

## Tench (*Tinca tinca*) broodstock management in breeding station under conditions of pond culture: a review

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**Abstract.** A basic assumption for carrying out the breeding work is to have well-organised broodstock management. Taking into account the number of populations bred and the limited number of ponds available, it is impossible to avoid rearing several juvenile or adult breeds in mixed stocks. Fish of individual breeds are group-marked with regular renewal of the freeze-branded mark and broodstock adults should be marked individually. To minimise inbreeding or losses in genetic variation, at least 120 fish per strain are reared and when the strain is restored, factorial crosses of at least 15 females and 25 males should be applied. All activities are individually registered in the “Evidence 2003” data-recording software and data can be sorted by the origin of the fish population, of the strain or of individual fish; reproductive and performance parameters are recorded individually also. Further breeding operations with tench comprise the selection after over-wintering and rearing of fish before, during and after the reproductive season.

### Introduction

Tench, *Tinca tinca* (L.), is a supplemental species for pond aquaculture in several European countries (Steffens 1995) or in extensive open water fisheries. In Europe, tench is utilised for food, for leisure purposes such as angling and as an ornamental fish. Its domestication started only recently (Billard and Flajšhans 1995), reflecting the strategy of studying new candidate fish species for diversification of aquaculture production. Apart from Europe, the aquaculture production of tench has increased dramatically in China since 1998 (Wang et al. 2004). In the Czech Republic, tench accounts for 1% (150–200 tons) of total annual fish farm production, ranking fourth after common carp (*Cyprinus carpio*; 87% of total production), salmonids (*Oncorhynchus* sp., *Salmo* sp.; up to 5%) and herbivorous fish (*Hypophthalmichthys molitrix*, *Ctenopharyngodon idella*; up to 4%) (Czech Fish Farmers Association (<http://rybsdr.fishnet.cz/>)) (Table 1). Tench is produced semi-intensively as supplemental fish in pond polyculture with other cyprinids, mainly for export on the European market.

Table 1. Production of marketable tench in the Czech Republic.

Year	2000	2001	2002	2003
Total fish production (tons)	15,815	16,693	15,996	16,609
Tench production (tons)	202	150	158	181
Proportion of tench (%)	1.28	0.90	0.99	1.09

Our attention has been focused on tench since the 1970s. Research activities have been carried out to assess its genetic parameters using selective breeding and testing programmes (Kvasnička and Linhart 1990), to conserve genetic resources of tench breeds as a part of the national heritage (Flajšhans et al. 1999; Flajšhans, 2002), to extend the existing bank of cryopreserved sperm of tench breeds (Rodina et al. 2004b), to assess effects of genome manipulations as reviewed by Flajšhans et al. (1995, 1998, 2004) and Buchtová et al. (2003, 2004), as well as to improve the protocols of artificial propagation and off-spring rearing (Linhart et al. 2000a, b, 2003a, b; Gela et al. 2003; Rodina et al. 2004a).

The basic structure of breeds, individual breeds, colour mutations and inheritance of colours are characterised in the papers by Gela et al. (1998) and Kvasnička et al. (1998).

### Goals and strategy of tench farming

In general, the goal of tench farming in the Czech Republic is aimed to optimise market production of this species, both qualitatively and quantitatively, under reasonable economic conditions. For these reasons, tench is considered an economically important freshwater aquaculture species, whose farming and selective breeding is regulated by the Czech Animal Breeding Act No. 154/2000 on farm animal breeding and data recording. These activities are controlled and guaranteed by the Czech Fish Farmers Association *via* its expert bodies. In particular, the Department of Fish Genetics and Breeding of RIFCH USB Vodňany and its breeding station were entrusted with the development of the breeding programme for tench and its practical application in fish farming.

The basic tools of fish breeding comprise selection, hybridisation and *in situ* conservation of genetic resources in live gene banks. Therefore, the strategies are diversified as follows.

#### *Farming of breeds according to principles of genetic resources conservation*

Five breeds of wild (olive-greenish) phenotypes, characterised by Gela et al. (1998; Vodňany, Hluboká, Tábor, Mariánské Lázně and Velké Meziříčí) and two colour variants (golden and blue; Kvasnička et al. 1998) are kept in live gene banks in order to maintain their genetic variability and to minimize

inbreeding effects (Flajšhans et al. 1999). Foreign imported and acclimated breeds (Hungarian, Romanian and German – Königswartha) as well as a newly generated Alampic colour variant (Kvasnička et al. 1998) are maintained under the same conditions.

#### *Selection programme*

The golden-white colour variant is aimed to suppress the occurrence of dark melanin spots on these fish with golden dorsal and white ventral body parts.

#### *Hybridisation programme*

This is focused on enhancement of selected performance traits (e.g. survival rate of offspring, growth rate) by producing  $F_1$  hybrids, as described by Kvasnička and Linhart (1990) and Kohlmann et al. (1998).

#### *Testing programme*

This is based upon performance tests of purebreds and crossbreds in ponds, following the same methodology as those for common carp (Linhart et al. 2002; Kocour et al. 2003) with control lines consisting of tench of different colour phenotype from the fish tested.

When farming individual breeds, adequate numbers of young brood fish and broodstock are needed. Fish are marked and/or tagged in order to prevent mistaken identifications and replacement. Group marking by means of freeze-branding can be performed on fish of 3–4 cm length (Figure 1). Passive Integrated Transponder (P.I.T) tagging (125 kHz TX1400L by Destron, USA or 134.2 kHz ID162 by AEG, Germany) is employed for three-year-old fish and data are simultaneously recorded into a national computer database for fish breeds “Evidence 2003” (Figure 2), a Microsoft Access application.

#### **Breeding cycle of tench broodstock in a closed farming system**

The Department of Fish Genetics and Breeding of RIFCH USB Vodňany employs a complete system of tench farming, i.e. the production of yolk-sac larvae from its own broodstock, rearing the juvenile and sub-adult categories until young brood fish are selected and classified into the broodstock. This approach is applied specifically for each breed, taking into account the results of isozyme and/or microsatellite DNA analyses assessing the genetic variation and inbreeding per generation (Šlechtová et al. 1998; Šlechta et al. 2000; Tong et al. 2002). To prevent increasing the rate of inbreeding, selected genetically



*Figure 1.* Group marking of tench yearlings by branding with an aluminium matrix frozen in liquid nitrogen. Photograph by Nicolas Caille, Vodňany (2004).

typed spawners of the same breed are introduced from other fish farms, if reasonable (Šlechtová et al. 1995).

#### *Pre-spawning preparation in spring*

The first step for successful artificial reproduction is spring selection of enough females and males in good condition without visible health problems, performed at the harvest of over-wintering ponds in late March and early April. Selected spawners are stocked, separately by sex, into earthen ponds ( $400 \text{ kg ha}^{-1}$ ) for “pre-spawning preparation”. This period of preparation is especially important for tench, as substantial quantitative developmental changes in tench gonads, particularly in ovaries, take place with increasing temperature during spring (Linhart and Billard 1995). For this reason, it is essential to provide the fish daily with good quality feed (pellets for cyprinids) up to 5% of fish biomass, as well as to enhance the development of natural food and to check the physical properties of the water in the ponds.

Spawners are fished out from preparatory ponds 2 days before the artificial propagation and the ripe females selected for propagation. Selection is based

The screenshot shows a software window titled "Evidence ryb , vybraná databáze : Databáze vurh.mdb". The menu bar includes "Soubory", "Data", "Výběry", "Tisky", and "Nastavení". The main area is titled "Hledání" and contains a list of specimen IDs on the left and a detailed data entry form on the right.

**Specimen List:**

- 6/171
- 6/28
- 6/29
- 6/41
- 6/42
- 6/46
- 6/50
- 6/52
- 6/57
- 6/6
- 6/87
- 6/89
- 6/1
- 6/10
- 6/11
- 6/118
- 6/135
- 6/139
- 6/147

**Data Entry Form:**

- Species:** Lin obecný
- Breed:** Lin vodnanský
- Breeding group:** LVO-VO-98/1
- Individual ID:** 6/139
- Date of tagging:** 01.04.2003
- Registration No.:** LVO-VO-98/1-6/139
- Fin-clip marking:** (empty)
- Sex:** F
- Other marking:** 6
- ID:** 6/139
- Weight (g):** 493
- To date:** 12.6.2003
- Selection:** A
- To date:** 1.4.2003
- Art. propagation:** A
- To date:** 12.6.2003
- Eggs (g):** 30,6
- To date:** 12.6.2003
- Sperm (ml):** (empty)
- To date:** (empty)

Figure 2. Main table of the locally developed software "Evidence 2003" to record breeding data on individual spawners.

on visual examination (body cavity filled with eggs) or upon ovarian biopsy aimed to assess the degree of oocyte maturation, if appropriate. The latter approach is highlighted in Dettlaff et al. (1993).

#### *Artificial reproduction of tench*

According to the public notice of the Czech Ministry of Agriculture No. 471/2000 for pure-breeding or cross-breeding, factorial crossing employs at least 15 females and 25 males. The selected fish are stocked into 6 m<sup>3</sup> holding tanks whose temperature and dissolved oxygen content can be controlled. Water temperature is maintained between 20 and 22°C. The spawners are hormonally stimulated as described by Linhart et al. (2000a). Males are intramuscularly injected with acetone-dried carp pituitary powder suspended in physiological saline at 1.5 mg kg<sup>-1</sup> body weight 36 h prior to sampling of sperm. Sperm is collected into syringes containing immobilising solution at a sperm:immobilising solution ratio of 1:2 (Rodina et al. 2004a; Linhart et al. 2006). Females are injected intramuscularly with gonadotropin releasing hormone (GnRH) analogue (Kobarelin, Institute of Organic Chemistry and Biochemistry, Acad. Sci. Czech Rep.), dissolved in physiological saline at a dose of 5 µg kg<sup>-1</sup> body weight. Ovulation occurs after 20–24 h at 21 °C (Linhart et al. 2000a). Ovulated eggs from individual females are stripped into separate dry bowls. Eggs

are pooled proportionally with diluted sperm in immobilising solution, allowing the process of gamete activation (Linhart et al. 2006).

The stickiness of eggs is removed prior to incubation in conventional Zuger jars by means of alcalase, an enzyme derived from *Bacillus licheniformis* (Calbiochem cat. No.126741) according to the procedure described by Linhart et al. (2000a, b; 2003a, b) and Gela et al. (2003).

#### *Tench nursing until the yearling stage*

Swimming-up yolk sac larvae are stocked into 0.02 ha ponds at 250 000 larvae  $\text{ha}^{-1}$ . Ponds are prepared in advance by the addition of manure (1.5 t  $\text{ha}^{-1}$ ) which aids the development of natural food. Usually, one breed is stocked into one pond while in the case of performance testing, the pure-bred or hybrid strain tested is stocked proportionally together with a control line differing in colour phenotype. Supplemental feeding (pollard or cereal meal) is applied *ad libitum* once the fish reach 3 cm long. Fish are harvested at the end of the season, weighed individually (min. 33 fish), survival rate calculated (usually 10–30%), and the group assessed statistically, and compared with other breeds or cross-breeds. Over-wintering is recommended as a monoculture in separate ponds.

#### *Rearing tench until 2 years old*

After group marking the fish by means of freeze branding, all groups reared can be stocked together with their identically marked control lines into a communal pond in triplicate for performance testing as described for common carp (Linhart et al. 2002; Kocour et al. 2003). Fish are fed appropriately sized common carp pellets (Grana Písek, Czech Rep.). The assessment of parameters after the growing season and/or over-wintering is the same as stated above.

#### *Rearing tench until classification as broodstock*

The two-year-old tench can be reared in polyculture ponds (400 kg  $\text{ha}^{-1}$ ) at a 2:1 ratio, with young brood common carp, herbivorous fish and/or with one- to two-year-old ornamental fish, e.g. koi carp, golden orfe (*Leuciscus idus*) or golden rudd (*Scardinius erythrophthalmus*). Feeding pattern and assessment of parameters after the harvest is the same as stated above.

When classifying the young tench into the broodstock, every breed is marked with a numeric freeze brand for group selection and an individual P.I.T. tag for selection of individuals and for data recording in the “Evidence 2003” software. Such a system requires large rearing capacities, highly skilled labour and expensive tagging. It is also time-consuming and requires a high degree of

knowledge. On the other hand, it provides conditions for keeping a high number of breeds in one fish farm, for handling them as live gene banks, for performing the breeding programme, and for minimising the danger of wrong identification of fish when carrying out the research.

### Acknowledgements

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**Erratum**

In the article, Tench (*Tinca tinca*) broodstock management in breeding station under conditions of pond culture: a review. Gela, D., Flajšhans, M., Kocour, M., Rodina, M. and Linhart, O. (this issue) there is incorrect citation and an incorrect reference given.

Please note that in the sub-section *Artificial reproduction of tench* (Paragraph 1) there are two instances where

Linhart et al. 2006      should read:      Linhart et al. 2005

Similarly, in the Reference section

Linhart O., Rodina M., Kocour M. and Gela D. (2006). Insemination and gamete management in tench, *Tinca tinca* (L.). Aquacult. Int. (this issue).

Should read:

Linhart O., Rodina M., Kocour M. and Gela D. (2005). Insemination and gamete management in tench, *Tinca tinca* (L.). Aquacult. Int. (this issue).

The publisher regrets these errors and any inconvenience that may have been caused.