University "Dunărea de Jos" from Galați Doctoral School of Mechanical and Industrial Engineering



DOCTORAL THESIS

Reproduction and rearing of walleye (*Sander lucioperca,* L., 1758) **in industrial aquaculture systems**

SUMMARY

Doctorand, Ing. DOBROTĂ GHEORGHE

Scientific leader

Prof. univ. Emerit dr. ing. CRISTEA VICTOR

Corresponding member of the Academy of Agricultural and Forestry Sciences

Secția I 4: Inginerie Industrială nr. 87 Galați 2022

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INTRODUCTION, THE OPPORTUNITY OF THE APROACHED SUBJECT

Aquaculture is the sector which targets the production of aquatic animals and plants for commercial purpose. In the last three decades, aquaculture registered the fastest growth on a global scale, compared to the rest of the agriculture sectors. The latest observed tendency in the aquaculture sector is to increase the production and to diversify products.

Given the fact that fish catches from the natural aquatic environment register stagnation, and many fish species are negatively influenced by overfishing and pollution, the development and introduction of new production species with economic value (sturgeons and other raptor species) in aquaculture systems has been imposed.

Romania has a legacy for practicing aquaculture in earthen pond production systems in most fish farms, therefore, the orientation towards these systems is inherent. Thus, new technologies can be applied within the aforementioned systems without the need to supplement with additional infrastructure investments, which consist 60% of the total investment cost.

Nowadays, an increased tendency for aquaculture products consumption and the reorientation of consumers towards species with economic value (such as sturgeon, trout, pikeperch etc.) had been observed. However, the national production does not cover this demand.

Through the approached subject, the present doctoral thesis contributes to the development of know-how, being the first research in Romania which approaches the assisted natural reproduction, fry development and first summer rearing of pikeperch. In this context, the thesis main objective was to elaborate a model for pikeperch fry production which can be delivered to fish farmers as a new, easy to implement technology in Romanian farms.

The main purpose of the present thesis is the elaboration of assisted natural reproduction technology, the rearing and development in the fry stage technology and the rearing of one summer old pikeperch (*Sander lucioperca*, Linaeus, 1758), in industrial aquaculture systems.

The final objective was to obtain biological material which can be populated without risks in the natural waters and multi-purpose ponds. The chosen theme complies with the characteristics of an applicative research, which can offer answers to questions raised during the last period of practicing pikeperch reproduction and rearing technologies in the stage of fry and one summer old, in order to obtain population material. In the light of the aforementioned, the main objective of the present thesis was to obtain useful data related to the most complex stages within the production process of pikeperch (assisted natural reproduction, fry development and one summer old rearing), with the possibility of knowledge technological transfer to the Romanian fish farms. The clarification of assisted natural reproduction technology, larval and fry development and fingerlings rearing technologies will allow the improvement of the technological and operational management in production unitswhich target to intensively produce this specie. In order to affirm that the technological coordinates for the production of a biologically valuable material (quantitative and qualitative) are known, the technologies for the assisted natural reproduction, fry development and one summer old rearing of pikeperch must be rigorously substantiated.

The present thesis is structured within six main chapters, as it follows:

The first chapter approaches "General data related to the aquaculture sector" the state of the art of global and national aquaculture, general data related to the evolution of

the quantity of commercialized aquatic products, as well as their origin (fish catches or aquaculture).

The second chapter approaches "Material base, methods and working techniques" which were utilized along the experimental analysis, the description of used equipment and calculus techniques that generated the results obtained within the present thesis. The experimental part of the thesis is categorised in three main steps, presented in chapters three, four and five.

Chapter three is entitled "Experimental protocols related to the elaboration of assisted natural reproduction technology of pikeperch (Sander Iucioperca, Linaeus, 1758)", and it describes the achievement of assisted natural reproduction of pikeperch without and with female hormonal stimulation, with two types of hormons (natural – carp pituitary gland and artificial – Neristin 5A).

Chapter four of the thesis "The elaboration of fry development and rearing technology for pikeperch (*Sander lucioperca*, Lineaus, 1758), in intensive system" was channelled towards two directions, respectively: the influence of stocking density and the administered type of feed. The experiments were conducted in 3 different rearing seasons during the years 2018, 2019 and 2020 in "Evos" tanks, in protected spaces and in triplicate, in order to obtain conclusive results.

The main objective of this stage was to elaborate the technology for fry development and rearinf and the secondary objective were as it follows:

- 1. The determination of optimum density for fry development and rearing in the postembryonary stage, in protected spaces, such that at the end this density will give information related to superior performance for growth rate, feed conversion efficiency, survival percentage, population homogeneity etc.
- 2. The identification of optimum feed for this predator specie in this stage such as a positive correlation between growth rate and feed conversion efficiency is obtained, as well as the registration of superior performance indexes.

Chapter five of the thesis presents "Experimental protocols for the elaboration of the rearing technology of one summer old pikeperch Sander lucioperca-L., 1758, in earthen ponds" realised within the experimental Base at the Fish Culture Research and Development Station of Nucet, which had the main objective to elaborate the rearing technology of one summer old pikeperch (*Sander lucioperca*, L. 1758), in earthen ponds, in monoculture, with the purpose to obtain one summer old population material which has a proper size (is no longer a target for predators and is capable of obtaining food) in order to restock natural fish populations and the introduction as a specie in aquaculture

The last chapter of the thesis is represented by "Final conclusions and personal contributions" which were drawn after the approach of the assisted natural reproduction technology, the fry development and rearing technology and one summer old pikeperch rearing technology, by presenting the achieved objectives and those that were not fully achieved.

Through the obtained results, the fundamental and experimental research will contribute to the thorough knowledge of the specie biology of *Sander lucioperca* in general, as well as the reproduction technologies, fry development and one summer old pikeperch technologies. By

realising the proposed activities, important scientific results were obtained, at the fundamental level (the elaboration of assisted natural reproduction model, fry development and rearing model and one summer old rearing model, the elucidation of aspects related to cannibalism, the highlighting of feeding physiological mechanisms on stress) as well as the technological level (the elaboration of assisted natural reproduction, fry development and rearing, one summer old pikeperch rearing), which will contribute to the development of national knowledge in the aquaculture sector.

By synthetizing the bibliographic information related to the actual situation and the principal strategic directions undertaken in percidae rearing in general and in pikeperch rearing in special, it can be concluded that this specie presents a high potential for future exploitation and the growth performances obtained in the present, as well as those forecasted for the future, confirm the national and international interest manifested for this species.

CHAPTER I. GENERAL DATA REGARDING AQUACULTURE

Our country now has the lowest fish consumption per capita in the European Union (5 kg fish/capita/year). By comparison, in Portugal 76 kg of fish/capita/year are consumed, in Belgium -25 kg, in Croatia 20 kg, in the Czech Republic -9.5 kg, in France -35 kg, in Germany -14 kg, in Greece -20 kg, Italy –25 kg, Poland –12 kg, Spain –60 kg and Great Britain –19 kg. Icelanders consume the largest amount of fish in Europe per capita, 90 kg/year, and Japan consumes over 130 kg/year, this consumption being natural in Japan, because it is a peninsular area, it is in first place in world.

Romania has the potential and resources to increase fish production from aquaculture, under the conditions in which an economic environment compatible with that of the EU countries will be created. Capitalizing on the exceptional potential of aquaculture in the European context, harmonized with an appropriate legal and institutional framework, will increase the competitiveness of Romanian aquaculture.



Figure 1 (a-b). a) Research-Development Station for Fish Culture Nucet (original photo) b) Pilot station for the reproduction of fish species (original photo)

CHAPTER II. RESEARCH INFRASTRUCTURE

The entire experimental research activity was carried out at the Nucet Fisheries Research-Development Station and was carried out during the three years of study (2018, 2019, 2020) within the doctoral program, in the Experimental Base number one of Nucet, equipped with: pilot stations for reproduction, incubation and post-embryonic development of fish, rearing, prematurity, maturation, wintering and naturally-directed breeding pools and laboratories for: monitoring of environmental conditions, animal welfare and genetics and fish breeding (figure 1a).

CHAPTER III. NATURALLY-DIRECTED REPRODUCTION OF PIKEPERCH

3.1. The objective

The experimental works regarding the development of the reproduction technology of the pike (Sander lucioperca, Linneaus-1758) started from the existing reality, created by the new context in Romania, by its influence on the fishing sector and by the visible changes that appeared in the climate in Romania, with tendencies of heating and high temperatures, even during the reproduction period of predatory fish, including the shad.

The main objective of this experiment was to achieve natural-directed reproduction in the saddle, as well as to establish the type of hormone, which would have the maximum effect for achieving ovulation in ponds conditions, with minimal numerical losses.

There were two directions in which the experimental works were carried out, namely:

- natural-directed reproduction of the stallion without homonal stimulation of the females;
- natural-directed reproduction of the saddle with the stimulation of maturation and ovulation through the use of hormonal preparations.

For the induction of maturation and ovulation in the two variants of naturally-directed reproduction of the shad (Sander lucioperca, Linneaus-1758), the following hormonal products were used in the experiments: carp pituitary gland and Nerestin 5A, a commercial preparation of LHRH type.

Nerestin 5A is an LHRH hormone preparation and is part of a wider range of preparations intended exclusively for fish, and the 5A variant is generally used for sturgeon. The product is presented in liquid form, in 20 ml bottles, which contain 100 doses (0.2 ml/kg body).

The specific objectives of the experimental works

The experimental works for the development of the technology of natural-directed reproduction of pikeperch were carried out according to the following work stages:

- establishing the experimental variants;
- establishment and preparation of breeding pools;
- preliminary evaluation of breeders;
- preparing the mattresses and installing them in the breeding pools;
- fishing of breeders from prematurity pools and their assessment;
- stimulation of females before introduction into breeding pools;
- the introduction of breeders into breeding pools;
- follow-up of the bridge deposit;
- sampling the mattresses and introducing the nests with fertilized eggs into the incubators;
- evaluation of the quantity of eggs deposited in the cubes introduced for incubation;
- determining the fertilization percentage;
- following the process of incubation and hatching of the larvae;
- determination of hatching percentage;

- ensuring optimal conditions during the larval development period until the moment of packaging (delivery) of the larvae;
- evaluation of 7-8 day old larvae, their packaging and transport.

3.2. Experimental design

The saddle is a species in which artificial reproduction is very difficult, due to the particular sensitivity to manipulations and traumas, having as a direct consequence the recording of breeding mortality rates of up to 50%. In order to minimize these losses, naturally-directed breeding has been resorted to, breeding that involves less manipulation, which leads to less stress on breeders and higher survival.

The naturally-directed reproduction of the saddle is carried out as follows: the breeders in the advanced stage of maturation are introduced into breeding pools to deposit the eggs on the mats, after which they are harvested and incubated in breeding stations, in "Nucet" type incubators. It presents the advantage that the breeders are not manipulated and traumatized, at the time of harvesting the sexual products, the losses of breeders are minimal.

The groves where the natural-directed reproduction of the saddle takes place must have a maximum surface area of 1000 m², a depth of 1.5-2.0 m and be completely devoid of vegetation. Because the saddle is very sensitive to handling and transport, it needs a longer period of adaptation to a new habitat and careless handling when populating breeding pools can lead to their inhibition, failure to lay eggs or even the loss of breeders. That's why it is recommended that the wintering of saddle breeders should be done near the heleste intended for reproduction. Spawning and fertilization takes place on special mats made of willow root whiskers. The norm of the population of the reproduction helešte is one family per 40-50 m² and one mattress is installed for each family. A family consists of a female and 1 or 2 males. The mattresses are installed in the sled at a water temperature of 12-14°C and checked daily. The nests with the mattresses with embryonated eggs are transferred to incubators fed with a water flow of 3-4 l/min. After hatching, the larvae remain in the incubator for 4-5 days, after which they are transferred to the nursery, in the parking basins, where they are fed with algae and rotifers. At the age of 7-9 days, the larvae are suitable for packaging (in polyethylene bags, with water and oxygen under pressure) and transported to their destinations.

3.2.1. Establishment of experimental variants

The variants established for the experimental works of natural-directed reproduction of pikeperch (Sander lucioperca, Linneaus-1758) were:

- reproduction without hormonal stimulation of females (control variant);
- reproduction with hormonal stimulation of females with freeze-dried carp pituitary (CPE);
- reproduction with hormonal stimulation of females with Nerestin 5A.

The males weren't hormonally stimulated. The period of the experimental works was in the years 2018-2019-2020.

3.2.2. Installing the mattresses and introducing the pikeperch spawners into the breeding ponds

Before the introduction of saddle breeders for breeding, mats with bundled willow roots ("whiskers" - Salix babylonica) were installed in the nine pools, for depositing the seminal elements, 5 mats/pool. The mattresses were installed at a distance from the shore of approximately 2.5 m (figure 2 a-b), with a distance between them of 8-10 m, 5

mattresses/breeding pool, taking into account that for 1.0 ha pool of reproduction, the population norm is relative, 50 families/ha can be introduced.

After installing the mattresses, the established work schedule was as follows:

- fishing of the pikeperch spawners from prematurity pools and temporary parking in maturation ponds;
- evaluation of the reproductive status of females;
- introducing them into the breeding pools as follows:
 - females without hormonal stimulation in pools B1, B2 and B3;
 - females with hormonal stimulation by injection with lyophilized carp pituitary gland (3.5 mg/kg body) in pool B4, B5 and B6;
 - females with hormonal stimulation with Nerestin 5A (0.15 ml/kg body) in basin B7, B8 and B9;
- fishing, injection and introduction into the breeding pools was done in the morning in the time interval 600-800, and the water temperature had values between 12.0-14.0°C during the three years of the study.

After evaluating the reproductive status, females that are in the advanced stage of maturation (grade IV) were selected for breeding. They also launched themselves into the breeding pools and the males, which produced a few drops of sperm upon a light massage of the abdomen. Another criterion for choosing the breeders, in addition to the degree of maturity, was the homogeneity of the batch in terms of waist.

3.2.3. Establishing hormone doses

To stimulate maturation and ovulation in the saddlefish, hormones from the carp pituitary gland and Nerestin 5A were used, the latter preparation being used for the North American sturgeon Polyodon spathula. We selected these hormones because the determining factor was taken into account, namely the reproduction temperature, which is very close in the two species.



Figure 2 (a-b). Installation of mattresses for directed reproduction of the saddle (original photo)

Depending on the mentioned considerations, the doses of exogenous hormones that were used in the experimental works on the reproduction of the shad (Sander lucioperca, Linneaus-1758) were:

- freeze-dried pituitary gland of carp: total dose 3.5 mg/kg female body (2 doses at 12-hour intervals, 0.35 mg/kg body preparatory dose and 3.15 mg/kg body decisive dose);
- Nerestin 5A: total dose 0.15 ml/kg female body (2 doses at 12-hour intervals, 0.015 ml/kg body preparatory dose and 0.135 ml/kg body decisive dose);

Doses of exogenous hormones used in the naturally-directed reproduction of the pike species (Sander Iucioperca, L., 1758) in the years 2018-2019-2020 are presented in table no. 1.

Hormone used	Females total dose			
normone used	Recommended	Used		
Carp pituitary (mg/kg body)	2,0-5,0	3,5		
Nerestin 5A (ml/kg body)	0,12 - 0,3	0,15		

Table no. 1. Doses of exogenous hormones used in the experimental works of natural-directed reproduction of the species (Sander Iucioperca, L. 1758)

Before the hormonal treatment, biometric measurements (figure 3 a-b) were performed for each specimen of reproductive saddle (\bigcirc + \bigcirc), determining the amount of hormonal substance to be injected.

3.2.4. Scheduling and administration of hormonal injections

The technology of reproduction, in fish in general and in salmon in particular, usually involved two injections for the females with the chosen hormonal preparation.

The initial (preparatory) injection is separated from the second (decisive) dose by an interval of 12-24 hours depending on the species and water temperature.

In the experimental works, the programming of the injections was carried out according to the stage of maturation, the physiological state and the state of health of the breeders. For females, dose I (preparatory) represented 10% of the total dose, and dose II (decisive) the difference of 90%.

The injection schedule of saddle females was as follows:

• 2000: fishing and selection of females from prematurity pools (parking lot) with the application of dose I (preparatory) and introduction into the maturation pools;

• 2000: fishing and selection of males from prematurity pools (parking) and introduction into maturation pools (all specimens in one pool);

• after 12 hours (8:00 a.m.), fish the females from the maturation pools, apply the hormonal injection, the second dose (decisive) and re-introduce them to the reproduction pools. During the ripening period, the water temperature fluctuated between 11-14°C, and the amount of dissolved oxygen did not drop below 6-8 mg/l.





Figure 3 (a-b). Biometric measurements at pikeperch (original photo)

Figure 4. Application of the injection for hormonal pikeperch stimulation to saddle females (original photo)

In the experiments carried out, taking into account the experience accumulated over time in breeding works for carp, Asian cyprinids, sturgeon (Polyodon spathula), underwater injection was practiced as a working method for the hormonal stimulation of pikeperch spawners, the breeders being introduced during injection into tarpaulin stretchers with water. The injection was made after the second dorsal fin (figure 4). For the most correct application of the injection, the syringe needle was inserted at an angle of 45°, and when it was withdrawn, the operator pressed his finger on the puncture site for a few seconds, to prevent the phenomenon of counter pressure, which could have led to elimination of the injected solution, after which, as an additional measure, a light massage was applied to the injection site.

With the first dose to the females, males were fished and selected from the prematurity (parking) pools and introduced into the maturation pools (all specimens in one pool). After the second dose of females, males were fished out of the maturation ponds and introduced into the natural-directed breeding ponds

3.3. Results and discussion

3.3.1. Determining prolificacy and introducing spawners into spawning ponds

In order to determine the average prolificacy of , pikeperch females selected in order to carry out experimental natural-directed reproduction works, biometric measurements were performed regarding individual weight, total length, circumference, etc.

For each experimental variant, the gonado-somatic ratio (RGS) was calculated in females; its average was 10% of body mass.

According to (Bucur C. et al., 2006) and according to their own determinations made during the three years of experimental work, the number of eggs/g was set at 1200-1320 eggs/g. For the calculation of the fertility of the females introduced for reproduction, the average of the two values of 1260 eggs/g was taken into account.



Figure 5 (a-b). Female pikeperch breeder fished from the prematurity pools (parking lot) introduced into the breeding pool (original photo)

A number of 90 breeders were selected for the experimental works, of which 45 were females and 45 were males. The 45 females were divided into three lots of 15 females/lot, and before the temporary parking in the prematurity pools, the following works were performed:

• a batch of 15 females without being hormonally stimulated for breeding pools B1, B2 and B3;

• a group of 15 females was hormonally stimulated with carp pituitary in a total dose of 3.5 mg/kg body for breeding pools B4, B5 and B6;

• a batch of 15 females was hormonally stimulated by injection with Nerestin 5 in a total dose of 0.15 ml/kg female body for breeding pools B7, B8 and B9.

Before breeding, the three batches of females (15 ex/lot) were parked separately in maturing pools. The males were all parked in a maturation pool without being hormonally stimulated. The distribution in these basins was done in the evening, in the time interval 1900-2100, at a water temperature of 11-16°C. On the following day, females from variants 2 and 3 were injected with the first dose of pituitary/synthetic hormones, and the second dose was injected 12-14 hours later. Simultaneously with the second dose, the breeders were introduced into the breeding pools (5 Q+5 d /pool, figure 5 (a-b)). The sex ratio was 1/1.

3.3.2. Monitoring of the physico-chemical parameters of water in breeding ponds

At the time of introduction of the biological material for reproduction, determinations were made of the temperature and dissolved oxygen in the water, recordings continued throughout the reproduction.

During reproduction, there were no sudden changes in the environmental conditions in terms of large variations in the temperature and oxygen content of the technological water which would influence reproduction, in the sense of inhibiting, accelerating or slowing down the duration of spawning in nests.

3.3.3. Observations on the directed reproduction of the saddle

From the observations made from the shore in the area of the breeding pools, it was found that after 12-24 hours, the breeding pairs were formed in the pool, by their presence above the mattresses, this being an indication that the moment of spawning is approaching.

As a rule, the laying of the litter took place in the early hours of the morning. The observations being made along the perimeter of the spawning pools from the shore, allowed the identification of the mats on which the spawn was deposited by following the breeding pairs that sat above the nests on the mats, the nests on which the eggs were laid being easy to identify, due to the transparency of the water and the depth of 70-80 cm at which they were installed, but also their location near the shore.

No.	Basin	Count the mattresses installed	Number of families	Variant	No. harvested mattresses	Mature females	Maturation percentage (%)
1	Total V1	45	45	V1	31	31	68,9
2	Total V2	45	45 45 V2 35		35	77,8	
3	Total V3	45	45	V3	43	43	95,6

 Table no. 2. The results obtained in the natural-directed reproduction of the shad regarding the number of nests with fertilized shad eggs harvested in the three experimental variants/year

where: V1 - no hormonal stimulation; V2 - carp pituitary hormone stimulation (CPE); V3 - hormonal stimulation with Nerestin 5A

After laying the eggs on the mats (figure 6 a-b), the nests with fertilized eggs were harvested by carefully detaching them with a knife from the nytal net of the mat, placed in trays with water and taken to the incubation station.

Since spawning was not done at once by all females, after spawning, the nests with eggs were removed as quickly as possible, so as to avoid infestation with fungi and other pests in the pools. Also, the operation of removing the mattresses from the water, detaching the nests and transporting them to the station in order to put them in the incubators, was carried out quietly

and with great care (saddle eggs are very sensitive), avoiding stressing the breeders who did not lay bridge



Figure 6 (a-b). Detachment of nests with fertilized roe and nest of fertilized roe before being introduced into the Nucet incubators (original photo)

In table number 2 shows the results obtained during the natural-directed reproduction of the salla concerning the number of nests with fertilized eggs, harvested in the three experimental variants/year, as well as the percentage of females maturing.

3.3.4. Incubation eggs

After harvesting the nests with fertilized eggs from the breeding pools, they were very carefully introduced into the nytal boxes in the Nucet incubators (figure 7 a-b), where a permanent supply of water was ensured, at a flow rate of 8 l/min. The capacity of the Nucet incubator is 140 litres, surface feeding and bottom emptying, it creates a vertical circular water current, providing the spawn with fresh, well-oxygenated water continuously.

The technological water supply of the breeding station is carried out by gravity, with spring water from a settling basin, filtered through 0.05 mm nytal filters.

The incubation period was 6-8 days depending on the water temperature.

3.3.5. Monitoring the physico-chemical parameters of the water during the incubation period

During embryonic and larval development, there were no sudden changes in the environmental conditions, in terms of large variations in the temperature and oxygen content of the technological water, which would influence the normal development of the embryo in the sense of accelerating or slowing down the duration of embryogenesis, so that the number of abortions found after hatching was almost non-existent, or to trigger mortalities during the larval period.

Water quality monitoring, by studying the dynamics of physical-chemical parameters, is particularly important, whose values within normal limits lead to the creation of a favorable environment for embryonic and larval development. At the same time, when deviations from normal values are observed, in one or more parameters, specific measures can be taken, so that the situation returns to normal.

During the embryonic and larval development of the shad, the environmental conditions were monitored by collecting samples at intervals of 3-4 days, the water temperature was determined at three hours (hours 3:00, 6:00, 9:00, 12:00, 15:00, 18:00, 21:00 and 24:00) and the concentration of dissolved oxygen daily or whenever needed, acting through intervention measures such as running water, so that the value of this parameter did not fall below the limit of 5-6 mg/l.



Figure 7 (a-b). Nests with fertilized roe in the Nucet hatchery (original photo)

3.3.6. Embryonic development

From the specialized data and from the observations made, it could be established that the duration of the embryonic development process in the saddle directly depends on the temperature of the technological water at which the embryonic development takes place..

In the 2018-2019-2020 experimental variants of natural-directed reproduction of the shad, embryonic development took place at water temperatures of 12.0-15.0°C, the average duration of this process being 167 hours, respectively 7 days (2019), minimum duration 144 hours/6 days (2020), maximum 187 hours/7-8 days (2018).

From the macro/microscopic observations, it was found that, shortly after fertilization (1-2 hours), the diameter of the roe varies between 1.2 and 1.4 mm. After 48-50 hours, under the conditions of embryonic development at water temperatures of 12-14°C, the embryo is in the gastrula stage.

After 70-72 hours, the formation of the body of the embryo (in which 21 myomeres can be distinguished) and which half surrounds the yolk sac was observed. The eyes are slightly pigmented, the heart and blood circulation can be seen.

After 4-5 days, a visible growth of the embryo became visible with the naked eye, whose body surrounds the yolk sac almost one and a half times, microscopically about 34 myomeres are visible. The eyes of the embryo are pigmented and have a brown color. The primordia of the nasal orifice, the heart and the blood circulation can be distinguished, and inside the egg it is easy to observe the wriggling movements of the embryo, a sign that the moment of hatching is close.

After the 7th to the 8th day, at the time of hatching, it was observed that the embryo surrounds the yolk sac twice, the eyes are strongly pigmented. With binoculars, small brown chromatophores were visible in the head area, on the yolk sac and to a lesser extent on the rest, while the digestive tube, the pink heart and blood and the fin crease were also visible. Figure 8 shows the larva of the saddle before hatching.

An aspect of great importance that was monitored during incubation was the prevention of the emergence and infestation of fungi. During the incubation period, the eggs were bathed with a 37% formaldehyde solution (concentration 1.0-1.8 ml formaldehyde /1 liter of water) to prevent the appearance and infestation with fungi. The first treatment was administered 24 hours after introducing the eggs to the incubator. The exposure time depended on the water temperature (10 min at a water temperature of 10-12°C or 15 min at a water temperature of 13-15°C). The process was repeated every 12 hours, until the embryo surrounded the entire yolk sac, the tail reached the eyes, the pigmentation is accentuated, the movements of the embryo became more intense and the heart pulsations were observed.

The incubation period was 6-8 days at the average daily water temperature of 12-15°C.



Figure 8. Larva of sloe before the moment of hatching (original photo)

No	Biotechnological indicators	Total average on experimental variants			
		V1	V2	V3	
1.	Number of families	45	45	45	
2.	Sex ratio	1/1	1/1	1/1	
3.	Females	45/97,9	45/96,4	45/97,8	
4.	Mature females	36/77,3	38/81,6	36/78,3	
5.	Maturation percentage	71,1	77,7	95,5	
6.	Average prolificacy	265230	278320	274820	
7.	Eggs for incubation	2,8287	3,2481	3,9379	
8.	Fertilization percentage	90	90,6	91,3	
9.	No fertilized eggs	2,548	2,942	3,596	
10.	Hatching percentage	73,5	73,9	76,4	
11.	Larvae to hatch	1,8712	2,1739	2,7472	
12.	Larvae 7- 8 day	1,7825	2,0635	2,6489	
13.	Larvae 7-8 day / ♀ mat	0,1672	0,1767	0,1848	
14.	Larvae 7-8 zile kg/ \cap mature	0,0794	0,08	0,0847	
15.	Survival percentage fertilized eggs/larvae 7 - 8 days	63	63,5	67,2	

Table no. 3. The total/average values of the technological indicators/experimental variants of natural-directed reproduction in the saddle in the years 2018-2019-2020

where: V1 - no hormonal stimulation; V2 - carp pituitary hormone stimulation; V3 - hormonal stimulation with Nerestin 5A.

After hatching, the remains of willow roots were removed from the Nucet incubators, the sallow larvae were kept in the incubators, depending on the water temperature, until the age of 7-8 days, during which time the hatchery feed rate was reduced to 4-5 l/minute. Taking into account the fact that the larvae are sensitive and swim hard, 2-3 frames of nytal were placed in the incubators for them to rest and rest.

The data regarding the comparative analysis of the total and average values of the main technological indicators realized in the experimental works of natural-directed reproduction of the saddle for each variant in the period 2018-2019-2020 is presented in tables no. 3.

3.3.7. Biotechnological indicators achieved

Maturation percentage of reproductives (figure 9a)

- 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 is 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%).

Total number of eggs obtained (figure 9b)

- the highest number of spawn 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 number of spawn was obtained in V3 (3.9043 million), and the lowest number was obtained in V1 (2.6649 million), respectively 3.3214 million in V2;
- in 2019, the highest number of spawn was 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 number of spawns was obtained in V3 (3.8402 mil.), and the lowest number was obtained in V1 (3.0076 mil.), respectively 3.0256 mil. in V2.



Figure 9 (a-b). a) Variation of the maturation percentage in female saddlefish – b) Variation of prolificacy (number of eggs/female)

Fertilization percentage of eggs (figure 10a)

> 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;

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

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

Egg hatching percentage (figure 10b)

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

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

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

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



Figure 10 (a-b). a) Variation of fertilization percentage - b) Variation of hatching percentage

Total number of 7-8 day old larvae obtained (figure 11a)

> the highest number of 7-8 day larvae was obtained in 2019 in V3 (2.7519 million), and the lowest number of 7-8 day larvae was obtained in 2018 in V1 (1.6851 million);

 \succ in 2018, the highest number of 7-8 day larvae was obtained in V3 (2.7415 million), and the lowest number of 7-8 day larvae was obtained in V1 (1.6851 mil.), respectively 2.1100 mil. in V2;

> in 2019, the highest number of 7-8 day larvae was obtained in V3 (2.7519 million), and the lowest number of 7-8 day larvae was obtained in V1 (1.8038 mil.), respectively 2.1948 mil. in V2;

 \succ in 2020, the highest number of 7-8 day larvae was obtained in V3 (2.4533 million), and the lowest number of 7-8 day larvae was obtained in V1 (1.8587 million .), respectively 1.8858 million in V2.

Number of 7-8 day old larvae/kg mature female (figure 11b)

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

> in 2018, the highest number of 7-8 day old larvae/kg mature female was obtained in V3 (0.0885 million), and the lowest number was obtained in V1 (0.0797 million), respectively 0.0800 mil in V2;

> in 2019, the highest number of 7-8 day old larvae/kg mature female was obtained in V3 (0.0852 mil.), and the lowest number was obtained in V1 (0.0810 mil), respectively 0.0814 million in V 2;

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

<u>The percentage of survival from the stage of fertilized spawn to larvae 7-8 days</u> (figure 12)

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

> in 2018, the best percentage of survival 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 percentage of survival was obtained in V3 (67.6%), and the lowest percentage was obtained in

V1 (64.1%), respectively 64.6% in V2;

 \succ in 2020, the best percentage of survival was obtained in V 3 (63.9%), and the lowest percentage was obtained in V1 (61.8%), respectively 62.3% in V2.



Figure 11 (a-b). a) Variation in the number of 7-8 day old larvae/kg mature female by years and experimental variants; b) Larvae of 7-8 days per kg mature female



Figure 12. Variation of survival percentage from fertilized spawn to larvae 7-8 days across years and experimental variants

CHAPTER IV. DEVELOPMENT OF POSTEMBRYONAL DEVELOPMENT TECHNOLOGY AT PERCH

4.1. The objective of the breeding period

The objective of the breeding period was to develop the technology of post-embryonic development of the pike species (Sander luciopercaLinneaus, 1758) in monoculture, in an intensive system, in protected spaces. This aims to obtain stocking material in summer I, with medium weights and sizes where the species is no longer the major target for predators and is able to easily obtain food, in order to restore natural populations and introduce and expand it in aquaculture.

The experimental work on the growth and development of the fry during the hatching period was carried out over a period of 3 years 2018-2019-2020, for 40 and 42 days respectively, in growth units suitable for the species, "Evos" type tubs.

The main biotechnological indicators followed were:

- ✓ population density (ex/m3);
- ✓ growth period (days);
- ✓ growth parameters: average mass (g/ex); average length TL (mm/ex);
- ✓ percentage of survival (%);
- ✓ feed conversion coefficient (FCR).

4.2. The technical-material basis

Intensive culture in protected spaces has the advantage that the growth environment can be fully controlled, from the point of view of environmental conditions, and the food administered regardless of the type (live food/supplementary feed) is available throughout the respective period, the administration is easy and it can be automated, drugs and biostimulators can be introduced. Diseases and parasites can also be controlled.

The experimental works on the growth and development of the flounder larvae during the hatching period were carried out in the pilot growth station in glass fiber tubs ("Evos tubs"), with a useful volume of 1000 liters, installed in the hatchery station no. 1. The "Evos" ponds are round or square in shape with rounded walls and are fed through an external pipe, the water falls freely from about 25 cm, thus facilitating oxygenation, and the evacuation is carried out centrally, creating a circular current. The optimal height of the water layer is 0.40-0.60 m. The water supply flow of the tubs was 7-15 l/minute, depending on the temperature. The evos ponds were colored blue and green (figure 13), colors that reduce the stress of the saddle in intensive growth systems (Grozea, 2016).

4.3. Establishment of experimental variants

The experimental works of growth and development in an intensive system of the saddle during the breeding period, were carried out in two directions, where:

I. the variable factor was "population density";

II. the variable factor was "type of food".

The degree of repeatability was 3 times, in triplicate and was carried out in 2018, 2019 and 2020.

In order to obtain the most conclusive results, the two experimental models of postembryonic development were carried out in the same premises (pilot station no. 1 within the Nucet Experimental Base), the water supply was done by gravity, with spring water from the river Ilfov, filtered through a 0.5 mm mesh nytal sieve, through a common PVC pipe, in "Evos" type tubs with a useful volume of 1000 liters.



Figura 13. Pilot plant ("Evos" basin) for the growth and development of shad larvae during the rearing period (original photo)

4.4. Experimental work on growth and development in an intensive system of shad (Sander lucioperca, L., 1758), during the breeding period, in different densities

4.4.1. The objective

The purpose of the rearing stage is that at the end of it, the chicks can be released into the natural environment or in anthropogenic pools for controlled growth, at a suitable size so that they can face the climatic and food risks and even predators. Consequently, the management of this stage is dependent, to a certain extent, on that of embryonic development, because it is absolutely necessary that the larvae, in the days following hatching, be populated in an environment rich in plankton of adequate size and devoid of predators.

The main objective of this experiment was to determine an optimal density of growth and development in the post-embryonic period, in protected spaces, so that there is a positive correlation between growth rate and feed utilization efficiency, as well as a high survival rate. Synthesizing the data presented from the specialized literature, the hypothesis of the experiment was outlined, namely that high growth densities influence the growth performance, produce stress on the biomass and lead to the occurrence of the phenomenon of cannibalism.

4.4.2. Experimental design

The "Evos" ponds were washed and disinfected with lime chloride solution, after which they were washed again with technological water and dried, and 12 hours before the time of population with larvae they were fed with water up to the level of 0.25-0.30 m. During the growth and development of the biological material, the technological water level was also increased, reaching 0.5-0.55 m on the 20th day of breeding.

Stocking density is a particularly important technological parameter for raising fish, in all stages of development, and is specific to the species, age and applied technology.

When establishing the population density, several factors were taken into account, such as:

- temperature and oxygen regime in the water;
- the behavioral characteristics of the crop species;
- the size that is aimed to be reached at the end of the breeding period;
- the quantity and quality of the administered food.

The work methodology considered the following aspects:

3 experimental variants were established in terms of population density;

- "Evos" type pools / experimental version: 3;
- degree of repeatability: 3;

• type of food, in 3 stages: natural food in a live state in stage I; mixed (live food/fodder) in stage II and with fodder in stage III;

• mode of administration: "ad-libitum", only during the day (12 hours), with permanent control of food consumption and monitoring of the main physico-chemical parameters of the technological water (temperature, oxygen content, pH, organic matter, etc. .);

duration of the breeding period: 40 days.

The experimental variants regarding population density were (table no. 4):

- variant 1 marked with V1: 1000 ex/basin;
- variant 2 marked with V2: 2000 ex/basin;
- variant 3 marked with V3: 3000 ex/basin.

No.	The experimental variant	Basin volume (I)	Ex. / basin	Nr. basin	Ex. total/year	No. years
1	V 1	1000	1000	3	3000	3
2	V 2	1000	2000	3	6000	3
3	V 3	1000	3000	3	9000	3

Table no. 4. Popularity of "Evos" tubs with 7-8 day old larvae

4.4.3. Results and discussions

4.4.3.1. Evaluation of the dynamics of water quality parameters from the source and from the growth modules

During the period of growth and development of the brine shrimp in the "Evos" ponds, the environmental conditions were monitored by taking samples at intervals of 3-4 days, and the water temperature and dissolved oxygen concentration were determined daily, acting in such a way that, the value of this parameter did not fall below the limit of 5-6 mg/l, through intervention measures such as: running water, sanitizing bathtubs by removing the remains of uneaten food and excreta eliminated after digestion. The concentration of nitrite (N-NO2-), nitrate (N-NO3-) and the concentration of ammonium ion (N-NO4+) were determined weekly. During the entire period of the experimental works in the premises of the pilot station no. 1, where the "Evos" ponds were placed, daytime lighting was ensured.

4.4.3.2. Amount and mode of administration of food

In order to increase the efficiency of feed use by the fry in the first days of feeding, the size of the feed particles did not exceed 80 microns. During the first 2-3 days, 40% of their weight was administered, after which the ration increased progressively as the fish grew. The feed was distributed during the day, at intervals of 2-3 hours; the frequency of meals plays a special role in ensuring weight gain and increasing feed efficiency (the smaller the fish, the more often it must be fed).

In the stage of post-embryonic development in the saddle in the experimental variants where the variable factor was the population density, the following feeding strategy was adopted and realized for a period of 40 days:

• the shad fingerlings were fed both live food and fodder;

• during the first 10 days, the fingerlings were fed with live food (Artemia salina nauplii) obtained from directed culture within S.C.D.P Nucet;

• after the 10 days of live food administration, it was switched to mixed feeding (in equal percentages live food + feed with a crude protein content of 60%) also for a period of 10 days, the type in which the shad fry begin to get used to consuming feed together with live food. Live food being reduced daily in the ration, so that at the end of the 10 days it represented 10-15% of the total feed;

• 20 days of feeding almost exclusively with fodder, live food being administered sporadically, once every three days, then until the age at which the achievement of growth parameters was observed and the phenomenon of cannibalism was observed (40 days) only with fodder;

daily feed rations in the form of live feed, mixed feed (live feed + feed) or only feed, were stable according to consumer mass evaluated at 5 days for each experimental variant;

• in all stages of feeding, both live food and fodder were administered separately or together, according to the "ad libitum" system at 1-2 hour intervals;

• the amount of feed was gradually increased according to the increase in weight of the fry and their numerical approximation to the data of the control fisheries.

The establishment of the feed requirement for each experimental variant was based on the assessment every five days of the mass of consumers existing in the "Evos" tubs, by weighing 30-50 specimens, the established mass then extrapolated to the relative number of specimens existing in the tubs .

From the observations made by the researchers from S.C.D.P Nucet regarding the intensity of food consumption during 24 hours by the larvae of some fish species (carp, Asian cyprinids, Polyodonsapthula, catfish) and from the specialized literature (Billard, 1995; Andrei and others, 1985, 1986, 1987; Ivanov, 1980) it was found that the nutrition of the fry is more intense between the hours of 8^{oo}-22^{oo}, after which the feeding intensity decreases during the night because in the hour interval 4^{oo}-6^{oo} the feeding approaches zero. It was also found that in the interval 8^{oo}-12^{oo}, the feeding intensity reaches up to 40-50% of the ration, increasing further up to 80% between 12^{oo}-18^{oo}, decreasing then, gradually towards the night.

Considering the mentioned, the distribution of the daily food ration in the three forms (live food, live food mixture + fodder, fodder) was made in 5 equal rations, at 8^{oo}, 11^{oo}, 14^{oo}, 17^{oo} and 20^{oo}.

It has been observed that, in the first days, the fry consume food from the surface of the water, then from the layer of water immediately below the surface film of the water, and as the age increases, almost entirely from the water mass.

4.4.3.3. Evaluation of the growth performance of saddle fry

The experimental development works during the rearing period in the saddle, in an intensive system, in which the variable factor was the stocking density, were carried out in three experimental variants.



Figure 14 (a-b). Biometric measurements at the end of the breeding period (original photo)

The duration of growth and development during the breeding period was 40 days (560-640 degree days), the end of the period being determined by the observation of the occurrence of the phenomenon of cannibalism.

Ensuring and complying with the mentioned conditions were aimed at if, at quasi-identical growth conditions, but at different population densities, the parameters of growth and development register significantly equal or strongly differentiated values (table 5).

The biotechnological indicator regarding the number of larvae per experimental variant being primarily dependent on the population density, it is obvious that at a higher population density and the number of larvae is higher or vice versa, even in the conditions where the population density of also influenced the survival rate.

					,	,				
The year	Vari- ant	Basin	SV (%)	W (g)	LT (mm)	Sr (g)	GR (g/zi)	PER (g/g)	FCR (g /g)	SGR (%/zi)
	V1	B1+B2+B3	65,8	1,581	56	1038,2	26,0	2,49	1,61	16,1
2018	V2	B4+B5+B6	50,3	1,012	49	1015,8	25,4	1,95	2,06	14,3
	V3	B7+B8+B9	44,3	0,755	46	997,1	24,93	1,54	2,06	13,3
	V1	B1+B2+B3	66	1,522	54	1002,2	25,05	2,45	1,63	16
2019	V2	B4+B5+B6	49,9	0,969	49	964,2	24,11	1,9	2,11	14,2
	V3	B7+B8+B9	43,3	0,792	46	1021,5	25,54	1,57	2,55	13,4
2020	V1	B1+B2+B3	66,2	1,566	56	1035,3	25,88	2,56	1,56	16
	V2	B4+B5+B6	49,5	0,978	50	967,1	24,18	1,87	2,15	14,2
	V3	B7+B8+B9	42,7	0,805	46	1028,1	25,7	1,57	2,55	13,4

Table no. 5. The situation of the main biotechnological indicators realized in the experimental works of growth and development in the saddle during the breeding period (average values/experimental variants/vears)

Meaning: W: average mass (g/ex); LT: average length (mm/ex); SV: survival percentage (%); Sr: real growth rate; GR: daily growth rate; PER: protein conversion factor; FCR: feed conversion factor; SGR: specific growth rate.

<u>Average mass (W) - g/ex</u>. The highest average weight of the fry after 40 days was obtained in the experimental variant V2 in 2019 of 1.555 g/ex, and the lowest was obtained in the variant V3 from the year 2018 of 0.821 g/ex (figure 15a).

In 2018, the highest average weight was obtained in the V1 variant of 1.414 g/ex, the lowest in the V3 variant of 0.821 g/ex, and in the V2 variant the average weight of 1.078 g/ex was obtained. In 2019, the highest average weight was obtained in the V1 variant of 1.555 g/ex, the lowest in the V2 variant of 0.969 g/ex, and in the V3 variant the average weight of 0.792 g/ex was obtained. In 2020, the highest average weight was obtained in the V1 variant of 1.489 g/ex, the lowest in the V3 variant of 0.880 g/ex, and in the V2 variant the average weight of 1.110 g/ex was obtained.

In conclusion, the highest average weight was obtained in all years of study in the V1 variant, followed by the V2 variant, and the lowest average weight in the V3 variant.

<u>Survival rate – Sv (%).</u> After 40 days of hatching, the highest survival percentage was obtained in the experimental variant V1 in the year 2020 of 66.2%, and the lowest was obtained in the variant V3 in the year 2018 of 42.7% (figure 15b).

In 2018, the highest percentage of survival was obtained in the V1 variant of 65.8%, the lowest in the V3 variant of 44.3%, and in the V2 variant a survival percentage of 50.3% was obtained.



Figure 15 (a-b) a) Average weight variation; b) Variation in survival rate (Sv)

In 2019, the highest percentage of survival was obtained in the V1 variant of 66%, the lowest in the V3 variant of 43.3%, and in the V2 variant a survival percentage of 49.9% was obtained.

In 2020, the highest percentage of survival was obtained in the V1 variant of 66.2%, the lowest in the V3 variant of 42.7%, and in the V2 variant a survival percentage of 49.5% was obtained.

In conclusion, the highest average survival percentage was obtained in all the years of study in the V1 variant, and the lowest in the V3 variant. This demonstrates that in high densities, the falcon manifests a state of stress, and from the age of approximately 35 days, the higher the density, the more intense the manifestation of cannibalism.

The feed conversion ratio (FCR) fell within the range of 1.61 – 2.55 as follows:

• in 2018, the lowest conversion coefficient was obtained in the V1 variant of 1.61 and the highest in the V2 variant and the V3 variant of 2.06;

• in 2019, the lowest conversion coefficient was obtained in the V1 version of 1.63, the highest in the V3 version of 2.55, and in the V2 version it was 2.11;

• in 2020, the lowest conversion coefficient was obtained in the V1 version of 1.56, the highest in the V3 version of 2.55, and in the V2 version it was 2.15.

The slightly high values of the food conversion coefficient are due to the fact that the administration at this age was done ad-libitum, so that all specimens have access to food, in order to avoid disproportionate growth and cannibalism.

The value of the higher conversion coefficient in variants V2 and V3 is due to the fact that in the first 35 days of breeding, all the biomass consumed food, after these 35 days the phenomenon of cannibalism in the two experimental variants manifested itself intensely, which led to a low survival, implicitly at an increased FCR value compared to the V1 variant (figure 16a).

<u>**Real growth rate (Sr).**</u> The actual growth rate had close values in all experimental variants, being between 964.2-1038.2 g. It was influenced by the survival percentage and the average weight of the biological material obtained at the end of the breeding season (figure 16b).

Daily growth rate (GR). The daily growth rate had close values in all experimental variants, without major differences, being influenced by the real growth spurt, which recorded close values and indirectly by the survival percentage (figure 17).



Figure 16 (a-b). a) Variation of the feed conversion coefficient (FCR); b) Variation of the real growth rate (Sr)



Figure 17. Variation of daily growth rate (GR)

Correlation between body mass and total length

By plotting the power regression of individual lengths and masses from the tested batches, slight differences are observed in terms of the allometric factor, respectively the condition of the specimens from the tested batches.

The mass length regression was carried out by means of simple linear regression analysis, and the confirmation and validation of the model was carried out with the help of descriptive statistics.

The correlation between total length (cm) and body mass (g) (Lt-M) was determined based on the data obtained from the biometrics performed at the end of the experiment, for the fish from the boiling experimental variant, using linear regression and the logarithmic equation (Chapter 2).

The fish populations obtained within the experimental variants had values that showed homogeneous populations. There were small variations that did not significantly influence the final results. At this stage of growth this correlation and not only have very sensitive deviations, weights of 0.1-0.4 g having strong influences in obtaining erroneous mathematical results.

The coefficient of determination R2 recorded values between 0.869 and 0.939 which signifies a good correlation between the studied variables and leads to the statement that the increase in body mass can be attributed to increases in length and varies from 86.9% in the V3 variant . to 93.9% in the V1 variant (table no. 6).

Correlation weight length										
The		Correlation	Coef.							
year	Variant	type	Pearson							
	V1	Positive	0,999							
2018	V2	Positive	0,999							
	V3	Positive	0,998							
	V1	Positive	0,999							
2019	V2	Positive	0,999							
	V3	Positive	0,997							
	V1	Positive	0,888							
2020	V2	Positive	0,94							
	V3	Positive	0,999							

Table number6. Correlation between length-weight within the experimental variants in the year 2018-
2019-2020

Analyzing the values of the "b" coefficient, it was observed an isometric increase of the biological material obtained in the V1 variant in all the years of the study, where b had values between 2,906 and 3,068, and a negative allometry in the experimental variant V2 where it was also recorded between values between 2,422. 2.722, and in the V3 variant, it increases in length and disfavors that of body mass, the b coefficient registering values between 2.098 and 2.421.

From the overall analysis of the coefficient of variability (CV) in what can be seen the variable body mass, it can be seen that the administration of food with a high percentage of protein led to the accentuation of the variability of the fish batches.

It is also worth noting that in the case of batches with high density, the percentage of survival and the average weight, recorded low values compared to the variant where the stocking density was lower (1000 ex/basin).

The	Variant	No. ex.	No. ex.		Weight	: (g)	CV(94)
year	Valialit	people	harvested	M min.	M max.	M med.±SD	CV (78)
	V1	3000	1975	1,203	1,92	1,581 ± 0,16	9,60 ± 0,013
2018	V2	6000	3020	0,553	1,745	1,012 ± 0,18	5,38 ± 0,015
	V3	9000	3986	0,436	1,068	0,755 ± 0,16	4,78 ± 0,013
	V1	3000	1980	0,953	2,295	1,552 ± 0,2	7,53 ± 0,017
2019	V2	6000	2995	0,505	1,789	0,969 ± 0,19	5,02 ± 0,016
	V3	9000	3895	0,306	1,211	0,792 ± 0,18	4,39 ± 0,015
	V1	3000	1986	1,107	2,192	1,565 ± 0,2	7,82 ± 0,016
2020	V2	6000	2971	0,552	1,36	0,978 ± 0,15	6,3 ± 0,013
	V3	9000	3847	0,521	1,397	0,805 ± 0,15	5,48 ± 0,012

Table no. 7. Minimum, maximum, average values, standard deviation and coefficient of variability of body mass, between the experimental variants at the end of the breeding season

In all the study years 2018-2019-2020, a positive correlation (according to the Pearson coefficient >0.5) is highlighted between the length and the average weight of the individuals stored in the growth and development pools, in the case of each experimental variant (V1, V2 and V3). This phenomenon highlights the homogeneous growth and development of the biological material, which proves that the food was sufficient in all experimental variants.

The value of the coefficient of variability in each experimental variant is below the value of 20%, thus all tested batches are considered homogeneous (table no. 7).

4.5. Experimental work on the growth and development of pikeperch (Sander lucioperca, L., 1758), in an intensive system, during the breeding period, in which the variable factor was the type of food administered

4.5.1. The objective

The main objective of this stage was the development of a technology for the growth and development of the salmon during the rearing period and the identification of the most suitable type of food for this predatory species at this stage, so as to achieve a positive correlation between the growth rate and valorization of administered food.

4.5.2. Experimental design

The "Evos" basin were washed and disinfected with lime chloride solution, after which they were washed again with technological water and dried, and 12 hours before the time of population with larvae they were fed with water up to the level of 0.25 -0.30 m. During the development of the biological material, the technological water level was also increased, reaching 0.5-0.55 m on the 20th day of breeding.

The experiments were carried out over 3 years: 2018, 2019 and 2020. Three experimental variants were established, each variant carried out in triplicate, being different from the point of view of the type of food administered.

Population density was set at 1000 ex/basin ("Evos" basin). This population density was chosen taking into account the fact that the cultivated species is a predatory species that hunts in the body of water and in which the predatory instinct is triggered quickly, easily manifesting cannibalism. The variable parameter was the type of feed administered. Food is a particularly

important technological parameter for growing fish, in all stages of development, and is specific to the species, age and applied technology.

The work methodology considered the following aspects:

 \succ three experimental variants were established, each variant made in triplicate, differentiated by the type of food administered;

- "Evos" basin type pools/experiential variant: 3;
- degree of repeatability: 3;
- feeding: natural food in live state or fodder;

➤ mode of administration: "ad libidum", only during the day (12 hours), with permanent control of food consumption and monitoring of the main physico-chemical parameters of the technological water (temperature, oxygen content, pH, organic matter, etc.);

duration of the breeding period: 42 days.

The experimental variants regarding the type of food administered:

- Option 1 feeding with zooplankton (Daphnia sp.);
- Option 2 feeding with fish larvae (carp);
- Option 3 feeding with fodder.

4.5.3. Results and discussions

4.5.3.1. Monitoring the environmental conditions of the water from the source and from the growth modules

During the development period in the "Evos" basin of the saltwater fry, with different types of food, the environmental conditions were monitored by collecting samples at intervals of 3-4 days, and the water temperature was determined daily three times/day (hours 800, 1400 and 2000) and the concentration of dissolved oxygen, acting in such a way that the value of this parameter did not fall below the limit of 5-6 mg/l, through intervention measures such as running water, sanitizing the bathtubs by removing the remains of unconsumed food and excreta eliminated by fry after digestion. Daytime lighting was ensured during the entire period of the experimental works in the room where the tubs were located.

The water temperature had a normal evolution, characteristic of the period in which the experimental works were carried out, with values that fell between 16.2 °C and 19.8 °C.

4.5.3.2. Amount and mode of administration of food

In the stage of post-embryonic development in the saddle, in the experimental variants where the variable factor was the type of food administered, the following feeding strategy was adopted and realized for a period of 42 days:

• the shad larvae were fed in variants V1 and V2 with live food and in variant V3 with granulated fodder;

• for the first 10 days, the larvae were fed with boiled egg yolk mixed with crushed feed "Aqua Garant Start 0.4" (50% egg yolk - 50% Aqua Garant Start), for all three variants;

• 5 meals per day were administered in the time interval 800-2000. The "Aqua Garant Start 0.4" feed and boiled egg yolk were crushed in powder form and administered in the water mass, where the biological material was;

• after the 10 days according to the protocol, it was switched to feeding on experimental variants, as follows:

- Option 1 feeding with zooplankton (Daphnia sp.);
- Option 2 feeding with fish larvae (carp);

• Variant 3 – feeding with granulated feed (from this date it was administered as such, because it was observed that the fry fryare developed and can ingest the feed with a size of 0.4 mm).

• in variants 1 and 2, food was administered twice a day (8 am and 3 pm), at the second meal, being administered only where it was found that live food was consumed;

• in variant 3, automatic belt feeders were placed, the feed being administered during the day between 800-2000;

• the amount of feed was gradually increased according to the increase in weight of the fry and their numerical approximation on the date of the "control fishing";

• after 40 days it was observed that the phenomenon of cannibalism appears and the fry were moved for growth in the first summer.

During the first ten days, the amount of 20 g of feed, composed of 10 g of boiled egg yolk + 10 grams of finely chopped "Aqua Garant Start 0.4" feed, was administered in the water mass, where the larvae were. During feeding, the larvae were observed to swim in the water mass after fine food particles, and after about 60 seconds, a yellow to brown coloration of the larvae's intestine could be observed, a sign that they had ingested the administered food. The frequency of meals during this period was 5 meals per day, being administered at 8⁰⁰, 11⁰⁰, 14⁰⁰, 17⁰⁰ and 20⁰⁰.

The amount of food administered during the 10 days was 200 g / tub "Evos" composed of 100 g boiled egg yolk + 100 g feed "Aqua Garant Start 0.4". The amount administered was identical in all experimental variants. The total amount of food administered during the 10 days per total experiment was 1800 g, composed of 900 g of boiled egg yolk + 900 grams of "Aqua Garant Start 0.4" feed.

Since the variable parameter was the type of feed administered, the amount of feed administered in the first decade was not taken into account when determining the feed conversion ratio.

After the ten days, it was switched to differentiated feeding, at first in smaller quantities, the ration gradually increasing depending on the feeding capacity of the fry. The feeding period was divided into three decades, the first and second decades being 10 days each and the third decade being 12 days.

In the works on the experiments carried out on the development of flounder larvae during the hatching period, the feeding in the V3 version was done with an Aqua Garant Start 0.4 feed, the same as when developing the development technology of the flounder during the hatching period in different densities.

The distribution of the daily food ration in two forms (live food and fodder) was carried out during the day, for variants V1 and V2 in two meals at 8°-15° and for variant V3, feed was administered in 5 equal meals at 8°0, 11°, 14°, 17° and 20°.

It was observed that, in the V3 version, the fry of the shad consume food in the first days from the surface of the water, then from the layer of water below the immediate vicinity of the surface film of the water, and as the age increases, almost entirely from the water mass. It was also found in all the experimental variants that the shalau consumes 85-90% of the food during the day during the light and 10-15% during the night.

In variants V1 and V2, the consumption of live food was achieved in the first part of the day in the time interval 8-11⁰⁰, in the proportion of 65-70%, in the second part of the day in the time interval 11-2000 in the proportion of 20-25% and only 10% was achieved at night. It should be noted that, during the night, the space where the breeding was carried out was not illuminated.

4.5.3.3. Evaluation of the growth performance of saddle fry

The experimental development works during the rearing period in the saddle, in an intensive system, in which the variable factor was the type of feed, were carried out in three experimental variants. They took place over 3 years, during 2018, 2019 and 2020, under almost identical conditions both from a technical and technological point of view.

The larval food source consisted of two components:

 natural food consisting of live food such as zooplankton (Daphnya sp.) and fish larvae (carp - Carassius auratus gibelio), obtained through natural - directed reproduction and growth at S.C.D.P Nucet;

• additional feed by administering the feed "Aqua Garant Start 0.4" ad libidum.

The duration of growth and development during the breeding period was 42 days (588 – 670 degree days), the end of the period being completed when the cannibalism phenomenon was observed (figure 18).

The experimental work on the development of the sow during the rearing period, in which the variable factor was the administered feed, was carried out under technical and technological conditions, similar. There were, however, insignificant variations regarding the physico-chemical parameters of the growth medium.



Figure 18. The phenomenon of cannibalism in the post-embryonic period (original photo)

Ensuring and complying with the mentioned conditions were aimed at quasi-identical growth conditions, but with different types of food administered, the parameters of growth and development quantified by: average mass W (mg/ex), total length LT (mm/ex) and survival, register significantly equal or strongly differentiated values.

Ensuring and complying with the mentioned conditions were aimed if, at identical growth conditions and stocking densities but different type of food, the parameters of growth and development register significantly equal or strongly differentiated values (figure 19 a-b).

Table no. 8. The situation of the main biotechnological indicators realized in the experimental works of growth and development in the saddle during the breeding period (average values/experimental variants/vears)

	variants/years/									
Anul	Vari- anta	Bazinul	SV med. (%)	W mediu (g)	LT mediu (mm)	Sr (g)	GR (g/zi)	PER (g/g)	FCR (g furaj/g spor biomasă)	SGR (%/zi)
	V1	B1+B2+B3	61,8	2,545	65	1571,1	37,41	1,5	1,97	16,37
2018	V2	B4+B5+B6	65,2	4,103	75	2673,4	63,65	1,38	2,18	17,59
	V3	B7+B8+B9	46,9	1,927	59	902	21,48	3,58	1,67	14,95
	V1	B1+B2+B3	60,8	2,479	65	1505,5	35,85	1,5	1,93	16,26
2019	V2	B4+B5+B6	65	4,08	77	2652,2	63,15	1,41	2,18	17,58
	V3	B7+B8+B9	47,3	1,961	60	925,4	22,03	3,59	1,68	15,17
	V1	B1+B2+B3	61,4	2,477	64	1520,3	36,2	1,37	2,3	16,36
2020	V2	B4+B5+B6	65	4,094	76	2658,5	63,3	1,55	1,97	17,55
	V3	B7+B8+B9	46,8	1,962	60	916	21,81	3,65	1,83	15,01

Average mass (W) - g/ex. The highest average weight of fry after 42 days was obtained in experimental variant V2, pool B5, in 2018 of 4.154 g/ex., and the lowest was obtained in variant V3, pool B9, from 2020 of 1.878 g /ex;

In 2018, the highest average weight was obtained in the V2 variant of 4.103 g/ex., the lowest in the V3 variant of 1.927 g/ex., and in the V2 variant the average weight of 2.545 g/ex. was obtained;

In 2019, the highest average weight was obtained in the V2 variant of 4.08 g/ex., the lowest in the V3 variant of 1.961 g/ex., and in the V1 variant the average weight of 2.479 g/ex. was obtained;

In 2020, the highest average weight was obtained in the V2 variant of 4.094 g/ex., the lowest in the V3 variant of 1.962 g/ex., and in the V1 variant the average weight of 2.477g/ex. was obtained;

in conclusion, the highest average weight was obtained in all the years of study in the V2 variant, followed by the V1 variant, and the lowest average weight in the V3 variant (table no. 8, figure 20 a).



Figure 19(a-b). Biometric measurements at the end of the rearing period (original photo)

<u>Survival rate (%).</u> After 42 days of hatching in the three years of the study, the highest survival percentage was obtained in the experimental variant V2, pool B4, in 2019 of 66.8%, and the lowest was obtained in the variant V3, pool B7, in 2020 by 45.6%;

In 2018, the highest percentage of survival was obtained in the V2 variant of 65.2%, the lowest in the V3 variant of 46.9%, and in the V2 variant a survival percentage of 61.8% was obtained;

In 2019, the highest percentage of survival was obtained in the V2 variant of 65.0%, the lowest in the V3 variant of 47.3%, and in the V1 variant a survival percentage of 60.8% was obtained;

In 2020, the highest percentage of survival was obtained in the V2 variant of 65.0%, the lowest in the V3 variant of 46.8%, and in the V1 variant a survival percentage of 61.4% was obtained;

In conclusion, the highest average survival percentage was obtained in all the years of study in the V2 variant, and the lowest in the V3 variant. This demonstrates that when the food is different from its specific food, the sable shows a state of stress. After the age of about 35 days, the shad switches to active predator feeding, consuming food that moves in the water mass such as zooplankton (Daphnia sp.), but especially fish larvae that actively swim in the water mass. During the studies, it was observed that the movements of fish larvae in the water mass instantly trigger their attack instinct. This instinct can be easily triggered by the swimming of specimens in front of it, and it no longer differentiates between species or size, specimens of the same size attacking each other causing numerical losses, implicitly a low survival. It was also observed that when fed with live food (zooplankton, fish larvae), the manifestation of cannibalism is reduced (figure 20 b).



Figure 20 (a-b). a) Average weight variation; b) Variation in survival rate

Feed conversion ratio (FCR) The best feed conversion ratio was obtained in the experimental variant V3 pool B9, in 2019, of 1.630, and the highest was obtained in variant V1, pool B2, in 2019 of 2.4.

In 2018, the lowest feed conversion coefficient was obtained in the V3 variant of 1.67, the highest in the V2 variant of 2.18, and in the V1 variant a feed conversion coefficient of 1.97 was obtained.

In 2019, the lowest feed conversion coefficient was obtained in the V3 variant of 1.68, the highest in the V2 variant of 2.18, and in the V1 variant a feed conversion coefficient of 1.5 was obtained.

In 2020, the lowest feed conversion coefficient was obtained in the V3 variant of 1.83, the highest in the V1 variant of 2.3, and in the V2 variant a feed conversion coefficient of 1.97 was obtained (figure 21 a).

In conclusion, the lowest feed conversion coefficient obtained in all the years of the study was in the V3 variant, and the highest in the V2 variant, with the exception of 2020 when an average coefficient was obtained. The low value of the feed conversion coefficient obtained in the V3 variant is due to the fact that the feed used to feed the fish material was qualitative, but also to the high percentage of crude protein it contained (60%).

It can be stated that the values obtained in variants V1 and V2 (where live food was administered) is an estimate, because the live food was administered as such, with a high water content. When calculating the different indices where the crude protein value was used, it was estimated for live feed at 30%.

Real growth rate (Sr). The actual growth rate had different values, so in the V1 variant they fell within the range of 1505-1571 g, in the V2 variant they were 2652-2673, and in the V3 variant 902-925. Within the variants they had close values. The differences between the variants show that the administration of food as close as possible to that in the natural environment gives it a higher growth boost in terms of total and individual quantity. It was also influenced by the percentage of survival and the average weight of the biological material obtained at the end of the breeding season (figure 21 b).



Figure 21 (a-b). a) Variation of the feed conversion coefficient (FCR); b) Variation of the real growth rate (Sr)



Figure 22. Variation of daily growth rate (GR)

Daily growth rate (GR). The daily rate of growth had different values, so in the V1 variant they fell within the gap of 35.85-37.41 g/day, in the V2 variant they were 63.15-63.65, and in the V3 variant of 21.48-22.03. Within the variants they had close values. The differences between the variants were influenced by obtaining different average weights and total amounts of biomass between the experimental variants. It was also influenced by the percentage of survival obtained at the end of the breeding season (figure 22).

Correlation between body mass and total length

By plotting the power regression of individual lengths and masses from the tested batches, slight differences can be observed in terms of the allometric factor, respectively the condition of the specimens from the tested batches.

The regression of length and body mass was carried out by means of simple linear regression analysis, and the confirmation and validation of the model was carried out with the help of descriptive statistics.

The correlation between total length (cm) and body mass (g) (Lt-M) was determined based on the data obtained from the biometrics performed at the end of the experiment, for the fish from the boiling experimental variant, using linear regression and the logarithmic equation (Chapter 2).

The fish populations obtained within the experimental variants had values that showed homogeneous populations. There were small variations that did not significantly influence the final results. At this stage of growth and development this correlation and not only, have very sensitive deviations, weights of 0.1-0.4 g having strong influences in obtaining erroneous mathematical results (figure 23a-b;24a-b).

The coefficient of determination R2 recorded values between 0.861 and 0.927 which means a good correlation between the variables studied and leads to the statement that the increase in body mass can be attributed to increases in length and varies from 86.1% in the V3 variant to 92.7% in the V1 variant.

		No ex	No ex		The we		
The year	Variant	people	harvested	M min.	M max.	M med.±SD	CV (%)
2018	V1	3000	1854	2,022	2,989	2,545 ± 0,2	13,00 ± 0,016
	V2	3000	1956	3,706	4,587	4,103 ± 0,20	19,98 ± 0,017
	V3	3000	1407	1,348	2,201	1,927 ± 0,16	12,19 ± 0,013
	V1	3000	1824	2,069	2,98	2,479 ± 0,24	10,28 ± 0,02
2019	V2	3000	1951	3,603	4,489	4,080 ± 0,26	15,94 ± 0,021
	V3	3000	1419	1,508	2,314	1,961 ± 0,26	10,59± 0,015
	V1	3000	1843	2,087	2,992	2,477± 0,17	14,62 ± 0,014
2020	V2	3000	1949	3,717	4,584	4,094 ± 0,21	19,24 ± 0,017
	V3	3000	1404	1,408	2,581	1,962 ± 0,18	10,47 ± 0,015

Table no 9. Minimum, maximum, average values, standard deviation and coefficient of variability of body mass, between the experimental variants

Table no. 10. Correlation between length-weight within the experimental variants in 2018

	Correla	tion weight len	gth
The		Correlation	Coef
year	Variant	type	Pearson
	V1	Positive	786
2018	V2	Positive	798
	V3	Positive	581
	V1	Positive	0,995
2019	V2	Positive	0,991
	V3	Positive	0,989
	V1	Positive	0,881
2020	V2	Positive	0,999
	V3	Positive	0,758

Analyzing the values of the "b" coefficient, an isometric increase of the biological material obtained in the V2 variant was observed in all the years of the study, where b had values between 2.896 and 3.056, a slightly negative allometry in the experimental variant V1 where b recorded values between 2.711 and 2.888, and in the V3 variant the increase in length favors that of body mass, the b coefficient registering values between 2.651 and 2.786.

From the overall analysis of the coefficient of variability (CV) with regard to the variable body mass, it can be seen that administration of different food led to the accentuation of the variability of the fish batches (table no. 9).

It is also worth noting that in the case of batches with live feed administration, especially crucian carp larvae, the percentage of survival and average weight recorded higher values compared to the version where granulated feed was administered.

In all the study years 2018-2019-2020, a positive correlation (according to the Pearson coefficient >0.5) is highlighted between the length and the average weight of the individuals stored in the growth and development pools, in the case of each experimental variant (V1, V2 and V3). This phenomenon highlights the homogeneous growth and development of the biological material, which proves that the food was sufficient in all experimental variants (table no. 10).



Figura 23 (a-b). a) Larvae pikeperch 7-10 days; b) Young pikeperch 28 days (original photo)



Figura 24 (a-b) Young pikeperch at the end of the postembryonic period (original photo)

CHAPTER V. EXPERIMENTAL WORKS REGARDING THE DEVELOPMENT OF THE TECHNOLOGY OF GROWING THE SALAD (SANDER LUCIOPERCA – LINAEUS, 1758), IN SUMMER I, IN GROUND PONDS

5.1. The objective of experimental works

The experimental works carried out within the Nucet Experimental Base (S.C.D.P. Nucet) have as their objective the development of the breeding technology of the pike species (Sander lucioperca, L. 1758), in the first summer, in earthen pools, in monoculture, in order to obtain material of popular for one summer, with the size (average weight and size) at which the

species is no longer a major target for predators and is able to easily obtain food, with a view to restoring natural populations and introducing and expanding the species in aquaculture.

5.2. Experimental design

The research was carried out in 2018-2019-2020 at the Nucet Aquaculture Research-Development Station. The experimental pools for growth in summer I are located in the main bed of the Ilfov stream, downstream of the Ilfoveni reservoir dam.

5.2.1. Establishment of experimental variants

To carry out the experimental growth works in the first summer of the barn, two experimental variants were considered where the variable factor was the type of food administered. The degree of repeatability was 3 times, in triplicate, their release period was between the years 2018, 2019 and 2020.

The experimental variants of stability were:

- variant V1: feeding with fodder;
- variant V2: feeding with live fish (crucian carp).

5.2.2. Preparing the basins for population

Before use, the rearing basins were left dry and disinfected with lime chloride, more intensively in wet areas. Also, in the basins intended for the experimental variant V2, at the base of the slope on the long side of the basins, a bed of artificial grass with a width of 1.0 m was inserted as a support for the laying of bridges by the caras (r.+ R.), 12 days before the time of stocking, the basins were supplied with water until the optimal level of exploitation (fish level) was achieved, thus ensuring average water levels between 1.6-1.8 m. The supply and the water is discharged individually for each basin, through the mentioned hydrotechnical installations. The water supply was made from a common slab channel that derives from the weir located upstream of the Nucet body basins on the llfov stream (supply source), and the water discharge is also done in the llfov stream. At the feeding and exhaust installations, gratings were installed on which metal sieves with a 4 mm mesh side were mounted, in order not to allow the entry of other species of fish or the escape of the stocked fish material. Considering the fact that the experimental pools are supplied with water from the same source, we can say that, under the aspect of the environmental conditions given by the physical-chemical characteristics of the water, a relatively high degree of homogeneity of the growth environment was ensured. During the period of growth and development of the fish in the experimental basins, a supply flow of 3-4 I/sec/ha was ensured to ensure the supply of water and the compensation of water losses due to water infiltration and evaporation.

5.2.3. The popularity of fish material

Establishing the population density took into account the fact that the cultured species (pikeperch) is a predatory species, which feeds actively in the water mass and whose predatory instinct is triggered quickly manifesting itself with great intensity, and in the absence of adequate food and enough can go on to consume its own smaller fellows (the phenomenon of cannibalism). Consequently, the population density was set at 750 ex/experimental pool (S:1,000 m2).

The variable parameter within the experimental variants was the type of food administered, food being a particularly important technological parameter for the directed growth of fish regardless of the fish species that is the object of growth in all stages of development with a well-defined degree of specificity for each species, age and applied technology.

The work methodology considered the following aspects:

- establishing the two experimental variants, each variant multiplied three times, differentiated by the type of food administered;

- establishing the number of soil basins/experimental variant: 3;

- degree of repeatability: 3;

- type of food administered: live natural food (carp) or fodder;

- mode of feeding/administration: "ad libidum", only during the day, with permanent control of food consumption;

- monitoring of the main physico-chemical parameters of technological water (temperature, oxygen content, pH, organic matter, etc.);

- duration of the growth period: average 120 days.



Figure 25(a-e). Biometric measurements on the stocking of experimental rearing ponds (original photo)

			2020				
The			People				
year	Variant	Basin	Nr. Ex.	Wmed.	Amount		
				(g/ex)	(kg)		
		B1	750	1,93	1,45		
	V1	B2	750	1,82	1,37		
		B3	750	1,97	1,48		
2018		B4	750	2,57	1,92		
	V2	B5	750	2,85	2,14		
		B6	750	2,62	1,97		
		B1	750	1,98	1,49		
	V1	B2	750	2,14	1,61		
2019		B3	750	1,95	1,46		
		B4	750	2,25	1,68		
	V2	B5	750	2,46	1,84		
		B6	750	2,24	1,68		
		B1	750	1,89	1,42		
	V1	B2	750	2,24	1,68		
2020		B3	750	2,05	1,53		
		B4	750	2,49	1,87		
	V2	B5	750	2,56	1,92		
		B6	750	2,56	1,92		

Table no. 11. The situation of the populations in the two experimental varieties for the period 2018-2019-

The popularity of the experimental variants was made with saddle aged 48-50 days, with average weights between 1.823 - 2.850 g/ex, as follows:

variant V1: 750 specimens/pool, where granulated fodder was administered to feed the biological material in the experimental pools: B1, B2 and B3;

variant V2: 750 specimens/pool, where live food (carp) was administered, in pools B4, B5 and B6 (table no. 11, figure 25a-e).

The choice of crucian carp for feeding the saddle was made for several reasons, namely:

- \checkmark is a very prolific species that reproduces several times in a vegetative season;
- \checkmark is not picky about the quantity but especially the quality of the food consumed;

 \checkmark it is the most resistant species in terms of environmental conditions in aquatic ecosystems regardless of their type;

 \checkmark has high survival in all stages of development and growth.

The method of carrying out the experimental work on the main work phases (stocking, feeding, harvest fishing) was as follows:

in 2018, populations were made on June 5 in both experimental variants, the feeding period was until October 8, resulting in a number of 120 feeding days (V1), and harvest fishing for both variants experimental was carried out on October 10;

in 2019, populations were made on June 9 in both experimental variants, the feeding period was until October 5 (V1), resulting in a number of 117 feeding days, and harvest fishing for both variants experimental was carried out on October 8;

in 2020, populations were made on June 11 in both experimental variants, the feeding period was until October 9 (V1), resulting in a number of 120 feeding days, and harvest fishing for both variants experimental was carried out on October 12;

in the V2 variant, the feeding of the saddle fry with live food had a permanent character.

In both experimental variants of each year of implementation, no feed was administered on Sundays (for the saddle fry and for the crucian carp).

In the V2 variant, before the pre-developed bream fry were stocked in the experimental pools B4, B5 and B6, crucian carp larvae obtained from the natural-directed reproduction within the S.C.D.P. Nucet as the first food support for broiler chickens immediately after hatching. For each pool, about 100,000 5-day-old larvae were stocked for each pool. For each of the three experimental pools (B4, B5 and B6), 10% of their surface was allowed to grow aquatic vegetation as additional support for the directed reproduction of the crucian carp.

Also in the three mentioned basins, after the flooding, 10 days before the stocking, the amount of 50 kg crucian carp (breeders and breeders), with an average weight between 50-80 g/ex, aged 1-2 years, was populated .

5.2.4. Ensuring food requirements

The feeding of the sallow during directed growth in monoculture in earthen basins (heleste) is a less frequently used method, with a novelty character, but it is being developed.

Granulated feed of the type for salmon (Salmo salar), rainbow trout (Oncorhynchus mykiss) and European sea bass (Dicentrarchuslabrax) can be used for artificial feeding of the salmon. These forages must have the property of "slow sinking" so that when they remain in the water mass, the shad have the opportunity to consume them. As mentioned, the feeding of the growing saddle in the first summer consisted of feed live and from granulated feed. Live food was crucian carp, and granulated feed was fed with "Aqua garant start 0.7" and "Aqua garant start 1".

In the V1 version, in each of the three years of the experimental works (2018-2019-2020) for the feeding of the fish material, starter granulated feed **"Aqua Start 0.7"** with slow sinking, variable granulation of 0.6-0.8 was administered mm, especially for chickens of 1-5 g/ex,

obtained by a special technology - marbling, having a high nutritional value, with high stability in water, microencapsulated to protect nutrients and preserve water quality. Crude protein was 55% and the feed was given during the first 30 days after laying. After 30 days, the fish stock was fed with **"Aqua Start 1"**, having the same characteristics as "Aqua Start 0.7", the difference being the grain size "Aqua Start 1" having the grain size of 0.9-1.1 mm.

In the V2 version, 850 kg of feed/basin/growing season were administered for feeding the crucian carp, the amount established based on the following calculation methodology:

- ✓ amount of crucian carp stocked per experimental pool: 50 kg;
- ✓ estimated multiplication rate for the growth period: 2.5 times;
- ✓ estimated amount of crucian carp in autumn fishing: 125 kg;
- \checkmark growth spurt: 125 kg 50 kg = 75 kg;
- ✓ survival of crucian carp larvae until autumn: 5.0%;
- ✓ estimated number of larvae in autumn fishing: 5,000 ex;
- ✓ estimated average weight: 10 g/ex;
- ✓ quantity: 5,000 ex x 10 g/ex = 50 kg;
- \checkmark total increase in production: 75 kg + 50 kg = 125 kg;
- ✓ specific consumption: 5% of total gain/day = $6.25 \text{ kg} \approx 6.0 \text{ kg}$;
- \checkmark average duration of feed administration: 120 days;
- ✓ required amount of feed: 120 days x 6.0 kg/day = 720 kg;
- ✓ 15% of the quantity required for the crucian carp fry resulting from the directed reproduction left unconsumed by the saddle 15% x 720 kg = 108 kg ≈ 110 kg;
- ✓ total feed: 720 kg + 110 kg = 830 kg ≈ **850 kg**.

The combined feed administered for the carp species was prepared at the unit level (S.C.D.P. Nucet) by grinding and mixing the feed components, in the chosen combination of feed components and the amount/component per 100 kg module, a crude protein content of \approx 26.0%.

Considering that the stocking with crucian carp in the experimental ponds had a heterogeneous character in terms of age and average weight, feeding was imposed by the need to predict the growth of specimens of smaller age and size (1-2 years) and maintain the state of maintenance for crucian carp breeders, ensuring in these conditions the increase in the prolificacy and quality of the seminal elements, which would allow the reproduction of this species, and the resulting offspring serving as live food for the bream.

Feeding for both experimental variants (V1- for bream and V2 - for crucian carp) started the day after breeding. Feed administration was done at four tables for each of the pools of the experimental version V2, and the fodder for the saddle in the water mass on almost the entire surface of the pools.

In the V1 version, the amount of feed administered was different from one day to another and from one pool to another, depending on the appetite of the fish material. From the point of view of the frequency of the meals, there were two a day, in the morning at 900 and in the afternoon at 1430. The feed was administered *ad-libitum*, taking into account the feed guide provided by the manufacturer, which establishes the daily amount of feed (kg feed/100 kg consumers/day) depending on the size and individual weight of the biological material and the water temperature.

In variant V2, the feed was administered in a single ration, in the morning at 900, as mentioned, the amount of feed administered was constant, 850 kg for each pool and for each year in which the experimental works were carried out (2018- 2029-2020).

5.3. Results and discussion

5.3.1. Monitoring the environmental conditions of water from the source and from the growth basins

Monitoring the quality of technological water by taking samples, determining and studying the values of the physico-chemical parameters is particularly important because maintaining them within normal limits leads to the existence of a favorable environment for the growth of fish in general and of salmon in particular in the experimental variants made. At the same time, when deviations from normal values were observed for one or more parameters, specific measures were taken so that the situation returned to normal.

Year	Vari- ant	Basin	SV (%)	W (g)	LT (mm)	Sr (g)	GR (g/zi)	PER (g/g)	FCR (g /g)	SGR (%/zi)
2019	V1	B1,B2,B3	70,7	132	22,7	68,6	572	4	1,39	2,28
2010	V2	B4,B5,B6	89,5	294	29,7	195,4	1628	1,9	1,61	2,73
2010	V1	B1,B2,B3	64,3	142	24,7	67	558	4,3	1,28	2,27
2019	V2	B4,B5,B6	91,6	297	31	202,3	1686	1,8	1,63	2,74
2020	V1	B1,B2,B3	66,2	150	24,9	73,2	610	4,3	1,29	2,3
2020	V2	B4,B5,B6	91,0	296	29,9	200,2	1668	1,7	1,72	2,73

Table no. 12. The average values per year/experimental variants of the main bioindicators of performance realized in the experimental growth works in summer I at the saddle



Figure 26. Determining the average weight of individual shad in harvest fishing (original photo)

During the growth period in the first summer of the sallow, the environmental conditions were monitored by collecting samples once a week, and the water temperature and dissolved oxygen concentration were determined daily, acting in such a way that the value of the latter parameter it did not fall below the limit of 5-6 mg/l, through intervention measures such as running water.

As a general conclusion, it follows that from the point of view of the quality of the growth medium (technological water) from the feed source and the experimental basins for the analyzed period 2018-2019-2020, it corresponded to the quality standards of fish waters, being included in the normative acts found in force (table no. 12).

5.4. Obtained results

The experimental breeding works in the first summer of the pike species (Sander lucioperca - L., 1758) were carried out during 3 years, in the period 2018-2019-2020, in two experimental variants, with food as a factor of variability administered.

From this point of view, the feed of the gerbil fry during the growth period/experimental variants was:

- live food consisting of crucian carp larvae of different ages (larvae from naturallydirected reproduction, pre-developed fry, fry);

- granulated feed "Aqua Garant Start 0.7" and "Aqua Garant Start 1" for free consumption.

Duration of the growth period on average 121 days (124 days/2018, 119 days/2019 and 120 days/2020).

Ensuring and complying with the mentioned conditions were aimed if, at identical growth conditions and stocking densities but different type of food, the parameters of growth and development register significantly equal or strongly differentiated values (table 21).

Survival rate (%). In the three years of the study, the highest survival percentage was obtained in the experimental variant V2, basin B6, in 2020 of 95.1%, and the lowest was obtained in variant V1, basin B3, in 2019 of 59.9%.

In all the years of the study, the survival percentage obtained was higher in variant V2 with values ranging from 84.9-95.1%, compared to variant V1 where it recorded values between 59.9-74% (figure 27 of).



Figure 27 (a-b). a) Variation in survival percentage b) Variation in mean weight

Average mass (W) - g/ex. The highest average mass of the salamander after the first vegetative season was obtained in the experimental variant V2, in 2018, basin B6 and in 2020 basin B5 having the value of 318 g/ex., and the lowest was obtained in variant V, from 2018, pool B3 of 130 g/ex;

In 2018, the highest average weight was obtained in the V2 variant of 294 g/ex., the lowest in the V1 variant of 132 g/ex.

In 2019, the highest average weight was obtained in the V2 variant of 297 g/ex., the lowest in the V1 variant of 142 g/ex.

In 2020, the highest average weight was obtained in the V2 variant of 296 g/ex., the lowest in the V3 variant of 150 g/ex.

In conclusion, the highest average weight was obtained in all years of study in the V2 variant, and the lowest average weight in the V1 variant (figure 27 b).

The total amount of biomass per surface unit (kg) - kg/ha. The total amount of biomass obtained relative to the surface unit (kg/ha) falls within the gap of 665-818 kg/ha for the V1 variant and with values between 1926-2099 kg/ha for the V2 variant.

The average values of the total amount and per surface unit (kg/kg/ha) are higher in the experimental variants V2 compared to those in the experimental variants V1 every year, the highest value of this bioindicator in the variant V2 is 2099 kg/ha in 2020 in the B5 basin and lower in 2018, in the V1 variant of 665 kg/ha in the B1 basin.

The multiplication rate of the average weight is at higher values in the experimental V2 variants compared to those in the V1 variants, thus the multiplication rate has the highest value in the V.2 variants in 2019 (128.0 (x), the lowest, 108.2 (x) in 2018 compared to variants V.1 where the highest value is 77.6 (x) in 2018 and the lowest, 70.8 (x) in 2019;

Analyzing the data, we can state that in the basins where the same experimental variant was carried out, the obtained values were close, large differences were obtained between the two variants, approximately 2.5–3 times smaller in the V1 variant compared to the V2 variant (figure 28 a).



Figure 28 (a-b). a) Variation of the amount of biomass per surface unit (kg/ha) b) Variation of the feed conversion coefficient (FCR)

The (estimated) feed conversion ratio (FCR)

In 2018, the lowest feed conversion coefficient was obtained in the V1 variant of 1.39, compared to the V2 variant where a feed conversion coefficient of 1.61 was obtained.

In 2019, the lowest feed conversion coefficient was obtained in the V1 variant of 1.28, compared to the V2 variant where a feed conversion coefficient of 1.63 was obtained.

In 2020, the lowest feed conversion coefficient was obtained in the V1 variant of 1.29, compared to the V2 variant where a feed conversion coefficient of 1.72 was obtained (figure 28 b).

In conclusion, the lowest feed conversion coefficient obtained in all the years of study was in the V1 variant, and the highest in the V2 variant. The low value of the feed conversion coefficient obtained in the V1 variant is due to the fact that the feed used to feed the fish material was qualitative, but also to the high percentage of crude protein it contained (55%).

It can be stated that the FCR values obtained in the V2 variant (where live food was administered) are estimated values, because it is not known exactly the amount of crucian carp

that was consumed by the saddle. This amount of populated crucian carp reproduced and developed in such a way that the fish had food available, hence the obtaining of higher indices such as average weight, percentage of survival, total amount of biomass obtained, etc.. When calculating the different indices where it was crude protein value was used, this was estimated for live feed at 30%.

<u>**Real growth rate (Sr).</u>** The real increase in growth had different values, so in the V1 variant they fell within the gap of 65.4-80.6 kg compared to the V2 variant, they were 191.2-208.3 kg. Within the variants they had close values. The differences between the variants show that the administration of food as close as possible to that in the natural environment gives it a higher growth boost in terms of total and individual quantity. It was also influenced by the percentage of survival and the average weight of biological material obtained at the end of the growing season in summer I (figure 29 a).</u>



Figure 29 (a-b). a) Variation of real growth rate (Sr) b) Variation of daily growth rate (GR)

Daily growth rate (GR). The daily growth rate had different values, so in the V1 variant they fell within the gap of 544.8–672.0 g/day, compared to 1593.1–1736.2, in the V2 variant. Within the variants they had close values. The differences between the variants were influenced by obtaining different average weights and total amounts of biomass between the experimental variants. It was also influenced by the survival percentage obtained at the end of the growing season in the first summer (figure 29 b).

	800	iy maee, ar	nong ino oxp	erimental t	aname for gre		
Voor	Variant	No. ex.	No. ex.		The weigl	nt (g)	CV(94)
rear	Valialit	people	harvested	M min.	M max.	M med.±SD	CV (78)
2019	V1	2250	1590	108	157	132 ± 10,92	12,08 ± 0,89
2010	V2	2250	2003	270	329	294 ± 17,93	16,39 ± 1,46
2010	V1	2250	1433	116	159	142 ± 11,97	11,83 ± 0,98
2019	V2	2250	2055	259	328	297 ± 16,08	18,46 ± 1,31
2020	V1	2250	1478	112	182	149± 14,55	10,27 ± 1,19
2020	V2	2250	2040	256	337	296 ± 19,4	15,26 ± 1,58

Table no. 13. Minimum, maximum, average values, standard deviation and coefficient of variability of body mass, among the experimental variants for growth in summer I

<u>Correlation between body mass and total length</u> By plotting the power regression of individual lengths and masses from the tested batches, slight differences can be observed in terms of the allometric factor, respectively the condition of the specimens from the tested batches.

The regression of length and body mass was carried out by means of simple linear regression analysis, and the confirmation and validation of the model was carried out with the help of descriptive statistics.

The correlation between total length (cm) and body mass (g) (Lt-M) was determined based on the data obtained from the biometrics performed at the end of the experiment, for the fish from the boiling experimental variant, using linear regression and the logarithmic equation (Chapter 2).

The fish populations obtained within the experimental variants had values that showed homogeneous populations. There were small variations that did not significantly influence the final results.

The coefficient of determination R2 recorded values between 0.874 and 0.946 which signifies a good correlation between the variables studied, which leads to the statement that the increase in body mass can be attributed to increases in length and varies from 87.4% to 94.6%.

Analyzing the values of the "b" coefficient, an isometric increase of the biological material obtained in the V2 variant was observed in all the years of the study, where b had values between 2.968 and 3.126 and a slightly negative allometry in the experimental variant V1 where b recorded values between 2.531 and 2.701, the increase in length disfavors that of body mass.

Correlation weight length						
Year	Variant	Correlation type	Coef. Pearson			
2018	V1	Positive	0,956			
	V2	Positive	0,966			
2019	V1	Positive	0,898			
	V2	Positive	0,878			
	V1	Positive	0,921			

Table number 14. Correlation between length-weight within the experimental variants in the year 2018-2019-2020

From the overall analysis of the coefficient of variability (CV) regarding the variable body mass, it can be seen that administration of food with a high percentage of protein in the version of feeding with granulated fodder and species-specific food in the version of feeding with crucian carp, led to increasing the variability of fish stocks (table no. 13).

Positive

0,942

2020

V2

It is also worth noting that in the case of the groups fed with granulated fodder, the percentage of survival and the average weight recorded low values compared to the variant where the feed was specific to the barn.

In all the study years 2018-2019-2020, a positive correlation (according to the Pearson coefficient >0.5) between the length and the average weight of the individuals stored in the growth and development pools is highlighted, in the case of each experimental variant (V1 and V2). This phenomenon highlights the homogeneous growth and development of the biological material, which proves that the food was sufficient in all experimental variants (table no. 14)

CHAPTER VI. GENERAL CONCLUSIONS AND PERSONAL CONTRIBUTIONS

The need to introduce new economically valuable species into the culture comes as a result of the increase in market demand for increasing quantities of fish products and the diversification of the consumption of aquatic products.

In our country, the growth of the pikeperch takes place in classic systems as a complementary species alongside the cyprinids, playing the role of sanitizing the ponds by consuming fish that do not have an appropriate growth rate and species without economic value that accidentally entered in ponds along water inflow.

The purpose of this work was to develop the technology of natural-directed reproduction, growth and development during the breeding period and growth in the first summer, for one of the economically valuable fish species, with great appreciation on the domestic and foreign markets , with wide export prospects and much sought after by sport-recreational angling enthusiasts, namely the pikeperch (*Sander lucioperca*, Linneaus, 1754).

The purpose of the development of these technologies is to support aquaculture specialists, who have or want to have as their activity the production of stocking material from the pikeperch species, necessary for the stocking /restocking of natural waters and fish ponds.

Starting from the current state of existing knowledge, both nationally and internationally regarding the reproduction, growth and intensive development in protected spaces of the pikeperch during the fry period and growth in the first summer, the research program and the experimental works carried out at the Fish Culture Research and Development Station Nucet, in the period 2018-2019-2020, included a set of activities of a fundamental nature, aiming at reproductive biology, gametogenesis, embryonic and post-embryonic development, as well as growth in the first summer of this species, under the general conditions of Romania and specific from Nucet.

The experimental works have an applied, competitive and pre-competitive nature and have solved the following main objectives:

→ the development of technology in the field of natural-directed reproduction of the pikeperch species with and without hormonal stimulation of the genitors;

→ developing the technology of intensive growth and development of the pikeperch during the fry period, with the administration of live food and/or granulated fodder, as well as establishing the optimal stocking densities;

→ the development of the growth technology in the first summer, in monoculture, with the administration of live food and/or granulated fodder, in order to obtain biological material with medium weights and dimensions at which the species can be stocked in natural habitats or in fish farms, with as few technological losses as possible.

In order to achieve these objectives, a series of experiments were carried out, with the aim of natural-directed reproduction with the achievement of a maximum percentage of mature females and the evaluation of the growth performance of the pikeperch during the fry period and the first summer, in the context of the manipulation of some essential factors of technologicalnature, such as: type of feed and stocking density.

The results obtained following the conduct of the experiments lead to a series of relevant conclusions, from a technological and application point of view, as follows:

I. The experiments regarding the development of the natural-directed reproduction technology of the pikeperch species, with and without hormonal stimulation of the genitors, highlight the following:

from the experiments performed, the best results were obtained in the V3 variant (stimulation with the synthetic hormone Nerestin 5A), in all three years of the study (2018-2019-2020), where the average percentage of female maturation was 95,6%. In the other experimental variants, the average maturation percentage values were 77.7% in the V2 variant (stimulation with carp pituitary gland) and 71.1% without hormonal stimulation;

the choice of the hormone is made according to the obtained technological indices, but also the price of the product and market availability. Nerestin 5A is a synthetic hormone, the price is below that of CPG (carp pituitary gland), it is easy to use and store unlike carp pituitary gland which is difficult to prepare and has a very short shelf life. Without hormonal stimulation, reproduction can be achieved with more modest results, situation in which the costs for the hormonal substance no longer exist, and part of the manipulations of the biological material in terms of injection no longer takes place;

the results of the tracked indicators from nest harvesting to hatching, were similar due to the good quality of the technological water used in the incubation process, the preventive treatments carried out against fungal infestation, as well as the optimal temperature interval in which the incubation was carried out;

the survival percentage of larvae up to the age of 7-8 days was between 63.9% (2020) and 70.2% (2018). Good results were also obtained in the variant without hormonal stimulation, the survival percentage of larvae up to the age of 7-8 days was between 61.8% (2020) and 64.1% (2018);

due to the fact that the pikeperch eggs were incubated in an optimal temperature range (11-15°C), the hatching time was average (7-8 days), resulting in homogeneous batches, a fact that was later reflected in the results obtained in larval growth.

II. The experiments regarding the elaboration of the development technology during the fry period in the pikeperch species bring out the following conclusions:

→ Regarding the effect of stocking density on the growth and development of thepikeperch during the fry period, the researches that have been carried out emphasize the negative effect of high stocking densities on the growth performance, not as a consequence of the decrease of water quality, but due to the installation of physiologically stress with the development of the fry, manifesting an aggressive behavior. Thus it can be stated that not all the mechanisms by which stocking density affects growth performance are known.

As previously mentioned, the main purpose of this experiment was the elaboration of the technology for the growth and development of the pikeperch during the fry period and, as a subsidiary, the determination of an optimal stocking density, under the conditions of a protected aquaculture system, so as to obtain biological material that can be stocked without the risk of significant numerical losses.

Following the growth and development experiment during the fry period with the influence of the stocking density effect, a series of conclusions resulted:

the physico-chemical parameters of the water recorded values that are within the optimal technological interval, without significant differences between the used stocked densities;

from the analysis of the growth performance indicators, it can be seen that the pikeperch in the stocking density of 1000 ex/tank, recorded better growth than the other two stocking densities of 2000 ex/tank and 3000 ex/tank;

from the point of view of other indicators such as: daily growth rate [GR], real growth rate [Sr], specific growth rate (SGR), they recorded similar values, without significant differences;

within this experiment we can specify that the stocking densities approached did not lead to the appearance of variability in the crop biomass, obtaining homogeneous populations in proportion of 86.9-93.9%;

the lack of obvious differences in the feed conversion rate, as well as its slightly high values, is justified by the fact that in all experimental variants the feed was administered ad-libitum, so that the biomass has easy access to it, because it is the most difficult period of growth and development, but also to avoid the phenomenon of cannibalism;

from a statistical point of view, the fish populations obtained within the experimental variants had values that showed homogeneous populations. There were small variations that did not significantly influence the final results;

the coefficient of determination R^2 recorded values between 0.869 and 0.939 which signifies a good correlation between the variables studied and leads to the statement that the increase in body mass can be attributed to increases in length and varies from 86.9% in the V3 variant to 93.9% in the V1 variant;

analyzing the values of the "b" coefficient, an isometric increase of the biological material obtained in the V1 variant was observed in all the years of the study, where "b" had values between 2.906 and 3.068, and a negative allometry in the experimental variant V2 where "b" a recorded values between 2.422 and 2.722, and in the V3 variant the increase in length favors that of body mass, the "b" coefficient recording values between 2.098 and 2.421;

from the overall analysis of the coefficient of variability (CV) in terms of variable body mass, it can be seen that administration of feed with a high percentage of protein led to an increase in the variability of the fish batches;

also, it is noteworthy that, in the case of batches with high density (2000 ex/tank and 3000 ex/tank), the percentage of survival and average weight recorded low values compared to the variant where the stocking density was lower (1000 ex/tank);

in all the study years 2018-2019-2020, a positive correlation (according to the Pearson coefficient >0.5) is highlighted between the length and the average weight of the individuals stored in the growth and development ponds, in the case of each experimental variant (V1, V2 and V3). This phenomenon highlights the homogeneous growth and development of the biological material, which proves that the food was sufficient in all experimental variants;

analyzing the boxplot graphs presented previously, for the three experimental variants, along the three distinct experimental periods, it can be stated that V1 presented the most symmetrical distribution among the experimental specimens, both in terms of length and individual average weight recorded at the end of the analyzed production cycle;

the cluster analysis highlights 3 distinct groups, which include the biometric data strings and individual biomass values of the experimental specimens, related to each of the three experimental variants. This fact highlights the growth differences associated with the specimens of each experimental variant. The clusters explain over 70% of the data variation in the case of the experimental period 2018 and 2020, respectively over 80% in the case of the 2019 period;

the principal components analysis (PCA) highlights 3 main components, which explain more than 70% of the data variation, in the case of the three experimental variants (V1, V2, V3). The strong correlations recorded between the average individual length and the average individual biomass, at the level of the fish material within the experimental variants, are also confirmed within the PCA;

the ensemble mathematical models based on algorithms with training sets of randomly constructed trees - random forest (RF), confirm the good ability to predict the total individual length associated with the fish specimens within the experimental variants, using the average individual biomass as the main predictor. It should be noted that, in the training of the models, the reduction of the percentage of data overlap as much as possible was taken into account and the validation of the obtained results was carried out by using a volume of approx. 30% of the total database considered. Also, the characterization of the degree of precision of the parameters is dependent highlighted by the good RMSE values, namely 0.034 in the case of V1 and 0.047 in the case of V2, respectively 0.035 in V3;

Solution concluding, we can say that the best results were obtained in the V1 variant with the lowest density. Due to the fact that the pikeperch is a predatory fish, when the stocking densities are high, the attack instinct is triggered very quickly, even if the food is abundant and the biological material is of the same size, after 30-35 days of growth, the phenomenon of cannibalism appears. The significant differences in the percentage of survival and individual weight in variants V2 and V3 compared to variant V1, show that in the pikeperch species, they are influenced by the stocking density. It can be said that the pikeperch can hardly bear the high density during the rearing period.

→ Regarding the influence of the type of food on the growth and development of cultured fish during the rearing period, it has been studied for a long time by aquaculture specialists, less studied being in the pikeperch. The research carried out underlines the fact that the type of food administered to this species has an effect on growth performance, in terms of individual weight and on the percentage of survival, through the behavior of the species, which after 30 days has an increasingly acute tendency to catch the food which is in motion. Thus, it can be stated that not all mechanisms by which the type of food affects growth performance are known.

during the experiment, the quality of the technological water did not influence the aquaculture population, no significant differences were observed between the values of the physical and chemical parameters of the water, because the supply water flow was sufficient, its characteristics classifying it as a good water for aquaculture;

with regard to the individual mass of the pikeperch obtained at the end of the growth experiments, significant differences were observed between the experimental variants, thus the very good values recorded in the V2 variant, where Prussian carp(*Carassius gibelio*) larvae were administered, were highlighted, good values in the V1 variant, where zooplankton was administered and lower but not negligible values in variant V3, where granulated feed was administered;

the biotechnological indicator regarding the number of fry obtained per experimental variant, being primarily dependent on the type of food administered, it is obvious

that in the variant where live food was administered, the percentage of survival was positively influenced;

from the point of view of the specific growth rate [SGR], similar values were recorded, without significant differences;

in terms of other indicators such as: daily growth rate [GR], real growth rate [Sr], protein efficiencyrate [PER], survival percentage [SV] and feed conversion rate [FCR], they registered different values, which shows that the administration of food as close as possible to that in the natural environment gives it an advantage in terms of plasticity and at the same time minimizes the tendency to manifest the phenomenon of cannibalism;

the appetite of the pikeperch in variants V1 and V2 (with live food administration) was higher, a fact observed during the experiments and confirmed at the end of the postembryonic period by the higher average weight obtained in these variants;

from a statistical point of view, the fish populations obtained within the experimental variants had values that showed homogeneous populations. There were small variations that did not significantly influence the final results;

the coefficient of determination R^2 recorded values between 0.861 and 0.927 which means a good correlation between the studied variables and leads to the statement that the increase in body mass can be attributed to the increase in length and varies from 86.1% in the variant V3 at 92.7% in the V1 variant;

analyzing the values of the "b" coefficient, an isometric increase of the biological material obtained in the V2 variant was observed in all the years of the study, where "b" had values between 2.896 and 3.056, a slightly negative allometry in the experimental variant V1 where "b" a recorded values between 2.711 and 2.888, and in the V3 variant the increase in length favors that of body mass, the "b" coefficient recording values between 2.651 and 2.786;

in all the study years 2018-2019-2020, a positive correlation (according to the Pearson coefficient >0.5) is highlighted between the length and the average weight of the individuals stored in the growth and development ponds, in the case of each experimental variant (V1, V2 and V3). This phenomenon highlights the homogeneous growth and development of the biological material, which proves that the food was sufficient in all experimental variants;

analyzing the presented boxplot graphs, for the three experimental variants, along the three distinct experimental periods, it can be stated that V1 and V2 presented the most symmetrical distribution among the experimental specimens, in terms of the average individual biomass recorded at the end of the analyzed sequences from the production cycle. Also, the distribution of the values associated with the average body length of the experimental specimens reveals a weak symmetry, a fact that can be attributed to the phenotypic differences that manifest at the level of the biological material, from the first stages of development;

the cluster analysis highlights 3 distinct groups, which include the biometric data strings and individual biomass values of the experimental specimens, related to each of the three experimental variants. This fact highlights the phenotypic and growth performance differences associated with specimens of each experimental variant. The clusters explain over 68% of the data variation in the case of the experimental period 2018 and 2020, respectively over 69% in the case of the 2019 period;

the principal components analysis (PCA) highlights 3 main components, which explain more than 68% of the data variation, in the case of the three experimental variants (V1,

V2, V3). The strong correlations recorded between the average individual length and the average individual biomass, at the level of the biological material within the experimental variants, are also confirmed within the PCA, especially within V3 in 2018, V1 in 2019 and V2, V3 in 2020;

thus, in 2018, models based on RF algorithms provide an accurate prediction of individual length within the variation range of average individual biomass [0-2.77], while in 2019 the optimal prediction range is [0-2, 88]. In the year 2020, ranges of average individual biomass were determined that provide a low degree of prediction [2.1-2.23; 2.69-2.81]. The data sets from the years 2018 offer the most accurate prediction, a fact also confirmed by the Rsq value (0.934), compared to 0.937 in the case of 2020 and 0.941 in the case of 2019. Also, the characterization of the degree of precision of the dependent parameters is highlighted by the good values of RMSE, namely 0.034 in the case of 2018, 0.047 in the case of 2019, respectively 0.035 in 2020;

the main conclusion regarding the **influence of the type of feed**, which emerges from the results obtained in the experimental works on growth and development in the pikeperch during the rearing period, showed that the species can also be fed with granulated feed, even if the best results were obtained by administering live food (Prussian carplarvae).

→ In both growth experiments, <u>in different densities and with different feed</u> <u>administered</u>, after about 35 days the phenomenon of cannibalism began to manifest. The pikeperch procures its food with the help of sight and attack speed. Even if you have created very good growth conditions through: the quality of the technological water and the superior quality and quantity of the administered feed, the predatory instinct is triggered very quickly especially when the feed is administered (the fish population is on the move) and does not hesitate to attack peers, even if they are the same size or larger. Such episodes were observed more often after the age of 35 days, the bite in 95% of cases was in the head area or near it. In some cases, after the attack, it cannot swallow the prey, it remains with its mouth stuck on the prey for about 1-2 minutes, thus 80-90% of the attacked specimens died, due to the caused injuries.

→ The growth and development in an intensive system of pikeperch (*Sander lucioperca*, L., 1758), during the fry period, is superior to other systems practiced until now, for the following reasons:

- requires small areas;

- allows high stocking densities;
- continuous control over water quality;
- control over the consumption of administered food (live food/fodder);
- control over the growth and health of the biological material;
- higher growth rate.

The observations and data on the growth and development of pikeperch larvae in the post-embryonic period in intensive system in the "Ewos" type tanks and the morpho-physiological characteristics observed macro and microscopically are:

I. At the age of 7-10 days:

- existing yolk sac but with small dimensions;
- gill buds without being covered by opercula;
- transparent body through which the brain and the primordia of the spine are observed;
- the larvae swim easily.

II. At the age of 17-18 days:

- less transparent body with a tendency to colorate to yellowish, with pigment spots that cover almost entirely the surface of the body;

- well-developed mouth located in the ventral position;

- teeth are observable on the lower jaw in the incipient phase of formation;

- the intestine formed from the larval stage has a more accentuated curvature in the ventral area of the stomach;

- the larvae swim easily in the mass of the water in search of food;

- food consisting mainly of supplementary food and juvenile forms of plankton (algae, rotifers, cladocerans, less adult forms, organisms entered into the water with the supply water from the settling tank through the filtration system).

III. At the age of 27-28 days:

- the body is no longer transparent, the pigment spots of brown color, cover almost the entire surface of the body;

- the fins are almost entirely formed, which allows the larvae to swim quickly in search of food;

- the stomach is well individualized;

- intestinal contents that highlight the presence of live food, that of additional feed and fragments of cladocerans from the settling tank water.

IV. At the age of 40 days:

- body shape resembling to that of the adult;

- specific color (green-gray back, less often yellow-gray, the sides are gray-silver, with darker stripes, arranged transversely, the abdominal region with a lighter color);

- body completely covered with scales;

- alert swimming specific to predatory fish species;

- tendency to manifest cannibalism phenomenon;

- the stomach content reveals the presence of the feed in proportion of 90% in different phases of digestibility and the remaining 10% live food.

III. For the third experiment regarding the growth of pikeperch in first summer, in monoculture, in earthen ponds, the following aspects were concluded:

the research carried out underlines the fact that the type of food administered to this species has an effect on growth performance, in terms of individual weight and on the percentage of survival, through the superior results obtained in the V2 variants compared to the V1 variant;

the main bioindicators monitored during the growth experiments in the first summer, in the pikeperch (*Sander lucioperca*, L. 1758) were: individual weight (W g/ex); correlation between body mass and total length; real growth rate (Sr); daily growth rate (GR); feed conversion rate (FCR); protein efficiency rate (PER) and survival percentage (Sv),these recording different values. This was due to the fact that the fry prefers live food and consumes it with pleasure, being a complete, attractive food intended for this active predatory species;

Solution which gives it an advantage from this point of view;

the fish populations obtained within the experimental variants had values that showed homogeneous populations. There were small variations that did not significantly influence the final results;

the coefficient of determination R^2 recorded values between 0.874 and 0.946 which signifies a good correlation between the variables studied and leads to the statement that the increase in body mass can be attributed to increases in length and varies from 87.4% to 94, 6%;

analyzing the values of the "b" coefficient, an isometric increase of the biological material obtained in the V2 variant was observed in all the years of the study, where "b" had values between 2.968 and 3.126 and a slightly negative allometry in the experimental variant V1 where "b" recorded values between 2.531 and 2.701, the increase in length disfavors that of body mass.

The research carried out at S.C.D.P. Nucet, had appreciable results and may have a special technological and economic importance for the development and diversification of the aquaculture activity in Romania.

The results obtained and their implementation in practice respond to the national aquaculture development priorities through:

• the quantitative and qualitative increase of aquaculture production, in order to ensure on the Romanian and EU market, with quality products, in accordance with national legislation aligned with European norms (91/493/EEC and 93/53/EEC);

promoting responsible aquaculturetowards the environment and its components;

• ensuring the degree of profitability of aquaculture farms in the context of the competitive mechanism of the market economy.

The results obtained during the research and experimental works were disseminated both through the means specific to the research activity: works and articles presented at scientific events and published in specialized journals, as well as through the utilization of a quantity of stocking material obtained as a result of the reproduction, the development and growth of the species.

Summarizing the previously written results, it can be concluded that through the topic addressed, the present paper constitutes an important step in the development of pikeperch aquaculture in Romania. As a result of the researches addressed, a series of appreciable and novel information emerges.

Thus, at S.C.D.P Nucet, the activity of documentation, research and the development of experimental works was carried out, which contributed to the preparation and writing of the doctoral thesis and having the following personal contributions:

• experimental works of natural-directed reproduction of the pikeperch. Hormonal stimulation of pikeperch females was carried out with carp pituitary gland (CPE) and Nerestin 5A under pond conditions;

• experimental works of growth and development during the fry period in the pikeperch, in an intensive system, in protected spaces ("Ewos" type tanks), based on the criterion of stocking density and the type of food administered;

• experimental works on the growth of pikeperch in the first summer, in monoculture, in small earthen ponds, according to the criterion of the type of food administered.

The results of the experimental work on the "**Reproduction and growth of pikeperch** (*Sander lucioperca* L., 1758) in industrial aquaculture systems" are a continuation of the tradition and experience in the field of reproduction and growth during the fry period of some fish species with high economic value : common carp, Asian cyprinid species, *Polyodon spathula* and predatory species, carried out over time at the Fish Culture Research and Development Station Nucet by many researchers, who had a particular impact on the development of aquaculture in Romania.

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