TRAINING MANUAL ON THE ARTIFICIAL PROPAGATION OF CARPS

A handout for on-farm training workshops on artificial propagation of common carp and Chinese major carps in Central and Eastern Europe, the Caucasus and Central Asia

Second revised edition
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László Horváth
FAO Consultant

Gizella Tamás
Biologist

André G. Coche
FAO Senior Fishery Resource Officer

Éva Kovács
Aquaculture Officer, FAO-REU

Thomas Moth-Poulsen
Fisheries Officer, FAO-REU

András Woynarovich
FAO Consultant

Food and Agriculture Organization of the United Nations
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FOREWORD

FAO has always played a leading role in the publication of practice-oriented technical papers and training materials on fisheries and fish culture. One of the objectives of producing this huge wealth of ready-to-use technical information is to support the sustainable development of fish culture all over the world.

Following these principles, two very successful, richly illustrated practical technical guides were produced in the mid-1980s on the mass production of eggs, early fry,¹ advanced fry and fingerlings of common carp. Owing to profound political, social and economic changes, the production of fish ponds and small water reservoirs in many countries of Central and Eastern Europe (CEE) and in the Caucasus and Central Asia (CCA) has declined considerably in recent decades. In order to restart and increase carp production in these regions, hands-on training courses supported with handouts has proved useful in the practical training and self-education of concerned and interested fish farmers.

The present handout builds on an earlier FAO publication.² The original watercolour illustrations used in this publication are the work of László Horváth, father of the senior author. Although the set of these pictures is practically complete, in order to prepare an even more understandable technical reference, additional black and white illustrations from other publications, as well as tables and text, have been inserted where sources were correctly indexed and listed under References.


¹ Today, they are called feeding larvae.
1. INTRODUCTION

It is widely admitted that common carp (*Cyprinus carpio*) has one of the most complex artificial propagation technologies among commercially valuable freshwater fish species. Consequently, a fish farmer skilled in the artificial propagation of common carp will have no problem in adopting this technology to the propagation of other valuable carps of the region such as Chinese major carps, e.g. grass carp (*Ctenopharyngodon idella*), silver carp (*Hypophthalmichthys molitrix*) and bighead carp (*Aristichthys nobilis*).

Following the above-mentioned concept, this present handout was prepared as a detailed guide to the artificial propagation of common carp with supplements for similar propagation of Chinese major carps. Accordingly, chapters detail key aspects, data and know-how about the artificial propagation of common carp, while the attached annex contains specific information and data needed for the artificial propagation of Chinese major carps as well.

In addition to the annex, two appendixes are also attached introducing the long-distance transport of feeding larvae of carps, and how to collect and preserve carp hypophysis.

This handout endeavours to use simple language and widely known technical words and expressions. However, in sections where lesser-known technical terms and expressions were unavoidable, they appear in italics and are marked with an asterisk (*) to indicate that they are explained in the Glossary.
2. BIOLOGY AND METHODS OF CARP PROPAGATION

This chapter provides on the biology and basic aspects of the propagation of carps in general and common carp in particular. This information is essential to the understanding and successful large-scale production of fish seed under controlled fish farm conditions.

2.1 Biology of reproduction

Fish culture practices are based on the biology of the species under cultivation. Reproduction control is particularly important. Here are some of the basic principles for common carp.

2.1.1 Life cycle of carp

The entire life cycle:
(1) “In adult fish reproduction cycle initiates in the gonads* with the development of sexual products: eggs and sperm (milt).” This ensures successful spawning,
(2) Fertilization of eggs.
(3) Eggs stick to the vegetation.
(4) Eggs develop and hatch into larvae, which first, in a non-feeding larval stage, also stick to the vegetation.
(5) Start of external feeding after larvae gulp air and start to swim horizontally.
(6) Development of advanced fry.
(7) Advanced fry stage.
(8) Development of fish (one-summer-old, two-summer-old, etc. fish).
(9) Adult age – sexually mature fish mates.

Development of a fertilized egg:
(1) Fertilization
(2) Swelling
(3) Start of cell division
(4) Two, four, eight, etc. cell stages
(5) Morula stage
(6) Blastula stage
(7) Gastrula stage
(8) Embryo
Development of larvae:
(1) Hatched larvae
(2) Just feeding larvae
(3) Early stage of advanced fry
(4) Advanced fry

2.1.2 Egg production in females

In the first few months, the fibre-like initials of the gonads are already being formed. At this stage, the ovary already contains the primitive early egg cells from which eggs will develop when females reach sexual maturation.

Evidence of sexual maturation is when the 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 a successful reproduction. This starts with the transformation of a batch of the primitive egg cells in the ovary into primary oocytes. → A follicle* forms around each primary oocyte. The process called oogenesis continues:

(1) – (5) Egg development goes through several stages during which single cells grow to 800–1 000 microns in size. At the end, yolk accumulates. This final process is called vitellogenesis.

(5) By the end of this stage, eggs are ready but remain dormant* in females until conditions for spawning become favourable.

(6) In cases of conditions favourable for spawning, eggs become fully ripened (maturation).

(7) – (8) Ovulation and fertilization of eggs are the last stages.

Tip: Vitellogenesis takes place after spawning/propagation when the new batch of eggs develops. Therefore, females should be fed properly during this period. See details in Section 3.5.

Ripe eggs ovulate as a result of a long neurohormonal process.

(A) In a temperate climate, carp spawn once a year.
(B) In a tropical climate, carp spawn several times

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3 Called oogonium, ovogonium or archovogonium.
2.1.3 Spawning of carp

Preconditions for further development of dormant eggs in the ovary are:
1. Suitable water temperature.
2. Enough dissolved oxygen in the water.
3. Light.
4. Enough natural food.

Favourable conditions for spawning are:
- A suitable ground covered with spawning substrate.
- High enough water temperature.
- Flooding of rivers.
- Presence of males. They produce *pheromones* that females are able to sense.

Spawning starts early in the morning because of the *diurnal rhythm*.

Carp spawn in groups on shallow grounds covered with vegetation. Released eggs stick to the vegetation regardless of their fertilization.

1. Male and female spawn side by side. Released eggs are fertilized by males.
2. Ripe eggs released into the water are surrounded by a cloud of moving sperm released by males.
3. Sperm enters in the micropyle, which is open for about 30–60 seconds after coming into contact with water.
4. Later, the micropyle closes regardless of whether a sperm has entered into the egg or not.
2.2 Propagation methods

All propagation methods are based on the reproductive biology of fish. These methods are:

- Imitation of favourable spawning conditions.
- Interference in the neurohormonal control of reproduction.

*Controlled spawning*, *induced spawning*, *induced ovulation* and their combinations can be distinguished accordingly.

Natural reproduction of carp depends on two groups of environmental factors. These are:

(A) Basic factors:
   1. Temperature: 18–24 °C.
   2. Dissolved oxygen: 5–0 mg/litre.
   3. Light.

(B) Stimulating factors:
   1. Favourably changing atmospheric pressure.
   2. Presence of males.
   3. Vegetation to spawn on.

Factors controlling the spawning of females with dormant eggs differ according to the propagation technique applied.

Semi-artificial reproduction (orange arrows) is controlled by the same basic (A) and stimulating (B) factors listed above, except that injection of gonadotropin hormone (4) becomes a major stimulating factor.

Artificial reproduction (red arrows) has only two basic factors that success. These are:

1. Water temperature.
2. Dissolved oxygen content.

2.2.1 Natural-like propagation

In natural-like propagation, only basic environmental conditions are ensured:

- Ponds with freshly flooded grassy areas can be used for natural-like propagation of carp.
- Mature broodfish are stocked. About 3–4 females per hectare, and 2–3 males per female.
2.2.2 Dubish method

This method simulates through ensuring essential environmental conditions:

1. Small floodplain-like ponds (100–1,000 m²).
2. (3) One or two sets of breeders (2 females and 3 males per set) are stocked. Imitation of flooding induces spawning.
3. (4) After spawning, breeders are removed.
4. (5) Developing eggs and larvae can easily be observed.

2.2.3 Mixed methods

Catching wild carp before spawning:

1. Mature, ready-to-spawn broodfish are captured.
2. Ovulated eggs are stripped.
3. (4) Eggs are fertilized and taken into a hatchery.

Incubation of eggs under sheltered conditions:

1. (2) Eggs are stripped and placed into baskets.
2. Eggs develop in the baskets.
3. Larvae swim out of the baskets.

2.2.4 Semi-artificial propagation

In semi-artificial propagation, one single hormone injection helps to induce spawning. The most frequently used techniques are the synchronization of spawning in grassy ponds, hapas or on kakabans.

Synchronized spawning in grassy ponds:

1. Broodfish are injected (3 mg of hypophysis per kilogram of BW*) when the water temperature is > 18 °C.
2. Broodfish are transported to spawning ponds.
3. Broodfish are stocked into a freshly inundated grassy pond.
4. It is to be expected that the broodfish will spawn on one of the following mornings.
Synchronized spawning in breeding hapas:
(1) – (2) Injected broodfish are placed into a breeding hapa (dimensions: $1 \times 1 \times 2$ m, mesh size: 1 mm), where they spawn.
(3) After spawning, broodfish are removed, while fertilized eggs remain in the hapa, which serves as a shelter during the incubation period.
(4) Larvae hatch in the hapa.

Synchronized spawning on kakabans:
(1) – (2) Injected broodfish are stocked into small, freshly flooded ponds where spawning substrates made out of plant fibres (kakabans) are placed.
(3) Eggs stick to the substrate.
(4) – (5) Spawning substrates with eggs are placed into a new pond or into wire-meshed boxes.
(6) Larvae are stocked from the old pond into a new one (where this was not done earlier).

2.2.5 Artificial propagation
Artificial propagation of fish in general and of common carp in particular is when propagation is fully programmed and each phase is completed under controlled hatchery conditions.

Key steps of artificial propagation are:
(1) Injection of suitable broodfish with gonadotropic hormones.
(2) Stripping of sexual products (eggs and sperm).
(3) Fertilized eggs treated against stickiness are incubated in hatchery jars.
(4) Hatched larvae are placed and reared in large jars.
(5) As soon as larvae start to feed, they are stocked into nursery ponds.
2.3 Comparison of natural reproduction and artificial propagation of carp

Advantages of artificial propagation of common carp compared with its natural reproduction are:

(A) Reproduction in nature:
   
   (1) Need for male spawners is increased.
   
   (2) – (4) During incubation, eggs are exposed to adverse environmental conditions.
   
   (5) – (7) Newly hatched larvae are not protected against their enemies; therefore, they will have less chance of survival.

(B) Artificial propagation:

   (1) Need for male spawners will be about 4–6 times less.
   
   (2) – (4) During incubation, it is possible to protect the eggs against parasites and water fungi as well as against bad weather conditions and predators.
   
   (5) – (6) Newly hatched larvae can also be better protected against their enemies; therefore, their survival is further enhanced through their controlled first feeding.
   
   (7) Stocking of feeding larvae in well-prepared ponds ensures better growth and survival.
3. BROODSTOCK MANAGEMENT OF CARP

This chapter deals with the management of broodstock, which covers three particular aspects of the rearing process:

- Selection of fish with desirable hereditary qualities typical of improved strains such as rapid growth potential, higher resistance to dissolved oxygen deficiency and adverse water quality, strong appetite or omnivorous feeding regime.
- Selection of fish with well-developed sexual organs.
- Rearing of selected fish to produce healthy potential spawners with well-developed dormant eggs in females.

3.1 Selection of future carp breeders

At the selection of future broodfish general body shape, scale distribution, state of health and development of sexual organs should be observed. In particular, selected fish:

(1) Should be in good health.
(2) – (3) Should have no wounds or parasites.
(4) Should have a scale distribution typical to strain.
(5) – (6) Should have no fin or body deformation.
(7) – (8) Should have a body with required shape and proportions, being neither too fat, nor too thin.

3.2 Differences between male and female carp

Females and males can be differentiated by shape: In females (♀), the body is plump and the genital opening is situated above the genital papilla. In males (♂), the body is slender and the genital opening is found behind the genital papilla.

**Tip:** External signs of maturity of females and males are distinct, so it is easy to check whether they have reached maturity (presence of dormant eggs or sperm) and should be selected for artificial propagation. For this reason, the belly and genital papilla should be examined carefully:

- A mature female (♀) has a well-rounded and soft or semi-soft belly; its genital papilla is erect and reddish; its anal opening is enlarged and protruding.
- A mature male (♂) will release milt under a slight abdominal pressure; its belly is not blown up but rather slim; sometimes it has callosities on the head.
3.3 Criteria of a good broodstock pond

Broodstock ponds are 0.5–1 ha large and about 1–2 m deep. Dykes should be protected by vegetation. Control of water should be easy. Access by road and protection against poaching are also important. Stocking rate varies from 100 to 300 breeders/ha.

In the broodstock ponds, it is also advisable to stock about 200–400 smaller (100–200 g/fish) carnivorous fish per hectare. This is sufficient to control unwanted wild fish or fry with wild spawning origin.

3.4 Annual cycle of broodstock management

Broodstock management is a year-round procedure:

(1) In temperate zones, late spring spawners used in the hatchery are stocked into ponds.

(2) They should be well fed during summer and autumn.

(3) Overwintering takes place in the same pond. The following spring, breeders are captured and selected by sex for later propagation.

(4) Broodfish selected for propagation should be kept separately by sex in small storage ponds.

(5) During the propagation season, breeders are taken from storage ponds to the hatchery.

3.5 Feeding of broodstock

Broodstock feed varies according to season:

(1) In spring, after dormant eggs have formed and breeders are waiting for spawning, the feed of broodfish should have a high (30–40 percent) protein content to prevent accumulation of fat in their gonads.

(2) After stripping, breeders form new eggs, which develop up to dormant stage. Therefore, they should eat a mixture of 50 percent natural food rich in proteins and 50 percent supplementary feeds with high carbohydrate content (e.g. maize, wheat).

Tip: If it is not possible to purchase fish feed with a high protein content, feeding of germinated wheat can be very useful because of its high vitamin E content. However, do not feed dry wheat, because fat will deposit in the liver and ovary, and, as a result, females will not be able to propagate.
3.6 Selection and transport of broodfish for propagation

Selection of breeders for artificial propagation should be done about 24–30 hours before planned stripping.

**Tip:** During the process of fishing, besides paying attention to fish and gentle handling of fish, also try to avoid oxygen shortage in nets where fish are crowded and in containers while they are transferred to the hatchery.

(1) Selected breeders are taken into the hatchery.
(2) Fish not selected are returned to storage pond.
(3) Those specimens that are definitely unsuitable for propagation should be sold as food fish.

**Tip:** Desired soft, deep belly of breeders should be checked by gentle touching before taking them out of water. Appropriate fish should be lifted out of water and their genital papilla examined more closely.

One of the most widely used means of transferring broodfish between ponds and the hatchery is a double (two compartments) or single-space hammock made of an iron or wooden frame and a waterproof canvas or similar materials.

Broodstock transport may also be mechanized by using a fibreglass container and oxygen. In a fibreglass tank with oxygen diffusion, about 20–30 breeders/m³ of water can be transported safely for several hours.

**Tip:** Avoid using woven polypropylene grain bags or similar containers to carry broodfish, as this material will wound the skin and/or scales of fish. If a hand container is used for carrying broodfish individually, always do it in water as illustrated (Woynarovich and Woynarovich, 1998).

When handling carp breeders, the best option is to use a scoop net with a strong but soft knotless mesh with an open end, which makes it easy to release the fish.
4. HATCHERY OPERATIONS OF CARP PROPAGATION

4.1 Tools and equipment in the hatchery

In the hatchery where stripping and artificial fertilization of eggs take place, various items of equipment should be available to ensure efficient working conditions.

(1) Large storage tanks with a good water supply are used for hormone treatment of breeders.
(2) Fish are easily transported in a fibreglass tank on wheels.
(3) Many plastic containers of various sizes are necessary to handle and treat stripped and fertilized eggs:
   - 3–4-litre bowls;
   - 15-litre bowls (for swelling eggs);
   - 10-litre buckets and 25–30-litre bins (for fertilization of eggs).
(4) In a smaller room, equipment such as clean towels, measuring jugs/cylinders, plastic tubing, needles and thread, syringes, etc. are stored together with chemicals and a stock of dried hypophyses. See more details in Annex 1.

4.2 Weighing, tagging and anaesthetizing broodfish

4.2.1 Weighing and tagging

Females should be weighted and tagged in a scoop net with both ends open when they are taken into the hatchery (Woynarovich et al., 2011; Woynarovich and Woynarovich, 1998). This is because the weight of females defines the required dose of hormone. With enough practice, the weight of males may be estimated.

**Tip:** After weighing females, tag them according to their weight with yarns of different colours. For example, red, blue, white, yellow, etc. will mark females of 3, 4, 5, 6, etc. kilograms. The easiest is if a piece of yarn about 5–10 cm of the selected colour is fixed by the help of a needle into the dorsal fin around the first backward serrated hard ray. Do not forget to remove the tag before the females are returned to the fish pond.
4.2.2 Anaesthetizing

(1) – (2) For smooth handling during injection or suturing, broodfish should be anaesthetized with authorized products.

(3) – (4) Anaesthetized breeders should be watched to see whether their opercula keep moving. If they do not, the fish should quickly be removed and placed under a current of fresh water.

Of authorized fish anaesthetizers, clove oil is one of the best (1 ml/10 litres of water for common carp). It is both cheap and very reliable.

Tip: If many broodfish are narcotized in one batch, oxygen diffusion, or at least compressed air, should be ensured.

4.3 Hormone treatment

Injections of gonadotropic hormone found in the dry pituitary gland induce final maturation and ovulation of dormant eggs. These injections replace stimuli otherwise needed from the environment. As a result, only two basic environmental factors have to be at optimum level:

- Water temperature.
- Dissolved oxygen content of water.

Table 1 shows the dosing of both hypophysis and physiological solution, which is a 0.65 percent salt solution.

Steps in preparing a hypophysis suspension*:

(1) The weight of hypophyses is calculated on the basis of the weight of breeders taken into the hatchery.

(2) Dried glands are ground into a fine powder in a mortar.

(3) This fine powder is then carefully mixed with the calculated volume of physiological solution.

(4) The hormone suspension is ready to be injected into the fish. As this suspension settles rapidly, if a long time passes between filling the syringe and administration, the syringe should be flicked to stir it.

Tip: At Step 3 of the preparation, first, only a few drops of physiological solution should be added to the powder. As a result, a thick paste appears, which should be rubbed with the stick of the mortar. This will ensure the best possible extraction of the hormone content. After this, the total calculated quantity of physiological solution should be added gradually under continuous stirring. When the suspension is ready, it can further be homogenized by putting it in and then pressing it out from the syringe. For this process, use the same needle and syringe with which you will inject the fish.
### Table 1: Dosing hypophysis and physiological solution for common carp brooders

<table>
<thead>
<tr>
<th>Sex</th>
<th>First dose per kg BW</th>
<th>Decisive dose per kg BW</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Hypophysis</td>
<td>Physiological solution</td>
</tr>
<tr>
<td>Females (♀)</td>
<td>0.3–0.35 mg</td>
<td>0.25–0.5 ml</td>
</tr>
<tr>
<td>Males (♂)</td>
<td>–</td>
<td>–</td>
</tr>
</tbody>
</table>

**Injections of females:**

- First (preparatory) injection initiates the final maturation of eggs*
- Second (decisive or provoking) injection results in the final maturation and ovulation* of eggs.

Interval between the first and second doses is 12–24 hours.

Injection can be administered in two different ways, of which the second one (B) is safer than the first one:

(A) Intramuscular injection. After the injection, the muscle should not be rubbed because this could press the hormone out of the muscle.

(B) Intraperitoneal injection, which is actually the only way injecting hormone into scaly carps.

**Injection of males:**

- Before the second dose for females.

At, but still before the second dose, the genital opening of females should be closed in order to prevent any loss of eggs. A needle and a strong but soft cotton thread are used for this purpose, as shown in steps (1)–(4).

**Tip:** For the suture, use a curved surgical needle and needle pincers.

After injection, the females are placed into a large tank, where well-aerated, continuously changing water should be provided.

The optimal water temperature ranges between 22 and 24 °C although higher (24–26 °C) water temperature is also acceptable. A lower (20 °C) temperature can result in partial ovulation (fewer eggs).

**Attention:** Concentration of injected females under the inflow of fresh water indicates oxygen shortage. More precisely, there is not enough dissolved oxygen in the water of the tank to support oxygen-demanding processes in the ovary. This may cause failure in ovulation. In this case, follicles do not split off but the hydration (growth in size) of eggs continues. The belly grows and becomes swollen but eggs cannot be stripped. These females will die within days owing to sepsis caused by a massive spoiling. Diffusion of pure oxygen into the tank of females after the second injection can effectively prevent oxygen shortage, hence such a failure of stripping and loss of female fish.
Injected females are put back into the tank.

(1) As the ovulation period approaches, females with fully ripe eggs start to look for a place to spawn, moving along the sides of the tank.

(2) If a smaller “indicator” male is added, the rippest females will start to simulate spawning with it. About 15–20 minutes after the first spawning splashing, ovulation of eggs will be completed, hence picking and stripping of ripe females can start.

The length of time between the second dose and complete ovulation is closely related to water temperature. Therefore, it should be measured hourly and noted after the second dose. When the sum of hourly measured water temperatures reaches 240–260 °H (hour degree*), females are ready to be stripped.

If the water temperature remains nearly constant, a graphic method may help calculations. In the graph (1) the water temperature (°C) is indicated that correlates to the number of hours necessary for ovulation, displayed by (2).

At 24 °C, ovulation takes place between 10 and 11 hours after the second dose, but at 20 °C this time will be 12–14 hours or longer.

*Tip: To note key data of hormone treatment, incubation of eggs and larval rearing, use the register shown in Annex 1.*

4.4 Stripping sexual products (eggs and milt)

(1) When females are ready for stripping, they should be concentrated into one end of the tank with a sliding net frame. From here, fish can easily be transferred into a smaller container where they are anaesthetized.

(2) After stripping, females are gently returned into the larger section of the tank.

Females anaesthetized and ready to be stripped should:

(1) Be gently dried.

(2) Have their sutures removed.

(3) Have the free genital opening closed with a finger.

Then, stripping should be done gently.
Smaller females are usually stripped while being held in the hands while larger ones are often stripped on a stripping table.

*Tip:* Weight and note the amount of stripped eggs in order to be able to calculate the number of them!

Taking milt (sperm) of males can be done parallel of stripping females or it can be stripped directly onto the dry eggs. Use milt of two males for each female (a total of 10 ml for 1 kg of egg).

*Attention:* Both stripped eggs and sperm should remain dry until they are used at fertilization!

### 4.5 Fertilization and treatment of eggs

Artificial fertilization should be carried out under dry conditions because:

**Females (♀):** as soon as eggs come into contact with water, they start to swell and their micropyle closes within a minute.

**Males (♂):** as soon as spermatozoids come into contact with water they become highly motile but only for about 30–60 seconds.
With common carp, fertilizing solutions should be used to prevent eggs sticking together.

First fertilizing solution: 40 g common salt (NaCl) and 30 g urea (carbamide) (CO(NH₂)₂) dissolved in 10 litres of well-aerated hatchery water.

If only the first solution is used, the eggs should be stirred continuously until they are fully swollen; otherwise, they stick together.

To save continuous stirring of eggs, a second solution should be used: 40 g common salt and 160 g urea dissolved in 10 litres of water (Woynarovich and Woynarovich, 1980).

Dry eggs and milt should be gently but well mixed for a few seconds. Then, the first fertilizing solution should be added while still mixing it gently with a plastic spoon or strong feather.

As dry eggs come into contact with the first fertilizing solution, they begin to swell.

**Tip:** Because of certain advantages, skilled specialists may also use clean, well-aerated water instead of the first fertilizing solution for the actual fertilization of eggs.

The egg-swelling process lasts 1–1.5 hours. Within this period, 1 litre of dry eggs (A) will become 6–9 litres of swollen eggs (B).

The fertilizing solution should be added during swelling, but not much more than the amount just covering the eggs. If the dilution is too rapid, it will cause a strong stickiness of the eggs.

If only the first fertilizing solution is used, it should be changed three to four times during the entire swelling process.

Where the second fertilizing solution is also used, stirring of eggs with the first solution should continue only for about 10–15 minutes, and then it should gradually be replaced by the second solution. As soon as the first solution is fully replaced, eggs should only be stirred from time to time.

To entirely remove stickiness, 5 g/10 litres tannin solution should be used when eggs are fully swollen, which takes about 1–1.5 hours after fertilization.

About 1 litre of tannin solution is used to treat 4–5 litres of swollen eggs. As soon as the solution is added, eggs should be gently but quickly stirred and then diluted with 5–10 litres of clean, well-aerated hatchery water. After eggs have settled, the diluted solution should be poured off. The entire procedure should be repeated 2–3 times.

After these procedures, the eggs should be placed into incubation jars.

**Tip:** Do not use old, deep dark tannin as this solution may enter into the egg and damage the zygote*. Therefore, use only light yellow tannin dust purchased from pharmacies or specialized chemical shops.
4.6 Incubation of eggs

Traditionally, incubation of fertilized eggs of common carp is done in large zoug jars (7–8 litres) made of thick glass.

In fish farms where such devices are not available, larger incubation jars used for Chinese major carps can also serve well (see Annex 1).

In any case, a continuous flow from the bottom of the jar is needed in order to roll the eggs.

Incubation of eggs in 7-litre zoug jars starts with half filling them with water.

1. About 1.5–2 litres of swollen carp eggs, equivalent to 250–300 g of dry eggs should be put in each jar.

2. Eggs settle on the bottom of the jars.

3. Water flow should be opened (about 0.6–0.8 litres/min.).

4. Eggs are gently moved around in the funnel-shaped part of the jars.

During incubation, the water flow should be adjusted to the actual development stage of the eggs.

1. For the first 10 hours, about 0.6–0.8 litres/min. of water should pass through each jar.

2. When the blastula stage starts, the water flow should be increased to 1–1.2 litres/min.

3. When the tail, the eyes and the pigmentation of the embryos become visible, the water flow should be increased to 1.5–2 litres/min.

Unfertilized eggs become white and easily come into contact with water fungi called *Saprolegnia*. As these eggs, which accumulate gradually above fertilized ones, endanger fertilized eggs, they should be removed through the following steps:

1. Stop water flow.

2–3. Good eggs settle at the bottom while the white ones remain at the top.

4. White eggs should be carefully siphoned out of the jars. Then, reopen the water flow.

**Tip:** The above procedure should be followed at about the eyed-embryo stage, and be repeated if necessary.

To prevent fungal infections of common carp eggs (and bacterial ones in the case of Chinese major carps), a light solution of formalin (1 ml of 36 percent formalin dissolved in 10 litres of hatchery water) can be used daily.
Attention: Previously, earlier malachite green was used against fungal infections of incubated carp eggs. Now, the use of malachite green is prohibited in the European Union (Member Organization), and fish and fish products with its residues are refused entry to its markets.

The duration of the incubation period is closely related to water temperature.
On the basis of the daily average of water temperatures, a day-degree* (°D) should be calculated. For common carp, it is 60–70 °D.
It can be estimated from a simple graph showing days (2) of the incubation period and average water temperatures (°C) (1).
The best water temperature for the incubation of common carp eggs is 22–24 °C.
4.7 Hatching and rearing of larvae

When hatching starts, first some, but soon a good deal, about 2–3 percent of free-swimming larvae appear in the jars. At this time, the water flow should be reduced to a minimum and the eggs siphoned gently.

1 – 2) At siphoning, adjust the water pressure by lowering the bowl as eggs are siphoned.

3 – 4) Conduct siphoned water along the inner side of the bowl to avoid hard dropping and direct collision.

1) Bowls containing the eggs are left standing for a maximum of 10 minutes.

2) Larvae are then climbing along the wall of the bowl.

3) Care should be taken to monitor this process very closely because it is the reduction of the amount of dissolved oxygen in the water that stimulates embryos to swim free from the eggs.

(A) Newly hatched larvae are put in large rearing jars with an average density of about 2 500 larvae per litre of water.

It is important to equilibrate even small (0.5–1 °C) differences in the temperature of water in the hatching bowls and rearing jars.

(B) Cleaning of the sieve should only be done from outside. Never do it from inside.

**Tip:** Backwashing the sieve from outside through a ¼-1/2 inch pipe is very effective.

Larval rearing lasts about 4 days (60–70 °D). There are three successive development stages:

1) For about 1.5 days, newly hatched larvae attach vertically on the wall.

2) During the next 1.5 days, free-swimming larvae first move vertically up and down, then gradually assume a more horizontal position and finally swim up to the water surface and gulp air to fill their air bladder.

3) Once the air bladder is full and functional, larvae already swim horizontally, have a developed digestive tract, and are able to eat exogenous food.

On reaching this last stage, larvae should be fed and transferred to well-prepared ponds as soon as possible.

**Tip:** To provide enough surface area for hanging larvae, place a loose, long piece of clean plastic raffia or narrow strips of grain bag woven from polypropylene fibre into the jars.
In order to prevent bacterial infections of developing larvae of common carp (and Chinese major carps) a light solution of formalin (1 ml of 36 percent formalin dissolved in 10 litres of hatchery water) should be used daily.

The first exogenous food for larvae in the hatchery should be hard-boiled eggs. There are two ways to prepare them, which are discussed as follows:

Hard-boiled eggs are finely ground and mixed with 0.5 litres of water. From this mixture, 5–6 tablespoons should be given in every 2–3 hours.

The yolk of a hard-boiled egg is pressed through a fine plankton sieve (150–200 μ). The yolk in the sieve is directly washed into the jars at 2–3 hour intervals.

Attention: Hard-boiled eggs are used for two purposes: to indicate that the feeding of larvae has already started, and to teach them how to hunt for food. As hard-boiled eggs are not a balanced diet and can cause unhealthy fat deposits in the liver, do not feed larvae with them for longer than a maximum of 1–2 days.

4.8 Removing feeding larvae from the incubation jars

The rearing of larvae ends with their removal from the hatching jars, when they are siphoned into a special container.

(1) A mobile fibreglass tank is the most convenient container if many millions of larvae are to be removed.

(2) – (3) A fine-mesh container is placed into the tank. This container consists of a light metal frame, inside of which a fine-mesh (0.2–0.3 mm) rectangular bag is hung.

(4) The water in the mobile tank is regulated to the desired level by inclining the mobile external outlet pipe.

(A) How to drain larval-rearing jars.
(B) How to remove feeding larvae from the fine-mesh container.

- Fish larvae are carefully concentrated into one corner of the fine-mesh container, which should constantly remain under water.
- During the concentration process, the fish larvae are washed with a bowl into the corner.
- The same bowl serves for scooping out larvae from corners of the container.

If only a few million larvae are produced in a production shift, larvae from the jars could be siphoned into large buckets equipped with fine-mesh sieves. Their preparation and use are demonstrated below.

Windows are cut in the wall of a simple 15–30-litre plastic bucket and covered with a fine (250–300 μm) sieve. Then the bucket is placed into a larger container, which ensures a constant water level in the bucket where larvae are concentrated.
GLOSSARY

BW – Abbreviation of body weight.

Controlled spawning is a fish propagation technique where the best possible environmental conditions are created or ensured in order to stimulate spawning of ripe females and males.

Day degree (°D) is the sum of the daily average water temperatures.

Diurnal rhythm is the daily periodical change of physiological and behavioural functions.

Dormant egg is the expression for a development stage of fish eggs that allows females to wait for favourable environmental conditions for spawning. The length of the stage when fish can successfully be propagated depends on the actual water temperature. It varies between 4 and 6 weeks.

Final maturation of egg – It starts when the nucleus of the egg migrates close to the micropyle and the first hydration takes place, leading to the ovulation of eggs.

Follicle is a pouch-like tissue where single eggs develop and remain until final maturation and ovulation.

Gonads are the ovaries (♀) and testes (♂) of different sexes of fish.

Hour degree (°H) is the sum of temperatures measured once in every hour.

Induced ovulation of carps is the result of a technique where hormone is administered into ripe fish of both sexes in order to induce the final maturation and ovulation of eggs in females and sperm release in males. For this purpose, dry carp pituitary (hypophysis) glands or different hormone products¹ are used.

Induced spawning is a fish propagation technique where the spawning of ripe males and females is synchronized with a hormone injection.

Ovulation is the process where follicles break up and release mature eggs.

Pheromones – Sexual pheromones are chemical compounds that are released into the environment by males in order to attract females.

Suspensions are those mixtures of a liquid and a solid material in which particles of the solid material are dispersed throughout the liquid. A suspension is different from a colloid or solution as particles are larger and visible under a microscope, some can even be seen with the naked eye. This is why particles in a suspension are likely to settle if left undisturbed.

Zygote is the scientific name for a fertilized egg.

¹ Producers have their own brands, which trade under different names.
LIST OF REFERENCES

During the preparation of this handout on artificial propagation of carps, an earlier FAO training material published in 1985 was updated and adopted to the needs and conditions of fish seed production of today. This publication is:


Other publications referred are those listed below:

1 Earlier versions of this handout had already been used and tested in Georgia (2011) and Albania (2012), where it was shown in practice that such training materials translated into national languages are very useful.
Annex 1

SUPPLEMENT TO THE BROODSTOCK KEEPING AND HATCHERY OPERATIONS FOR COMMON CARP AND CHINESE MAJOR CARPS

Temperate-zone carp hatcheries can be used for the propagation of many different fish species from early spring to early summer. The season starts with pike and continues with pike-perch, common carp, tench, then European catfish, and finishes with Chinese major carps. The propagation period occurs once a year, during spring and early summer and lasts about 16–18 weeks.

Artificial propagation of common carp is followed by the propagation of Chinese major carps, using practically the same devices and techniques. Tables A1-1, A1-2 and A1-3 summarize the similarities and differences between the propagation of common carp and Chinese major carps.

Table A1-1: Comparative list of similarities and differences about broodstock management of common carp and Chinese major carps

<table>
<thead>
<tr>
<th>Tasks</th>
<th>Common carp</th>
<th>Chinese major carps</th>
</tr>
</thead>
<tbody>
<tr>
<td>Transferring broodfish into small storage ponds:</td>
<td>Autumn or early spring.</td>
<td>Late spring.</td>
</tr>
<tr>
<td>Separation of different species:</td>
<td>Done when broodfish are transferred into small storage ponds.</td>
<td>Not necessary. Males and females can be kept together.</td>
</tr>
<tr>
<td>Separation of breeders by sex before the propagation season:</td>
<td>Done in early spring at the latest.</td>
<td>With grass carp, breeders should be fed daily with fresh terrestrial plants (grass and clover).</td>
</tr>
<tr>
<td>Feeding of broodfish in the storage ponds:</td>
<td>Protein-rich feeds are given.</td>
<td></td>
</tr>
<tr>
<td>Behaviour of broodfish when handled and treated:</td>
<td>Calm.</td>
<td>Nervous and jumping.</td>
</tr>
<tr>
<td>Keeping and feeding of broodfish after propagation:</td>
<td>It is done in polyculture,¹ where common carp should receive good-quality feed² while grass carp need terrestrial green plants. Feeding of silver and bighead should be ensured through manuring/fertilization of pond water.</td>
<td></td>
</tr>
</tbody>
</table>

¹ After propagation, females and males are kept together. The number of stocked brooders of common carp and Chinese major carps may vary between 300 and 500 fish/ha (1 200–2 000 kg/ha) where there is enough water to change in the pond. Otherwise, the number of stocked brooders should not exceed 200–250 fish/ha (800–1 000 kg/ha). Stocking 0.1–0.3 kg European catfish (200–300 fish/ha) will keep the pond clean from wild spawning of common carp as well as from low-value fish (Horváth, Tamás and Tölg, 1984).

² After breeding, energy and protein-rich (25–30 percent) feeds at a rate of 1–3 percent of BW/day are important for the newly developing eggs. Manuring and fertilization of pond water ensures the natural food (Horváth, Tamás and Tölg, 1984).

Table A1-2: Comparative list of similarities and differences in the hormone treatment of common carp and Chinese major carps

<table>
<thead>
<tr>
<th>Tasks</th>
<th>Common carp</th>
<th>Chinese major carps</th>
</tr>
</thead>
<tbody>
<tr>
<td>Optimal water temperature during hormone treatment:</td>
<td>22–24 °C</td>
<td>22–26 °C</td>
</tr>
<tr>
<td>Weighing fish before hormone treatment:</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>Use of clove oil:</td>
<td>Yes: at injection and stripping.</td>
<td>Yes: at tagging, injection and stripping.</td>
</tr>
<tr>
<td>Use and quantity of clove oil:</td>
<td>1 ml/10 litres</td>
<td>0.4–0.5 ml/10 litres</td>
</tr>
<tr>
<td>Types of hormone used:</td>
<td>Hypophysis of carps and different artificial hormone products.¹</td>
<td></td>
</tr>
<tr>
<td>Quantity of hypophysis and physiological solution at 1⁰ injection:</td>
<td>0.3–0.35 mg/kg BW 0.25–0.5 ml/kg BW</td>
<td>0.35–0.45 mg/kg BW 0.25–0.5 ml/kg BW</td>
</tr>
<tr>
<td>Quantity of hypophysis and physiological solution at 2⁰ injection:</td>
<td>3–3.5 mg/kg BW 0.25–0.5 ml/kg BW</td>
<td>3.5–4.5 mg/kg BW 0.25–0.5 ml/kg BW</td>
</tr>
</tbody>
</table>

¹ After breeding, energy and protein-rich (25–30 percent) feeds at a rate of 1–3 percent of BW/day are important for the newly developing eggs. Manuring and fertilization of pond water ensures the natural food (Horváth, Tamás and Tölg, 1984).
Table A1-3: Comparative list of similarities and differences of fertilization and incubation of eggs and hatching and rearing of larvae of common carp and Chinese major carps

<table>
<thead>
<tr>
<th>Tasks</th>
<th>Common carp</th>
<th>Chinese major carps</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fertilization of eggs:</td>
<td>1st salt-carbamide solution</td>
<td>Clean hatchery water.</td>
</tr>
<tr>
<td>Swelling of eggs:</td>
<td>Happens before placing eggs into hatchery jars.</td>
<td>Happens in the hatchery jar as within 5–10 minutes fertilized eggs are placed into the jars where they reach their final size.</td>
</tr>
<tr>
<td>Tannin treatment of eggs:</td>
<td>Yes.</td>
<td>No.</td>
</tr>
<tr>
<td>Best devices for egg incubation:</td>
<td>7–9 or 20 litre zoug jars.</td>
<td>50 or 200 litre jars.</td>
</tr>
<tr>
<td>Best devices for larval rearing:</td>
<td>50 or 200 litre jars.</td>
<td></td>
</tr>
</tbody>
</table>
| Phases of larval development in hatchery jars: | 1) Hanging  
2) Zigzag swimming  
3) Horizontal swimming after gulping air | 1) Vertical upward swimming then sinking  
2) Sinking to the bottom of the jar  
3) Horizontal swimming after gulping air |
| First food of feeding larvae:              | Hard-boiled eggs.                                                          |                                                                  |

Time of ovulation, different at each species, can be expected after the second injection as shown in the graphs below. They also show how time requirements change not only according to species but also on the basis of the water temperature at which females reach ovulation stage.

Heat sums necessary for ovulation of common carp and Chinese major carps

![Common carp graph](image)

![Silver carp graph](image)
Heat sums necessary for ovulation of common carp and Chinese major carps

![Heat sums for Bighead carp](image1)

![Heat sums for Grass carp](image2)

*Source: Horváth (1978).*

If new jars are needed, the shape and dimensions of the jars below can be copied. In such jars, 2 500 larvae/litre can be reared. Their capacities are about 50 000, 150 000 and 500 000 feeding larvae.

**Modern devices for egg incubation and larval rearing**

![Modern devices](image3)

Hatchery jars of 20, 60 and 200 litres. They have a body (1) and a large removable sieve (2). Water enters at the bottom through a banded pipe (3) which breaks the water current. (Dimensions are in millimetres.)
Sources: Woynarovich et al. (2011); Woynarovich and Woynarovich (1998).

Preparation of the necessary notes and calculations of hatchery operations both useful and an integral part of the results. Accordingly, size of broodfish and quantity of gained eggs should be measured while fertilization rates and proportion of hatched larvae should be estimated. The form below is an example of how it could be organized (Woynarovich et al., 2011; Woynarovich and Woynarovich, 1998).

Table A1-4: Key data of artificial propagation of Common carp and Chinese major carps

<table>
<thead>
<tr>
<th>Description</th>
<th>Common carp</th>
<th>Silver carp</th>
<th>Bighead carp</th>
<th>Grass carp</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sexual maturation of females (years)</td>
<td>4</td>
<td>5</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>Sexual maturation of males (years)</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>6</td>
</tr>
<tr>
<td>Size of mature females (cm)</td>
<td>30</td>
<td>40</td>
<td>40</td>
<td>60</td>
</tr>
<tr>
<td>Size of mature males (cm)</td>
<td>25</td>
<td>30</td>
<td>40</td>
<td>60</td>
</tr>
<tr>
<td>Size of mature broodfish (kg)</td>
<td>2.5</td>
<td>3</td>
<td>4</td>
<td>6</td>
</tr>
<tr>
<td>Water temperature at propagation (°C)</td>
<td>16</td>
<td>22</td>
<td>21</td>
<td>23</td>
</tr>
<tr>
<td>Sex ratio at propagation (♂:♀)</td>
<td>2:1</td>
<td>1:1</td>
<td>1:1</td>
<td>1:1</td>
</tr>
<tr>
<td>Percent of ovulation in females after hormone treatment (%)</td>
<td>60</td>
<td>90</td>
<td>60</td>
<td>80</td>
</tr>
<tr>
<td>Ovulation after the decisive (2nd) dose (H°)</td>
<td>230</td>
<td>260</td>
<td>210</td>
<td>220</td>
</tr>
<tr>
<td>Number of eggs per 1 kg of female BW</td>
<td>100 000</td>
<td>200 000</td>
<td>60 000</td>
<td>80 000</td>
</tr>
<tr>
<td>Diameter of dry eggs (mm)</td>
<td>1</td>
<td>1.5</td>
<td>0.7</td>
<td>1</td>
</tr>
<tr>
<td>Diameter of swollen eggs (mm)</td>
<td>1.5</td>
<td>2.5</td>
<td>3.7</td>
<td>5.3</td>
</tr>
<tr>
<td>Description</td>
<td>Common carp (from</td>
<td>to</td>
<td>Silver carp (from</td>
<td>to</td>
</tr>
<tr>
<td>-------------------------------------------------</td>
<td>------------------</td>
<td>----</td>
<td>------------------</td>
<td>----</td>
</tr>
<tr>
<td>Number of eggs/1 kg dry eggs</td>
<td>700 000</td>
<td>1 000 000</td>
<td>900 000</td>
<td>1 100 000</td>
</tr>
<tr>
<td>Number of eggs/1 litre swollen eggs</td>
<td>80 000</td>
<td>120 000</td>
<td>18 000</td>
<td>22 000</td>
</tr>
<tr>
<td>Rate of fertilization (%)</td>
<td>80</td>
<td>95</td>
<td>70</td>
<td>90</td>
</tr>
<tr>
<td>Hatching of fertilized eggs (%)</td>
<td>90</td>
<td>95</td>
<td>75</td>
<td>85</td>
</tr>
<tr>
<td>Survival of larvae up to taking air (%)</td>
<td>90</td>
<td>95</td>
<td>80</td>
<td>90</td>
</tr>
<tr>
<td>No. of feeding larvae from 1 kg of dry eggs</td>
<td>500 000</td>
<td>700 000</td>
<td>500 000</td>
<td>600 000</td>
</tr>
<tr>
<td>Length of egg incubation (°D)</td>
<td>60</td>
<td>70</td>
<td>24</td>
<td>30</td>
</tr>
<tr>
<td>Length of non-feeding larvae phase (°D)</td>
<td>60</td>
<td>70</td>
<td>60</td>
<td>70</td>
</tr>
<tr>
<td>Size of feeding larvae (mm)</td>
<td>6</td>
<td>7</td>
<td>6</td>
<td>6.5</td>
</tr>
<tr>
<td>Size at starting species specific feeding (mm)</td>
<td>25</td>
<td>30</td>
<td>30</td>
<td>35</td>
</tr>
<tr>
<td>Amount of eggs in a 7–9 litre zoug jar (g)</td>
<td>100</td>
<td>200</td>
<td>40</td>
<td>50</td>
</tr>
<tr>
<td>Amount of swollen eggs in a 7–9 litre zoug jar</td>
<td>1</td>
<td>2.5</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>Amount of swollen eggs in a 60 litre jar (g)</td>
<td>100</td>
<td>200</td>
<td>100</td>
<td>150</td>
</tr>
<tr>
<td>Amount of swollen eggs in a 60 litre zoug jar</td>
<td>1</td>
<td>2.5</td>
<td>5</td>
<td>10</td>
</tr>
<tr>
<td>Number of larvae in a 60 litre jar</td>
<td>80 000</td>
<td>120 000</td>
<td>80 000</td>
<td>120 000</td>
</tr>
<tr>
<td>Number of larvae in a 200 litre jar</td>
<td>250 000</td>
<td>400 000</td>
<td>250 000</td>
<td>400 000</td>
</tr>
</tbody>
</table>


Attention: It is not recommended to use 7-9 litre large hatchery jars for egg incubation of Chinese major carps.
LONG-DISTANCE TRANSPORT OF FEEDING LARVAE

For long-distance journeys, bags made of strong plastic (0.3–0.5 mm thick) are preferable.

(1) These bags (55–60 cm in diameter and 80–90 cm deep) should be filled with 20 litres of clean water from the hatchery.

(2) Put 100 000 feeding larvae into this water, and fill up the free space above the water surface with oxygen.

(3) The bag should then be tightly closed with a strong rubber band, and checked for possible leaks. For further safety, it is a common practice to use two plastic bags placed one inside the other.

For short distances to ponds close to the hatchery, a fibreglass water tank (200 × 100 × 80 cm) equipped with a supply of compressed oxygen may be used.

In such tanks, about 1 000 000 larvae/m³ can be safely transported to ponds. To avoid injuries, they should be either siphoned from the tank directly into pond water, using for example a rubber pipe with a diameter of 6–8 cm, or released through a 20 cm flexible pipe attached to the bottom of the tank.

Transport of larvae in containers and in plastic bags

<table>
<thead>
<tr>
<th>Species</th>
<th>Temperature of transporting water</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>10 °C</td>
</tr>
<tr>
<td>Common carp (No.)</td>
<td>–</td>
</tr>
<tr>
<td>Chinese major carps (No.)</td>
<td>–</td>
</tr>
</tbody>
</table>

In fish-transport containers under continuous oxygen diffusion (1 m³ water). Duration of transport: 2–6 hours.

<table>
<thead>
<tr>
<th>Species</th>
<th>Temperature of transporting water</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>20 000–400 000</td>
</tr>
<tr>
<td>Common carp (No.)</td>
<td>–</td>
</tr>
<tr>
<td>Chinese major carps (No.)</td>
<td>–</td>
</tr>
</tbody>
</table>

In plastic bags with pure oxygen (30 l water and 30 l oxygen). Duration of transport: 2–12 hours.

Source: Antalfi and Tölg (1971).
COLLECTION AND STORAGE OF CARP HYPOPHYSES

Ovulation of female carps can be induced with different natural or artificial hormones. Common carp hypophyses is one of the most frequently used natural hormones for the artificial propagation of carps, and it is also excellent for inducing ovulation in many other fish species all over the world. Therefore, it is important to know how to collect and preserve them.

Where possible, hypophyses (pituitary glands) should be collected from mature, freshly killed carp. To store them for a long period, they should be dehydrated. Dried hypophyses are used as hormonal extract to be injected into carp breeders to induce ovulation of eggs and release of sperms.

To remove hypophysis from freshly killed carp:

1. A special drill bit should be used in an electric drill.
   - The head of the fish should be firmly held vertically between two pieces of wood. Proceed as follows:
     - Draw an imaginary line perpendicular to the lateral side of the head from both eyes.
     - Place the centre of the drill at the meeting point of these lines.
     - Adjust the drilling angle so that it is 90°.
     - Drill through the top of the skull to the brain and the base of the skull, down to the mouth cavity.
     - Take out the drill bit together with the small cylinder made of bones and tissues.
2. With the help of a wooden finger fixed to the table, remove this cylinder from the drill bit.
3. Carefully cut it into two and lift the brain tissue (including the hypothalamus) lying at the base of the skull. Pick out the hypophysis with forceps. Place it into a bottle containing acetone together with other hypophyses collected at the same time.

Attention: It is important to pay special attention to protect the drill and its user from electric shock!

Tip: If selling fish on the market, the hole in the forehead may reduce the market value of fish. Leaving a hole in the forehead can be avoided by removing the hypophysis through the roof of the mouth.

(1) – (2) Before storage, hypophyses are dehydrated in acetone in three steps, using eight hours for each period. New acetone should be used at the beginning of each period.

(3) After this treatment, the hypophyses are hard and should be dried on blotting paper.

(4) Then, they should be placed into small glass containers and pressed down with a ball of dry fine cotton. Containers of conserved glands:
   - should be tightly sealed;
   - should be labelled with key information, 1) date and place, 2) number and total weight of glands;
   - sealed containers should be kept either in a plastic bag or in a desiccator in the presence of a water absorbent such as silica gel or calcium chloride.

If the preservation and storage of hypophyses are done properly, acetone-dried hypophyses can be stored for at least 5 years without refrigeration even in tropical regions.
Owing to profound political, social and economic changes, the production of fish ponds and small water reservoirs in many countries of Central and Eastern Europe (CEE) and in the Caucasus and Central Asia (CCA) has declined considerably in recent decades. In order to restart and increase carp production in these regions, hands-on training courses supported with handouts has proved useful in the practical training and self-education of concerned and interested fish farmers.

This manual is a basic guide to successfully practice seed production of different fish. Although the guide concentrates on the propagation of cyprinids it serves as a good basis for the reproduction of a number of other species as well. The aim of this training material is to guide the reader through the necessary technical information, related practical solutions and the steps of preparation for the propagation of cyprinid species, thus providing a basis also for seed production of other species.