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RESEARCH ARTICLE

Growth and survival in earthen ponds of different sizes of juvenile pike reared in recirculating aquaculture systems

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Abstract. The aim of the study was to determine the growth and survival of juvenile pike, *Esox lucius* L., reared in recirculating aquaculture systems (RAS) and then released into earthen ponds. Three different sizes of fish were used in the experimental releases of stocking material with mean body weights of 1.5 g (group S), 7.0 g (group M), and 18.5 g (group L) which were reared in a recirculating aquaculture system and fed formulated feed exclusively. Before the fish were released, they were tagged with visible implant elastomer (VIE) tags. The highest final body weight was attained by the fish in group S, the mean body weight of which was 85.5 g, which was significantly statistically higher than in the other groups ($P < 0.001$). No differences in survival were noted among the groups. Positive biomass increases were only exhibited in group S (37 kg ha^{-1}), and it was highly statistically significantly greater than it was in the other groups ($P < 0.001$). The results of the experiment could indicate that the suitability of juvenile pike for stocking depends on its size at release. Extending the rearing period in RAS resulted in poorer stocking results in the ponds.

Keywords: pike, stocking, RAS, survival, VIE tags, elastomer

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Introduction

Pike, *Esox lucius* L. is a species of considerable significance in aquatic ecosystems. In many waters it is the primary predator, and as such plays a significant ecological role in regulating populations of other fish species, especially of cyprinids. Because of its wide range of occurrence and abundance, it is of significant importance to fisheries management. In many countries, because of deteriorating natural spawning conditions for this littoral species, populations of pike are often protected by various types of conservation measures including the implementation of minimum landing lengths, closed fishing periods, and stocking (Paukert et al. 2001, Sutela et al. 2004). In Poland pike is the most frequently stocked fish species in terms of both quantity and financial outlay (Mickiewicz 2011). Stocking is usually performed with the youngest

Table 1
Basic composition of diets used in the experiment with juvenile pike

Feed	Total protein (%)	Raw fat (%)	Carbohydrates (%)	Digestible energy (MJ kg-1)	Pellet size (mm)
Nutra Amino Balance 2.0	54	18	8	19.5	0.6-1.0
Nutra Amino Balance 1.1	54	18	8	19.5	0.8-1.4
Nutra 1.5	54	18	8	19.5	1.5
Nutra 1.9	52	20	8.5	19.9	1.9

developmental stages, e.g., larvae, or with material obtained through extensive pond cultivation with summer and autumn fingerlings. The use of larvae carries with it the very high risk of mortality when environmental conditions are disadvantageous, which is why it is preferable to stock older developmental stages of this species (Margenau 1999).

In recent years it has become increasingly common to produce stocking material in recirculating aquaculture systems (RAS) and techniques for doing so are improving. The rearing conditions in these systems are controlled which ensures optimal fish growth and high survival rates. Thanks to advancements in rearing technologies, it is now possible to conduct intensive rearing of juvenile pike in RAS using commercial formulated feed (Wolnicki and Górny 1997, Kucska et al. 2005, Szczepkowski 2009). One of the obstacles to the wider use of stocking material originating from intense rearing is the lack of documented data on the fate of such material after it has been released into the natural environment, or, in other words, the effectiveness of stocking with this type of material is unknown.

The aim of the current study was to determine the growth rate, condition, and survival of three size groups of juvenile pike reared in RAS (body weight range from 1 to 20 g) and then released into earthen ponds.

Materials and methods

Origin and rearing of stocking material

The study material was juvenile pike obtained from rearing in a recirculating system. In spring 2011 wild pike spawners caught in Lake Dgał Wielki (Mazurian Lake District, northeastern Poland) were used for artificial reproduction. Standard procedures were used to obtain sex products and to fertilize and incubate the eggs (Szczepkowski and Szczepkowska 2008). Larval and juvenile stages for experimental stocking were reared exclusively on commercial formulated feed from the Nutra line (Skretting, France): Nutra 2.0, Nutra 1.1, Nutra 1.5, Nutra 1.9 (Table 1). The feed was delivered 24 h per day by an automated band feeder (FIAP, Germany). The fish were reared in a recirculating system with a total water volume of 60 m^3, in square basins made of synthetic materials, each of which had a working volume of 1 m^3. The mean water temperature during rearing in the RAS was 18.2°C. The oxygen content, pH, total ammonia nitrogen (CAA = NH_4^+-N + NH_3-N), and nitrites (NO_2-N) were monitored. The values of these parameters were measured at the rearing tank outflows and were as follows: oxygen content was above 5.2 mg dm^{-3}, water pH was from 6.77 to 6.91, the maximum contents of CAA and nitrites were 0.30 mg CAA dm^{-3} and 0.24 mg NO_2-N dm^{-3}, respectively. The rearing tanks were illuminated 24 h per day. The stocking density during rearing did not exceed 15 kg m^{-3}.

Tagging procedure and tag reading

During rearing, the pike were tagged according to three size groups: mean body weight of 1.5 g (group S; fish age 32 days post hatch (DPH); 7.0 g (group M; fish age 53 DPH); and 18.5 g (group L; fish age 85 DPH) (Table 2). All of the fish were tagged with visible implant elastomer (VIE) tags manufactured by Northwest Marine Technology Inc. (USA). The tags

Table 2
Characteristics of three size groups of juvenile pike reared in RAS and used for the experimental stocking of ponds (mean values ± SD; n = 100)

Parameter	Fish size group		
	group S	group M	group L
Body weight (g)	1.5 ± 0.3	7.0 ± 1.6	18.5 ± 3.8
Body length (cm)	5.7 ± 0.4	9.5 ± 0.6	12.8 ± 0.9
Total length (cm)	6.4 ± 0.4	10.6 ± 0.7	14.3 ± 1.0
Body weight coefficient of variation CV (%)	19.9	22.8	20.4
Age of fish post-hatch (days)	42	63	96
Period of rearing fish in RAS (days)	34	55	88
Elastomer tag color	red	yellow	green
Date ponds stocked	1 Jun 2011	22 Jun 2011	25 Jul 2011
Number of fish released into ponds (ind.)	507	375	254

were fixed in place subcutaneously on the head with a manual injector. Each group received a different colored tag (Table 2), and in order to increase visibility, the tags used were fluorescent. The tags were read *in vivo* under UV light during examinations of the harvested fish.

Stocking and harvesting the ponds

Three earthen ponds with surface areas of 0.5 ha each and mean depths of 1.5 m were used for the experimental stocking. Each pond was stocked with all three groups of juvenile pike, which were released subsequently as they grew in the RAS (Table 2). The ponds were stocked on the following dates: group S – June 1, group M – June 22, group L – July 25. Each pond was stocked with a total of 1,136 juvenile pike. The ponds used in the stocking experiment were drained in the winter and then filled in spring. In order to ensure a food base for the juvenile pike, the ponds were stocked with cyprinid spawn in April, followed by tench, *Tinca tinca* (L.), spawners in June. The mean water temperature during the period from stocking to harvest was 17.7°C at a range of 11.0-23.9°C. The ponds were harvested in October at a water temperature range of 12.5-14.0°C. The fish were harvested when the water was being drained, and they were collected in harvesting pens located behind the outlet box or in the ponds. In all of the ponds harvested in addition to juvenile pike there were also abundantly occurring cyprinids including roach, *Rutilus rutilus* (L.), rudd, *Scardinius erythrophtalamus* (L.), and tench.

Measuring procedures and statistical analysis

To determine the body weight, body length, and total length of the fish before release into the earthen ponds, 100 individuals were measured from each group. After the autumn harvest, all of the fish were counted and segregated into the three size groups according to the elastomer tags. Each individual was also evaluated to identify any visible anomalies. Measurements of from 150 to 250 fish from each pond were taken. Body weight was determined to the nearest 0.1 g, and length was measured to the nearest 1 mm. The digestive tracts of a randomly selected sub-sample of from 20-50 individuals from each pond were dissected to determine what the fish fed on. Before all measurements, tagging, and tag reading, the fish were anesthetized by placing them in an aqueous solution of Propiscin (IFI Olsztyn, Poland) at a concentration of 1-2 cm^3 dm^{-3} water (Kazuń and Siwicki 2001).

Table 3
Growth performance, condition, production, survival, and number of pike with body deformations reared at three stocking size groups in earthen ponds. Group S – initial body weight 1.5 g; group M – initial body weight 7.0 g; group L – initial body weight 18.5 g. Data are mean values ± SD, n = 3. Groups tagged with different letter indexes in the same row differ significantly statistically (P < 0.05)

Parameter	Fish size group		
	group S	group M	group L
Final body weight (g)	85.5 ± 15.5^{a}	23.4 ± 5.7^{b}	23.2 ± 1.4^{b}
Specific growth rate SGR (% d^{-1})	3.10 ± 0.17^{a}	1.10 ± 0.18^{b}	0.30 ± 0.07^{c}
Body weight coefficient of variation CV (%)	94.3 ± 24.6^{a}	35.1 ± 9.6^{a}	53.8 ± 38.3^{a}
Condition coefficient (K)	0.77 ± 0.02^{a}	0.72 ± 0.01^{b}	0.70 ± 0.02^{b}
Stock biomass increase (kg ha^{-1})	37.0 ± 4.1^{a}	-0.8 ± 1.4^{b}	-5.4 ± 1.5^{b}
Survival (%)	45.4 ± 8.9^{a}	26.0 ± 9.5^{a}	34.0 ± 13.6^{a}
Number of individuals with body anomalies (% of all fish in a group)	11.5 ± 0.8^{b}	14.2 ± 0.8^{ab}	18.3 ± 3.4^{a}

The analysis of the results was based on calculating the following cultivation indexes:

- specific growth rate, SGR (% d^{-1}) = 100 × (ln final body weight (g) – ln initial body weight (g)) × rearing time^{-1} (days);
- body weight coefficient of variation, CV (%) = 100 × (body weight standard deviation (g) × mean body weight^{-1} (g));
- condition coefficient, K = 100 × (body weight (g) × body length $^{-3}$ (cm));
- stock survival, S (%) = 100 × (final abundance (individuals) × initial abundance $^{-1}$ (individuals));
- fish biomass increase in ponds, B (kg ha^{-1}) = (biomass of harvested fish (kg) – biomass of released fish (kg)) × pond surface area (ha^{-1}).

Statistical calculations were performed with Statistica 5.0PL (StatSoft Inc.). Single factor analysis of variance (ANOVA) was used to confirm the significance of differences. The significance of differences among experimental variants was performed based on mean values of replicates within a given variant. Tukey's multiple range test was used to evaluate the significance of differences. Differences were considered to be statistically significant at $P \leq 0.05$.

Results

The small size of the elastomer tags and their colors meant that they were not clearly visible without using floorescent lights. Additionally, the tags shifted position on the fish heads. With highly differentiated fish body weights ranging from several to several hundred grams, only in a small portion of the fish, approximately 20% and mainly smaller individuals, were the tags visible on the outer side of the fish body. In all the other fish, the tags were visible on the inner side of the operculum.

The mean body weight of all the groups of pike harvested in autumn together was 63.7 g with a range of 57.4 to 72.1 g in all three ponds (Table 3). The total length was 21.6 cm at a range of 17.3 to 24.3 cm. The mean survival in the ponds from release to harvest was 36.4% at a range of 30.0 to 41.7%. Biomass increases ranged from 26.3 to 37.4 kg ha^{-1} at a mean of 30.8 kg ha^{-1}. The effectiveness of tagging was high with tags confirmed in 96.8 to 98.8% of the individuals from the three ponds.

The analysis of the results of the different pike size groups stocked into the ponds indicated that growth rates differed ($P < 0.05$; Table 3). The highest final body weight was noted in the fish from group S, in which the mean body weight was 85.5 g. In the other two groups it was similar at 23.4 g in group M and 23.2 g in group L. The specific growth rate (SGR) in group S was nearly three-fold higher than that in group M and nearly ten-fold higher than that in group L ($P < 0.05$: Table 3). The value of the body weight coefficient of variation (CV) ranged from 35.1% (group M) to 94.3% (group S; $P > 0.05$; Table

3). The value of the condition coefficient in group S was 0.77, which was significantly statistically higher than that in group M (0.72; $P < 0.05$) and in group L (0.70; $P < 0.01$). Fish survival in the different groups ranged from 26.0% in group M to 45.4% in group S ($P > 0.05$). A significant difference was confirmed in the increase in fish biomass in the different size groups. Only in group S did biomass assume a positive value at 37 kg ha^{-1}, which was significantly higher than in the other groups ($P < 0.05$; Table 3). The number of individuals with visible physical anomalies increased with the size of the fish stocked. In group S it was 11.5% of all individuals, while in group L it was 18.4% ($P < 0.05$; Table 3). The anomalies were largely noted in mouth structure and one- or two-sided opercular twisting. Among the pike harvested, 88.3% were confirmed to have prey in their stomachs at a range of 86.0 to 90.0% in the different ponds. No incidents of cannibalism were confirmed.

Discussion

The intensive rearing of fish in RAS is of increasing economic importance, especially in the case of juvenile stages (Bergheim et al. 2009). Some advantages, among others, of this system is the possibility of producing significant quantities of material of similar condition and size and the opportunity of selecting environmentally optimal periods to release stocking material into open waters. The rearing conditions in RAS differ significantly from those in the natural environment, and this can pose problems when using the fish produced in them for stocking.

The current experiment indicated that the juvenile pike stocking material reared in a RAS can be used to stock ponds. Its survival until autumn was similar to or higher than the results when using larval stocking material (Wright and Giles 1987, Szczerbowski 1993, Szczepkowski 2011), and the final biomass was also within the range of values obtained in earthen ponds (Bry and Souchon 1982, Szczerbowski 1993). It should be emphasized that in the current experiment an increase in the final stock biomass was only noted in the case of one of the three size groups released into the ponds. Density dependence in growth rates is often considered to result from a lack of food resources for the fish (Pierce 2012). Most of the harvested fish (90%) were found to have fish prey in their stomachs. This is an indication that the pike reared in the RAS were able to adapt to consuming natural food and shifted from feeding on granulated feed that was delivered continually to actively hunting for and capturing fish prey.

When stocking is performed with fish reared using traditional, extensive methods, stocking effectiveness is greater the larger the fish are (Marshall and Johnson 1971, Farrell and Werner 1999, McKeown et al. 1999). This is linked with the choice of conditions ensure feeding and environmental conditions are better during stocking (Mesing et al. 2008) as well as the fact that larger fish are less susceptible to predation (Wahl and Stein 1989). In the current study no such simple dependence was observed. The best results were obtained from stocking the smallest, youngest pike forms with body weights of approximately 1.5 g. The fish from this group attained the highest final body weight, which was three-fold higher than that in the other groups, and the greatest relative increase in body weight. Only in the group of the smallest pike was a positive increase in fish biomass noted, and this was within the range of values obtained in earthen ponds (Szczerbowski 1993). It is noteworthy that the different results obtained from stocking the ponds with pike of different initial sizes did not result from different survival rates, for which there were no statistically significant differences among the groups, but because of the different growth rates of the fish in the analyzed size groups. This might have resulted from differences in how well the pike from the various size groups adapted to pond conditions that differed from those in the tanks and also how the fish adapted to catching live prey. Confirmation of this might be the very low weight gain among the largest pike stocked, even though the availability of food was ensured for each of the three size groups with prey of various sizes. Interesting study from ponds has provided new evidence that social stresses can be just as important (Edeline et al. 2010). Authors stocked pike at different densities into experimental ponds and manipulated the amount of food the fish were receiving so that feeding rates were similar. Increased pike density significantly depressed individual pike growth in both length and weight. Edeline et al. (2010) concluded that social stress can alone drive demographic density dependence without any significant change in the consumptive effects of interference or exploitative competition.

Observations conducted under laboratory conditions of muskellunge, *Esox masquinongy* Mitchill, indicated that the fish fed feed consumed a similar number of fish-prey that were given to them and that their growth rates were similar to those of individuals that were fed only fish all the time (Szendreya and Wahla 1995). Differences were noted, however, after they were released into the natural environment where the survival of individuals that had been fed feed was lower; this stemmed from the fact that these fish were more susceptible to predation by other fish (Gillen et al. 1981, Szendreya and Wahla 1995).

The results of the current experiment correspond to other observations of intensive rearing of juvenile pike in recirculating systems (M. Szczepkowski, unpublished data). It was confirmed that the initial rearing phase (until weights of 5-10 g) is characterized by high fish growth rates and feed utilization, but that as the fish grow significant changes occur in fish behavior that reduce efficiency substantially (Szczepkowski 2009). It should be emphasized that these changes do not result, as one might presume, from increased mutual aggression from the fish since cannibalism is a sporadically observed phenomenon in the intensive rearing of older juvenile pike. This phenomenon is linked to the limited tendency to consume feed, and it might stem from the fact that formulated feed has wholly different physical and chemical properties than the food pike consumes in the wild, which means this feed is not readily accepted by these fish. It was confirmed, however, that under laboratory conditions pike weighing 24 g and which had been reared exclusively on formulated feed had difficulty catching live fish that were given to them. This suggests that these fish lacked the ability to catch fish-prey (Szczepkowski and Szczepkowska 2004). These studies indicated that pike lose their predatory instinct to a much greater degree than do pikeperch, *Sander lucioperca* (L.), but to a lesser degree than do Wels catfish, *Silurus glanis* L.

An interesting fact noted during the current study was that there were no incidences of cannibalism, even though this phenomenon can be exhibited among pike in significant proportions (Giles et al. 1986, Bry et al. 1992), and increased cannibalism has been observed among pike that is the same size as in the current experiment (Bry et al. 1995). When catching larger prey, the fish are susceptible to cannibalistic attack from other pike in the moment before the prey is swallowed (Nilsson and Brönmark 1999). One of the factors that might have prevented cannibalism could also have been the stocking density which did not exceed 9.4 individuals 100 m^{-2} during harvest. In wild lakes cannibalism was not noted at a density of up to 4.7 individuals 100 m^{-2} (Sutela et al. 2004), but it was noted at densities of 6.8-13.3 individuals 100 m^{-2} (Korhonen 1995, cited in Sutela et al. 2004). It is also possible that cannibalism occurs only during the initial period of stocking, especially since the density of fish stocked at 22.7 individuals 100 m^{-2} was substantially higher than that reported by Wright and Giles (1987) as the upper limit for stocking density (5 fry individuals 100 m^{-2}) at which increased mortality is not noted. The limited losses to cannibalism in this critical period could also have been because the material released was of similar size, as it had been sorted earlier during rearing in the RAS. It is widely known that size differences in the case of pike is one of the most common causes of cannibalism and low fish survival in the 0+ age group (Skov et al. 2003). It is plausible, however, that this conclusion could refer to the fish from one size group since it is not known what the size relations were when subsequent groups of pike were released into the ponds.

The results of the experiment presented in this paper indicate that the size of the pike stocking material reared in RAS and stocked into earthen ponds can be significant for the effectiveness of stocking. The optimal size that guarantees the highest stocking effectiveness is material with a body weight of approximately 1.5 g. The results obtained could be significant for strategies to use pike stocking material reared in RAS. The possibility of using smaller forms of stocking material is important economically as it permits shortening the rearing period and reducing costs. There appears to be no justification for releasing larger sizes of pike even though rearing them is relatively easy (Szczepkowski 2009) even at high stocking densities (exceeding 100 kg m^{-3}, Szczepkowski unpublished data). The number of anomalies that occur in subsequently larger pike size groups is also noteworthy, and their increasingly frequent occurrence in longer periods of rearing in RAS might indicate technological errors in the rearing of older fish. Identifying and eliminating these could improve the effectiveness of using them as stocking material.

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