

Rearing of Burbot, *Lota lota* (Pisces, Teleostei), Larvae with Zooplankton and Formulated Microdiets

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Abstract

Different feeding methods were tested for burbot, *Lota lota*, larvae. In small scale experiments with 300 larvae per treatment grinded artemia flakes, enriched artemia flakes, artemia flakes supplemented with dried algae (*Chlorella* sp., *Spirulina* sp.) and formulated microdiets consisting of different combinations of fishmeal, fish oil, soybean lecithin, casein, dextran, and artemia flakes were fed over a period of 30 days. These foods were compared to live zooplankton food collected from the nature. After 30 days, only feeding with live zooplankton resulted in high survival rates > 80%. No survival was observed with artificial microdiets. The 15 d survival of larvae was significantly lowest with agar agar bound microparticles and with formulated diets containing $\geq 7\%$ soya lecithin and $\geq 3\%$ fish oil. The live zooplankton feeding method was also tested in a large scale experiment with 25,000 larvae per tank for a period of 100 d. After 100 d the larvae survival rate was > 65 %, and the body length had increased for circa 6-fold.

Keywords: larvae, *Lota lota*, nutrition, zooplankton, formulated diets

1. Introduction

In nature newly hatched fish larvae feed on zooplankton (Poulet & Williams, 1991; Hsieh & Chiu, 2002). For intensive culture conditions zooplankton is generally not available and live food (mainly *Brachionus calyciflorus* and *Artemia* sp.) is used which is produced under controlled conditions (Lubzens, Tandler, & Minkoff, 1989; Policar et al., 2007). Alternatively larvae of fresh water fish are reared under semi-extensive or extensive conditions in natural ponds where they feed on naturally growing zooplankton sources (Ludwig, 1999; Milstein, Valdenberg, & Harpaz, 2006). Larvae are also reared in net cages which are placed in plankton containing water systems (Žiliukienė, 2005; Žiliukienė & Ziliukas, 2006; Paragamian, Laude, Cain, Barron, & Jensen, 2011). Formulated microdiets were tested with variable success as starter food for larvae of white sturgeon (*Acipenser transmontanus*) (Gawlicka, McLaughlin, Hung, & de la Noüe, 1996), grass carp (*Ctenopharygodon idella*) and bighead carp (*Aristichthys nobilis*) (Rottmann, Shireman, & Lincoln, 1991), zebrafish (*Danio rerio*) (Carvalho, Araújo, & Santos, 2006), pike-perch (*Sander lucioperca*) (Ostaszewska, Dabrowski, Czuminiska, Olech, & Olejniczak, 2005), and loach (*Misgurnus anguillicaudatus*) (Wang, Hu, Cao, Yang, & Wang, 2008). Combination of live food and formulated, commercially available diets were successfully used for feeding the larvae of goldfish (Kaiser, Endemann, & Paulet, 2003), chub (*Leuciscus cephalus*), ide (*Leuciscus idus*), orfe (*Leuciscus idus*) and dace (*Leuciscus leuciscus*) (Kwiatkowski et al., 2008) and of vimba bream (*Vimba vimba*) (Hamáčková et al., 2009).

The culture of the burbot, *Lota lota* has high importance as it is endangered or extirpated in many European regions (Van Houdt, Hellemans, & Volckaert, 2003; Van Houdt, de Cleyn, Perretti & Volckaert, 2005). However, the larvae are difficult to rear as they are one of the smallest in fresh water. Only few data are available on this topic. Shiri Harzevili et al. (2003) reared *Lota lota* larvae with *Brachionus calyciflorus* and *Artemia* sp. with a survival rate of circa 70%. Paragamian et al. (2011) reared them in pond cages resulting in an average survival rate of 18 %.

In the present study we investigated alternative feeding methods for *Lota lota* larvae. We tested dried *Artemia* sp., *Chlorella* sp. and *Spirulina* sp., as this food is easy to prepare and to handle. We tested also formulated microdiets which composition bases on recipes of Carvalho et al. (1997), of Carvalho, Araújo, & Santos (2006), and of Robin & Vincent (2003). These feeding regimes were compared with live zooplankton feeding which was harvested from a natural lake population.

2. Method

2.1 Broodstock Fish and Egg Collection

Lota lota derived from a wild population from lake Mondsee (47°49'N, 13°24'E). The fish were kept in the fish farm Kreuzstein in flow through concrete tanks with a water supply of 2 - 3 l s⁻¹ and under a natural photoperiod. They were fed with dead rainbow trout fingerlings at a rate of circa 10% of their body weight every second day. Spawning was initiated by a decrease in temperature from 5°C to 1.5°C in the beginning of February. For spontaneous spawning 15 males and 15 females were transferred into a spawning tank (1.5 x 2 x 0.5 m – length x width x height) with a false bottom consisting of a stainless steel mesh to prevent egg cannibalism 3 weeks before the expected date of spawning. Eggs were collected and incubated in Zug charrs at a temperature of 1.7 ± 1.2°C until the stage of ready to hatch embryos. To synchronize and shorten the hatching period the temperature was raised to 9°C. Larvae were drained out of the Zug charrs using PVC tubes and stocked in rectangular tanks with a volume of 200 l. The tanks had initially a water height of circa 10 cm to facilitate the up-swimming of larvae and the inflation of the swimbladder. Five days after hatching was finished, the tanks were gradually filled to 200 l.

2.2 Larvae Feeding Experiments

The feeding experiments are shown in the following flow chart. Feeding was started 10 days after hatching (dph).

SMALL SCALE EXPERIMENTS	
Set up: 300 larvae/experiment, each experiment in duplicate, 4 feedings per day, feeding period 30 d; density: 60 larvae/l. Quantities of food ingredients are reported in parenthesis in % (w : w)	
EXPERIMENT 1: <i>Artemia</i> sp. and algae (300 mg/ feeding)	(1) grinded artemia flakes (2) grinded artemia flakes enriched with 0.5% (w/w) Selco presso (3) grinded artemia flakes (75), <i>Chlorella</i> (25) (4) grinded artemia flakes (75), <i>Spirulina</i> (25) (5) live zooplankton (5000 animals per feeding)
EXPERIMENT 2: Formulated diets I (300 mg/ feeding)	(a) fish meal (76), soya lecithin (13), fish oil (5) ¹ (b) fish meal (76), soya lecithin (13), fish oil (5) in agar agar matrix ¹ (c) fish meal (84), soya lecithin (7), fish oil (3) in agar agar matrix ¹ (d) fish meal (91), soya lecithin (2), fish oil (1) in agar agar matrix ¹ (e) live zooplankton (5000 animals per feeding)
EXPERIMENT 3: Formulated diets II (based on results of experiment 2) (300 mg/ feeding)	(I) fish meal (91), soya lecithin (2), fish oil (1) ¹ (II) fish meal (56), casein (35), soya lecithin (2), fish oil (1) ¹ (III) fish meal (75), dextrin (18), soya lecithin (0.8), fish oil (0.2) ¹ (IV) fish meal (56), artemia flakes (35), soya lecithin (2), fish oil (1) ¹ (V) live zooplankton (5000 animals per feeding)
↓	
LARGE SCALE EXPERIMENT	
Set up: Best feeding regime from small scale experiments, food type: natural zooplankton, 25.000 larvae per tank, experiment in duplicate, feeding period 100 d	
EXPERIMENT 4 (Natural zooplankton)	10 – 30 days post hatch (dph): size class 100 – 200 µm, 400 ± 50 animals per feeding, 4 feedings per day 31– 50 dph: size class: 200 – 400 µm, 450 ± 50 animals per feeding, 7 feedings per day 51– 100 dph: size class: > 400 µm, 360 ± 40 animals per feeding

¹contains 4% vitamin mix and 2% mineral mix

The small scale experiments were performed in modified Zug char glasses (Figure 1) at a flow through rate of 10 ml/min and at natural photoperiod. Three-hundred larvae were stocked in each glass. Feeding was performed at 8, 11, 15, and 19 h using 300 mg of formulated microdiet food or 50 ml zooplankton (animal density = 100,000/l).

Zug charrs were cleaned daily from food remnants and dead larvae. Larvae survival was counted after 15 and 30 d. After 15 days 20 larvae were sampled from each feeding regime for morphometric investigations (see viability determinations).

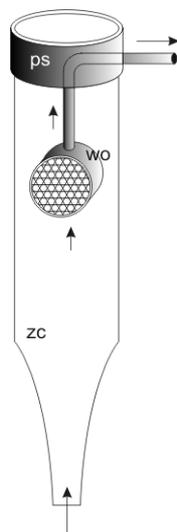


Figure 1. Modified Zug charr (zc) for on-feeding of *Lota lota* larvae. Water outlet (wo) occurred via a 10 cm plastic ring covered with a 200 µm mesh size plankton net to avoid settling of larvae in the drainage. ps – plastic sleeve for fixation of the water outlet. Arrows indicate direction of water flow

In the large scale experiments circular tanks with a diameter of 90 cm and a height of 45 cm were used for 10 – 30 dph larvae. Water inlet and outlet were peripherally at opposite sides of the tanks, the inlet at the bottom and the drainage at the surface. Settling of larvae at the drainage (as it is the case for circular tanks with a central bottom outlet) was avoided by this construction and zooplankton kept floating in the water body. Larvae were fed 3 times per day at 9, 13, and 18 h. After 30 dph the larvae were transferred into 5 m³ rectangular tanks (5 x 1.6 x 0.6 m, length x width x height) having the water inlet at one broadside and the water outlet at the opposite one. For larvae > 30 dph feeding was performed 7 times per day from 8 - 18 h in 1.5 h intervals. The experiment was terminated 100 dph. Details of the feeding regimes are shown in Table 1. The feeding experiment was made in duplicate in separate tanks, each with 25,000 larvae (determination of larvae numbers see below). For morphometric measurements and for control on malformations 50 larvae, respectively, were sampled from each tank at 0, 20, 30, 50, and 100 dph.

Table 1. Tank and feeding parameters used in the large scale experiment. *weight of zooplankton separated from water

	10 – 30 dph	31– 50 dph	51– 100 dph
Tank parameters			
Tank volume (l)	200	5000	5000
Stocking density (larvae/l)	125 ± 5	4 ± 1	3.5 ± 0.5
Flow through rate (l/min)	0.6 l/min	15 l/min	15 l/min
Feeding parameters			
Zooplankton fraction (µm)	fine (100–200)	medium (200–400)	coarse (> 400)
Feedings per day	3	7	7
Zooplankton in g per day*	30	175	560
Zooplankton in organisms per day (x 10 ³)	1200 ± 150	3150 ± 350	2520 ± 390
Organisms per feeding per larvae	16 ± 2	23 ± 2.5	21 ± 8
Organisms per feeding per liter water	2500 ± 310	100 ± 10	75 ± 8

2.3 Preparation of Formulated Microdiets

Artemia flakes, dried *Chlorella* and *Spirulina*, and Selco presso enrichment suspension (according to manufacturer instructions it contains high concentrations of docosahexaenoic acid and eicosapentaenoic acid, minerals, trace elements, and vitamins) were obtained from www.artemia-shop.de/. Fish meal, casein, and fish oil were obtained from a local angler shop, agar agar, soya lecithin, dextrin and the vitamin mix from a local pharmacy.

For experiment 1 *Artemia* flakes were grinded with a com mill to a particle size of 100 – 120 µm and mixed with *Chlorella* or *Spirulina* powder as indicated in the flow chart. For enrichment, artemia flakes were soaked with a 0.5% suspension of Selco presso and dried in an incubator at 70°C. Thereafter, the flakes were grinded as described above.

The formulated diets used in experiments 2 and 3 were prepared as follows: All ingredients were grinded to a particle size < 30 µm in a ball mill. The proteinous ingredients indicated in the flow chart were mixed with water to form a dough and heated to 100°C for circa 15 min under constant stirring. Thereafter, the dough was cooled to circa 70°C. Fish oil, soya lecithin (in form of a 5 % solution), and vitamin mix were added in the required amounts and the components were mixed for additional 5 min. This nutrient mixture was processed in two ways. For experiment 2 the ingredients were bound in agar agar matrix. A 2% agar agar solution was added to the 70°C warm nutrient mixture in a ratio of 1 : 1 (w : w) and mixed for 10 min. Then the mixture was spread on a tin foil in a 1 – 3 mm thick layer and dried in an incubator at 70°C for 24 h. For experiment 3 the nutrient mixture was dried without matrix embedding. Finally, the dry microdiets were grinded in a corn mill to a particle size of 100 -150 µm and stored air tight in a refrigerator at 4- 5°C until use. Four % vitamin mix (w/w) and 2 % mineral mix (w/w) was added to all formulated diets described in experiment 2 and 3. The vitamin mix contained per kg 20 g ascorbic acid, 1 g biotine, 200 g choline chloride, 0.5 g cholecalciferol, 1 g cyanocobalamin, 2 g D-calcium pantothenate, 0.1 g folic acid, 1 g menadione, 30 g meso-inositol, 1 g niacin, 0.3 g pyridoxin, 0.34 g retinyl acetate, 400 mg riboflavine, 100 mg thiamin, 10 g α-tocopherol. The mineral mix contained per kg 215 g CaCO₃, 500 g CaHPO₄·2H₂O, 20 mg CoSO₄·7H₂O, 3 g CuSO₄·5H₂O, 20 g FeSO₄·7H₂O, 90 g KCl, 40 mg KL₄O, 124 g MgSO₄·7H₂O, 3 g MnSO₄·H₂O, 40 g NaCl, 1 g NaF, 4 g ZnSO₄·7H₂O

2.4 Collection of Live Zooplankton

Live zooplankton was collected from wild populations from lake Mondsee with plankton nets (AquaTech, Kitzbühl, Austria). The nets were dredged floating behind a boat in a depth of 10 – 15 m and at a cruising speed of 0.5 miles/h. The dredging depth was adjusted depending on weather conditions and season. Using a sieve netting procedure three size fractions were collected. Sieve nets with 100 µm as lower and 200 µm as upper mesh size limit were used to collect a “fine” zooplankton fraction for feeding the 10 – 30 dph old fish. The fine zooplankton fraction contained organisms with a width of 195 ± 30 µm and a length of 610 ± 110 µm (measured in subsamples fixed in 4 % formaldehyde, n = 250 from 10 different sampling dates).

Nets with a lower mesh size of 200 µm and an upper mesh size of 400 µm were used to collect a “medium” sized zooplankton fraction for the 31 – 50 dph fish. These organisms had a body width of 305 ± 55 µm and a body length of 940 ± 110 µm (n = 375 from 15 sampling dates). A plankton net with a mesh size of 400 µm was used to collect a coarse zooplankton fraction for > 50 dph fish. Organisms in this fraction had a width of 470 ± 260 µm and a length of 1190 ± 420 µm (n = 400 from 16 different sampling dates).

The quantities of collected zooplankton depended on the mesh size of the nets. With the 100 – 200 µm sieve nets 0.08 – 0.12 kg zooplankton were collected per hour (weight of zooplankton separated from water). This represents an amount of (3.2 – 4.8) x 10⁶ animals (number of animals counted in a 10 mg subsample and extrapolated with the total zooplankton mass). With the 200 – 400 µm sieve net 0.2 – 0.5 kg zooplankton (= [3.6 – 9.0] x 10⁶ animals) were collected per hour, and with the 400 µm net 1.7 – 1.9 kg (= [10.2 – 11.4] x 10⁶ animals).

The collected zooplankton was washed out of the nets in 20 l buckets and diluted to a density of 100.000 animals per litre. Preliminary adjustment was made visually by optical density estimations and exact adjustment by counting the animal numbers in 1000 µl subsamples in Petri-dishes. Subsamples of the zooplankton fractions were fixed in 4% formaldehyde in 2 day intervals and the species composition and development stages were determined.

2.5 Larvae Viability Evaluations

The number of viable fish was determined 10 dph (= initial stocking rate before the onset of feeding), 30 dph, and 100 dph. Ten dph and 30 dph the total number of larvae was determined based on density determinations. Larvae were homogenously distributed in the tanks by gentle mixing and four 1 l samples were taken. Larvae numbers per liter were counted and extrapolated to the total water volume of the tank. The numbers of juvenile fish was determined gravimetrically. The fish were transferred out of the tank and weighted. Thereafter the individual

weight of 20 fish was determined and the total number of fish was calculated as total fish weight divided through the average individual fish weight. The 30 and 100 dph survival rate was calculated as the percentage value of fish in relation to the number of stocked fish.

For the morphometric investigations the larvae were killed by an overdose of MS222 and photographed. in a stereomicroscope at 2 to 5-fold magnification depending on the fish size. In the small scale experiment the larvae total length was measured, in the large scale experiment the larvae total length, head width and body width.

2.6 Statistics

The survival rate of fish was expressed as percentage of surviving fish relative to the total number of fish and reported as mean \pm standard deviation. Morphometric data are presented as mean \pm standard deviation, too. For statistical analysis, percentage data were transformed by angular transformation ($\arcsin\sqrt{P}$) and morphometric data by a logarithmic transformation. Survival rates were corrected for larvae numbers sampled for the morphometric determination. To determine if the experimental treatments resulted in significantly different survival rates, analysis of variance (ANOVA) with Waller Duncan posthoc test for the percentage data and Tukey-B posthoc test for the morphometric data was used.

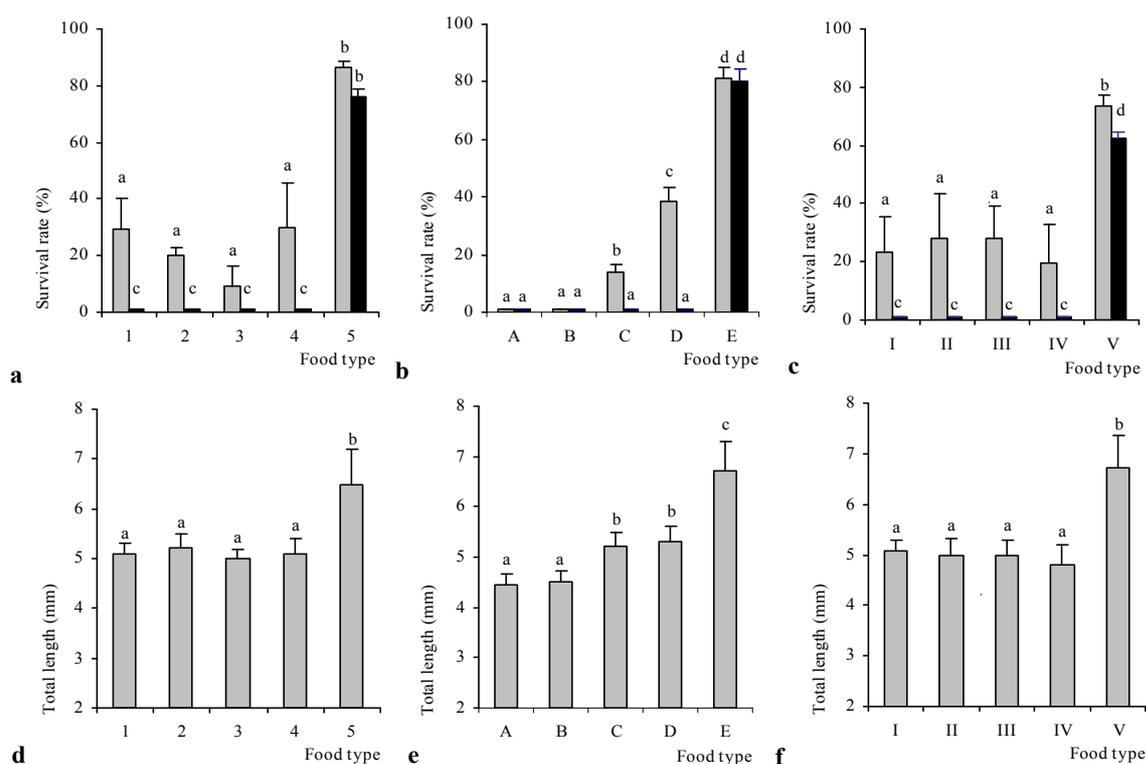


Figure 2. Survival and total length of *Lota lota* larvae after feeding with formulated diets and live zooplankton. Data are mean \pm S.D., those with different superscripts are significantly different ($P < 0.05$). For survival rates sample number (n) = 2 (2 tanks each with 300 larvae) and for total length n = 40 (20 larvae from each tank).

Quantities of food ingredients are expressed in % weight

(a) Survival rates and (d) total length obtained in experiment I: 1: grinded *Artemia* flakes (100), 2: grinded *Artemia* flakes (75) & *Chlorella* (25), 3: grinded *Artemia* flakes (75) & *Spirulina* (25), 4: enriched *Artemia* flakes (100), 5: live zooplankton.

(b) Survival rates and (e) total length obtained in experiment II: A: Fish meal (76), soya lecithin (13), fish oil (5) B: similar to (1) and in agar agar matrix, C: fish meal (84), soya lecithin (7), fish oil (3) in agar agar matrix, D: Fish meal (91), soya lecithin (2), fish oil (1) in agar agar matrix, E: live zooplankton

(c) Survival rates and (f) total length obtained in experiment III: Food types: I: Fish meal (91), soya lecithin (2), fish oil (1), II: fish meal (56), artemia flakes (35), soya lecithin (2), fish oil (1), III: fish meal (56), casein (35), soya lecithin (2), fish oil (1), IV: fish meal (75), dextrin (18), soya lecithin (0.8), fish oil (0.2) V: live zooplankton.

3. Results

3.1 Small Scale Experiments with Artificial Microdiets and Live Zooplankton

In all 3 experiments the survival rates and the growth (total length) were high with live zooplankton (Figures 2a,b, c). With artificial microdiets in some treatments a percentage of up to 40% of the larvae survived for circa 15 days. Thereafter all larvae died.

In experiment 1 there were no differences in 15 d survival rates and in total length of larvae after 15 d (Figure 2d) between grinded artemia flakes (food type 1), enriched artemia flakes (type 4), grinded artemia flakes mixed with *Chlorella* sp. (type 2), and grinded artemia flakes mixed with *Spirulina* sp. (type 3) (Figure 2a).

In experiment 2 the 15 d survival of larvae and the total length of larvae after 15 d was significantly lower with agar agar bound microparticles than with microparticles having no agar agar matrix (compare food type A with type B) (Figures 2b, 2e). Also microparticle food with $\geq 7\%$ soya lecithin and $\geq 3\%$ fish oil (types B and C) resulted in lower larvae survival and in lower larvae total length than microparticle food with only 2 % soya lecithin and 1 % fish oil (type D) (Figures 2b, 2e).

Experiment 3 showed that the supplementation of the microparticle food (food type I) with additional protein sources as grinded artemia flakes (type II) or caseine (type III) had no effect on the larvae survival and on the larvae total length after 15 d (Figures 2c, 2f). Also the supplementation of food type I with carbohydrates in form of dextran (type IV) did not affect the larvae survival rates and the larvae total length after 15 d (Figures 2c, 2f).

3.2 Large Scale Experiment with Live Zooplankton

The composition of the zooplankton fractions in aspects of species composition and development stages is shown in Figure 3. The 30 dph survival rate of *Lota lota* was 83 % for tank 1 and 71 % for tank 2 (Figure 4a). The 100 dph survival rate was 78 % and 66 %, respectively (Figure 2a). The growth rates did not significantly differ between tank 1 and tank 2 and therefore the pooled data set is presented. During the first 100 days of feeding the body length of *Lota lota* increased for circa 6-fold from 4.6 to 28.0 mm (Figure 4b), the body width for circa 5.5-fold from 0.8 to 3.1 mm, and the mouth width for almost 13-fold from 0.24 to 3.10 mm (Figure 4c). The percentage of larvae malformations was $< 2\%$.

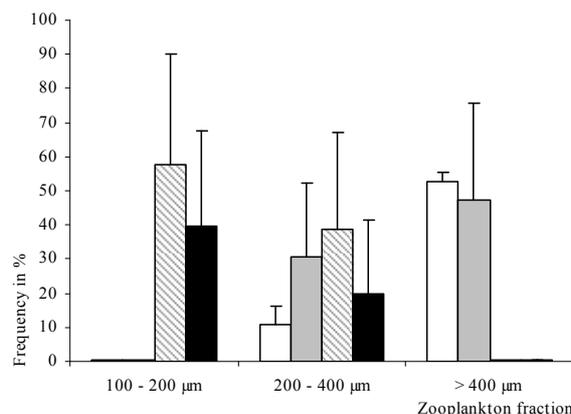


Figure 3. Species composition of the zooplankton fractions used for *Lota lota* feeding. White bars: adult cladocera (*Daphnia* sp.), grey bars: adult copepods (*Cyclops* sp. and *Diaptomus* sp.), shaded bars: copepodites, black bars: nauplii. Data are mean \pm standard deviation. N=20 for the $<200\ \mu\text{m}$ fraction (sampling period: 10–30 dph), n=15 for the 200–400 μm fraction (sampling period: 31–50 dph), and 16 for the total fraction (sampling period 51–100 dph)

4. Discussion

The present study shows that not any of the tested artificial diets was successful for first feeding of *Lota lota* larvae. Only feeding with live zooplankton resulted in high survival rates and growth rates. The food ingredients used in experiment 1 were artemia flakes and artemia flakes in combination with dried algae. Although these ingredients derived from organisms which are generally used as larvae food for *Lota lota* and for other fish species (Lubzens et al., 1989; Paragamian et al., 2011), they were not suitable for *Lota lota* larvae in the dried form. This

could have the following reasons: (a) Drying and processing could have decreased the nutritional value. (b) Food particles might be unstable in water and dissolve quickly and (c) food particles might not be ingested by the larvae. The latter two hypotheses are unlikely as food particles were observed sinking down from the water surface for up to 30 min and as ingestion of the food particles by larvae could be observed (unpublished data).

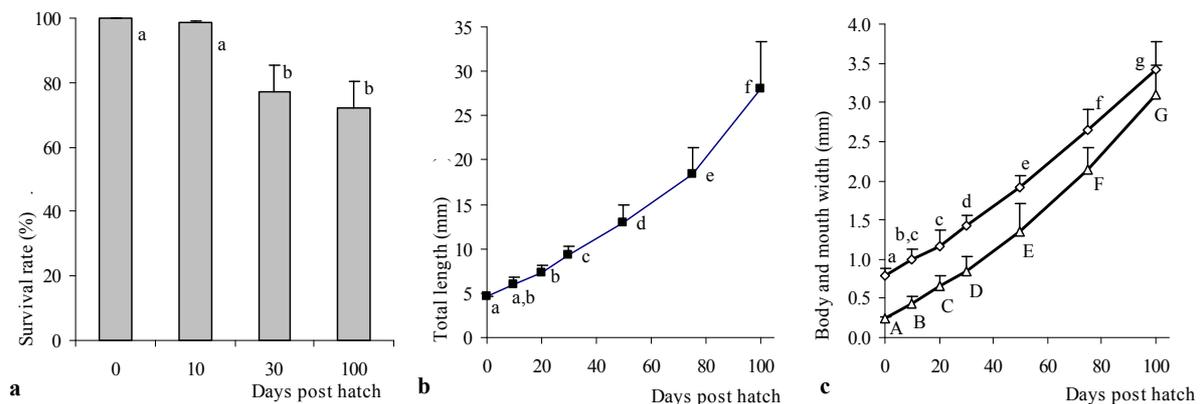


Figure 4. (a) Survival rates, (b) total length, (c) body width (rhombes) and mouth width (triangles) of *Lota lota* larvae during on-feeding with live zooplankton. Data are mean \pm S.D. In (a) $n = 2$ for 2 tanks, in (b) and (c) data from the 2 tanks were pooled as there existed no significant differences ($n = 100$)

The formulated diets used in experiments 2 and 3 contained 62 – 97 % fishmeal, 2 – 13 % soy bean lecithin, and 0.1- 5 % fish oil. Therefore, they were similar in their qualitative and quantitative composition to the diets used for onfeeding of *Sander lucioperca* larvae (74% fish meal, 14% hydrolyzed fish meal, 0 – 13 % soy bean lecithin, 0 – 13% fish oil – Hamza, Mhetli, Ben Khemis, Cahu, & Kestemont, 2008). Relatively similar mixtures were also tested for *Sparus aurata* larvae (69% protein and protein hydrolysate, 12% soy bean lecithin, 5% fish oil - Robin and Vincent 2003). While in *Sander lucioperca* the tested microdiets resulted in a survival of circa 35 % after 34 days post hatch (Hamza et al., 2008), no survival was obtained in *Lota lota*. Thus might indicate different food requirements for the two species. Although all onfeeding experiments with artificial diets were successful in *Lota lota*, the following three conclusions can be drawn from the experiments. (1) Fifteen d survival of larvae was significantly lower with agar agar bound microparticles than with microparticles having no agar agar matrix. This might be due to an indigestibility of agar agar for *Lota lota* larvae. As the particles are dehydrated for grinding, they swell quickly when immersed in water and might exceed the size limit for *Lota lota* larvae. Also the attractivity of the microbound particles might be low, as ingestion was observed only rarely. (2) Formulated diets with high lipid content were inadequate, as concentrations of $\geq 7\%$ soya lecithin and $\geq 3\%$ fish oil resulted in lower larvae survival than 2 % soya lecithin and 1 % fish oil. This is in contrast to results on the pike perch where lipids and phospholipids in the described concentration range had no effect. For onfeeding of zebrafish larvae lipid concentrations of 1 – 4% and phosphatidylcholine concentrations of 2 – 4% were successfully used (Carvalho et al., 2006). (3) The supplementation of the formulated diets with additional protein sources as grinded artemia flakes or casein and with carbohydrates in form of dextran did not improve the larvae survival.

The present study demonstrates that rearing of *Lota lota* larvae with live zooplankton collected from wild populations resulted in high fish survival rates of $> 70\%$ in the small scale and in the large scale experiments. Therefore, zooplankton feeding is also practicable under intensive culture conditions in fish farming. When adequate facilities (lakes or ponds) for zooplankton collection are available the feeding method has advantages as high numbers of food animals can be collected and simultaneously separated in size fractions with low effort.

In the present experiment the number of food animals administered for on-feeding was circa 15 – 25 organisms per larvae and the food was adjusted to the requirements of the growing fish after 30 and 50 dph by feeding bigger organisms (bigger zooplankton size fractions). Shiri Harzevili et al. (2003) used 140 *Brachionus calyciflorus* per *Lota lota* larvae. After 10 days, nutrition was changed to circa 25 *Artemia* per larvae. In both studies the survival rate of *Lota lota* was circa 70% and the growth rate circa 200 % after 35 days post hatch. When *Lota lota* larvae were stocked in pond cages the survival from stocking to final recovery in September was 18 % and the average total length 49 mm (Paragamian et al., 2011).

A zooplankton analysis demonstrated, that composition in aspects of species and development stages revealed daily qualitative and quantitative variability which is consistent with earlier observations on plankton communities (e.g. Ferrara, Vagaggini, & Margaritora, 2005; Lahnsteiner, Kletzl, Weismann, 2009). The smallest zooplankton fraction of 200 - 400 µm, which was used as starter food for *Lota lota* larvae, contained mainly copepodites and nauplii while smaller organisms as rotifers and algae passed the 200 µm net. Generally, rotifers are considered as optimal for on-feeding of small fish larvae (*Lota lota*: Shiri Harzevili et al., 2003; *Ctenopharygodon idella*, *Aristichthys nobilis*: Rottmann et al., 1991; *Gobio gobio*, *Perca fluviatilis*: Awaiss, Kestemont, & Micha, 1992; different ornamental fish: Lim & Wong, 1997; Lim, Dhert, & Sorgeloos, 2003).

In summary the present investigations show, that on-feeding of *Lota lota* larvae with the tested artificial microdiets is impossible. As the larvae survival rates were zero in all experiments the diet composition is far away from the optimum and the development of suitable diets difficult. Onfeeding of *Lota lota* larvae with live zooplankton collected from wild populations is possible with high efficiency and is therefore a method suitable for fish farms and for intensive culture conditions. Possibly the molecular composition of natural zooplankton could serve as a basis to develop suitable formulated microdiets for *Lota lota* larvae.

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