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Intensive rearing of African sharptooth catfish (Clarias gariepinus)

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1. INTRODUCTION

1.1. Taxonomy and biological characteristic

African sharptooth catfish, *Clarias gariepinus* (Burchell 1822; Fig. 1) belongs to the class Actinopterygii (ray-finned fish), order Siluriformes (catfish), family Clariidae (labyrinth catfishes) which includes about 100 species in 13 genera (Hanel & Novák, 2004). Czech name for this species is not steady. In Czech literature, several synonyms can be found – e.g. sumeček africký (Adámek, 1994; Hamáčková *et al.*, 2007), keříčkovec červenolemý (Hanel, 1997), klarias africký (Kůrka *et al.*, 2000), sumčík africký (Pokorný *et al.*, 2004; Hamáčková *et al.*, 2007). Present valid Czech name is keříčkovec jihoafrický (Hanel & Novák, 2004).

Species of the family Clariidae inhabit stagnant freshwaters of Syria, Southeast Asia (Philippines and Java), Malaysia, Africa and Madagascar. Outside the African continent, the African sharptooth catfish occurs in Asian countries of the Mediterranean coast. Northern boundary of its distribution is southern Turkey (Viveen *et al.*, 1986). It was introduced and occurs in the wild as well as in Florida in USA (de Graaf & Janssen, 1996).



Fig. 1. African sharptooth catfish, Clarias gariepinus (Burchell 1822). Photo by J. Kouřil

African sharptooth catfish populations occurring in various parts of Africa were originally named by different names: *Clarias mossambicus* (eastern part), *Clarias lazera* (northern and central part), *Clarias senegalensis* (western part) and *Clarias gariepinus* (southern part). However, i tis always one and the same species (Teugels, 1984).

In natural habitats, the African sharptooth catfish is a species well adaptable to diverse environmental conditions. It occurs in various types of African inland waters, both standing and slowly flowing, with an average temperature of 25 °C. It succeeds in

both shallow and muddy waters and in deep lakes with relatively clean water. During the rainy season, it migrates to spawn into the shallow tributaries (Hecht *et al.*, 1988).

African sharptooth catfish body is torpedo-elongated without scales. Dorsal and lateral parts are dark gray to olive colour, ventral part is white colour. There are also individuals with bright spots or bright colour throughout the body. The head is dorso-ventrally flattened with strong bone structure of the skull. There are 4 pairs of long barbels around the mouth. The dorsal fin extends to the caudal peduncle with 68–79 soft fin rays. The first fin rays of the pectoral fins are hard and serrated on the inside (Hamáčková *et al.*, 2007).

The family Clariidae is characterized by occurrence of suprabranchial accessory airbreathing organ formed by arborescent extensions of mucosa of the branchial cavity above the gill arches (Baruš & Oliva, 1995). The arborescent organ allows the survival of labyrinth catfishes in waters with low or zero oxygen content (adaptation to drought, when the water remains only in the deepest places of periodically flooded areas on places of its natural distribution). The ability to breathe also atmospheric oxygen is one of the major reasons why African sharptooth catfish was successfully introduced into intensive aquaculture (Hamáčková *et al.*, 2007).

Due to the construction of its body and adaptability, this species is able to intake a wide range of food from tiny zooplankton to fish prey reaching almost a half of its body length. Its short and extended esophagus allows it to eat even larger prey. In the stomach, the food is diluted and continues into the intestine which is simple, thin and relatively short. Due to this fact, African sharptooth catfish is dependent on intake of high-protein food. The digestive system develops relatively quickly, it allows early transfer to dry starter food in case of intensive reared fish, unlike some other fish species (Hecht *et al.*, 1988).

Because of the great adaptability and wide range of inhabited waters, the information about growth rate of African catfish differ considerably. In nature, under optimal conditions, it reaches usually about 200-300 mm of total length at the first year of life. In subsequent years, the yearly growth is 80-100 mm. In nature, it grows to the maximum total length of 140 cm and weight of 40-60 kg. The largest specimens are found primarily in large turbid rivers (Hecht *et al.*, 1988).

The African sharptooth catfish is characterized by mostly evening and nocturnal activity of food intake. In nature, it feeds especially predatory on plankton and bentos (various invertebrates and their developmental stages) and amphibians. Juveniles and adults intake mostly smaller fishes, including dead fish (Hecht *et al.*, 1988; Yalcin *et al.*, 2000).

In experimental conditions, Adámek *et al.* (1989) studied food selectness of African sharptooth catfish of weight about 220 g, originally bred in intensive rearing. From eight offered food fish species in individual total length about 12-22% of length of African sharptooth catfishes, catfishes preferred primarily Belica, *Leucaspius delineatus* (Heckel,

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1843) and Rudd, *Scardinius erythrophthalmus* (Linnaeus, 1758). Conversely, negative selection was demonstrated in Nile tilapia, *Oreochromis niloticus* (Linnaeus, 1758) and Stone maroko, *Pseudorasbora parva* (Temminck & Schlegel, 1846). Generally, intensity and efficiency of predation was quite low with regard to the predation strategy in African sharptooth catfish. After multiple attacks the injured or dead prey was eaten during the night. Achieved feed conversion ratio (FCR) in African sharptooth catfishes fed by live fishes was 4.73 and specific growth rate (SGR) 0.39%.day⁻¹. These information partly refuted a hypothesis about the utility African sharptooth catfish breeding in polyculture with Nile tilapia, where the African catfish should partly correct the number of fish and population density of fast reproducing Nile tilapia (Adámek *et al.*, 1989).

Sexual dimorphism is strongly developer in African sharptooth catfish. Males are characterized by longer sexual papilla of conical shape, females have star-shaped papilla and visibly enlarged abdominal part before spawning period (Hamáčková *et al.*, 2007) – see Fig. 2 and 3. In nature at the beginning of the rainy season, the broodstock migrate into vegetated shallow tributaries where they spawn on plant substrate. Parental care was not recorded, generation fish return back into their original habitat. Several months after hatching, the progeny stays in vegetated, shallow waters and migrates downstream into larger streams and lakes at the beginning of dry season.



Fig. 2. Detail view of organization of male and female genital papilla in African sharptooth catfish (A and B, respectively). Photo by J. Kouřil



Fig. 3. Male (left) and female (right) of African catfish. Photo by J. Kouřil

1.2. Rearing methods

Studies of Dutch authors (Hogendoorn, 1979, 1980, 1981; Hogendoorn & Vismans, 1980; Hogendoorn *et al.*, 1983; Viveen *et al.*, 1986) were crucial for development of African sharptooth catfish breeding. Soon, is intensive rearing was spread from Netherlands to many other countries in Europe (especially into Hungary, but also in Germany and Poland) and elsewhere in the World (Fig. 4 and 5). Main reasons for its rearing in aquaculture are high adaptability to the environment (except low temperature), low demands on oxygen, high stocking density, great growth rate and high muscle quality (high dietetic value, excellent taste and lack of muscle "Y" bones). It was introduced in current place of the Czech Republic in 1989 (Pokorný *et al.*, 2004).



Fig. 4. Intensive farming of African sharptooth catfish in Szarvas (HAKI – Research Institute for Fisheries, Aquaculture and Irrigation) in Hungary: view in the nursery hall outside (A), tank with farmed fishes (B). Photo by J. Kouřil



Fig. 5. Exterior (A) and interior (B) of originally agricultural building which was adapted and rebuilt a farm with intensive rearing of African sharptooth catfish (firm "Krolestwo ryb") in Pielgrzymowice in Poland. Photo by J. Kouřil

Rearing of African sharptooth catfish is not possible in outdoor tanks with natural water temperature during the greater part of the year, due to low water temperatures. However, either year-round breeding in water-flow or closed-water systems with water temperatures above 20 °C, or a combination of summer rearing in ponds (Adámek, 1994; Adámek & Sukop, 1995) with aforementioned breeding in systems with heated water is feasible. Thanks to possibility of African sharptooth catfish breeding in high stocking densities, this fish belongs among the most suitable species for breeding in

recirculation aquaculture systems (hereinafter RAS; Hamáčková et al., 2007). Intensive fish rearing RAS represents important alternative to intensive fish production in flow water systems and pond rearing (Kouřil et al., 2008). As evidence, the development of African sharptooth catfish production in Hungary may be considered, where it reaches about 15% of total fish aquaculture production. In RAS optimal rearing conditions can be maintained in terms of both water quality and dosing feed. Recirculation aquaculture systems are systems with partially or completely closed water circulation. They are independent on external environment, they have low demands on amount of water and limited build-up area. In such devices, all water, or at least part of water used for fish farming is purified and modified in such way that it can be reused. Fish excrements and any feed remnants are removed from the water by sedimentation and mechanical filtration. End product of protein metabolism - ammonia is oxidized by biological way (not chemical) with using of biological nitrification filters. Ammonium is converted to nitrites and subsequently to nitrates by nitrification process. Through denitrification process, these ions can be converted up to molecular nitrogen which escapes into the atmosphere. Next product of metabolism – carbon dioxide have to be removed from water with using of outgassing (with aeration or oxygenation). In this way, the dissolved oxygen in water is also replenishment. Water constantly circulates in RAS. Only, a small amount of circulating water is replaced by fresh water. Usually, it is a volume from 0.1% to 10% of total volume of the RAS. That's way, the RAS are characterized by a high fish production with use of very small built-up area and need for low inflow of water (Kouřil et al., 2008).

The African sharptooth catfish is very resistant fish species which is capable to inhabit waters with very low oxygen level thanks to its accessory air-breathing organ. After Britz & Hecht (1987), temperature of 26-32 °C is an ideal for its intensive rearing. Outside this thermal range, temperatures reduce its growth. The African sharptooth catfish can be reared in water with higher salinity. Salinity up to 0.5% is acceptable for rearing of its fry. Regarding the survival rate, the salinity of 0.75% is suitable for this species (Britz & Hecht, 1989). According Hamáčková et al. (2007), the oxygen saturation is also important for fry before the start of breathing atmospheric oxygen. In this period, oxygen saturation above 90% is appropriate. In any case it must not fall below 40%. The pH should be maintained between 6.5 and 8.0. Mortality occurs when pH value exceeds 11 and conversely, it decreases when pH value is below 4. After Adámek (1994), the African sharptooth catfish survives without consequences the short-term drop of temperature below 12 °C. However, the fish are affected by fungus and die during long-term decline below 15 °C. The upper lethal temperature is very high (above 40 °C). Several days after hatching (in dependence on water temperature), yolkfeeding larvae search cover and accumulates in the dark parts of the tank. Therefore, overshadow above inlet part of rearing tank is recommended (Viveen et al., 1986; Hamáčková et al., 2007). In comparison to permanently illuminated environment, dim

conditions cause higher survival rate of fry (Britz & Pienaar, 1992; Appelbaum & Kamler, 2000). Also, rearing of fish in market size takes place either in the shadow or complete darkness.

In conditions of intensive rearing, the reproduction of African sharptooth catfish is carry out by hormonally induced artificial spawning with using of carp pituitary or synthetic hormonal preparations based on GnRH (*gonadotropine releasing hormone*). Females are highly fertile and their spawning is quite easy. The relative working fecundity (number of spawned eggs per kg of female weight) averages 100-150 thousands eggs (Adamek, 2001; Brzuska *et al.*, 2004). Females sexually mature at age of 6 to 7 months. In terms of spawning and subsequent fry rearing, the best results are achieved in females at the age of 2-3 years. Males become sexually mature at the age of 1.5-2 years. Both sex of broodstock fish can be kept together in one tank. Optimal water temperature for rearing of broodstock is 23-25 °C (Hamáčková *et al.*, 2007).

Good feed conversion of intensively reared African sharptooth catfish is dependent on quality of food supply. The African sharptooth catfish is omnivorous fish species characterized by high activity of digestive enzymes (amylase, lipase and protease; Fourie, 2006). Feeding experiments performed by Hecht *et al.* (1988) demonstrated the value of feed conversion ratio (FCR) about 1.0 with prerequisite for further improvement.

Although, the African sharptooth catfish is classified as omnivorous fish species, its intenstine is simple, thin and relatively short, which means that it is dependent on a diet rich in protein. Based on previous feeding experiments, the best parameters of food conversion were observed with using of diet containing of 38-42% crude protein and 8-12% fat (Hecht *et al.*, 1988). De Graaf & Janssen (1996) recommend diet with 35-42% of protein and 12 kJ.g⁻¹ of digestible energy.

Based on later research of de Graaf & Janssen (1996), the optimal nutrient content in the dry matter of feed was defined for individual age categories of African sharptooth catfish. Optimal content of 35-40% digestible protein and 12-16 kJ.g⁻¹ of digestible energy is equally recommended for fry and broodstock. Market size fish should be fed by food containing of 30-35% of digestible protein and 10-14 kJ.g⁻¹ of digestible energy. As well, the recommended range of calcium (Ca) and phosphorus (P) content achieves 0.8-1.5% of Ca and 0.6-1.0% of P in food for fry and broodstock. In the case of fish in market size, it should be 0.5-1.8% and 0.5-1.0% (Ca and P, respectively). Also, minimum requirements for the content of some amino acids were studied. Methyonin and cystyne concentrations are about 1.2% in fry, 0.9% in market size fish and 1.0% in broodstock. Lysin content should reach 2.0 % in fry, 1.6% in market size fish and 1.8% in broodstock.

Water temperature is an important factor influencing growth and feed conversion. It significantly affects intensity of feed intake in African sharptooth catfish. Hogendoorn *et al.* (1983) recommend feed rations in % of total biomass for individual

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weights (from 1 to 200 g) in dependence on temperature (range 21-33 °C) with using of commercial feeding (protein content of 50%) for Rainbow trout, *Oncorhynchus mykiss* (Walbaum, 1792).

After the above study, the highest feed rations were recorded at temperatures in range of 27-29 °C. Thereafter, Adamek (2001) refined these values (see Tab. 1) for rearing of African sharptooth catfish fry and juveniles (fish average weight 1-150 g). The table is supplemented with temperature, weight of reared fish and presumed relative daily weight gain. Adamek (2001) also recommended daily feed ratios for breeding of market size fish of African sharptooth catfish (Tab. 2).

Tab. 1. Recommended relative daily feed rations (in % of the stocking biomass per day) and presumed growth rate (in % of weight gain of fish per day; numbers in brackets) in rearing of African sharptooth catfish fry and juveniles in dependence on water temperature and individual weight of fish (Adamek, 2001).

Water tempera			Average i	ndividual w	reight (g)		
	1	5	15	25	50	100	150
20	4.8 (3.5)	4.3 (3.0)	3.6 (2.5)	2.4 (1.4)	1.4 (0.7)	0.9 (0.3)	0.7 (0.3)
21	5.4 (4.3)	4.8 (3.8)	4.3 (3.2)	3.0 (2.0)	1.9 (1.1)	1.2 (0.5)	1.0 (0.4)
22	5.9 (5.2)	5.4 (4.6)	4.9 (4.0)	3.7 (2.7)	2.6 (1.5)	1.7 (0.8)	1.5 (0.7)
23	6.3 (6.1)	6.0 (5.5)	5.5 (4.9)	4.4 (3.4)	3.3 (2.1)	2.2 (1.2)	2.0 (1.1)
24	6.8 (7.1)	6.4 (6.4)	6.1 (5.8)	5.1 (4.2)	3.9 (2.7)	2.7 (1.6)	2.2 (1.4)
25	7.2 (7.9)	6.9 (7.3)	6.5 (6.6)	5.7 (5.0)	4.5 (3.3)	3.1 (2.0)	2.4 (1.8)
26	7.5(8.7)	7.2 (8.1)	6.9 (7.3)	6.1 (5.6)	5.0 (3.8)	3.4 (2.3)	2.4 (2.0)
27	7.7 (9.3)	7.4 (8.6)	7.1 (7.9)	6.4 (6.1)	5.2 (4.2)	3.5 (2.5)	2.4 (2.0)
28	7.8 (9.8)	7.6 (9 0.)	7.3 (8.2)	6.4 (6.3)	5.2 (4.3)	3.5 (2.5)	2.3 (1.9)
29	7.8(10.0)	7.6 (9.2)	7.2 (8.3)	6.3 (6.2)	5.0 (4.1)	3.2 (2.2)	2.1 (1.6)
30	7.8 (10.0)	7.4 (9.1)	7.0 (8.2)	5.9 (5.9)	4.5 (3.7)	2.8 (1.9)	1.8 (1.3)
31	7.8 (9.7)	7.2 (8.8)	6.7 (7.8)	5.4 (5.4)	3.9 (3.2)	2.3 (1.4)	1.5 (0.9)

Individual weight of fish (g)							
	20	22	24	26	28	30	32
100-300	1.2	2	2.5	3.2	3.5	3.2	3
300-800	1	1.7	2.2	2.8	3.1	2.9	2.8

Tab. 2. Recommended daily feed rations (in % of fish biomass) in dependence on water temperature during rearing of African sharptooth catfish in market size.

Unlike the vast majority of the other fish species, the African sharptooth catfish is characterized by muscle (meat) typically red in colour with very little fat and high protein content. From culinary point of view, its muscle has excellent taste properties (Hamáčková *et al.*, 2007). Osibona *et al.* (2009) studied meat composition of African sharptooth catfishes purchased from local fishermen in Lagos (Nigeria) with representation of 18.8% protein, 9.3% fat and 1.2% ash. Fourie (2006) enumerates various supply options of African sharptooth catfish for consuming purposes: whole fish (only killed), gutted fish (removed viscera and head, but fins and skin remain), processed body (without viscera, head and fins), steaks (slices 20-25 mm wide prepared by a transverse cut through processed body) and fillets (with skin or skinless).

The African sharptooth catfish is native in Africa, where it is the second most important fish species in intensive aquaculture after Nile tilapia. Demand for fish continues to increase and the production of African sharptooth catfish is an important part of the national economy of countries as Nigeria, Kenya, Cameroon, Mali and South Africa. Primarily, African sharptooth catfish is reared in small ponds in the one-year to two-year production cycle, it often is reared in polyculture with Nile tilapia. Exceptionally, African sharptooth catfish is reared in cages (Hecht et al., 1988). Cage culture of African sharptooth catfish in heated waters is also realized in Bulgaria (Kouřil, unpublished). Especially, Nigeria, Netherlands, Hungary, Kenya, Syria, Brazil, Cameroon, Mali and South Africa belong among the major global producers of African sharptooth catfish. At the beginning of the 90s of last century, several farms in Belgium, Germany and Hungary reached up production of African sharptooth catfish from 5 to 200 tons per year. Some farms started to specialize in reproduction and rearing of fry, others in production of market size fish. Based on the FAO (Food and Aariculture Organization of the United Nations) statistics, African sharptooth catfish production in aquaculture has grown exponentially over the last thirty years. In 1980, the global production of African sharptooth catfish was about 50 tons, in 1990 already 1.5 thousand tons, in 2000 it was 5.5 thousand tons and in 2010 even 191 thousand tons (see Fig. 6, Pouomogne, 2012 - FAO statistics).



Fig. 6. Worldwide annual production (thousand tonnes per year) of market size African sharptooth catfish in aquaculture in the years 1970-2011 (Pouomogne, 2012 – FAO statistics).

Artificial spawning of African sharptooth catfish females was usually induced by hormonal stimulation with using of Common carp, *Cyprinus carpio* (Linnaeus, 1758) and exceptionally synthetically produced combined hormonal preparations were used. Carp pituitary is once injected intramuscularly or intraperitoneally at a dose of 2 to 3 mg.kg⁻¹ as a suspension of crushed pituitary and dissolved in physiological saline solution. At temperature of 25 °C, the spawning occurs after 11 h from using of carp pituitary. Water temperature significantly influences the length of latency interval (Adamek, 2001; Hamáčková *et al.*, 2007). Length of latency period (i.e. time from injection till the egg ovulation) and length of incubation period of eggs is dependent on water temperature (see Tab. 3).

Tab. 3. Length of latency interval (period from injection till spawning) and length of incubation
period (time interval from egg fertilization till hatching) in African sharptooth catfish at different water
temperatures (18–30 °C; Adamek, 2001).

Temperature (°C)	Length of latency interval (h)	Length of incubation period (h)
18	21	57
21	18	46
22	15	38
23	13	33
24	12	29
25	11	27
26	10	25
27	9	23
28	8	22
29	7.5	21
30	7	20

Recently, synthetic hormonal preparations are used more and more for induction of artificial spawning in various fish species in the Czech Republic. Utilization of GnRH - Kobarelin (D-Ala⁶, Pro⁹ NHEt-mGnRHa) and Lecirelin (D-Tle⁶, Pro⁹ NHEt-mGnRHa), as well as combined preparation, e.g. Ovopel (firm AgroFish, Hungary) containing above mentioned analogue GnRH and dopaminergic inhibitor metoclopramide is proved. Hungarian preparation Ovopel is applied once intramuscularly or intraperitoneally in doses of 10-40 µg.kg⁻¹ (1 pellet per 1 kg of female). After Ovopel injection, female ovulation occurs in 12-13 h at temperature of 24-25 °C (Kouřil *et al.*, 2011).

After hormonal injection, it is absolutely necessary to retain the females separately (one fish per tank) in a perfectly covered tanks, because of their increased aggressiveness and trying to escape from the tank. Males can be kept together before the spawning. They should not be fed 1-2 days before the planned injection. Optimal water temperature for reproduction is 25-27 °C. Before the artificial spawning, females have to be anaesthetized using clove oil (at a dose of 0.04-0.05 ml.l⁻¹ of water) or 2-phenoxyethanol (at a dose of 0.3-0.5 ml.l⁻¹ of water). Before a hand stripping of female, the ventral body part and fins have to be dried. Relative weight of total spawned eggs is 10-20% of female weight before spawning. Spawned eggs are from yellow-green, green to brown-green colour. Weight of egg is about 1.4 mg, i.e. 1 kg of

dry mass of spawned unswollen eggs contains approximately 700 thousand eggs (Hamáčková *et al.*, 2007; Kouřil *et al.*, 2011).

Milt is obtained from just killed males by preparation of gonads. Mature gonads should be white or cream colour. Dissected gonads have to be dried and then cut by scissors. Consequently, pieces of gonads will be sieved through a dry sieve or inert textile directly on the mass of eggs which are divided after 200-300 g separately into dry containers. Equally, milt can be firstly collected into glass containers and subsequently used. Egg fertilization is carried out in containers which were used during spawning. When eggs and sperm are mixed together, water is poured and mixture of water and gametes is mixed again. After a further 2-5 min, fertilized eggs are washed with water and impure water with sperm remnants is rapidly decanted. Thereafter, fertilized eggs should be properly spread in incubation tank, so that they stick on the submerged sieve.

African sharptooth catfish eggs can be also incubated in Zuger jars. In this case, eggs have to be unsticked. However, there are fish hatcheries where water composition allows egg incubation without unsticking. Clay or tannin suspension (tannin concentration 0.7-1 g.l⁻¹ of water) can be used for egg unsticking. Before preparation of the suspension, tannin is firstly dissolved in a little volume of warm water. In this solution, the unsticking of eggs is realized by two short baths, both for 20 seconds. Between these baths, as well as after completion of unsticking process, the eggs are decanted by sufficient volume of water. Then, the eggs are placed into incubation jars and the water flow is adjusted (Hamáčková *et al.*, 2007; Kouřil *et al.*, 2011).

Because of African sharptooth catfish is a tropical fish species, there are numerous modifications of its rearing methods. In countries of natural distribution, it is usually reared in non-flow ponds or various sophisticated breeding systems when it is fed by fish, waste of various origins or fodder mixtures. Currently, it is reared in countries of temperate climate zone (i.e. less suitable climate areas) in various breeding systems. In these countries, rearing of African sharptooth catfish can take place seasonally in ponds (only in summer) and in tanks and cages in flow-water systems with heated water (thermal water, cooling water from industry). Its controlled reproduction is usually provided using hormonal stimulation with subsequent artificial spawning and egg incubation. Fry rearing is a separate section of African sharptooth catfish breeding for first two months after hatching that is more difficult due to higher requirements for sufficient amount of dissolved oxygen before the onset of additional breathing, rearing hygiene and adequate nutrition. Due to cannibalism, a loss of fish can occurs till individual weight of 200-300 g. Up to its market size, African sharptooth catfish can be reared in extremely stocking densities (up to 300-400 kg.m-3) in flow-water or recirculating systems at relatively low oxygen level and high organic load. This is a difference from other intensively reared fish species, for which the stocking density

generally must not exceeds 100 kg.m³ during their rearing. The African sharptooth catfish grows rapidly, efficiently utilizes the food, it is characterized by high quality of product without intermuscular bones. These properties make it a very perspective fish species for intensive farming. Its production cycle is shown in Fig. 7.

By determining of consumer value of African sharptooth catfishes (an average weight of 340 g), Krupka (1998) recorded yield of fillets without skin about 52%, the relative weight of body without head, fins and viscera reached 70% and the relative weight of body without head, fins, viscera and skin was about 62%.



Fig. 7. Production cycle (after Viveen et al., 1986).

Successful hybridization between African sharptooth catfish, *Clarias gariepinus* (Burchell 1822) and vundu catfish, *Heterobranchus longifilis* (Valenciennes, 1840) (Hecht & Lublinkhof, 1985; Legendre *et al.*, 1992). Legendre *et al.* (1992) found that these hybrids are viable and their survival is comparable to native species. Growth rate of vundu catfish and its hybrids with African sharptooth catfish is faster than growth rate of pure line of African sharptooth catfish. However, neither vundu catfish nor hybrids with African sharptooth catfish are not reared for commercial purposes in the Czech Republic. Next species of genus *Clarias* Philippine catfish, *Clarias batrachus*

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(Linnaeus, 1758) (Fig. 8) inhabits Southeast Asia. It is characterized by high variability in colour, including the frequent occurrence of albinos in natural conditions. However, it grows to smaller size than African sharptooth catfish.



Fig. 8. Philippines catfish – Clarias batrachus (Linneaus, 1758). Photo by M. Kořínek – www.biolib.cz

2. AIM

The aim of this publication is a publication of eleborated technological procedure of intensive farming in African sharptooth catfish in heated water, especially recirculating aquaculture systems in the Czech Republic. Including, breeding of broodstock, their artificial reproduction, manipulation with gametes, egg incubation, fry and market size fish rearing. The present technology also includes the results of testing the production efficiency of commercially produced feed with impact on product quality (with using of these methods – yield, organoleptic assessment, basic chemical analysis of meat).

3. FACILITIES FOR AUTHENTICATION OF TECHNOLOGY

Athentication of technology take place at three workplaces. Part of experiments focused on artificial reproduction, manipulation with gametes and fry rearing was carried out in the experimental hall of the Research Institute of Fish Culture and Hydrobiology, Facutly of Fisheries and Protection of Waters (hereinafter RIFCH FFPW USB) in Vodňany (Fig. 9). Some experiments relating to fry and market size fish rearing took place at farm of company BaHa, Ltd. in Mydlovary (Fig. 10). Most of experiments on artificial reproduction, manipulation with gametes, fry rearing and feeding experiments take place in aquarium roon of the Institute of Aquaculture, Faculty of Fisheries and Protection of Waters (hereinafter IA FFPW USB) in České Budějovice (Fig. 11 and 28).



Fig. 9. Experimental hall of RIFCH FFPW USB in Vodňany. Photo by J. Kouřil



Fig. 10. Rearing hall of farm in Mydlovary (currently, rented by company BaHa, Ltd.). Photo by J. Kouřil

4. DESCRIPTION OF TECHNOLOGY

4.1. Rearing of broodstock

4.1.1. Technological procedure

Broodstock were reared in two gray fiberglass tanks of square shape with rounded corners (volume: 500 litres, depth: 0.5 metres) connected in one recirculation aquaculture system where mechanical and biological purification of water was ensured by combined cellular filter from company Alcedor, Ltd. in Zliv. Tanks were covered with massive self-supporting plastic lid which covered two thirds of the surface. Remaining third was covered by a plexiglass lid with hinges. This part was burdened by few kilograms things, so that the reservoir was sufficiently secured against an escape of fish (Fig. 11).



Fig. 11. Recirculation aquaculture system for rearing of African sharptooth catfish broodstock (aquarium room IA FFPW USB). Photo by B. Drozd

Both sexes (females and males) of individual weight 1.0–5.5 kg were reared together at stocking density of 50–150 kg.tank⁻¹ (125–375 kg.m⁻³) and fed mainly by food EFICO Alpha 717 6.0 (firm BioMar, Denmark). As supplementary food CatCo GROWER – 12 EF, CatCo GROWER – 13 EF, CatCo SELECT – 13 EF (firm Coppens, Netherlands) and possibly Harcsa- és Pisztráng nevelőtáp (firm Haltáp, Hungary) were used. Broodstock were fed in two or three daily doses. At the serving of food, fish usually respond by the violent reaction, so the fast covering of the tank is necessary to prevent water loss.

4.1.2. Results

Based on fish feeding activity, using the daily feed ration was verified in the range of 0.5–0.1% of the current biomass of fish. Before any manipulation with broodstock (even only at catching of fish), cleaning of tank etc. feeding of fish had to be decreased for one to two days or completely omitted at the first day. In case of insufficient coverage of rearing tanks, fish can escape at any time (especially in the morning and at distraction of fish). Therefore, tanks have to be covered and burdened by heavy things. Before harvesting of fish, it is necessary to reduce the water level in well covered tank to a minimum. Then, it is possible to catch the fish. Otherwise, there is a risk of jumping out of the tank or possible injury.

Mortality of broodstock is exceptional. In most cases related to injuries caused by their jumping and falling, or in connection with the manipulation during artificial spawning, sorting etc. Individual annual weight gain of broodstock is about 1–2 kg. The largest broodstock (females) reached individual weight of 4–5.5 kg at the age of five years.

4.2. Hormonally induced artificial reproduction

4.2.1. Technological procedure

Artificial spawning of females was primarily focused on verification of using combined hormonal preparation Ovopel (firm Agrofish, Hungary) at different temperatures with aim to determine the dependence of latency interval on water temperature. Broodstock from own breeding (see Chapter 4.1.) were selected after their capture from tank. Before harvesting of broodstock, fish were not fed for one day. Females of 1–4 kg at the age of 1–3 years were artificially spawned. Selected females were separately placed into thermoboxes (volume: 20 liters) with aeration and soft inner wall preventing injury of fish. After a few hours, water temperature was modified to desired values (19.1–31.5 °C). Simultaneosly, three females were prepared for each individual artificial spawning at all temperatures. Water temperature in thermoboxes was recorded each four hours. Any temperature deviations were modified by exchanging small amounts of water (thanks to good isulating properties of thermoboxes the temperature deviations were minimal). Continuously, the thermoboxes with females were covered during all experiment. And their covers were prevented to potential attempts to escape by jumping of fish. From several hours to one-day adaptation to the desired temperature (in dependence on temperature difference from the temperature at which broodstock was preciously bred), females were injected by hormonal preparation.

Injection was performed in anaesthetized females. Fish anesthesia (Fig. 12) was induced by clove oil (Eugenol preparation, company Dr. Kulich Pharma Ltd., Hradec Králové) at a concentration of 0.06–0.10 ml.l⁻¹ of water, an exposure time corresponded to level of anesthesia 2b (Hamáčková *et al.*, 2003). Temperature significantly affects the anesthesia process (higher temperature lead to shortening of this interval) and the variability of individual susceptibility of fish to the used anesthetics. At a temperature of 23–25 °C, level of anesthesia 2b occurs after 2–5 min.

Artificial stimulation of egg ovulation in African sharptooth catfish females was induced by hormonal product Ovopel (company AgroFish, Hungary). This product is supplied in pressed white pellets. Each pellet contains two active ingredients: 20 µg synthetic GnRHa and 2 mg of dopamine inhibitor – metoclopramide. Recommended dose (Horvath *et al.*, 1997; Kouřil *et al.*, 2011) is one pellet per 1 kg of female weight for all species for which this prepatate is adequate. Pellets of hormonal preparation Ovopel were stored in a plastic bottle with waterproof lid in darkness at room temperature.



Fig. **12.** Anaesthesia (A) and manipulation with anaesthetized female (B) of African sharptooth catfish. Photo by J. Kouřil

Before female injection, the required number of pellets was collected. Then, pellets were crushed with a pestle in a dry mortar. Ovopel pellets are harder than the carp pituitary which is homogenized in similar way. Therefore, greater power is necessary for crush the pellets. Simultaneously, the mortar have to be covered (e.g. by food foil) to avoid loss of material during the crushing. After that, physiological solution, which was stored in its original packaging (sterile packaged) in a refrigerator, was added to crushed pellets in a quantity corresponding to the dose and concentration required for stimulation of fish (Fig. 13). Homogenizing of the mixture was carried out in a mortar

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with a pestle. Generally, recommended dose 1 pellet per 1 kg of female weight (at dilution: 1 pellet per 0.5 ml of physiological solution) was used. Immediately after that, injection of females was performed. However, if preparation of hormonal suspension carried out a few hours in advance, suspension was stored in a glass breaker (covered with aluminium foil) in a refrigerator at +5 °C. Suspension of homogenized hormonal preparation was sucked into a disposable syringe in required volume (with respect to weight of female) separately for each individual fish. Then, mortar and pestle were properly washed by hot water and dried, ready for further use.





Top row (left to right): beaker, graduated cylinder, physiological solution (original packaging), middle row: pestle and mortar with pellets of hormonal preparation Ovopel; bottom row: syringes with needles (diameter 0.6 mm).

Subsequently, females were taken out from the anesthetic and intramuscular injection was carried out (Fig. 14). Then, the fish were washed by water to remove residual anesthetic from their surface and they were give back into thermoboxes. About 2 h before expected ovulation, females were visually or by hand palpation controlled in half-hourly intervals. If the first eggs where found out on walls or at the bottom of thermoboxes, the artificial spawning had been immediately performed

(comparison of female just before and after the artificial spawning – see Fig. 15). Time to ovulation or time of the artificial spawning, was recorded with an accuracy of fifteen minutes. Eggs were spawned (Fig. 16) into a dry container of pre-known weight and marking. Each female was hand-stripped into a separate container (Fig. 17).



Fig. 14. Injection into the dorsal muscle in African sharptooth catfish female. Photo by B. Drozd



Fig. 15. Ventrolateral view of African sharptooth catfish female just before the artificial spawning (A; C – detail of lateral part) and after spawning (B; D – detail of lateral part). Photo by B. Drozd



Fig. 16. Artificial spawning of African sharptooth catfish female. Photo by J. Matoušek



Fig. 17. Freshly spawned eggs of African sharptooth catfish have greenish-yellow to greenish-brown colour. Photo by Foto B. Drozd

Abdominal area or other body parts of females were treated with a weak solution of hypermanganate (potassium permanganate concentration of: 3 grains per 500 ml of water). Then, females were given back into thermoboxes. Water exchange was performed twice a day with gradual transition back to the water temperature in broodstock rearing tank. Due to control of health condition of females, as well to prevent aggressiveness of just spawned fish in the broodstock tank, artificially spawned females were placed back into the broodstock tank after 2–3 days.

Sperm (milt) for egg fertilization was obtained from just killed males using the procedure introduced by Hamáčková et al. (2007). Usually, eggs were fertilized by a mixture of sperm from two males. Males were caught from the tank a few hours before planned artificial spawning of females. They were placed separately into covered boxes. Just before the estimated time of ovulation and consequent artificial spawning of females, the males were taken out from the water. After their killing, the gonads (testes) were removed using surgical stainless steel scissors so as to avoid contamination of the gonads with water or blood (Fig. 18). Thereafter, the gonads (Fig. 19) were dried on a filter paper and pput on a square of a dry inert technical textile (net with mesh size 0.5–1.0 mm) with an area of 25 x 25 cm. Male gonads were cutted to about 1 cm pieces with using of dry stainless scissors (Fig. 20A). Textile was held above the glass or plastic container, in which the sperm was dripped. After that, the textile with cutted male gonads was gently pressed by fingers from the outside and another portion of seminal fluid containing sperm dripped was obtained (Fig. 20B). The sperm was temporarily stored in a covered dish to prevent the contamination of water in the refrigerator +5 °C. During the next 10 min to 1 h sperm was used for insemination of eggs.



Fig. 18. Dissection of gonads (testes; marked by a star) from the body cavity of African sharptooth catfish males. Photo by J. Matoušek



Fig. 19. Dissected gonads (testes) of African sharptooth catfish. Photo by B. Drozd



Fig. 20. Obtaining of the sperm for egg fertilization by cutting (A) and pushing through an inert technical textile (B) of dissected gonads of African sharptooth catfish male. Photo by J. Matoušek

Egg insemination and fertilization (Fig. 21) was performed according to procedure presented by Adamek (2001) and Hamáčková et al. (2007). For short term, spawned eggs were kept in containers covered by wet, properly wrung out textile which did not touched the eggs. Containers with unfertilized eggs were placed on the floor (temperature of eggs was about 20 °C). Before insemination, eggs were stored for 10 min. to 1 hour. Before egg insemination, eggs were divided into 200–300 g portions into separate containers. Egg fertilization was carried out with 2-5 ml of sperm (Fig. 21A). Then, the mixture of gametes were slightly mixed with dry plastic or rubber spatula. Followed by fertilization of eggs, which was performed by pouring of water till the gamete mixture was completely under water and above their surface was about 0.5–1 cm layer of water (Fig. 21B). The resulting mixture was stirred approximately 1 min. Then, the container with sex products and water was left for 2 min at rest. In the next 2-4 minutes, the fertilized eggs were washed with water. By repeatedly mixing and pouring of water on the eggs, the elimination of sperm residues or infrequantly occurring white (fertilization unable) eggs was implemented. After that, the external adhesive layer of eggs was activated. Therefore, the eggs were immediately poured on incubation sieves which were submerged under water and well attached to the wall of the plastic trays. This activity was carried out as quickly as possible and in such a way that the eggs were well spread over the whole surface of the textile to prevent their greater accumulation and subsequent formation of lumps and clumps of aggregated eggs stuck together. Technival (inert) textile Uhelon of 0.5 mm mesh size proved to be the best for production of incubation sieves.



Fig. 21. Egg insemination by sperm (A) and egg fertilization (B) of African sharptooth catfish. Photo by J. Matoušek

Incubation trough (Fig. 22), or several throughs, was/ were part of a separate recirculation system with a plastic reservoir tank under it/ them. From incubation tanks/ s, the water gravitationally spilled down into the reservoir tank/ s and then the water was pumped up into a plastic pipe using submerged pump. Aquarium UV lamp proved to be useful part of pressure pipe/ s that broutgh the water to the incubation trough (troughs). Perfect purity of all used components was necessary for the prevention of any water contamination during egg incubation.

4.2.2. Results

Generally, 40 females were injected during 10 separate experiments ehich were carried out at different water temperatures in range of 19.1–31.5 °C. Ovulation was induced and artificial spawning was performed in a total of 39 females (achived success of 97.5% – see Tab. 4).

Tab. 4. Length of latency interval and number of injected and ovulated (artificially spawned) females of African sharptooth catfish at different temperatures in the range of 19.1-31.5 °C.

Temperature (°C)	Length of latency	Number of fish				
	interval (h)	injected	ovulated			
19.1	26.75	4	4			
19.5	26.75	4	4			
21.4	19.00	4	4			
21.5	18.75	4	4			
22.5	17.25	4	4			
23.7	15.75	4	3			
25.4	14.00	4	4			
27.0	11.75	4	4			
29.7	9.25	4	4			
31.5	7.00	4	4			

Comparison between lengths of latency interval after using of Ovopel and carp pituitary preparation in dependence on water temperature 18–32 °C is recorded in Tab. 5 (for use in hatchery practice). Latency interval length induced by Ovopel was observed by authors of this technology. Data from Tab. 4 were adjusted by

extrapolation and interpolation with subsequent rounding of numbers. Data with using of carp pituitary extract were taken from Adamek (2001).

Tab. 5. Dependence of latency interval length (h) on water temperature at hormonal induction of ovulation in African sharptooth catfish females using a single injection of Ovopel (data observed by authors of this technology) in comparison with a single injection of carp pituitary extract – CPE (results published by Adamek, 2001).

Temperature °C	: 18	19	20	21	22	23	24	25	26	27	28	29	30	31	32
Ovopel (h)	-	27	23	20	18	16	15	14	13	12	11	10	9	8	7
CPE (h)	21	-	-	19	18	15	13	12	11	10	9	8	7,5	7	-



Fig. 22. Trough for incubation of eggs and newly hatched fry of African sharptooth catfish. Photo by J. Kouřil. A: Trough with yolk-feeding larvae; B: Inlet part of the trough with externally fed larvae (overshadowing of tank).

4.3. Temperature effect on storage of artificially spawned eggs before fertilization

4.3.1. Technological procedure

In total, three females were artificially spawned and their eggs were used for this experiment. After Ovopel injection, females (see Chapter 4.2.) were stored till artificial spawning (during latency period) at water temperature 23.7 °C. From each female, part of spawned eggs (about 200 g) was immediately taken place into environment at different temperature. Containers with eggs were covered with wet, wrung out textile and separately stored in thermoboxes (Fig. 23). There was a temperate (required) water bath at the bottom of thermoboxes. Different temperatures were monitored and maintained (through addition of ice cubes, or a little amount of warm water) at 5, 10, 15, 20, 25 and 30 °C (in compliance with maximum deviations of 0.1 °C from desired value) in thermoboxes. Gradually, samples of eggs stored at various temperatures were removed with a dry plastic spoon after 1/4, 1/2, 1, 2, 4, 6 and 8 h from spawning, respectively. Then, these egg samples (about 100 eggs per one sample; alqays three replicates for each time interval), including a sample collected immediately after the spawning, were placed into dry glass container temperated to 25 °C. Furthermore, eggs were inseminated with sperm mixture from two males and after that they were fertilized by pouring of water at identical temperature (25 °C). Subsequently, the mixture of eggs and sperm was stired with plastic spoon so that unsticked eggs spread over the surface of container. After 10 min from fertilization, water from container with fertilized eggs was repeatedly decanted and poured. Eggs were washed with clean water to eliminate the remnants of sperm and ovarian fluid. Containers with fertilized eggs were placed together on table at air temperature of 25 °C (Fig. 24). During egg incubation, water was twice exchanged in containers. After 12 hours, fertilized (developing) and white (unfertilized, or not developing) eggs were counted. Based on these data, fertilization rate of eggs was determined in %.



Fig. 23. Storage of artificially spawned eggs of African sharptooth catfish in thermobox. Photo by J. Kouřil

4.3.2. Results

During experiment, significant effect of different temperatures on length of fertilization capability was found out in African sharptooth catfish. In control groups (eggs fertilized immediately after spawning), average fertilization rate about 90% was achieved. For storage of just spawned unfertilized eggs of African sharptooth catfish, temperature range between 15 and 20 °C can be considered (see Tab. 6). After 6 hours from spawning, the fertilization rate of eggs still reached about 75% and more (*note: this fertilization rate was chose by authors as arbitrary limit for spawning which can be regarded as successful from hatchery point of view). At lower temperatures, the fertilization ability decreases significantly faster. Already after 1.5 hours, the drop of egg fertilization below 75% was observed at 10 °C. After 0.5 hour, the fertilization rate decreased after mentioned limit at 5 °C. On the contrary, the less decline of fertilization ability was found out at higher temperatures. After 4 h, the fertilization rate of eggs was above 75% at 25 °C. After 2 hours, the mentioned arbitrary limit of fertilization rate was reached also at temperature of 30 °C. Based on the results, temperatures of 15-20 °C (or even 25 °C) can be recommended for storage of spawned unfertilized (uncontaminated with water) eggs of African sharptooth catfish up to 4 h (at temperature of 15 and 20 °C up to 6 h). At these temperatures and time intervals, no significant reduction (decrease below 75%) of fertilization ability was observed.

Temperature (°C) Length of egg storage (h)										
	0.5	1	1.5	2	3	4	6	8		
5	72	68	65	48	51	16	24	9		
10	79	85	68	71	63	54	43	6		
15	82	85	77	78	77	77	78	37		
20	90	84	85	77	90	81	75	29		
25	84	91	84	84	75	76	68	19		
30	83	84	82	79	53	33	1	0		

Tab. 6. Temperature effect (°C) and storage length (h) of unfertilized eggs of African sharptooth catfish without water contamination on their fertilization ability (%).

* **Bold and italic** are the combinations of temperature and storage length, in which the egg fertilization rate of 75% and more was reached.


Fig. 24. Incubation of African sharptooth catfish eggs in glass containers during experiments focused on study of environment temperature effect on storage of artificially spawned eggs before insemination and length of water contamination effect on rate of micropyle closure. Photo by J. Kouřil

4.4. Effect of length of water contamination of eggs on rate of micropyle closure

4.4.1. Technological procedure

Generally, two indicative experiments on indirect determination of time interval of unfertilized egg micropyle closure after contact with water were carried out. Artificially spawned eggs from three or four females were used during first and second experiment, respectively. After Ovopel injection (see Chapter 4.2.), females were kept at 23-24 °C till the spawning (during latency period). Always, amount about 100 eggs (from each female) was placed into every marked glass container using a plastic spoon. Overall 20 experimenal variants were realized (see below) and always one control with three containers was used for each variant during all experiments. Immediately after spawning, eggs were inseminated and then activated by water (in close time interval) in all control groups. Water was added to eggs in containers at 30 second intervals. Subsequently, mixture of fresh sperm (from two males) was added to each container using a syringe at 1-minute intervals. Immediately after addition of sperm, the mixture of gametes and water was gently mixed. This procedure was used in each of experiments. In total, 20 variants with different water contamination length (0.5-10 min. before sperm addition) of eggs were performed. Like in the experiment mentioned in Chapter 4.3., water from every container was repeatedly decanted and poured after 10 min. from sperm addition. Always, the eggs were washed by clean water to eliminate sperm and ovarian fluid remnants. Similarly, fertilization rate of eggs was determined after 12 h from beginning of the experiment. During egg incubation (Fig. 24), all containers were placed on a table in room at air temperature 25 °C. During incubation, water was exchanged twice. After 12 h from hatching, egg hatchability was evaluated in %.

4.4.2. Results

In the control groups (sperm was added to eggs earlier than water), egg fertilization fluctuated in repetitions (n = 7) between 43–95% (75.13 \pm 20.21%; mean \pm S.D.) and fry hatchability ranged 20–71% (47.00 \pm 19.93%). During water contamination of eggs (at indirect micropyle closure after contact with water), very rapid reductions of egg fertilization rate and subsequent hatching rate were observed with increasing length of presence of eggs in water (see Fig. 25). Already after 1 min. from water contamination, egg fertilization rate and hatching rate decreased to half. After 2 min. from water contamination, both parameters decreased to a quarter of values that were recorded in the control group. After 3 min. from water contamination, the fertilization

rate decreased below 10%. In connection with this, also hatching rate decreased. From 2.5–3 min. (and more), no hatchilings were recorded (hatching rate dropped to 0%).

Based on the indicative experiment, it can be concluded that water contamination of African sharptooth catfish eggs (e.g. at inadequately professionally controlled artificial spawning) has adverse effect on egg fertilization and hatching rate. This effect increases with increasing length of presence of eggs in the water without sperm contamination. This effect can be prevented thorough a perfect protection from water contamination of eggs (e.g. well dried abdominal area of female and safe storage of spawned eggs – covering with curefully wrung out textile). Neverthelles, if water contamination will occur during or after the spawning, this problem can be eliminated with immediate egg insemination (to 1 min. from contact of eggs with water) by sperm and subsequent activation with water (sperm and water have to be prepared in advance), but without a guarantee of sure success. Risk of water contamination of eggs may be reduced by separate spawning of eggs from each female into different dry containers.



Fig. 25. Fertilization and hatching rate (mean ± S. D.; %) of African sharptooth catfish eggs in dependence on time interval length of water contamination of unfertilized eggs till their fertilization (addition of sperm). □ Fertilization rate ■ Hatching rate

4.5. Effect of temperature and egg incubation on hatching and early development

4.5.1. Technological procedure

Series of experiments was aimed on effect of water temperature on ontogenetic development: length of incubation period, hatching period, onset of intake of external feeding, size and survival of African catfish larvae. Experiments were carried out under laboratory conditions in non-flow aquariums and glass containers placed in temperate baths where the temperature was continuously monitored and regulated using thermostats and electric heaters (range: 19–33 °C, tolerated deviation: maximum 0.1 °C from required temperature.

4.5.2. Results

It was observed that the length of incubation period (interval from egg fertilization to hatching) and hatching period (interval from start of hatching till the end of hatching) is dependent on water temperature. Both parameters decreased with increasing water temperature (see Tab. 7). At optimal temperatures (19–31 °C), length of incubation and hatching period was 19–39 h and 3–5 h, respectively.

Size (total body length and wet weight) of freshly hatched individuals called larvae (in interval from hatching till full yolk absorption) was dependent on water temperature range 23–33.5 °C. Size and developmental stage of larvae decreased with increasing water temperature, because just hatched individuals which were incubated at higher temperatures reached lower size and ontogenetic stage, than the fish hatched at lower temperatures. Freshly hatched larvae of African sharptooth catfish reared at optimal temperature conditions (23–30 °C) were 4–5 mm long and weighed 1.2–1.6 mg. Yolk volume of just hatched larvae correlated with size of spawned eggs (yolk volume of hatched individuals increased with rising egg size). At optimum water temperatures (23–30 °C), yolk volume achieved 0.8–2 μ l.

Tab. 7. Length of incubation period (interval from fertilization till hatching; indicated for moment of						
hatching of 50% of individuals; h) and hatching period (interval from beginning till the end of						
hatching; h) in dependence on water temperature (°C) in African sharptooth catfish.						

Water temperature (°C)	19	21	23	25	27	29	31	33
Length of incubation period (h)	70	48	39	29	24	20	18	16
Length of hatching period (h)	2	6	5	4	3.8	3	2.8	1.6

Generally, African sharptooth catfish is characterized by highly variable and often quite low fertilization rate and subsequent hatching rate (i.e. a number of surviving individuals at the time of hatching). Within optimum temperature range (23–30 °C), the hatching rate reach to 95%. However, significantly lower values could be achieved in dependence on spawning conditions, broodstock quality and professional experience. On average, hatching rate about 50–70% can be achieved, but values about 25% are not exceptional (see Tab. 8).

Tab. 8. Hatching rate (% of hatched larvae from the total number of controlled fertilized eggs) in dependence on water temperature (°C) in African sharptooth catfish.

Data in brackets represent the minimum and maximum average values achieved during different spawnings in 2009–2011.

Water temperature (°C)	Hatching rate (%)
18	0
19	29
21	75 (58; 91)
23	73
24	81 (70; 95)
25	78 (25; 88)
27	64 (40; 77)
29	35 (34; 37)
30	28 (25; 42)
31	14 (13; 21)
33	11 (10; 18)
35	0

Length of endogenous feeding period, i.e. time from hatching till the onset of external food intake (mixed feeding period), was dependent on incubation temperature. It decreased with increasing water temperature (see Tab. 9). Under optimal temperature conditions (23–30 °C), larvae of African sharptooth catfish had started to intake external food after about 40–80 h from hatching at size of 6–8 mm and wet weight 2–4.5 mg.

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Tab. 9. Length of endogenous feeding period (i.e. from hatching till the onset of intake of external food; mean, h) in dependence on water temperature (°C) in African sharptooth catfish.

	,	, ,		,			
Water temperature (°C)	21	23	25	27	29	31	
Length of endogenous feeding period (h)	93	77	65	50	43	34	

Length of yolk absorption period (i.e. from hatching till the full yolk absorption when the larvae completely resorbed their yolk reserves and they started to use only external food as the only energy source), was dependent on water temperature. This parameter decreased with increasing water temperature. Under optimum temperatures (23–30 °C) and feeding conditions (*ad libitum* feeding), full yolk absorption occured. The transition to only external feeding averaged about 10–26 days from hatching (see Tab. 10). If the larvae were not fed with external food, a depletion of yolk reserves occured after 2.1–4.6 days from hatching (*these data serve as information to select the suitable time for the beginning of brine shrimp nauplii or starter feed application*).

Tab. 10. Length of period from hatching till full yolk absorption (mean, days) in dependence on water temperature (°C) in fed and unfed larvae of African sharptooth catfish.

Water temperature (°C)	21	23	25	27	29	31
Length of yolk absorption period in <i>fed</i> larvae (days)	33.8	25.8	20.4	15.4	12.5	9.0
Length of yolk absorption period in <i>unfed</i> larvae (days)	6.8	4.6	4.0	2.7	2.2	2.0

After full yolk absorption, size of fed larvae (total body length and wet weight), which were kept without external food, did not differ statistically in thermal range of 23–30 °C (it was not dependent on water temperature). At optimum water temperatures (23–30 °C), fed larvae were 6.5–7.5 mm long and weighed 2–3 mg. However, size of fed larvae significantly varied in dependence on water temperature after full yolk absorption. In this case, size exhibited inversely proportional dependence on incubation temperature (it decreased with increasing temperature). Larvae reared at optimal temperature conditions were 10–20 mm long (about doubled in comparison with experiments without external feeding) and weighed 15–50 mg.

Size of fed larvae (total body length and wet weight) was temperature dependent after full transition to exogenous feeding (yolk was fully absorbed). Larval body length and weight decreased with increasing water temperature and averaged between 10–20 mm and 15–50 mg, respectively (*body length of unfed larvae reached 6.5–7.5 mm and weighed 2–3 mg*).

Survival rate after hatching (interval from hatching till the full yolk absorption) ranged up to 75% at optimal temperatures, but on average it was 50% (see Tab. 11).

Tab. 11. Average survivale rate after hatching (interval from hatching till the transition to exogenous feeding/ full yolk absorption; %) in dependence on water temperature (°C) in African sharptooth catfish.

Water te	mperature (°C)	Survival rate (%)
21		16
23		48
25		48
27		51
29		47
30		52
33		12

Fry should be reared at water thermal range of 27–30 °C in shallow flow-water throughs with brine shrimp nauplii - *Artemia salina* (Linnaeus, 1758) initial feeding with subsequent transition (after several days) to starter feeding (Fig. 26 and 27). Condition of rearing success is closely related to high purity of environment and good water quality.



Fig. 26. Rearing of African sharptooth catfish fry. Photo by J. Kouřil



Fig. 27. Detail of African sharptooth catfish fry at external feeding by starter food. Photo by B. Drozd

4.6. Testing of feed for breeding fish in market size

4.6.1. Technological procedure

Generally, three consecutive feeding tests were carried out in African sharptooth catfish in market size. Overall, 10 different commercially produced feeds were tested as potentially applicable for African sharptooth catfish. Some of them were directly produced for catfishes (African catfish). Several feeds were tested only once, others repeatedly.

Fish fry on average weight 200–300 g were used in feding tests. Fry was reared in same type of tanks and fed with EFICO Alpha 714 feeding (company BioMar, Denmark) before the experiments. Each test started with three-week adaptation period. At the beginning of test, tanks were harvested and current stocking densities were joined in one reservoir tank and then same numbers of fish were given into all experimental tanks under following principles: individual weight (with accuracy of 1 g), sex determination (half-representation of both sex in each tank), well developed fish without cannibalism or damage, relatively low size variability (less than 1% of the

average biomass) and about the same total weight (biomass) of fish stocks. In the first three-week period, the fish were getting used to the tested food which they received during experimental period. Daily food ration (DFR) corresponded to this adaptation. First day after stocking, the fish were fed with one guarter of DFR. Daily food ration was gradually increased recommended amount of DFR. In dependence on feed, the adaptation lasted from a few days to 1-2 weeks. Then, the fish began to feed in required quantity (full amount of DFR) with respect to the average individual weight and current stocking biomass. Daily food rations were divided into six daily doses (at 8, 10, 12, 15, 18 and 20 h). Relative amount of DFR (in relation to estimated current biomass) was determined in advance, according to the recommended DFR. Alternatively, DFR was corrected according to eaten amount of feeding in the previous day. There was also an effort to follow the same DFR for all experimental groups. However, if apparent differences were observed in acceptance of various feeds, the principles have had to be gradually changed. For these reasons, relative DFRs were slightly diversified. Current DFR was calculated according to actual stocking biomass. On the following day, relative DFR was determined (in % stocking biomass) and accordingly the absolute DFR was calculated (in grams) for each tank. If the full amount of DFR was eaten by fish, the weight of feed have been added to the original weight of biomass and then the theoretical stocking biomass have been calculated for next day. In the case that DFR was not eaten by fish during one day, the weight of remaining feed was deducted from DFR and only the really amount of eaten feed was recorded. Rarely, the increasing of DFR was performed when the fish exhibited a higher willingness to accept the feed. Usually, it occurred after the day when incomplete DFR was served to fish for any reason. In that case, determination of absolute DFR was carried out in opposite way. Determined DFR had to be increased by the weight of unconsumed feed more. This procedure was followed throughout all 20 feeding days. Following day (21st experimental day), harvesting of stocks, individual weight and sex determination of all fish were performed. Fish were re-stocked to the experimental tanks. Before and after the harvesting day, fish were not fed.

After twenty-day feeding period, weight change of fish biomass was found in all stocks (the difference between stocked and harvested fish weight in grams, called also absolute weight gain). From this value and average between stocked and harvested biomass, the relative daily gain of biomass (in %.d⁻¹). Also, a total absolute weight of really eaten feeds was determined after twenty-day feeding period. From the absolute gain of fish biomass and real feed consumption, the food conversion ratio (FCR) was calculated. It express the feed consumption per unit of weight gain, e.g. kilograms.

Fromula for calculating of FCR: **FCR** = $(W_t - W_o)$

- **F** feed consumption during tested period
- W_t stocking weight at the end of experiment
- Wo stocking weight at the beginning of experiment

Real relative DFR was calculated from average stocked biomass, harvested biomass and absolute DFR. Real relative DFR was used for determination of initial relative DFR for following sub-feeding period. Based on price, absolute weight of eaten feed and absolute weight gain of fish biomass, costs of one kilogram weight gain of biomass were calculated in selected perspective feeds. Cost determination was carried out summarily for the entire rearing period (for period of several partial twenty-day tests). Feed prices used in the cost calculation are based on retail prices for small farmers (i.e. customers with consumption of 20-25 kilograms bags of feed per month.

The first feeding experiment took place in aquariums placed in two-decker metal stand (Fig. 28A). In recirculation system, the continuous water flow and outlow was maintained in aquariums. Recirculation system was composed of sedimentation tank (placed on the floor under stand) and upper tank with submerged biological filter (placed at the top, above experimental aquariums). Aeration was installed into the aquariums. Water flow ensured water exchange about once every 1–2 hours. Aquarium walls and glass were cleaned with foam sponge in every day. Sludge was extracted twice during rearing and once at fish harvesting. Sedimentation tank and biological filter were purified as needed, they were cleaned more frequently at the end of rearing in relation to increasing biomass and pollution.

Next two feeding experiments took place in cylindrical tanks from white plastic material with usable volume of 315 l (Fig. 28B), upper inlet and lower outlet of water. Water outlet flowed into sedimentation tank, then the water was pumped about two meters above into shallow supply tank, from where it uniformly flows into the trickle biological filter. Under biological filter, the water was trapped into the collecting tank and then gravitationally flowed through the pipe into other tanks. Aeration was installed into each tank. Water flow ensured exchange of water about once per every 3–4 hours. Each week, tank walls were cleaned using foam sponge (twice a rearing and once a fish harvesting).Sedimentation tank and biological filter were purified as needed, they were cleaned more frequently at the end of rearing in relation to increasing biomass and pollution.



Fig. 28. Interior of aquarium room of the Institute of Aquaculture FFPW USB in České Budějovice: experimental aquariums (A) and circular tanks (B) in recirculating aquaculture systems with course of feeding tests in African sharptooth catfish. Photo by J. Kouřil

Both systems were daily refilled by warm tap water. Water temperature in recirculating aquaculture systems was maintained thanks to sufficient heating of aquarium room. If necessary, desired water temperature was achieved using electrical heaters. African sharptooth catfishes were reared to market size (a total individual body weight of 800–1 500 g).

In the first experiment, in total six different feeds (in three repetitions) were tested in 18 aquariums. Five salmonid feeds – Aqua Focus (Aller, Poland), EFICO Alpha 714 (BioMar, Denmark), Skretting F-2P B40 (Skretting, Norway), Troco Supreme-22 a Troco Prime-18 (Coppens, Netherlands) and one feed for trouts and African sharptooth catfishes – Harcsa-és Pisztráng nevelőtáp (Haltáp, Hungary) were tested. Specification of individual tested feeds are listed in Tab. 12. During the first experiment, one adaptation and four experimental feeding periods were realized. Total length of experimental feeding test lasted about 84 days. Average water temperature was 26.0 °C.

Feed Haltáp	Aqua Focus <i>A</i>	EFICO Alpha 714	Skretting F-2P B40 S	Troco Supreme-2	Troco 22 Prime-	18
Granule size (mm)	4.5	4.5	4	4.5	4.5	5
Protein content (%)	37	42–46	41	44	42	48
Fat content (%)	12	13–16	12	22	18	6.4
Ash content (%)	7	6.4	6.5–8	7.1	7.1	
Fiber content (%)	4	6	2.5–3	1.8	2.9	1.8
N content in dry matter (%)	6.5					
P content in dry matter (%)	1.2	1	0.85–1.4	0.9	1	1.3
Ca content in dry matter (%)		0.86		1.3	1	1.4
Na content in dry matter (%)				0.3	0.2	0,3
Mn content in dry matter (mg.kg ⁻¹))	30				
Cu content in dry matter (mg.kg ⁻¹)			6			
Amino acid content (%)			2.5			6
Vitamin A (IU.kg ⁻¹)	2 500		5 000	10 000	10 000	1 400
Vitamin D3 (IU.kg ⁻¹)	500			3 000	2 000	140
Vitamin E (mg.kg ⁻¹)	150		150	200	150	70
Gross energy (ths kJ.g-1)	19.5	20–22		22.4	21.4	
Digestible energy (ths kJ.g-1)	15.3	15.5	17.6	20	19.2	
Price (CZK.kg ⁻¹)	31	35	37	42	45	32

Tab. 12. Specifications of tested feeds used during the first feeding experiment in African sharptooth catfish (feed from Haltáp company was used also during the third feeding experiment).

* IU means international units

In the second experiment, in total four feeds were tested in 12 tanks (each feed in three repetitions). Three feeds for African sharptooth catfish keříčkovce – CatCo GROWER – 12 EF, CatCo GROWER – 13 EF and CatCo SELECT – 13 EF (Coppens, Netherlands) and one salmonid feed – Dibaq Trout Evolution (Dibaq, Spain) were tested. Specifications of tested feeds are listed in Tab. 13. During the second experiment, one adaptation and four feeding periods were carried out. Length of experimental feeding test lasted 84 days. Average water temperature was 26.0 °C.

Feeds	CatCo gROWER-12	CatCo 2 EF gROWER-1	CatCo 3 EF SEIECt-13	Dibaq trout EF Evolution
Granule size (mm)	4.5	4.5	4.5	5
Protein content (%)	45	42	42	38–40
Fat content (%)	12	13	13	24
Ash content (%)	8.7	7.4	8.5	8.5
Fiber content (%)	1.9	2.7	1.9	1.8–2.2
P content in dry matter (%)	1.1	1	1.1	0.85
Cu content in dry matter (mg.kg ⁻¹)				7
Vitamin A (IU.kg ⁻¹)	10 000	10 000	10 000	7 500
Vitamin D3 (IU.kg ⁻¹)	2 000	2 000	2 000	1 000
Vitamin E (mg.kg ⁻¹)	200	200	200	150
Gross energy (kJ.g-1)	19.9	20.2	20.0	
Digestible energy (kJ.g ⁻¹)				
Price (CZK.kg ⁻¹)	18.1 51	18.1 51	18.1 51	48

Tab. 13. Specifications of tested feeds used during the second feeding experiment in African sharptooth catfish (feeds CatCo GROWER – 12 EF, CatCo SELECT – 13 EF were used also during the third feeding experiment).

* IU means international units

In the third experiment, three different feeds were tested in nine tanks (each feed in three repetitions). They were special feeds produced for rearing of catfishes. They were tested already in two previous experiments – CatCo GROWER – 12 EF (Fig. 29A), CatCo SELECT – 13 EF (Fig. 29B; Coppens, Netherlands) and Harcsa-és Pisztráng nevelőtáp (Haltáp, Hungary; Fig. 29C). During the third experiment one adaptation and

three feeding periods were realized. Total length of feeding experiment was 63 days. Average temperature was 24.5 $^{\circ}\mathrm{C}.$



Fig. 29. Special tested feeds for catfishes were from Durch company Coopens: CatCo GROWER – 12 EF (A), CatCo SELECT – 13 EF (B) and Hungarian company Haltáp: Harcsa- és Pisztráng nevelőtáp (C) at production of African sharptooth catfish in market size during third feeding experiment. Photo by O. Houda

4.6.2. Results

In the first experiment, the best feed conversion ratio (FCR) was reached in both feeds intended for salmonids from company Coopens – Troco Supreme-22 (FCR = 1.19) and Troco Prime-18 (FCR = 1.26). The third best feed conversion ratio was observed in feed developed for salmonids and African sharptooth catfish from company Haltáp – Harcsa-és Pisztráng nevelőtáp (FCR = 1.45), followed by a feed for salmonids from company Aller – Aqua Focus (FCR = 1.58) and company Skretting – Skretting F-2P B40 (FCR = 1.74). At the same time, the highest increase of biomass was recorded in feed from company Haltáp. The highest feed conversion ratio was surprisingly achieved in feed EFICO Alpha 714 from company BioMar (FCR = 1.97).

Then, costs of one kilogram weight gain were observed in relation to using of various feeds. The lowest cost was reached in feed from company Haltáp (45 CZK.kg⁻¹), followed by feeds Aqua Focus (46 CZK.kg⁻¹) and TROCO SUPREME-22 (50 CZK.kg⁻¹). In other feeds the costs significantly exceeded 50 CZK.kg⁻¹, which can be taken as arbitrary limit of profitability of African sharptooth catfish rearing in RAS under small-farm conditions. Costs for other feeds were: Troco Prime-18 (56 CZK.kg⁻¹), Skretting F-2P B40 (57 CZK.kg⁻¹) and EFICO Alpha 714 (66 CZK.kg⁻¹).

In the second feeding experiment, the lowest feed conversion ratio achieved in Dutch feed developed for African catfish rearing from company Coppens – CatCo SELECT-13 EF (FCR = 0.85). Simultaneously, this feed produced the highest specific growth rate (1.30%). This led also to the lowest cost per kilogram of weight gain (43 CZK. kg⁻¹).

In the third feeding experiment, the most favourable feed conversion ratio was achieved with using of feed CatCo GROWER-12 EF (FCR = 0.82). Followed by previously proven feed – CatCo SELECT-13 EF (FCR = 0.88). The worst result was observed with using of feed from company Haltáp (FCR = 1.37). In this trial, the costs per one kilogram weight gain were relatively balanced. Despite the high price of feed (51 CZK.kg⁻¹), the lowest cost was observed at feed CatCo SELECT-13 EF (42 CZK.kg⁻¹), due to its prosperous feed conversion ratio. Followed by feeds Haltáp (44 CZK.kg⁻¹) and CatCo GROWER-12 EF (45 CZK.kg⁻¹).

In repeatedly tested special feeds for African sharptooth catfish, quite balanced costs per one kilogram of weight gain were achieved. The best tested feed was CatCo SELECT-13 EF (costs per 1 kg of weight gain were 41 and 42 CZK) which was also the most expensive feed. On the contrary, the worst was the cheapest feed Haltáp (costs per 1 kg of weight gain were 45 and 44 CZK). Also, the feed Haltáp had a less favourable physical properties, as a considerable amount of dust particles and easy disintegrates in water. If the feed ration is immediately consumed by fish, the large feed disintegrating is not critical. Both of these factors can have adverse effect on higher content of suspended solids in water. It induce higher need of capacity of mechanical

filters or sedimentation tanks, and increases a risk of impaired function of biological filters. This need for any intensified exchange of supplementary water requires higher operating costs (higher water and energy consumption for its heating). This state unequivocally favors at African sharptooth catfish rearing in RAS with using of feed CatCo SELECT-13 EF. If this feed will be used under operating conditions in compliance with good feeding technique and less frequent harvesting which disturbs the fish (in comparison to annotated results in the experiments), further potential reduction of feed conversion ratio to 0.7–0.8 and thereby reduction of feeding cost per one kilogram of weight gain to 35–40 CZK can be expected.

4.7. Fillet yield and product quality

4.7.1. Technological procedure

Fillet yield (Fig. 30 and 31) was determined as weight percentage of fillets without skin and without pectoral fins (Fig. 32). During fish processing, all visceral organs, pectoral fins, head, as well as skin and spine (Fig. 33). Next part of experiment was determination of protein and fat contents in dry matter of fillets provided in Laboratory of Genetics, Breeding and Nutrion at Faculty of Agriculture USB (Dipl.-Ing. Jaromír Kadlec, Ph.D.). At sensory evaluation, ten people assessed the consistency, smell, taste and aftertaste in order to determine whether there are any differences among tested feeds, and if their composition also influences the quality (in terms of existence of possible positive or negative taste characteristics) of African sharptooth catfish meat (Fig. 34 and 35).



Fig. 30. Detail gutting (A) and filleting (B) of market size African sharptooth catfish. Photo by J. Kouřil



Fig. 31. Gutting of African sharptooth catfish. Photo by J. Kouřil



Fig. 32. Detail of African sharptooth catfish fillets. Photo by J. Kouřil



Fig. 33. Skinning (A), the spine with dorsal, tail and anal fins (B), cut part of abdominal area with pelvic fins (C), gonads (D), fillets (E) and head with pelvic fins (F) of African sharptooth catfish. Photo by J. Kouřil

4.7.2. Results

Performed basic chemical analysis of fillet composition showed about 17% protein and 6% fat contents in dry matter. The highest average values of fillet yield (43%) were detected in feed CatCo SELECT-13 EF. However, the average value of fillet yield slightly exceeded 40% (in all feeds). It did not differ neither between sexes, nor among tested feeds (no statistically significant difference at a significance level of 5%). In terms of quality of final product, no statistically significant differences were detected in organoleptic muscle (meat) characteristics of African sharptooth catfish fed by various tested feeds (as the tastiest sample CatCo SELECT-13 EF was evaluated by 38% of assessors). It follows that, in terms of product quality, all feeds used in feeding tests are useful and appropriate for African sharptooth catfish.





A: Homogenization of fillets into cubes of approximately 3 x 3 cm;

B: Closed and labelled glass jars with sliced cubes of flesh before cooking.



Fig. 35. Organoleptic assessment of heat treated samples of African sharptooth catfish muscle by assessors. Photo by T. Zajíc

5. ECONOMICAL BENEFIT OF TECHNOLOGY FOR BUSINESS SUBJECT

Technology is designed for specialized farms engaged in fish rearing in recirculation systems with intensive breeding of African sharptooth catfish to efficient production of marketable fish. The purpose of technology is to help to solve some problems with rearing of this fish species. In particular, the use of artificial reproduction and optimisation of utilization of commercially produced feeds that will be convenient for African sharptooth catfish.

User with applying of the technology can expect annual gross profit of 400 thousand CZK. This assumption is based on the usual price of marketable fish (80 CZK.kg⁻¹), feed prices (40 CZK.kg⁻¹), feed conversion ratio (FCR = 0.8) and estimated proportion of feed costs in amount of 50% from total costs. Next costs include the costs for rearing and artificial spawning of broodstock, egg incubation and fry rearing, including starter feeds, energy, water, transport, property depreciation, personnel and insurance costs. In the case of annual production of 25 t of marketable African sharptooth catfish in total price of 2 million CZK, the annual feed consumption in the amount of 20 t can be expected and thus feed costs reach 800 thousand CZK. Total annual costs of rearing can assume 1600 thousand CZK. Estimated annual gross profit (the difference between revenue from sales of marketable fish and total costs) farms with annual production of 25 t of marketable African sharptooth catfish should be 400 thousand CZK.

6. APPLICATION OF TECHNOLOGY IN PRODUCTION OF BUSINESS SUBJECT

Technology summarizes the practical experience and the results of a series of partial experiments focused on the problems of broodstock breeding, methods of artificial reproduction, egg incubation and marketable fish production (including effects of comercially produced feeds on fillet yield and meat quality). The aim of technology is to provide, to specialized fish farms, a sophisticated technological procedure and necessary information for effective intensive farming of African sharptooth catfish to efficient production of marketable fish. This species is very perspective for rearing in recirculating aquaculture systems of various sizes using heated water. The main advantages of its rearing can include short production cycle and high meat quality. African sharptooth catfish breeding allows not only to increase species diversity of fish market, but also to retain or increase market production of Czech aquaculture.

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