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Attempts to increase the growth rhythm of juvenile Northern pike (*Esox lucius*, L.) by adding enzyme based ingredients into dry feed

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Abstract. Northern pike's rearing in recirculation systems is limited because of increasing FCR together with size of fish. The reason is apparently connected to the disability of pike to secrete sufficient digestive enzymes to process any offered dry feed. During feeding on natural food, the pike gets the necessary enzymes from the prey's body. This is the reason why we added to feeds digestive enzymes and even "fish juice". We used commercially available enzymes such as ®Colebil and ®Triferment in different dozes. We also used supplements of live feed. The results are encouraging since we noticed a higher growth rate in feeding the pikes fingerlings with dry feed enriched with protease enzymes.
Key Words: Northern pike, enzyme, growth, recirculation system.

Rezumat. Creşterea ştiucii în sisteme recirculante este limitată din cauza Ratei de Conversie a Hranei (FCR) ce creşte odată cu dimeniunea peştelui. Motivul se pare că este legat de incapacitatea ştiucii de a secreta suficiente enzime digestive pentru procesarea oricărei hrane granulate oferite. În cazul hrănirii cu hrană naturală, ştiuca îşi procură enzimele necesare digestiei chiar din pradă. De aceea în experimentul nostru am adăugat în furaje enzime digestive, dar şi „suc de peşte". S-au folosit enzime disponibile pe piaţă, sub formă de medicamente precum ®Colebil şi ®Triferment în diferite doze. Am administrat, de asemenea, şi suplimente de hrană vie. Rezultatele sunt îmbucurătoare, remarcându-se un ritm de creştere mai ridicat în cazul folosirii de enzime proteolitice în furajarea puietului de ştiucă.
Cuvinte cheie: ştiucă, enzime, creştere, sistem recirculant.

Introduction. Northern pike is becoming more popular among fish farmers, since the sport fishing market is increasing. The northern pike is a very popular sport fish, therefore increasing the production is necessary (Kucska et al 2002a, 2002b; Bodis et al 2003). Intensive farming for pike has limitations because the pike seems to stop growing at certain sizes, and continuing to feed with dry feeds becomes less economic. Previous research (Muscalu et al 2012) based in adding supplements during feeding showed a better growth rate at pike juveniles. It is very clear that the dry feed alone cannot supply the pike's digestive system with enough enzymes, therefore growth is slow. This fact was also observed by other scientists (Poczyczyñski 1996) that stated that pike larvae need the enzymes from their prey in order to have a normal growth (Engstrom 2005). Since larvae have already this problem, it is fair to conclude that also fingerlings and furthermore adults have the same problem. In our experiments we have used some pharmaceutical products containing digestive enzymes, one product containing lipases and other proteases.

Material and Method. Research was carried in a closed recirculated system, consisting in 5 fiberglass tanks, 1250 liters each, connected to a water treatment unit. The water is passing through a drum filter with 50 micron mesh, a moving bed biofilter, an oxygen contactor (15 meter deep underground U-tube). The water exchange rate in the tanks was 2 times per hour. The illumination was natural, although wooden shades were used

to decrease the light intensity. The time intervals of our trials were between 20th of July and 2nd of November 2012, a period of 105 days.
Twenty net cages were made from plastic mesh (a=4 mm) with equal shape and size (40cm x 40cm x 40 cm), submerged 30 cm in water. Four tanks were equipped with 4 cages, and one with 2 cages.

The two cages were populated with 100 pikes each, consisting in the control lot (duplicate). Then, six variants (three cages each) were installed, populating each cage with 100 pikes. In total, 2000 pike fingerlings were used. The pikes were weaned on dry feed using consecrated methods (Kucska et al 2005). At the beginning of the experiment, the pikes were 55±2 mm long and weighing 1.8 gram. All fish were initially fed on dry feed containing 56% Protein and 11% Crude Fat, with pellet size of 1.3-2 mm. Later, when pikes exceeded 100 mm, feed was switched to 45% Protein and 15% Fat, and pellet size 3 mm. By the end of experiment, when pikes exceeded the size of 160 mm, the pellet size was increased to 4.5 mm. The experimental variants are shown in Table 1.

Table 1

Experimental variants of diets for pike

Variant	*Additive to dry feed*	*Ammount of additive*	*Frequency of administration*
control	only dry feed	-	continuously
V1	fish juice	10%	continuously
V2	lipase (®Colebil)	1 mg g^{-1} of feed	continuously
V3	lipase (®Colebil)	5 mg g^{-1} of feed	continuously
V4	protease (®Triferment)	5 mg g^{-1} of feed	continuously
V5	protease (®Triferment)	10 mg g^{-1} of feed	continuously
V6	live fish	100%	one full day per week

The fish juice was made by mincing 100 gram of carp fry (1-5 gram size) with a kitchen blender. The resulting juice was mixed in a bowl with 1 kg of dry feed. After the dry feed absorbed most of the liquid it was put to dry-off. The resulted feed was kept in the refrigerator, and used for maximum 5 days, when it was replaced by freshly prepared one.

The pharmaceutical products were used simply by mincing the pills into a fine powder and the mixed with dry feed. Some water was added. After drying, the products were completely absorbed into the feed pellets. This feed was prepared every 5 days.

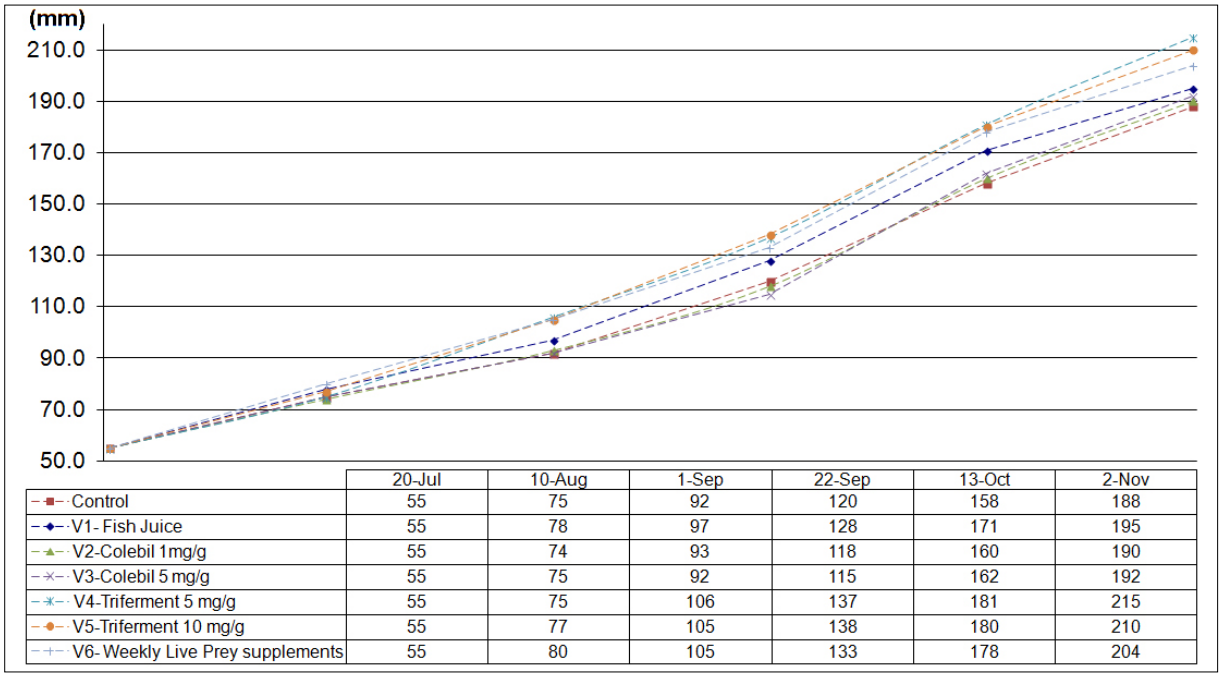
All pellets, with or without additives were distributed in a ratio of 5% BW/D, only by hand feeding. The feeding was made in up to 12 times per day, removing each time the wooden cover. Of course, not all of the feed was taken by pikes, therefore, the FCR was not calculated.

Regarding the V6, every Sunday morning, the pikes were feed ad-libidum with carp fry and fingerlings. The remaining carps were removed from the experimental cages each Sunday night. In the coming Monday, it was noticed in the morning a lower appetite for dry feed, but the situation changed by evening.

Every three weeks, the variants were evaluated, by making control harvest from each cage. Fish were randomly picked for length and weigh measurements. However, counting was made every time. Up to 25 fishes were measured each time from each cage.

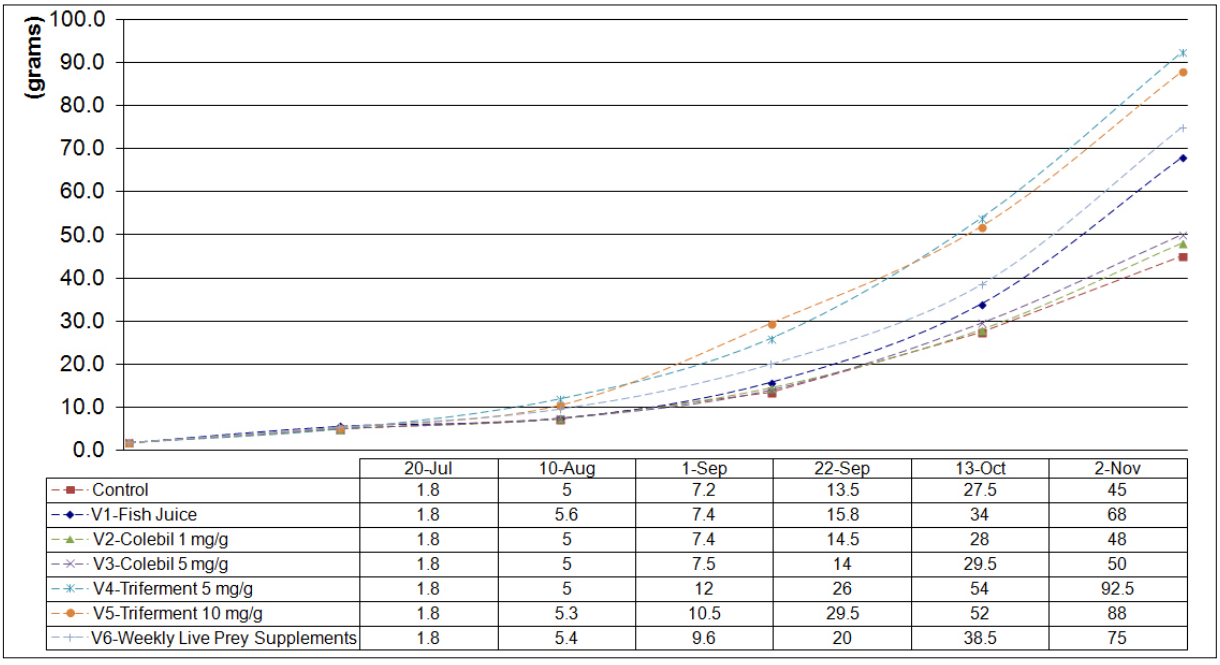
Results and Discussion. The pikes were measured every three weeks, and the population was evaluated for cannibalism rate. It was interesting that no pike was found dead. The losses were only caused by cannibalism. Every time we noticed a pike swallowing another pike, we eliminated both the prey and the predator, and recorded as two losses. At each evaluation, after counting, we sometimes recorded less fish than expected, without seeing a bigger pike between the others. In this case we noted only the missing pikes as losses. The idea was to record the survival rate as pikes able to be reared in intensive system, able to eat dry feed (even if some of them were occasionally

cannibals). Figures 1 and 2 show the evolution of standard length and body weight for the studied variants.



	20-Jul	10-Aug	1-Sep	22-Sep	13-Oct	2-Nov
Control	55	75	92	120	158	188
V1- Fish Juice	55	78	97	128	171	195
V2-Colebil 1mg/g	55	74	93	118	160	190
V3-Colebil 5 mg/g	55	75	92	115	162	192
V4-Triferment 5 mg/g	55	75	106	137	181	215
V5-Triferment 10 mg/g	55	77	105	138	180	210
V6- Weekly Live Prey supplements	55	80	105	133	178	204

Figure 1. Evolution of standard length for pike fingerling fed on different additive diets.



	20-Jul	10-Aug	1-Sep	22-Sep	13-Oct	2-Nov
Control	1.8	5	7.2	13.5	27.5	45
V1-Fish Juice	1.8	5.6	7.4	15.8	34	68
V2-Colebil 1 mg/g	1.8	5	7.4	14.5	28	48
V3-Colebil 5 mg/g	1.8	5	7.5	14	29.5	50
V4-Triferment 5 mg/g	1.8	5	12	26	54	92.5
V5-Triferment 10 mg/g	1.8	5.3	10.5	29.5	52	88
V6-Weekly Live Prey Supplements	1.8	5.4	9.6	20	38.5	75

Figure 2. Evolution of body weight for pike fingerling fed on different additive diets.

Although the different diets resulted in a relative constant development of standard length, the situation is much clearer for body weight. Figure 2, clearly shows that diets with additives of lipases had almost no effect on the growth rhythm of pike. Very small difference in growth is seen on the end of the experiment where lipases additives determined a growth of 6.6% for lipases concentration of 1 mg g^{-1} of feed and 11,1% for concentration of 5 mg g^{-1} of feed. Although there is a difference, we consider that added lipases are not determinant factors in successful intensive pike farming.

In contrast, the proteases seem to have a much higher impact in the growth of pike fingerlings. Even from the early stages of experiment, the differentiation was obvious. The proteases, at both concentrations, have a high impact of growth. The size of

pikes is almost double in weight than the size of the control variant (fed with simple dry feed). There is a 95% and 101% higher growth rate on feeding with protease additives at concentrations of 5 mg g^{-1} and 10 mg g^{-1}, respectively. We consider that 5 mg g^{-1} of feed is enough in order to have a significant result in pike farming.

Also the live fish supplements administrated weekly had a strong impact on growth, as demonstrated before (Muscalu et al 2012). The result seems to be in the middle, between control or lipase additives and adding proteases. The growth rate was increased with 66%.

As expected, also the fish juice had a high effect on growth, similar to the live fish supplements, with a growth rate higher with 51% than the control.

The survival rate was also determined on the experimented trials, as the Figure 3 illustrates.

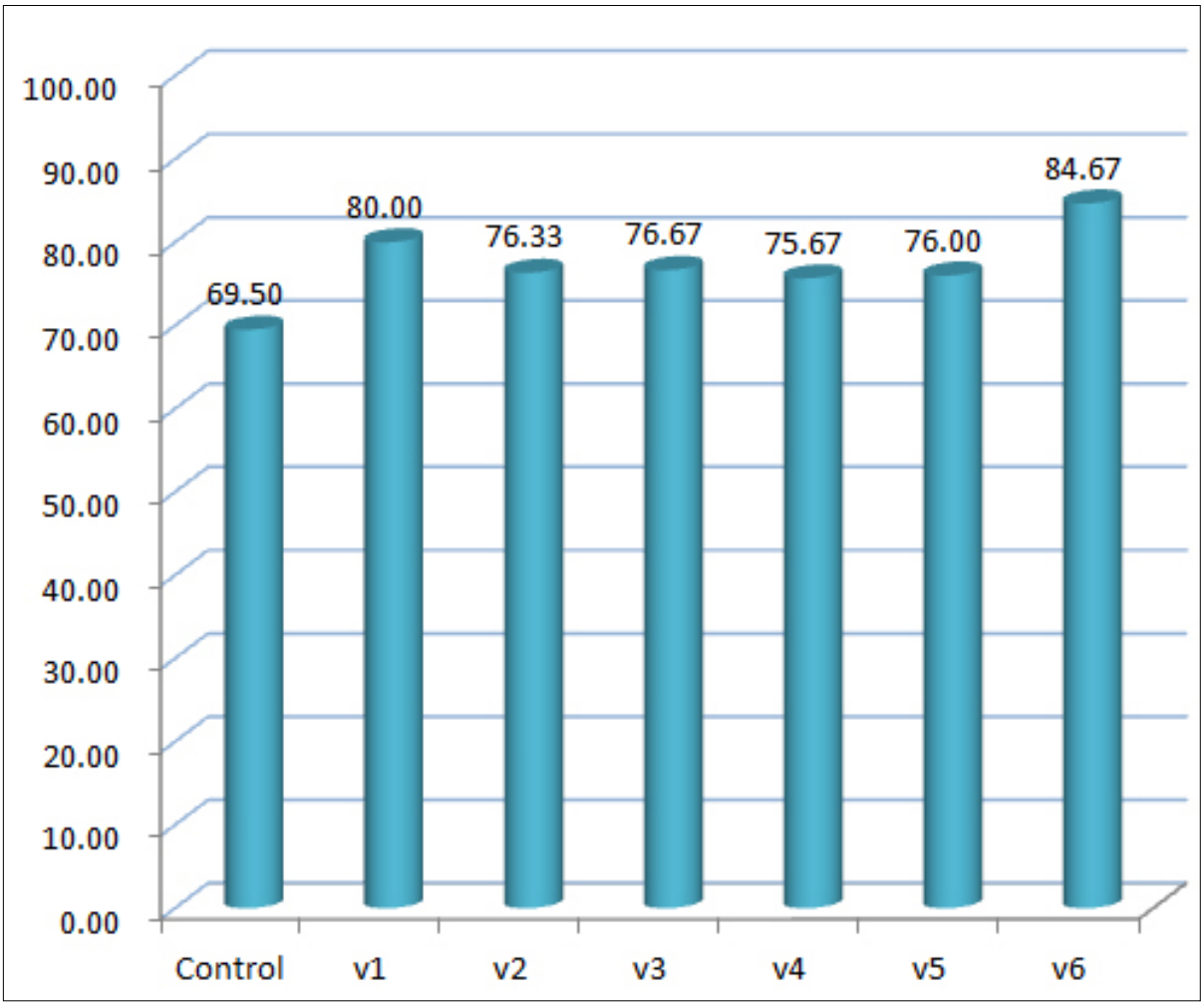


Figure 3. Survival rate for pike fingerling fed on different diets.

The highest survival rate was recorded in the trials where live feed or natural additives (fish juice) were used. The highest survival rate was at fingerlings fed with live fish once a week (84.67%) and next to the fish fed with dry feed soaked in fish juice (80%). Then, survival was almost the same in experimental trials, except the control, where it was registered the highest cannibalism rate. Here survival was only 69.5%. The high cannibalism rate for the lowest growth rate is explainable because usually cannibalism occurs more frequently at smaller pikes. It seems that together with increased growth rhythm, the risk of cannibalism is increasing also, maybe because the feeding rate was not enough in our experiments, and pikes were always hungry. Probably further experiments on this will eliminate also the high cannibalism rate.

From practical point of view, we suggest that using fish juice and live fish as prey in intensive, large scale farming is not feasible due to high polluting effect of this ingredient and risk of infestation with pathogens, although the encouraging survival rate would tempt any farmer. We used these variants in order to strengthen the belief that pikes needs some elements from live prey in order to grow normally. Using proteases, from pharmaceutical products (for example ®Triferment) as additives could be very profitable in spite of the lowest survival rate.

Further experiments are necessary in order to establish precisely the enzyme responsible to faster growth rate, since the pharmaceutical product ®Triferment contains a mixture of proteolitic enzymes. Perhaps histological studies would emphasize the digestive tract modifications from one variant to another. Moreover, since the FCR couldn't be calculated also the feed intake from one variant to another was not determined. What is certain and proved is that the protease is responsible to a higher growth rate to pike fingerlings up to the size of 90 grams. This article represents partial results, the work is continuing also next year when the experiments will be continued up to market size pike.

Conclusions. Using live fish supplements in the diet of Northern pike brings an increased growth rhythm with 66% that of control variant, and a survival rate of 84.67%.

Using fish juice as additive in dry feed results in increasing the growth rate with up to 51%, and has a survival rate of 80%.

Lipases bring an increased growth rate of 6.6% and 11% compared to control at concentrations of 1 mg g^{-1} and 5 mg g^{-1}, respectively. The cannibalism in this case brings a survival rate of just above 76%.

Protease from pharmaceutical products gives an increase rate of 95-101% compared to control. The survival rate in this case is low, only 75-76%.

Although the lowest survival rate due to cannibalism was noticed when proteases were used, we recommend the use of these additives for better growth rate for Northern pike.

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