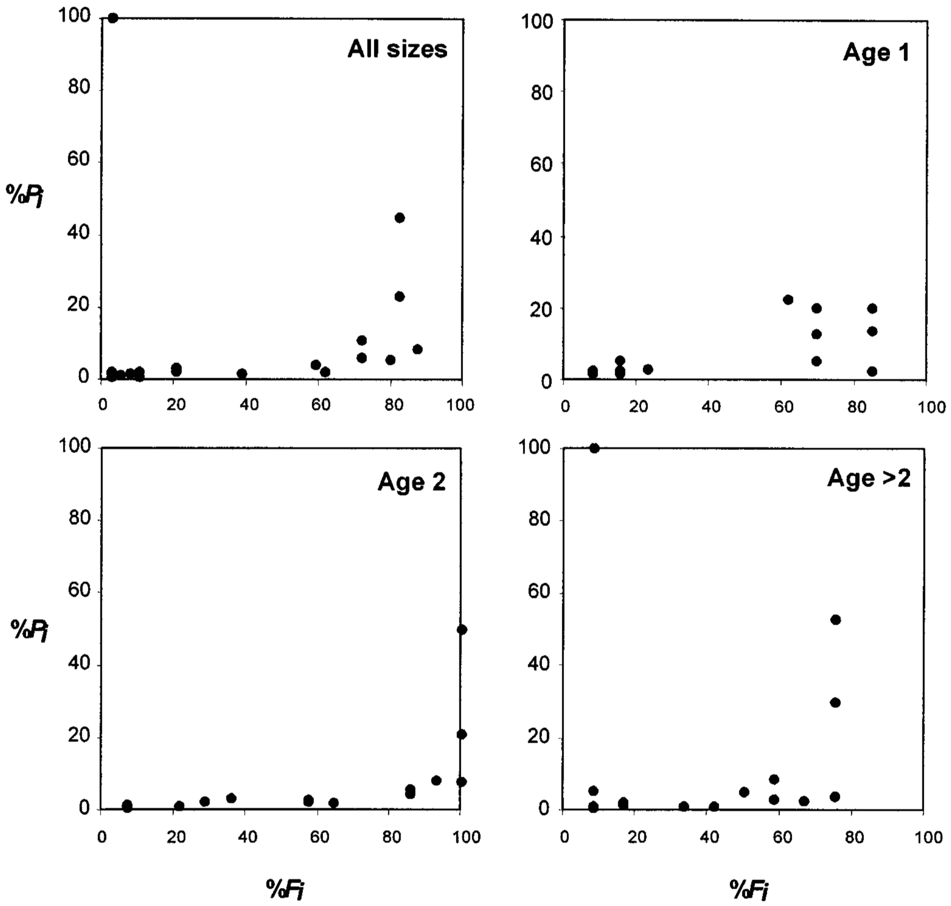

Fig. 3. Feeding strategy plots of the different rainbow trout age classes in the Urederra River (May 1995). Points represent different prey items.

both rainbow trout and other salmonids (Tippets & Moyle 1978, Ferriz 1994, Oscoz et al. 2000). In the Urederra River age 1 trout had a more benthic diet than older trout, probably because feeding at the bottom of the river implies less risk from predators than feeding at the surface (Vollestad & Andersen 1985, Haugen & Rygg 1996). In older trout there was a higher consumption of terrestrial invertebrates, indicating that they mainly feed on drifting invertebrates. Elliott (1967) suggested that these differences in diet composition between age classes could reduce trophic competition, somewhat facilitated by habitat segregation. This could be the explanation why smaller trout use prey not available to older trout (Jonsson & Gravem 1985). Likewise, Mollusca and fishes were only used by older trout because their bigger size requires a bigger mouth size, and a greater swimming ability in the case of fish predation (Easton & Orth 1992, Keeley & Grant 1997). This also allows older trout to have a bigger number of potential preys. Furthermore, the larger stomach size in older trout would allow ingesting a higher number of preys (Neveu & Thibault 1977).

Observed prey selection in analyzed rainbow trout would be related to prey availability and its energetic value (Vinyard 1980). Those prey easier to capture or with higher energetic value (such as Ephemeroptera, Diptera and Trichoptera) would be consumed more (Penczak et al. 1984), while those of lower energetic value, smaller or those that can camouflages themselves or hide under the substratum (e.g. Plecoptera) would be difficult to detect (Ware 1973, Rajasilta & Vuorinen 1983, Rincón & Lobón-Cerviá 1999), and as a result be consumed less.

Our results indicate that rainbow trout can affect autochthonous fish populations, like brown trout and Eu-

Table 2. Prey selection by rainbow trout in the Urederra River (May 1995). %Di: availability, %Ai: relative abundance, Wi: Savage index. ns: P>0.05; *: P<0.05; **: P<0.01. The significance levels were obtained applying Bonferroni's correction ((/number of categories).

	Benthos	Age 1			Age 2			Age >2		
	%D_i	**%A_i**	**W_i**		**%A_i**	**W_i**		**%A_i**	**W_i**	
Hidracarina	0.18	0.14	0.809	ns	-	0.000	ns	0.24	1.336	ns
Gammaridae	0.57	0.29	0.512	ns	0.12	0.211	ns	-	0.000	ns
Isopoda	-	-	-		-	-		0.12	-	
Lymnaeidae	-	-	-		-	-		8.00	-	
Baetidae	2.54	22.14	8.724	**	14.93	5.884	**	6.81	2.683	**
Heptageniidae	1.46	5.93	4.064	**	3.23	2.209	**	5.26	3.601	**
Leuctridae	40.52	0.14	0.004	**	-	0.000	**	-	0.000	**
Nemouridae	0.14	2.89	20.645	**	3.11	22.157	**	0.96	6.817	**
Perlodidae	-	-	-		0.12	-		-	-	
Hydroptilidae	-	0.43	-		0.12	-		0.12	-	
Limnephilidae	-	-	-		-	-		0.48	-	
Rhyacophilidae	0.20	0.72	3.564	*	1.91	9.415	**	0.84	4.119	**
Chironomidae	27.58	30.54	1.107	ns	48.39	1.755	**	59.62	2.162	**
Limoniidae	0.05	-	0.000	ns	0.36	6.740	**	0.12	2.247	ns
Simuliidae	5.26	16.21	3.079	**	10.99	2.088	**	4.90	0.930	ns
Stratiomyidae	0.05	0.87	16.328	**	1.31	24.713	**	0.48	8.987	**
Dytiscidae (A)	0.01	0.14	9.978	ns	0.12	8.238	ns	-	0.000	ns
Haliplidae (A)	-	-	-		0.12	-		-	-	
Nematoda	0.53	19.54	37.075	**	15.17	28.794	**	12.07	22.899	**
Other invertebrates	20.91	-	0.000	**	-	0.000	**	-	0.000	**

ropean minnow, because it is a predator of smaller specimens. In addition, previous works in the nearby Larraun River showed that European minnows and 0+ brown trout use the same prey items with a positive preference for Chironomids (Oscoz et al. 2000, 2001, Oscoz 2003). This competition could be the explanation for the low density of European minnow and 0+ brown trout observed in this reach by Oscoz et al. (1999). Nevertheless, a more precise study would be necessary in order to quantify the effect of rainbow trout on autochthonous fish populations.

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