



Influence of extracts of krill and chironomus larvae as feed attractants on growth, survival and feed utilisation in fry of European perch *Perca fluviatilis* Linnaeus, 1758

IVAYLO SIRAKOV AND STEFKA STOYANOVA

Faculty of Agriculture, Trakia University, Studentski Grad, 6000 Stara Zagora, Bulgaria
e-mail: ivailo_sir@abv.bg

ABSTRACT

Technological stages of European perch (*Perca fluviatilis* Linnaeus, 1758) farming and feeding norms satisfying its nutritional requirements are not well studied. A critical moment in farming of this species in recirculation systems is the transition from live feed to commercial pelleted feed. The aim of current research was to evaluate the influence of two attractants viz., extracts from krill and chironomus larvae as feed supplements for European perch (*P. fluviatilis*) fry on their survival, growth and feed utilisation. European perch fry were distributed in three experimental groups, viz., F₀: fed non-supplemented control feed; F_k: fed krill extract supplemented feed and F_{ch}: fed feed supplemented with chironomus extract. Survival of European perch supplemented with krill extract was 46% as compared to 26% in control group and 30% in fish supplemented with chironomus extract (p<0.05). The highest final weight and specific growth rate were observed in perch fed feed supplemented with chironomus extract but the differences between the krill-supplemented and control groups were not significant (p>0.05). Feed conversion ratio (FCR) in fish supplemented with chironomus extract was 1.91 which was significantly lower by 32.7% than control fishes and by 29.5% lower than fish fed with krill extract supplemented feed (p<0.01). The addition of krill extract to the feed of European perch fry improved survival rate of fry, while higher growth performance was demonstrated in fish fed with chironomus extract supplemented feed.

Keywords: Attractant, Chironomus, European perch, Feed utilisation, Growth, Krill

Introduction

During the past few decades, extensive research has been conducted at a global scale for improvement of feeding and reproductive performance of predatory fishes. European perch (*Perca fluviatilis* Linnaeus, 1758) is a species highly valued in the European market. It is a pelagic daytime predator and the larvae feed during the light part of the day (Dabrowski, 1982; Kestemont and Melard, 2000). The main concern for the fry is the transition from live feed to pelleted feed. For the different fish species, this stage is associated with a high risk as it is accompanied by high mortality (Szkudlarek and Zakes, 2002; Molnar *et al.*, 2004a, b; Policar *et al.*, 2013). The successful adaptation to artificial feed under controlled fish farming depends on the quality of pelleted feed, the proper formulation of daily rations and the age category of fish (Kestemont *et al.*, 1996; 2003; Policar *et al.*, 2013). The use of live feed in the early stages of feeding results in a more expensive produce (Kolkovski *et al.*, 2000). This is one of the reasons for the interest in the subject. One of the options to avoid using live feed in the diet of perch larvae is the supplementation of feed with attractants. According to Gaber (2005), feeding stimulants help to mask diverse

feeding deterrents which would lower the palatability of diet. It has been demonstrated that feed attractants usually have low molecule weight, could be dissolved in water, have acid active reaction and contain nitrogen (Nunes *et al.*, 2006). Other compounds which are found in attractants are free amino acids, nucleotides, nucleosides and ammonium bases (Kolkovski *et al.*, 2000), small peptides, amines, free amino acids as well as taurine and aniline, proline, sarcosine and glucosamine (Shimizu *et al.*, 1990; Paul and Yan, 2012).

The use of attractants in the feed of fry and fingerlings has resulted in good growth performance with respect to fish weight and body length, high survival rate and low feed conversion ratio (Polat and Beklevik, 1999; Molnar *et al.*, 2000; Molnar *et al.*, 2004a, b; Altun *et al.*, 2008). There are no studies on the use of feed attractants in the ration of European perch (*P. fluviatilis*) which is the subject of present study. Reports are available only for yellow perch, by Kolkovski *et al.* (2000) demonstrating a positive effect of supplements on the survival and growth performance of fish.

Krill and chironomus larvae are often used during weaning phase in fry and fingerlings of carnivorous fish

(Kubitza and Lovshin, 1997; Bodis *et al.*, 2007; Hubenova *et al.*, 2014). It has been reported that for Antarctica alone, the biomass of krill is estimated to be more than 440 million t (Hewitt *et al.*, 2004). Chironomid larvae can be reared at high densities under laboratory conditions in 20-22 days (Nandi *et al.*, 2014). Because of aforementioned facts, krill and chironomids are easily accessible for the sector and the possibility to be used for production of fingerlings of high value fish, make their use strongly rationale.

The aim of the current research was to evaluate the influence of two attractants *viz.*, extract from krill and extract of chironomus larvae supplemented to the feed of European perch (*P. fluviatilis*) fry on their survival, growth and feed utilisation.

Materials and methods

European perch

European perch fry were caught from the Sladak Kladenetz Dam and transported to experimental aquaculture base in Trakia University, Stara Zagora (Bulgaria). The fish fry consumed only natural feed before the start of the experiment. The weight of fish was measured using an electronic balance (with accuracy of ± 0.001 g) and fry with similar weight were transferred to tanks of the experimental recirculation system (RAS). The fish were separated into three experimental groups *viz.*, fish fed: control feed without addition of attractants (F0); feed sprayed with extract from krill as attractant (Fk); and feed sprayed with extract from *Chironomus* larvae as attractant (Fch). Commercial extracts from larvae krill (S-carp) and *Chironomus larvae* (Sensas) were used.

The initial average weight of European perch from different experimental groups were: F0 - 0.8 ± 0.002 g; Fk - 0.8 ± 0.009 g; and Fch - 0.77 ± 0.004 g. The initial weight of fish from the different experimental groups were not statistically different ($p \geq 0.05$). The stocking density of perch fry was 1 no. l⁻¹.

Experimental feeding

The tanks used in current research had 35 l volume. Water in recirculation aquaculture system (RAS) was treated by means of two mechanical filters and one moving bed biofilter each with 800 l volume. The moving bed biofilter consisted of 0.7 m³ biorings augmented with bacteriological culture from *Nitrosomonas* and *Nitrobacter* genera (Seranitrivec®). During the trial, the nitrate concentration was lowered by daily replacement of 10% of water in RAS with freshwater. The flow rate in every fish tanks was maintained at 5 l min⁻¹. During the trial, water quality parameters *viz.*, temperature (°C),

pH and oxygen (mg l⁻¹) were measured with a portable meter equipped with appropriate probes (HQ40D, Hach Lange®).

During the experimental trail, European perch fry were fed extruded commercial feed (Aqua Garant) having 0.4 mm granule size, for a period of 60 days. Nutritional content of the experimental feed used is presented in Table 1. In the current trial, addition of the tested attractants, *viz.*, extracts of krill and chironomus larvae, to commercial diet was carried out according to Telli *et al.* (2014) with a slight modification. The attractants were homogenised in sunflower oil (1% from daily feed amount) and sprayed on commercial feed granules. In the control feed, only oil was sprayed on granules @ 1% from daily feed amount instead of tested attractants. The sprayed feed was left to dry for 12 h. The fish were fed manually 7 times per day at every two hours between 07.00 to 19.00 hrs.

Table 1. Nutritional content of commercial feed used in the trial (according to Aqua Garant)

Parameters	Content
Crude protein (%)	64.00
Fat (%)	12.00
Dietary fibre (%)	0.50
Ash (%)	13.00
Calcium (%)	1.90
Phosphorus (%)	1.50
Sodium (%)	0.60
Digestible energy (MJ kg ⁻¹)	17.00
Vitamin A (IU kg ⁻¹)	10000
Vitamin D ₃ (IU kg ⁻¹)	1000
Vitamin E (mg kg ⁻¹)	480
Vitamin C (mg kg ⁻¹)	400
Mn (mg kg ⁻¹)	12
Cu (mg kg ⁻¹)	10
Zn (mg kg ⁻¹)	70
I (mg kg ⁻¹)	3
Se (mg kg ⁻¹)	0.5
Co (mg kg ⁻¹)	1

During experimental feeding, daily the number of dead fry was counted and at the end of the trial, the survival was calculated using the following equation:

$$\text{Survival (\%)} = \left[\frac{\text{Number of fish at the end of trial}}{\text{Initial number of fish}} \right] \times 100$$

Specific growth rate (SGR, % day⁻¹) in the experimental fish was calculated using the equation:

$$\text{SGR} = \left[\frac{(\ln W1 - \ln W2)}{D} \right]$$

where, $\ln W_1$ = natural logarithm of fish weight at the end of the trial; $\ln W_2$ = natural logarithm of fish weight at the beginning of the trial; D = no. of experimental days.

The feed conversion ratio (FCR) was calculated using the equation:

$$FCR = \left[\frac{F}{W_f - W_i} \right]$$

where, FCR = feed conversion ratio, F = amount of feed fed (g), W_f = weight of perch fry at the end of the trial (g). W_i = initial weight of perch fry (g).

The data were analysed by means of single-factor ANOVA (Statistica).

Results and discussion

During the feeding trial, water temperature ranged between 22.0 to 24.7°C and the average oxygen content varied from 7.40 to 8.5 mg l⁻¹ and were found to be within the optimum range for the species (Melard *et al.*, 1995; Fiogbe *et al.*, 1996).

In our trial, the average survival rate for perch fry fed krill extract-supplemented feed (Fk) was 45.7%, while survival of control fish (F0) and fish supplemented with extract of chironomus larvae (Fch) was considerably lower, being 25.7 and 30% respectively. The differences between groups F0 and Fk as well as between groups Fk and Fch were statistically significant ($p < 0.05$) (Fig. 1). Experiments with pikeperch have shown higher survival rates when the diet was supplemented with frozen chironomus (Bodis *et al.*, 2007; Hubenova *et al.*, 2014). In our study, the highest survival rate was obtained in the group that received krill extract supplemented feed (Fig. 1). This was in line with the results of Kolkovski *et al.* (2000).

In the beginning of the trial, the average live weight of perch from the three groups ranged between 0.77-0.8 g. There were no statistical differences between average

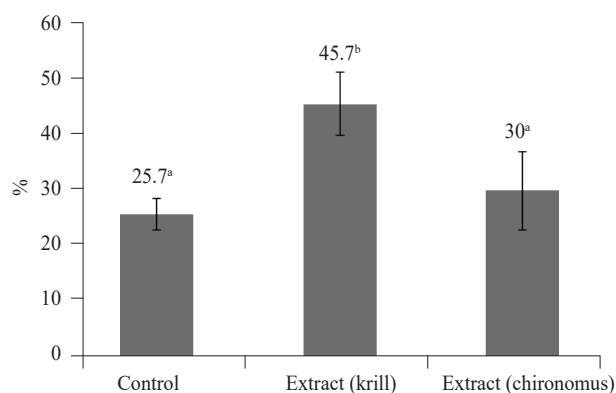


Fig. 1. Survival of European perch (*Perca fluviatilis*) fed feed with and without attractants

weights in the three groups ($p \geq 0.05$) (Fig. 2a). The final live weights of perch from the three experimental groups are presented in Fig. 2b. Fish from experimental group Fch had 10.3% higher final average live weight than that of fry in group F0 and 16.8% higher than fishes of group Fk ($p \leq 0.05$). The results corresponded to the findings for another predatory fish species *i.e.*, pikeperch, whose final live weight was higher in the groups supplemented with chironomus larvae and krill extract during weaning (Bodis *et al.*, 2007; Hubenova *et al.*, 2014).

At the end of the trial, SGR was the highest in perch from group Fch with 12.5% higher SGR than that of control fishes and 20.31% higher SGR as compared to krill-supplemented fish, but without statistically significant differences ($p > 0.05$) (Fig. 2c). These results corresponded to the data reported by Bodis *et al.* (2007), evidencing higher specific growth rate in pikeperch fed chironomus supplemented feed. Kolkovski *et al.* (2000), established higher, though statistically insignificant growth rates in yellow perch supplemented with krill ($p > 0.05$).

At the end of the experiment, average FCR in perch supplemented with Fch was 1.91 which was lower than that of control fish and Fk by 32.7 and 29.5% respectively

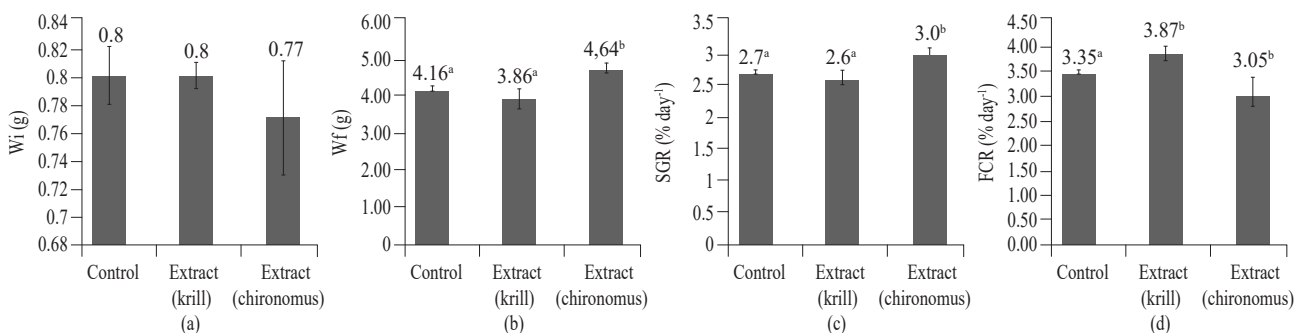


Fig. 2. Growth parameters in European perch (*Perca fluviatilis*) during the experimental trial: (a) Initial weight of perch (g); (b) Final weight of perch (g); (c) SGR (% day⁻¹) and (d) FCR

(Fig. 2 d). The differences in FCR between group F0 and Fch and between Fk and Fch were significant ($p < 0.01$). This agreed with reported results in other percid fish species (Kolkovski *et al.*, 2000; Rowland *et al.*, 2005; Bodis *et al.*, 2007).

Our observation showed that during the experiment, the feed consumption was faster with Fk, followed by Fch and the most slowly consumed feed was F0. Food attractiveness and stimulation of ingestion of fish by attractants results from physical and chemical stimulation (Kolkovski *et al.*, 2000). The higher speed of consumption of krill and chironomus supplemented feeds by perch fry during the current experiment could be the result of high level of water soluble protein and free amino acids present in krill extract (Hansen, 2011) and high protein level (up to 55.7%) and polyunsaturated fatty acids, PUFA (34.03%) in chironomus larvae (Bogut *et al.*, 2007). Other possible reason for the attraction of perch fry towards feeds supplemented with extracts of krill and chironomus larvae could be the change in colour of the granulated feed. These conclusions are confirmed by Shimizu *et al.* (1990) who stated that chemical stimulation is related to olfactory and gustatory responses of feed, while physical stimulation is related to the colour of feed.

According to the results of the present study, the supplementation of feed with krill extract, considerably improved the survival rates during the weaning of perch fry to dry pelleted feed, while supplementation with extract of chironomus larvae improved specific growth rate and feed conversion ratio. Probably, krill extract facilitated the transition of perch larvae to dry pelleted feed while the effect of supplementation of chironomus larvae extract was manifested when the fry has already accustomed to dry feed and it would have improved its utilisation. Therefore, it will be interesting to investigate whether the combined use of both supplements would act synergistically, at the time of weaning European perch fry to dry feed.

Acknowledgements

This research work was carried out with the support of Faculty of Agriculture and financed from projects 5AF/16.

References

- Altun, T., Ozluer, A., Gungor, E., Celik, F. and Polat, A. 2008. Transition of wild-caught juvenile pikeperch, *Sander lucioperca* to dry feed using different types of food. *J. Anim. Vet. Adv.*, 7(1): 5-10.
- Bodis, M., Kucska, B. and Bercsenyi, M. 2007. The effect of different diet on the growth and mortality of juvenile pikeperch (*Sander lucioperca*) in the transition from live food to formulated feed. *Aquac. Int.*, 15: 83-90. DOI:10.1007/s 10499-006-9063-0.
- Bogut, I., Has-Schon, E., Adamek, Z., Rajkovic, V. and Galovic, D. 2007. *Chironomus plumosus* larvae - a suitable nutrient for freshwater farmed fish. *Poljoprivreda*, 13(1): 159-162.
- Dabrowski, K. R. 1982. The influence of light intensity on feeding of fish larvae and fry: II. *Rutilus rutilus* L. and *Perca fluviacalis* L. *Zool. Jahrb. Physiol.*, 86: 353-360.
- Fiogbe, E. D., Kestemont, P., Melard, C. and Micha, J. C. 1996. Effects of dietary crude protein on growth of the Eurasian perch (*Perca fluviatilis*). *Aquaculture*, 144: 239-249. DOI:10.1016/S0044-8486(96)01293-8.
- Gaber, M. M. A. 2005. The effect of different levels of krill meal supplementation of soybean-based diets on feed intake, digestibility and chemical composition of juvenile Nile tilapia *Oreochromis niloticus*, L. *J. World Aquac. Soc.*, 36: 346-353. doi.org/10.1111/j.1749-7345.2005.tb00338.x.
- Hubenova, T., Zaykov, A., Katsarov, E. and Terziiski, D. 2014. *The influence of stocking density on the growth and survival of pikeperch (Sander lucioperca L.) during weaning from natural to dry feed.* *Jivotnovydni Nauki, Agricultural Academy, Sofia, LI*, 4:36-40.
- Hewitt, R. P., Watkins, J., Naganobu, M., Sushin, V., Brirley, A. S., Demer, D., Kasatkina, S., Takao, Y., Goss, C., Malyshko, A., Brandon, M., Kawaguchi, S., Siegel, V., Trathan, P., Emery, J., Everson, I. and Miller, D. 2004. Biomass of Antarctic krill in the Scotia Sea in January/February 2000 and its use in revising an estimate of precautionary yield. *Deep Sea Research Part II: Topical Studies in Oceanography*, 51: 1215-1236. doi.org/10.1016/j.dsr2.2004.06.011.
- Kestemont, P., Melard, C., Fiogbe, E. D., Masson, G. and Vlavanou, R. 1996. Nutritional and animal husbandry aspects of rearing early life stages of Eurasian perch *Perca fluviatilis*. *J. Appl. Ichthyol.*, 12(3-4): 157-165.
- Kestemont, P. and Melard, C. 2000. *Aquaculture*. In: Craig, J. F. (Ed.), *Percid fishes, systematic, ecology and exploitation*, Blackwell Science Publishing, Oxford, p. 191-224.
- Kestemont, P., Houbart, M., Jourdan, S., Melard, C., Paspatis, M., Fontaine, P., Kentouri, M. and Baras, E. 2003. Size heterogeneity, competition and cannibalism in cultured predatory fish larvae: abiotic and biotic influences. *Aquaculture*, 227: 333-356. DOI:10.1016/S0044-8486(03)00513-1.
- Kolkovski, S., Czesny, S. and Dabrowski, K. 2000. Use of krill hydrolysate as a feed attractant for fish larvae and juveniles. *J. World Aquac. Soc.*, 31: 81-88. DOI:10.1111/j.1749-7345.2000.tb00701.
- Kubitza, F. and Lovshin, L. L. 1997. The use of freeze-dried krill to feed train largemouth bass (*Micropterus salmoides*): feeds and training strategies. *Aquaculture*, 148(4): 299-312. doi.org/10.1016/S0044-8486(96)01426-3.

- Melard, C., Kestemont, P. and Grignard, J. C. 1995. Intensive ongrowing of European perch juveniles (*Perca fluviatilis*): zootechnical parameters and growth. In: Kestemont, P. and Dabrowski, K. (Eds.), *Short communications of the Workshop on the Aquaculture of Percids*, 23-24 August 1995, Vaasa, Finland, p. 34-37.
- Molnar, T., Hancz, Cs., Molnar, M. and Stettner, G. 2000. Investigation on technological parameters of pikeperch (*Stizostedion lucioperca*). *Agriculture*, 6(1): 126-128.
- Molnar, T., Hancz, Cs., Molnar, M. and Horn, P. 2004a. The effects of diet and stocking density on the growth and behaviour of pond pre-reared pikeperch under intensive conditions. *J. Appl. Ichthyol.*, 20: 105-109. DOI:10.1046/j.1439-0426.2003.00529.
- Molnar, T., Hancz, Cs., Bodis, M., Muller, T., Bercsenyi, M. and Horn, P. 2004b. The effect of initial stocking density on growth and survival of pike-perch fingerlings reared under intensive conditions. *Aquac. Int.*, 12: 181-189. DOI:10.1023/B:AQUI.0000032079.62056.8.
- Polat, A. and Beklevik, G. 1999. The importance of betaine and some attractive substances as fish feed additives. *CIHEAM - Options Mediterraneennes*, p. 217-220.
- Policar, T., Stejskal, V., Kristan, J., Podhorec, P., Svinger, V. and Blaha, M. 2013. The effect of fish size and stocking density on the weaning success of pond-cultured pikeperch *Sander lucioperca* L. juveniles. *Aquac. Int.*, 21(4): 869-882. DOI:10.1007/s10499-012-9563.
- Rowland, S. J., Allan, G. L., Mifsud, C., Nixon, M., Boyd, P. and Glendenning, D. 2005. Development of a feeding strategy for silver perch, *Bidyanus bidyanus* (Mitchell), based on restricted rations. *Aquac. Res.*, 36: 1429-1441. DOI:10.1111/j.1365-2109.2005.01364.
- Shimizu, C., Ibrahim, A., Tokoro, T. and Shirakawa, Y. 1990. Feeding stimulation in sea bream *Pagrus major*, fed diets supplemented with Antarctic krill meals. *Aquaculture*, 89: 43-53. doi.org/10.1016/0044-8486(90)90232-C.
- Szkudlarek, M. and Zakes, Z. 2002. The effect of stock density on the effectiveness of rearing pikeperch *Sander lucioperca* (L.) summer fry. *Arch. Pol. Fish.*, 10: 115-119.
- Telli, G. S., Ranzani-Paiva, M. J., Dias Dde, C., Sussel, F. R., Ishikawa, C. M. and Tachibana, L. 2014. Dietary administration of *Bacillus subtilis* on haematology and non-specific immunity of Nile tilapia *Oreochromis niloticus* raised at different stocking densities. *Fish Shellfish Immunol.*, 39: 305-311. doi.org/10.1016/j.fsi.2014.05.025.