



CHAPTER 4

TECHNOLOGY OF BROWN TROUT CULTURE IN CONTROL CONDITIONS FOR RUNNING WATERS STOCKS PRODUCTION

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TECHNOLOGY OF BROWN TROUT CULTURE IN CONTROL CONDITIONS FOR RUNNING WATERS STOCKS PRODUCTION

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1. AIM OF THE NEW TECHNOLOGY

The purpose of the described technique is to provide fisheries management with information about the method of breeding and management of local brown trout (*Salmo trutta m. fario* L.) populations in controlled conditions. Implementation of this technique should increase and stabilize the production of high quality indigenous brown trout stock genetic lines for the stocking of natural waters, and also result in the subsequent reduction in transport of stock between different regions and countries. Furthermore, stocking of indigenous genetic lines will help to support original wild trout populations, preserve the intraspecies genetic variability and will prevent genetic contamination by allochthonous material. The proposed technique must comply with the standards of sustainable production of stock with preserved wild population characteristics.

2. TECHNOLOGY DESCRIPTION

INTRODUCTION

In the Czech Republic, brown trout (*Salmo trutta m. fario* L.) is one of the most commercially important fish species in running waters. However, in recent years there has been a significant decline in population numbers due to many factors. The reasons and possible solutions of trout population decline have been discussed in both scientific and non-professional communities. The Czech Fishing Union, an organisation responsible for the management of open waters, is often criticised for insufficient stocking of rivers. However, the situation is more complicated, and satisfactory solutions can be achieved only by adopting complex measures. Such measures have to be based on a thorough analysis of the situation in every particular river. It is important to know that successful natural reproduction is crucial for population development, and under optimal conditions, it ensures the preservation of genetic diversity and purity and thus ensures the stability of the population. Successful natural reproduction requires sufficient amount of brood stock. Numbers of parent fish (and fish in general) are affected mainly by the river morphology (Harsányi a Aschenbrenner, 2002; Turek et al., 2009), hydrological conditions (Rogers et al., 2005), intensity of the predation pressure of fish-eating predators (Mareš and Habán, 2003; Spurný, 2000, 2003a, b), water contamination (Kolářová et al., 2005), fishery management, and finally by fishing pressure on the river (Lusk et al., 2003).

In order to prevent further weakening of trout populations, it is necessary to implement enhanced protection measures, such as the prevention of unreasonable amendments of river beds, improvement of river morphology, reduction of predatory activity, and change in angling rules and fishery management of the river etc.

Brown trout stocking is a relatively easy and practicable way of supporting wild populations. There has been an increased tendency among fisheries to intensify the support of native populations by stocking. However, numbers of stocked fish is limited by the numbers of stripped brood fish. Brood stock is usually caught in rivers during the pre-spawn period, but the numbers of available brood fish are declining. In order to obtain sufficient numbers of brood stock, it is necessary to fish in longer and more valuable stretches of Czech rivers. In some parts of the country the numbers of fry for stocking are still insufficient and stock is imported from other regions or even from abroad. Stocking of different genetic breed lines coming from an intensive breeding process has become common practice (e.g. Ital, Kolowrat).



Photo 1. *Brown trout.*

Intensive farming practice is increasingly used for the production of stock due to the need for higher production efficiency. Stocking of fish from hatcheries is often criticized because of its negative effect on wild fish populations (L'Abée-Lund, 1991; Einum and Fleming, 2001). Stocked fish can affect local wild populations by higher aggressiveness, predation, food competition and the spreading of disease etc. (Hedenskog et al., 2002; Petersson and Järvi, 2003; Huntingford, 2004). There is also a potential risk of genetic contamination in cases where stocked fish crossbreed with the native population (Weber and Fausch, 2003). On the other hand, there is reduced adaptability in stocked fish resulting in higher mortality rates after stocking. Farmed fish often become preferred prey for piscivorous predators after stocking due to the lack of experience, and reduced anti-predatory behaviour (White et al., 1995; Weiss and Schmutz, 1999). There are also difficulties with the adaptation to the natural food source. Searching for food and defending already used food territories poses high energetic expenditure for fish which have been fed in hatcheries. This often leads to exhaustion and in combination with other factors (such as predators, diseases, fishing) it can lead to eradication of stocked fish in the river (Ersbak and Haase, 1983; Johnson, 1983; Bachman, 1984; Nicholls, 1985; Lachance and Magnan, 1990).

Uncontrolled stocking of fish of a different origin can have a negative effect on genetic characteristics of local wild populations and can lead to a significant decline or even extinction of genes and genotypes which are best adapted to the local conditions. Uncontrolled stocking can thus weaken the local fish populations, and in such cases restoration is very difficult.

Stocking still remains an effective way of supporting wild fish populations, especially in the localities where the natural reproduction is limited. However, genetic characteristics of stocked fish must be similar to those of wild fish populations living in stocked water and stocked fish must have high adaptability to the local conditions. In order to produce such stock, it is necessary to have high quality brood fish with the required genetic characteristics.

Successful technology of brood fish rearing in controlled conditions can be a suitable solution for stabilizing, or even increasing brown trout stock production. This can be done without the elimination of natural reproduction as there is less need for catching parent fish for stripping. A breeding technique must involve methods that do not significantly affect genetic characteristics of fish even from the long term perspective. For the preservation of the original gene pool of local fish populations and their intra species diversity it is therefore necessary to work with these native fish populations from the particular region.

This technology describes veterinary aspects, principles and methods of brown trout brood stock production in controlled conditions and rearing of the particular age classes (fry, fingerling, juveniles, brood fish).

METHODS OF BROOD STOCK ESTABLISHMENT AND REARING OF DIFFERENT BROWN TROUT AGE CLASSES

Fisheries designed for rearing brown trout should have a strong all year round supply of high quality water, the temperature of which does not exceed 18 °C, even on hot summer days. The rearing system should be designed as a flow-through system.

BROOD STOCK ESTABLISHMENT

Rearing of brood fish in controlled conditions should be carried out from the fry stage. A wild trout population from the local area is the best source of parent fish to use for brood stock establishment. Ideally it is best to obtain fish egg of brood fish from as many local rivers in the same catchment area as possible. Such rivers should have limited influence of fishery management (mainly brown trout stocking) and fish should naturally spawn (e.g. fish protected areas or parts of rivers with no fishery management).

Stripping must ensure maximum genetic variability of fry. Common methods where large amounts of eggs (from tens of females) are fertilised by the sperm of many males (polysperm fertilization) has proved to be inadequate for genetic variability preservation. Several studies have shown that polysperm fertilization causes a competition in spermatozoa, with most eggs being fertilised by only a small number of males originally used for stripping. Different methods are thus recommended for preservation of genetic variability. (Kašpar et al., 2008). The following recommended technology can be used in conditions under which most of the hatcheries in the Czech Republic operate.

The most suitable method is dry fertilization, where eggs are stripped straight into the dry bowl together with ovarian fluid. Eggs can also be stripped to the sieve where the ovarian fluid is left to drip off and eggs are then carefully moved into a dry plastic bowl.

Each female should be stripped separately, i.e. eggs from each female are stripped into one dry plastic container or sieve. Quality of eggs should then be visually checked for the presence of blood, clumps of eggs, white eggs or evidently damaged or low quality eggs. A defined volume of eggs (approximately an average number of eggs from one female) is then transferred by a gauge (ladle) into a bigger plastic container (container A). The aim of this procedure is to put similar numbers of eggs from each female into the container. The rest of eggs (if there is any) are put into another container (container B). If the quality of eggs is evidently poor, it is better to put these eggs straight into container B, or discard immediately.

The eggs are thus collected into containers A and B during the entire stripping process. It is necessary to cover both containers with a damp cloth. The eggs should never be exposed to sun radiation during the entire process, as the temperature can rise well above the incubation temperature. It is also important to prevent eggs from getting into contact with water, as the presence of water in eggs before fertilization considerably decreases their fertilization success rate.

When all the females are stripped, eggs from container A are carefully mixed and then separated (if possible) into as many smaller parts as possible. Each part is then fertilized by milt from different males. The number of males used for fertilization of one partial part of eggs should be equal to the proportion of available males and to the proportion of containers with eggs. It means that every male is used for the fertilization of 1 partial group of eggs only. The optimum number of males used for fertilization of one group of eggs should be 1–6. The fewer males are used for the fertilization of one partial group of eggs, the higher are the chances of achieving maximum genetic variability. But it is recommended to use at least two males for every part of eggs as it eliminates the loss of eggs if the sperm of one male is not fertile. Milt is added straight onto the eggs, which should be protected from any contact with water by drying off the urogenital area of stripped fish, including the anal fin, with a damp cloth. The egg is then mixed and water is added to activate gametes and initiate the process of fertilization. The eggs are then carefully mixed again using a suitable tool, e.g. clean spatula or trowel (plastic, rubber or wooden). The source of water must be the same throughout the whole process. To prevent excessive diluting of milt which could result in lower fertility, the water level in the bowl should not be higher than 1–2 cm above the eggs. Containers are left to stand for about 2–3 minutes for the process of fertilization to finish. The eggs are then carefully rinsed with water, and all groups of eggs which were originally in container A are poured together again into one container and mixed with caution. This step of mixing all the eggs together is important for homogenisation of eggs originating from many different parent fish before placing them into individual incubation apparatus. When the rinsing with clear water is finished, as much water as possible is added to the container(s) and eggs are left to rest for about 2–3 hours. During this stage the eggs swell and are very sensitive to shaking. Once the eggs have swollen, it is time to place them into the incubation apparatus.

The same procedure is repeated with the eggs in container B. The same males used for the container A are used for insemination. It is necessary to mark individual incubation apparatus to distinguish eggs from containers A and B. If the stripping takes several days, groups of eggs (A and B) are produced by the method described above for every stripping day separately. Approximately the same volume of eggs from each female will be transferred into container A throughout the whole stripping season (i.e. the same gauge will be used throughout the season). If the stripping in one day takes more than several hours and the number of stripped fish is high, the procedure of stripping and fertilization is repeated several times during the day.

Brood fish which have been stripped are put into a potash bath for a short period of time (Kolářová and Svobodová, 2009) before being released back into the river as soon as possible.

Brood stock should always be preferably established from offspring originating from container A. It is necessary to ensure maximum genetic variability of the future brood stock. If stripping was carried out over different time periods, then the same amount of fry has to be taken from every stripping process (i.e. from each container A in a given season). Fish from the container B are used only when there is not enough fish produced from the container(s) A.

In order to produce one brood female about 100 fertilized eggs are needed. The optimum number of brood stock is about 100–200 females. Thus for brood stock of 100 females some 10,000 fertilized eggs are needed. These eggs should come from as many parents as possible (minimum of 20–30 pairs).

To prevent major phenotype and genetic changes of reared brood stock and subsequently of their offspring (Fleming and Einum, 1997; Einum and Fleming, 2001; Verspoor, 1988; Hanák, 2008), it is always necessary to raise brood stock from offspring of wild fish acquired by the method described above.

Using only the first generations of artificially raised fish ensures maximum preservation of the original characteristics of parent populations. For the same reason it is advisable to keep only females (or only a small number of males as their presence may positively affect maturation of females) and their eggs fertilized by the milt of wild males taken from a suitable river (for example a fish protected area).



Photo 2. *Catching of brown trout brood stock in natural habitat.*

Optimally, every brood stock rearing fish farm should have its own fish protected area as a source of native brood fish and wild males for the fertilization of eggs from reared females. Any Fish Protected Area (FPA) must comply with the following requirements:

- *The character of the river section chosen as a fish protected area should be as close to natural conditions as possible and unpolluted by industry and communal waters.*
- *The whole section of FPA should allow migration for fish and migration barriers should not be in the connecting parts of the river.*
- *The size of the area should allow the occurrence of several hundreds of brood fish.*
- *No selection measures are recommended within FPA and its tributaries. No stocking should take place in such areas (especially reared stock from hatcheries) and restocking of fish should only be based on natural spawning.*
- *To ensure successful natural reproduction, it is necessary to leave part of the brood stock (c. 1/3) in the river when catching brood fish. It is therefore advised to put young fish at their first spawning season back to the river.*

INCUBATION OF EGGS

The most suitable incubation device is the classic Rückel-Vacek apparatus. Its construction enables the setting of circular (sideways), or bottom water flow by the easy shifting of the inside plate with eggs. The apparatus is usually set to the circular flow during the incubation (but bottom flow is also possible) and to the bottom flow after hatching to prevent damage to the yolk sac. The eggs in the apparatus should be placed in one row only. The capacity of one incubation apparatus is thus around 8–10,000 eggs.

The incubation apparatus used for hatching has to be clearly marked. After the fertilization and placement of eggs into the apparatus, it is necessary to carefully remove dead (white) eggs. Incubating eggs are very sensitive to shaking, manipulation and light until they reach the stage of eyed eggs (220–230 days degree /D/ from fertilization). The length of incubation period depends on the water temperature, usually between 350–500°D.



Photo 3. *Incubation of brown trout eggs in Rückel-Vacek apparatuses.*

Preventive disinfection bath treatment of the eggs during incubation is possible (Kolářová and Svobodová, 2009). The fertilization success rate usually exceeds 95% and the mortality during incubation is up to 10% provided that good quality water is available and rules of hygiene and careful manipulation are kept. High losses can be caused especially by improper manipulation with the eggs before the stage of eyed eggs. Hatching takes place in the apparatus. It is necessary to remove egg shells and set the apparatus to the bottom flow during the hatching because of higher oxygen demand of fry.

REARING OF FRY AND YEARLING

After hatching, yolk sac fry stay on the bottom of the apparatus absorbing nutrients from its yolk sac (so called quiescent phase). This stage ends when the fry has absorbed about 1/2–2/3 of its yolk sac and starts to swim-up. This stage usually lasts for about 150–200 °D (c. 3 weeks). At the end of this stage it is possible to start with initial feeding directly in the apparatus.

At the beginning of the next (so called active) phase of rearing, when the fry is remarkably active and changes its diet from endogenic to exogenic, the fry must be transferred into shallow troughs (usual size 4 x 0,4 x 0,2 m) where it is necessary to start early feeding. The fry from one apparatus should be placed into one trough. Rearing troughs must be shaded and protected from direct sunlight. It is recommended to feed fry only with compound feed. The feed for rainbow trout with lower fat content from reputable producers have proved to be suitable for trout fry. The size of granules used at the early feeding should be about 0.5–0.6 mm and should not be floating on the surface but slowly sinking. Ideally, the trout should be fed small portions at a higher frequency (6–10 times per day) by hand over the whole area of the trough. Later, when fish readily take the food, it is possible to use automatic feeders (for example operated by a clock), preferably two feeders per trough. The feed rations should be at the bottom bounds of the rations recommended for the rainbow trout by the manufacturers of the compound feeds. The size of granules should be adequate to the size of raised fish according to the manufacturer catalogue. At the beginning the water level in the troughs should be kept low (c. 10 cm). Feeding of plankton increases the risk of contamination and slows the process of becoming accustomed to compound feed. Plankton can be used only temporarily if there is a problem with compound feed intake at the stage of early feeding. In such cases it is advisable to feed both plankton and compound feed together. The mortality at the first stages of rearing is usually up to 10%.

After 4–6 weeks the fry are transported to larger tanks, usually rectangular troughs or circular basins, where rearing generally continues to the stage of yearling. The stocking rate depends on the size of tank and the water oxygen content, the usual stock rate is between 1,000–2,000 fry per 1 m³ of water. The oxygen saturation should not fall below 60% in the tank outlet. It is possible to use oxygenators or oxygen apparatus, which enable an adequate increase in the stocking rate. During the rearing the fish are separated into more tanks according to the growth rate. The stocking rate at the end of yearling rearing is usually between 300–600 individuals per m³. The mortality during the rearing of yearling is usually about 20%.



Photo 4. Trough room for rearing brown trout fry.

It is important to carry out regular preventive inspections of the fry for the presence of parasitic infections (in minimum intervals of two weeks, in case of higher mortality immediately) and the rearing facility exceptionally tidy (i.e. remove excessive food remains, excrement and dead individuals). When there is a high risk of parasitic infections (summer months) it is advisable to use preventative baths (Kolářová and Svobodová, 2009).

The habituation of brown trout fry to granulated feeds at an early stage gives the opportunity for continuation of rearing in controlled conditions and to successfully raise required brood stock. This technique also solves the problem of obtaining plankton for feeding, allows regular food supply and eliminates the risk of parasitic infections.

REARING OF JUVENILES AND BROOD FISH

The technology of rearing brood fish juveniles (1–3 years old) has to be adapted to the quality of water supply. Ground ponds are not recommended if the water is supplied from a river as there is a potential risk of pathogen transmission. Flow-through tanks with a high flow and a hard bottom (concrete store-ponds, trench ponds, channels, flumes etc.) have been proved suitable in such cases. If there is a source of good quality water with no fish stock available, it is possible to use ground ponds. It is optimal to keep particular age categories (1–2 year, 2–3year, parent fish) in the separated tanks. Three year old fish can be placed together with the brood stock. The oxygen saturation should not fall below 60% in the outlets.

Juveniles of brood fish prosper well in flow-through tanks with the water volume up to 10 m³. The stocking rate of yearling trout is about 100–300 fish per m³. After one year (two year old fish) it is appropriate to reduce the stocking rate to 30–50 individuals per m³. At the same time the fish should be re-sorted and smaller individuals placed among younger fish categories. It is not recommended to do any selection, except for removing ill or deformed individuals. Fish which appear to be “outsiders” in fish farm conditions can bear important genetic characteristic for survival in natural conditions. The mortality during this stage of rearing is usually about 10%.



Photo 5. Example of suitable rearing tank for juveniles and brood trout.

The brood stock can be reared in the flow-through tanks holding several tens of cubic metres of water. The stocking rate is about 10 fish per m^3 . The mortality during the brood stock rearing is between 10–30% per year, the highest being in the post spawning period.

If a source of brood male fish from the wild is available (e.g. from FPA) for fertilizing stripped eggs, it is advisable to remove most of the males before placing juveniles to the brood stock. This selection should be done in the autumn, when males are easily distinguishable (tab. 1). Only a few (usually 10–20) males are left in each tank with the brood stock. The presence of males in the tank probably improves maturation of females. In the spawning period, males tend to fight each other if there are more of them in the tank. Injured fish are a source of bacterial and fungal infections which can spread to other fish weakened by the stripping.



Photo 6. Trout brood stock raised in controlled conditions.

If there is no source of wild males available it is necessary to rear them in controlled conditions in sufficient numbers. In this instance the selection is avoided and males are kept in the tanks together with females. The old (big) males can become very aggressive so they should be removed from the rear. It is also necessary to regularly check the fish health condition (especially in the post spawning period), remove all individuals with high levels of fungal infection and treat them accordingly in a bath or with antibiotics. It is not recommended to keep males separately from females as they tend to fight more and injuries can cause high and very often total losses of the stock.

Table 1. Overview of external morphologic characteristics with significant sexual dimorphism, i.e. typical for particular gender of mature fish.

| Characteristic | Male | Female |
|-----------------------------------|------------------------------|-----------------------------|
| Enlarged abdomen | indistinctive | distinct |
| Stimulation of abdomen releases | sperm (milt) of white colour | Eggs – just before spawning |
| Colour of abdomen | dark | pale |
| Urogenital orifice | slit shaped | oval, swollen |
| Maxilla extends | beyond eye | up to the eye |
| Lower jaw | hooked (older males) | straight |
| Front part of upper jaw (rostrum) | straight (sharp) | rounded |
| Body colour (spawning) | distinct | less distinct |

It is recommended to use only high quality rainbow trout compound feed. Younger categories are fed with less intensive feed (less fat content) and mature fish with specially designed feed for this category. The daily ration should be at the bottom bound of the ration recommended for the rainbow trout by the manufacturer of the feed. The size of granules should be appropriate to the size of fish. The granules should not be floating on the surface but should be slowly sinking. It is possible to use either hand feeding or the automatic feeding apparatus.



Photo 7. Sexual dimorphism of reared brown trout brood (top – male, down – female).

RESTOCKING OF BROOD STOCK

The spawning season does not significantly differ from wild populations, which in the territory of the Czech Republic usually begins in September and lasts until mid October. The manipulation of fish from wild and reared populations must be done with caution to prevent their shuffling. Shortly (c. 1 week) before stripping, the brood fish are transported from the rearing tanks to smaller manipulation tanks with males and females separated. If there is a river with a fish protected area or other suitable source of wild brood stock, the required numbers of males are caught and placed into a separate manipulation tank. Before spawning, females are sorted and only mature fish (abdomen cranially from urogenital orifice soft, urogenital orifice swollen, pressure to abdomen releases eggs) are used for stripping. Immature fish are put back to the manipulation tank and are checked at weekly intervals.

The hormonal stimulation is not necessary for the stripping. It is suitable to use anaesthetics to prevent damage of bigger brood fish during manipulation (Kolářová et al., 2007). Immediately after stripping it is advisable to put fish into a potash bath (Kolářová and Svobodová, 2009) and release them back to the rearing tanks or the river, depending on where the fish came from. The eggs from artificially reared fish should be inseminated by the sperm of wild males from the ancestral population. For the maximum preservation of genetic variability, the process of stripping for stocking of natural waters must be done by the same way as described in previous part. If however, all offspring are going to be sold to other fisheries, it is not necessary to separate fertilized eggs to parts A and B. If females from open waters are also stripped, the eggs of these fish have to be incubated separately from the eggs of reared females and the incubation apparatus should be carefully marked. The parameters for estimating future egg production from reared brood stock are as follows:

- *absolute fertility (number of stripped eggs per female) about 1,000 eggs;*
- *relative fertility (number of eggs per kg of female weight) 1,500–2,000 eggs;*
- *40–60% of females mature in their third year, the rest of fish mature later. Males usually mature one year earlier than females, so they can be taken out from the rear in their 2nd or 3rd year.*

Farmed fish will usually live longer (about 5–8 years) than fish in the wild. Reared fish thus grow bigger and have more eggs than in natural conditions. Weight of females used for stripping is in most cases between 300–1,000 g. Fish usually undertake 3–5 strippings during its life. The post-spawning mortality of the young brood fish (1st and 2nd stripping) is minimal. The fish reared for longer periods can develop morphological (reduced fins) or behavioural (loss of shyness, reactions to feeding) changes as an adaptation of the organism to the new environment and impact of unnatural conditions. However, if the above described method is used, these changes are not likely to be transferred to offspring as they are not usually genetically fixed.

One very useful tool for fish identification can be tagging. This can be done either for the group of fish (e.g. VIE system based on implantation of coloured visible implant elastomers under the upper layer of skin) or individuals (microchips, VIA systems based on implantation of visible coloured alphanumeric plates under the skin). Tagging can be done at any time provided that the size of fish matches the requirements of the method used (more information on tagging can be found at www.nmt.us). It is recommended to use anaesthetics during tagging (Kolářová et al., 2007). It is not advisable to tag fish at the juvenile stage as tagging of small sized fish can result in high losses of applied tags. Chip systems ensure clear individual identification and minimum losses.

Offspring of females artificially reared by the above described method have a potential for successful adaptability in natural conditions. The best method is to plant yolk sac fry or fed fry (4–6 weeks) into the brood streams or straight into the salmonid fishing grounds. In order to support native populations it is not recommended to plant older stock (more than 1 year old).

HEALTH ASPECTS OF REARING

A good health condition is essential for successful rearing of the brown trout brood stock. The health condition depends on many factors, such as the observance of hygienic rules, good quality of food and effective prevention against importing infection into the hatchery. Nevertheless caution must be taken and the fish farmer must be ready to tackle an unexpected infection outbreak. The following overview describes some of the most significant diseases of the brown trout. Detailed description, treatment and prevention of individual diseases have been published in Kolářová and Svobodová (2009) and Kouřil et al. (2008).

VIROSES

There are basically 4 viroses which can occur in salmonids and which are listed as Notifiable (serious) diseases in the Czech Republic and the EU: **Viral Hemorrhagic Septicaemia (VHS)**, **Infectious Pancreatic Necrosis (IPN)**, **Infectious Haematopoietic Necrosis (IHN)** **Infectious Salmon Anaemia (ISA)**. ISA has not been recorded so far in brown trout populations either in the salmonid fisheries in the Czech Republic or world-wide. IPN affects young fish (up to 5 cm) and it has been diagnosed in brown trout, as well as VHS. Brown trout is more immune to IHN than rainbow trout, but can act as a disease vector.

The symptoms of all described diseases are very similar in the first stage of illness – darkening of the body, exophthalmus (bulging eyes), torpidity, swimming malfunction, loss of reflexes. Veterinary care should therefore be sought everytime suspicion arises. Exact diagnosis of salmonid viruses is only feasible by laboratory tests which are carried out by the National Reference Laboratories of The State Veterinary Administration (SVS ČR) for fish viruses: The Veterinary Research Institute (VÚVeL Brno) for Moravia and Silesia area and The Reference Laboratory of the SVS ČR for fish diseases, The State Veterinary Institution (SVÚ) České Budějovice for the Czech area. About 10–15 live fish with symptoms should be sent to the laboratory for testing. The water temperature should not exceed 10 °C during transportation.

Treatment of viral diseases is not carried out.

The liquidation of serious disease is carried out according to the public notice No. 299/2003 Coll. The breeder whose animals have symptoms of dangerous disease is obliged to ensure that suspicious animals do not leave until the arrival of an official veterinary surgeon.

Prevention. Veterinary Act No. 286/2003 Coll. imposes obligations for breeders who supply salmonids to the market to examine salmonid fish for dangerous diseases IHN, IPN, VHS and ISA. The legislation classifies brown trout as receptive species to all the above named diseases. Fisheries with production of stock for open waters provide ovarial liquid taken from individual species of stripped fish for virological examination. During the sampling it is advisable to cooperate with the responsible veterinary surgeons.

BACTERIAL DISEASES

All three undermentioned bacterioses are listed as fish diseases on list III. of the European legislative (public notice No. 299/2003 – list III. and n. 381/2003 Coll. in the Czech Republic). If such infection occurs, an epicentre of infection and protection zone is declared by the veterinary authority. If after disinfection and re-stocking of the fishery with the sensitive species and age category the disease or suspicion does not re-occur during the observation time determined by the veterinary authority, the disease is considered to be defeated.

Bacterial diseases are treated with antibiotics (Kolářová and Svobodová, 2009). Suitable antibiotics are chosen according to the results of the bacteriological test and the test of sensitivity of the causative agent. Fish are tested at the State Veterinary Institutions and The University of Veterinary and Pharmaceutical Sciences Brno. The fish must be alive when sent for examination. The most frequent bacterial diseases are considered to be Furunculosis, Enteric Redmouth Disease and Bacterial Kidney Disease.

Furunculosis

Furunculosis is a globally widespread disease including the Czech Republic. All salmonids are sensitive to this disease, with brown trout being more sensitive than rainbow trout. The causative agent is the bacterium *Aeromonas salmonicida* subsp. *salmonicida*. The disease develops mainly during higher water temperature (15–21 °C). In temperatures below 7 °C the disease develops in a latent form. The disease can also develop in peracute form without the specific symptoms (it causes rapid mortality). An acute form is characterised by neurotic symptoms and anal prolapse. Typical changes of skin (abscesses – furuncles) are developed only in a subacute and chronic form.

Enteric Redmouth Disease (ERM)

A causative agent is the bacterium *Yersinia ruckeri*. Brown trout are among the most sensitive species to this infection. The disease develops in fish in their first year when water temperature is between 13–15 °C. Infected fish are dark, lethargic, and blood stains develop at the base of fins, jaws, upper roof of the mouth and operculum.

Bacterial Kidney Disease (BKD)

A causative agent of the disease is the bacterium *Renibacterium salmoninarum*. Brown trout are more sensitive than rainbow trout. The disease usually develops at spring when the water temperature rises to 13–18 °C. Fish generally get infected in their second year of life, soft and alkaline water worsens the disease progress. Infected fish are dark and have neurotic symptoms.

PARASITICAL DISEASES

Parasitoses are treated with an antiparasitic bath and antiparasitic medications added to the food (Kolářová and Svobodová, 2009). Parasitological diseases can be endoparasitic or ectoparasitic.

Ectoparasitic diseases

A causative agent of parasitosis is located on the skin and gills and causes necrosis of the affected area. Parasitoses develop especially during high concentrations of fish and a lack of light. Mature trout are most commonly infected by the protozoan *Chilodonella piscicola*. It survives without the host fish for several days at water temperatures between 3–5 °C, but dies without a host after 1 hour in 20 °C temperature. Sunlight and light negatively affects the reproduction of the parasite. Another common parasitic protozoan is *Ichthyophthirius multifiliis* (white spot disease). The developmental cycle of this protozoan is 35 days in water temperatures off 10 °C and 3–4 days in 21–24 °C. The developmental stage released from the cyst (theront) dies within 2–4 hours without a host. Theronts die in light about 2–3 times faster than in the dark and do not survive in pH 5.5. Contamination is caused by infected fish or by the water containing theronts. Skin and gills are also often invaded by flukes *Monogenea*, family *Gyrodactylus* and *Dactylogyrus*.

Endoparasitic diseases

Salmonids are threatened by the parasite *Hexamita salmonis*, especially in their first year of life. This protozoan does not attach itself to the organs surface, but floats in liquids and affects the caudal part of the intestine, cholecyst and bile duct. It causes digestion disorders followed by a loss of weight and death. Intestines of salmonids are also attacked by flukes (class *Trematoda*), thread worms (*Nematoda*) and tapeworms (*Cestoda*), such as *Proteocephalus neglectus* reaching lengths of 100–150 mm. Intermediate hosts are copepods. Adult tapeworms attach themselves to the mucous membrane of pyloric appendixes with only the body protruding to the intestine. The mucous membrane is mechanically damaged and this causes an inflammation and necrosis along with malfunction of physiologic function of the intestines. Affected fish lose weight and can die, with massive invasions causing intestinal obstruction.

3. COMPARISON OF „TECHNOLOGY NOVELTY“

Traditional and widely used methods of obtaining brown trout stock for open waters consist of fishing for the brood fish from native rivers, their stripping and rearing of fry. Rearing can be done in extensive conditions (brooks, ponds) or in controlled conditions to the stage of 1 or 2 year old fish which are then stocked into the fishing grounds. However, numbers of brood fish in open waters are declining for a number of reasons. To obtain sufficient numbers of brood stock, it is necessary to fish in longer and most valuable stretches of rivers. This of course has a negative effect on the natural reproduction which is the basis for the survival of the species and conservation of its genetic variability. There is a lack of brood stock in some parts of the country, and this situation is often dealt with by importing and stocking of fish originating from different regions or even abroad. A relatively common practice is stocking of intensively farmed fish and breed lines of a different genetic origin (e.g. Ital, Kolowrat). Such methods can threaten the stability of local wild populations. Fry production from traditional methods varies considerably from year to year because its dependence on seasonal conditions. Because of this instability, purchasers secure the stock with supplies from other sources as they cannot rely on one fishery production. Expenses related to fishing for brood fish, transport and rearing of fish often exceed the profit from sale of the produced stock. On many rivers the negative impact of brood stock fishing and their stripping on a native population is evident.

Implementation of new techniques will help to increase and stabilise production of brown trout stock on the level needed by organisations responsible for management of rivers in the Czech Republic. The creation of the system of regional fry production facilities working with local populations of trout will help to move back from stocking of allochthonous fish from other regions or abroad. Self-breeding of brood fish and abundance of the above mentioned method will fulfil the requirement of long term sustainability and stability of production of high quality stock with a characteristic maximally similar to wild populations. At the same time this technology will enable us to abandon massive fishing for brood fish from natural waters, which will support their natural reproduction.

4. APPLICATION OF THE TECHNOLOGY

This technology is designed especially for the brown trout stock production facilities, such as angling unions or small private hatcheries. The aim of the implementation of this technique should be to increase and stabilise the production of high quality brown trout stock of native genetic origin for natural waters, stocking and reduction of stock transport between different regions and from abroad. Stocking of indigenous genetic lines will increase the support of the wild population, preservation of the gene pool and intraspecies variability and will prevent genetic contamination from stocking of allochthonous stock.

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REFERENCES

- Bachman, R.A., 1984. Foraging behaviour of free-ranging wild and hatchery brown trout in a stream. *T. Am. Fish. Soc.* 113, 1–32.
- Einum, S., Fleming, I.A., 2001. Implications of stocking: Ecological interactions between wild and released salmonids. *Nord. J. Freshwat. Res.* 75, 56–70.
- Ersbak, K., Haase, B.L., 1983. Nutritional deprivation after stocking as a possible mechanism leading to mortality in stream-stocked brook trout. *N. Am. J. Fish. Manag.* 3, 142–151.
- Fleming, I.A., Einum, S., 1997. Experimental tests, of genetic divergence of farmed from wild Atlantic salmon due to domestication. *ICES J. Mar. Sci.* 54, 1051–1063.
- Harsányi, A., Aschenbrenner, P., 2002. Development of the stock and reproduction of grayling (*Thymallus thymallus*) in lower Bawaria (in Czech). *Bulletin RIFCH Vodňany* 3, 99–127.
- Hanák, R., 2008. Interaction between wild and stocked fish in open waters (in Czech). *Bulletin RIFCH Vodňany* 44 (1), 3–20.
- Hedenskog, M., Petersson, E., Järvi, T., 2002. Agonistic behavior and growth in newly emerged brown trout (*Salmo trutta* L.) of sea-ranched and wild origin. *Aggressive Behav.* 28, 145–153.
- Huntingford, F.A., 2004. Implications of domestication and rearing conditions for the behaviour of cultivated fishes. *J. Fish Biol.* 65, 122–142.

- Johnson, M., 1983. An evaluation of stream trout stocking in Langlade, Lincoln and Marathon counties. Wisconsin Department of Natural Resources, Fish Management Report 114, p. 7.
- Kašpar, V., Vandeputte, M., Kohlmann, K., Hulák, M., Rodina, M., Gela, D., Kocour, M., Linhart, O., 2008. A proposal and case study towards a conceptual approach of validating sperm competition in common carp (*Cyprinus carpio* L.), with practical implications for hatchery procedures. J. Appl. Icht. 24 (4), 406–409.
- Kolářová, J., Velíšek, J., Nepejchalová, L., Svobodová, Z., Kouřil, J., Hamáčková, J., Máchová, J., Piačková, V., Hajšlová, J., Holadová, K., Kocourek, V., Klimánková, E., Modrá, H., Dobšíková, R., Groch, L., Novotný, L., 2007. Anesthetics for fish. Methodology edition, RIFCH USB Vodňany, no. 77, 19 pp
- Kolářová, J., Svobodová, Z., 2009. Therapeutical and preventive procedures in fish rearing (in Czech). Methodology edition, RIFCH USB Vodňany, no. 88, 30 pp.
- Kolářová, J., Svobodová, Z., Žlábek, V., Randák, T., Hajšlová, J., Suchan, P., 2005. Organochlorine and PAHs in brown trout (*Salmo trutta fario*) population from Tichá Orlice River due to chemical plant with possible effects to vitellogenin expression. Fresen. Environ. Bull. 14/No.12a-2005, 1091–1096.
- Kouřil, J., Mareš, J., Pokorný, J., Adámek, Z., Randák, T., Kolářová, J., Palíková, M. 2008. Rearing of salmonids, grayling and coregonids (in Czech). RIFCH USB, 142 pp.
- L'Abée-Lund, J. H., 1991. Stocking of hatchery-reared fish an enhancement method? Fauna 44, 173–180.
- Lachance, S., Magnan, P., 1990. Performance of domestic, hybrid, and wild strains of brook trout, *Salvelinus fontinalis*, after stocking: the impact of intra- and interspecific competition. Can. J. Fish. Aquat. Sci. 47, 2278–2284.
- Lusk, S., Lusková, V., Halačka, K., Smutný, M., 2003. Anglers' catches as an indicator of fish population status. Ecohydrology and Hydrobiology 3 (1), 113–119.
- Mareš, J., Habán, V., 2003. Mareš, J., Habán, V., 2003. Impact of otter and cormorant oversized presence on development in fishing areas of Moravian Anglers Union (in Czech). In: Textbook of report of conference "Fisheries and predators". Czech Angler Union, Praha, 36–40.
- Nicholls, A.G., 1985. The population of a trout stream and the survival of released fish. Mar. Freshw. Res. 9, 319–350.
- Petersson, E., Järvi, T., 2003. Growth and social interactions of wild and sea-ranched brown trout and their hybrids. J. Fish Biol., 63, 673–686.
- Rogers, M.H., Allen, M.S., Jones, D., 2005. Relationship between river surface level and fish assemblage in the Ocklawaha River, Florida. River Res. Appl. 21, 501–511.
- Spurný, P., 2000. Predation pressure of cormorant on fish populations (in Czech). Rybářství 7, 304–305.
- Spurný, P., 2003a. Effect of predators on fish populations of salmonid waters (in Czech). In: Textbook of report of conference "Fisheries and predators". Czech Angler Union Praha, 41–47.
- Spurný, P., 2003b. Deterioration of the fish community of the salmonid Dyje River cause by overwintering cormorant (*Phalacrocorax carbo*). Acta Scientiarum Polonorum 2 (1), 247–254.
- Turek J., Randák T., Velíšek J., Hanák R., Sudová E., 2009. Comparison of fish abundance and biomass in part of small brook with different morphological and discharge condition (in Czech). Bulletin RIFCH Vodňany 45 (1), 18–25.

- Verspoor, E., 1988. Reduced genetic variability in 1st generation hatchery populations of atlantic salmon (*Salmo salar*). Can. J. Fish. Aquat. Sci. 45 (10), 1686–1690.
- Weber, E.D., Fausch, K.D., 2003. Interactions between hatchery and wild salmonids in stream: differences in biology and evidence for competition. Can. J. Fish. Aquat. Sci. 60, 1018–1036.
- Weiss, S., Schmutz, S., 1999. Performance of Hatchery-Reared Brown Trout and Their Effects on Wild Fish in Two Small Austrian Streams. Trans. Am. Fish. Soc. 128, 302–316.
- White, R.J., Karr, J.R., Nehlsen, W.N., 1995. Better roles for fish stocking in aquatic resource management. In: Sachrann, H.L., Piper, R.G. (Eds.), Uses and effects of cultured fishes in aquatic ecosystems. Am. Fish. Coc. Symp. 15, Bethesda, Md. 527–547.

LIST OF PUBLICATIONS PRECEDING THIS TECHNOLOGY

- Kašpar V., Vandeputte M., Kohlmann K., Hulák M., Rodina M., Gela D., Kocour M., Linhart O., 2008. A proposal and case study towards a conceptual approach of validating sperm competition in common carp (*Cyprinus carpio* L.), with practical implications for hatchery procedures. J. Appl. Icht. 24 (4), 406–409.
- Kouřil, J., Mareš, J., Pokorný, J., Adámek, Z., Randák, T., Kolářová, J., Palíková, M., 2008. Rearing of salmonids, grayling and coregonids (in Czech). RIFCH USB, 142 pp.
- Prokeš, M., Randák, T., Peňáz, M., Baruš, V., Žlábek, V., 2004. Post-hatching genesis of brown trout (*Salmo trutta* m. *fario* L.): Comparative analysis of effect of wild and hatchery reared mature fish (in Czech). Textbook of VII. Czech ichthyological conference, RIFCH USB, Vodňany, pp. 219–223.
- Randák, T., Kocour, M., Žlábek V., Policar, T., Jarkovský, J., 2006. Effect of culture conditions on reproductive traits of brown trout *Salmo trutta* L. Bull. Fr. Peche Piscic., 383, 1–12.
- Randák T., 2006. Possibility of stocks production increasing of brown trout (*Salmo trutta* m. *fario* L.) and European grayling (*Thymallus thymallus* L.) for free waters stocking (in Czech). Ph.D. thesis, University of South Bohemia, Faculty of Agriculture. 132 pp.
- Randák, T., 2002. Performance of artificially reared stocks of brown trout (*Salmo trutta* m. *fario*) and grayling (*Thymallus thymallus*) in free waters conditions (in Czech). Textbook of V. Czech ichthyological conference, Mendel university, Brno, pp. 139–145.
- Randák, T., 2002. Methods of initial fattening of brown trout (*Salmo trutta* m. *fario*). Textbook of conference "Production of fish and crayfish stock, RIFCH USB, Vodňany, pp. 34–39.
- Randák, T., Pokorný, J., 2001. Basic morphological and reproductive parameters of brown trout (*Salmo trutta* m. *fario*) in Blanice Vodňanská river (in Czech). Textbook of conference NP a CHKO Šumava, Vimperk, pp. 206–209.
- Randák, T., Žlábek, V., 2004. Comparison of reproductive parameters artificially reared and wild population of brown trout (*Salmo trutta* m. *fario* L.) (in Czech). Textbook of VII. Czech ichthyological conference, RIFCH USB, Vodňany, pp. 111–114.
- Randák, T., Žlábek, V., 2004. Possibility of stocks production increasing of brown trout (*Salmo trutta* m. *fario* L.) in Šumava area (in Czech). Textbook of conference CHKO Šumava, pp. 224–229.
- Turek J., Randák T., Velíšek J., Hanák R., Sudová E., 2009. Comparison of fish abundance and biomass in part of small brook with different morphological and discharge condition (in Czech). Bulletin RIFCH Vodňany 45 (1), 18–25.

