

Response of larval and juvenile rudd *Scardinius erythrophthalmus* (L.) to different diets under controlled conditions

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ABSTRACT: The growth and survival of rudd *Scardinius erythrophthalmus* (L.) were evaluated in a laboratory at 25°C. In 20-day Experiment 1, first-feeding larvae at the age of 4 days post-hatch (initially: TL = 5.7 mm, BW = 0.9 mg) were fed live *Artemia* nauplii or commercial dry feed (Aller Futura Larvae, AFL) or combinations of both. Even the longest period of initial feeding of nauplii (6 days) was insufficient to obtain satisfactory larval growth after weaning to AFL (TL = 12.4 mm and BW = 17.7 mg vs. TL = 18.9 mm and BW = 68.5 mg for the nauplii-fed fish, significant differences). Nauplii-fed older larvae (24 days post-hatch) were then used in Experiment 2, in which they were fed AFL or Ewos AgloNorse (EAN) dry feeds for 40 days. The EAN diet proved to be significantly ($P \leq 0.05$) superior to AFL regarding the final fish growth (TL = 36.5 mm and BW = 506.8 mg vs. TL = 33.4 mm and BW = 392.0 mg), final survival rates (97.6% vs. 100%) and the incidence of spinal deformities (0% vs. 13.5%).

Keywords: rudd; larvae; juveniles; feeding; growth; survival; body deformities

The rudd, *Scardinius erythrophthalmus*, is a cyprinid fish species inhabiting either stagnant waters (littoral zone of lakes, dam reservoirs, ponds) or the lower reaches of slowly-flowing streams and rivers throughout Europe and western Asia (Załachowski, 2000). In Europe, this fish occurs commonly, but its populations rarely become abundant enough to dominate other fish species (e.g. Ravera and Jamet, 1991). Lelek (1987) designated *S. erythrophthalmus* as an endangered European freshwater fish species.

The first attempts at the controlled propagation and larval rearing of *S. erythrophthalmus* were probably performed in the mid-1960s in the Netherlands (Klein Breteler, 1979). Since then, considerable knowledge has been amassed regarding the reproductive biology and techniques to stimulate the reproduction of this species (e.g. Kucharczyk et al., 1997; Bonisławska and Winnicki,

2002; Kopiejewska et al., 2004). In contrast, much remains unknown about larval rearing under controlled conditions, and this might be a bottleneck in the mass production of juveniles for stocking natural waters.

The aim of the present study was: (1) to evaluate the growth and survival of *S. erythrophthalmus* larvae from the first feeding in response to a dry commercial diet used exclusively or after initial feeding with live food; (2) to assess growth, survival, and the incidence of body deformities in older larvae and early juveniles, in response to two dry commercial diets.

MATERIAL AND METHODS

Aged 4 days post-hatch the first-feeding *S. erythrophthalmus* larvae (the pooled offspring of 3 female

Table 1. Feeding schedule employed in Experiment 1 with first-feeding *S. erythrophthalmus* larvae

Experimental group	Feeding schedule
L20F0	days 1–20 – live <i>Artemia</i> nauplii
L6F14	days 1–6 – live <i>Artemia</i> nauplii days 7–20 – Aller Futura Larvae dry feed
L4F16	days 1–4 – live <i>Artemia</i> nauplii days 5–20 – Aller Futura Larvae dry feed
L2F18	days 1–2 – live <i>Artemia</i> nauplii days 3–20 – Aller Futura Larvae dry feed
L0F20	days 1–20 – Aller Futura Larvae dry feed

and 3 male spawners) of initial total length of 5.7 ± 0.2 mm (mean \pm SD) and mean wet body weight of 0.9 mg were used in Experiment 1. To stimulate ovulation in the females, mGnRH analogue Ovopel was used according to the standard propagation technique (Horváth et al., 1997).

A 20-day experiment was conducted in ten 10 dm³ glass flow-through aquaria with a density of 50 larvae per dm³. The experimental design comprised five feeding groups, all in duplicate (Table 1). The larvae were fed manually every 3 hours between 08:00 and 20:00. The live *Artemia* nauplii (EG grade, INVE Aquaculture B.V., Belgium) were fed in noticeable excess considered *ad libitum*. The daily feeding rates of the dry commercial diet for cyprinid fish (Aller Futura Larvae, Aller Aqua, Denmark; crude protein 64%, crude fat 12%, carbohydrates 5%, ash 11% as declared by the producer), always slightly excessive, were based on the visual assessment of larval appetite. The finest grade of the dry diet ("000") was used from the onset of the experiment until D13, and from D14 onward grade "00" was applied (particle size of 0.1–0.2 mm and 0.2–0.3 mm, respectively).

Experiment 2 was conducted on *S. erythrophthalmus* larvae aged 24 days after hatching with the initial total length of 18.9 ± 1.3 mm and body weight of 68.5 ± 15.9 mg (mean \pm SD). Prior to Experiment 2, the larvae were reared for 20 days exclusively on live *Artemia* nauplii in Experiment 1 (group L20F0, Table 1). The fish were stocked in four glass flow-through aquaria of 10 dm³ volume at a stocking density of 16.5 individuals per dm³. Two commercial dry diets, Aller Futura Larvae (grade

"0"; particle size 0.3–0.4 mm) and feed for marine fish larvae (AgloNorse Standard, Ewos, Norway; grade "1", particle size 0.4 mm; crude protein 59%, crude fat 20%, carbohydrates 5% and ash 12%, manufacturer's data) were evaluated in duplicate for 40 days. The feeding technique was the same as in Experiment 1. The daily rates of dry feeds were based on the visual assessment, and maintained at about satiation level.

In both experiments, the aquaria were continuously supplied with filtered, aerated water from the recirculation system. The temperature of 25°C (range $\pm 0.5^\circ\text{C}$), which is an optimum temperature for larvae and juveniles of most cyprinid fish species (Wolnicki, 2005), was kept in the aquaria throughout the experiments. The dissolved oxygen content in the aquaria was maintained at 70–90% of air saturation. Total ammonia and nitrites were kept at about 0.1 mg/dm³ and below 0.02 mg/dm³, respectively, and pH was 7.5–7.8. Temperature and oxygen were monitored systematically, whereas the remaining parameters twice a day. Fluorescent illumination of the aquaria (around 700 lx at the water surface) was provided between 08:00 and 21:00 (13L:11D). The aquaria bottom was cleaned of uneaten food and faeces twice a day. The dead fish were removed and counted during and after the cleaning of aquaria.

In Experiment 1, an initial random sample of 50 larvae was collected at the onset of day one (D1). At the end of D20, samples of 40 larvae were taken from each aquarium, and then all the remaining fish were counted. The final sample collected from group L20F0 was the initial sample for Experiment 2 ($n = 80$). Samples taken on the final day (D40) of Experiment 2 numbered 50 juvenile individuals per treatment. All of the fish sampled were preserved in 4% formalin.

In both experiments the individual total length (TL; measured to the nearest 0.01 mm) and individual wet body weight (BW; 0.1 mg) of the fish were determined in all samples (except the initial sample in Experiment 1, when mean BW was determined on the basis of the total fish biomass). The final survival rates and percentages of deformed individuals were determined as well. Duncan's multiple range test was used to compare the mean values of TL and BW. Survival percentages and percentage shares of deformed individuals were normalized using angular transformation, according to Sokal and Rohlf (1969). The level of significance was set at $P \leq 0.05$.

Table 2. Initial and final characteristics of *S. erythrophthalmus* fed live food or dry feed or their combinations for 20 days in Experiment 1

Parameter	Experimental group				
	L20F0	L6F14	L4F16	L2F18	L0F20
Initial total length (mm ± SD)			5.7 ± 0.2		
Final total length (mm ± SD)	18.9 ± 1.3 ^a	12.4 ± 1.1 ^b	10.9 ± 0.9 ^c	9.9 ± 0.8 ^d	9.0 ± 0.6 ^e
Initial body weight (mg)			0.9		
Final body weight (mg ± SD)	68.5 ± 15.9 ^a	17.7 ± 5.7 ^b	10.8 ± 3.2 ^c	8.2 ± 2.1 ^d	5.7 ± 1.6 ^e
Final survival rate (%)	98.7 ^a	44.8 ^d	94.5 ^c	96.7 ^b	96.3 ^{bc}

data for fish size are mean ± SD except the initial BW (only mean), initial $n = 50$, final $n = 80$; for the survival rates, initial $n = 1\ 000$; within rows, data with different superscripts are significantly different at $P \leq 0.05$

RESULTS

In Experiment 1, all the differences in the final *S. erythrophthalmus* mean length and weight were significant (Table 2). The fastest growth was noted in the larvae fed exclusively live food (group L20F0; TL = 18.9 mm, BW = 68.5 mg). The final survival rate recorded for these larvae (98.7%) was also significantly the highest. Larvae fed exclusively dry feed (group L0F20) grew the slowest (TL = 9.0 mm, BW = 5.7 mg). Fish fed the combinations of live food and dry feed grew faster when the initial feeding period with the live diet was longer. In group L6F14, there were massive losses between D11 and D20 resulting in the lowest final survival rate of 44.8%.

During the first 10 days of Experiment 2, all the fish completed the larval period and became juveniles, which was indicated by the scale cover.

In Experiment 2, fish fed Ewos AgloNorse grew significantly better than those fed Aller Futura Larvae diet (final TL = 36.5 mm v. TL = 33.4 mm, respectively), significantly higher was also their final survival rate (100% v. 97.6%, respectively) (Table 3). The latter diet also resulted in a significantly higher (13.5% vs. 0.0%) incidence of body deformities (spinal curvature).

DISCUSSION

In most cyprinid fish species the larval ability to utilize dry formulated diets from the first feeding is low (Wolnicki, 2005). For example, satisfactory growth performance and/or survival were not achieved when exclusively dry diets were fed to larval asp, *Aspius aspius* (Wolnicki, 2005), gudgeon,

Table 3. Initial and final characteristics of *S. erythrophthalmus* fed Ewos AgloNorse (EAN) or Aller Futura Larvae (AFL) dry feeds for 40 days in Experiment 2

Parameter	Experimental group	
	EAN	AFL
Initial total length (mm ± SD)		18.9 ± 1.3
Final total length (mm ± SD)	36.5 ± 3.0 ^a	33.4 ± 2.7 ^b
Initial body weight (mg ± SD)		68.5 ± 15.9
Final body weight (mg ± SD)	506.8 ± 135.9 ^a	392.0 ± 95.8 ^b
Final survival rate (%)	100.0 ^a	97.6 ^b
Final share of deformed fish (%)	0.0 ^b	13.5 ^a

data for fish size are mean ± SD, initial $n = 80$ and final $n = 50$; for the survival rates, $n = 330$; for the share of deformed individuals, $n = 50$; within rows, data with different superscripts are significantly different at $P \leq 0.05$

Table 4. Relative growth rates for wet body weight (*RGR*, %/day) and survival rates (*S*, %) of larval cyprinids fed exclusively live *Artemia* nauplii from the first feeding at 25°C. Fish density 40–60 indiv/dm³; duration of feeding 20–21 days; data shown in decreasing order of *RGR*

Species	Initial BW (mg)	Final BW (mg)	<i>RGR</i> (%/day)	<i>S</i> (%)	Source
<i>Cyprinus carpio</i>	1.2	95.1	24.4	95.0	Wolnicki (2005)
<i>Leuciscus cephalus</i>	1.4	108.4	24.3	93.0	Wolnicki and Myszkowski (1999a)
<i>Scardinius erythrophthalmus</i>	0.9	68.5	23.9	98.7	Present paper
<i>Vimba vimba</i>	2.3	110.9	21.4	99.2	Wolnicki (1996a)
<i>Aspius aspius</i>	2.6	118.1	21.1	99.0	Wolnicki and Myszkowski (1999b)
<i>Chondrostoma nasus</i>	6.3	168.0	17.8	98.0	Wolnicki and Myszkowski (1998)
<i>Leuciscus leuciscus</i>	3.0	81.2	17.0	99.9	Kujawa (2004)
<i>Leuciscus idus</i>	3.0	78.4	16.8	98.6	Kujawa (2004)
<i>Barbus barbus</i>	9.5	121.1	12.9	99.3	Kujawa (2004)

relative growth rates (Myszkowski, 1997) were calculated according to the formula: $RGR = 100 (e^G - 1)$ where $G = (\ln BW_f - \ln BW_i) n^{-1}$; BW_i and BW_f = the initial and final body weight, respectively; n = the number of feeding days

Gobio gobio (Awaïss et al., 1992), chub, *Leuciscus cephalus* (Wolnicki and Myszkowski, 1999b; Shiri Harzevili et al., 2003), ide, *L. idus* (Shiri Harzevili et al., 2004; Hamáčková et al., 2007), dace, *L. leuciscus* (Lepičova et al., 2002; Shiri Harzevili et al., 2005), European minnow, *Phoxinus phoxinus* (Stalmans and Kestemont, 1991; Kestemont and Stalmans, 1992) or tench, *Tinca tinca* (Wolnicki and Górny, 1995b; Wolnicki, 2005). Considerably improved rearing results were attained when the first-feeding larvae were initially fed natural food before being

weaned to dry formulated feeds (Wolnicki, 2005). The supplementation of dry diets with natural food turned out to be another similarly effective solution (Wolnicki and Korwin-Kossakowski, 1993; Wolnicki and Górny, 1995a).

In contrast to all the aforementioned species, only a few cyprinids demonstrate relatively fast growth and very good survival when fed dry feeds exclusively from the very beginning of exogenous feeding: barbel, *Barbus barbus* (Wolnicki and Górny, 1995c; Fiala and Spurný, 2001; Policar et

Table 5. Relative growth rates for wet body weight (*RGR*, %/day) and survival rates (*S*, %) of larval cyprinids fed exclusively dry formulated feeds from the first feeding at 25°C. Fish density 40–60 indiv/dm³; duration of feeding 15–20 days; data shown in decreasing order of *RGR*

Species	Initial BW (mg)	Final BW (mg)	<i>RGR</i> (%/day)	<i>S</i> (%)	Source
<i>Chondrostoma nasus</i>	6.3	100.0	14.8	98.0	Wolnicki and Myszkowski (1998)
<i>Vimba vimba</i>	2.3	20.9	11.7	99.1	Wolnicki (1996a)
<i>Leuciscus cephalus</i>	1.3	8.5	9.8	13.3	Wolnicki (2005)
<i>Scardinius erythrophthalmus</i>	0.9	5.7	9.7	96.3	Present paper
<i>Tinca tinca</i>	0.7	4.4	9.6	82.4	Wolnicki et al. (unpubl. data)
<i>Leuciscus idus</i>	3.0	10.0	8.4	41.0	Wolnicki and Górny (1995a)
<i>Barbus barbus</i>	12.0	36.0	7.6	99.0	Wolnicki and Górny (1995c)

RGR calculations see Table 4

Table 6. Relative growth rates for wet body weight (*RGR*, %/day) of juvenile cyprinids fed exclusively dry formulated feeds at 25°C. Fish density 4.0–16.5 indiv/dm³; duration of feeding 20–40 days; data shown in decreasing order of *RGR*

Species	Initial BW (mg)	Final BW (mg)	<i>RGR</i> (%/day)	Source
<i>Barbus barbus</i>	89.6	510.0	6.0	Wolnicki (1997)
<i>Leuciscus idus</i>	58.0	182.3	5.9	Wolnicki (1996b)
<i>Scardinius erythrophthalmus</i>	68.5	506.8	5.1 ¹ 4.5 ²	Present paper
<i>Vimba vimba</i>	110.9	467.3	4.9	Wolnicki (1996a)
<i>Chondrostoma nasus</i>	168.0	573.0	4.2	Wolnicki and Myszkowski (1999a)

¹on Ewos AgloNorse; ²on Aller Futura Larvae
RGR calculations see Table 4

al., 2007), goldfish, *Carassius auratus* (Abi-Ayad and Kestemont, 1994; Kaiser et al., 2003), nase, *Chondrostoma nasus* (Wolnicki and Myszkowski, 1998; Spurný et al., 2004), roach, *Rutilus rutilus* (Köck and Hofer, 1989) and vimba, *Vimba vimba* (Wolnicki, 1996a).

In the present study, when *S. erythrophthalmus* larvae were fed live food from the onset of exogenous feeding, they were among the fastest growing cyprinids, with a relative growth rate in wet weight of 23.9%/day (Table 4). On the other hand, when fed dry formulated feed exclusively, the larvae of this species grew very slowly in comparison with the other cyprinids in the same period of life (Table 5). However, although larvae in Experiment 1 grew poorly on the dry diets, their final survival rates were high in most of the experimental groups. Mass larval losses recorded for group L6F14 (Table 2), as the only exception, were difficult to explain.

The results of the present study suggest that *S. erythrophthalmus* larvae may require a relatively long period of the initial feeding of live food to improve their ability to utilize Aller Futura Larvae for growth. The major reason may be that this dry diet does not meet all the nutritional needs of the larvae. Larval cyprinids most often need about 5 days of the initial feeding of natural food, but in the case of some species and some dry diets, not less than 8–12 days are necessary (Kujawa, 2004; Wolnicki, 2005). As indicated by the current results, 6 days of the initial feeding of *Artemia* nauplii (Experiment 1) may be insufficient. On the other hand, a 20-day period of feeding live food, preceding Experiment 2, proved to ensure growth rates on both dry feeds evaluated in the present work that

were comparable to those noted for the cyprinids that utilized dry diets most effectively, such as *B. barbus* or *L. idus* (Table 6).

In conclusion, the fast growth, maximum survival, and the lack of body deformities in older larvae and early juveniles fed Ewos AgloNorse in Experiment 2 prove the usefulness of this diet for *S. erythrophthalmus*, and most likely for other cyprinid species as well. This finding is noteworthy because this diet (1) is for marine fish species, (2) contains as much as 20% fat. According to many experimental data (Wolnicki, 2005; Kamler and Wolnicki, 2006), feeding to young cyprinids high fat diets, i.e. containing more than 10%, usually results in a gradual increase of fat content in the fish body, a decrease in body minerals, and finally disruption in the process of bone mineralization, resulting in further skeletal malformations.

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