CHAPTER 4

SUPPLEMENTARY FEEDING OF CARP WITH MODIFIED CEREALS

Másílko, J., Hlaváč, D., Hartman, P., Bláha, M., Hartvich, P., Hůda, J., Všetičková, L., 2013. Supplementary feeding of carp with modified cereals. Verified Technology, Edition of Methods, FFPW USB, Vodňany, No. 143, 27 pp. (in Czech)

It was allowed by publisher on 22nd April, 2014 to include the manuscript in this Ph.D. thesis.

SUPPLEMENTARY FEEDING OF CARP WITH MODIFIED CEREALS

J. Másílko[1*], D. Hlaváč [1], P. Hartman[1], M. Bláha[1], P. Hartvich[1], J. Hůda[2], L. Všetičková[3]

[1]University of South Bohemia in Ceske Budejovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses, Institute of Aquaculture, Husova trida 458/102, 370 05 Ceske Budejovice, Czech Republic
[2]Rybářství Třeboň Hld. a.s., Rybářská 801, 37901 Třeboň
[3]Ústav biologie obratlovců AV ČR, Květná 8, 603 65 Brno

**Corresponding author's email: masilj00@frov.jcu.cz*

Introduction to the problem

The common practice of pond aquaculture with carp as the "major fish" anticipates providing proper nutrition of the stock based on natural feed and supplementary feeding. Feed for carps must be efficient as well as environmentally friendly, with respect to effects on water, which is seen as "precious possessions" (Directive 2000/60/ES, 2000 of the European Parliament and of the Council). Water, including retained water, is a factor of production in fish farming, but is not the subject of property. Companies involved in aquaculture are in the state of "limited resources and capabilities" and their prosperity is related to their level of economy income.

Breeding of market carp should fulfil the purpose of the production of whole foods, while respecting the laws in force and using scientific and technical knowledge. The certified technology summarizes new findings related to the efficiency of a traditional treated animal feed – cereals, which have been applied successfully in the nutrition of carp. The technology describes a technique of feeding and the influence of the physiological process of conversion carp feed on the balance of phosphorus in water, as the main element of eutrophication. Monitoring feed efficiency is the subject of controlling water quality and the welfare of farmed fish.

1.1. Supplementary feeding of carp

1.1.1. Importance of natural and supplementary feeding in carp farming

Fish production in ponds in the Central Europe is achieved mostly by semi-intensive farming combining natural and supplementary feeding, mainly cereals (Hepher and Pruginin, 1982; Moore, 1985; Horváth et al., 1992; Kaushik, 1995; Bauer and Schlott, 2006; Adámek et al., 2010; Mareš et al., 2012). Natural food plays an irreplaceable role in the diet of carp. In terms of nutritional levels, it contains easily digestible proteins, mainly from representatives of zooplankton and zoobenthos. This natural food is of good quality and contains all the components necessary for normal growth of fish (Jirásek et al., 2005). It is known that aquatic invertebrates contain large amounts of protein (55–70% of dry matter), necessary for good growth of older carp which need usually 25–30% of protein (Hepher, 1979; Wieniawski, 1983; Kaushik and Preface, 1995; Jirásek et al., 2005). It is apparent that protein from natural feed is not always fully economically used for the growth of fish. Supplementary feeding in ponds creates an increase from 25–30% (Adámek et al., 2008) to 50% (and more) of biomass of fish (Tacon and De Silva, 1997; Szumiec, 1999). The rest of the production arises from natural feed. Supplementary feeding is a useful source of nutrients and energy for carp and adds the necessary components for better growth and production of fish (Abdelghany and Ahmad, 2002). The semi-intensive farming system uses feed of plant origin, such as rye, triticale, maize, wheat and barley (Hůda, 2009; Jankovic et al., 2011). However, such feed does not cover all needs necessary for nutrition and growth of farmed carp although the cereals are cheap and an easily available source of energy (Ghosh et al., 1984; Turk, 1994, 1995; Markovic and Mitrovic-Tutundzic, 2003; Hůda, 2009; Mráz and Picková, 2009).

Starch is an essential component of cereals (60–70%), its digestibility for carp is of about 70% if the cereals are unprocessed (Cirkovic et al., 2002) however, it can be increased to up to 90% by a heat treatment (Przybyla and Mazurkiewicz, 2004). A specific enzymatic system with high amylase and maltase activity allows the carp to use high amounts of carbohydrates (Steffens, 1985). Thanks to their high digestibility, carbohydrates are one of the most valuable sources of energy in feeds for carp and enable better utilization of protein for fish growth (Sadowski and Trzebiatowski, 1995). Total protein content in the grains of cereals varies depending on the type and quality in the range of 7–15% (Füllner et al., 2000; Dordević and Dinić, 2007). On the other hand, the protein composition is poor in essential amino acids (Przybyl and Mazurkiewicz, 2004; Másílko and Hartvich, 2010). Hofer and Sturmbauer (1985) mention that wheat and some other cereals contain anti-nutritional substances (albumin, etc.), which reduce the α-amylase activity. These substances may reduce the digestion of starch. Anti-nutritional factors mainly include protease inhibitors, phytoestrogens giotrogeny, antivitamins, phytates, various oligosaccharides and antigenic proteins – allergens (Tacon and Jackson, 1985; Hendricks and Bailey, 1989; Macrae et al., 1993; Liener, 1994; Anderson and Wolf, 1995; Friedman, 1996; Alacrón et al., 1999). The effects of anti-nutritional factors are undesirable because they reduce feed intake and bioavailability of nutrients from the diet, thus they cause the reduction of the growth and higher water pollution by excreted faeces (Van der Ingh et al., 1996; Refstie et al., 1998; Alacrón et al., 1999; Arndt et al., 1999). A fundamental reason is the absence of enzymes. As for example, phytic acid, the stored form of phosphorus in plant feed, creates complex compounds called phytates with some elements (e.g., Ca, Mg, Zn). The proportion of phytate phosphorus, which is significantly indigestible for monogastric animals, of total phosphorus in the wheat grain is 73% (Kudrna, 2004). To be used by fish, this phosphorus must be released from the complexes by enzymatic hydrolysis through the phytase enzyme. The feed of plant origin contains an insufficient amount of phytase and fish is not able to produce this enzyme. Phytases produced by microorganisms may be added in feed or feed mixtures. Higher utilization of phosphorus from plant components leads to a reduction in the need to add inorganic phosphates in feed mixtures (Rodehutscord and Pfeffer, 1995; Oliva-Teles et al., 1998).

1.1.2. Stability of production in pond aquaculture

Research on and implementation of nutritional strategies to reduce the production of metabolites in ponds is one of the factors that influences the sustainability and stability of pond aquaculture production. Steffens (1985) dealt with the assessment of protein from feed and their conversion into biomass of carp under operating conditions, showing that the deposition of proteins in two-year old carp fed industrially produced feed mixtures based on cereals is in the range of 27–32%, which is relatively low. Similar results were reported by Máchová et al. (2010). In our study, in order to verify the technology, the quality of the feed, its appearance and the applicability of this technology to increase carp stocks have been analysed recently (Hossain et al., 2001, Másílko et al., 2009, Davies and Gouveia, 2010). Type, composition and production method of feed have a significant impact both on the retention of nutrients in fish biomass and, reciprocally, on the amount of metabolites produced by fish stocks in the ponds (NRC, 1993; Jirásek et al., 2005). Improving the quality of feed in order to retain phosphorus in the fish biomass is one of the main objectives in order to reduce the impact of aquaculture on the environment (Gavin et al., 1995, Satoh et al., 2003). From the perspective of excessive nutrient loading of pond ecosystems used for fish farming, it will be important to adjust the amount of feed and fertilizer to reach a zero balance of phosphorus in ponds (TP in feed + TP in fertilizer + TP in fish stock = TP in harvested fish). In such case, all of the phosphorus supplied to the pond in connection with fish farming would be removed from the water again by the biomass of fish. No "extra" phosphorus would be supplied increasing its concentration in the surface water and thus the water eutrophication (Knösche et al., 2000; Duras and Potužák, 2012; Hartman, 2012). Due to the thermal instability of anti-nutritional substances (e.g. lectins and protease inhibitors), it is possible to reduce, limit, or inactivate their function without a depreciation of the feed material using a heat treatment (Másílko and Hartvich, 2010). Some anti-nutritional substances may be present in the hull of the cereals. Therefore, removal or disruption of hull and subsequent heat treatment of certain feed can significantly reduce the impact of these factors (Robaina et al., 1995; Burel et al., 1998; Refstie et al., 1998; Glencross et al., 2007). In accordance with these conclusions, it is expected that thermal or mechanical processing of feed cereals before using them in carp ponds can help to increase the digestibility of such modified cereals and to reduce the load on the pond environment by undigested or poorly digested feed supplements (Jovanovic et al., 2006; Hlaváč et al., 2014), and thus relieve the nutrient balance of the ponds.

1.1.3. Supplementary feeding of stocks in relation to current legislation

Pond aquaculture from the sixties up to the nineties of the 20th century was focused on stocking and feeding with granular feed mixtures containing a proportion of animal protein. The current pond aquaculture returned to the original method of the supplementary feeding of carp with cereals. The reason for feeding cereals is their characteristic stability against nutrient leaching in opposite to industrially produced feed mixtures in the form of granules.

The significantly different digestibility and retention of protein, fat and carbohydrates from natural feed and supplementary cereals in the biomass of carp is equally important. Schäperclaus and Lukowicz (1998) report a recoverability for protein up to 90% and fat based on unsaturated fatty acids up to 95% from zooplankton, while the protein utilization of cereal (depending on fibre content) is in the range of 30–45% by common carp. For the above mentioned, the efforts for more efficient utilization of cereals by increasing their digestibility and the nutrient conversion are completely justified, both in terms of fish farming, and for the interests of the protection of surface waters (Knösche et al., 2000; Duras and Potužák 2011; Hlaváč et al., 2014). Currently used methods in feed production are very effective, in order to improve dietary characteristics, digestibility, eliminate anti-nutritional substances and to extend the shelf life (Kudrna, 2004). The available treatments for carp feed were evaluated in 2009–2012 including thermal treatment (hygienization), pressing of cereals, heat treatment simultaneously with pressing and grinding of grains (Hartvich and Urbánek, 2007; Urbánek, 2009; Hůda, 2009; Másílko et al., 2009, 2010) together with the influence of supplementary feeding on the nutrient balance in ponds (Hartman, 2012; Hlaváč et al., 2013).

2. Aim

The primary concern in the management in pond aquaculture with fish production is the efficient utilization of material inputs, such as feed and nutrients for the development of pond biocenosis. This is also important in regard to the requirement of an environmentally-sound cultivation practices of the surface water, which results in sustainable development of fish ponds.

The aim of the technology is to provide expertise about the effectiveness of modified cereals compared to unmodified cereals in pond common practice of carp farming.

It is necessary for sustainable fish farming in ponds to prevent cumulating of nutrients in ponds or pond basins due to fish increment. On the other hand, it is desirable to convert the nutrients into the biomass of the fish growth and to eliminate them from the water.

3. Place where technology was certified

The technology was certified in 2009–2012 in Rybářství Třeboň a.s. The evaluation was done in the storage ponds in Třeboň (Figure 2) – objects providing a controlled environment without flow, excluding the impact of basin or minimizing the effect of nutrient sources in sediments and in four flow-tight ponds (Figure 1) – in pond systems Naděje, near Frahelž (2012), Horák (2,2 ha), Baštýř (1.7 ha), Fišmistr (2.8 ha) and Pěšák (2.7 ha). Management in those ponds matched operating conditions of normal pond practice, but without fertilization.

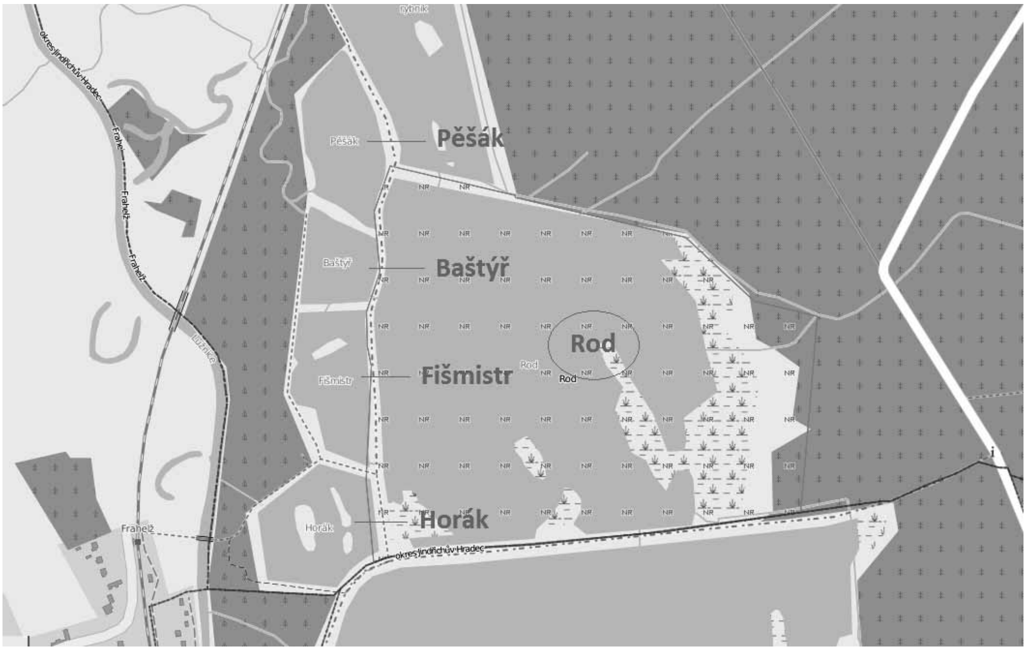

Figure 1. *Ponds near the village of Frahelž (source: www.openstreetmap.org).*

Figure 2. *Preparation of holding ponds for evaluations in Třeboň (Photo: D. Hlaváč)*

4. Description of the technology and results

4.1. Evaluation of the „novelty of the procedure"

A digestibility between 60–86% of untreated cereals by two and three-year old carps was achieved under suboptimal water temperature (19–22°C) but optimal oxygen content (70–75%). However an absorption of nutrients into the muscles of carp reached a maximum of 32% in operating conditions (Steffens, 1985).

The search for options how to ensure a better digestibility and absorption of cereals by fish is therefore justified primarily in terms of economic efficiency of feed and also for the reduction of metabolism products of carp stocks with regard to the protection of the aquatic environment. A carp digests most of fibre and some other polysaccharides and plant proteins with difficulties because of the absence of a stomach and its functions (Jirásek et al., 2005). The current method of traditional pond aquaculture is based on the supplementary feeding of untreated whole cereals. Earlier the only possible treatment of cereals consisted of soaking the cereals in a vessel with a volume of water equal to the weight of the grains (for legumes usually a volume of water to 2.5 times the weight is used). The technology compares whole cereals without modification and heat-treated cereals (Figure 7) labelled as hygienized, pressed cereals, cereals pressed and simultaneously heat-treated cereals and rough grinding. Heat treatment of feed in principle means to apply an action of heat or heat combined with moisture to the feed. Starch, which is present in abundance in the cereals begins to swell at a temperature of 50–60 °C (Doležal et al., 2006). The temperature up to 120 °C with 20% moisture is more appropriate for greater degree of gelatinous. This results into better digestibility of feed (starch is partially hydrolyzed and more easily accessible for digestive enzymes).

For that reason, the heat treatment was used in our evaluation. The heat treatment is executed with hot steam at the temperature of 95–100 °C followed by 60–90 seconds in the hygienizer at a temperature ranging from 75 to 85 °C and under the pressure of 0.2 MPa. After cooling the cereals are placed in a container for further dispatch. Hygienisation secures an increase in the digestibility of polysaccharide – mainly of the starch by gelatizing of 60–90% of the original content. This method is known as the HTST (High Temperature Short Time). The feed is processed gently, without adversely affecting the natural shell of the grains and thus its "water resistance". The heat treatment of cereals (legumes or oilseeds) took place in a continuous process (Kudrna, 2004) and used an equipment manufactured by Bühler AG (Switzerland – Figures 3 and 4).

The effect of **hygienisation of feed** is the preparation of feed with improved dietary properties. This is done primarily by increasing the digestibility of each of the components, reducing the content of anti-nutritional substances and elimination of undesirable microorganisms. It leads also to extend the shelf life of the feed.

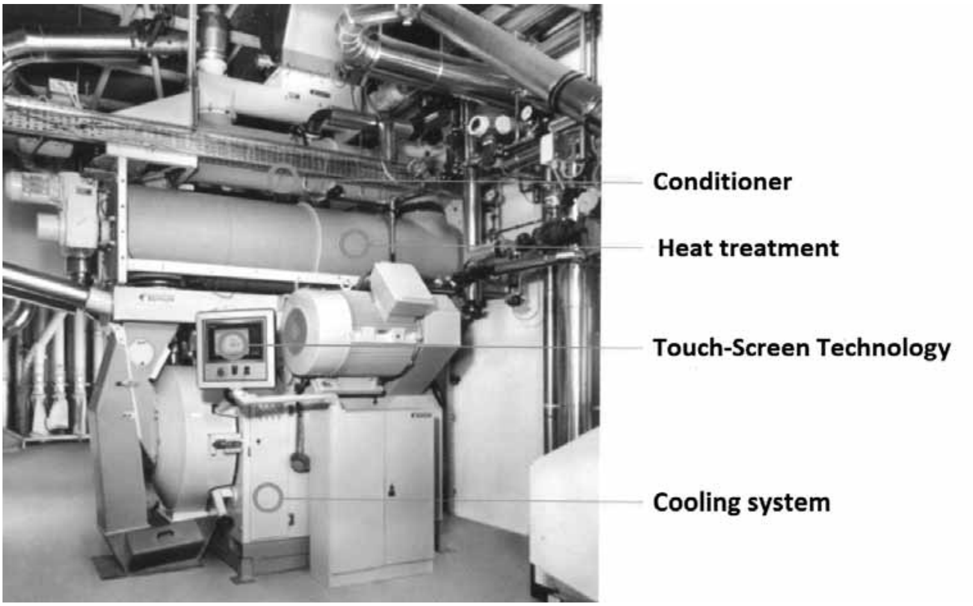

Figure 3. *Hygienisation system (heat-treatment of cereals) according to technical documentation of Bühler AG, Switzerland.*

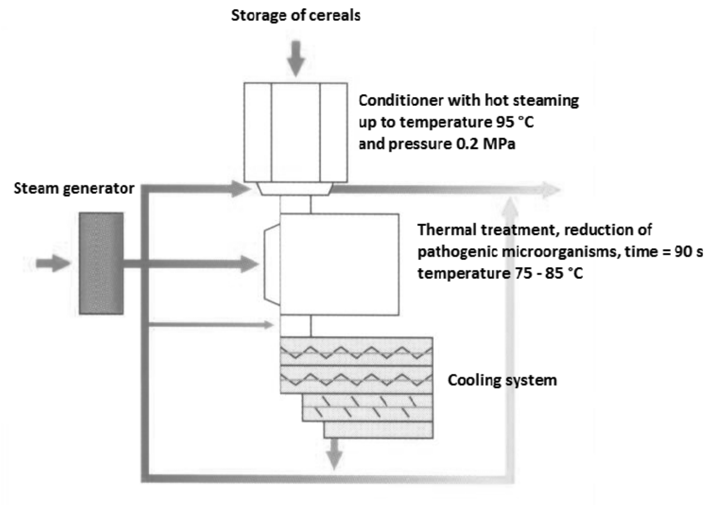

Figure 4. *Scheme of the heat treatment of cereals according to technical documentation of Bühler AG, Switzerland.*

Pressing of cereals is executed by smooth rollers, moving against each other, the distance can be adjusted as needed pressure on the grains. Squeezing undermines the integrity of the grains by compression (Figure 8), thus facilitating its digestibility especially for carps of lower ages (farming of K1 to K2) with weights up to 200 to 250 g per carp. Pressing of cereals can be considered as a first stage of grinding which generally represents a major mechanical treatment of the grain.

Grinding is a violation of surface packing of a grain, which leads to the reduction of undesirable anti-nutritional substances which are contained in the grain (Tacon and Jackson, 1985). The size and particle size balance influences the efficiency of feed and the amount of processing costs. Preparation and treatment of grain feed is very demanding on the required energy input. Therefore, the appropriate solution in order to save energy is very important. In practice, the feed includes three levels of grinding with the following particle sizes: coarse (> 2.0 mm), medium (1.0 to 2.0 mm) and fine (< 1 mm) (Másílko et al., 2009). Grinding of animal feed provides better feed intake and increases the digestibility (Urbánek, 2009). Čítek et al. (1998) describe that the goal of grinding is the adjustment of particle size according to the size and needs of the fish fed with the supplementation. The positive effect of grinding (Figure 9) is the improvement of the digestibility. Some losses occur due to dissolution in the water (up to 30% or more) and leaching of elements (up to 50%) from the grounded grains. Solved particles then decrease water quality and increase the eutrophication of a pond.

4.2. Material and Methods

4.2.1. Supplementary feeding of carp by technologically modified cereals

Feed was given to monoculture stocks of K_3 with the initial weigh of 1 ± 0.3 kg per individual, three times a week (Monday, Wednesday and Friday) from May to September (160–170 days). Stocking density was 363 $ks.ha^{-1}$ of weight-balanced individuals of carps; corresponding to 350 $kg.ha^{-1}$ of biomass. Cereals (wheat, triticale and rye), unmodified, thermally treated, pressed, grinded, or treated with a combination of heat treatment and pressing were used. The tests were done in storage ponds in Třeboň and in four flow-tight ponds in Naděje, a pond system near the village of Frahelž. Annual (vegetation) feed ration of cereals accounted for 3 kg of tested feed per 1 kg of carp during stocking. Current feed ration of feed administration was at the level of 2–5% with respect to the growing season, water temperature, oxygen content and the actual weight of the stocking found by the previous control catching. The level of development of natural feed was also taken into account in the ponds. The nutritional value of feed was determined by the producer and an independent accredited laboratory in the respect of crude protein, Nitrogen-Free Extract (NFE), fat, digestible energy (DE) and total phosphorus (TP).

Before the beginning and after the end of each experiment, the total weight of the fish stock was assessed. Subsequently, the feed conversion ration **FCR** rate was calculated by the following formula:

$$\mathbf{FCR} = \frac{F}{(w_t - w_o)}$$

w_t .... weight of fish stock at the end of the experiment [kg]
w_o .... weight of fish stock at the beginning of the experiment [kg]
F .... amount of feed during the period [kg]

To evaluate the efficiency of treated feed in relation to the untreated feed, the following calculation was then used to calculate U_k increase in conversion in % (feed efficiency in %) according to the FCR:

$$U_k = \frac{(FCRU + FCRUk_n) \times 100}{FCRN_{k1} + FCRN_{kn}}$$

$FCRU_{k1} - FCRU_{kn}$ .... FCR modified feed during the monitoring *1* up to *n*
$FCRN_{k1} - FCRN_{kn}$ .... FCR unmodified feed during the monitoring *1* up to *n*

4.2.2. Calculation of the phosphorus balance

To calculate the phosphorus balance (as a major nutrient causing the development of eutrophication and algal blooming), it is necessary to know the input of these elements by supplementary feeding of cereals. Monitoring included an analysis of the feed nutrients to focus on the TP content in the dry matter of feed.

According to the analysis of feed (2012), the untreated wheat contained 3.15 $g.kg^{-1}$ of TP. The heat treatment increased the content to 3.25 $g.kg^{-1}$ of TP. The difference in the content of total phosphorus in treated wheat was caused by partial loss of water during the heat treatment. To calculate the balance is also important to determine the content of the main element in the biomass of fish in order to quantify the output of nutrients by the fish production.

The analysis was based on data showing that 1 kg of fish biomass contains 8.4 g of TP (Hlaváč et al., 2013). The method of the assessment of the balance of nutrients (by Duras and Potužák, 2012) is shown in Figure 5. The calculation used data of the pond from 2012. The balance was calculated with respect to the input and output of TP in feed and fish biomass without taking into account the inputs and outputs of the inlet and outlet of the water.

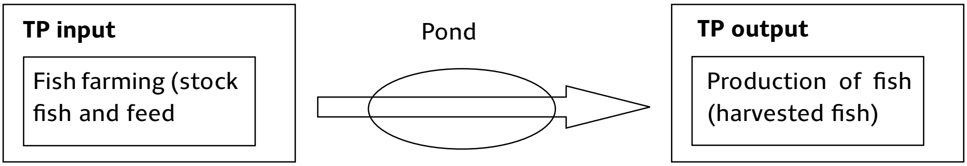

Figure 5. *The basic shceme for evaluating the balance of matter.*

The critical value of the feed conversion rate (FCR_k) was calculated by the proportion of the TP content in the feed and in fish biomass according to Hejzlar et al. (2007), which shows the ratio between the concentration of TP in the fish biomass and the nutrient concentration present in the feed according to the following formula:

$$FCR_k = \frac{Cr}{Ck}$$

Cr .... concentration of the element in fish biomass
Ck...................... concentration of the element in the feed

Example of calculating the balance of phosphorus

The retention of total phosphorus in the fish biomass is shown in Table 1. The output of phosphorus in harvested fish was always higher than the input by feed and the initial fish stock. The highest retention (-2985 $g.ha^{-1}$), i.e. output of phosphorus in fish biomass was recorded in control group (only on natural feed) with zero input of phosphorus by the feed. The fish stocks supplementary fed heat-treated wheat had the highest retention (-1877 $g.ha^{-1}$) compared to the other supplementary fed groups of fish. The lowest retention was recorded in ponds where fish were supplementary fed by wheat without modification (-1301 $g.ha^{-1}$). Furthermore, the value of the critical threshold for the feed conversation rate was calculated (FCR_k = ratio exmashing the content of TP in the growth of the fish to TP content in the feed). The critical factor of feed for wheat without treatment was 2.67 (8.4 $g.kg^{-1}$ of TP in fish / 3.15 $g.kg^{-1}$ of TP in wheat) or wheat after heat treatment is 2.58. According to the obtained coefficients of unmodified wheat, the real FCR was 1.94 and 1.69 for wheat after heat treatment in the ponds. A rational supplementary feeding of the fish stock was performed with regard to the balance of phosphorus in the ponds, without exceeding the critical value of the feeding rate for TP could be concluded.

Table 1. *Example of calculating the balance of phosphorus in the ponds in 2012.*

Feed	**Input** initial fish stock [g.ha^{-1}]	**Input** feed [g.ha^{-1}]	**Output** harvested fish [g.ha^{-1}]	**Total balance** [g.ha^{-1}]	**FCR**	**Total weight gain of fish** [kg.ha^{-1}]
Heat-treated wheat	2290	3562	7729	**-1877**	1.69	644
Untreated wheat	2397	3441	7139	**-1301**	1.94	563
Control	2426	–	5411	**-2985**	–	355

4.2.3. Physical and chemical properties of water and natural feed of fish

Since 2009, the physical and chemical water parameters temperature, oxygen and pH were monitored in the experimental objects (storage ponds and ponds) at fortnightly intervals in the storage ponds and 5 times during the growing season in the other ponds during the feeding experiment. To measure the physical and chemical properties of the water the YSI Professional (Yellow Spring, USA) was used.

Water properties in the storage ponds and the normal ponds are shown in Tables 2 and 3. According to the values observed all water quality parameters were suitable for good growth of carp during the experiment.

Table 2. *Physical and chemical parameters of water (holding ponds in Třebon, 2009–2012).*

Year	**Water temperature (°C)**	**Oxygen (mg.l^{-1})**	**pH**
2009	20.6 ± 2.9	5.4 ± 0.7	7.3 ± 0.9
2010	21.0 ± 2.7	6.2 ± 1.6	7.2 ± 0.4
2011	20.6 ± 2.7	7.0 ± 1.0	8.1 ± 0.5
2012	20.6 ± 2.4	6.3 ± 1.1	8.1 ± 0.6

Table 3. *Physical and chemical parameters of water (ponds 2012).*

Pond	**Feed**	**Water temperature (°C)**	**Oxygen (mg.l^{-1})**	**pH**
Horák	Heat-treated wheat	20.2 ± 5.3	8.2 ± 1.8	7.9 ± 0.8
Fišmistr	Untreated wheat	22.2 ± 5.0	8.6 ± 1.8	8.2 ± 0.8
Baštýř	Control	20.5 ± 5.3	9.7 ± 0.8	8.4 ± 0.5
Pěšák	Heat-treated wheat	22.0 ± 4.5	7.7 ± 1.7	7.8 ± 0.7

Zooplankton samples were collected in the ponds used for the experiment for quantitative and qualitative analysis. For the collection of the zooplankton in the storage ponds a plankton throw net of 80µm mesh size was used. Samples were made with a five-metre lines and preserved in 4% formaldehyde. The analysis was performed in the Sedgwick-Rafter cell counting chamber. The sample of zooplankton from the ponds was filtered through a sieve with a mesh size of 700 µm. The resulting fraction was transferred to a graduated cylinder. After 30 minutes of sedimentation the zooplankton was condensed to the volume of one litre. To calculate the weight and dry weight the following relation was used: 1ml of the zooplankton = 0.5 g of fresh weight. In the Naděj pond system, quantitative samples of macrozoobenthos

(Figure 6) were collected as well in 2012. Samples were collected by the Ekman-Birge bottom sampler with grasping area of 225 cm^2. Four samples were collected in each of the ponds and individually rinsed on a mesh size of 0.5 mm immediately, and the remaining sediment with benthic organisms was preserved as a sample in 4–6% formaldehyde. The density and biomass of the zoobenthos was recalculated for one square meter area of the bottom. Monitoring of the macrozoobenthos in storage ponds was not done because due to the mineral substrate of the bottoms (sand), no significant presence of higher aquatic invertebrates occurred.

Feed Supply – zooplankton (storage ponds in Třeboň, 2009-2012)

Storage ponds 2009

Average abundance of the zooplankton (Rotifera, Cladocera, Copepoda) was to 488 ± 294 ind.l^{-1}, the lowest content was observed in September (82 ind.l^{-1}). Throughout the season the dominant group was Daphnia (Cladocera 69%), mainly the *Daphnia longispina*. The amount of *Daphnia* larger than 0.7 mm during the growing season was highly variable and fluctuated from a maximum of 250 ind.l^{-1} in July to 3 ind.l^{-1} in August.

Storage ponds 2010

A mean value of 204 ± 127 ind.l^{-1}, of zooplankton was observed. *Daphnia* (mainly *Bosmina longirostris*) dominated throughout the whole season (83%). Copepods (Copepoda) accounted for about 13% of the total plankton communities and were represented mainly with the dominant copepod species *Thermocyclops crassus*. Rotifers (Rotifera) represented only 4% of the total abundance. The quantity of *Daphnia* larger than 0.7 mm were in the order of a few specimen recorded in June (3 ind.l^{-1}) and a maximum in August (20 ind.l^{-1}) with *D. longispina* as the dominant species.

Storage ponds 2011

The average value of total the zooplankton was 181 ± 245 ind.l^{-1}. The lowest amount was observed in August (43 ind.l^{-1}), when the community was dominated by daphnia (mainly *D. longispina*). The highest content of zooplankton was found in September (798 ind.l^{-1}), when the copepods dominated the zooplankton with a proportion of 83%. The abundance of daphnia larger than 0.7 mm represented mainly by species *D. longispina*, was in the order of several specimen (average of 5 ± 2 ind.l^{-1}).

Storage ponds 2012

The mean value of the zooplankton was 84 ± 82 ind.l^{-1}. According to the proportion of each group, the population was dominated by copepods with 60%, represented mainly by cyclopoida. Daphnia accounted for 34%, with *D. longispina* as the dominant species. Rotifers accounted for only 6% of the total abundance of the zooplankton. The highest amount was found in July (189 ind.l^{-1}), while the lowest amount of the zooplankton was recorded in September (26 ind.l^{-1}). *Daphnia* greater than 0.7 mm were predominantly represented by *D. longispina* (9 ± 9 ind.l^{-1}).

The quantity the zooplankton (especially individuals larger than 0.7 mm) was lower in the control (no supplementary feeding), while in the ponds with different types of supplementary feed a significantly higher quantity of zooplankton was found, although this also fluctuated in different years.

Feed supply – zooplankton and zoobenthos (experimental ponds 2012)

Specimen bigger than 0.7 mm were represented in all experimental ponds by *Daphnia galeata* during the major part of the growing season, to a lesser extent also by species of *Ceriodaphnia sp*. and *Daphnia pulicaria* (Table 6). The ponds provided enough natural feed for almost the whole season and an almost optimal feed conversion was achieved. The pond of Baštýř (without supplementary feeding) clearly showed lower presence of the bigger zooplankton specimen (Table 4).

Table 4. *Abundance of the zooplankton in ponds, 2012 ($g.m^{-3}$).*

Feed	zoopl. > 0.7 mm ($g.m^{-3}$) průměr	zoopl. > 0.7 mm ($g.m^{-3}$) minimum	zoopl. > 0.7 mm ($g.m^{-3}$) maximum
Heat-treated wheat	1.72	0.69	3.67
Untreated wheat	1.83	0.23	5.50
Control	1.03	0.23	3.67

Macrozoobenthos in the ponds in 2012 consisted of 23 taxa, ten of which were found in the control pond, 15 of them were found in the ponds with supplementary feeding. All 10 taxa occurring in the control pond were common to all samples. Differences in the number of taxa identified in individual samples were not significant between the samples.

The density and biomass of benthic invertebrates was higher in samples from the ponds with supplementary feeding compared to the control pond. Differences were found when comparing the index of saprobity (SI) – while its value by macrozoobenthos showed worse alfa-mezo-saprobity in the control pond (SI 3.46 ± 0.17) ponds with supplementary feeding indicated water quality by about a half degree better (SI 2.79 to 2.98). The diversity index of macrozoobenthos reported no differences; the values were, however, overall, slightly lower in ponds with supplementary feeding (Table 5).

Table 5. *Analysis of benthos in ponds in 2012.*

Feed	Control	Untreated wheat	Heat-treated wheat
density [$ind.m^{-2}$]	411 ± 154	907 ± 353	863 ± 561
biomasss [$g.m^{-2}$]	1.89 ± 0.83	2.89±1.37	3.68 ± 2.74
Number of taxa	5.6 ± 1.7	7.2 ± 2.6	6.7 ± 3.1
Sum of taxa	10	15	15
SI	3.46 ± 0.17	2.98 ± 0.51	2.79 ± 0.54
H´	1.32 ± 0.23	1.27 ± 0.41	1.10 ± 0.45
The dominant taxa	Tubifex tubifex Limnodrilus hoffmeisteri Chironomus plumosus Ceratopogonidae	*Tanytarsus* sp. Tubifex tubifex *Limnodrilus* sp. Ceratopogonidae	*Tanytarsus* sp. *Chaoborus* sp. *Limnodrilus* sp. Ceratopogonidae

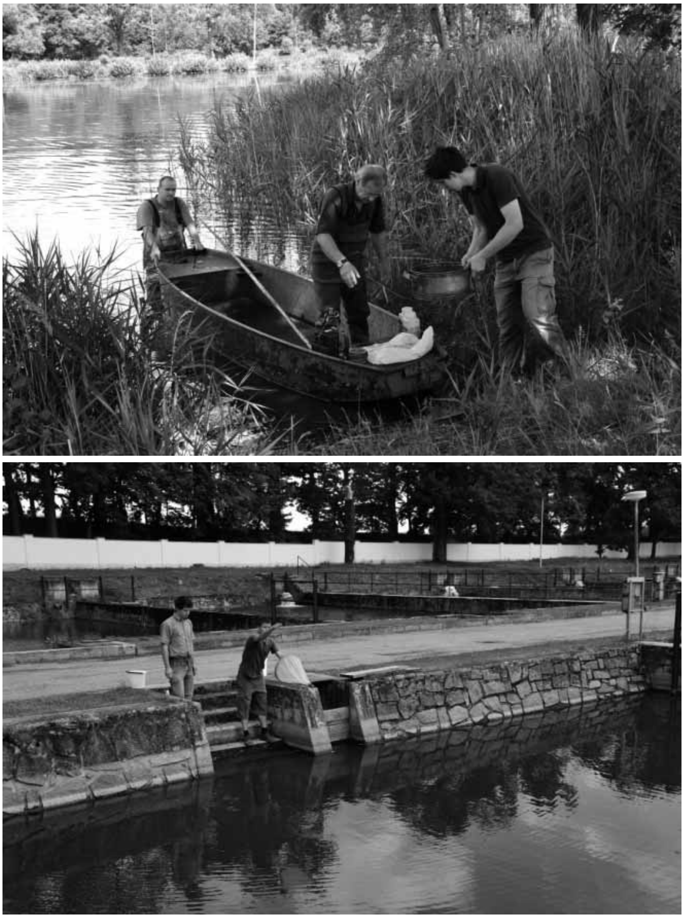

Figure 6. *Sampling of plankton in storage ponds of Třebon and sampling of benthos in ponds of Naděj system (Photo: D. Hlaváč and M. Podhradská).*

4.3. Results of supplementary feeding of carp with treated cereals

The aim of aquaculture today is to ensure an adequate production of fish and aquatic animals. One option for achieving this goal is a technological modification of cereals in order to increase the digestibility and the nutritional value while preserving the natural shell of the cereals, ensuring their water resistance until their consumption. The research was carried out in storage ponds, which are characterized by flow-tightness and relatively low natural production without being affected by input of nutrients, and in normal ponds (Table 6).

In the experimental storage ponds, the efficiency of heat treated cereals resulted in an improvement of the feed conversion of 9.73% at a relatively low natural production level of 185 $kg.ha^{-1}$ in average. The results were reported after seven tests. In contrast, the heat-treated cereals in the ponds showed to improve feed conversion by nearly 13%. However, this can probably be attributed to the higher natural feed production in these ponds compared to the natural feed production in the storage ponds (371–372 $kg.ha^{-1}$). Heat-treated cereals in accordance with a sufficient supply of natural food are suggested to be quite positively for the increase of fish biomass.

Heat treated and pressed cereals resulted in an improved feed conversion of 7.93%, after three tests. This was marginally lower compared to heat-treated cereals. Also pressed cereals improved the feed conversion rate (FCR) by 10.34% compared to untreated whole grains. Since pressing the cereals is not expensive, but unfortunately reducing the shelf life, these treatment can be used especially for K_1–K_2 (age categories of carp). Supplementary feeding by grinded cereals reported insignificantly better feed conversion expressed as more efficient FCR by 2.93% (with a medium to coarse grinding after eight tests in holding ponds in 2009–2012). The research of supplementary feeding by grinded cereals was not realized in ponds due to insufficient soaking of gristle and resolving of the cereals in the water.

Table 6. *Summary of the average values of the FCR (± SD) and the effectiveness of technologically modified feed (U_k) in the storage ponds and ponds in 2009–2012.*

Experimental objects	**Heat-treated**	**Untreated**	**Pressed**	**Untreated**	**Heat-treated and Pressed**	**Untreated**	**Grinded**	**Untreated**
Storage ponds	2.23 ± 0.13	2.47 ± 0.09	1.77 ± 0.50	2.00 ± 0.40	2,24 ± 0,14	2,43 ± 0,10	1,85 ± 0,73	1,91 ± 0,64
U_k FCR [%]	**9.73**		**10.34**		**7.93**		**2.93**	
*Ponds	1.69 ± 0.0	1.94**	–	–			–	–
U_k FCR [%]	**12.89**		–		–		–	

* FCR scored on the ponds was recalculated using the weighted average in relation to their area (ha)
** Without repetition

An example of the calculation of efficiency (U_k) of technologically modified cereals (Fm) is given below (coefficients of modified feed cereals in the numerator and untreated cereals (Fs) in the denominator are in brackets)

$$\text{Example.: } \boldsymbol{U_k} \text{ (storage pond)} = 100 - \frac{(2.05 + 2.35 + 2.04 + 2.23 + 2.43 + 2.2 + 2.29) * 100}{(2.55 + 2.63 + 2.36 + 2.38 + 2.45 + 2.45 + 2.45)} = 9.73\%$$

Figure 7. *Heat treatment – hygenisation of cereals. This method of cereal treatment is indistinguishable from the whole grains. The only chance to detect heat treated cereals is to check the germination. The cereals are not able to germinate after hygenisation process. (Photo: J. Másílko).*

Figure 8. *Mechanical treatment of cereals by pressing. (Photo: J. Másílko).*

Figure 9. *Mechanical treatment of cereals by grinding. (Photo: J. Másílko).*

4.4. Assessment of supplementary feeding of carp with modified cereals

1. Heat treated cereals showed a more efficient conversion of nutrients during the growth of carp in both storage ponds and also in the normal ponds. This confirms that the carp as an agastric animal can use hygienized cereals significantly more efficiently by about 10–13% compared to cereals without modification. Improved dietary characteristics of the heat treated cereals together with a sufficient supply of natural food resulted in a more efficient conversion of nutrients to fish biomass. Humid heat treatment of cereals by the hygienisation process showed an increased digestibility and an increased conversion of nutrients by the fish. Finding new ways to tailor the feed and the confirmation of the feeds suitability for pond aquaculture production seems to be promising for the future.

2. Pressing of cereals, which disrupts the natural protective shell – a husk of caryopsis, makes nutrients in the endosperm better available and partially removes anti-nutritional properties of a grain. This treatment resulted in positive results in the storage ponds, increasing the feed conversion up to 10%. The ponds can be affected in particular by the possibility of dissolving in the water, similarly to cereals modified by grinding. This method of treatment is therefore more appropriate in smaller ponds and fingerling ponds with the recommendation to feed into feed frames.

3. Feeding heat-treated and pressed cereals together resulted into improved conversion by nearly 8% (7.94%) in holding ponds and corresponded with the pressing of cereals. However, applying this adjustment for market carp feeding in the main ponds is at least questionable with regards to possible dissolving in the water and increased costs.

4. Coarsely and medium grinded cereals were observed to be insignificantly positive (3%) for the conversion of nutrients by the fish in the storage ponds. The experiments were not tested in the ponds because of the known dissolving of such treated cereals in the water.

5. Balanced nutrient management in ponds is achievable assuming the exploitation of natural food and an adequate supplementary feeding at up to twice the level of the natural output. To focuse on the nutrient balance in the ponds is not only a tool to optimize their use, but also an opportunity to improve the management of the entire pond system. Different modifications of cereals listed in this technology may be a framework for the sustainable management of pond resulting in an improved phosphorus balance in the pond systems.

4.5. Store and dealing with treated cereals

Storage is a very important aspect for mechanically treated cereals. A long-term storage leads to a wide variety of undesirable biochemical, chemical or even physical changes (Skalický et al., 2008). Disrupting the integrity of the caryopsis enables easy contamination by the surrounding environment. Therefore, it is particularly important to store mechanically processed cereals in clean, dry and ventilated area. Mechanical treatment of cereals is subject to the current need for feeding fry and stocks. It is not recommended to store the modified cereals as reserves for very long time. Furthermore, pressed cereals have a lower specific weight. According to Mareš et al. (2011) pressing reduces the specific weight of up to 30%.

Reducing the specific weight increases the demands on the application for example feeding into enclosed frames.

Cereals treated by hygienisation leave the production process chilled with a lower water content stabilized against external influences during storage. Due to the reduced moisture cereals are protected against for exampleagainst fungal diseases orgermination of grain caused by increased digestibility of the polysaccharides due to the reduced moisture. Logistics related to the storage, and application is comparable to the standard cereal feeds.

5. Economic benefits of technology to the business entity

The cereals treated by hygienisation resulted into a significant reduced FCR (Food Conversion Ratio) = the ratio between the consumed food and fish growth) of 9.73% in the storage pond in a controlled environment with low natural production and up to 12.89 % compared to untreated cereals in ponds with relatively high natural production of appropriate conditions in Třeboň area. Tests were done at K3 to K4 ages of carp with normal stocking densities and supplementary feeding intensity. Evaluating the economic efficiency of heat-treated cereal respected the change in protein and polysaccharide content, due to the reduced water content, which resulted in an increase of the original content of protein and polysaccharide in the order of a few percent. The costs of the technological adjustments, mechanical or thermal were in the order of tens of crowns for 1 q (1 q = 100 kg) of cereals. Technological modifications by pressing or crushing were more3 expensive by about 10 to 15 CZK / q; the heat treatment costs were about 20 to 30 CZK / q.

Třeboň Fisheries currently consumes 100 t of heat-treated cereals annually. Increasing the growth by 10–12% can cause the financial effect of 90–100000 CZK per year. The extension of the use of heat-treated feed can save feed costs in the order of several hundred thousand CZK

Reducing the load of ponds with indigestible and non-absorbable nutrients, especially phosphorus (total phosphorus, TP) is an effect whose financial impact is not precisely quantifiable. Annually, it would be possible to expect a direct saving of tens of kilograms of the TP in the case of Třeboň Fisheries according to the above data, which is released into the Lužnice basin. A further reduction the TP input into the aquatic environment is due to the higher digestibility and utilisation of the heat-treated cereals to considerable parts of fish stocks.

6. The application of technology in production

The use of pressed, heat-treated, grinded cereals or a combination of heat-treatment and pressing is utilized in bigger companies for pond aquaculture. In comparison with granular feed, processed cereals are attractive because of their economic availability, a significantly higher stability in water (excluding grinded cereals) and have an almost equivalent level of digestibility of individual components (proteins nitrogen-free extracts, digestible energy, etc.). This method of treatment is generally available for most aquaculture companies as the feed could be purchased from production and supply enterprises also involved in the sale and distribution of feed for livestock.

The Method of using modified cereals increases the conditions of effective supplementary feeding for a wider age range of carp. While the unmodified cereal feeds are given mostly to carp in breeding ages K_2 to K_3 and K_3 to K_4; the modified cereals, can be used for K_r to K_1 and K_2 for a considerable part of the growing season with very beneficial results due to physiological availability of nutrients.

7. References

Abdelghany, E.A., Ahmad, M.H., 2002. Effects of feeding rates on growth and production of Nile tilapia, common carp and silver carp polyculture in fertilized ponds. Aquac. Res. 33, 415–423.

Adámek, Z., Berka, R., Hůda, J., 2008. Carp as a traditional food fish from pond aquaculture of the Czech Republic. World Aquaculture 39, 52–54/68.

Adámek, Z., Helešic, J., Maršálek, B., Rulík, M., 2010. Applied Hydrobiology. FFPW USB, Vodňany, 350 p. (in Czech)

Alacrón, F.J., Moyano, F.J., Diaz, M., 1999. Effect of inhibitors present in protein sources on digestive proteases of juvenile sea bream (*Sparus aurata*). Aquat. Living Resour. 12, 233–238.

Anderson, R.L., Wolf, W.J., 1995. Compositional changes in trypsin inhibitors, phytic acid, saponins and isoflavones to soybean processing. J. Nutr. 125, 581–588.

Arndt, R.E., Hardy, R.W., Sugiura, S.H., Dong, F.M., 1999. Effect of heat treatment and substitution level on palatability and nutritional value of soy defatted flour in feeds for coho salmon, *Oncorhynchus kisutch.* Aquaculture 180, 129–145.

Bauer, C., Schlott, K., 2006. Reaction of common carp (*Cyprinus carpio* L.) to oxygen deficiency in winter as an example for the suitability of radio telemetry monitoring the reaction of fish to stress factors in pond aquaculture. Aquac. Res. 37, 248–254.

Bühler AG, Switzerland, Project documentaion – Hygienisier – und Kompaktiertechnologie HYSYS, 8 pp. (in German)

Burel, C., Boujard, T., Corraze, G., Kaushik, S.J., Boeuf, G., Mol, K.A., Van Der Geyten, S., Kuhn, E.R., 1998. Incorporation of high levels of extruded lupin in diets for rainbow trout (*Oncorhynchus mykiss*): nutritional value and effect on thyroid status. Aquaculture 163, 325–345.

Cirkovic, M., Jovanovic, B., Maletin, S., 2002. Fisheries. Univerzitet u Novom Sadu, Poljoprivredni fakultet, Serbia. (in Serbian)

Čítek, J., Krupauer, V., Kubů, F., 1998. Fish pond management. Informatorium, Praha, 306 p. (in Czech)

Davies, S.J., Gouveia, A., 2010. Response of common carp fry fed diets containing a pea seed meal (*Pisum sativum*) subjected to different thermal processing methods. Aquaculture 305, 117–123.

Directive 2000/60/EC of the European Parliament and of the Council of 23 October 2000.

Doležal, P., Zeman, L., Kopřiva, A., 2006. Technological treatments of feeds. In: Zeman, L., Kopřiva, A., Mrkvicová, E., et al. (Eds), Nutrition and feeding of livestock. Profi Press, pp. 147–194. (in Czech)

Dordević, N., Dinić, B., 2007. Hrana za životinje. Cenzone tech-Europe, d.o.o. Arandelovac. pp. 257–259, 261,265, 464, 466, 467, 469–471, 473.

Duras, J., Potužák, J., 2012. First results obtained by monitoring of phosphorus mass balance of several fish ponds. Vodní hospodářství, 6, 210–216. (in Czech)

Friedman, M., 1996. Nutritional value of proteins from different food sources. A review. J. Agricult. Food Chem. 44, 6–29.

Füllner, G., Langner, N., Pfeifer, M., 2000. Ordnungsgemässe Teichbewirtschaftung im Freistaat Sachsen,Sachsisches Landesanstalt für Landwirtschaft, Referat Fischerei Königswarta, 66 pp.

Ghosh, S.K., Mandal, B.K., Borthakur, D.N., 1984. Effects of feeding rats on production of common carp and water quality in paddy-cum-fish culture. Aquaculture 40, 97–101.

Glencross, B.D., Booth, M., Allan, G.L., 2007. A feed is only as good as its ingredients – a review of ingredient evaluation strategies for aquaculture feeds. Aquacult. Nutr. 13, 17–34.

Hartman, P., 2012. Nutrition model of pond biocoenosis in relation to total phosphorus. In: Urbánek, M. (Ed.), Fish farming and water quality. Czech Fish Farmers Association, České Budějovice, 24. 1. 2012, pp. 33–48. (in Czech)

Hartvich, P., Urbánek, M., 2007. Influence of cereals feeding on production parameters in market carp farming to reduce feed costs. Verified technology, FA USB, 9 p. (in Czech)

Hejzlar, J., Duras, J., Komárková, J., Turek, J., Žaloudík, J., 2007. Water-supply reservoir Mostiště: Evaluation of monitoring water-supply reservoir and catchment area 2006. Biology Centre of the ASCR, v.v.i., České Budějovice, 71 pp.

Hendricks, J.D., Bailey, G.S., 1989. Adventitious toxins. In: Halver, J.E. (Ed.), Fish Nutrition, Academic Press, London, pp. 605–651.

Hepher, B., 1979. Supplementary diets and related problems in fish culture. Proc. World Symp. on Finfish Nutrition and Fishfeed. Technology, Vol. I. Hamburg, Germany.

Hepher, B., Pruginin, Y., 1982. Tilapia culture in ponds under controlled conditions. In: Pullin, R.S.V., McConnel, R.H.L. (Eds), The biology and culture of tilapias. International Conference on the Biology and Culture of Tilapias, Bellagio (Italy), 2-5 Sep. pp. 185–203.

Hlaváč, D., Hartman, P., Adámek, Z., Másílko, J., Bláha, M., Pechar, L., Baxa, M., 2013. Effect of supplentary feeding in carp ponds on water quality and nutrient balance. In: Urbánek, M. (Ed.), Fish farming and water quality II. Czech Fish Farmers Association, České Budějovice, pp. 21–30.

Hlaváč, D., Adámek, Z., Hartman, P., Másílko, J., 2014. Effects of supplementary feeding in carp ponds on discharge water quality: a review. Aquacult. Int. 22, 299–320.

Hofer, R., Sturmbauer, C., 1985. Inhibiton of trout and carp α-amylase by wheat. Aquaculture 48, 277–283.

Horváth, L., Tamás, G., Seagrave, C. (Eds), 1992. Carp and Pond Fish Culture. Fishing News Books, Blackwell Scientific Publications Ltd., UK, 154 pp.

Hossain, M.A., Focken, U., Becker, K., 2001. Effect of soaking and soaking followed by autoclaving of Sesbania seeds on growth and feed utilisation in common carp, *Cyprinus carpio* L. Aquaculture 203, 133–148.

Hůda, J., 2009. Production efficency of cereals in carp farming. Dissertation, FA USB, 159 pp. (in Czech)

Jankovic, S., Jovanovic, R., Cirkovic, M., Ljubojevic, D., Rakic, S., Milosevic, N., 2011. Importance and use of grains in fish nutrition. In: Z. Markovic (Ed.), 5th International Conference Aquaculture and Fishery. Belgrade (Serbia), 1–3 Jun 2011, pp. 103–109.

Jirásek, J., Mareš, J., Zeman, L., 2005. Nutrition requirements and tables of fish feed nutritive value. Final report, MZLU Brno, 68 pp. (in Czech)

Jovanovic, R., Milisavljevic, D., Sredanovic, S., Levic, J., Duragic, O., 2006. Proizvodnja hrane za ribe različitih fizičkih karakteristika. Biotechnol. Anim. Husband. 22, 339–349.

Kaushik, S.J., 1995. Nutrient requirements, supply and utilization in the contex of carp culture. Aquaculture 129, 225–241.

Kaushik, S., Preface, D., 1995. Fish nutrition in practice. In: Kaushik, S.J., Luquet, P. (Eds), Practice Proceedings of the IV International Symposium on Fish Nutrition and Feeding, 24–27 June 1995, Bairritz, France, INRA Les Colloques, n. 61. Institut National De LA recherche Agronomique, Paris, France, pp. 15–16.

Knösche, R., Schreckenbach, K., Pfeifer, M., Weissenbach, H., 2000. Balances of phosphorus and nitrogen in carp ponds. Fish. Manag. Ecol. 7, 15–22.

Kudrna, V., 2004. Proccessing of feed, term of their safety and efficacy. Scientific Committee for Animal Nutrition, Prague, 56 pp. (in Czech)

Liener, I.E., 1994. Implication of antinutritional components in soybean foods. Crit. Rev. Food Sci. 34, 31–67.

Macrae, R., Robinson, R.K., Sadler, M.J. (Eds), 1993. Encyclopaedia of Food Science, Food Technology and Nutrition. Academic Press, London, UK, pp. 2964–2965.

Máchová, J., Valentová, O., Faina, R., Svobodová, Z., Kroupová, H., Mráz, J., 2010. Water pollution by carp originating from different pond management systems. Bull. VÚRH Vodňany 46, 31–38. (in Czech)

Mareš, J., Kopp, R., Brabec, T. 2011. New methods in fish farming. In: Hulík, M. (Ed.), Intesive rearing of fish and quality and protection of waters. Třeboň, Rybářství Třeboň a.s., pp. 5–13. (in Czech)

Mareš, J., Kopp, R., Brabec, T., 2012. Common carp meat quality – nutritional and sensory parametres. In: Urbánek, M. (Ed.), Fish farming and water quality. Czech Fish Farmers Association, České Budějovice, pp. 73–80. (in Czech)

Markovic, Z., Mitrovic-Tutundzic, V., 2003. Gajenje riba. Zaduzbina Andrejevic. Beograd, Serbia, 138 pp.

Másílko, J., Hartvich, P., 2010. Utilization of modified cereals in market carp farming (a review). Bull. VÚRH Vodňany 46, 35–43. (in Czech)

Másílko, J., Urbánek. M., Hartvich, P., Hůda, J., 2009. Effective supplementary feeding of mechanically modified cereals in market carp farming at Fisheries Třeboň. Edition of Methods, FFPW USB, Vodňany, No. 98, 11 pp. (in Czech)

Moore, L.B., 1985. The role of feeds and feeding in aquatic animals production. GeoJournal 10, 245–251.

Mráz, J., Picková, J., 2009. Differences between lipid content and composition of different part sof fillets from crossbred farmed carp (*Cyprinus carpio*). Fish. Physiol. Biochem. 35, 615–623.

NRC, 1993. U.S. National Research Council. Nutrients requirements of fish. National Academy Press, USA, 114 pp.

Oliva-Teles, A., Pereira, J.P., Gouveia, A., Gomes, E., 1998. Utilisation of diets supplemented with microbial phytase by seabass (*Dicentrarchus labrax*) juveniles. Aquat. Living Resour. 11, 255–259.

Przybyl, A., Mazurkiewicz, J., 2004. Nutritive value of cereals in feeds for common carp (*Cyprinus carpio* L.), Czech J. Anim. Sci. 49, 307–314.

Refstie, S., Storebakken, T., Roem, A.J., 1998. Feed consumption and conversion in Atlantic salmon (*Salmo salar*) fed diets with fish meal, extracted soybean meal or soybean meal with reduced content of oligosaccharides, trypsin inhibitors, lectins and soya antigens. Aquaculture 162, 301–312.

Robaina, L., Izquierdo, M.S., Moyano, F.J., Soccoro, J., Vergara, J.M., Montero, D., Fernandez-Palacios, H., 1995. Soybean and lupin seed meals as protein sources in diets for gilthed seabream (*Sparus aurata*): Nutritional and histological implications. Aquaculture 130, 219–233.

Rodehutscord, M., Pfeffer, E., 1995. Effect of supplemental microbial phytase on phosphorus digestibility and utilization in rainbow trout (*Oncorhynchus mykiss*). Water Sci. Technol. 31, 143–147.

Sadowski, J., Trzebiatowski, R., 1995. Fish feeds. Pasze Polskie 1/2, 110–118. (in Polish)

Satoh, S., Hernández, A., Tokoro, T., Morishita, Y., Kiron, V., Watanabe, T., 2003. Comparison of phosphorus retention efficiency between rainbow trout (*Oncorhynchus mykiss*) fed a commercial diet and a low fish meal based diet. Aquaculture 224, 271–282.

Schäperclaus, W., Lukowicz, M., 1998. Lehrbuch der Teichwirtschaft, 4., neubearbeitete Auflage, Parey Buchverlag Berlin, Germany, 590 pp.

Skalický, J., Kroupa, K., Branda, J., Pastorková, L., 2008. Ošetřování a skladování zrnin ve věžových zásobnících a halových skladech. Metodická příručka MZe, Výzkumný ústav zemědělské techniky, v.v.i. Praha, 88 pp.

Steffens, W., 1985. Grundlagen der Fischernährung. VEB Gustav fisher Verlag Jena, Germany, 226 pp.

Szumiec, J., 1999. Intensive rearing of carp in the ponds. Bulletin VÚRH Vodňany, 4, 165–167. (In Czech).

Tacon, A.G.J., Jackson, A.J., 1985. Utilisation of conventional and unconventional protein sources in practical fish feeds. In: Cowey, C.B., Mackie, A.M., Bell, J.G. (Eds), Nutrition and Feeding in Fish, Academic Press, London, UK, pp. 119–145.

Tacon, A.G.J., De Silva, S.S., 1997. Feed preparation and feed management strategies within semiintensive fish farming systems in the tropics. Aquaculture 151, 379–404.

Turk, M., 1994. Croatian freshwater fisheries in 1993. Ribarstvo, Zagreb, 52, 119–132.

Turk, M., 1995. Croatian freshwater fisheries in 1994. Ribarstvo, Zagreb, 53, 105–118.

Urbánek, M., 2009. Influnece of cereals feeding on production parameters and quality of fish flesh in market carp farming. Dissertation, FA USB, 179 pp. (in Czech)

Van der Ingh, T.S.G.A.M., Olli, J., Krogdahl, Å., 1996. Alcohol-solube components in soybeans cause morphological changes in the distal intestine of Atlantic salmon, *Salmo salar* L. J. Fish Dis. 19, 47–53.

Wieniawski, J., 1983. Ziwienie karpi. Gospodarka rybna 35, 15–18.

Internal referee
Assoc. Prof. M.Sc. Zdeněk Adámek, CSc.
University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Center of Aquaculture and Biodiversity Hydrocenoses,
Zátiší 728/II, 389 25 Vodňany, Czech Republic, www.frov.jcu.cz

External referee
Assoc. Prof. Dipl.-Ing. Lukáš Kalous, Ph.D.
Czech University of Life Sciences Prague, Faculty of Agrobiology, Food and Natural Resources, Department of Zoology and Fisheries
Kamýcká 129, 16521 Praha 6 – Suchdol, Czech Republic, www.af.czu.cz

Verification and application of the technology in the year 2013
User of the technology: Rybářství Třeboň, a.s., company headquarters Rybářská 801, 379 01 Třeboň, Czech Republic

Adress of authors
Dipl.-Ing. Jan Másílko (masilj00@frov.jcu.cz)
Dipl.-Ing. David Hlaváč (hlavac@frov.jcu.cz)
Dipl.-Ing. Pavel Hartman, CSc. (hartman@frov.jcu.cz)
Dipl.-Ing. Martin Bláha, PhD. (blaha@frov.jcu.cz)
Assoc. Prof. Dipl.-Ing. Petr Hartvich, CSc. (hartvich@frov.jcu.cz)
University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, South Bohemian Research Center of Aquaculture and Biodiversity Hydrocenoses,
Zátiší 728/II, 389 25 Vodňany, Czech Republic, www.frov.jcu.cz

Dipl.-Ing. Jan Hůda, Ph.D. (jhuda@rybarstvi.cz)
Rybářství Třeboň Hld. a.s. Rybářská 801, 379 01 Třeboň, Czech Republic

Dipl.-Ing. Lucie Všetičková, Ph.D. (lucka.kocanda@seznam.cz)
Institute of Vertebrate Biology, Academy of Sciences of the Czech Republic, v.v.i., Květná 8, 603 65 Brno, Czech Republic

In the edition of Methodologies (Technological series) issued by University of South Bohemia in České Budějovice, Faculty of Fisheries and Protection of Waters, editorial board: Prof. Dipl.-Ing. Martin Flajšhans, Dr.rer.agr., Dipl.-Ing. Blanka Vykusová, CSc., Zuzana Dvořáková, number of copies: 200 pcs, published 2014, Graphic design and technical realization: JENA Šumperk Publishing.