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In cooperation with **Leibniz-Institute of freshwater ecology and inland Fisheries**

A COMPARISON OF THE EFFECTS OF A COMMERCIAL AVAILABLE FEED AND A HERMETIA ILLUCENS (LINNAEUS 1789) BASED FEED ON THE DEVELOPMENT–DYNAMICS OF CLARIAS GARIEPINUS (BURCHELL 1822) IN AQUAPONIC-SYSTEMS

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ABSTRACT

A feeding test in a floating aquaponics system was conducted over the course of six weeks involving two groups of African catfish, *Clarias gariepinus* (BURCHELL 1822). The two groups were fed different feeds with an emphasis on the main protein source to observe the resulting differences in development dynamics. While the control group received classic feed with fishmeal, the test group's feed contained black soldier fly meal, *Hermetia illucens* (LINNEÉ 1758), as a main protein source. Fish fed with the control feed showed more adequate results in the observed characteristics length, weight and condition factor but differences were only partially significant. Overall significances were only observed for weights of fish starting after five weeks of testing. It is, however, not with complete certainty to state that these discrepancies are only resulting from the different feeds, as environmental influences should also be considered.

In einer schwimmenden Aquaponik-Anlage wurde über sechs Wochen ein Fütterungsversuch an zwei Gruppen afrikanischer Welse, *Clarias* gariepinus(BURCHELL, 1822) durchgeführt. Den beiden Gruppen wurden unterschiedliche Futtermittel zugeführt, die sich vor allem durch ihre Hauptproteinquelle unterschieden, um dadurch die unterschiedliche Auswirkung der jeweiligen Proteinquelle auf die Entwicklungsdynamik der Fische zu beobachten. Die Kontrollgruppe bekam Futter mit klassischem Fischmehl, während das Futter der Testgruppe Soldatenfliegenmehl der *Hermetia illucens* (LINNEÉ 1758) als Hauptproteinquelle enthielt. Fische die mit dem Kontrollfutter gefüttert wurden zeigten bessere Ergebnisse in den beobachteten Charakteristika, Länge, Gewicht und Konditionsfaktor, die Unterschiede waren allerdings nur teilweise signifikant. Erst anch fünf Wochen der Testphase konnten signifikante Unterschiede beim Gewicht der Fische festgestellt werden. Des Weiteren ist nicht mit völliger Sicherheit zu sagen, dass diese Unterschiede alleine als Resultat der unterschiedlichen Futtermittel sind, da auch Umwelteinflüsse eine Rolle gespielt haben können.

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1 Introduction

1.1 Clarias gariepinus

Catfish or *Siluriformes* are part of the bony fish and belong to the superorder *Ostariophysi*, which contains about 64% of all freshwater fish. An important characteristic of the *Ostariophysi* is the Weberian apparatus, which is used for sound perception. The Weberian apparatus is a modification of the fish's anterior four or five vertebrae and likely evolved as a consequence of the poor visibility found in common habitats of the *Ostariophysi* (Guy G. Teugels, 1996, p. 10).

As of 1996, the *Siluriformes* represent 33 families, approximately 412 genera and 2584 species(Guy G. Teugels, 1996, p. 12).

Figure 1 *Clarias gariepinus* (BURCHELL 1822)

Clarias gariepinus (BURCHELL 1822), as seen in Figure 1, belongs to the family *Clariidae.* The family contains 14 genera and about 168 species. The Asian *Clarias batrachus* (LINNEÉ 1758), *Clarias microcephalus* (GÜNTHER 1864) and the African *Clarias gariepinus* are the most studied of these species. Furthermore, they are of great importance to fisheries and aquaculture (Guy G. Teugels, 1996, pp. 16–17).

1.1.1 Physiology

The external physiology of *Siluriformes* varies greatly, making it difficult to describe the whole order. Nevertheless, *Siluriformes* do share some general characteristics. Most are found with scales, but possess barbels. There can be up to four pairs of these barbels on a catfish, which are covered in taste buds and are used to detect food. G.G. Teugels (1983) describes *Clariidae* as typically having an elongated body as well as long dorsal and anal fins. The dorsal fins are always without a spine. Some genera have an adipose fin with elongated neural spines, while the pectoral fin has a strong spine. Specimen with sizes of up to 130 centimetres in length and weights of 12.8 kg are reported (Guy G. Teugels, 1996, p. 10; 16–17).

Clariidae have a suprabranchial organ that allows them to breathe atmospheric air, which makes them obligated air breathers according to De V. Pienaar (1968) as cited by Olaniyi & Omitogun (2013). This suprabranchial organ is formed from Clariidae's second and fourth epibranchial, which also enables some species to cover distances of several hundred meters on land by using their pectoral spines to move. They are also ground dwellers (Olaniyi & Omitogun, 2013, pp. 314–315; Teugels, 1986, pp. 16–17). According to Shepherd & Bromage (1988, p.33) Catfish can generally live in water with low values of dissolved oxygen below 2 mg/L.

1.1.2 Feeding Behaviour

Clarias gariepinus is an omnivorous predator and its exact feeding behaviour appears to depend on several factors such as the fish's development stage, habitat, available feed and time of day. De Graaf & Janssen (1996) describe different observations of these feeding behaviours cited from M. N. Bruton (1979), Micha (1976) Munro (1967) and Spataru, Viveen, & Gophen (1987) and shows that *Clarias gariepinus* feed on aquatic insects, fish and higher plant debris. However, they have also been observed consuming terrestrial insects, molluscs, crustaceans, arachnids and even fruits. While one study's observations showed that the amount of consumed zooplankton rose with size of the fish, another showed that 81% of feed consisted of preyed fish. To cover such a high variety of food sources, *Clarias gariepinus* has numerous adaptations. Its wide mouth has a great capacity for vertical

displacement, which is accompanied by its long gill rakers on its five branchial arches and pharyngeal teeth. This results in a high filtering capability while also making *Clarias gariepinus* an efficient predator. Its predatory skills are supplemented by its barbels, which the fish uses in its slow, methodical hunting technique (de Graaf & Janssen, 1996, pp. 9–11).

1.1.3 Distribution

Catfish have a wide geographical distribution, comprising the Americas, Europe, Asia, Africa and Australasia. Almost all *Siluriformes* are freshwater species, as is the family of the *Clariidae* (Guy G. Teugels, 1996, p. 10).

According to De V. Pienaar (1968), Guy G. Teugels (1996) and Clay (1979) as cited by Olaniyi & Omitogun (2013) and de Graaf & Janssen (1996) respectively, *C. Gariepinus* is mostly common in Africa up to Syria, but also in southern Turkey and Southeast Asia. Its preferred habitats are floodplain swamps and pools. Additionally, this species can be found in stagnant water bodies like lakes, ponds or around dams, along with streams and rivers. Many of their inhabited water bodies can be seasonally dry or reduced to muddy remnants. Therefore, *C. gariepinus* is euryoecious and can handle a very wide range of different environmental conditions. Their optimum growth rate occurs at temperatures around 28°-30° Celsius, but they can survive in a range between 8°-35° Celsius with a pH range of 6.5-8.0 (de Graaf & Janssen, 1996, p. 9; Olaniyi & Omitogun, 2013, pp. 314–315).

1.2 Aquaculture

Figure 2 shows the development over past few decades in the aquaculture sector, during which it experienced an immense growth. From 1980 to 2010, the industry for food fish production in aquaculture grew to twelve times its size, with an average growth rate of 8.8 per cent per year. Although this trend was stronger in the 1980s and 1990s with respective growth rates of 10.8 per cent and 9.5 percent, its current growth rate at 6.3 per cent is still notable. As a result, the proportion of worldwide produced food fish for humans from aquaculture increased from only 9 per cent in the 1980s, to 47 per cent in 2010. A majority of the production takes place in Asia, representing 89 per cent of the global volume of aquaculture production, while China alone is responsible for 61.4 per cent of the worldwide production. Additionally, while the percentage of freshwater fish production in aquaculture increased in Asia, all other continents showed a decline. For example, in Europe the amount of brackish and marine water production rose from 55.6 per cent in 1990 to 81.5 per cent in 2010. Norway is the biggest producer of fish in Europe, accounting for 1.009.010 tonnes in 2010, which represents 39.95 per cent of the 2.523.179 tonnes produced in Europe that

year. However, in a worldwide comparison, Europe produces only 4.2 per cent of all aquaculture-produced food fish (FAO, 2012, pp. 25–28).

In 2010, carp production comprised the majority of worldwide freshwater fish production, accounting for 71.9 per cent of the share, or 24.2 million tonnes. Catfish, with the exception of *Pangasius*, had a worldwide production rate of just below two million tonnes. Of these, 73.3 per cent are yielded in Asia and 12.3 per cent in African countries where the production of *Clarias gariepinus*, the North African catfish, dominates (FAO, 2012, p. 36; 38).

Lucas & Southgate (2012) cite from New (1999) that depending on the intensity of the operation, aquaculture can be a threat to its surrounding environment. These negative effects are mostly a direct consequence of poor planning, inappropriate site selection or management procedures, lack of attention to environmental protection and the intense increase in production that coincides with the onset of new, profitable industries. Some possible impacts from land-based aquaculture are the destruction of natural habitats, eutrophication and sedimentation as a result of effluents, overuse of resources or negative influence on native fisheries or biodiversity. However, in developed countries, effluents from aquaculture are not generally a major contributor to water pollution, as opposed to other industries or agriculture (Lucas & Southgate, 2012, pp. 84–85).

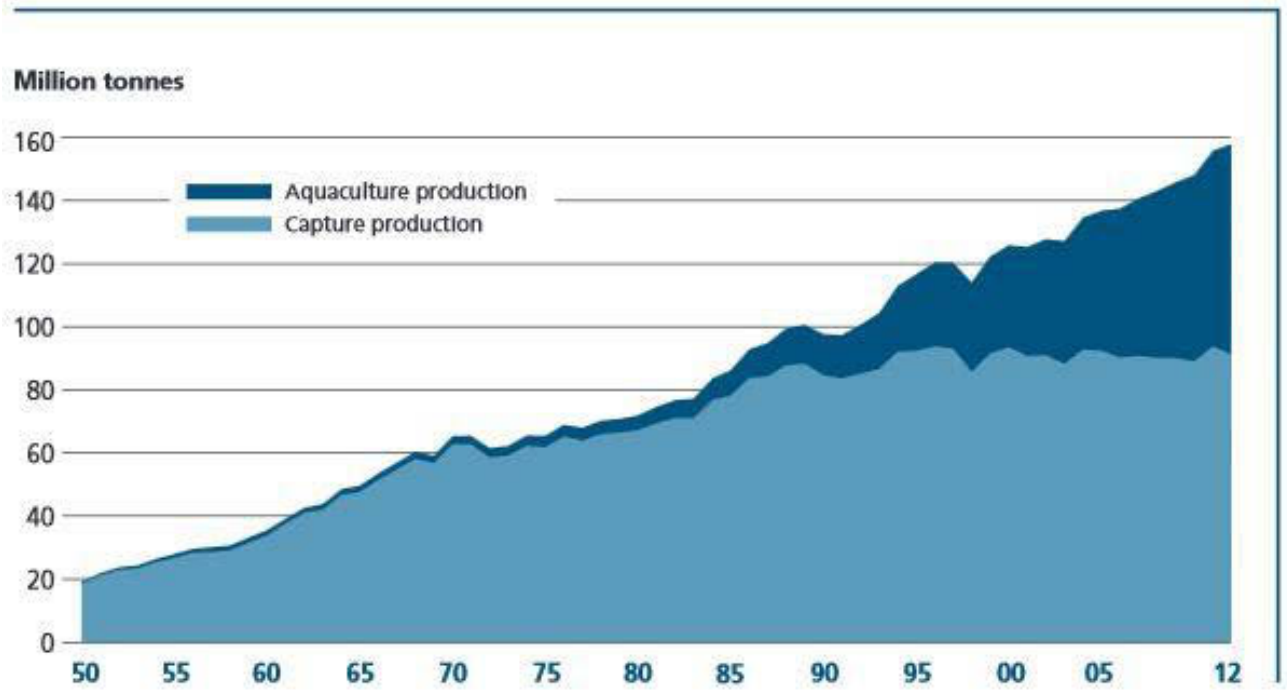


Figure 2 World fisheries and aquaculture production (FAO, 2014)

1.2.1 Classification of Aquaculture

Lucas and Southgate (2012) differentiate between aquaculture projects on three main criteria: type of structure, the amount of water exchange and the intensity of the culture. All

three have a strong influence on the way the aquaculture is conducted. Commonly used structures in aquaculture include ponds, tanks, and nets.

Ponds, like in Figure 3, are the most commonly used structure and also the oldest technique, providing an option for simple aquaculture. The structure itself does not have to be much more than a hole in the ground, and the water inlet and outlet is often gravity-driven. Its requirements to work are few: a consistent source of good quality water and soil that allows for the construction of a pond and with enough nutrients to support the pond ecosystem. This simple way of operating an aquaculture is also cost-effective (Lucas & Southgate, 2012, pp. 18–19).

Figure 4 shows fish tanks, which are the second most frequently used structures in aquaculture and can be used both outside and inside. They are often made of concrete or synthetics but can be made of other materials. One advantage of using tanks is that it allows for aquaculture in areas where the construction of a pond is not possible. Tanks can come in different sizes and shapes, with several ways of functioning. These, once again, are dependent on the kind of aquaculture that is conducted (Lucas & Southgate, 2012, pp. 22–23). Tanks are also used for recirculating aquaculture and aquaponic systems, as they allow more control over certain water characteristics than ponds. These characteristics include the amount of water and its quality.

Water exchange describes the amount of water that gets in and out of the system, and consists of four main categories: static, open, semi static or recirculating (Lucas & Southgate, 2012, p. 18).

Figure 3 Trout farm in Lower Austria
Source: http://www.blauerkreis.at/unsere-projekte/freunde-und-partner/

Static systems do not have any planned exchange of water during production time, with the exception of evaporation and precipitation. The system of static water exchange, or non-water exchange, is mainly used in pond systems. Consequently, the system's water body is more vulnerable to quality decline resulting from large biomasses and is therefore only appropriate for extensive aquaculture. Whereas the static system has almost no water exchange, the open system has a great deal of it. Open systems use natural water bodies such as the ocean or lakes for aquaculture. The organisms of interest are caged or kept in these water bodies other suitable ways, such as on long lines for bivalves. In open systems, the water quality is maintained by the environment through tides or natural currents, and therefore the fish farm operator has little to no control over it. Costs are therefore small and are limited to capital costs along with costs for feeding, labour and eventually lease. On the other hand, suitable spaces for open system aquaculture are often also of interest for other uses such as fishing or tourism. Open systems are also more vulnerable to predation or diseases than closed systems. Aquaculture with open systems appears in combination with extensive to intensive operations. A key example of an intensive open aquaculture system is Atlantic salmon production (Lucas & Southgate, 2012, pp. 32–33).

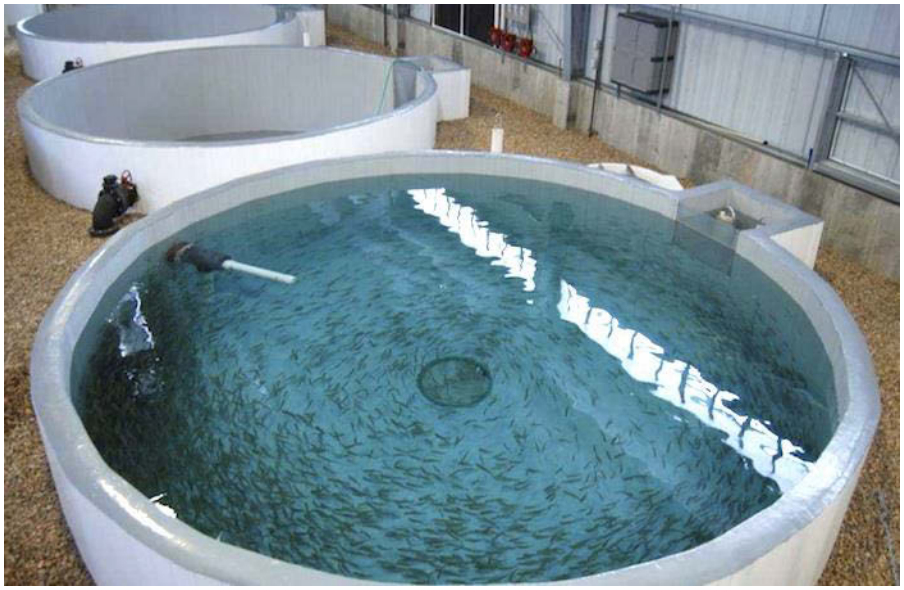


Figure 4 Tanks in a recirculating aquaculture system (RAS) © http://web.octaform.com/blog/topic/recirculating-aquaculture-systems

Semi-closed systems consist of parts from both aforementioned systems. Ponds or tanks are used as a structure to nurse the organisms like in static systems, but there is a higher water exchange. Usually a nearby water source is used as a constant water inlet to the system and the outlet is also noticeably greater than in static systems. It does not reach as high an amount of water exchange as the open system and is still regulated by the fish farm operator. This technique can lead to a higher productivity because of the higher water exchange. It also exposes the system to more threats from outside (Lucas & Southgate, 2012, pp. 33–34).

The most regulated kind of aquaculture system is the closed or recirculating aquaculture system (RAS), as seen in Figure 4. These systems have only little water exchange and water losses. As only small amounts of water are lost and gained in these systems, the water has

to be recycled somehow after usage. The filtration of water can be achieved through different techniques: one kind of RAS, which usually use a combination of mechanical and ecological filtration, is aquaponics (Shepherd & Bromage, 1988, pp. 98–100).

The third characteristic to differentiate between types of aquaculture is the intensity of the culture. This describes the amount of organisms per unit of space or area. Intensity of a venture can be intensive, semi-intensive or extensive(Lucas & Southgate, 2012, p. 18)

The amount of cultured fish is mainly dependent on two factors: the supply of the organisms with sufficient feed and the upkeep of the needed water quality for the cultured species. Intensive aquacultures meet those needs and therefore have the potential for the highest yields per area. To do so requires a greater effort, which usually also results in higher costs than in other kinds of systems for reason such as larger amounts of required feed. In intensive aquaculture, the operator introduces all energy input as feed. Other costs include measures for water exchange. What threshold of fish per area is characterised as an intensive aquaculture may differ between species (Lucas & Southgate, 2012, pp. 27–28).

In contrast to intensive aquaculture, there is extensive aquaculture, which depends on a natural ecosystem to provide feed and water quality. In this case, the operator's main task is to provide the habitat and to stock the fish at the beginning of the season. Ongoing work is mainly regarding maintenance, such as controlling harmful aquatic plants or giving additional feed. In these cases, the operation could also be considered semi-extensive aquaculture. Extensive aquaculture is often used in ponds. In general, extensive systems produce visibly less biomass than intensive systems, with abundances usually below 500 kg per ha. However, they also have much lower costs in construction and upkeeping, therefore both systems can produce profits. In a global comparison, extensive aquaculture is in the majority (Lucas & Southgate, 2012, pp. 28–29).

The type of aquaculture chosen depends on different factors: Different species may have very different demands. Whereas carp is an ideal species for pond systems with warm and stagnant waters, trout prefer cold water with a certain current. Fish also show different requirements as they develop over time. Additionally, sometimes the type of aquaculture is a matter of space and funding.

1.3 Aquaponics

The previous chapters mainly described classic aquaculture. However, as mentioned in the introduction, classic aquaculture often fails at being sustainable and can pose a major threat to its environment. Thus, what differentiates aquaponics from "classic" aquaculture?

Aquaponics is a combination of recirculating aquaculture and hydroponics – a soilless system for crop production. Hydroponically grown plants are used to extract dissolved waste

from fish via the system's recirculating water. According to Love et al., the technique was first introduced in the 1970s by different sources (Love et al., 2015, p. 67).

Every aquaponics (AP) system is also a recirculating aquaculture system (RAS). RAS allows for reusing up to more than 90 per cent of the water, which is especially attractive for areas with water shortages. Furthermore, in such systems, it is also possible to control the environmental and water quality parameters. These contain, among others, the control of heat and diseases. However, with these advantages come the disadvantages of heightened costs. Heaters, aerators, pumps and other devices might be necessary to keep the system functional depending on the species being raised. The reuse of the water also requires several techniques to clean the water. On the other hand, the higher investments might also lead to higher incomes, resulting from higher growth rates (Shepherd & Bromage, 1988, pp. 98–100).

Aquacultural wastes can be difficult to be manage, as they are suspended or dissolved in water, thus aquaponics offers a way of solving this problem. It is also one of the few techniques that can extract low amounts of nitrogen and phosphorus from the water. This occurs as the wastewater from the aquaculture tanks flow past the plants, which use the nutrients in it for nourishment. These raised crops can lead to a further income for the fish farmer (Buzby & Lin, 2014, p. 39).

Despite their advantages, RAS and aquaponics systems have some unavoidable drawbacks. It is not an easy task to run hatchery – or small animal operations. The reasons for these problems vary from operation, species and stage of fish development. Additionally, water quality can be a problem, with the most common problem being the presence of solids. Other problems can be oxygen and carbon dioxide. One major problem is that these systems are still seldom cost effective, which results from different factors. As mentioned, one such factor is the higher amount of initial costs in comparison to classic aquaculture. These costs result from necessities like pumps or water filters. Tanks and pipes also result in extra necessary costs. These investments stand in contrast to flow-through systems or even extensive systems, which in some cases have only feed as investments. In addition to investment costs, RAS also requires higher amounts of maintenance and therefore, again, higher costs. As a result, it takes an average of eight years after implementing this type of system until the initial costs are recovered (Badiola, Mendiola, & Bostock, 2012, pp. 29–31).

Nevertheless, in northwest Europe, there is a clear trend from flow-through systems to RAS. On the Faroe Islands, for example, all seven of its hatchery systems are now RAS (Bergheim, Drengstig, Ulgenes, & Fivelstad, 2009, p. 47).

While in common RAS, only mechanical filters are used to clean used water, aquaponic systems also support this by natural processes with the cultured plants, making the cleaning procedure more effective.

How Aquaponics Work

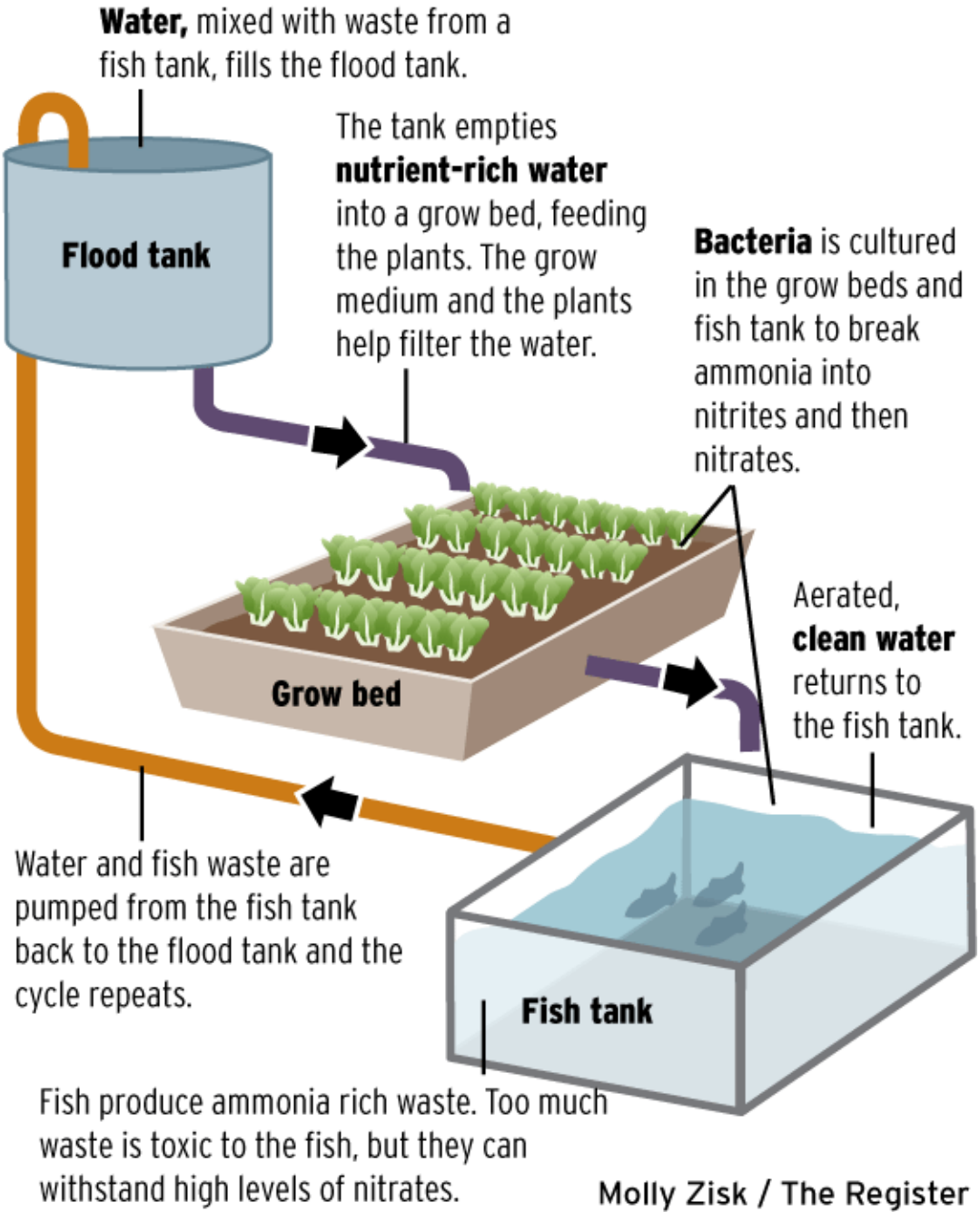


Figure 5 One circle Aquaponics-System Source: http://www.howtoaquaponic.com/wp-content/uploads/2015/04/what-is-aquaponics-cycle-2.gif

1.3.1 One Circle System versus Two Circle System

Two major kinds of aquaponics systems exist today. A system with a single water cycle (OCS) like in Figure 5 contains water, which recirculates the system as a whole. The second kind is a two water cycle system (TCS), which has a one sided connection between aquaculture and hydroponics. This means that while in OCS-cleaned wastewater recirculates directly back from the hydroponics to the aquaculture part of the system, this does not occur in TCS. Figure 6 shows a schematic of TCS. Even as the concept of OCS was a major advance from classic aquaculture regarding sustainability, some problems remain. Whereas most fish and bacteria used for *nitrification* prefer pH values of around 7 to 9, this is not the case for most plant species used in hydroponics. These plants usually prefer pH values of 5.8 to 6.2. Therefore in OCS, one part of the system must have a disadvantage resulting from suboptimal pH values, while with TCS this problem can be solved. Additionally, water is constantly filtered in OCS, which prevents the accumulation of higher amounts of nutrients in the system. This prevented previous aquaponics systems from providing optimal conditions for the rearing of high nutrient-requiring crops (Kloas et al., 2015, p. 180; Rakocy, J., Shultz, R. C., Bailey, D. S., & Thoman, 2003, p. 2).

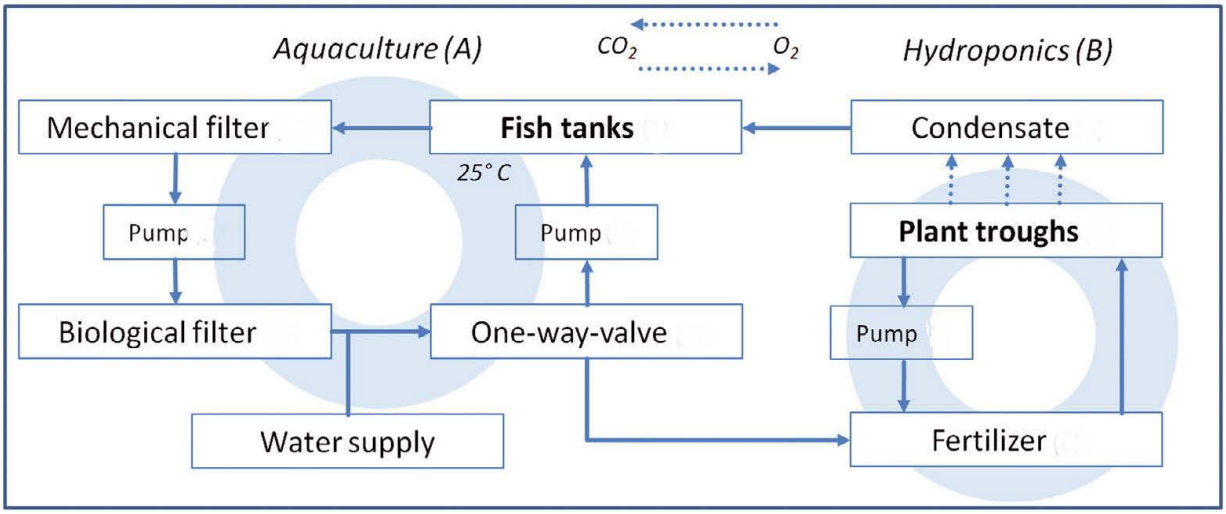


Figure 6 Two-circle aquaponics system ASTAF-PRO (Kloas et al., 2015)

1.4 African Catfish in Aquaculture and Aquaponics

Clarias Gariepinus is a common fish in aquaculture for several reasons.

As mentioned, *C. gariepinus* are air breathers, which allow keeping them in higher densities, as they are less dependant on the oxygen amounts in the water compared to other species. This is an important factor for intensive and artificial aquaculture (Teugels, 1986, pp. 16–17).

To run a functioning aquaponics project, a certain amount of nutrients provided by the fish is necessary. The higher the amount of nutrients, the higher the amount of crops that can be reared. Furthermore, crops with higher nutritional prerequisites can be used. Therefore, a higher density of fish equals a higher density of crops. This leads to an overall higher production of the whole system.

C. gariepinus also tends to have sluggish behaviour when it is cared for properly. This behaviour leads to a low necessity of upkeep for the fish. This leads to additional important factors: African catfish have an efficient feed utilisation and a high growth rate. These factors are vital for every aquaculture or aquaponics installation, whether the operation is being conducted for commercial reasons with a main focus on profits or if it is mainly used to support a self-sustaining food production. Furthermore, *C. gariepinus* can produce usable eggs and sperm throughout the year if its climatic needs are met (Huisman & Richter, 1987, pp. 9–10). However, this might not be the case in Central Europe under normal conditions during winter and fall. Nevertheless, it might be possible to provide these requirements in indoor aquaponics facilities. Additionally a heightened aggressive behaviour was observed in *C. gariepinus* when kept in lower densities compared to fish kept in higher densities (Boerrigter, Bos, Vis, Spanings, & Flik, 2015, p. 12). This provides additional support for the use of intensive aquaculture.

Catfish are not only used in classical aquaculture, but in aquaponics as well. In a survey conducted by Love et al., responses showed that 25 per cent of farmers were raising catfish. Although the survey was addressed to an international audience, 81 per cent of respondents were located in the United States. The survey, however, does not explicitly mention what species of catfish were used by the farmers (Love et al., 2015, p. 70). In consideration of the responses, it is not surprising that *Clarias gariepinus* replaced tilapia for the first time as the most reared fish in Sub-Saharan aquaculture in 2004. Nigeria, with the greatest yield of catfish in Africa, even imports catfish feeds from regions in Europe (FAO, 2012, p. 33).

1.5 Fish Nutrition

Digestion refers to the process of extracting nutrients for organisms to use them. This process mainly occurs in organisms' intestines, but is prepared by chewing. After digestion, nutrients are transported to the blood or lymphs through different ways of transportation such as diffusion. The mouth, pharynx, esophagus, stomach and intestines belong to the catfish's digestive tract, and are complemented by the pancreas, liver and gall bladder. The pH level of catfish stomachs range from 2 to 4, whereas the intestine has a pH value of 7 to 9 (Robinson, Li, & Maning, 2001, pp. 1–2).

As stated in the first law of thermodynamics, energy remains constant, but may be altered or reassigned inside a system. Living organisms have to obtain energy from certain sources.

While autotroph organisms can use their surroundings, like sunlight and nutrients, medium heterotroph organisms accomplish this through the intake and utilisation of food. This obtained energy is again transferred to other states to keep the organism working. The processes occurring are *endergonic* and *exergonic*: The exergonic processes describe a loss of energy and happen during metabolism, while endergonic processes cannot work without an interchange with exergonic processes and ultimately have an exergonic effect. This interchange happens, for example, through the compound adenosine triphosphate (ATP). As mentioned, these processes are overall exergonic. The energy that cannot be used for the organism is lost, for example, through warmth. The amount of heat that is produced through this process is always a result of the amount of energy that entered the organism. Therefore, the ways in which energy is transferred is not as important as the energy content of the processed compounds (Murray et al., 2012).

According to Baxter (1990) as cited by Halver & Hardy (2003), gross energy, or GE, is the amount of energy in the whole diet. GE is commonly represented with a plus sign (+). Even as the GE depends on a specific substance, there are mean values for the most important nutrients, lipids, proteins and carbohydrates. These are, 39.5, 23.6 and 17.2 kJ/g respectively. Minerals (ash) have no energy content, as they are not combustible. These and other nutrients must be digested and absorbed by the organism to make their energy content useable. Some parts of feed cannot be digested, however, and leave the organism as feces. The unused energy is egested as feces and is referred to as feces energy (FE). The difference between these two units – GE and FE – is called digestible energy (DE) (Halver & Hardy, 2003, pp. 8–9).

For aquatic organisms, determining the DE can be more challenging than for terrestrial organisms. As these tests must be conducted through feed intake, the medium of water poses a problem. The solution of nutrients in water, affecting the nutritional intake and therefore their egestion, cannot be completely controlled (Robinson et al., 2001, pp. 1–2).

There are at least 40 known nutrients necessary for a catfish’s metabolic function to work properly, with the most important ones presented in this chapter.

Carbohydrates are used as a source of energy, a component of tissue and also play a role in metabolic processes. Therefore, sources of carbohydrates are not only found in plants, but in animals as well, although in smaller abundances. It is not necessary per se for fish to take in carbohydrates as part of their diet, because animals can also produce carbohydrates from lipids and proteins. Therefore, the ability to process carbohydrates and use them as an energy source differs in different fish species. In general, warm and freshwater fish are more capable to do so than cold water and marine fish. One reason may be that warm water fish have a higher intestinal amylase activity. While there are known enzymes for processing

carbohydrates in fish, the hormonal and metabolic processes involved remain unclear. Whereas catfish are able to use polysaccharides quite well, mono- and disaccharide are more challenging. Catfish seem to process glucose the same way as mammals, but less effectively. This may result from an absence of enzymes or endocrine systems capable of such processing (Robinson et al., 2001, pp. 5–6).

Fats and oils, also known as lipids, are a very effective source of energy and deliver more than twice the amount of energy as carbohydrates. Lipids also play a major role in several parts of metabolism. For example, lipids are the only way for fish to get essential fatty acids (EFA), which can't be synthesised by the organism itself. In general, the essential EFA for fish are omega-3 acids. Catfish need only small amounts of these omega-3 acids. Additionally, some, but not all non-essential fatty acids can be synthesised by catfish (Robinson et al., 2001, p. 6).

Table 1. Amino Acids
(De Silva & Anderson, 1995, p. 69;
FAO, n.d.)

Essential	Non-essential
Arginine	Alanine
Histidine	Asparagine
Isoleucine	Aspartate
Leucine	Cysteine
Lysine	Glutamate
Methionine	Glutamine
Phenylalanine	Glycine
Tryptophan	Proline
Valine	Serine
	Tyrosine

Protein serves as transport for essential and non-essential amino acids. These amino acids, and not the protein itself, are of main importance in fish nutrition. A list of both essential and non-essential amino acids in fish is shown in Table 1. The protein source must contain sufficient amounts of all essential and of most non-essential amino acids (De Silva & Anderson, 1995, p. 72).

1.6 Dietary requirements of *Clarias gariepinus*

Information on necessary percentages of nutrients in *Clarias gariepinus* nutrition varies in literature. As an omnivorous predatory species, high amounts of protein are required, although the inclusion of higher amounts of carbohydrates is possible as *C. gariepinus* also feeds on plants. Whereas protein levels of 50 to 55 per cent are advised in very early development stages, during the grow-out stage, these values can be reduced to 40 to 43 percent. Of these the highest amounts of essential Amino Acids are needed of Arginine, Leucinine, Lysine and Phenylalanine, as marked in Table 1. Corresponding lipid values are at around 9 per cent in early stages and with a relatively high range, depending on the source, from 8 to 17 percent. The intake of carbohydrates is recommended from 15 to 35 percent(FAO, n.d.).

Table 17 in the Appendix shows this list of advised nutrient-compositions.

1.7 Fish Feed

Catfish, especially in aquaponics and other intensive aquacultures, are feed-dependent. This means that the use of fish feed is essential in aquaponics. This is especially relevant, as fish feed is the most expensive part of an on-going aquaculture operation. Depending on the raised species, the demands for ingredients and therefore costs vary. The most important nutrients are carbohydrates, proteins and lipids.

Cho & Kaushik (1990) as cited by Halver & Hardy (2003) state that the digestibility of the feed is an important factor and must therefore be compiled for the aquatic species for which the feed is used (Halver & Hardy, 2003, p. 9).

According to Pillay (1993) and as cited by him from Cowey & Sargent (1972) and Stickney & Shumway (1974) Carbohydrates are the least expensive part of aquaculture feed. Although research on carnivorous species raises doubts about the value of carbohydrates in fish feeds, practical experience shows that they can be used when kept in balance with the other nutrients. The utilisable amount may vary from up to 3.8 kcal per gram in easy digestible sugars to almost zero for indigestible cellulose. The ability to digest starch from carbohydrates depends on the production of the enzyme amylase. Amylase occurs in herbivores species through the entire digestive tract. In addition to these dietary reasons, carbohydrates also function as binders in fish feed (Pillay, 1993, p. 95).

In natural diets, the share of lipids can be up to 50 per cent. In commercial aqua feeds, however, they usually make up less than 10 per cent, which is roughly the amount of lipids in regular fishmeal. Tests suggest that higher shares of lipids do not sustain higher growth in fish. Furthermore, too many lipids can lead to diseases like fatty liver (Pillay, 1993, pp. 99–100).

Protein is the most expensive part of artificial fish feed. It is also the primary source for amino acids and nitrogen. As fish have an energy value of 4.5 kcal per gram, which is comparatively higher than that of mammals and birds, a high-protein diet is possible. The main reason high-protein diets do not occur in aquaculture is their cost inefficiency since protein is the costliest part of fish feed. The most commonly used source for protein is fish meal (FM) (Pillay, 1993, p. 95).

The Food and Agriculture Organization of the United Nations (FAO) estimates that in 2008, 46.1 per cent of all 31.7 million tonnes of aquacultures (including water plants) were feed-dependent. In 2008, there were 708 million tonnes of industrial compound animal feed produced and 29.2 million tonnes of these were aqua feeds. Additionally, an estimated that 18.7 million to 30.7 million tonnes of aqua feeds was produced on farms.

In recent decades, the production of fishmeal and fish oil was characterised by major changes. From the second half of the 1970s to the mid-1990s, their production grew significantly. By 1995, 29.5 per cent of the total marine catch worldwide was used to produce fishmeal and fish oil. In both percentage and absolute terms, this trend has decreased from 1995 onward, while proportionally the worldwide catch decreased to 20.2 percent. This trend seems as though it will continue: In recent years, the amount of fisheries by-products for FM and fish oil (FO) has increased by up to 25 per cent of total production. Although the overall production of these products is decreasing, its use in aquaculture is rising. From 1995 to 2008, it increased from 1.87 million tonnes (with a peak of 4.23 million tonnes in 2006) to 3.73 million tonnes of FM and from 0.46 to 0.78 million tonnes FO. However, a continuation of the overall decrease is expected. Reasons include the drop in caught fish and the replacement of FM with more cost-effective alternatives. While the use of FM is expected to decline over the next years, FO seems as though it will increase over a longer time period (FAO, 2012, pp. 172–179).

	Diets (%)		
Ingredients	D1	D2	D3
Corn	11	11	11
Soy bean meal	34	39	43.5
Fish meal	25	12.5	-
Maggot meal (HFLM)	-	12.5	25
Blood meal	10.3	10	10
Wheat bran	11	8.0	6.0
Palm oil	5.2	3.5	1.0
Bone meal	3.0	3.0	3.0
Vitamin/Mineral premix*	0.3	0.3	0.35
DL-Methionine	0.2	0.2	0.15
Total	100	100	100
Nutritional composition (in % of dry matter)			
Dry matter	90.45	90.78	90.13
Crude Protein	40.76	40.59	40.74
Ether extract	9.2	8.98	8.51
Crude fiber	4.1	4.87	5.22
Ash	10.59	9.10	8.0
M.E (kcal/kg)	2,795.8	2,794.5	2,737.3
M.E.(MJ/kg)	11.7	11.69	11.45

* Biomix fish vitamin/mineral premix providing per kg of diet at 5kg per tonne inclusion: 20,000 i.u, Vitamin A, 2000 i.u,Vit. D3, 200 mg Vit E, 8mg Vit K3, 20mg Vit B1, 30mg Vit B2, 12mg Vit B6, 50 mg Pantothenic acid, 0.8mg Biotin, 150 mg Niacin, 0.05mg Vit B12, 4.0mg Folic acid, 500mg Vit C, 600 mg Choline chloride, 200mg Inositol, 200mg Betaine, 2.0mg Cobalt, 40mg Iron, 5.0mg Iodine, 30mg Manganese, 4mg Copper, 40mg Zinc, 0.2mg Selenium, 100mg Lysine, 100mg Methionine, 100mg Anti-oxidant.

Figure 7 Feed formulation from (Aniebo et al., 2009)

Still, there are alternatives to using conventional fishmeal as a protein source.
These include different plants protein, meat meal and bone meal. Because of its nutritional value, the soybean is a prominent source for plant protein. However, the use of unprocessed soybean meal proved problematic resulting to its anti-nutritional constituents. This can be resolved by turning soybean into soybean protein concentrate (Dersjant-li, 2002, p. 541).

1.7.1 *Hermetia illucens* – meal in fish feed

As fishmeal is, from a nutritional point of view, very effective in fish diets, it is of interest to find substitutions with similar profiles of nutrients and amino acids.

A potential substitute as protein source is insect meal. Insects are a natural component of many fish species' diets, meaning it is reasonable to assume that they might be useable in

aquaculture. Possible species include the black soldier fly (BSF), *Hermetia illucens* (LINNEÉ 1758), the common housefly, *Musca domestica* (LINNEÉ 1758) and several others. BSF feed on manure and are able to convert this into adequate amounts of protein and lipids. When the larvae are further fed with fish wastes, the amount of lipids, including omega-3 acids, can rise. BSF along with other *Diptera* species also show a very amino acid profile to fishmeal's (Barroso et al., 2014; Sheppard, Newton, Thompson, & Savage, 1994, pp. 277–278).

The black soldier fly, of the order *Diptera*, belongs to the family *Stratiomyidae*. Tomberlin & Sheppard (2002) describe its occurrence in tropical and moderate areas. *Hermetia illucens* develops on decomposing organic matter and only feed during larval stages of their life-cycle (Jeffery K Tomberlin, Adler, & Myers, 2009, p. 930).

Table 2 Amino acid profile of fishmeal, Hermetia illucens larvae meal (HIL) and Hermetia illucens pupae meal (HIP)

	Fishmeal	HIL	HIP
ARG%	7,42	8,24	8,05
HIS%	7,86	5,29	5,16
ILE%	5,04	5,76	5,34
LEU%	7,81	6,87	6,83
LYS%	8,78	7,6	7,31
MET%	2,93	1,5	3,26
PHE%	5,38	6,88	6,22
PRO%	4,76	6,16	5,56
THR%	6,26	5,39	4,95
TYR%	3,91	6,35	7,14
VAL%	5,56	6,31	6,34

Over the last decade, several papers have discussed insects in fish feed. (Barroso et al., 2014) presented variations in the amount of crude protein ranging from 36.2 per cent to 40.6 percent. At the same time, all of the presented cases show a much higher percentage of lipids in insect meal. For *Hermetia illucens* larvae, this value is approximately 18 per cent and might be influenced by its development stage, the age and diet. Other sources report 45 per cent protein and 35 per cent lipids, and other similar compositions (Barroso et al., 2014, p. 195; van Huis, 2011, pp. 567–568).

As a comparison, fishmeal generally has a crude protein percentage of 73%, compared to lipids at around 8%. This illustrates the differences regarding protein and lipid compositions in fishmeal and BSF meal (Figure 8). Other insects also show higher percentages in protein and lower amounts of lipids. What qualifies *Hermetia illucens* as an alternative protein source in fish diets is its amino acid profile, seen in Table 2, which is very similar to that of fishmeal. This is also the case in other *Diptera* species (Barroso et al., 2014).

Henry, Gasco, Piccolo, & Fountoulaki (2015) present several ways of handling the high amount of lipids, drawing on different papers. Several techniques allow for defatting the

meal, which also increases the percentage of crude protein in the insect meal (Henry et al., 2015, p. 16).

Aniebo et al. show that a diet without fishmeal is possible for *Clarias gariepinus*. In a trial, three different feed formulations were produced. One of these formulations, Figure 7, abstained completely from the use of fishmeal and instead had housefly maggot meal in it. After the trial, there was no significant difference in important factors such as specific growth rate, feed conversion ratio and protein efficiency ratio. All diets were in compliance with nutrient requirements of *C. gariepinus* (Aniebo, Erondu, & Owen, 2009).

Figure 8 Hermetia illucens meal © Filzwieser 2015

1.8 Hydrochemical Introduction

Of further interest for aquaponics are the nutrient cycles of nitrogen and phosphorus. These and other nutrient cycles, require some basic prerequisite conditions to function in aquatic systems. This includes organisms as sources and users of nutrients, and water as a solution and way of transportation. This results in particular nutrients being dissolved in the water. Under these conditions, certain processes taking place: the bioactivity of organisms, interaction between water and sediments as nutrient storage, and the loss of nutrients in several manners. The sum of this production is described as the trophic level (Schwörbel & Brendelberger, 2013, pp. 142–143).

In natural waters, nitrogen can occur in several different forms. Sources of nitrogen vary from precipitation to biological activity.

According to Alexander (1965a, 1965b), Kuznetsov (1970) and O'Neill & Wilkinson (1977), ammonium (NH_4^+) enters the system through the gills of fish. It is toxic in higher concentrations and is turned to nontoxic nitrate (NO_3^-) through nitrification. In this process, the reduced nitrogenous compounds are transformed to a more oxidised state. Nitrate can

then be used as nutrients by plants. For ammonium to be turned to nitrate, aerobic bacteria are necessary, which turn ammonium into nitrite (NO_2^-) first. The first step, transforming ammonium into nitrite, occurs mainly through nitrosomonas, whereas the nitrite is turned into nitrate mainly by nitrobacter (Wetzel, 2001, pp. 215–126).

In balance with NH_4^+, the highly toxic NH_3 appears. The amount of NH_3 in the water depends on the total ammonium, based on NH_4^+ and NH_3 as well as the pH value and temperature of the water. The lower the pH and temperature are, the lower the proportion of NH_3. Therefore at a pH value of 7 and below, ammonia is almost untraceable. However, very low amounts of about 0.02 mg/L are considered the maximum value for vulnerable fish like salmonids (Shepherd & Bromage, 1988, p. 34).

The literature varies on what amounts of nitrite are threatening to fish's well-being. Helfrich, Libey, & Tech, (1990) suggest values below 0.5 mg/L in aquaculture whereas Molleda, Fe, & Thorarensen, (2007, p. 14) cite a value of 1.0 mg/L from Pillay & Kutty, (2005). Siikavuopio & Sæther, (2006) also show that values of up to 4.99 mg/L show reduced weight gain in Atlantic cod *Gadus morhua (LINNAEUS 1758)* after only 32 days of exposure.

Lethal amounts of nitrate range at values of up to 1000 mg/L according to Timmons et al. (2002) as cited by Molleda et al., (2007, p. 14) for certain fish species.

Phosphorus is usually the main limiting factor in natural waters as its availability is very low. Phosphorus can occur in three different forms: inorganic P, mainly orthophosphate, soluted organic P and in particular, organic P. Phosphorus is integrated in the food chain through the consumption of photoautotroph organisms (Schwörbel & Brendelberger, 2013, p. 129; Wetzel, 2001, pp. 239–240).

1.9 Standard Weight in Fish

The standard weight in fish describes the expected weight of a fish corresponding to a certain body length. This value is dependent on the fish species. The following formula as cited by Davies, Tawari, & Kwen (2013) from Ricker (1973) can be used for calculations:

$$W = a\,L^b$$

W and **L** describe the weight and length, respectively. Usually grams and centimetres are used as units of measurement. The other variables describe the regression constant with **a** and the slope with **b** (Davies et al., 2013, p. 325).

According to LeCren, as cited by Blackwell, Brown, & Willis (2000), there are two explanations for the use of the standard weight in fish. The first is to mathematically describe the relationship of the two characteristics, weight and length, with the main objective to predict one from the other. The second application is to assess the well-being, fatness or

gonad development of the fish considering the expected values (Blackwell et al., 2000, pp. 1–2).

The standard weight also describes how growth develops in fish. When the **b** value is at 3, fish have an isometric growth. Whereas values below or above 3 describe a negative or a positive allometric growth, respectively (Khaironizam & Norma-Rashid, 2002, p. 21).

Additionally, there are several further ways of describing fish well-being. One such method is Fulton's condition factor. This condition factor is a commonly used tool to assess the overall fitness of fish by putting its weight and length in relation. Fulton's condition factor is formulated as:

$$K = 100(W/L^3)$$

Again, W stands for weight and **L** for length in gram and centimetres. 100 is a constant that is used to transform the **K value** close to 1 (Anderson & Neumann, 1996, p. 455).

2 Research Questions and Hypotheses

In consideration of the previous chapters, the following research question was formulated for this thesis.

2.1.1 Main Question

- **What is the growth reaction of *Clarias gariepinus* fed with feed containing *Hermetia illucens* meal as a main protein source, in comparison to feed with fishmeal as a main protein source on the development dynamic of two groups of *Clarias gariepinus* in identical aquaponics systems?**

2.1.2 Hypotheses

The main question is specified by three hypotheses:

H1: The development dynamic of *Clarias gariepinus'* body length shows no significant differences between the two feeds.

H2: The development dynamic of *Clarias gariepinus'* body weight shows no significant differences between the two feeds.

H3: The development dynamic of *Clarias gariepinus'* condition factor shows no significant differences between the two feeds.

3 Material and Methods

3.1 Construction of the System

Figure 9 First setting of the floating elements © Filzwieser 2015

The aquaponics system was constructed on the compound of the Leibniz-Institute of Freshwater Ecology and Inland Fisheries (IGB), at Müggelseedamm 310, 12587 Berlin. The swimming system was placed in one of the fishponds behind the Aquarienhalle. The components used are shown in Table 3.

The system was placed on an island made of JETFLOAT floating elements. As seen in Figure 9, holes were included in the construction. These were used to put the fish tanks, as well as the sedimentation tanks, further down. This was necessary, as the construction had limited space in both horizontal and vertical directions resulting from a nearby greenhouse.

The distribution of the floating elements, shown in Figure 9, would later prove to not be optimal, and was therefore changed to dimensions of 7 by 3.5 metres.

At this stage, the island was fixed at the “Monk” in the fishpond, as it was not heavy enough to damage it. Later, it was fixed at two trees on opposite sides of the pond. The system would stay at this place for the rest of the testing period, as the Monk was also used as a way to physically access the system.

To provide the system with more stability, a wooden platform that was used as a fundament was constructed, as seen in Figure 12. The parts of the platform are listed in Table 3. Every part of the used wood was painted with three layers of water-resistant paint. The logs were connected with iron brackets and rustproof screws. This allowed for building a 2.5 by 6.0 metre structure. Afterwards, the plates were fixed at the logs with rustproof screws.

For the fish tanks, CEMO tanks with a volume of 550 litres were chosen. Each fish tank consisted of two nest able tanks. Between those tanks, 20 centimetres of Styrofoam was placed at the bottom and as much fitting foam that could fit in the structure as possible to use was used as insulation for the walls. The insulation was necessary as because the tanks were in constant contact with the pond water.

The sedimentation tanks were constructed from PVC plates with a thickness of 5 millimetres. To prepare the plates for PVC welding, the edges had to be cut at an angle. These angles were later used as the points of connection during the welding process. The sedimentation tanks were constructed with a light slope to allow the sedimentations to slip down to the deepest part of the tank without assistance. As with the fish tanks, the sedimentation tanks also had to be insulated. Following the same principle as the fish tanks, the sedimentation tanks were also planned and constructed as nestable constructions. In this case, only Styrofoam was used, as there was no difficulty reaching all the necessary areas with it. Between the bottom parts, 10 centimetres of Styrofoam was used and a 5-centimetre layer insulated the walls.

Figure 10 Biofilter © Filzwieser 2015

After all tanks were checked for leak tightness, they were added to the structure.

For the biological filtration, as shown in Figure 10, a rain barrel with a volume of 500 litres was used to make a “trickle filter”. In a trickle filter, biocarriers, as seen in Figure 11, are sprinkled with water through a spray bar and drained by gravitation. Necessary *nitrosomonas* and *nitrobacter* cultures establish on these biocarriers.

Figure 11 Biocarriers in trickle filter © Filzwieser 2015

Because of limited space, the filter was placed above the fish tank. This hindered accessibility to fish but the stability of the industrially-produced CEMO tanks surpassed that of the sedimentation tanks. Additionally, the CEMO tanks had much wider edges and were therefore better suited for additional structures.

The trickle filter was placed on a plate in a shape that left as much space as possible to reach the fish tank. This plate was made from the same wood as the fundament of the system and was also painted with three layers of water-resistant paint.

Table 3 Construction parts of the Aquaponics-system

Item	Dimensions/Volume	Performance	Amount
Greenhouse	250*600 cm	////	1
CEMO tanks	550L	////	4
Rain barrels, big	500L	////	3
Rain barrels, medium	30L	////	2
Rain barrels, small	25L	////	2
Material for biological filters	-	////	-
JETFLOAT floating elements	100*50*40 cm	////	47
Old Pumps for Aquaculture	////	4600 Watt	2
New Pumps for Aquaculture	////	6200 Watt	2
Sedimentation Tanks	~ 500L	////	2
Pumps for Hydroponics	////	2000 L/h	2
Pipe, s big	50 mm	////	-
Pipes, small	30 mm	////	-
Hoses	2 Inch	////	2
Pellet mill	////	-	1
Wooden plates	250*600*3 cm	////	-
Wooden logs	10 cm thick	////	-

To install the plate for the trickle filter, three threaded bars were installed on the edges of the fish tanks. The bars were kept in place with rust-free screw nuts. The same was done at the plate itself. This was a simple solution for fixing the plate and also allowed for a fast change in the height of the filter.

For the filter itself, several holes were drilled into the barrel: one at the centre of the bottom and several others around the barrel. The hole at the bottom was large enough that a pipe with a diameter of 5 centimetres could be inserted. This pipe was later used to transport the water from the filter back to the fish tank. The upper side of this pipe, on the inside of the barrel, had to let through water but keep out the plastic parts that were used as a biological medium. To do so, the upper side of the pipe was closed with a cap. Afterwards, small holes were drilled in the cap as well as the upper part of the pipe.

Short pipes with a 45-degree angle were inserted into the holes on the side. These holes were used so that air could leave the filter and therefore facilitate the flowing of water.

Figure 12 Construction of the systems fundament © Filzwieser 2015

At the centre of the lid, a 2-centimetre-wide hole was drilled to allow for the insertion of a pipe. This pipe was used to pump water from the sump into the filter. To reach an optimal amount of nitrification, the outlet of this pipe was rotating. Its movement was solely a result of water pressure.

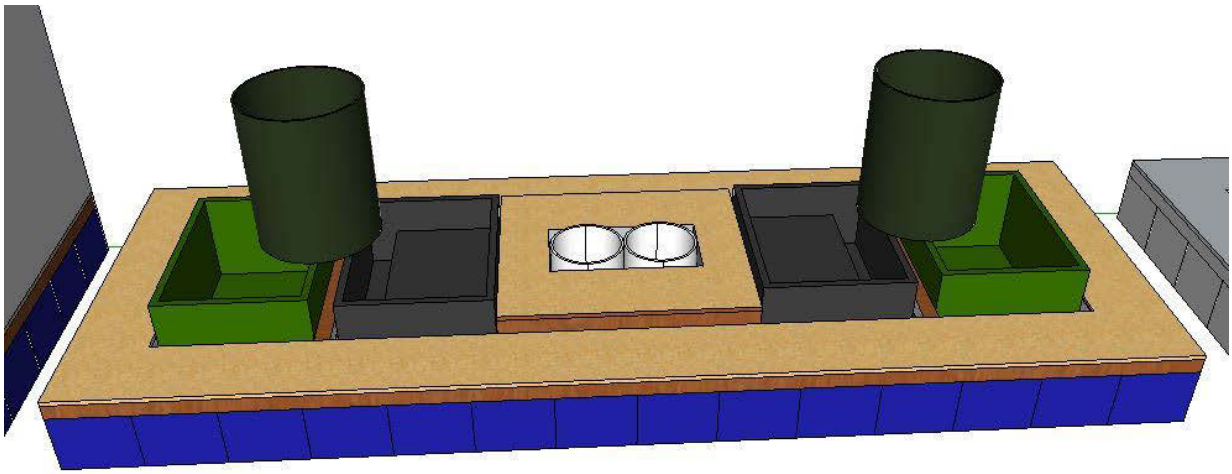


Figure 13 Sketch of the system, without hydroponics and greenhouse © Jeutner 2015

At the centre of this structure was a 50-centimetre-long threaded bar, surrounded by the aforementioned pipe. At the end of this pipe at 90 degrees was another pipe. This pipe had a diameter of 2 centimetres and was long enough to cover the length of the barrel. It was closed with caps on both ends. On both arms of the pipe, holes were drilled on one side, opposite to each other. The threaded bar in the middle was attached on the lower pipe, as well as on the top of the barrel's lid. To keep the structure in place, two screw nuts and two support plates were used at the top and the bottom of the threaded bar. The support plates' round sides faced each other and were enclosed by the nuts, which were tightened in each other's direction. The second nut was necessary, as without its use, the movement of the system would have caused the single nut to unscrew itself. The plates ensured a better movement altogether.

A 25-litre barrel was used as the sump. The sump is the lowest part of the system, which ensured that all water in the system could only get there through gravity, while water will be pumped out of it. The sump was placed in a hole in one of the JETFLOAT floating elements. The sump was also put further down to not take up too much space. A net for physical filtration was installed in front of the opening of the sump with a net size of 200 micrometres. Additionally, the sump was also where the 2000-watt heaters were placed – one for each subsystem.

All parts of the system were connected using pipes with a diameter of 10 centimetres. The pipe between the sump and filtration ran to a narrower pipe to heighten the water pressure for the rotation of the filter. The pressure turned out to be so high that the pipes were separated, requiring a hose to be put between the two pipes. The hose had a diameter of 2 centimetres and was fixed with a water hose clip on the narrow pipe.

The used pump had a power of 4600 watts, which turned out to not be strong enough to keep the biological filter at an adequate movement. A stronger one with 6200 watts subsequently replaced the pump.

A third-party company installed the 250 by 600 centimetre greenhouse. At its highest point, it was 2.5 metres high.

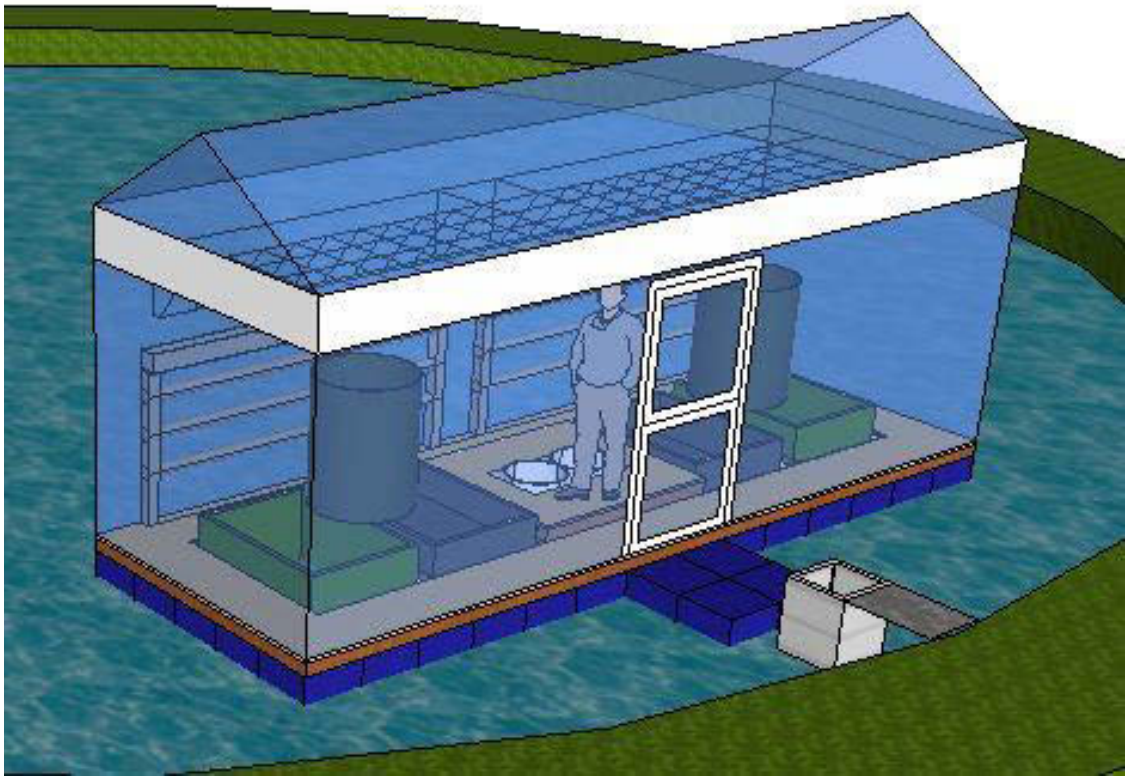


Figure 14 Sketch of the completed system © Jeutner 2015

For the hydroponics part of the system, a barrel with a volume of 30 litres was used as a water reservoir. The water was manually added from the aquaculture circle. A pipe with a diameter of 150 millimetres was used as a medium for the plants and was fixed on the upper part of the greenhouse. Eight holes with a diameter of 75 millimetres each were made and were also planned to house one tomato plant each. The water pump inside the reservoir was connected with the big pipe via a hole of 2 centimetres. The pipe for the plants was installed in a way that the water could return to the reservoir solely through gravity.

Above the whole system, a net of nylon ropes was installed to allow for further plant growth. Figure 13 and Figure 14 show sketches of the system at different stages of construction.

3.2 Physical and Biochemical Water Tests

Before starting the main biochemical testing period, drop tests from “Tetra” were used to ensure functionality of the subsystems. While drop tests were used, only nitrate and nitrite values were measured. These measurements occurred every Tuesday and Friday. These tests were continued until more precise testing began on the 1st of September 2015.

From this time on, biochemical testing was conducted at IGB’s Laboratory facilities at Müggelseedamm 301. Testing continued twice per week, always on Tuesdays and Fridays.

Table 4 shows a list of all monitored nutrients as well as the tools used for biochemical testing.

Physical tests of the subsystems' water included pH value, temperature and oxygen content. To do this, a testing probe was used.

Table 4 Observed nutrients and used Tools

Nutrients	Tool	Designation
K, Mg, Ca, Fe, Cu, Mo, Zn	ICP OES	ICAP 6000 Series – Thermo Scientific
Phosphorus	Spectrophotometer	Cary 1E UV-Visible Spectrophotometer
NH_4^+, NO2-, NO_3^-	FSA	Skalar
NO_3^-, SO_4^{2-}, Cl-	Ion chromatograph	Dionex ICS-2000
DOC, DN	DOC	Analytic Jena multi N/C 3100

Table 5 Nutrient composition of Control-feed

	ME-2 Meerval Start
Rohprotein (%)	49
Rohfett (%)	11
Kohlenhydrate (%)	19,8
Rohasche (%)	11
Rohfaser (%)	1,2
Phosphor (%)	1,6
Kupfersulfat (mg/kg)	3
Verdauliche Energie in MJ/kg	17,6
Vitamin A I.E./kg	10.000
Vitamin E mg/kg	150

3.3 Formulation of feed

To produce the feed, the "Alleinfuttermittel für Wels" was used as the basis for the control feed. The control feed was standard feed produced by the company, Skretting. The composition of the purchased feed can be seen in Figure 15 and Table 5. Some ingredients were not purchasable in time for the tests, while others were purposefully refrained from. The nutrient composition of the used test feed can be seen in Table 6.

40 kilograms of *Hermetia illucens* meal was purchased from Hermetia Deutschland GmbH. The

company specialises in the production of beneficial insects.

The Humboldt University of Berlin's Albrecht Daniel Thaer-Institute of Agricultural and Horticultural Sciences provided the wheat. The mill at the teaching and research facility at Berlin-Dahlem was also where the wheat was grinded. The grain size was 1 millimetre.

The fava beans were purchased at an online gardening shop, and came as whole beans. Therefore, they also had to be grinded at the teaching and research facility. As with the wheat, the fava beans were grinded into 1-millimetre grains. As an intermediate step, they were first ground into 4-millimetre grains.

Fish oil, specifically salmon oil, was purchased at a generic pet store.

A 20-kilogram free sample of gluten was provided by Skargill. It arrived as a meal and was therefore immediately ready for use.

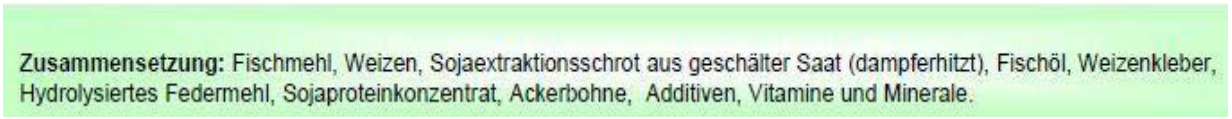


Figure 15 Components of control feed

Table 6 Nutrient composition of the test feed original substance (OS) and dry matter (DM)

Nutrient	g/kg OS	% in DM
Total	954	100,00%
Protein	489	51,26%
Lipids	109	11,43%
Ash	53	5,56%
Fiber	67	7,02%
Phosphor	9,3	0,97%

The desired outcome was for the control feed to have the same percentage of nutrients as the Skretting feed. Table 7 shows the combination of ingredients needed for the required nutrient combination for 10 kilograms of feed.

Two attempts were necessary to determine the correct formulation. In the first attempt, the percentage of gluten was too high and therefore the mass was too sticky making in unsuitable for the production of pellets.

The final produced mass was next placed in the pellet mill (machine-type SKM) from Alexanderwerk, which produced pellets with a size of 3 millimetres. The pellets were kept in several boxes, always in a single layer, and were air-dried for two days. Once dry enough, they were filled into several small plastic bags and kept at the cooling chamber of the IGB. The finished feed is shown in Figure 16.

Table 7 Combination of test-feed

Phosphorus (g/kg)	Crude fibre (g/kg)	Crude ash (g/kg)	Crude fat (g/kg)	Crude protein (g/kg)	Dry-matter		**Amount in kg**	**Percentage**
8,61	63,96	46,13	54,74	373,92	579,33	**Hermetia illucens meal**	6,15	61,5%
0,30	1,80	1,42	1,24	11,9,7	77,14	**Wheat**	0,89	8,9%
0,0	0,0	0,05	49,57	0,0	49,62	**Fish oil**	0,50	5,0%
1,08	14,16	6,15	2,66	44,20	149,35	**Fava bean**	1,72	17,2%
0,19	0,27	,72	1,88	60,0	69,75	**Gluten**	0,75	7,5%
10,18	80,19	54,46	110,08	490,09	925,19	**Sum in dry matter**	10	

3.4 Conduction of Feeding Tests

The system was separated into Subsystem A and Subsystem B. Both systems were stocked with 40 individuals of *Clarias gariepinus*. Before introducing the fish into the system, they were kept together in a tank inside the IGB's fish hall. The 80 test fish were chosen out of 120 purchased individuals. The subjects were chosen randomly in groups of five. For each group, every individual fish was weighed using an electronic scale with an accuracy of one decimal point. To ensure this, a bucket with a random amount of water was put on the scale. Before introducing the single fish, the scale was set to 0.0 grams. All further weighings were conducted with the same scale and method. The first group of five was selected for Subsystem A, the next for Subsystem B, then back to Subsystem A again, and so on. When this system led to an unsatisfactory distribution of weight, some fish were intentionally witched with each other. This led to an almost identical net weight of the fish in each system. Subsystem A contained 2077.9 grams of fish and Subsystem B had 2076.3 grams. The fish were introduced into the aquaponics system on the 4th of August 2015. Both systems continued to be fed using the standard Skretting feed, with an amount of three per cent of the net weight of the separate subsystems. This led to a daily feed amount of 62.3 grams per subsystem in the first week. The feeding test started on the 14th of August 2015. On this day, the fish were weighed and their lengths were measured. To measure length, the ruler in Figure 1 was used. Table 8 shows the measurements that were taken on the first day of the feeding test. Additional measurements are listed in the Appendix. From this day onward, the fish in Subsystem B were fed with the test feed described in Chapter 3.2, while the Skretting feed continued to be used for Subsystem A. In both subsystems, the amount of feed introduced per day was equal to three per cent of the net weight of fish. This percentage was chosen at the advice of

Figure 16 Test feed © Filzwieser 2015

the control feed's producer, as seen in Figure 17. For the feed introduction, an automatic feeder with a feeding duration of 12 hours was used. The feeding process was identical for both subsystems. The fish were always weighed and measured on Fridays and the amount of feed was kept the same until the next weighing. Regular visual inspection showed that fish in both subsystems accepted the fed feed very well.

Fütterungsempfehlung: tägliche Futtermenge in % vom Fischlebendgewicht :

Korngrößen	Durchm (mm)	Fisch-gewicht (g)	(Empfohlener Sauerstoffgehalt: 7 mg/l im Auslauf) Wassertemperatur (°C) 25-26
ME-2 Meerval Start	2,5	20-40	5
ME-3 Meerval Start	3	40-150	3,5
ME-4,5 Meerval 44-14	5	120-300 300-600 >600	3 2 1,5

Figure 17 Feed recommendation Skretting

Table 8 Weight and length measurements at beginning of feeding test

Subsystem A			Subsystem B		
Sequential number	**Length per fish [mm]**	**Weight per fish [g]**	**Sequential number**	**Length per fish [mm]**	**Weight per fish [g]**
1	190	53,7	1	143	23,5
2	180	44,7	2	220	74,1
3	200	67,1	3	195	53,8
4	180	51	4	210	63,2
5	215	68,9	5	205	68,5
6	200	66,8	6	190	61,9
7	230	100,9	7	230	103,6
8	195	61,1	8	190	51,4
9	215	78,5	9	180	42,3
10	208	66,4	10	195	53,3
11	170	36,9	11	180	40,5
12	245	116	12	190	49,3
13	160	34,3	13	210	70,3
14	205	79,9	14	180	38,3
15	200	57,2	15	185	47,8
16	160	26,3	16	175	40,4
17	210	74,1	17	215	76,5
18	195	47,1	18	190	55,9
19	205	62,2	19	200	60,7
20	190	50,4	20	215	72,9
21	185	49,2	21	205	61,7
22	180	43,7	22	180	35
23	192	45	23	180	43,9
24	190	48,4	24	210	66,3
25	188	62,8	25	185	46,9
26	188	40,5	26	180	38,5
27	188	56,3	27	240	99,9
28	192	48,6	28	190	49,6
29	190	47,8	29	170	30,8
30	185	48,9	30	215	72,4
31	185	48,9	31	215	58,1
32	208	65,7	32	155	26,4
33	173	36	33	195	51,5
34	150	27,2	34	190	58
35	188	58,2	35	190	51,6
36	162	35,9	36	210	66,6
37	185	47,7	37	200	51,1
38	190	49,1	38	195	53,2
39	195	50,4	39	160	35,1
40	195	65,6	40	230	105,7
40	**191,55**	**55,49**	**40**	**194,83**	**56,26**
Number of fish	**Mean length Subsystem A**	**Mean weight Subsystem A**	**Number of fish**	**Mean length Subsystem A**	**Mean weight Subsystem A**

3.5 Statistical Methods

During statistical analysis, several procedures were conducted that were used to test differences in the development dynamic of the two subsystems.

Observed data was adjusted for analysis by removing extreme values. This occurred for weight and condition factor with values above 275 grams and 1.5, respectively.

To determine the homogeneity of the characteristics' length, weight and condition factor influenced by time and fed feed, an independent samples median test, with a significance level of 0.05 was used.

Additionally, Pearson correlations were used to test the differences between the characteristics. These were again further split into measuring date and feed.

A regression analysis was constructed in several combinations. First, using length as the independent variable and using weight as the dependent variable. These were further grouped by measuring date and fed feed. The, second regression analysis used measuring date as the independent variable, in combination with weight, length and condition factor of fish as dependent variables. These were also grouped by fed feed.

For all statistical analyses, IBM SPSS Statistics 21 was used.

4 Results

4.1 Water Data

The highest risk for the well-being of the test subjects was shown by water temperature, ammonium and nitrite values. All other water-related factors only posed a secondary threat, as they did not have a direct influence on fish health. Physical water quality was adequate over most of the testing period, with temporary exceptions for temperature and pH. Biochemical values developed mainly as expected and reached adequate amounts soon after the introduction of test fish.

4.1.1 Physical Values

Tested physical characteristics of the water in the system consisted of temperature, oxygen content and pH value. Corresponding data is presented in the following chapter. All values for physical water data can be found in the Appendix.

4.1.1.1 Temperature

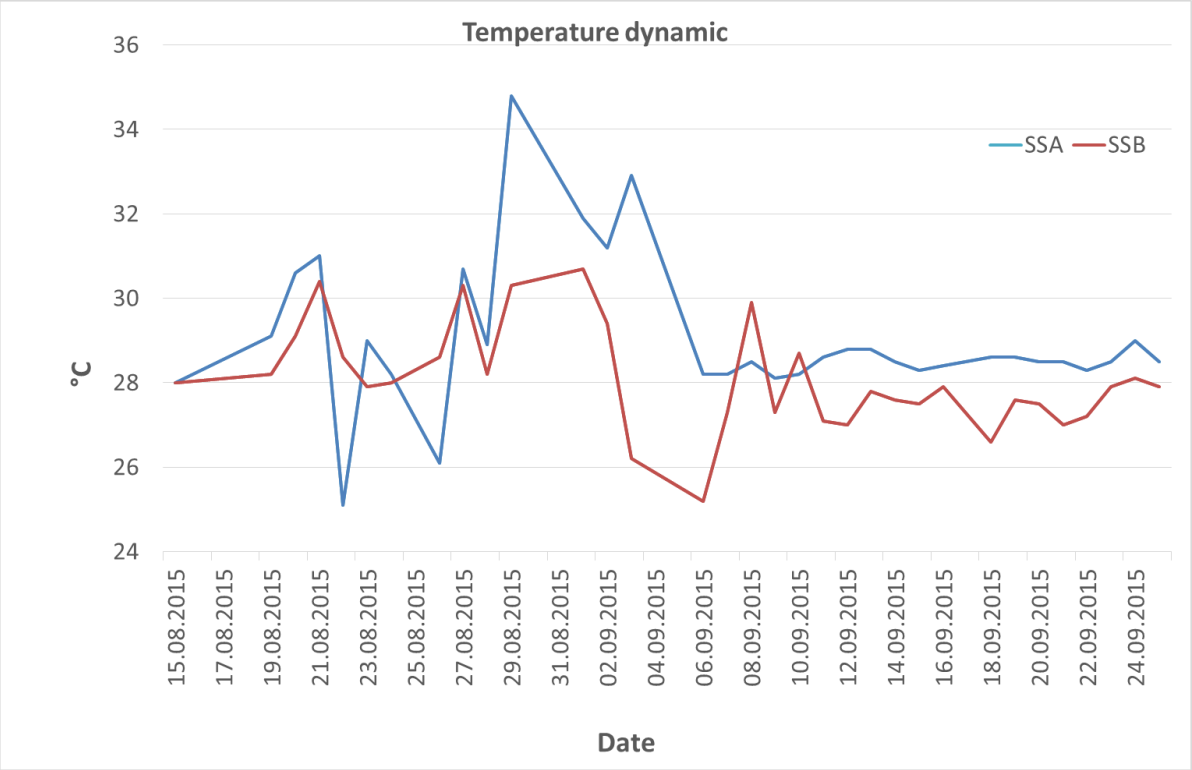


Figure 18 Temperature development of Subsystems A (SSA) and B (SSB)

Figure 18 shows the dynamic of the temperature for both subsystems. SSA shows a temporary rise above 30° Celsius, with one day reaching almost 35° Celsius. SSB, on the other hand, experienced a single day with a temperature a little below 25° Celsius. The

unusually high temperatures were caused by improper conduction of the heater in Subsystem A. Other drops, especially in SSB, resulted from differences in the regulation of the temperature, as the heaters' control devices were not precise enough. Over most of the testing period, however, the temperature was at stable range between 25° and 30° Celsius.

Table 9 shows the overall mean temperature and the day degrees of both subsystems.

Table 9 Mean temperature and day degrees in Subsystems A (SSA) and B (SSB)

	°C SSA	°C SSB
Mean Temperature	29,0	28,0
Day Degrees	958,6	927

This shows a difference of almost one and 31 degrees respectively. These differences are certainly influenced by the extreme values at the beginning of September and are therefore not applicable over the whole testing period. Nevertheless, the temperatures had an influence on feeding behaviour when these extremes occurred.

4.1.1.2 Oxygen

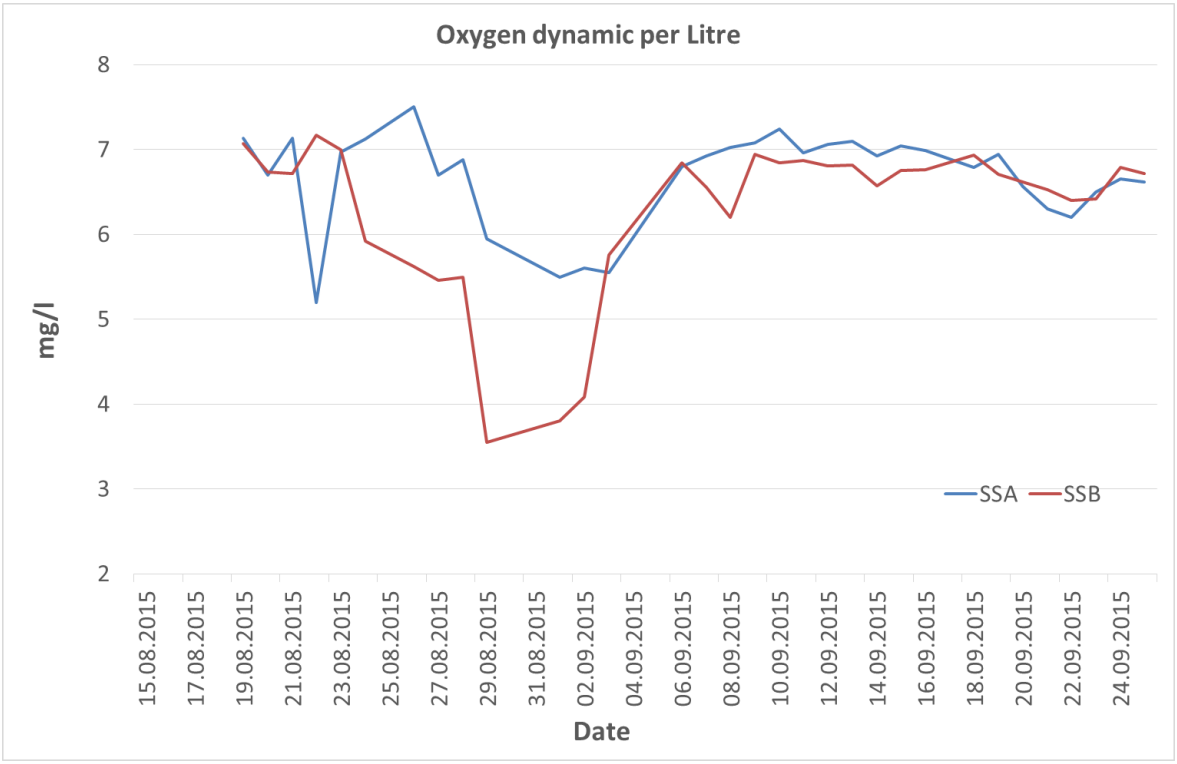


Figure 19 Oxygen development of Subsystems A (SSA) and B (SSB)

With the exception of a drop in late August, values for oxygen were generally stable, as seen in Figure 19. The values are around seven milligrams per litre and show a very similar dynamic for both subsystems. In the days before the sudden drop in oxygen on the 29th of August, the pumps' performance, started to drop – especially in Subsystem B, which led to

less effective water circulation. This again led to a lower amount of oxygen content in the water. In SSA, the final drop was very likely a result of the heightened water temperatures at the time. In SSB, the cause is mainly attributable to the malfunction of the pump. A normalisation of oxygen values started at the beginning of September, which was just after the installation of the new pumps and a resulting rise in water circulation.

4.1.1.3 pH Value

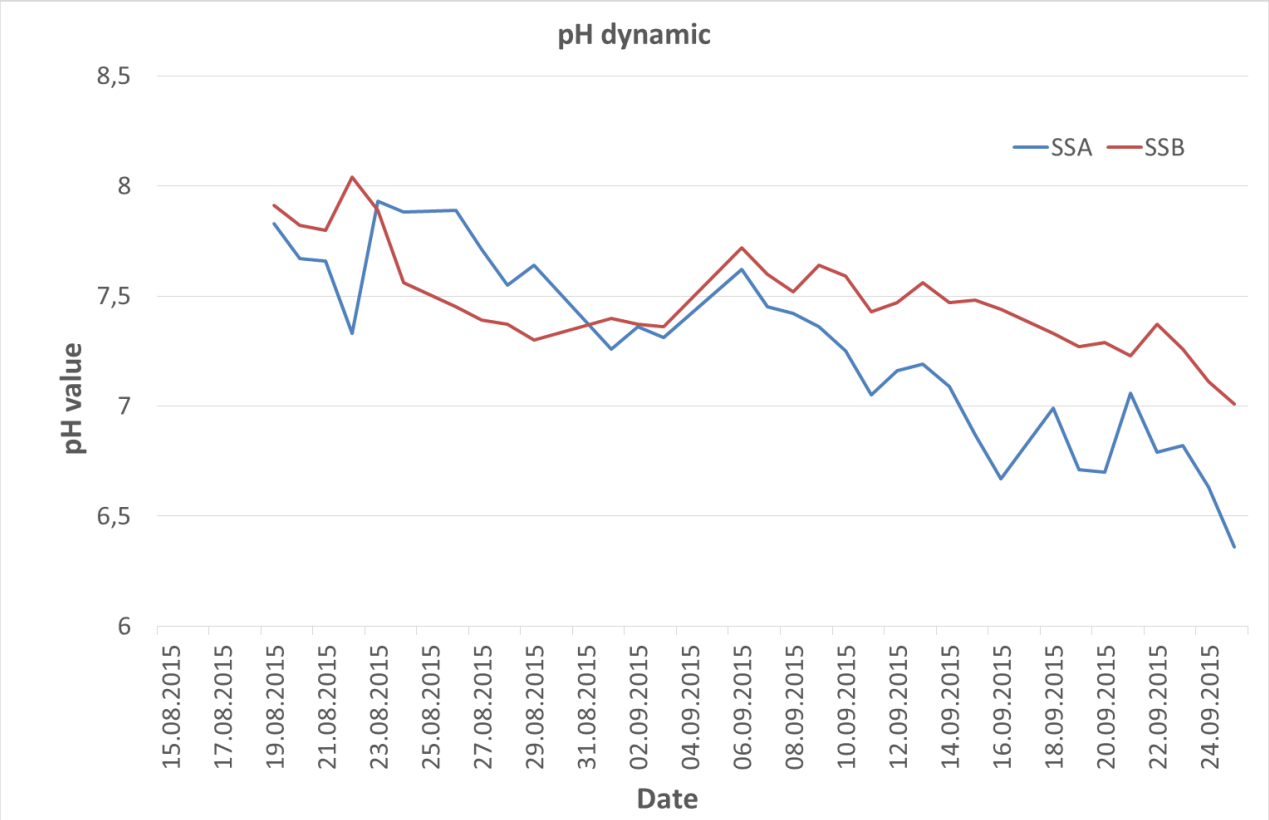


Figure 20 pH development of Subsystems A (SSA) and B (SSB)

The pH value, as seen in Figure 20, experienced a steady decline over the course of the project. At the beginning of testing, the values were roughly below 8 in both subsystems. At the last testing on the 25th of September, values of 7.01 for SSB and 6.36 for SSA were measured. The drop of the pH value was to be expected and is a regular occurrence. The faster drop in Subsystem A is most likely a result of the higher biomass and the accompanying faster nitrification. To counter this drop, irregular liming with 10 grams of chalk was conducted. All increases of pH were a direct result from this liming. As indicated by the more rapid decrease in SSA, it was also necessary to have higher rates of liming here. Only on the last day of testing did the pH value of this subsystem drop below the optimum range. However, the distance from the optimum is rather small, and the time at which it occurred

was late in the test period and had a short duration, so it is plausible that this had at most a negligible influence on the growth development of fish in the subsystem.

4.1.2 Biochemical Values

Table 10 Important biochemical value of Subsystems A (SSA) and B (SSB)

	NH_4^+ [mg/L]		NH_3[mg/L]		NO_2^- [mg/L]		NO_3^- [mg/L]	
Date	**SSA**	**SSB**	**SSA**	**SSB**	**SSA**	**SSB**	**SSA**	**SSB**
19.08.2015	0,063	0,063	0,003	0,003	9,854	9,854	-	-
21.08.2015	0,232	0,348	0,013	0,018	9,341	12,810	49,580	48,252
25.08.2015	0,063	0,063	0,003	0,003	0,319	1,981	-	-
01.09.2015	0,399	0,927	0,002	0,006	0,030	0,033	172,646	92,963
04.09.2015	0,541	0,644	0,003	0,003	0,030	0,033	203,633	119,524
08.09.2015	0,155	0,386	0,001	0,002	1,872	0,066	172,646	115,097
11.09.2015	0,322	0,373	0,002	0,002	0,030	0,033	234,621	128,377
15.09.2015	0,270	0,361	0,001	0,002	0,131	0,066	185,926	82,339
18.09.2015	0,695	0,554	0,004	0,003	0,030	0,033	273,577	97,390
22.09.2015	0,657	0,721	0,004	0,004	0,033	0,066	303,679	123,951
25.09.2015	1,829	0,889	0,010	0,005	0,033	0,066	271,806	113,326

Table 10 shows collected values of ammonium, ammonia, nitrite and nitrate in milligrams per litre.

Biochemical values were adequate after some time and left no cause for concerns. The only irregularity was the difference in nitrate development between the two subsystems. Nitrite reached its desired values after a few days. These values were first reached in Subsystem A on the 25th of August and in Subsystem B on the 1st of September.

4.1.2.1 Ammonium

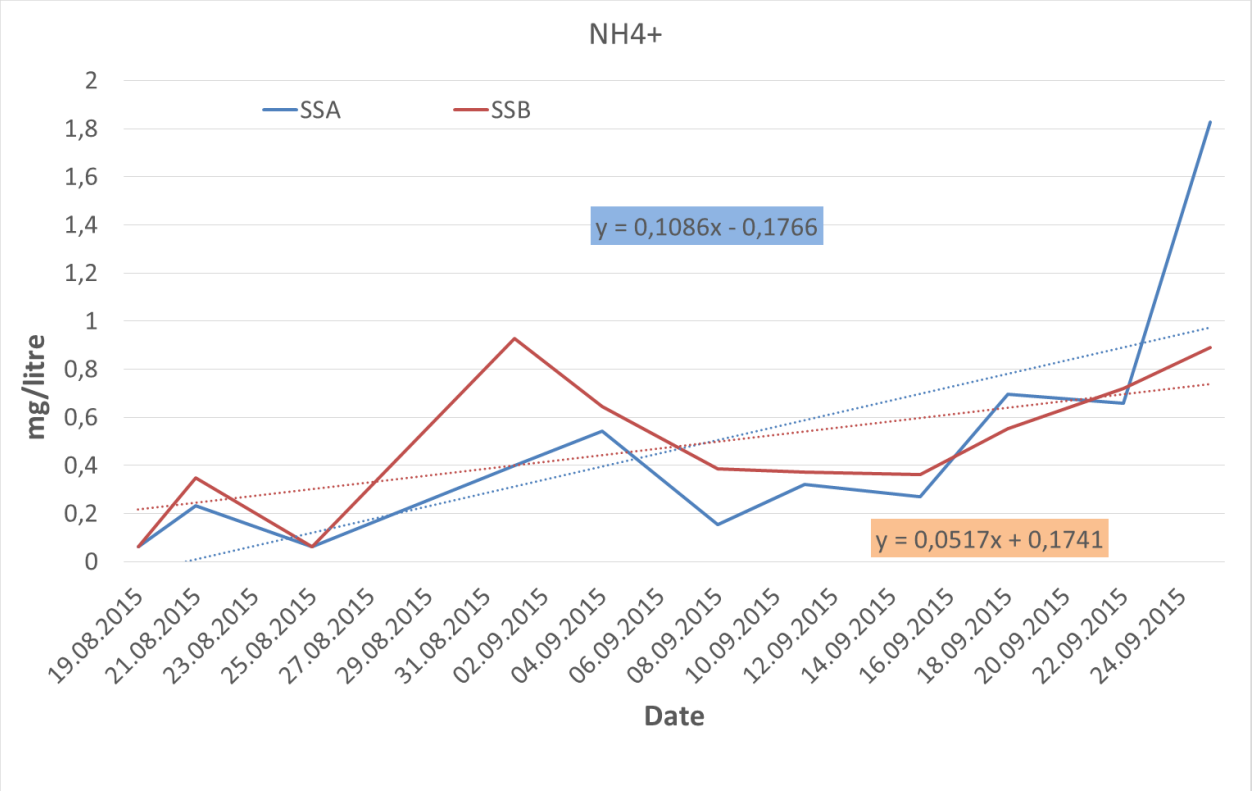


Figure 21 Ammonium development of Subsystems A (SSA) and B (SSB)

Figure 21 shows the development of ammonium, which is characterised by a steady rise in both subsystems that were likely corresponding to the gain of total biomass. The overall increase in SSA is steeper, but this is likely influenced by the sudden rise at the end of the testing period; the reason for this sudden rise is not clear. The values were in a non-threatening range at all times.

4.1.2.2 Nitrite

A sudden drop, as seen in Figure 23, characterised the development of nitrite after several days. This drop resulted from the establishment of nitrosomonas cultures in the system's biological filtration. Once the values were below 0.05 milligrams per litre, they were very constant. Subsystem A had two irregularities in which values of 0.1 and even 1.8 milligrams per litre were observed. Both values did not result in any obvious events and might be measurement failures.

4.1.2.3 Nitrate

Values for nitrate showed the biggest differences between the two subsystems. These differences were not only present in the amounts of measured NO_3, but also in its development. Subsystem A had higher total values in comparison to Subsystem B, which

contained the test group. These higher values were observable from almost the beginning of the testing. In addition to the difference in amounts of nitrate, the nitrate development showed also high deviations. Subsystem A clearly experienced faster increase, as seen in Figure 23.

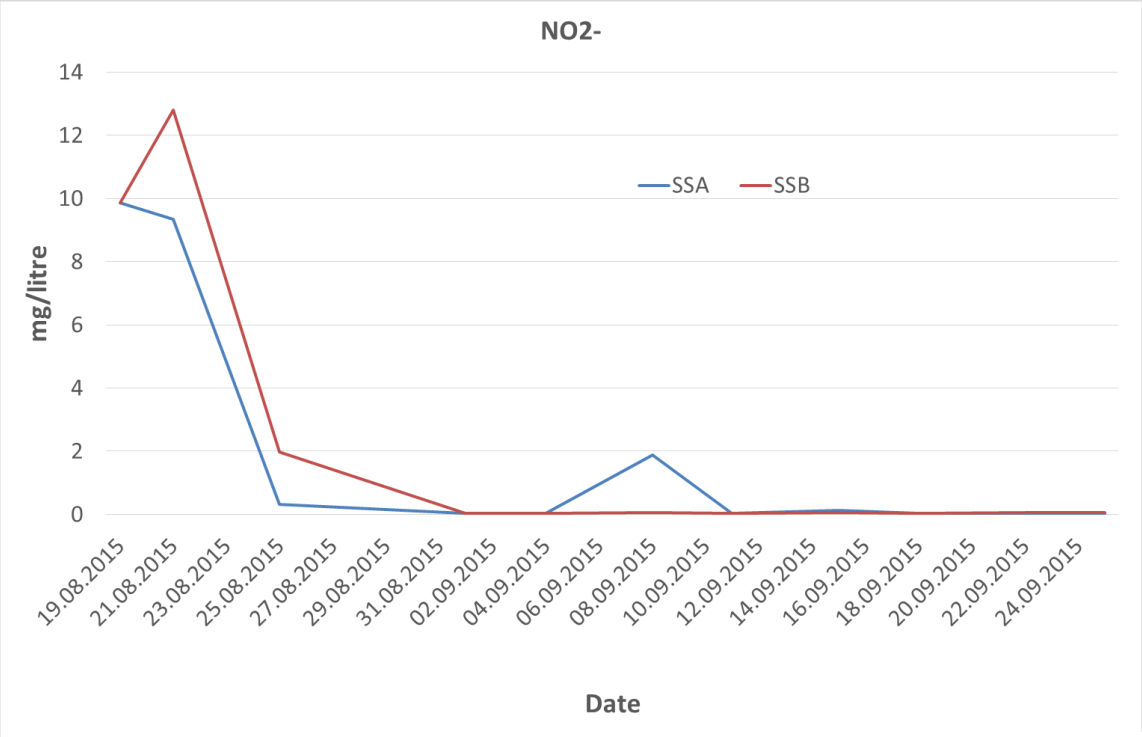


Figure 22 Nitrite development of Subsystems A(SSA) and B (SSB)

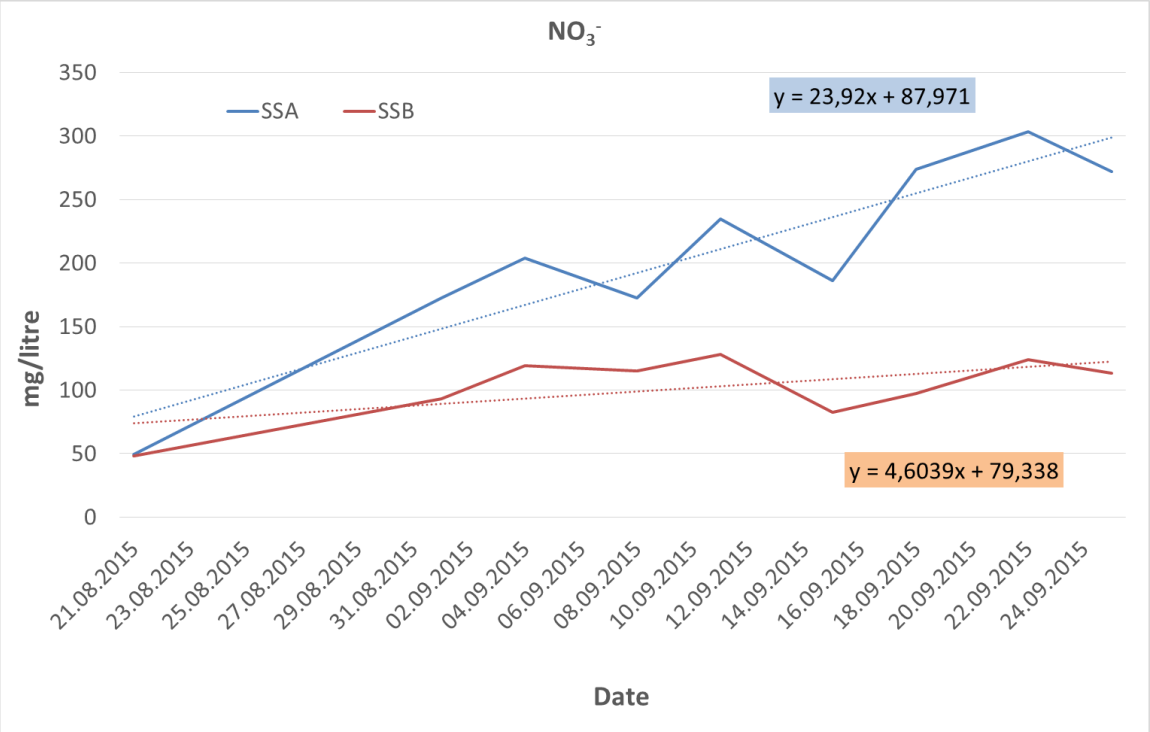


Figure 23 Nitrate development of Subsystems A (SSA) and B (SSB)

4.2 Development Dynamic

Observed indicators for development dynamics were length and weight of fish. To further examine the results, mean length and weight were calculated grouping by measuring date and feed. Additionally, standard weight development of fish, considering all weights and lengths over the time of testing, was calculated along with the mean condition factor according to Fulton. Both feeds reached satisfactory results according to development dynamics of *Clarias gariepinus* during testing.

4.2.1 Length

Over the course of the six-week testing period, both systems showed constant length growth. Figure 24 shows the mean length development over the whole testing period for both subsystems.

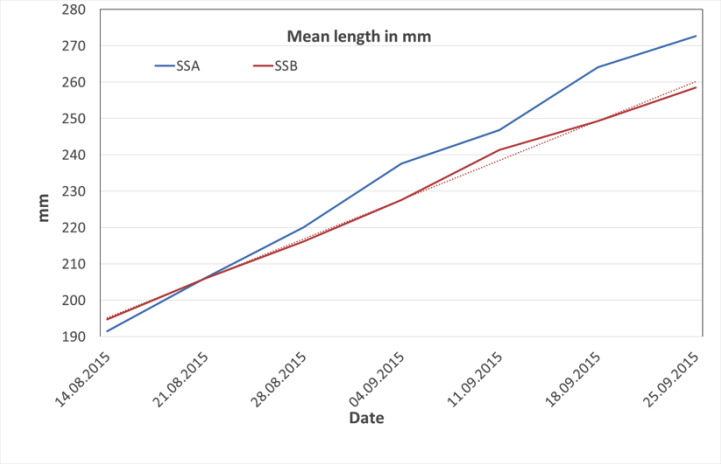


Figure 24 mean length developments of Subsystems A (SSA) and B (SSB)

It shows that the length development of fish fed with the control feed K reached higher values than fish fed with the test feed T over the period of testing. At the beginning of testing, Subsystem B had a slightly higher mean length. After one week, the values were identical. After two weeks, the mean length difference between the two subsystems was 4 millimetres, with Subsystem A at the higher value. Over the following weeks, the difference increased and decreased, but the values for Subsystem B did not overtake those of SSA again.

Figure 25 shows the corresponding development trends for both subsystems. SSA shows a more rapid increase then SSB, with the difference between the two at roughly 3 millimetres per week.

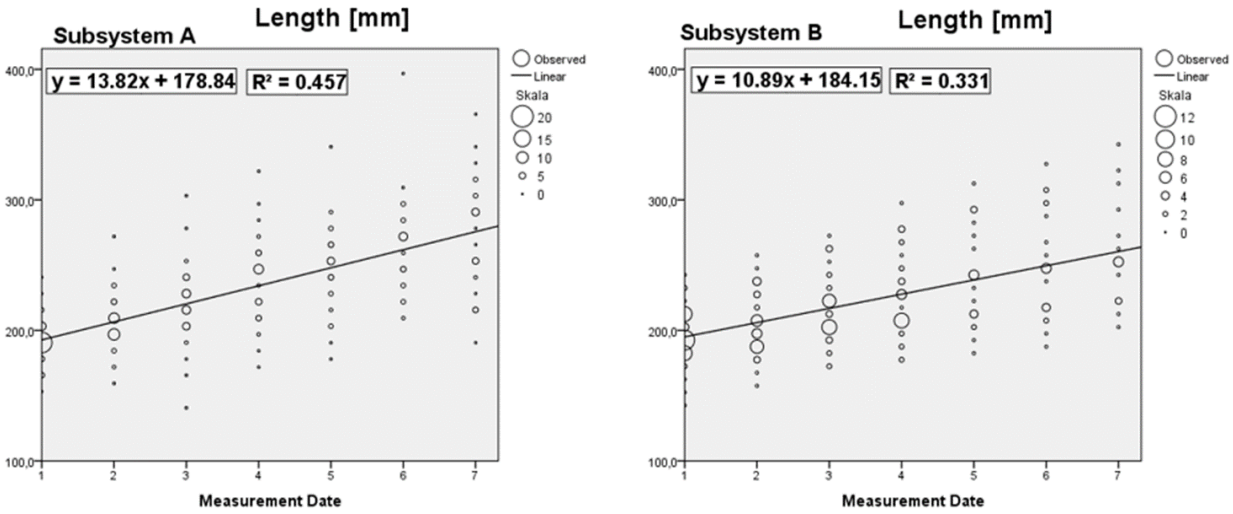
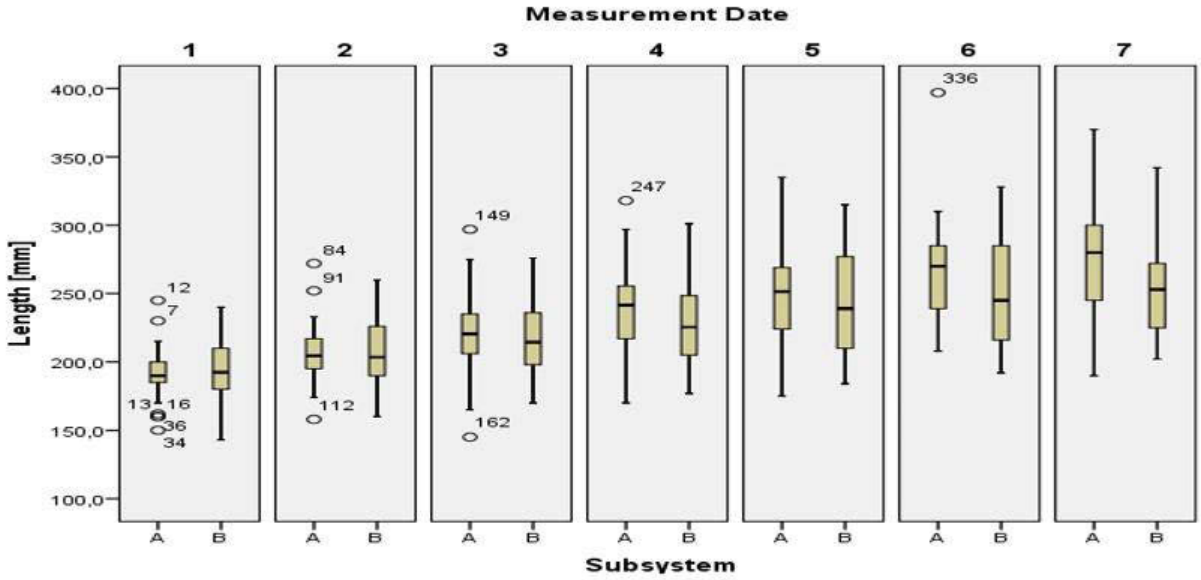


Figure 25 Development trends for length in Subsystems A and B

Figure 26 shows the development of the distribution in lengths of fish for both subsystems. The spread of values grows over time and neither of the two subsystems seems to spread significantly wider than the other one. From the p-values that originated from the independent samples median test, it is apparent that there are no significant differences between medians for lengths in SSA and SSB.



Chi²:	0.05	0.06	0.24	1.56	0.37	1.67	1.22
P:	**0.823**	**0.808**	**0.627**	**0.211**	**0.545**	**0.196**	**0.269**
Median total:	190	204.5	218	231	242	253	257

Figure 26 Distribution and significance of differences in medians for length in tested fish

4.2.2 Weight

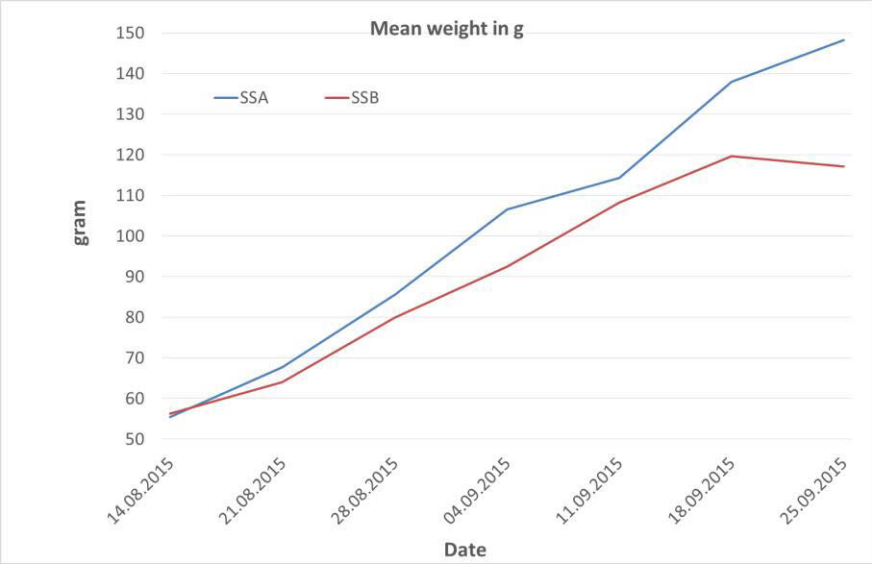


Figure 27 mean weight developments of Subsystems A (SSA) and B (SSB)

The development dynamic for the mean weight of the two subsystems in Figure 27 shows a similar situation as seen in the length dynamic of Figure 24. At the start of the feeding test, the values were almost identical but from week two onward, the values of SSA were higher. A constant increase in difference between the two subsystems started to decline after three weeks. This decline in difference lasted for one testing period and then changed again in an abrupt increase in difference.

The drop between the last two measurements in Subsystem B is the result of a statistical inconvenience. During measurements on the 18th of September, there was a single fish that was close to the threshold of statistical inclusion, but surpassed the threshold on the 25th. Therefore, it appears as if there was a decline in overall weight, when in reality this did not happen.

In Figure 28, the development trends for weight in the subsystems is illustrated. As with length, Subsystem A shows a steeper increase of values. Here the difference is at 4 grams

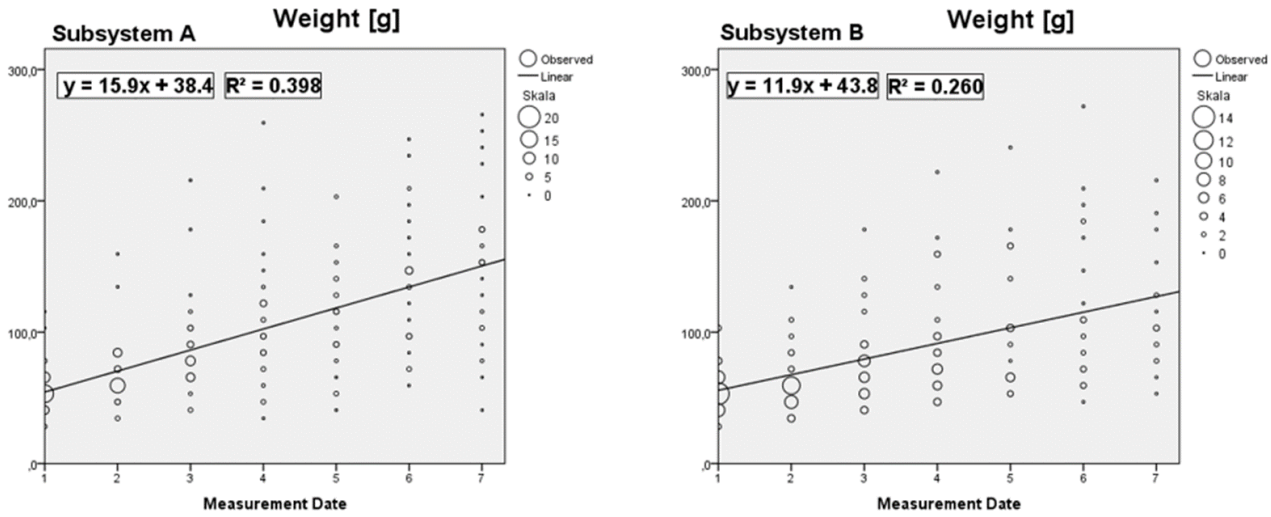
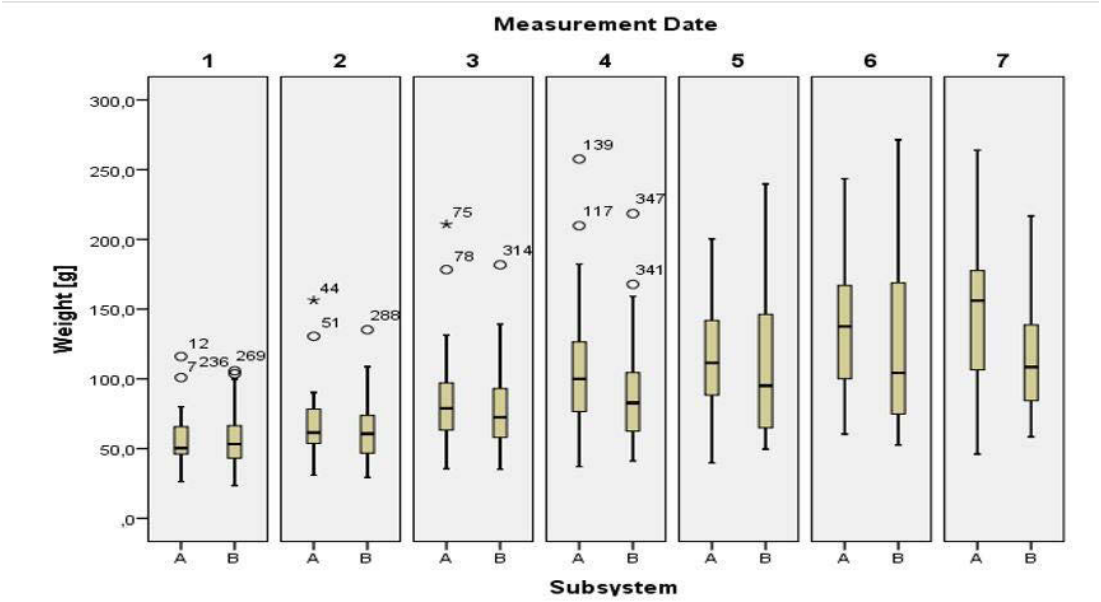


Figure 28 Development trends for weight in Subsystems A and B

As with the values in length, Figure 29 shows that distribution of weights increased over time. This distribution also appears to develop with roughly the same characteristics for both subsystems. According to the conducted independent samples median test, significant differences in medians occur after five weeks of testing.



Chi²:	0.45	0.06	1.47	1.56	1.67	3.98	4.38
P:	**0.502**	**0.808**	**0.225**	**0.211**	**0.196**	**0.046(S)**	**0.036(S)**
Median total:	51.55	61	77.65	90.1	104.8	121	124.7

Figure 29 Distribution and significance of differences in medians for weight in tested fish

4.2.3 Standard Weight

As established in Chapter 1.9 and as cited from Khaironizam & Norma-Rashid (2002), standard weight in fish is not only used to estimate weight in fish populations but can also describe its growth development. When using the formula $\mathbf{W = a\ L^b}$, b is the slope of the established trend line. Slopes at 3 describe isometric growth, while values below or above are indicators for negative and positive allometric growth. As can be seen in Figure 30, both feeds led to developments close to

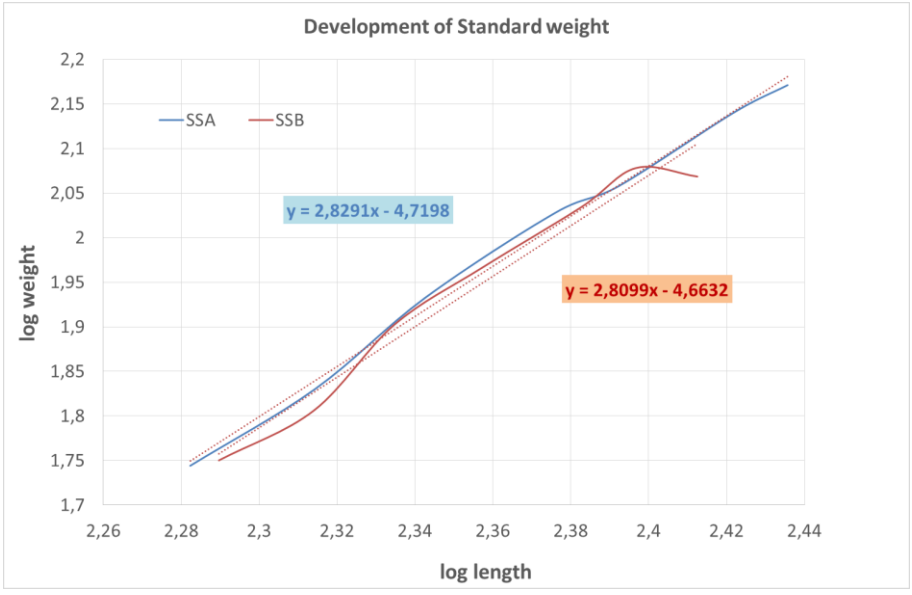


Figure 30 Development of standard weight of fish in Subsystems A (SSA) and B (SSB)

isometric growth. This is likely a result of the young age of the fish, for which isometric growth is not unusual. This can be seen in different other studies like Okogwu (2011) and Yalcin, Solak, & Akyurt, (2002). The failure during the last measurement is responsible for the drop observable in the graph for test feed.

4.2.4 Condition Factor

The development of the mean condition factor displayed in Figure 31 is also characterised by fluctuations over the course of the testing period for both development curves. Again, at the last week of testing, an erratic divergence occurs. A slight decline is observable.

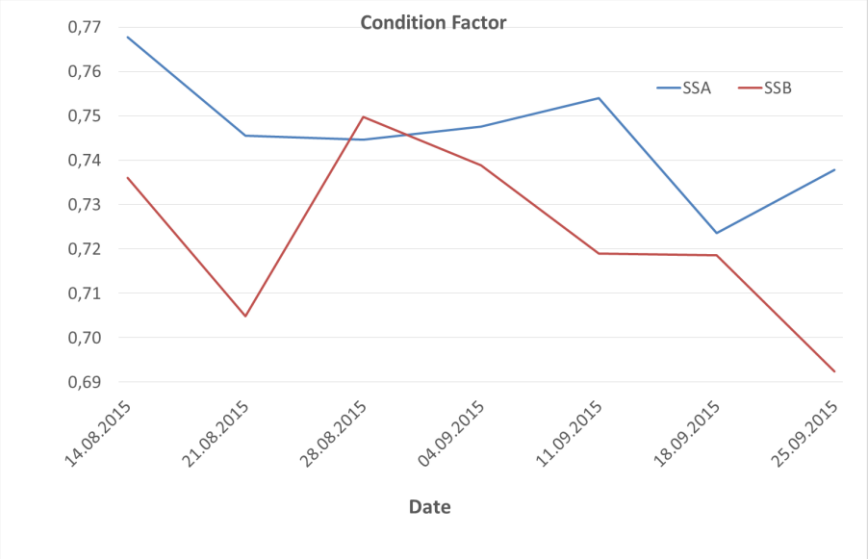


Figure 31 Development of mean condition factor of Subsystems A (SSA) and B (SSB)

This decline occurs in both subsystems and is slightly higher in SSA. As the decrease is so low, and the difference between the two is even smaller, it is unlikely that they have significant meaning. Therefore, the total decrease can be assumed as zero, which would mean that neither an increase nor a decrease in fish fitness occurred over the period of testing. The almost identical rates of development of condition factors allows for the assumption that both feeds had the same effects on overall fitness of fish. For further illustration, Figure 32 shows the development trends in Subsystem A and Subsystem B.

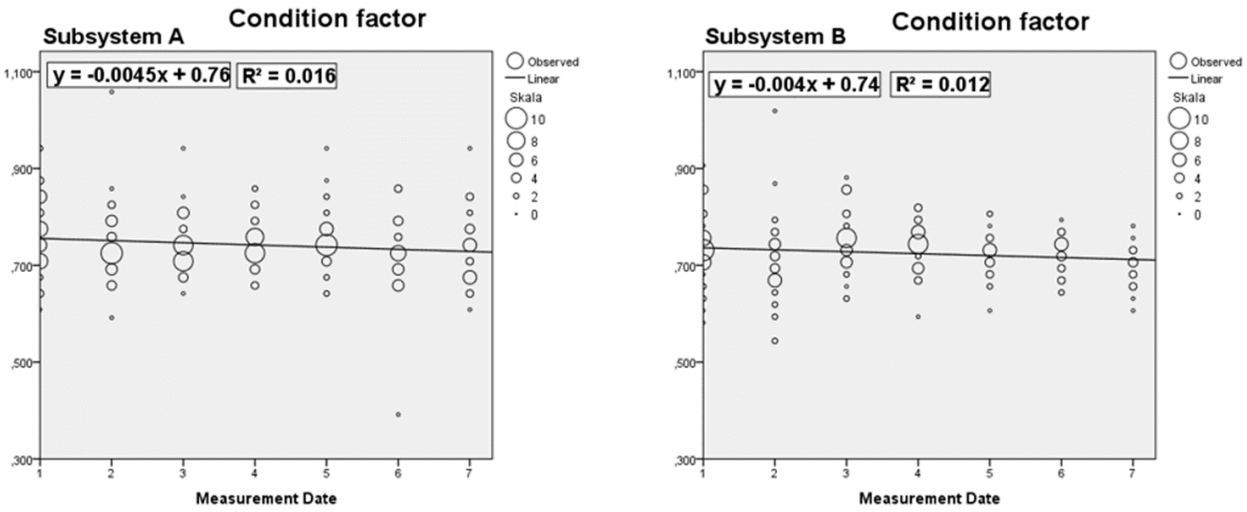
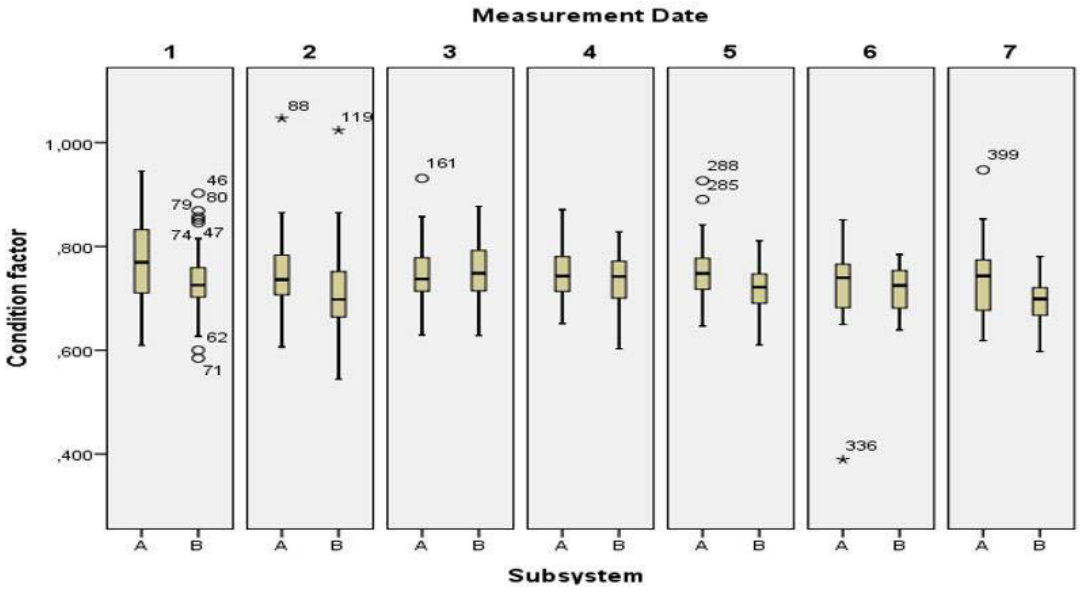


Figure 32 Development trends for condition factor in Subsystems A and B

In contrast to lengths and weights, the condition factor of fish did not spread further over time. As can be seen in Figure 33, the distribution stays fairly similar over the course of testing. Differences in medians can be observed at the second and fifth measurements.



Chi²:	2.45	4.77	1.81	0.06	3.86	0.08	1.49
P:	**0.118**	**0.029(S)**	**0.178**	**0.803**	**0.49(S)**	**0.78**	**0.22**
Median total:	0.75	0.72	0.74	0.74	0.73	0.73	0.71

Figure 33 Distribution and significance of differences in medians for condition factor in tested fish

5 Summary and Discussion

5.1 Construction of the System

Neither in scientific literature nor from other sources was it possible to find any records of other floating aquaponics systems. Therefore, the process had to be oriented by common terrestrial systems. As expected, construction on water was quite different from construction on land. First, limited space – both horizontal and vertical – was a problem. This problem mainly arose from the limitations of the greenhouse, which measured 2.5 metres wide and 2 to 2.5 metres high at its highest point. Therefore, it was necessary to plan the system in a way that ensured it was very compact while still offering enough space to enable daily procedures. Figure 34 shows the spatial limitations of the system.

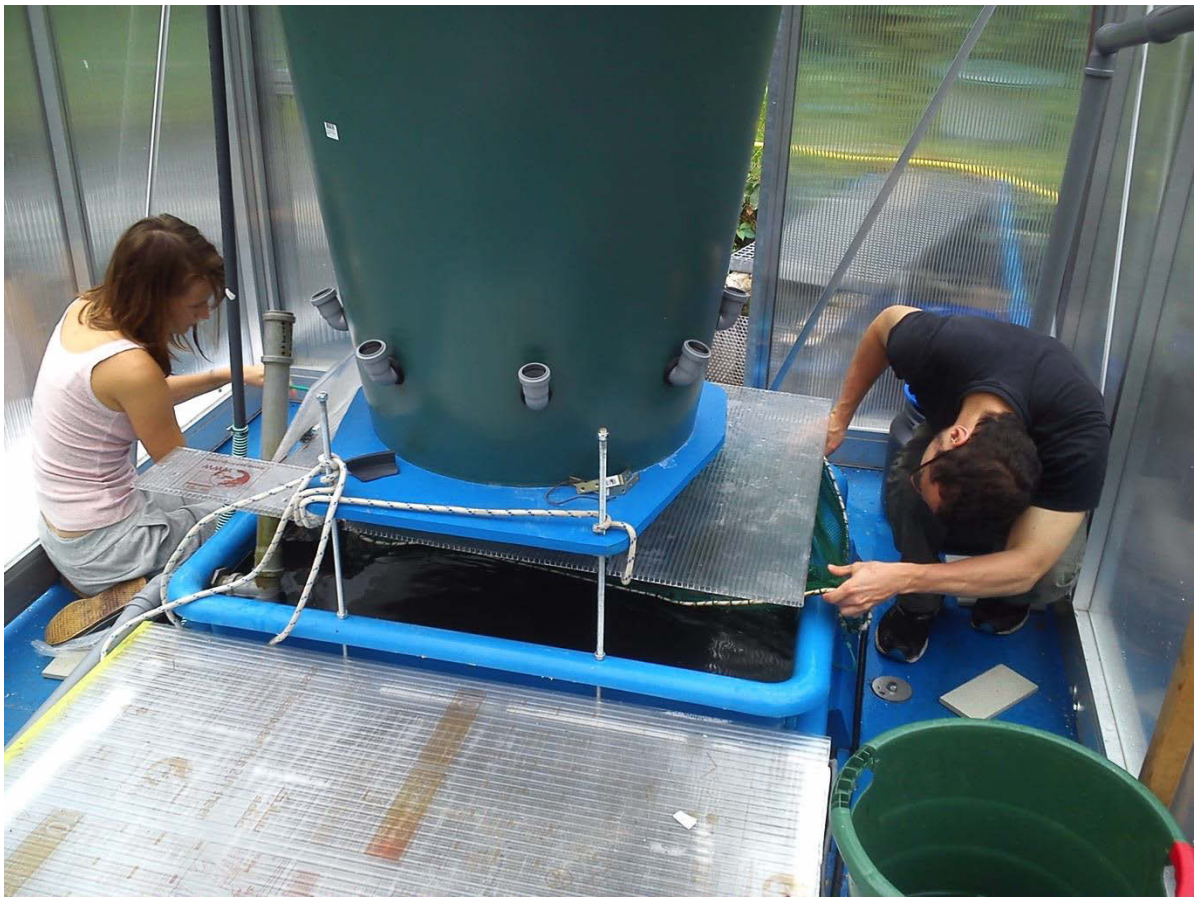


Figure 34 Withdrawal of fish for measurements © Filzwieser 2015

As mentioned in Chapter 3.1, holes were left out to help sink fish tanks and sedimentation tanks. This was mainly done to enlarge the vertical span of the system, but it was also used to ensure gravitational transport of water. As the tanks were below water level, they had to be isolated to ensure constant water temperatures in the system. Styrofoam was chosen as the main material as it is cheap, easy to handle and obtains satisfactory results. Fitting foam was only used in areas where Styrofoam couldn't fit because of space limitations. Another

main issue was stability of the system. On their own, the floating elements did not ensure enough stability to enable a trouble-free operation of the system; therefore, the wooden platform was constructed. It made fixing the single components of the system possible and also reduced the effects of water-induced movements. Both of these effects were of high importance for the greenhouse, as water movement could have compromised its integrity and its relatively big surface made it delicate to the wind.

The wooden platform was built in separate parts for two reasons. First, logs in the necessary length that would have covered the whole platform length were not available at the time. Ordering them would have resulted in a few weeks of waiting time. Second, the smaller parts made it possible to build them on land and then carry them onto the floating elements to later connect them there. As it turned out during construction, the shorter logs, combined with the non-symmetrical distribution of the floating elements, led to balance problems of the system. When the first tank was tentatively filled with water, it showed that the “island” had a slope from both ends to the middle. This problem could be solved by lifting the logs using screw clamps, a steel beam and by reinforcing the connections between the logs with iron plates that were fixed using several rustproof screws. Additionally, the floating elements were reorganised into a more symmetrical order. These measures were sufficient to deal with the limitations of the wooden platform.

To reach an optimal amount of surface in the biological filtration, the filter was designed with a spinning water sprinkler. The biological filter was also adjustable in height on the lid of the filter as well as through the plate on which it was placed. This allowed for altering the height difference between the lowest and the highest point of the system according to pump performance. While these measures were sufficient at the beginning of the system operation, it was later on necessary to implement pumps with higher power to keep the system efficiently working. Of further interest is that Subsystem A showed less technical problems then Subsystem B. Up to this point it, is not certain why the performance of the first pumps was eventually insufficient, nor where the difference of performance between the Subsystems originated, as both subsystems were built identically. A possible explanation can be found in the chronology of the system’s construction. SSB was constructed first and the lack of building experience might have had an influence on its performance.

5.2 Adequacy of test feed compared to control feed

Both feeds were very well accepted by their corresponding group of fish. This was to be expected for the control feed, however the palatability of the *Hermetia illucens* meal had not been tested before. The test group did not hesitate to consume the new feed and adjusted to it from the first feeding. This allows for the assumption that regarding palatability, the test

feed was at least satisfying. Also the size of the pellets, at three millimetres, was chosen as a size that could be consumed by all fish.

The stability of the feed was tested before introducing it to the system. It was able to contain its form for an adequate amount of time but had a considerably shorter period of stability compared to the control feed. Times of stability are around at least a day for the control feed and roughly an hour for the test feed. This might also be a result of the floating potency of the control feed, which the test feed did not have. These differences might have had an effect on the amount of consumed feed during testing. Another possible outcome regarding the feed's stability was the coloring of the water in Subsystem B, as seen in Figure 36. Over the testing period, the water in SSB turned the colour of the *Hermetia illucens* meal. This also hindered visual observation further into testing. It was, however, possible at all times to observe fish consuming feed.

As it was suspected that the pellet mill might agglutinate with fine material, it was decided to grind fava beans as well as wheat only to a grain size of one millimetre, shown in Figure 35.

Figure 35 Final grain size of fava bean © Filzwieser 2015

During production of the pellets, this concern was disproven. Other ingredients of the feed had considerably smaller grain sizes and the mill had no trouble processing them. The large grain size, on the other hand, might have led to other problems. It was observable that some traces of fava beans were left behind at the bottom of the tank after feeding. It is not clear if these were remainders of unstable feed or if the fish did not accept them. A non-acceptance could be a result of non-palatability of fava beans, but also the grain size. Unfortunately, it is not possible to determine if either were the cause. In the leftover parts of the feed, however, at least a certain amount of fava beans and therefore its corresponding nutrients, were not consumed by the test fish in Subsystem B. This is a point that must be reconsidered in interpreting the development dynamics of the fish.

Figure 36 Water samples of Subsystems A and B © Jeutner 2015

Additionally, it was relinquished to use any vitamin or mineral substitutes for the feed. It was, however, assumed that the combination of vitamins and minerals are sufficient without it.

5.3 Losses in the systems

Table 11 Number of individuals over time of testing in Subsystems A (SSA) and B (SSB)

Date	Number of individuals	
	SSA	SSB
14.08.2015	40	40
21.08.2015	34	34
28.08.2015	34	34
04.09.2015	32	32
11.09.2015	30	22
18.09.2015	29	22
25.09.2015	30	17*

* When emptied 21 individuals were found

Over the course of testing, Subsystem B showed much higher losses of individuals then SSA, as shown in Table 11. With the exception of one skull, it was not possible to find any remnants of dead fish in the tanks. However, two dead fish were found outside of the tanks. Therefore, two main theories are plausible about the losses.

First, as water levels in the tanks were rather

high it is possible that fish escaped the tanks in higher numbers, despite the weighted lid on the tanks. As the system was floating, these fish may have escaped into the pond below. Second, because of differences in sizes, the biggest individuals were drawn to cannibalism and consumed the smaller individuals without leaving any leftovers. As it is challenging to prove either of these theories, it is not possible to determine where the missing fish went.

Regardless of the reasons, the heightened losses in Subsystem B between the 4th and 11th of September are especially interesting. This could be a sign of heightened cannibalism in this time period, but also for careless handling of the closure of the tank.

5.4 Characteristics of Development Dynamic and Possible Influences

Both feeds that were used in the feeding test contained their main protein source as the main ingredient. Therefore, it is assumed that most of the differences in performance between the two feeds are also a result of the different sources of protein. Additional differences might occur from discrepancies in quality of feed production as well as quality and processing of ingredients. Additionally, the different stability of the feeds might have had an influence on intake availability. Other non-feed related reasons might also have had an effect on development dynamics.

5.4.1 Development Systematics

As seen in Table 12, the analysation of the data shows a high positive correlation between weight and length of test subjects. Therefore, fish with a higher weight are also longer.

Table 12 Pearson correlation of mean weight, mean length and mean condition factor

		Length_mm	Weight_g	Condition Factor
Length_mm	Pearson Correlation	1	,955**	-,031
	Sig. (2-tailed)		,000	,521
	N	430	424	427
Weight_g	Pearson Correlation	,955**	1	,152**
	Sig. (2-tailed)	,000		,002
	N	424	424	422
Fultonindex	Pearson Correlation	-,031	,152**	1
	Sig. (2-tailed)	,521	,002	
	N	427	422	427

**. Correlation is significant at the 0.01 level (2-tailed).

The condition factor as the third characteristic, which results from the two first characteristics, shows no correlation to length and only a marginal one to the weight of the fish. This probably means that the condition factor cannot be judged solely by weight or length, but rather in combination with each other.

Table 13 and Table 14 show that correlations are also the same for fish fed with the different feeds. This allows for the assumption that the two feeds have no difference in their influence on the systematics of the fish growth.

Table 13 Pearson correlation of mean weight, mean length and mean condition factor in Subsystem A

		Length _mm	Weight _g	Condition Factor
Length_mm	Pearson Correlation	1	,947**	-,065
	Sig. (2-tailed)		,000	,333
	N	229	224	226
Weight _g	Pearson Correlation	,947**	1	,137*
	Sig. (2-tailed)	,000		,041
	N	224	224	222
Condition Factor	Pearson Correlation	-,065	,137*	1
	Sig. (2-tailed)	,333	,041	
	N	226	222	226

**. Correlation is significant at the 0.01 level (2-tailed).

*. Correlation is significant at the 0.05 level (2-tailed).

Table 14 Pearson correlation of mean weight, mean length and mean condition factor in Subsystem B

		Length _mm	Weight _g	Condition Factor
Length _mm	Pearson Correlation	1	,966**	-,031
	Sig. (2-tailed)		,000	,657
	N	201	200	201
Weight_g	Pearson Correlation	,966**	1	,129
	Sig. (2-tailed)	,000		,068
	N	200	200	200
Condition Factor	Pearson Correlation	-,031	,129	1
	Sig. (2-tailed)	,657	,068	
	N	201	200	201

**. Correlation is significant at the 0.01 level (2-tailed).

A further in-depth inspection of the statistical results substantiates these claims. The correlations show variations depending on the measuring date, but they still tend to have similar results when comparing the feeds. These results can be found in Table 15.

Table 15 Correlation of length, weight and condition factor of tested fish in the subsystems over time

Date	Subsystem	Length- Weight	Length-Condition	Weight-Condition
14.08.2015	A	0,930**	0,033	0,023
	B	0,936**	0,039	0,348*
21.08.2015	A	0,946**	-0,026	0,233
	B	0,935**	-0,117	0,196
28.08.2015	A	0,870**	0,165	0,379*
	B	0,960**	0,301	0,497**
04.09.2015	A	0,963**	0,304	0,457**
	B	0,968**	0,051	0,233
11.09.2015	A	0,960**	0,135	0,358
	B	0,978**	0,01	0,161
18.09.2015	A	0,924**	-,377*	-0,018
	B	0,977**	0,082	0,234
25.09.2015	A	0,889**	0,207	,504**
	B	0,986**	-0,035	-0,082

* Correlation is significant at the 0.05 level

** Correlation is significant at the 0.01 level

5.4.2 Differences in Development Dynamics in Subsystems A and B

Chapter 4.2 displays a trend for all development characteristics. Control feed outperforms test feed after a while and the difference stays relatively constant. An independent samples median test of the data shows that these differences are generally not significant. A significant difference across development dynamics under reconsideration of different feeds is detectable for the condition factor, but without indication of regularity. These results are unlikely to show a trend, as the corresponding dates are distributed along the testing period. More specifically, the values were observed after one and four weeks of testing, with significances of 0.029 and 0.049, respectively. The second value was just under the threshold of significance. Additionally, the level of significance did decrease over time. Further comparisons of the mean values of the condition factor support the general systematic. Table 16 shows a post hoc comparison as per Duncan. Again, differences are very small, but values from fish fed with the test feed tend to be lower compared to those with control feed. The only value below 0.7 was found with fish in SSB on the last day of measuring. This is certainly a result of the high drop in mean weight between measuring dates six and seven. Additionally, it shows that values are separated in only four different

classes.

Table 16 Duncan Post hoc visualisation of condition factor

Date of measurement/ Feed	N	Subset for alpha = 0.05			
		1	2	3	4
7 / Test	17	,69241			
2 / Test	34	,70491	,70491		
6 / Test	22	,71864	,71864	,71864	
5 / Test	22	,71897	,71897	,71897	
6 / Control	28	,72355	,72355	,72355	
1 / Test	40		,73597	,73597	,73597
7 / Control	29		,73784	,73784	,73784
4 / Test	32		,73889	,73889	,73889
3 / Control	33		,74460	,74460	,74460
2 / Control	34		,74548	,74548	,74548
4 / Control	32			,74751	,74751
3 / Test	34			,74974	,74974
5 / Control	30			,75404	,75404
1 / Control	40				,76772
Sig.		,126	,057	,106	,142

Means for groups in homogeneous subsets are displayed.

a. Uses harmonic mean sample size = 28,908.

b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

For length of observed fish, there were no significant signs of differences between the two subsystems, nor was there any sign of a trend that would suggest that this would happen in the future if testing was continued.

Weight shows significance in differences of medians at the sixth and seventh measuring dates. Despite the first value of significance being 0.046, which is slightly below the necessary threshold, the second is already at 0.036. Additionally, the overall development of significance in differences of weight does not give any indications that this trend is likely to change.

- **Therefore: From a statistical point of view, these results show that fish tested in both subsystems have the same development of condition factor and length. The development of weight between the groups shows significant differences after five weeks of testing and an additional trend of further increase of mean values of weight is observable, which allows the assumption that these differences heighten over time.**

The main question that arises is where do these differences come from? Are they a result of the different feeds, or are other influences responsible for them?

To determine this, of further interest is the regression line of the condition factor. In contrast to weight and length development, it shows a negative growth. As seen in Figure 31 and Figure 32, this decline is only minimal and the difference between the two subsystems is even smaller. Therefore it might be acceptable to ignore it. Additionally, the amounts of decline observable for both subsystems are negligible, and therefore the difference in feed

cannot be their cause. Additionally, the differences in length are the same for both subsystems.

- **Is it possible that differences in growth performances were independent from fed feed?**

If feed is not responsible for differences in development dynamics, something else must be.

A possible explanation for differences might be found in the process of the test itself. Before the feeding test was started, all 80 test fish were fed the same feed. The used feed was the same that was later used for the control group. This means that the control group, kept in Subsystem A, was not exposed to a change in feed, while fish in Subsystem B had to adapt to a new feed. This might have led to reduced consumption at the beginning of testing. As mentioned in Chapter 5.2, visual observation did not support this assumption.

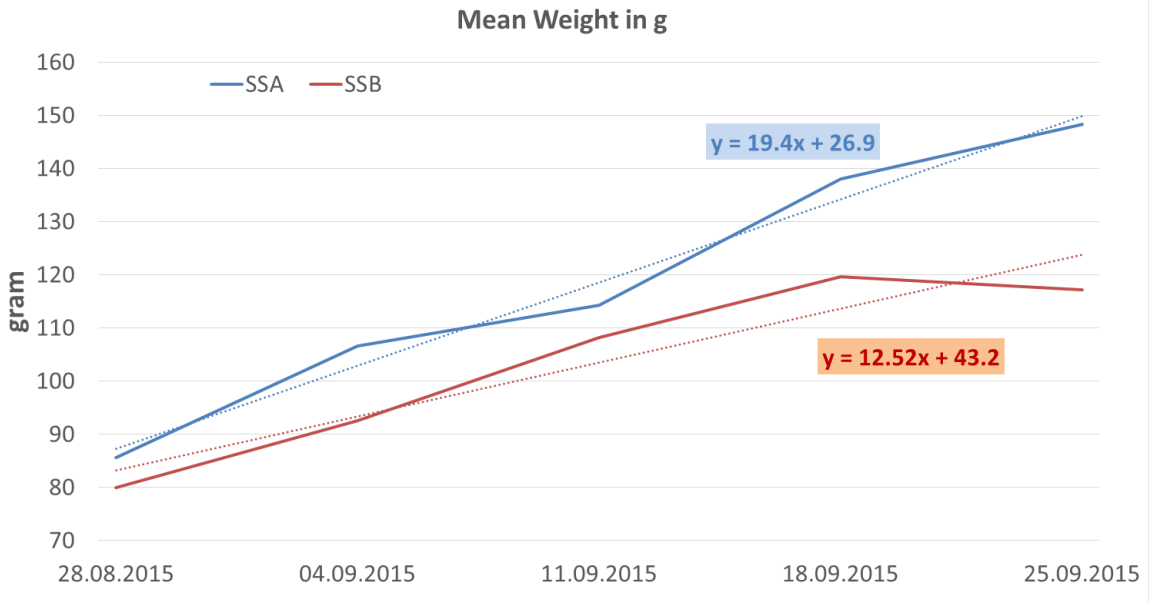


Figure 37 Regression line for mean weight development starting from 28.08.2015

If this would be the case, regression lines – starting after the first two measurements – should have a steeper increase for mean weight compared to the original regression lines for SSB but not for SSA. Figure 37 shows these alternate regression lines, which display an increase for the gradient in both subsystems. The increase is even higher for Subsystem A.

- **This suggests that the adjustment to the new feed for fish in Subsystem B did not have a grave influence on development dynamics.**

Another explanation could be that the amount of feed actively available in Subsystem B was below the desired three per cent of body weight per day. This is supported by the fact that in both subsystems, leftover feed was seldom found. What little leftovers were found were

mainly in the sedimentation tank of Subsystem B. As feed for Subsystem B was not floating, in contrast to SSA, and the drain for both subsystems were located at the bottom of the fish tanks, it was to be expected that SSB had higher amounts of leftover feed in its sedimentation tank.

- **This leads to the assumption that feed in SSA stays in the fish tank longer and is therefore longer available for the fish's consumption as well as also less likely to sink down to the drain.**

Additionally, physical and chemical conditions of the water could be a reason. From the observed physical characteristics of the water, temperature and pH values are mainly of interest. The oxygen content is negligible, as *C. gariepinus* do not depend very much on the water's oxygen for breathing. Out of these characteristics, only the temperature of SSA was close to concerning ranges at any time. Furthermore, chemical values of the water might have influenced the feeding behavior.

5.5 Water Data and its Influence on Development dynamic

It was possible to keep both the physical and chemical values of the water used in the system in optimum or satisfactory ranges most of the time. The small exceptions in physical values did not seem to have long-term negative effects on the development of the fish. While chemical values were eventually within satisfactory ranges, they varied in amounts of produced nitrate.

5.5.1 Physical

According to the optimum physical conditions of water for *Clarias gariepinus* shown in Figure 38 and Figure 39*,* values were mostly satisfactory during testing (de Graaf & Janssen, 1996, p. 9; Olaniyi & Omitogun, 2013, pp. 314–315).

pH values were constantly in the optimum range, while temperature was either in or close to the optimum range most of the time. Only twice did water in SSA reach unsatisfactory temperatures. In these cases, the water temperature rose up to 34.8° and 32.9° Celsius. These high temperatures resulted from failure of conduction in a heater. After both incidents, fish partially refused feeding. This was observable by inattention towards added feed, as well as higher amounts of leftovers than usual in the tank and mechanical filtration. A refusal of feed also occurred at least once in SSB but was a result of temperatures that were too low.

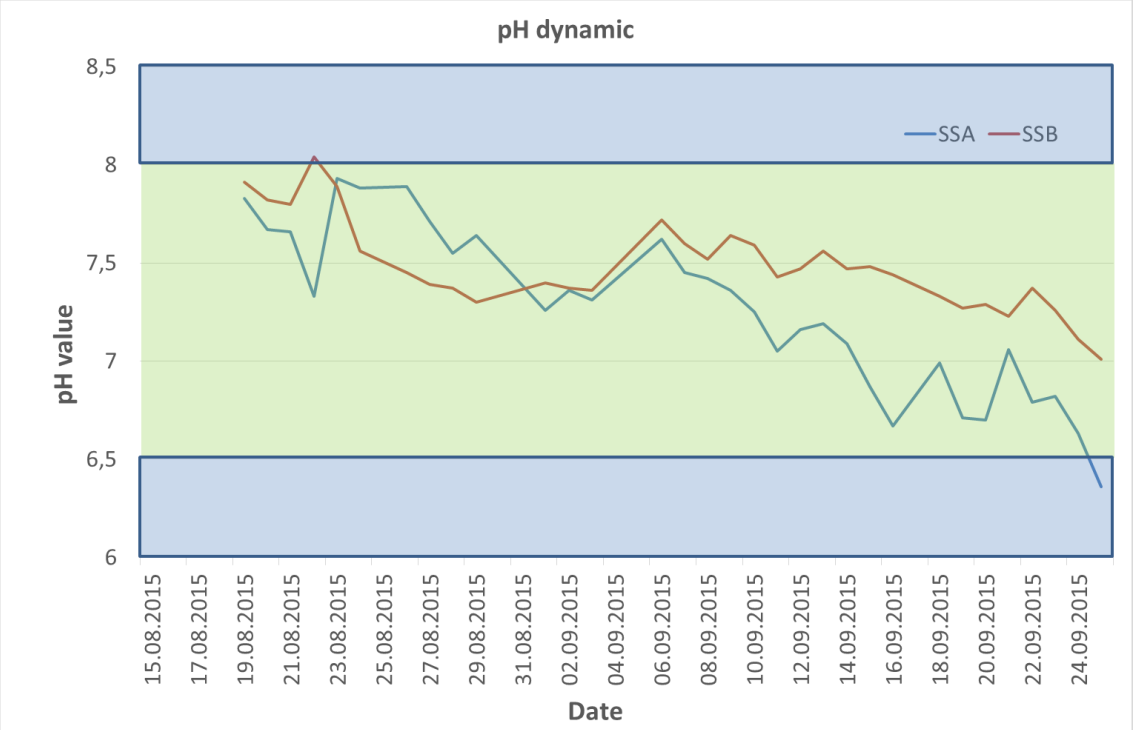


Figure 38 Optimum (green) and non-threatening (blue) ranges of pH for *Clarias gariepinus*

These incidents might have delayed the development of fish in both subsystems. As the corresponding weight and length gain in the corresponding timespan are still more then satisfactory, it is assumed that these incidents' negative influence was not too high. Therefore, it seems that the extreme values of physical water data in the system did not have a critical negative effect on overall development dynamics of fish in either of the two subsystems.

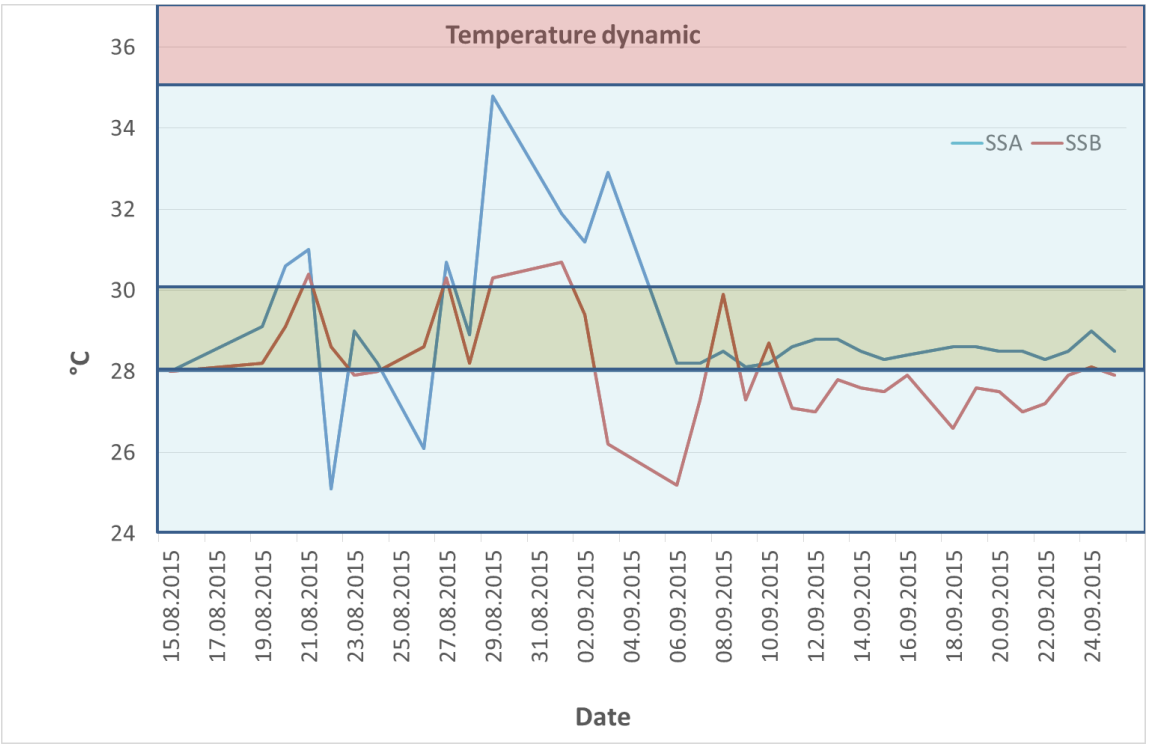


Figure 39 Optimum (green), non-threatening (blue) and threatening ranges of temperature for *Clarias gariepinus*

It is, however, observable that temperatures in Subsystem B were generally below temperatures in Subsystem A. As seen in Table 9 this led to a 1° Celsius difference in mean temperature over the course of testing of as well as to a difference of roughly 30 day degrees. Additionally, the water in Subsystem B was more frequently below the optimal temperature. It is possible that this difference in overall temperature had influences on the weight development of fish.

5.5.2 Chemical

The difference in technical performance between the subsystems likely led to a delayed establishment of nitrobacter and nitrosomonas cultures in Subsystem B. Therefore, water in SSA reached desirable nitrite, nitrate and ammonium values sooner than SSB. Both systems had non-threatening values after a reasonable time. From the 1st of September onwards, the nitrite values of both subsystems reached very similar values. At this point, differences were mainly observable in nitrate and ammonium levels. This means that Subsystem A had at least four more days of non-influential nitrite values regarding the development of the fish. It is possible that these four days had an overall influence on the differences of weight development of fish in the both subsystems.

NH_4^+ values were very similar over the course of testing. The higher amounts at the end of the testing period most likely resulted from the corresponding higher amount of biomass in SSA at the end of the testing period.

While the amount of ammonium allows for the assumption of a correlation with the biomass in the corresponding subsystem, the reasons for the NO_3^- values over the course of testing

for both systems are not clear. As ammonium is turned into nitrate, a strong positive correlation was expected. Statistical testing showed that values in SSA have a positive correlation of around 0.5, while SSB shows only a correlation of around 0.3. As the nitrate values are fairly constant in SSB, but have an overall steady rise in SSA, this weakens the theory that the differences might result from a later effective biofiltration in SSB. It might, however, result from a less effective biofiltration. This is also questionable, because after some starting problems, both systems worked equally effective.

It also seems unlikely that the amount of ammonium is heavily influenced by the two different feeds, as SSB had higher values then SSA at times.

6 Conclusion

As stated in Chapter 2, the main research question of this master thesis was:

- **What is the growth reaction of *Clarias gariepinus* fed with feed containing *Hermetia illucens* meal as a main protein source, in comparison to feed with fishmeal as a main protein source on the development dynamic of two groups of *Clarias gariepinus* in identical aquaponics systems?**

And was specified by three hypotheses:

H1: The development dynamic of Clarias gariepinus' body length shows no significant differences between the two feeds.

H2: The development dynamic of Clarias gariepinus' body weight shows no significant differences between the two feeds.

H3: The development dynamic of Clarias gariepinus' condition factor shows no significant differences between the two feeds

According to Chapters 4 and 5, Hypotheses H1 and H3 can be confirmed. Even as there are differences in the development dynamic between the two systems, and the test group has seemingly poorer results, these differences are in such small dimensions that they can't be proven significantly. Hypothesis 2, H2, however, must be refused.

H1: The development dynamic of Clarias gariepinus' body length shows NO significant differences between the two feeds.

H2: The development dynamic of Clarias gariepinus' body weight DOES SHOW significant differences between the two feeds.

H3: The development dynamic of Clarias gariepinus' condition factor shows NO significant differences between the two feeds

This leads to the answer of the main research question as:

- **The growth reaction of *Clarias gariepinus* fed with feed containing *Hermetia illucens* meal in comparison to feed with fishmeal as a main protein source on the development dynamic of two groups of *Clarias gariepinus* is the same for length and condition factor. The growth reaction of the development dynamic for weight established significantly better results for fishmeal-based feed after five weeks.**

Despite this, differences cannot be linked to the fed feed with complete certainty. The effect of the hydrochemical and physical differences in the two subsystems is not entirely clear, along with the differences in amounts of consumed feed. It is also not possible to completely reject the feed as the reason for the differences.

This leads to the assumption that feed with *Hermetia illucens* meal as a main protein source for *Clarias gariepinus* is not as ideal under the tested circumstances as the classic catfish feed. Nevertheless, it shows that it is generally possible to use such alternative feed compositions in floating aquaponics systems and probably in aquaponics in general, without a reduction in fish well-being.

To ensure more certainty, it is necessary to conduct further testing on the topic. For such testing, it might be reasonable to also test the test feed indoors as well as using classic aquaculture.

Fish were not tested on other possible consequences resulting from the use of *Hermetia illucens* meal in *Clarias gariepinus* feed. Therefore, it is possible that *Hermetia illucens* meal has further effects on *Clarias gariepinus*. These effects might be positive or negative.

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7.1.1 List of Tables

7.1.2 List of figures

8 Appendix

Table 17 Summary of dietary nutrient requirements of North African catfish Clarias gariepinus (requirement expressed for dry feed except where otherwise mentioned) Source: http://www.fao.org/fileadmin/user_upload/affris/docs/North_African_Catfish/English/table_4.htm

Nutrients	**Nutrient levels**			
	Life stage/size class			
	Larval rearing	**Nursery phase**	**Grow out**	**References**
	12–14 d	0.5–10 g	10–1 000 g	
Protein and amino acids				
Crude protein, % min	55 [1]	50 [e,20]	40-42 [2], 40 [3,24] 43 [2,4,6,16,17,25,26]	1,2,3,4,6,16,17,20,24,25,26
Least costed and or appetite feeding protein requirement			35 [16], 38 [2]	2,16
Amino acids, % min of dietary protein				
Arginine		4.5 [6], 4.45-4.50 [c,19]		6,19
Histidine		1.0–1.05 [5]	1.39 [17]	5,17
Isoleucine			1.56 [17]	17
Leucine			4.87 [17]	17
Lysine		5.7 [9]	4.49 [17]	9,17
Methionine	2.5 [1]		3.2 [8]	1, 8
Phenylalanine			4.56 [17]	17
Threonine			2.04 [17]	17
Tryptophan		1.1 [7]	2.59 [17]	7,17
Valine			2.08 [17]	17
Lipid and fatty acids				
Crude lipid, % min	9 [1]		8.2 [15], 10–12 [2],11.5 [3],13 [16],10–17 [17]	1,2,3,15,16,17
Essential fatty acids, % min				
18:2n-6				
20:4n-6				
18:3n-3				
20:5n-3				
22:6n-3				
n-3 : n-6 ratio	1:1 [1]			1
Carb., % recommended	21 [1]		15–35 [2,10,11, 12,15,16], 26-32 [17]	1,2,10,11,12,15,16,17

Energy				
Digestible energy, min kJ/g			14–16 [2], 12.7 [3, 21]	2,3, 21
Metabolisable energy, min kJ/g			13 [3]	3
Gross energy, min kJ/g			11–13 [4], 21 [3], 21.2 [15], 22-24 [17]	3,4,15,17
Protein to energy ratio, mg/kJ			20.5 [15], 26–29 [2], 31 [3], 31–36 [4], 21.5–23 [17]	2,3,4,15,17
Lipid to carbohydrate ratio (g/g)			2.47(lipid 13%, carbohydrate 33.42%) [16]	16
Minerals[a]				
Macroel. (%)				
Calcium		0.45 [18]	1.5 [2]	2,18
Phosphorus		0.45 [18]	0.5 [2]	2,18
Magnesium		0.04 [18]		18
Sodium				
Potassium		0.26 [18]		18
Microelements, mg/kg dry diet				
Iron		30 [18]		18
Sulphur				
Chlorine				
Copper		5 [18]		18
Manganese		≤2.40 [18]		18
Zinc		20 [18]		18
Cobalt				
Selenium		0.25 [18]		18
Iodine				
Molybdenum				
Chromium				
Fluorine				
Vitamins[a]				
Vitamin A IU/kg		1 000–2 000 [18]		18
Vitamin D IU/kg		500–1000 [18]		18
Vitamin E min mg/kg		25–50 [18]		18
Thiamine min mg/kg		1 [18]		18
Riboflavin min mg/kg		9 [18]		18
Pyridoxine min mg/kg		3 [18]		18
Pantothenic acid min mg/kg		10–15 [18]		18
Niacin min		33.1 [23]		23

mg/kg				
Folic acid min mg/kg		1.2 [18]		18
Choline min mg/kg		400 [18]		18
Biotin [b] min mg/kg			2.49 [13]	13
Ascorbic acid min mg/kg	150 [14], 500 [23]	11–60 [18], 50 [22]		14,18,22,23

Notes:

[a] Mineral and vitamin requirements are generally assumed to be the same as for *Ictalurus punctatus*.

[b] Biotin requirement determined for *Clarias batrachus*.

[c] For hybrids between *Clarias gariepinus* and C. *macrocephalus*.

[e] For hybrids between *Clarias gariepinus* and *Heterobranchus bidorsalis*.

Source:

[1] Uys and Hecht (1985)

[2] Uys (1989)

[3] Machiels and Henken (1985)

[4] Degani, Ben-Zvi and Levanon (1989)

[5] Khan and Abidi, (2009)

[6] Fagbenro, Nwanna and Adebayo (1999)

[7] Fagbenro and Nwanna (1999)

[8] Fagbenro, Balogun and Fasakin (1998)

[9] Fagbenro et al. (1998)

[10] Balogun and Ologhobo (1989)

[11] Heinsbroek, Van Thoor and Elizondo (1990)

[12] Fagbenro et al. (1993)

[13] Mohamed, Ravisankar and Ibrahim (2004)

[14] Merchie et al. (1997)

[15] Ali and Jauncey (2005a)

[16] Ali (2001)

[17] Pantazis (1999)

[18] Wilson and Moreau (1996)

[19] Singh and Khan (2007)

[20] Adebayo and Alasoadura (2001)

[21] Yilmaz et al. (2006)

[22] Adewolu and Aro (2009)

[23] Kuczynski (2002)

[24] Machiels and Henken (1987)

[25] Ali and Jauncey (2005b)

[26] Ali and Jauncey (2005c)

Table 18 Measurements 14.08.2015

Date: 14.08.2015					
Kreislauf A			Kreislauf B		
Fish number	Length per fish	Weight per fish	Fish number	Length per fish	Weight per fish
1	190	53,7	1	143	23,5
2	180	44,7	2	220	74,1
3	200	67,1	3	195	53,8
4	180	51	4	210	63,2
5	215	68,9	5	205	68,5
6	200	66,8	6	190	61,9
7	230	100,9	7	230	103,6
8	195	61,1	8	190	51,4
9	215	78,5	9	180	42,3
10	208	66,4	10	195	53,3
11	170	36,9	11	180	40,5
12	245	116	12	190	49,3
13	160	34,3	13	210	70,3
14	205	79,9	14	180	38,3
15	200	57,2	15	185	47,8
16	160	26,3	16	175	40,4
17	210	74,1	17	215	76,5
18	195	47,1	18	190	55,9
19	205	62,2	19	200	60,7
20	190	50,4	20	215	72,9
21	185	49,2	21	205	61,7
22	180	43,7	22	180	35
23	192	45	23	180	43,9
24	190	48,4	24	210	66,3
25	188	62,8	25	185	46,9
26	188	40,5	26	180	38,5
27	188	56,3	27	240	99,9
28	192	48,6	28	190	49,6
29	190	47,8	29	170	30,8
30	185	48,9	30	215	72,4
31	185	48,9	31	215	58,1
32	208	65,7	32	155	26,4
33	173	36	33	195	51,5
34	150	27,2	34	190	58
35	188	58,2	35	190	51,6
36	162	35,9	36	210	66,6
37	185	47,7	37	200	51,1
38	190	49,1	38	195	53,2
39	195	50,4	39	160	35,1
40	195	65,6	40	230	105,7
40	191,55	55,49	40	194,83	56,26
Number of fish	Mean length Subsystem A	Mean Weight Subsystem A	Number of fish	Mean length Subsystem B	Mean Weight Subsystem B

Table 19 Measurements 21.08.2015

Date: 21.08.2015					
Kreislauf A			Kreislauf B		
Fish number	Length per fish	Weight per fish	Fish number	Length per fish	Weight per fish
1	175	32,5	1	240	103,9
2	174	38,8	2	226	89,9
3	210	73,3	3	234	96,2
4	272	156,3	4	187	35,6
5	205	61,7	5	173	53
6	230	88,2	6	240	95,5
7	225	80,5	7	190	49
8	182	63,1	8	218	61,6
9	197	57,4	9	190	42,3
10	203	58,8	10	252	108,7
11	252	130,5	11	229	80,9
12	195	52,5	12	210	61,5
13	196	55,4	13	202	60,3
14	210	71,3	14	212	70,2
15	205	68,3	15	193	62,2
16	230	90,2	16	212	61,3
17	186	52,8	17	218	73,8
18	200	60,4	18	205	59,5
19	215	73,6	19	260	135,2
20	206	58,6	20	192	43,1
21	217	82,8	21	178	44,4
22	210	60,8	22	210	59,7
23	204	61,1	23	160	29,2
24	209	62,2	24	188	50,2
25	194	53,7	25	197	59,1
26	202	60,2	26	206	60,9
27	197	49,4	27	200	55,9
28	192	53,3	28	230	86,2
29	225	89,8	29	164	30
30	221	78,3	30	235	70,8
31	200	69,2	31	198	61,3
32	158	30,9	32	175	35,9
33	233	82,3	33	191	46,7
34	181	41	34	192	42
34	206,21	67,62	34	206,09	64,00
Number of fish	Mean length Subsystem A	Mean Weight Subsystem A	Number of fish	Mean length Subsystem B	Mean Weight Subsystem B

Table 20 Measurements 28.08.2015

Date: 28.08.2015					
Kreislauf A			Kreislauf B		
Fish number	Length per fish	Weight per fish	Fish number	Length per fish	Weight per fish
1	297	210,8	1	198	66
2	230	82,4	2	205	72,7
3	245	106,1	3	246	112,7
4	275	178,3	4	204	63,5
5	243	98,5	5	236	93,9
6	227	94,4	6	245	92,4
7	228	87,6	7	220	75,1
8	255	131,3	8	258	130,6
9	227	95,7	9	253	136,8
10	218	79,1	10	223	77,7
11	246	110,2	11	276	181,7
12	233	90,4	12	180	42,9
13	206	81,4	13	260	139,2
14	145	76,4	14	195	50,3
15	223	77,9	15	258	129
16	200	60,6	16	216	70,9
17	203	61,5	17	200	57,9
18	215	71,6	18	170	35,7
19	208	63,2	19	196	47,3
20	250	115,2	20	220	80,1
21	190	47,9	21	204	69,4
22	211	67,8	22	170	35,1
23	231	85,2	23	200	51,9
24	242	103,9	24	226	85,5
25	214	72,4	25	213	72,3
26	178	43,9	26	178	40,6
27	207	55,8	27	227	93
28	221	78	28	218	77,6
29	235	97,1	29	219	79,7
30	212	68	30	198	58,1
31	198	62,4	31	200	64,1
32	185	41,7	32	189	52,3
33	220	78,6	33	212	65,6
34	165	35,4	34	236	115,3
34	220,09	85,61	34	216,15	79,91
Number of fish	Mean length Subsystem A	Mean Weight Subsystem A	Number of fish	Mean length Subsystem B	Mean Weight Subsystem B

Table 21 Measurements 04.09.2015

Date: 04.09.2015					
Kreislauf A			Kreislauf B		
Fish number	Length per fish	Weight per fish	Fish number	Length per fish	Weight per fish
1	226	84	1	258	103,5
2	267	145,9	2	202	54,1
3	280	182,2	3	277	159,1
4	250	136,1	4	280	167,8
5	261	133,8	5	180	42,2
6	196	51	6	232	94,7
7	254	118,9	7	272	155,9
8	220	76,5	8	209	70,4
9	297	209,8	9	265	136,6
10	245	110	10	301	218,5
11	242	101,3	11	207	66,1
12	257	126,3	12	226	80,7
13	195	48,3	13	280	154
14	245	122,4	14	220	78,1
15	242	98,6	15	252	132,3
16	232	92,3	16	230	93,9
17	245	115,7	17	245	100,8
18	214	64,5	18	234	86,6
19	185	49	19	210	75,5
20	241	103,8	20	203	57,7
21	261	126,6	21	237	105,5
22	274	153,7	22	200	61,4
23	224	81,9	23	183	45,5
24	227	88,6	24	205	69,2
25	215	84,6	25	210	76,7
26	236	92,6	26	205	63,2
27	252	104,7	27	227	85,8
28	211	76,5	28	237	91,5
29	219	74	29	210	61,8
30	170	37	30	177	41,1
31	318	257,6	31	185	45,9
32	204	62,5	32	225	84,7
32	237,66	106,58	32	227,63	92,53
Number of fish	Mean length Subsystem A	Mean Weight Subsystem A	Number of fish	Mean length Subsystem B	Mean Weight Subsystem B

Table 22 Measurements 11.09.2015

Date: 11.09.2015					
Kreislauf A			Kreislauf B		
Fish number	Length per fish	Weight per fish	Fish number	Length per fish	Weight per fish
1	276	157,6	1	291	170,2
2	240	110,6	2	244	107,3
3	289	196,9	3	260	142,5
4	242	108,2	4	242	91,9
5	268	171,4	5	234	93
6	175	39,7	6	207	64,4
7	335	287,1	7	208	64,4
8	218	96	8	245	102,8
9	257	130	9	294	181,5
10	255	122,3	10	191	50,9
11	256	125,2	11	315	239,7
12	275	155,8	12	210	64,9
13	258	113	13	207	54,1
14	230	88,3	14	212	71,4
15	235	91,8	15	283	169,3
16	296	200,4	16	292	171,6
17	270	143,2	17	220	69,2
18	248	111,4	18	238	97,3
19	204	57,4	19	277	146,3
20	269	163,8	20	240	99,7
21	221	73,7	21	184	49,6
22	200	56,4	22	217	79,4
23	238	104,8			
24	259	135,8			
25	226	82,8			
26	195	55,7			
27	264	119			
28	209	66,9			
29	224	93,6			
30	272	141,9			
30	246,80	120,02	22	241,41	108,25
Number of fish	Mean length Subsystem A	Mean Weight Subsystem A	Number of fish	Mean length Subsystem B	Mean Weight Subsystem B

Table 23 Measurements 18.09.2015

Date: 18.09.2015					
Kreislauf A			Kreislauf B		
Fish number	Length per fish	Weight per fish	Fish number	Length per fish	Weight per fish
1	285	160,4	1	285	168,8
2	245	99,3	2	303	215,3
3	310	236,5	3	209	64,8
4	397	243,6	4	305	192,8
5	239	94	5	300	181,8
6	260	130,2	6	192	52,6
7	255	130,8	7	250	103
8	273	150,9	8	218	77,2
9	286	173,4	9	294	187
10	270	145,8	10	250	120
11	270	145,4	11	210	59,2
12	291	208,8	12	328	271,4
13	253	319,3	13	255	114,4
14	208	70	14	268	145,1
15	272	132,8	15	233	81,5
16	253	122	16	245	106
17	295	191,2	17	199	59,4
18	290	207,5	18	219	82,4
19	295	187,5	19	245	105,5
20	278	142,7	20	245	100,2
21	274	152,6	21	216	74,8
22	244	101,6	22	216	70,2
23	270	142,3			
24	238	108,5			
25	235	84,3			
26	218	71,8			
27	228	100,9			
28	218	68,9			
29	208	60,4			
29	264,07	144,26	22	249,32	119,70
Number of fish	Mean length Subsystem A	Mean Weight Subsystem A	Number of fish	Mean length Subsystem B	Mean Weight Subsystem B

Table 24 Measurements 25.09.2015

Date: 25.09.2015					
Kreislauf A			Kreislauf B		
Fish number	Length per fish	Weight per fish	Fish number	Length per fish	Weight per fish
1	285	172	1	222	85,4
2	314	264	2	272	152,6
3	220	75	3	319	216,7
4	300	228,3	4	248	106,6
5	300	205,9	5	255	108
6	190	46,1	6	224	80,6
7	297	168,7	7	310	192,1
8	291	168,7	8	342	294
9	245	97,6	9	202	58,4
10	231	78	10	297	177,5
11	256	112,5	11	251	111,2
12	266	142,7	12	264	124,7
13	310	247,9	13	215	59,4
14	329	275,6	14	253	108,8
15	221	85,1	15	225	83,2
16	255	157,1	16	257	122,3
17	336	285,2	17	239	86
18	370	359,1			
19	294	180,6			
20	314	236,7			
21	213	65,4			
22	239	112,5			
23	289	180			
24	287	175,1			
25	210	158,9			
26	250	124,9			
27	251	107,4			
28	275	156			
29	253	105,4			
30	290	150,9			
30	272,70	164,11	17	258,53	127,50
Number of fish	Mean length Subsystem A	Mean Weight Subsystem A	Number of fish	Mean length Subsystem B	Mean Weight Subsystem B

Table 25 Collected values of physical characteristics of water

	Subsystem A				Subsystem B			
Date	O^2 [mg/L]	pH	°C	mS	O^2 [mg/L]	pH	°C	mS
05.08.2015	9	7,87			10,21	8,08		
15.08.2015			28				28	
19.08.2015	7,13	7,83	29,1		7,07	7,91	28,2	
20.08.2015	6,7	7,67	30,6		6,73	7,82	29,1	
21.08.2015	7,13	7,66	31		6,72	7,8	30,4	
22.08.2015	5,2	7,33	25,1		7,17	8,04	28,6	
23.08.2015	6,97	7,93	29		7	7,89	27,9	
24.08.2015	7,12	7,88	28,2		5,92	7,56	28	
26.08.2015	7,5	7,89	26,1		5,62	7,45	28,6	
27.08.2015	6,7	7,71	30,7		5,46	7,39	30,3	
28.08.2015	6,88	7,55	28,9		5,49	7,37	28,2	
29.08.2015	5,95	7,64	34,8		3,55	7,3	30,3	
01.09.2015	5,49	7,26	31,9		3,8	7,4	30,7	
02.09.2015	5,6	7,36	31,2		4,08	7,37	29,4	
03.09.2015	5,55	7,31	32,9		5,76	7,36	26,2	
06.09.2015	6,8	7,62	28,2		6,84	7,72	25,2	
07.09.2015	6,92	7,45	28,2		6,55	7,6	27,3	
08.09.2015	7,02	7,42	28,5		6,2	7,52	29,9	
09.09.2015	7,08	7,36	28,1		6,94	7,64	27,3	
10.09.2015	7,24	7,25	28,2		6,84	7,59	28,7	
11.09.2015	6,96	7,05	28,6		6,87	7,43	27,1	
12.09.2015	7,06	7,16	28,8		6,81	7,47	27	
13.09.2015	7,1	7,19	28,8		6,82	7,56	27,8	
14.09.2015	6,92	7,09	28,5		6,57	7,47	27,6	
15.09.2015	7,04	6,87	28,3		6,75	7,48	27,5	
16.09.2015	6,99	6,67	28,4	1,65	6,76	7,44	27,9	1,37
18.09.2015	6,79	6,99	28,6	1,7	6,93	7,33	26,6	1,48
19.09.2015	6,94	6,71	28,6	1,89	6,71	7,27	27,6	1,43
20.09.2015	6,56	6,7	28,5	2,7	6,62	7,29	27,5	1,57
21.09.2015	6,3	7,06	28,5	2,13	6,53	7,23	27	1,61
22.09.2015	6,2	6,79	28,3	1,95	6,4	7,37	27,2	1,53
23.09.2015	6,5	6,82	28,5	2,18	6,42	7,26	27,9	1,58
24.09.2015	6,65	6,63	29	2,16	6,79	7,11	28,1	1,44
25.09.2015	6,62	6,36	28,5	2,14	6,72	7,01	27,9	1,51
26.09.2015	6,84	6,6	28,8	2,48	7,18	7,15	25,4	1,5
27.09.2015	6,83	6,56	28,4	2,4	7,23	7,16	25,1	1,53
28.09.2015	7,03	6,81	28,5	2,48	6,87	7,13	27,9	1,67
29.09.2015	6,61	6,72	28,6	2,51	6,82	6,89	27,9	1,66
30.09.2015	6,51	7,1	28,7	2,53	6,68	7,05	28,2	1,56
02.10.2015	6,94	6,54	28,5	2,5	7,23	7,02	26,5	1,66
03.10.2015	6,61	6,33	28,1	2,62	7,26	6,99	24,7	1,61
04.10.2015	6,74	6,51	28,9	2,72	6,98	7,04	27,9	1,68
05.10.2015	6,7	6,51	28,1	2,68	6,87	6,98	27,4	1,71
06.10.2015	6,86	6,62	27,9	2,7	6,97	7,01	27,2	1,73
07.10.2015	6,65	6,17	27,7	2,77	6,89	6,85	27,8	1,79
08.10.2015	7,02	6,48	26,6	2,79	6,88	6,98	26,3	1,8