# ASPECTS REGARDING THE CONTROLLED REPRODUCTION OF PIKEPERCH (*SANDER LUCIOPERCA* LINNE, 1758) IN INDUSTRIAL AQUACULTURE SYSTEMS

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#### Abstract

Pike-perch is a very active predatory fish in its natural environment, but extremely sensitive in aquaculture units. Usually, the frequent manipulations may cause many losses in the fish farms, especially during artificial or controlled reproduction. This paper presents the obtained results at S.C.D.P. Nucet, in 2018-2020 period, regarding the pikeperch controlled reproduction in ponds units. The fish were divided into three experimental variants, in triplicate:  $V_1$ -controlled reproduction, without hormonal induction,  $V_2$ -controlled reproduction with hormonal induction (carp pituitary),  $V_3$  -controlled reproduction (synthetic analogue hormone-Nerestin 5A). Each variant used 5 females and 5 males with an average body weight of 2 kg/fish. The best technological indicators were registered in  $V_3$  variant, which was based on hormonal induction with Nerestin 5A hormone (88-92% eggs fertilization rate, 71-78% eggs hatching rate, about 2.7 million 7-8 days larvae). The smallest brood stock losses were recorded in  $V_1$  variant (without hormonal induction).

Key words: controlled reproduction, pikeperch, pituitary, ponds, synthetic hormone.

# INTRODUCTION

The pikeperch (*Sander lucioperca* L., 1758) is present in most of continental waters on the Romanian territory, being a valuable species from an alimentary and commercial point of view. This species is a promising candidate for fish diversification (Fontaine et al., 2009; Pourhosein Sarameh et al., 2012; Dalsgaard et al., 2013).

Pikeperch artificial reproduction is difficult to achieve due to the spawners sensitivity to manipulation (Kucharczyk et al., 2007), as a consequence, the mortality registration rates could raise up to 50%. Along the time, few studies have been focused on the pikeperch conttroled reproduction. Physiological processes in fish knowledges has facilitated the use of hormonal substances that stimulate maturation and reproduction (Rinchard et al., 2005).

# MATERIALS AND METHODS

The researches were realised in the 2018-2020 period at the Fish Culture Research and

Development Station Nucet. The experimental basins are located in the major riverbed of the Ilfov brook, downstream of the Ilfoveni accumulation dam. For the pikeperch conttroled reproduction, are needed: wintering ponds, prematuration ponds, maturation ponds, spawning ponds and the hatchery station. Breeding mattresses, made of bundled willow roots ('mustaches') (*Salix babylonica* L.), caught on a nytal sieve and mounted on a wooden support with 1.0 x 1.0 m sides, were made for laying the eggs (Figure 1).



Figure 1. Pikeperch breeding mattress

The mattresses thus made are fixed directly on the bottom of the pond by fastening wooden poles or by fixing with weights, at 2-3 m from the shore with a 8-10 m distance between them. Starting from the pikeperch breeding biology particularities in general (stages of ontogenetic development), the technology of reproduction includes:

- ✓ setting up groups of spawners;
- ✓ spawners selection and prematurization;
- ✓ conttroled reproduction ponds preparation and breeding mattress installation;
- ✓ spawners stimulation with hormonal substances;
- ✓ ponds stocking;
- ✓ monitoring the spawning process and the breeding mattresses control;
- ✓ collecting nests with embryonated eggs and introducing them into incubators;
- ✓ incubation, application of antifungal treatments and hatching process;
- $\checkmark$  larval growth until the age of 7-8 days old.

The spawners wintering took place in two ponds. In March, when the water temperature constantly reached 7-8°C values, the pikeperch spawners were transferred to prematurity ponds, separated by gender (Figure 2).



Figure 2. Pikeperch spawner taken out from wintering pond

For the experimental works of conttroled pikeperch reproduction, were selected 3-4 years old females, with an average weight of about 2 kg/ex, whose oocytes are in the 4<sup>th</sup> maturation stage. The selected males were the same age as the females, with an average weight of about 1.9 kg/ex. When the spawners where transfered from the wintering ponds to the prematurity ones, the oocyte polarization index was also determined, using oocytes extracted by probing (Zarski et al., 2011) (Figure 3).



Figure 3. The determination of the maturation stage of oocytes

According to the scientific protocol, 3 experimental groups were formed, being selected 90 spawners (45 females and 45 males). The experiments were performed in nine breeding ponds with an average area of 1000 m<sup>2</sup>/ponds. Before spawning, the three groups of females (15 ex/batch) were stocked separately in maturation basins. The males were all stocked in a maturation pond. The distribution in these ponds was made in the evening time, between 19<sup>00</sup>-21<sup>00</sup>, at a 11-16°C water temperature. The females from the 2 and 3 variants were injected the next day with the first dose of pituitary / synthetic hormones, and the second dose was administered 12-14 hours away. Simultaneously with the second dose, the spawners were introduced into the spawning ponds (5  $\bigcirc$  + 5  $\bigcirc$  / pond).

The experimental variants were the following:

- Variant 1 (V1) without females hormonal stimulation; performed in triplicate (R1, R2, R3), in basins B1, B2 and B3;
- Variant 2 (V2) with females hormonal stimulation with carp pituitary hormone, performed in triplicate (R1, R2, R3), in basins B4, B5 and B6;
- Variant 3 (V3) with females hormonal stimulation with Nerestin 5A, performed in triplicate (R1, R2, R3), in the basins B7, B8 and B9.

For females stimulated with carp pituitary hormone, the total dose administered was 3.5 mg/ kg body weight, and for females stimulated with Nerestin 5A, the total dose administered was 0.15 ml/kg body weight. In this case males were not hormonally stimulated.

In each experimental variant, the gonadosomatic ratio (GSR) for females was calculated; its average being 10% of body weight.

# **RESULTS AND DISCUSSIONS**

During the experiments, physico-chemical parameters of the water were periodic monitored. The obtained results interpretation was performed in accordance with the " Classification norm of surface water quality" provisions, correlated with the specialty literature data for aquaculture waters (OMMGA no. 161/2006) (Table 1).

Pikeperch families formed in 12-24 hours after their introduction into the reproduction ponds. According to data published by Zak & Demska-Zak (2005), the waiting time for laying eggs for a female can vary from 10 to 70 h after an HCG injection.

In 2018, reproduction began in basins B7, B8 and B9 (V3), followed by basins B1, B2 and B3 (V1) and basins B4, B5 and B6 (V2). The spawning season took place between 7-11 April.

In 2019, reproduction started in basins B7, B8 and B9 (V3), followed by basins B4, B5 and B6 (V2) and basins B1, B2 and B3 (V1). The spawning season took place between 11-15 April.

In 2020, spawning began in basins B7, B8 and B9 (V3), followed by basins B1, B2 and B3 (V1) and basins B4, B5 and B6 (V2). The spawning season took place between 13-17 April.

After laying the eggs on the breeding mattresses, the nests with fertilized eggs were collected and transported to the hatchery station, in water containers (Figure 4).



Figure 4. Pikeperch nest with embryonated eggs

The nests were introduced in "Nucet" type incubators, where a permanent water supply was ensured, with 8 liters/minute flow rate (Figure 5).



Figure 5. Pikeperch nest with embryonated eggs in "Nucet" type incubator

The data regarding the average values of these measurements, the average prolificacy and the type of hormone administered are presented in Table 2.

During the incubation, the eggs were bathed formaldehvde with а 37% solution (concentration 1.0-1.8 ml 37% formaldehvde/ 1 liter of water) to prevent the appearance and infestation with fungi. The first treatment was given 24 hours after the eggs were introduced to incubate. The exposure time was depending on the water temperature (10 min/10-12°C or 15 min/13-15°C). The process was repeated every 24 hours, until the embryo surrounds the entire volk sac, the caudal reaches the eves, the pigmentation is accentuated, the movements of the embryo become more intense and the heartbeat is observed.

In Table 3 are presented the results obtained for the conttroled reproduction of the pikeperch in the three experimental variants for each year of research.

During the incubation period, the water temperature was daily recorded, the average number of eggs laid in the nests and introduced to the incubator was evaluated, as well as the fertilization percentage. The incubation duration was 7-8 days at an average daily water temperature of 13.5°C. After hatching, the fasciculated willow roots ("mustaches") were removed from the "Nucet" type incubators.

Fish larvae were kept in incubators up to the age of 7-8 days, until the end of the vitelus reserves resorption period. The main indicators recorded in the experiments are presented in Tables 4-6.

|       |  |   |            | Parameter                 |                             |  |  |  |
|-------|--|---|------------|---------------------------|-----------------------------|--|--|--|
| C.No. | 0.1101                                   | Chemical parameter                          |            | Source Experimental ponds |                             | Optimal according to quality standards |  |  |
|       |  |   |            | Average of t              | he years 2018 - 2019 - 2020 |  |  |  |
| 1     | pН                                       |   | pH unit    | 7.2                       | 7.5                         | 7-7.8                                  |  |  |
| 2     | Alkalinity                               |   | mg /l      | 146                       | 163                         | 200-400                                |  |  |
| 3     | Calcium (C                               | a <sup>2+</sup> )                           | mg/l       | 38.2                      | 46.8                        | 90-120                                 |  |  |
| 4     | Magnesiu (1                              | Mg <sup>2+</sup> )                          | mg/l       | 22.4                      | 20.8                        | 10-40                                  |  |  |
| 5     | Ca2+/ Mg2+                               | Ca <sup>2+/</sup> Mg <sup>2+</sup>          |            | 1.7                       | 2.25                        | 5                                      |  |  |
| 6     | Organic Matter                           |   | mg KMnO4/l | 16                        | 22.95                       | 20-60                                  |  |  |
| 7     | Oxygen                                   |   | mg/l       | 10.4                      | 8.8                         | 5-12                                   |  |  |
| 8     | Ammonia (                                | NH <sup>+</sup> 3)                          | mg/l       | missing                   | missing                     | missing                                |  |  |
| 9     | Nitrates (NO                             | D <sup>-</sup> 3)                           | mg/l       | missing                   | 0.21                        | 2.5-4                                  |  |  |
| 10    | Nitrogen (N                              | NO <sup>-</sup> 2)                          | mg/l       | 0.002                     | 0.004                       | 0.03                                   |  |  |
| 11    | Phosphates                               | Phosphates (PO <sup>3-</sup> <sub>4</sub> ) |            | missing                   | 0.06                        | 0.05-1.5                               |  |  |
| 12    | Clorides                                 | Cl -  | mg/l       | 8.83                      | 8.43                        | 30                                     |  |  |
| 12    | Ciorides                                 | Na Cl                                       | mg/l       | 14.61                     | 14.03                       | 20                                     |  |  |
| 13    | Ammonium (NH <sup>+</sup> <sub>4</sub> ) |   | mg/l       | missing                   | 0.014                       | 0.5-1                                  |  |  |
| 14    | Total hardness                           |   | (°D)       | 12.6                      | 14.4                        | 12                                     |  |  |

Table 1. Water physical and chemical indicators for the 2018-2020 period (average values)

Table 2. Weight and number of the eggs according to the variant

|      |              | Variant 1(without hormonal stimulation)             |               |                          |              |  |               |                          |              |  |               |                         |
|------|--------------|---|---------------|--------------------------|--------------|--|---------------|--------------------------|--------------|--|---------------|-------------------------|
|      | V1R1         |   |               |                          | V1R2         |  |               | V1R3                     |              |  |               |                         |
| Year | W<br>(g/ex.) | Estimated<br>amount<br>of eggs<br>Wi (g)            | Eggs/<br>gram | Prolificity<br>(milions) | W<br>(g/ex.) | Estimated<br>amount of<br>eggs Wi<br>(g) | Eggs/<br>gram | Prolificity<br>(milions) | W<br>(g/ex.) | Estimated<br>amount<br>of eggs<br>Wi (g) | Eggs/<br>gram | Prolificiy<br>(milions) |
| 2018 | 2150         | 215   | 1260          | 1.3545                   | 1890         | 189                                      | 1260          | 1.1907                   | 2305         | 230.5                                    | 1260          | 1.452                   |
| 2019 | 1840         | 184   | 1260          | 1.1592                   | 2260         | 226                                      | 1260          | 1.4238                   | 1990         | 199                                      | 1260          | 1.254                   |
| 2020 | 1960         | 196   | 1260          | 1.2348                   | 2140         | 214                                      | 1260          | 1.3482                   | 2410         | 241                                      | 1260          | 1.518                   |
|      |              | Variant 2 (stimulation with carp pituitary hormone) |               |                          |              |  |               |                          |              |  |               |                         |
|      |              | V2  | 2R1           |                          | V2R2         |  |               | V2R3                     |              |  |               |                         |
| 2018 | 1990         | 199   | 1260          | 1.2537                   | 2380         | 238                                      | 1260          | 1.4994                   | 2220         | 222                                      | 1260          | 1.399                   |
| 2019 | 2310         | 231   | 1260          | 1.4553                   | 2120         | 212                                      | 1260          | 1.3356                   | 2310         | 231                                      | 1260          | 1.455                   |
| 2020 | 2080         | 208   | 1260          | 1.3104                   | 2320         | 232                                      | 1260          | 1.4616                   | 2150         | 215                                      | 1260          | 1.355                   |
|      |              |   |               | ١                        | /ariant 3 (  | stimulation                              | with syntl    | hetic hormor             | le)          |  |               |                         |
|      |              | V3R1  |               |                          | V3R2         |  |               | V3R3                     |              |  |               |                         |
| 2018 | 2070         | 207   | 1260          | 1.3041                   | 2410         | 241                                      | 1260          | 1.5183                   | 2160         | 216                                      | 1260          | 1.361                   |
| 2019 | 1890         | 189   | 1260          | 1.1907                   | 2260         | 226                                      | 1260          | 1.4238                   | 2310         | 231                                      | 1260          | 1.455                   |
| 2020 | 2350         | 235   | 1260          | 1.4805                   | 2120         | 212                                      | 1260          | 1.3356                   | 2060         | 206                                      | 1260          | 1.298                   |

| C.no. | Basin    | Installed<br>breeding<br>mattresses | Families<br>number | Variant | Number of collected nets | Maturated<br>females | Maturation<br>percent (%) |
|-------|----------|-------------------------------------|--------------------|---------|--------------------------|----------------------|---------------------------|
|       | 1        | 1                                   | Yea                | r 2018  |                          |                      |                           |
| 1     | B 1      | 5                                   | 5                  | V1 R1   | 3                        | 3                    | 60                        |
| 2     | В2       | 5                                   | 5                  | V1 R2   | 4                        | 4                    | 40                        |
| 3     | В3       | 5                                   | 5                  | V1 R3   | 3                        | 3                    | 60                        |
| 4     | B 4      | 5                                   | 5                  | V2 R1   | 4                        | 4                    | 80                        |
| 5     | В 5      | 5                                   | 5                  | V2 R2   | 4                        | 4                    | 60                        |
| 6     | B 6      | 5                                   | 5                  | V2 R3   | 4                        | 4                    | 60                        |
| 7     | В 7      | 5                                   | 5                  | V3 R1   | 4                        | 4                    | 80                        |
| 8     | B 8      | 5                                   | 5                  | V3 R2   | 5                        | 5                    | 100                       |
| 9     | В9       | 5                                   | 5                  | V3 R3   | 5                        | 5                    | 100                       |
| То    | tal V1   | 15                                  | 15                 | V1      | 10                       | 10                   | 66,7                      |
| То    | tal V2   | 15                                  | 15                 | V2      | 12                       | 12                   | 80                        |
| То    | tal V3   | 15                                  | 15                 | V3      | 14                       | 14                   | 93.3                      |
|       |          |                                     | Yea                | r 2019  |                          |                      |                           |
| 10    | B 1      | 5                                   | 5                  | V1 R1   | 4                        | 4                    | 80                        |
| 11    | В2       | 5                                   | 5                  | V1 R2   | 3                        | 3                    | 60                        |
| 12    | В3       | 5                                   | 5                  | V1 R3   | 3                        | 3                    | 60                        |
| 13    | B 4      | 5                                   | 5                  | V2 R1   | 4                        | 4                    | 80                        |
| 14    | В 5      | 5                                   | 5                  | V2 R2   | 4                        | 4                    | 80                        |
| 15    | B 6      | 5                                   | 5                  | V2 R3   | 4                        | 4                    | 80                        |
| 16    | В 7      | 5                                   | 5                  | V3 R1   | 5                        | 5                    | 100                       |
| 17    | В 8      | 5                                   | 5                  | V3 R2   | 5                        | 5                    | 100                       |
| 18    | В9       | 5                                   | 5                  | V3 R3   | 5                        | 5                    | 100                       |
| То    | tal V1   | 15                                  | 15                 | V1      | 10                       | 10                   | 66.7                      |
| То    | tal V2   | 15                                  | 15                 | V2      | 12                       | 12                   | 80                        |
| То    | tal V3   | 15                                  | 15                 | V3      | 15                       | 15                   | 100                       |
|       |          |                                     | Yea                | r 2020  |                          |                      | -                         |
| 19    | B 1      | 5                                   | 5                  | V1 R1   | 4                        | 4                    | 80                        |
| 20    | B 2      | 5                                   | 5                  | V1 R2   | 4                        | 4                    | 80                        |
| 21    | В 3      | 5                                   | 5                  | V1 R3   | 3                        | 3                    | 60                        |
| 22    | B 4      | 5                                   | 5                  | V2 R1   | 4                        | 4                    | 80                        |
| 23    | В 5      | 5                                   | 5                  | V2 R2   | 3                        | 3                    | 60                        |
| 24    | B 6      | 5                                   | 5                  | V2 R3   | 4                        | 4                    | 80                        |
| 25    | В 7      | 5                                   | 5                  | V3 R1   | 5                        | 5                    | 100                       |
| 26    | B 8      | 5                                   | 5                  | V3 R2   | 5                        | 5                    | 100                       |
| 27    | В9       | 5                                   | 5                  | V3 R3   | 4                        | 4                    | 80                        |
|       | tal V1   | 15<br>15                            | 15                 | V1      | 11                       | 11                   | 73.3                      |
|       | Total V2 |                                     | 15                 | V2      | 11                       | 11                   | 73.3                      |
| То    | tal V3   | 15                                  | 15                 | V3      | 14                       | 14                   | 93.3                      |
|       |          |                                     |                    | otal    |                          |                      |                           |
|       | tal V1   | 45                                  | 45                 | V1      | 31                       | 31                   | 68.9                      |
|       | tal V2   | 45                                  | 45                 | V2      | 35                       | 35                   | 77.8                      |
| То    | tal V3   | 45                                  | 45                 | V3      | 43                       | 43                   | 95.6                      |

Table 3. The obtained results during the spawning in the pikeperch conttroled reproduction

| C.No. | Biotechnological Indicators                               | M.U.              | V1            | V2                   | V3            |
|-------|---|-------------------|---------------|----------------------|---------------|
| 1     | Pond  | -                 | В 1-3         | B 4-6                | В 7-9         |
| 2     | Surface   | ha                | 0.3           | 0.3                  | 0.3           |
| 3     | Number of mattresses installed                            | -                 | 15            | 15                   | 15            |
| 4     | Families number   | -                 | 15            | 15                   | 15            |
| 5     | Gender relation   | \$\\$             | 1/1           | 1/1                  | 1/1           |
| 6     | Females characteristics                                   | no.ex/g/ex        | 15/2115       | 15/2197              | 15/2213       |
| 7     | Males characteristics                                     | no.ex/g/ex        | 15/1880       | 15/2050              | 15/2017       |
| 8     | Female hormonal stimulation                               | -                 | -             | Pituitary<br>hormone | Nerestin 5A   |
| 9     | Dosage  | mg/kg body weight | -             | 3.5                  | 0.15          |
| 10    | Reproduction period                                       |                   | 07-11.04.2018 | 07-10.04.2018        | 07-11.04.2018 |
| 11    | Reproductive water temperature                            | °C                | 11-15         | 11-15                | 11-15         |
| 12    | Collected nets  | no                | 10            | 12                   | 14            |
| 13    | Maturated females   | ex/g/ex           | 10/20015      | 12/2197              | 14/2213       |
| 14    | Maturation percentage                                     | %                 | 66.7          | 80.0                 | 93.3          |
| 15    | Average prolificacy                                       | eggs/♀            | 266490        | 276780               | 278880        |
| 16    | Eggs for incubation                                       | mil               | 2.6649        | 3.3214               | 3.9043        |
| 17    | Fertilization rate  | %                 | 88.1          | 89.4                 | 92            |
| 18    | Number of fertilized eggs                                 | mil               | 2.348         | 2.969                | 3.592         |
| 19    | Hatch percentage  | %                 | 74.3          | 74.8                 | 77.8          |
| 20    | Hatched larvae  | mil               | 1.7444        | 2.2210               | 2.7946        |
| 21    | Incubation survival                                       | %                 | 96.6          | 95.0                 | 98.1          |
| 22    | Larvae of 7 to 8 days viable                              | mil               | 1.6851        | 2.1100               | 2.7415        |
| 23    | Larvae 7-8 days / 🌳 matured                               | mil               | 0.1685        | 0.1758               | 0.1958        |
| 24    | Larvae 7-8 days / kg ♀ matured                            | mil               | 0.0797        | 0.0800               | 0.0885        |
| 25    | Fertilized eggs survival of larvae<br>7-8 days percentage | %                 | 63.2          | 63.5                 | 70.2          |

Table 4. The main technological indicators in 2018

Table 5. The main technological indicators in 2019

| C.No | Biotehnological Indicators        | M.U.              | V1           | V2                   | V3           |
|------|-----------------------------------|-------------------|--------------|----------------------|--------------|
| 1    | Pond                              | -                 | В 1-3        | В 4-6                | В 7-9        |
| 2    | Surface                           | ha                | 0.3          | 0.3                  | 0.3          |
| 3    | Number of mattresses installed    | -                 | 15           | 15                   | 15           |
| 4    | Families number                   | -                 | 15           | 15                   | 15           |
| 5    | Gender relation                   | ₽/ð               | 1./1         | 1./1                 | 1./1         |
| 6    | Females characteristics           | no.ex/g/ex        | 15/2030      | 15/2247              | 15/2153      |
| 7    | Males characteristics             | no.ex/g/ex        | 15/1970      | 15/2040              | 15/2007      |
| 8    | Female hormonal stimulation       | -                 | -            | Pituitary<br>hormone | Nerestin 5A  |
| 9    | Dosage                            | mg/kg body weight | -            | 3.5                  | 0.15         |
| 10   | Reproduction period               |                   | 1115.04.2019 | 1114.04.2019         | 1115.04.2019 |
| 11   | Reproductive water<br>temperature | °C                | 1115         | 1115                 | 1115         |
| 12   | Collected nets                    | no                | 11           | 12                   | 15           |
| 13   | Maturated females                 | ex/g/ex           | 11./2030     | 12/2247              | 15/2153      |
| 14   | Maturation percentage             | %                 | 73.3         | 80                   | 100          |

| C.No | Biotehnological Indicators                               | M.U.   | V1     | V2     | V3     |
|------|--|--------|--------|--------|--------|
| 15   | Average prolificacy                                      | eggs/♀ | 255780 | 283120 | 271280 |
| 16   | Eggs for incubation                                      | mil    | 2.8136 | 3.3974 | 4.0692 |
| 17   | Fertilization rate                                       | %      | 90.6   | 92.1   | 91.8   |
| 18   | Number of fertilized eggs                                | mil    | 2.549  | 3.129  | 3.736  |
| 19   | Hatch percentage   | %      | 75.2   | 73.6   | 76.9   |
| 20   | Hatched larvae   | mil    | 1.9169 | 2.303  | 2.8726 |
| 21   | Incubation survival                                      | %      | 94.1   | 95.3   | 95.8   |
| 22   | Larvae of 7 to 8 days viable                             | mil    | 1.8038 | 2.1947 | 2.752  |
| 23   | Larvae 7 - 8 days / ♀ matured                            | mil    | 0.164  | 0.1829 | 0.1835 |
| 24   | Larvae 7 - 8 days / kg ♀<br>matured                      | mil    | 0.081  | 0.0814 | 0.0852 |
| 25   | Fertilized eggs survival of larvae 7 - 8 days percentage | %      | 64.1   | 64.6   | 67.6   |

Tabel 6. The main technological indicators in 2020

| C.No. | Biotechnological Indicators                                 | M.U.   | V1            | V2                   | V3            |
|-------|---|--|---------------|----------------------|---------------|
| 1     | Pond  | -  | В 1-3         | В 4-6                | В 7-9         |
| 2     | Surface   | ha   | 0.3           | 0.3                  | 0.3           |
| 3     | Number of mattresses installed                              | -  | 15            | 15                   | 15            |
| 4     | Families number   | -  | 15            | 15                   | 15            |
| 5     | Gender relation   | \$\\$  | 1/1           | 1/1                  | 1/1           |
| 6     | Females characteristics                                     | nr.ex/g/ex   | 15/2170       | 15/2183              | 15/2177       |
| 7     | Males characteristics                                       | nr.ex/g/ex   | 15/2003       | 15/1913              | 15/1917       |
| 8     | Female hormonal stimulation                                 | -  | -             | Pituitary<br>hormone | Nerestin 5A   |
| 9     | Dosage  | mg/kg body weight                                  | -             | 3.5                  | 0.15          |
| 10    | Reproduction period   |  | 13-17.04.2020 | 13-17.04.2020        | 13-16.04.2020 |
| 11    | Reproductive water<br>temperature                           | ° C  | 11-16         | 11-16                | 11-16         |
| 12    | Collected nets  | no   | 11            | 11                   | 14            |
| 13    | Maturated females   | ex/g/ex  | 11/2170       | 11/2183              | 14/2177       |
| 14    | Maturation percentage                                       | %  | 73.3          | 73.3                 | 93.3          |
| 15    | Average prolificacy   | thousands eggs / $\stackrel{\bigcirc}{\downarrow}$ | 273420        | 275060               | 274300        |
| 16    | Eggs for incubation   | mil  | 3.0076        | 3.0257               | 3.8402        |
| 17    | Fertilization rate  | %  | 91.3          | 90.2                 | 90.1          |
| 18    | Number of fertilized eggs                                   | mil  | 2.746         | 2.729                | 3.460         |
| 19    | Hatch percentage  | %  | 71.1          | 73.2                 | 74.4          |
| 20    | Hatched larvae  | mil  | 1.9524        | 1.9977               | 2.5743        |
| 21    | Incubation survival   | %  | 95.2          | 94.4                 | 95.3          |
| 22    | Larvae of 7 to 8 days viable                                | mil  | 1.8587        | 1.8859               | 2.4533        |
| 23    | Larvae 7 - 8 days / $\stackrel{\bigcirc}{_{+}}$ matured     | mil  | 0.1690        | 0.1714               | 0.1752        |
| 24    | Larvae 7 - 8 days / kg ♀<br>matured                         | mil  | 0.0779        | 0.0785               | 0.0805        |
| 25    | Fertilized eggs survival of<br>larvae 7 - 8 days percentage | %  | 61.8          | 62.3                 | 63.9          |

### Discusions

### 1. Spawners maturation percentage

• the best maturation percentage was obtained in 2019 in V3 (100%), and the lowest was obtained in 2018 in V1 (66.7%);

• in 2018 the highest maturation percentage was obtained in V3 (93.3%) and the lowest in V1 (66.7%), respectively 80% in V2;

• in 2019 the highest maturation percentage was obtained in V3 (100%) and the lowest in V1 (66.7%), respectively 80% in V2;

• in 2020 the highest maturation percentage was obtained in V3 (93.3%) and the lowest in V1, respectively V2 (73.3%).

Figures 6 and 7 show the variation of spawners maturity percentage on experimental variants and by years.

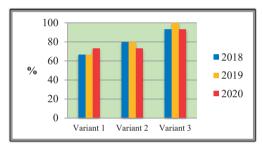


Figure 6. Variation of maturation percentage by experimental variants

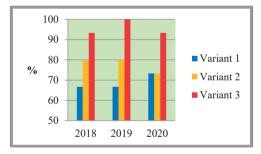


Figure 7. Variation of the maturation percentage per year

### 2. The total number of obtained eggs

• the highest eggsnumber was obtained in 2019 in V3 (4.0692 million), and the lowest number was obtained in 2018 in V1 (2.6649 million);

• in 2018, the highest eggs number was obtained in V3 (3.9043 million), and the lowest number was obtained in V1 (2.6464 million), respectively 3.3214 million in V2;

• in 2019, the highest eggs number of obtained in V3 (4.0692 million), and the lowest number was obtained in V1 (2.8136 million), respectively 3.3975 million in V2;

• in 2020, the highest eggs number was obtained in V3 (3.8402 million), and the lowest number was obtained in V1 (3.0076 million), respectively 3.0256 million in V2.

• Figure 8 shows the eggs number variation obtained per year and experimental variants.

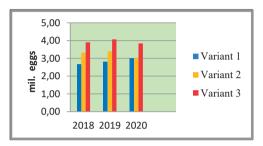


Figure 8. The eggs number variation obtained per year and experimental variants

#### 3. Eggs fertilization percentage

• the best fertilization percentage was obtained in 2019 in V2 (92.1%), and the lowest percentage was obtained in 2018 in V1 (88.1%);

• in 2018 the best fertilization percentage was obtained in V3 (92.0%), and the lowest in V1 (88.1%), respectively 89.4% in V2;

• in 2019 the best fertilization percentage was obtained in V2 (92.1%), and the lowest in V1 (90.6%), respectively 91.8% in V3;

• in 2020 the best fertilization percentage was obtained in V1 (91.3%), and the lowest in V3 (90.1%), respectively 90.2% in V2.

Figure 9 shows the fertilization percentage variation by years and experimental variants.

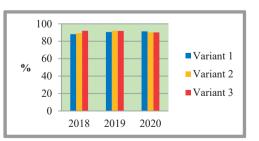


Figure 9. The fertilization percentage variation by years and experimental variants

#### 4. Eggs hatching percentage

• the best hatching percentage was obtained in 2018 in V3 (77.8%), and the lowest percentage was obtained in 2020 in V1 (71.1%);

• in 2018, the best hatching percentage was obtained in V3 (77.8%), and the lowest in V1 (74.3%), respectively 74.8% in V2;

• in 2019, the best hatching percentage was obtained in V3 (76.9%), and the lowest in V2 (73.6%), respectively 75.2% in V1;

• in 2020, the best hatching percentage was obtained in V3 (74.4%), and the lowest in V1 (71.1%), respectively 73.2% in V2.

Figure 10 shows the hatching percentage variation by years and experimental variants.

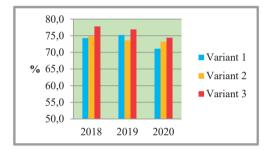


Figure 10. Hatching percentage variation by years and experimental variants

# 5. The total number of obtained 7 - 8 days larvae

• the highest number of 7-8 days old larvae of was obtained in 2019 in V3 (2.7519 mil.), and the lowest number of 7-8 days larvae was obtained in 2018 in V1 (1.6851 mil.);

• in 2018, the highest number of 7-8 days larvae was obtained in V3 (2.7415 mil.), and the smallest number of 7-8 days larvae was obtained in V1 (1.6851 mil.), respectively 2.1100 mil. in V2;

• in 2019, the highest number of 7-8 days larvae was obtained in V3 (2.7519 mil.), and the smallest number of 7-8 days larvae was obtained in V1 (1.8038 mil.), respectively 2.1948 mil. in V2;

• in 2020 the highest number of 7-8 days larvae was obtained in V3 (2.4533 mil.), and the smallest number of 7-8 days larvae was obtained in V1 (1.8587 mil.), respectively 1.8858 million in V2.

Figure 11 shows the 7-8 days larvae number variation per year and experimental variants.

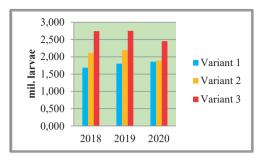


Figure 11. The number of days 7-8 larvae

# 6. Number of 7-8 days larvae/kg matured female

• the highest number of 7-8 days larvae/kg matured female was obtained in 2019 in V3 (0.0852 mil.), and the lowest number of 7-8 days larvae o/ kg of matured female was obtained in 2020 V1 (0.0779 million);

• in 2018, the highest number of 7-8 days larvae / kg of matured female was obtained in V3 (0.0885 million), and the lowest number was obtained in V1(0.0797 million), respectively 0.0800 million in V2;

• in 2019, the highest number of 7-8 days larvae / kg of matured female was obtained in V3 (0.0852 mil.), and the lowest number was obtained in V1 (0.0810 mil.), respectively 0.0814 million in V2;

• in 2020, the highest number of 7-8 days larvae / kg of matured female was obtained in V3 (0.0805 mil.), and the lowest number was obtained in V1 (0.0779 mil.), respectively 0.0785 million in V2.

Figure 12 shows the 7-8 days larvae/kg female matured number variation per year and experimental variants.

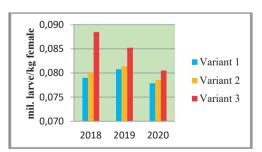


Figure 12. The number of 7-8 days larvae/kg female matur ed

# 7. Survival rate from fertilized egg stage to 7 - 8 day larval stage

• the best survival percentage was obtained in 2018 in V3 (70.2%), and the lowest percentage was obtained in 2020 in V1 (61.8%);

• in 2018, the best survival percentage was obtained in V3 (70.2%), and the lowest percentage was obtained in V1 (63.2%), respectively 63.5% in V2;

• in 2019, the best survival percentage was obtained in V3 (67.6%), and the lowest percentage was obtained in V1 (64.1%), respectively 64.6% in V2;

• in 2020, the best survival percentage was obtained in V3 (63.9%), and the lowest percentage was obtained in V1 (61.8%), respectively 62.3% in V2.

Figure 13 shows the survival rate variation from fertilized eggs to 7-8 days larvae per year and experimental variants.

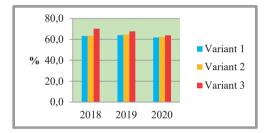


Figure13. The survival rate variation from fertilized eggs to 7-8 days larvae

# CONCLUSIONS

The technology of conttroled pikeperch reproduction, practiced at Nucet Research Center, includes the following successive stages: in the spring beginning, when the water temperature reaches the value of 8-10°C, the spawners are fished from the winter ponds and are separated by sex. Next, they are introduced into the prematurity basins, where they are kept for about 3-4 weeks, depending on the water temperature. At a 10-12°C water temperature, the advanced maturation stage females are hormonally stimulated (V1 and V2 variants). The unstimulated females from V1 variant and all the males are parked separately in maturation ponds. After the second dose of the hormone administered to the females, the spawners are introduced into natural-directed reproduction ponds, where the laying eggs mattresses have previously been introduced. The mattresses with fertilized eggs are collected and placed in "Nucet" type incubators in the breeding station. The advantage of this method is that the pikeperches are not manipulated when they are spawning their sexual products.

The best results were obtained in the synthetic hormone stimulation variant with (Nerestin 5A), in each of the three research years of (2018-2020). The females maturation percentage was 95.6%. The survival rate of larvae up to the age of 7-8 days was between 63.9% (2020) and 70.2% (2018). In this experiment, were also obtained good results in the variant without hormonal stimulation (V1), the survival rate of larvae up to the age of 7-8 days being between 61.8% (2020) and 64.1% (2018).

Due to the fact that the eggs were incubated in an optimal temperature range (11- $15^{0}$ C), the hatching time was relatively short, resulting in homogeneous groups, which was later reflected in the results obtained in larval growth.

The choice of the hormone is made depending on the technological indicators obtained, but also on the price of the product. Nerestin 5A is a synthetic hormone, accessible on the market, easy to get unlike the carp pituitary hormone which is much more expensive and difficult to procure.

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