GUIDE TO THE SMALL SCALE ARTIFICIAL PROPAGATION OF TROUT

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GYÖRGY HOITSY

PREFACE

This book aims to provide basic information on reproduction and propagation of trout, tries to describe most important hatchery activities and challenges and also provides advices on the rearing of trout fry in order to support small scale farmers and producers. As small scale fish farming is a substantial opportunity to generate income and also provides a valuable protein source it is important to afford knowledge about the proper technology.

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1 INTRODUCTION

Small scale trout farming is a realistic income generating alternative in the mountainous regions of Central and Eastern Europe, the Caucasus and Central Asia where income sources and employment opportunities are scarce or even missing.

Though all female eyed eggs of well performing pedigree trout strains can be purchased from many different locations of the world propagation of locally maintained trout can still be technically and economically feasible. Increased official opposing of introducing fish strains with genotypes new in a region is also an additional reason why practical knowledge of artificial reproduction of rainbow trout may be very useful.

It is expected that technical information presented in this book will help in the reproduction of other trout species. It may also successfully complement recently produced FAO publications such as Small scale rainbow trout farming, Small scale trout processing methods and Trout farming based angling tourism.

In order to satisfy interests for further details a glossary is compiled and tables and annexes are attached to this paper. For the sake of finding additional information asterisk symbols (*) are used after words in italics which are explained in the glossary.

2 REPRODUCTION OF TROUT IN THE NATURE

2.1 REPRODUCTION STRATEGY

Not only growth but also the successful reproduction of fish depends on the surrounding environment. For this reason different species of fish develop and practice different reproductive strategies which, in a characteristic way to species, ensure a successful offspring production even under fluctuating environmental conditions.

Reproductive strategy of fish includes reproductive age, fecundity according to size and age, size of eggs, reproductive behavior and the number of times spawning occurs in the lifetime of females (Bond, 1996).

2.1.1 Reproductive age and the number of times spawning occurs in a lifetime

In different fish species the length of time in which males and females become sexually mature may vary between a few months and several years. The actual sexual maturation of trout depends on species, sex and environmental conditions (water temperature, feeding conditions, etc.) in which fish live and develop (see Table 1).

Table 1: Reproductive age and period of selected trout species

Species	Sexual maturation (years)				Reproductive period (years)	
	In nature		Within farm conditions		Within farm conditions	
	Females	Males	Females	Males	Females	Males
Rainbow trout	3–4	2–3	2–3	(1)–2	4–6	6–7
Brown trout	3–4	2–3	3	2	4–6	6–7
Brook trout	3–4	2–3	2–3	(1)–2	2–3	2–3

2.1.2 Fecundity

*Fecundity** of egg-laying bony fish species depends on their *parental care** and the size of produced eggs. Fecundity of fish is expressed with the absolute and relative number and the weight of eggs produced by a female. Table 2 summarises the fecundity of widely and frequently produced trout species.

Source:
1. Bascinar and Okumus, 2004,

Table 2: Size at sexual maturation, fecundity and duration of egg and sack-fry incubation of selected trout species

Species	Size of broodfish		Fecundity of female (1000 eggs)		Duration (D°)		
					Incubation of egg		Swim-up
	Females (kg)	Males (kg)	Absolute (egg/fish)	Relative (egg/kg *BW**)	Up to eyed-egg stage	Total	
Rainbow trout	1–7	1–4	1–10	1.6–3.0	160[1]	310[1]	500[1]
Brown trout	1–6	1–4	0.5–8	1.6–3.6	195–273[1]	250–406[1]	380–610[1]
Brook trout	1–3	1–1.5	0.5–5	1.2–2.5	195–276[1]	235–444[1]	338–675[1]

2.1.3 Size of eggs

Egg sizes of fish indicate the size of yolk-sack in developing embryos and non-feeding larvae which are also called sack-fry (see Figures 9).

Salmonid eggs are the largest among the eggs of bony fishes. The size of their dry eggs varies between 3.7 and 5.2 mm (32–100 mg) (Hoitsy, 2002). Egg size of younger and smaller females may be smaller than that of the elder and larger ones. Eggs of 5–6 year-old females are the largest but the quality and quantity of feed also influences the actual size of them The volume of 1 000 eggs may be 79–90 cm^3.

The large size of trout eggs can be explained with the long duration of egg and sack-fry incubation.

2.1.4 Reproductive behavior

Reproductive behaviour is a complex sequence of sexually mature female and male fish which aims to ensure the highest possible survival of laid and fertilised eggs and hatched non-feeding larvae.

At trout reproductive behavior includes timing of spawning, selection and preparation of spawning ground, courting and mating with selected males.

By mating season eggs and milt should be ripe, ready for being fertilized (eggs) or to fertilize (milt). Spawning in the right place and time when both eggs and milt are ripe and all environmental conditions are favourable is the final response of the *endocrine** system of fish to environmental cues. These are physical (temperature, clearness, speed and depth of water) and chemical (clean water, rich in oxygen) qualities of water, light, day-length, suitability of mating ground and presence of the opposite sex (Woynarovich and Horvath, 1980 and Bond, 1996). In other words favourable environmental conditions stimulate production, development, final maturation and ovulation of eggs (see Box 1).

Development and release of milt in males is also influenced by environmental conditions but nature built less endocrine control into this process than into the one for females.

2.2 SPAWNING

Most of the trout species are *diadromous**, more precisely *anadromous** fishes. Still, those *riverine** and *lacustrine** trout species which habituated fully to freshwater environment, hence never move into the seas, also migrate for spawning to upper sections of rivers and their tributaries.

Shortening days and decreasing water temperature affect hormone concentration of brood fish. This causes rhe development of secondary and accessory sex characteristics, drives reproductive behaviour and triggers reproductive activities.

Light conditions have a very important role in the reproductive behaviour and activity of trout. Shortening days stimulate development of eggs and activate instincts of spawning migration. On large broodfish farms where the objective is a continuous, year-round propagation and eyed-egg production light conditions are also controlled in order to stimulate and synchronize ovulation in females.

Changes of water temperature also primarily stimulate and synchronise ovulation in females. If it decreases autumn spawning strains, while if increases spring spawning strains will prepare for spawning. If season specific trend of water temperature changes (increase in autumn and decrease in spring) preparation for spawning will stop.

Dissolved oxygen (*DO**) content of water is also a prime environmental condition which drives and influences the spawning of trout. Trout are mostly autumn spawning fishes since they are the most DO demanding fish species therefore a high DO is needed for the proper development of their embryos and sack-larvae. This environmental condition will more likely occur during winter as DO content is higher in colder waters. Swim-up of fry is usually finished by the end of the winter which reduces possible losses that melting snow and spring floods may cause.

Water current conditions, water depth and bottom types have less role in the final maturation and ovulation of eggs.

For the above described reasons trout spawn seasonally. Actual spawning season depends on species and strains hence it might be either in autumn or in spring. Spawning of trout takes place in the headwaters and tributary streams of rivers or in the inlet or outlet streams of lakes (Edwards, 1989).

Spawning of trout is wonderful which can be observed in cold mountain rivers and larger streams where they leave. Females and males swim up to the upper sections of rivers and streams and they easily pass rapids as high as 0.5–1 meter. They swim in the dark during evening and

Development and ovulation* of eggs in sexually matured bony fish

By about the first months after swim-up and starting external feeding of young fish fibre-like initials of the *gonads* (ovary* and testis*)* are already formed.
In this stage the ovary already contains the primitive egg cells (oogonium, ovogonium or archovogonium) from which the eggs will develop when the fish reaches sexual maturation. Evidence of sexual maturation is when a first production cycle of eggs suitable for ovulation and *fertilization** occurs. Development of egg batches in the ovary before each spawning is a precondition of successful reproduction. This process has different subsequence phases:

1. A batch of primitive egg cells (oogonia) in the ovary transforms into *primary oocytes**.
2. A *follicle** is formed around each primary oocytes.
3. Eggs undergo a quantitative development called *vitellogenesis** when yolk builds into the eggs in four subsequent steps. This phase is a longer procedure which is completed before the spawning season. During vitellogenesis a properly balanced feeding of females is essential.
4. Though eggs are ready for final maturation and ovulation when the vitellogenesis is completed still there is a *dormant (resting) stage** in their development. The reason for it is that the future of fertilized eggs depends on the timing of spawning when environmental conditions are most suitable for laid and fertilized eggs.
5. Dormant phase of eggs in the ovary ends when environmental conditions become favourable for spawning. At this time favourable environmental stimuli arrive into the brain of fish through sense organs. A complex *neuro-endocrine process** starts in the brain which results in the ovulation of eggs and spawning of fish.

Depending on fish species the above described procedure may be continuous within a well defined shorter or longer period of the year or it can be seasonal as it is in case of trout.

After: Woynarovich and Horváth, 1980

Box 1

night time in order to reach their spawning ground by dawn.

At the spawning ground females choose the most suitable location where the bottom is covered with pebbles and the water is swift. Here they make their shallow nests (also called redd) by cleaning an area of about 0.5–1 meter diameter. These hollows on the bottom of streams and rivers indicate the presence of an active trout population. When nests are ready their bright clean pebbles will contrast with surrounding bottom covered with greenish or brownish algae.

Males follow females. This time they are aggressive and fight for females. The strongest male occupies the nest with the female where they mate in the early hours of the day.

At mating the female stops in the front while the male stands behind her. Then the female bends in a "C" shape and presses the ovulated eggs out with a *peristaltic** movement of her muscles. When the female positions herself and is prepared to release the eggs the male joins. He also bands in a "C" shape close to the side of the female and fertilizes the released eggs. They have to be fast and accurate because only a very short period of time (sometimes even seconds) is available to fertilize the released eggs which are quickly drifted away by currents.

One spawning nest may be used by more than one couple. Trout do not guard their nests but males may remain around and chase away intruding fishes. Later they also leave the site.

Some publications of trout spawning describe trout as a fish which covers its eggs with fine pebbles or even by sand. This is incorrect because trout has the most oxygen-demanding eggs therefore any action to cover and hence cut them off from open, oxygen rich water currents is rather unlikely.

3. REPRODUCTION OF TROUT IN FISH FARMS

Trout becomes sexually mature in fish farms sometimes even earlier than in nature if water temperature is higher and feeding is better than there.

As many other fish species, trout also can reach sexual maturation in fish farms. Females and males will produce eggs and milt (*gametes**) but they can not spawn successfully under regular fish farm conditions unless all required favourable environmental conditions are simulated. Though, reliable production of large quantities of fertile eggs and hatched larvae would be both difficult and expensive on this way. Therefore, there are other more efficient ways of *artificial propagation** of trout which result in the ovulation of eggs stripped and fertilized with milt from males.

3.1 PREPARATION FOR PROPAGATION

On a fish farm the sign of the arriving propagation season is that sexually matured females and males gather at the inflow of their tank near to the water surface and often try to jump against the water current. This indicates that females and males are ready to migrate to spawning grounds. At this time females and males should be separated. Otherwise already ripened brood fish will spontaneously spawn while others will pick and consume released and fertilised eggs from the bottom of the tank where this uncontrolled spawning takes place.

In trout sexes can easily be distinguished during the propagation season (see Figure 1). Therefore separation of females and males can be safely completed:

Males are slimmer and their back is higher and humpy. Their colours are brighter. Their lower jaw is pointed and wedge shaped. At elder age lower jaw takes a hook shape and is covered with swollen growths. Their pointed urogenital papilla sticks out from which white milk-type sperm bursts if the abdomen is gently pressed.

Females are more round and filled at the belly because of their enlarged ovary. Their urogenital papilla sticks out about 1–2 cm and is top is rounded (see Figure 1).

Fishing and sorting of brood fish should be done gently with care in order not to stress and hurt the fish which are full of eggs and milt.

On large tout farms which are specialized on propagation, ovulation and spermiation are induced with hormones such as salmon *hypophysis** or Gonadotropin Releasing Hormone Analogues (*GnRH/A**). However, most of small scale trout farms do not use hormones to harmonise and complete ovulation in females and spermiation in males. These fish farms imitate favourable environmental conditions which stimulate both final maturation and ovulation of eggs in females. If a fish farm is successful in the simulation of most important favourable environmental conditions then less fishing and handling efforts will result in a higher quantity and better quality egg production. This will also save brood fish from unnecessary stresses when their readiness is checked.

The best way for simulation of favourable environmental conditions is when water level is lowered and speed of water is increased in freshly cleaned tanks where females and males are separated. If it is done properly, about 50–70 percent of females will be ready for stripping 7–10 days after their separation. In case of autumn spawning females a slight reduction, in case of spring spawning females a slight increase of water temperature is also an effective way to imitate environmental conditions which will advance ovulation.

Figure 1: Differences between females and males during reproductive season

Those females which do not react on the described manipulation of keeping conditions will perform a lengthy propagation period when a successful stripping will remain less predictable.

3.2 REARING AND FEEDING OF BROOD FISH

If eggs, fry or young fish are not purchased but are produced on a fish farm the entire success depends on the quality of brood stock which is kept and propagated there. Only those healthy females and males should be propagated which have a proven good life time performance and are able to inherit them. Most advantageous traits are size, utilization of feeds, growth rate and overall resistance to stress and diseases.

It is a general rule for a positive selection of brood fish that good *genotypes** should be searched among fish with good *phenotypes**. It means that when a fish is selected for propagation its appearance, physiological properties and individual performance should be considered.

It is advantageous to select future brood fish from a stock which is about 10 month old. The most properly looking, largest and healthiest specimens from the same age group should be selected for future propagations. From this time on selected and separated stocks of future brood fish should receive a different, less protein rich diet. If fish receive high protein diet their muscles and body will grow instead of their gonads. Today well balanced feeds are widely available for this purpose where all the proteins, energy, vitamins and minerals are contained in the right quantity and proportion needed for growing and grown-up brood fish. Sometimes also other ingredients are added in order to *change the colour of the fertilized eggs**.

Under farm conditions male trout become sexually mature in the second year, while females in the thirds year with the exception of brook trout. About 60–70 percent of brook trout females become sexually mature already by the second year. In nature both males and females need 1–2 more years to reach sexual maturity. About 6–7 brood fish should be stocked per 1 m^2 of tank area.

There are no specific criterions of a good brood stock rearing or keeping tank. It should be suitable for being cleaned and kept cleaned easily to avoid accumulation of faeces and unconsumed feeds. It should also be suitable to change water in it about 2–10 times per day.

On many fish farms males and females are kept separately during the entire year. Such practice has no considerable advantages if basic reproduction related physiological and ethological aspects are in focus. It is true that males and females should be separated before the propagation season. Still, some young males placed in the tank of females before propagation will help advancing and synchronizing ovulation in females. Even after the most proper *stripping** of a female a little portion of ovulated eggs will remain in the fish. If females cannot release these eggs and they remain in the fish they will negatively influence egg production next year and eggs remained in the female may decay and even cause mortalities. Therefore, if these females are kept together with males they will drive out even the last drops of eggs from them. Males are very aggressive during the propagation season. If females are mixed with males after stripping, aggressiveness of males will reduce. Therefore, losses due to fighting and wounds will reduce too.

3.3 PREPARATION OF THE HATCHERY

Parallel to separation and preparation of brood fish for propagation fish hatcheries should also be prepared. It means:

- Checking, repairing and cleaning of both water supply and drainage systems.
- Cleaning of brood fish tanks, trolleys, hatchery devices and equipment.
- Making ready weighing and measuring devices.
- Completing and cleaning stripping bowls and towels.

Disinfection of the water supply and drainage systems and hatchery devices can be done with formalin.

3.4 WORKS IN THE HATCHERY

Works in the hatchery include the choosing of suitable broodfish, stripping, fertilization and incubation of eggs and rearing of hatched fry.

3.4.1 Selection of suitable broodfish and stripping of eggs and milt

There are different techniques for removing ovulated eggs and milt from broodfish. Elaboration of the first technique of artificial propagation of trout copied natural spawning. It was done by a German fish farmer, Jacobi between 1763 and 1765. He stripped eggs into a jar of water. Today nobody follows this “wet” technique because the expectable rate of fertilization is as low as 20 percent.

In 1856 Vranszkij Russian fish culturist started a “dry” technique. Since this time this method has been successfully practiced because it ensures 98–100 percent fertilization. The essence of this technique is that first eggs then milt are stripped into a dry bowl where they are gently mixed before fertilization starts with the adding of water.

The first step is the separation of those females which already ovulated. Signs of ovulation are an enlarged and softened abdomen. Eggs in the female can be felt when gently touching and the urogenital opening sticks out about 1-2 cm (see Figure 1).

There are different techniques for stripping ovulated eggs. According to the Australian one air is pumped into fish with a syringe and this is what presses eggs out of the female. The Swedish technique uses a double walled rubber pipe. Female is placed into this tube then water is injected between the walls of the tube. The inner soft wall covers the entire body which gently presses out the ovulated eggs from the female.

A most widely practiced technique is hand stripping. This technique is simple. Head and tail of fish are wrapped into handtowels and the female is firmly but gently hold at her two “ends” on a way that head is 45 degree upward. This position of fish allows driving the ovulated eggs with a gentle massage with the thumb and forefinger toward the urogenital opening from where eggs will directly pour into the stripping bowl.

In case of rough handling and unprofessional stripping fish may be hurt or even injured. Therefore, instead of stripping everywhere from head to tail it should be restricted only to the lower section of the belly. The stripping hand should not start higher than an imaginary line between the dorsal and the pelvic fins. If fish is stripped above the mentioned imaginary line its internal organs such as spleen or liver may be damaged which can result in mortality. Another reason why females should be stripped at the lower section of their bellies is that the ovulation of eggs starts in the lower section of the ovary. Hence, there is no reason for stripping fish along the entire body.

Milt of males is stripped similarly to eggs. Gentle handling and stripping is very important here, too.

Box 2

Structure of trout egg and sperm

Egg: It has an *animal pole** and a *vegetal pole**. Egg shell of brown trout is about 33–37 microns thick while this is thinner at rainbow trout. On the egg shell about 1 micron pores can be observed which continue in narrow ducts. Microphyle through which sperm enters is situated on the animal pole of the egg. After the first sperm enters it closes but without the entering of any sperm the microphyle will also close within about a minute when the egg starts swelling. Perivitelline space separates shell from the cell which fills with fluid within 20–60 minutes after fertilization. This fluid filled space allows the embryo to rotate (gyrate) freely in the egg shell and to remain always in the right position. During swelling the volume of egg increases with about 12–20 percent. Trout egg is not adhesive but may stick to a substrate or to the wall of the hatchery devices until fully swelled.

Getting into the egg the *germinal disk**, the vitelline membrane and yolk can be found. The yolk is a dense yellowish liquid which contains *globulin** and oil droplets. These oil droplets accumulate in the upper animal pole of the egg in order to keep the germinal disk at the right place and in the correct position.

Sperm: Head of the sperm is 1.7 by 2 microns and its tail is about 25–35 microns long. Active life of sperm starts when ejaculated into water and lasts for about 24–40 and 40–50 seconds at brown trout and rainbow trout, respectively.

Figure 2: Hand stripping of a female

Figure 3: Stripping of milt onto eggs

Figure 4: Fertilization (mixing of eggs with milt)

Figure 5: Adding water to fertilized eggs

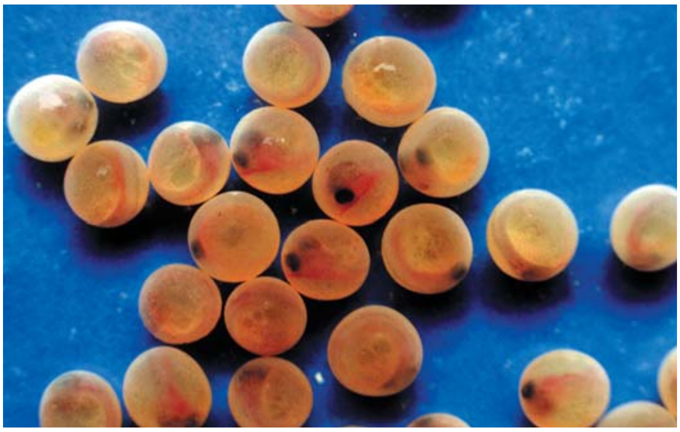

Figure 6: Eyed eggs

Figure 7: Hatched larvae

Quick and gentle stripping of eggs is essential. Where larger quantities of females are stripped MS 222 tranquilliser is used. Recently alternative, less expensive materials are researched and used, among others clove oil.

Not only brood fish but also stripped eggs should be handled with care. If they fall from a higher distance into the stripping bowl or they are steered and mixed with milt roughly they may loose their fertility and eggs will die.

Trout ovulate in batches with intervals. About 2-5 days after the first major ovulation when about 75-85 percent of the eggs can be stripped a smaller second ovulation occurs. For this reason it is important either to check the already stripped females and strip them again or to place them together with males. They will drive out later ovulated eggs from the females. Otherwise ovulated but not stripped or released eggs will decay in the fish. Even if this process would not necessarily cause mortality, still it will negatively influence the quality and quantity of egg production in the next year.

About 4-6 year old and 2.5-3.5 kg large females will produce the largest quantity and best quality eggs. Egg production of elder than 6-year-old females will gradually reduce both quantitatively and qualitatively because of the accumulated effect of different stresses experienced by fish.

It is very important to avoid the contact of eggs with water before fertilization. Therefore, before stripping fish - especially their urogenital opening - should be dried with a towel.

One portion of stripped eggs, about 5 000-10 000 pieces should be fertilised with the milt of at least 2 or 3 males. This will ensure proper fertilization of all eggs even if, for some reasons, one of the males is unfertile. Males can be stripped again after about 3-7 days. Therefore milt of good males can be used for the fertilization of eggs from many different females. 1 male is enough to fertilize the eggs of about 3-8 females.

After stripping, eggs and milt should be gently mixed still without water. If all eggs are covered with the film of milt they should be left for about 1-2 minutes in order to let them be fertilized. When eggs are fertilized a little fresh water should be added then eggs should be gently washed clean with adding more fresh water to them. During this procedure bad, unfertile (white) eggs should be removed. They are white because proteins coagulate in the unfertile eggs. After washing and cleaning eggs should be placed into hatchery devices.

	Brown trout *(Salmo trutta m. fario)*	**Rainbow trout** *(Oncorhynchus mykiss)*
Sexual maturation of females (years):	3	
Sexual maturation of males (years):	2	
Proportion of sexes:	3–8 ♀:1 ♂	
Propagation season:	November–January	November–March (depending on the strain)
Eggs per 1 kg BW (No.):	1 600–3 580	1 600–3 100
Eggs in 1 kg of dry eggs (No.):	12 500–16 500	10 000–18 200
Quantity of milt per male (cm^3):	5–27	
Quantity of sperm in 1 cm^3 milt:	16 000 000	20 000 000
Fertility rate of eggs (%):	95–100	85–100
Rate of hatching from fertilized eggs (%):	90–100	75–95
Length of embryogenesis at 10 °C (days):	40–42	30–34
Duration until eyed-egg stage at 10 °C (days):	20–21	18–21
Duration of sack-fry (none-feeding larvae) stage (days):	20–28	20–21

Table 3: Key propagation data of brown and rainbow trout

3.4.2 Incubation of eggs

During the last centuries many dozens of different types of incubation vessels were invented, developed and used. They were different both in materials (ceramic, glass, wood, metal or plastic) and shape. Today Californian, Sandfort and vertical tray incubators are used most widely (see Figure 8) but cylindrical Zuger jars can also be found in many trout hatcheries.

When eggs are placed into incubation vessels - about 10 000 eggs/0.2 m^2 - they are less sensitive and bad ones should be picked out for until about an additional 36 hours. Then the first sensitive period starts and lasts until eyed-egg stage. During this period multiplying cells in the eggs are very fragile. A bigger bump on the egg may cause a malformation in the developing embryo or even mortality. Therefore developing eggs should be kept undisturbed in hatchery devices. After eyed-egg stage until hatching eggs can be transported, hand or machine sorted again and bad, damaged eggs can also be picked out. It is because eggs are hardy during this period. Then from about 48 hours before hatching they become fragile again.

During the first fragile period of eggs the only prevention against *saprolegnia** is the use of formalin in a concentration of about 0.25 ml/l. The use of specific iodine containing products is also feasible against saprolegnia.

Since saprolegnia develops on dead and decaying eggs from which it can spread to healthy ones it is very important to remove damaged and dead eggs as soon as they can be touched, regardless if they are infected with saprolegnia or not.

There are contradicting opinions and publications about light sensitivity of trout eggs. It is sure that most of trout eggs exposed to direct sunlight for a few minutes will die. Therefore diffused light or even darkness in the hatchery is widely recommended.

Figure 8: Different incubation vessels for trout eggs and larvae
1. Californian hatchery,
2. Sandfort hatchery tray and
3. Vertical tray incubator

3.4.3 Hatching and development of sack-fry

The actual time of hatching depends on water temperature but oxygen content of water also greatly influences it. Hatching below 4 °C and above 15–18 °C can only be done with great losses.

Eggs of rainbow trout and brown trout will hatch within about 520 and 320 *day-degrees** (D°). If in nature water temperature is less than 2 °C, the development of embryos stops. This *diapause** causes great losses. It is especially important because in nature, even if all environmental conditions are optimal not more than 15–20 percent of eggs will hatch and only 0.5–1 percent of fertilized eggs will survive and grow to sexually mature adults.

After fertilization, at the start of cell-division, at pigmentation of the eyes and before hatching oxygen demand of developing embryos will especially increase. During these periods insufficient quantity of oxygen will result in higher mortality or at the end of the embryonic stage as a result of oxygen deficiency earlier hatching will occur.

Before hatching embryos gradually move (gyrate) more and more intensively. This movement mechanically thins the egg shell from inside. In addition, hatching larvae decompose the egg shell with an enzyme (hyaluronidase) which is secreted by a gland found in the head of the embryo.

Eggs stripped from different females but fertilized at the same time may hatch differently. This difference may be 2–3 days. Hatching larvae tear open the egg shell with their tail and literally reverse from it. Larvae which tear up the egg shell with their head often die because the egg shell remains stuck on their head and gills, therefore they die of suffocation.

Larvae are hatched with a large yolk-sack from which they feed until switching to external feeding. Yolk-sack might make up for 2/3–3/4 of the entire weight of hatched larvae.

Hatched larvae remain laying on the bottom of hatchery devices. Under normal conditions they evenly occupy these. During incubation of non-feeding larvae egg shells, dead larvae, floating grease and oil drops should be removed from water surface. Developing non-feeding larvae does not need any other care for about 2–4 weeks (see exact duration in Figure 9). During this period only dead larvae should be removed regularly from the devices.

It is important not to expose developing larvae to strong light since then they will try to hide under each other which may cause oxygen shortage, hence mortality. It is also important to keep the rearing devices clean. This is done by an appropriate exchange of fresh water and siphoning of dead and decaying larvae. During this period larvae are sensitive to chemicals including formalin, therefore a clean rearing environment is the only option for prevention.

As larvae advance in development their yolk-sack is gradually consumed and they start to feed externally. They also start to move until finally they swim up to the water surface and gulp air from the atmosphere. External feeding starts still before the yolk-sack is entirely consumed. This few days overlapping of internal (yolk-sack) and external feeding of larvae ensures a safe supply of nutriment for swim-up fry until they learn how to feed. By the time the yolk-sack is fully consumed fry learn how to feed from their environment.

Table 4: Length of the incubation period of trout eggs under different water temperatures

Water temperature (°C)	Brown trout		Rainbow trout		Brook trout	
	Days	D°	Days	D°	Days	D°
6	77	462	55	330	80	480
8	61	488	43	344	62	496
10	41	410	31	310	40	400
12	27	324	26	312	38	456

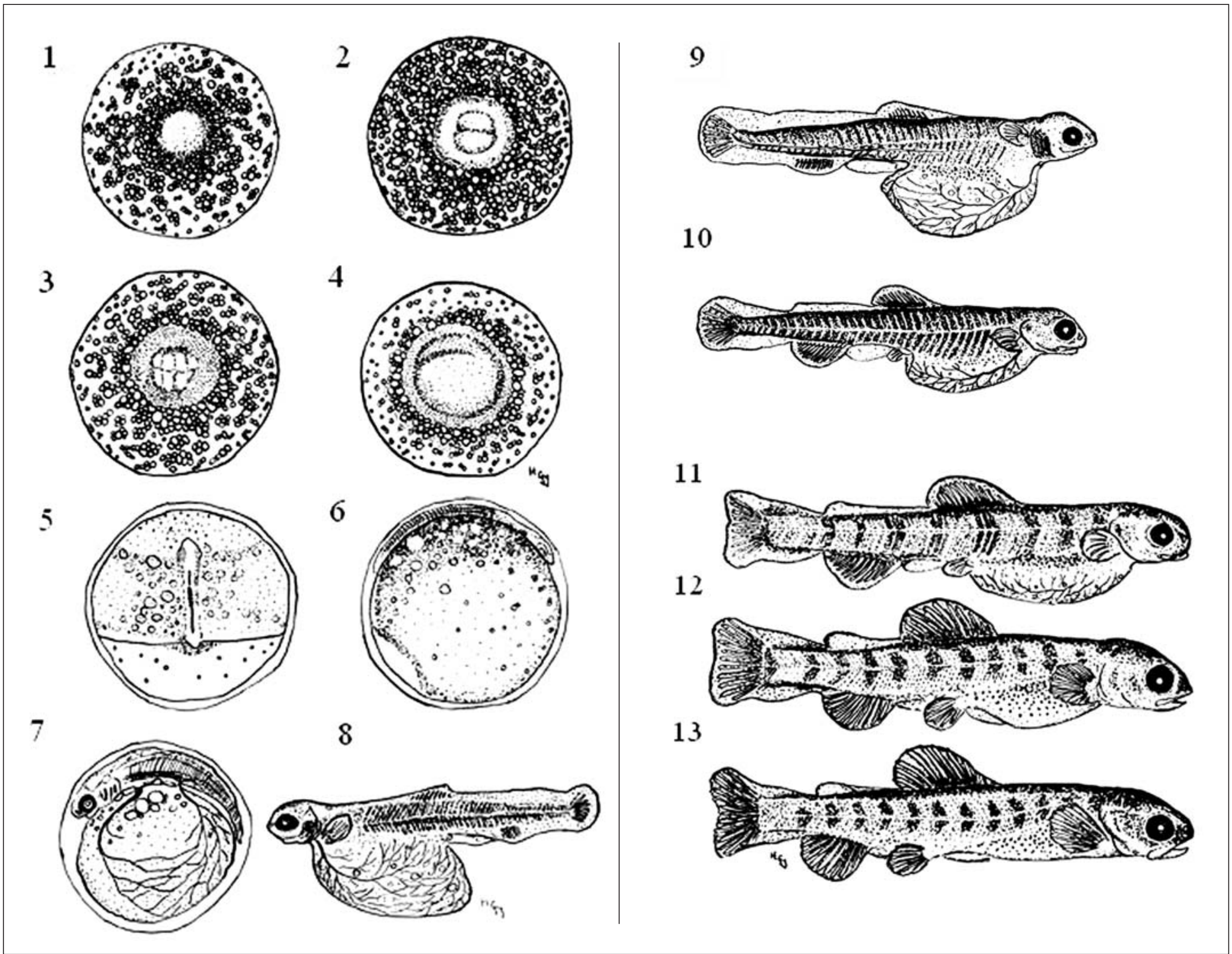

Figure 9:

Figure 9: Development of fertilised rainbow trout eggs and hatched larvae at about 10 °C

1. Moment of fertilization,
2. Early stage of blastula (7.5 hours),
3. Late stage of blastula (13.5 hours),
4. Start of gastrula stage (3.5 days),
5. Embryo is about 3.1 mm with the initiatives of eyes (7.5 days),
6. Embryo is 5 mm, the heart starts to beat (12 days),
7. Embryo is about 7.5 mm and the eye is pigmented (16 days),
8. Hatching, when the larva is about 14–14.5 mm (34 days)
9. Larva is about 18 mm and the initiative of adipose fin appears (42 days),
10. 2/3 of the yolk sack has already been consumed and larvae gulp air (52 days),
11. Length of the larvae is about 21 mm, when the edge of pectoral, pelvic and caudal fins become serrated (59 days),
2. Yolk-sack is almost entirely pigmented (70 days),
13. Yolk-sack is fully consumed (85 days).

3.4.4 ◗ Rearing of fry

After the swimming-up of fry their feeding should start with properly balanced dry feeds which contain the needed quality and quantity of proteins (50–60 percent), vitamins and minerals. Size of the applied feed is also very important. It should only be as large as the developing fry can easily grab and swallow. During the rearing of fry or as often called advanced fry, intervals between feedings should be about ½ hours. A sign of insufficient feeding is the increasing difference between individual sizes of developing fry. This may result in serious cannibalism.

Tanks of different shapes are used for rearing fry. Few meters long troughs, rectangular or round tanks are the most widely applied rearing devices. At the beginning depth of water may vary between 0.1 and 0.2 meters which should gradually be increased to 0.5–0.8 meters.

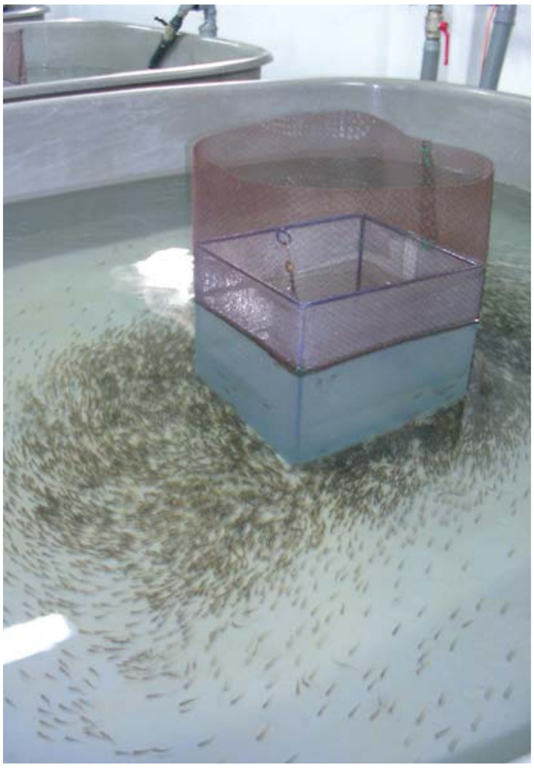

Figure 10:
Typical fry rearing fibreglass tank

Figure 11:
Typical fry rearing concrete tanks

Density of fry in rearing tanks may vary from 2 000 to 5 000 fry/m^2 and water supply should be about 0.5–1 l/sec (Hoitsy, 2002).

It is recommended to keep fry rearing devices in a closed place where air temperature can be controlled. This will ensure a continuous (uninterrupted) growth of fry. Otherwise daily and occasional sharp changes in temperature may also influence water temperature of the densely stocked rearing tanks.

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GLOSSARY

Anadromous fishes
Those fish species which migrate from marine to freshwater waters for spawning, like trout or salmon are anadromous fishes.

Animal pole
It is the more active, multiplying pole of an egg cell.

Artificial propagation or reproduction of fish
It is a collective term and comprises a wide range of different techniques and technologies which aim to produce young fish under controlled farm conditions.

Elements of artificial reproduction of fish may be:

- Stripping of wild males and females during spawning season.
- Rearing of broodfish (females and males).
- Synchronized, stimulated or induced spawning of females and males.
- Striping and fertilization of eggs through stimulated or induced ovulation.
- Incubation of embryo and non-feeding larvae (yolk-sack fry) under controlled fish hatchery conditions.

Depending on the produced fish species practiced techniques may include only some or all of the listed elements.

BW
It is the abbreviation of body weight.

Catadromous fishes
Those fish species which migrate for spawning from freshwater to marine water, like eel are catadromous fishes.

Changing the colour of fertilised eggs
On fish farms where removing of unfertile eggs is done with photo sensor based machines, carotenoids are added to the feed of females. Carotenoids change the colour of fertile eggs to dark orange. The machine can more easily distinguish this colour from the white unfertile eggs. Therefore, the efficiency of sorting machines will increase.

Chromosome
It is a thread-like structure found in all living cells that carry genetic information in the form of genes.

Daydegree (D°)
It is used to express the length of incubation or developmental period of non-feeding larvae. It is the summed up value of daily average temperature of water. If the actual water temperature is lower more, if it is higher less days are needed for the development of embryo or non-feeding larvae.

Diadromous fishes
Those fish species which migrate for spawning from marine to freshwater waters or vice versa are diadromous species. Fish like eel which migrate from freshwater to sea water are catadromous, while species like trout or salmon which migrate from the sea to the freshwater to spawn are anadromous fishes.

Diapause
Suspended development of an organism. It may occur if environmental conditions change to unfavourable.

DO
It is the abbreviation of **dissolved oxygen** in the water which ensures respiration of fish. The actual oxygen content of water depends on some important physical and chemical characteristics of water. Water can dissolve only a certain quantity of oxygen at a certain temperature. The possible maximum dissolved oxygen content of water (100 percent saturation) depends on the actual water temperature and the partial pressure of oxygen in the atmosphere. Dissolved oxygen content changes slightly with the quality and quantity of other dissolved materials. Altitude also modifies the oxygen content of water.

Dormant (resting) stage
At this development stage eggs are ready for final maturation and ovulation. During this stage females are waiting for favourable environmental conditions of spawning. When suitable conditions arrive this stage ends and final maturation and ovulation of eggs occur.

°C	DO (mg/l)	°C	DO (mg/l)	°C	DO (mg/l)	°C	DO (mg/l)	°C	DO (mg/l)
1	13.92	6	12.21	11	10.83	16	9.75	21	8.82
2	13.52	7	11.91	12	10.61	17	9.55	22	8.67
3	13.20	8	11.62	13	10.38	18	9.35	23	8.41
4	12.88	9	11.33	14	10.15	19	9.16	24	8.36
5	12.52	10	10.10	15	9.96	20	9.00	25	8.22

Table 5: Dissolved oxygen content of fully saturated water at different temperatures

Endocrine
It is the collective name of glands which produce hormones or *hormone** like products and secrete them directly into the blood system.

Fecundity
Egg production capacity of a female fish.

Fertilization
It is the act when the male reproductive product, in case of fish sperm enters into an egg and initiates the division of the cell.

Follicle
It is a sheath, protective covering and connective tissue around egg cells. Its function is to protect and nurture developing eggs, eventually become a double layer of eggs (Woynarovich and Horvath, 1980).

Gametes
They are mature *haploid** female (ovulated egg) or male (sperm) germ cells. They are able to unite into *zygote** with another reproductive cell of the opposite sex.

Genotype
It is the genetic constitution of living organisms.

Germinal disk
It is the location on the top of the yolk where an egg cell is fertilized and cell-division starts.

Globulins
These are simple proteins soluble in salt solutions.

GnRH/A
Gonadotropin releasing hormones are secreted by the hypothalamus when fish receive a favourable environmental stimuli for spawning through their receptors and brain. These hormones, as their name indicates stimulate the release of gonadotropin hormones in the pituitary. GnRH/A is the abbreviation of gonadotropin releasing hormone analogues which are artificially produced in order to induce ovulation and spermation in a female or male fish.

Gonads
In bony fishes gonads, ovary and testis are paired and in most forms there is no connection between the reproductive and urinary systems (Bond, 1996). In trout the two systems connect into one joint urogenital papilla where they empty.

The gonads develop from the peritoneum in which primitive egg cells can be found.

Habitat
It is the environment of an organism or community, characterized by its physical or biotic properties (Allaby, 1994).

Haploid
A haploid cell is a germ cell which has an unpaired, single set of *chromosomes**.

Hormone
It is produced by specialised glands in order to regulate action of organs such as gonads.

Hypophysis or pituitary
It is a very important endocrine gland under the brain. It produces different hormones, among others gonadotropins which stimulate and control the activity of gonads. This gland of sexually matured male and female preserved with alcohol or acetone are used for inducing ovulation or spermation in female and male fish.

Hypothalamus
It is a region of the brain under the thalamus through which fish receive and pass sensory information.

Lacustrine
A fish species is lacustrine if it is adapted to lake *habitat**.

Meiosis
It is a type of cell division that results in daughter cells with half of the number of chromosomes as the parent cell.

Mitosis
It is a type of cell division that results in daughter cells which have the same number and kind of chromosomes as the parent cell.

Neuro-endocrine process
It is the response of sexually matured fish to favourable environmental stimuli carried by the receptors to the brain. As a result the brain together with the endocrine system executes the development and release of eggs and sperms. The schematic sequence of information is: 1) Favourable environmental conditions → 2) Receptors → 3) Brain → 4) Hypothalamus that acts through gonadotropin releasing hormones (GnRH) → 5) Pituitary that acts through gonadotropin hormones (GtH) → 6) Gonads → 7) Secondary and accessory sex characteristics; reproductive behaviour and activity (Woynarovich and Horvath, 1980; Bond, 1996).

Ovary
It is the female reproductive organ which produces eggs.

Ovulation
It is the release of matured eggs from the ovary.

Parental care
Parental care of egg laying bony fishes may be indirect, direct and the combination of these two. At indirect parental care parents select the type of spawning ground where released and fertilized eggs and hatched larvae can safely develop and feeding larvae and developing fry can find both food and shelter. A characteristic of these fish species is that their fecundity is very high. Their females produce several hundred thousands of eggs. This ensures the survival of sufficient offsprings required for the maintenance of the species. Substrate spawning common carp, breams, pike and river spawning Chinese and Indian major carp and South American Carassius species are typical members of this group of cultured freshwater fishes. There are freshwater fish species which perform active parental care during incubation of eggs but then they abandon the hatched larvae. They wash out or build nests where developing eggs are ventilated and guarded. European catfish and pikeperch belong to this group of fish. Fecundity of these fish is also high. Fish species of active parental care produce less eggs. As a compensation they take intensive care of the developing eggs, hatched larvae and even the fry. The most well known representatives of substrate spawning and mouth breeder groups of freshwater fishes are tilapias.

Peristaltic
It is constrictions and relaxations of muscles of the intestine or other canal-type organs. This is an unintentional waive-like movement which pushes the content of the canal forwards.

Phenotype
It is the observable characteristics of interactions between genotype and environment.

Pituitary
See at hypophysis.

Primary oocytes
They are egg cells which develop from primitive egg cells with normal *mitosis**.

Riverine
A fish species is riverine if it is adapted to a river *habitat**.

Saprolegnia
The fungi responsible for saprolegniasis are secondary pathogens which appear and develop after handling or any traumatic damage caused to the skin of a fish.
Symptoms caused by Saprolegnia spp. are greyish-white patches on the skin which have a cotton wool-like appearance under the water.

Stripping
It is the act of removing ovulated eggs and milt from female and male fish.

Testis
It is the organ which produces sperm.

Vegetal pole
Also called vegetative pole. It is the less active yolky pole of an egg cell.

Vitellogenesis
It is a longer process before each reproductive season when the yolk builds into the eggs.

Zygote
It is the name of fertilized eggs

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