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Results and Outcomes of Induced Breeding and Fry Rearing of Zander (*Sander lucioperca* L.)

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Abstract

In this study data on successful propagation of zander (*Sander lucioperca L.*) from several spawning seasons are evaluated. Propagation was based on stripping of spawners after hormonal induction and rearing of fry in artificial ponds. Fish were injected with acetone-dried carp pituitary extract at a dose of 4.0 mg/kg homogenized in a fish physiological saline. When ovulation was detected, the eggs were stripped and weighed. Eggs were collected from each ovulated female and fertilized with milt from the male. Stickiness of zander eggs was removed before transferring them into Zuger jars. Data collected from four consecutive spawning seasons were analyzed statistically calculating individual and group-based pseudo-gonadosomatic index (PGSI) values. Reproduction capacity assays were grouped on the basis of three aspects: (1) examination of differences between years, (2) within a reproductive season, and (3) examination of the effects of females' size. During the four-year trial period eggs were gained from 121 hormonally treated females, 64.7% of the total 187. Based on the data of the investigated period the mean PGSI was 10.7%. At 95% significance level PGSI data of different years were considered to be similar. There were no significant differences between PGSI data collected from different periods of the reproductive season. Previous findings on the positive correlation between the amount of stripped eggs and body weight of females were confirmed (r^2=0.103). Correlation was not justified between PGSI and body weight (r^2=0.002). Advanced rearing of zander fry in a 2.0 hectare pond was carried out in six consecutive years. Before stocking the nutrient level of the pond was increased by organic manuring and the Copepods were eliminated by insecticides. To prevent cannibalism, four weeks after stocking the advanced fry of zander were harvested. On the basis of the results from six consecutive years, after rearing of advanced fry in a shallow, 2.0 hectare rearing pond for four weeks, the mean survival rate was 13.4±4.37% (mean±SD). By considering our results, breeders can calculate their hatchery's and pond's production in advance. These results will also be helpful in calculating the total weight of broodstock which must be held in order to produce a specific number of advanced fry.

Keywords: Zander, *Sander lucioperca* L., induced breeding, fry rearing.

Introduction

In recent years, market demand for Percid fishes including zander has significantly increased. According to the 2012 FAO database, the amount of fish caught from natural waters are highly variable and gradually decrease from year to year. Conventionally, in fish farms zander is produced as an associated species of carp ponds. Traditionally, this species is cultured on an extensive level in open fish ponds, predominantly in combination with common carp (*Cyprinus carpio*) and other warm water Cyprinids (Huet, 1986). As a recent development, zander is also produced in intensive aquacultures in a low but gradually increasing level. Recently, induced breeding under hatchery conditions based on stripping has come to the front (Zakes and Demska-Zakes, 2005).

To evaluate breeding success, it is important to know the reproductive potential of females. An appropriate indicator of the reproductive potential is the gonadosomatic index (GSI). The general formula for the GSI is: ovary weight × 100 / ovary weight + body weight. However, not all of the oocytes present in the ovary ovulate before spawning. Considering the real reproductive capacity of the female fish, more accurate estimation can be achieved by measuring the amount of eggs stripped from females. Pseudo-gonadosomatic index (PGSI) can be calculated as follows: weight of stripped eggs (g) × 100 / body weight of the female (g). Data of successful propagation based on stripping of zander females induced to ovulate in hatcheries had been collected during four breeding seasons.

On the basis of physiological demands of zander fry, two-phase rearing (a nursing period for 3-4 weeks and then the rearing of fingerling up to autumn) can secure higher survival rates and optimal body sizes (200-300 g / one-summer old individual). Because of the small size of the feeding larvae of zander the appropriate pond preparation and multiplication of small-sized food organisms is essential (Horváth *et al*., 1984). During pond preparation the chemical treatment eliminates cyclops and the rotifers can be stimulated by the use of fertilisers. When the plankton population is highly concentrated the small zander grow very quickly. After one month the fish are 40±5.0 mm long. At this stage the pond cannot supply enough food and cannibalism quickly becomes a problem. To prevent this, harvesting should be commenced.

Until now most of the stocks of advanced fry come from fry rearing earthen ponds. It is not only able to provide fry for intensive systems but is also a safe source for stocking fish ponds, reservoirs and natural waters (Tamazout, 2008). Pond rearing of zander carried out in our experiments was based on the selection of zooplankton. The technology is only effective if an accurate protocol of plankton management with numerous agrotechnical steps are employed. The most important phases of this nursing procedures are summarized by Horváth *et al*. (2002). Efficiency of this rearing protocol expressible in numbers is presented in our study on the basis of 6-year data of a 2.0 ha zander nursing pond located in Hungary.

Materials and Methods

Zander broodfish were kept in a wintering pond of Attala Fish Ltd., Hungary for 3 months before propagation. One day before ovulation induction, the fish were transferred to the hatchery with well saturated running water at 12.0±1.0°C. Experiments were conducted during the normal breeding season of zander in mid April. The number of stripped females and their mean weight, as well as the mean PGSI values for each season are indicated in Table 1.

Fish received hormonal treatment with acetone-dried carp pituitary extract at a dose of 4.0 mg/body weight kg. Four to ten pairs of treated fish were placed into a tank (3×3 m, water depth 0.3 m, oxygen saturation above 80%) with the same number of artificial nests. Behavior of zander pairs were being continuously monitored over the period of ripening. When ovulation was detected, the eggs were stripped and weighed. Eggs were collected from each ovulated female and fertilized with milt from the male. Stickiness of zander eggs was removed before transferring them into Zuger jars. For this reason zander eggs were hardened for about 1.0 to 1.5 hours in a fertilization solution (20 g NaCl and 15 g carbamide in 10 liter of water).

Collected data of different seasons were analyzed statistically (Graphpad Prism 4 for Windows) for calculating individual and group-based PGSI values. The minimum significance level was set at $P<0.05$. Reproduction capacity assay were grouped on the basis of three aspects: (1) For comparison of PGSI data from the four consecutive seasons a non-parametric Kurskal-Wallis Test and Dunns Multiple Test were used; (2) To examine differences within a reproductive season three different periods were created within a spawning season. Data followed normal distribution, therefore one-way ANOVA was applied; and (3) Examination of the effects of females' size: pairs were created from data of body weight and PGSI and then correlation analysis was applied.

The technology of two-phase zander fry rearing results in a reproducible and safe survival only by the application of an accurately employed protocol with numerous agrotechnical steps. Most important steps of the nursing protocol applied in present experiment are summarized by Horváth *et al*. (1984). After stocking, the small zander start a rapid development. The average temperature of the pond water was 16.0±2.0°C over the rearing period. Extent of development, composition and quantity of zooplankton were monitored by one sampling per week. In the warming spring environment within 3-4 weeks zander fry reached a body length of 3-5 cm. Due to a favorable survival and the rapid growth, the plankton stock of experimental pond gradually decreased. After the third week zander fry started to starve, consequently their growth rate decreased. In the third and fourth week cannibalism appeared. To prevent damages of cannibalism at the age of 3-4 weeks small zander were harvested within 1-2 days. During harvesting, in the crowded fingerling stock cannibalism could increase further so harvesting was done at night or early morning. Efficiency of this zander rearing protocol expressible in numbers is presented in our study on the basis of six-year data of

Table 1. Number of stripped females, their mean body weight (BW) in grams and the mean PGSI values (%) for each experimental season

Experimental seasons	No. of striped females	BW of females (mean±SD)	PGSI (mean±SD)
1st season	31	2307.3±519.6	10.2±4.4
2nd season	34	1852.9±231.2	11.6±6.0
3rd season	18	1916.7±191.7	10.5±7.2
4rd season	38	1852.6±159.4	10.5±2.9

a 2.0 ha zander nursing pond.

Results

During the four-year trial period eggs were gained by stripping from 121 hormonally treated females, 64.7% of the total 187. No eggs could be stripped from 60 zander females since they had already spawned before stripping on artificial nests placed in the tanks. Six females did not answer to the hormonal treatment. Based on the data of the investigated period the mean PGSI was 10.7%.

At 95% significance level PGSI data of different years were considered to be similar (Figure 1). There were no significant differences between PGSI data collected from different periods of the reproductive season (Figure 2). Previous findings on the positive correlation between the amount of stripped eggs and body weight of females were confirmed (r^2=0.103), although correlation were not justified between PGSI and body weight (r^2=0.002) (Figure 3).

Efficiency of our zander rearing protocol expressed in numbers is presented on the basis of six-year data of a 2.0 ha zander nursing pond located in Hungary (Table 2.). In each season the pond was stocked with feeding larvae produced by induced propagation in a neighboring hatchery in the same region. It is apparent from the data that in a relatively small pond area advanced zander fry can be produced in a commercial quantity. From the six nursing experiments, the average survival rate over the for-week nursing period was 13.4±4.37%.

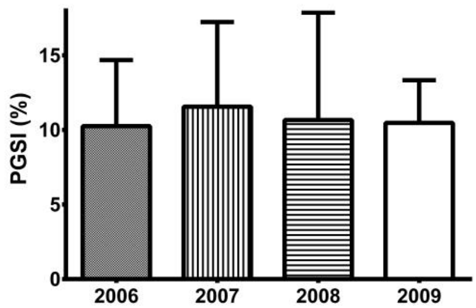

Figure 1. PGSI data (mean±SD) of the examined zander spawners of four different seasons.

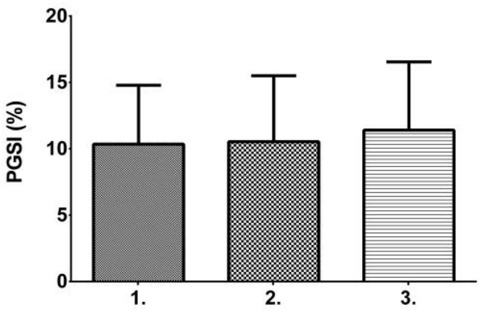

Figure 2. PGSI data (mean±SD) of the examined zander spawners in three different part of the spawning season. 1. First part of the season. 2. Middle part of the season 3. End of the spawning season.

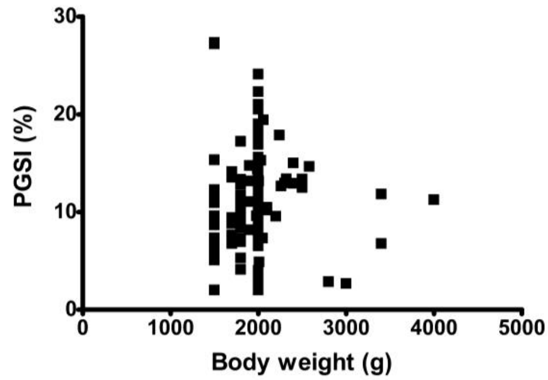

Figure 3. Relationship between PGSI and females size.

Discussion

Before the spawning season of zander under natural conditions, when the size of ripe oocytes is the largest in the gonads, the value of GSI can reach 14-19% (Lappalainen *et al*., 2002). The relative fecundity which is defined as the number of eggs per 1g of female varies between 150 and 400 eggs on average (Lappalainen *et al*., 2002). However, there is no data about the GSI of zander females cultured in farm conditions. As a summary of these trials, it could be concluded that the average value of PGSI of stripped females was 10.7%. At the same time no justifiable differences within or between seasons could be reported. Furthermore, no supportable connection between PGSI and body weight was declared, hence the success of propagation is only moderately affected by these parameters. On the other hand data collected from female zander bred in fish farms as an associated species can differ from egg production of zander stocks originated from natural waters or intensive systems.

The most important factor for offspring survival at natural reproduction of fish is the presence of living prey with adequate size at the start of exogenous feeding. It is also similar in case of rearing fry in ponds. A most relevant task of a fish breeder is to ensure the presence of natural food with a size proportional to that of fish larvae just starting to feed.

When fish species with small (4-7 mm) larvae start their exogenous feeding - e.g. most of Cyprinids peculiar in European stagnant waters and also zander, - in the first period due to their size are not able to eat most species of zooplankton living in their environment. Body sizes of most common zooplankton species (Cladocers and Copepods) are too big, many hundred microns. As a result, fish larvae in their first days of exogenous feeding are not able to capture them. For common carp and zander larvae the adequate size of zooplankton for their first food is small, 50-100 µm. In zooplankton rotifers, protozoa (mainly Ciliates) and nauplius larvae of Copepods have such body sizes.

Among primitive crustaceans there are certain species that serve as an important prey for fish but are also aggressive predators of fish larvae and small fry in their early periods of life (Tamás and Horváth, 1976). These mostly carnivorous Copepods attack and destroy all kinds of aquatic organisms which can not escape quickly enough or shake off their aggressors. Due to their large numbers these Copepod species are able to destroy whole stocks of fish fry.

In pond farms the zooplankton getting into the pond in the period of filling is fairly composite and usually the number of carnivorous Copepods is significant. If fish producers do not intervene and control the composition of zooplankton artificially their work will not have much result. No matter how much larvae is produced in hatcheries and stocked into the nursing ponds the number of copepods will always be larger and they will destroy a great ratio of fry produced with hard work and expense. Therefore, in the period of preparation for fry nursing fish farmers have to remove Crustaceans periodically and selectively. For this reason and mainly in fingerling rearing of Cyprinids insecticides belonging to the group of organic phosphoric acid esters has been applied for decades. These chemicals selectively remove Crustaceans belonging to arthropods but leave rotifers (Rotatoria) alive since these chemicals in such a weak solution are ineffectual for them. This treatment is called zooplankton selection (Tamás and Horváth, 1976).

In living waters containing oxygen of biological origin deriving from algal assimilation trichlorphon products decay quickly (within 24-36 hours), their phosphor content causes a slight phosphorus load in waters (Schmahl, 1991). This effect reckons as an advantage in nursing ponds since algal multiplication is required as a nutrient supply (algae) for rotifer stocks growing rapidly.

In nursing ponds cannibalism is especially significant in the period when the body length of fry hits 30-40 mm. For zander with such size the ratio of head length is big compared to that of torso length (nearly one-to-two). As a result, mouth opening is also disproportionately big, adequate for small zander to change from feeding on zooplankton to capturing of living fish fry. If it manages to catch a proper amount of small sized fish fry, as a result of sufficient protein-supply in the next weeks torso length will grow better. Therefore, mouth size is going to be proportionately smaller and cannibalism will also gradually decrease. Cannibalism – especially in case

Table 2. Data of stocking, harvesting and survival rate of zander fry from six consecutive rearing seasons. The size of the nursing pond was 2.0 ha

Years	No. of stocked larvae / ha	No of harvested fry / ha	Survival (%)
2005	2.0 million	283,900	14.2
2006	2.0 million	166,800	8.3
2007	2.0 million	237,500	11.9
2008	2.0 million	214,000	10.7
2009	2.0 million	421,500	21.1
2010	2.0 million	283,000	14.2
Mean	2.0 million	267,783+87,324	13.4+4.37

of belated harvesting – can cause a 50-60% loss in fingerling stocks so the preceding of its appearance is important in the prosperous survival of fry**.** From our nursing experiments, the average survival rate over the for-week nursing period was 13.4±4.37%. Survival rate was outstanding in 2009 probably because pray availability for zander larvae was optimal. The specific environmental factors however is hard to specify.

Zander has been described recently as a new candidate for intensive freshwater aquaculture (Wang *et al*., 2008). However, zander larvae are very difficult to grow on artificial feed (Zakes, 2012). Survival and growth of larvae are very low since their alimentary tract is undeveloped and enzymatic activity is very low compared to the adult fish. Better results were obtained by using mixed rearing of zander (first phase in ponds, and second – in tanks). Advanced fry of zander harvested from the ponds after several weeks of pre-rearing could be well adapted to the intensive rearing on artificial feed (Müller *et al*., 2011). Therefore, use of advanced fry of zander reared in ponds could be useful stocking material for intensive culture.

By considering our results breeders can calculate their production of zander feeding larvae as well as advanced fry in advance. These results will also be helpful in calculating the total weight of brood stock which must be held and wintered on the farm.

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