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# THE ADDITION OF SIMPLE BIOLOGICAL FILTERS OF DIFFERENT CAPACITY TO SEMI-RECIRCULATING FISH REARING SYSTEM AND ITS EFFECTS

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## Abstract

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Aquaculture is currently one of the fastest growing food-producing sectors, accounting for around 50% of the world's food fish production. Limited resources, together with climatic change, have stimulated the search for solutions to support and sustain the production of fish as a source of protein for human consumption. The integration of a biological filtration (BF) into a semi-recirculating fish rearing system can increase its carrying capacity and increase system efficiency compared to its' energy consumption with minimum changes of system composition and minimal costs. Question is the capacity of the BF installed to a system and how it affects water quality. Two different amounts of BF media (surface) added to semi-recirculating rearing system compared with the same system without BF were tested in case of this study. The results have shown that if the BF capacity is insufficient, BF can have negative effects to the quality of water environment. The insufficient amount of BF media caused 4 times reduction of ammonia nitrogen (N-NH4+) in system with BF compared to non BF system so it increased the system capacity for feed load 4 times. On the other hand it also increased nitrite nitrogen concentrations permanently more than 5.8 times for BF system compared to non BF system and increased rearing costs because the need of adding chlorides to the system to protect fish from nitrites toxicity. When the BF was dimensioned properly (next year) there were almost no N-NH4+ in a system  $(0.10 \text{ mg}.\text{l}^{-1})$  and the concentration of N-NO<sub>2</sub><sup>-</sup> was kept at low levels too  $(0.150 \text{ mg}.\text{l}^{-1})$ . The nitrates  $(N-NO_3^{-})$  concentration reached the level of 5.37 and 8.65 mg.l<sup>-1</sup> in 2012 and 2013 respectively.

Keywords: Water reuse, nitrification, ammonia, nitrite, rearing capacity, Bioblok, trout

## **INTRODUCTION**

With increasing lack of high quality water to breed fish for human consumption increasing of fish production intensity is a way how to get more fish without raising water consumption. About 50 % of fish for human consumption is already produced in aquaculture (FAO 2014). Unfortunately simple reuse of the water to increase the efficiency of water use have its limitation in buildup of high ammonia concentrations. The solution is in adding the BF to the system to turn the toxic ammonia (Heteša and Kočková, 1997; Luo *et al.*, 2016; Sinha *et al.*, 2014) to nitrates which are toxic only in extremely high concentrations (Davidson *et al.*, 2014; Learmonth and Carvalho, 2015; Luo *et al.*, 2016; van Bussel *et al.*, 2012) compared to toxic levels for ammonia and nitrates (Arillo *et al.*, 1981, Bartlett *et al.*, 1987, Neils *et al.*, 1998). Increasing the complexity of RAS can reduce the need of fresh water up to 100 times (MacMillan, 1992; Blancheton *et al.*, 2007).

Nitrification is the standard way how to get rid of toxic ammonia in oxic conditions. The nitrification is a two-step process which transforms ammonia at first to nitrites than to nitrates (Blancheton *et al.*, 2007; Carrosa *et al.*, 2012; Dodds a Whiles 2010). This process is affected by many environmental

conditions and it is relatively slow so it takes time to bacteria to "ingest," "digest" and release the compounds of the process. The bacteria of second step (nitratation) are more sensitive to environmental changes and every treatment than first step (nitritation) bacteria (Pedersen et al., 2015). That means, in conditions of RAS, usually with high rates of water flowing around and low concentrations of ammonia, the filtering efficiency is not 100% per pass but it is usually somewhere around 5-60% per one passage of water thru BF (Brian, 2006; Liu, 2013 - lower efficiencies are mostly observed in cold seasons or after some mistakes in water quality management). The filter dimensions are not calculated to clean up all ammonia which comes to them at once, but to deal with the ammonia loading per whole day.

The question is the dimension of the BF. Everywhere in the publications scientists all over the world write about how the environment and its' conditions are affecting the biological filtration, but I haven't found many mentions about the proper dimensions of biological filtration (Carrosa *et al.*, 2012; Pedersen *et al.*, 2015; Suhr and Pedersen, 2010; Summerfelt, 2006) and how the under dimensioned filter will work. If it will improve the conditions just a little or will it do more harm than good?

Different amounts of filtration surface added to a system which simply reused the water before were tested. In a first year to compare the effect of additional filtration to water quality in comparison of two identical systems one with (F) and second without BF (NF). Increased amount of BF media added to the system was tested to compare its performance after the first year results examination.

#### **MATERIALS AND METHODS**

The testing systems were a twin raceways with the volume of 120 cubic meters of water (90 m<sup>3</sup> of rearing area). Water circulation was propelled by two airlifts both at the beginning of rearing area of the system (water flow was oriented clockwise). Sedimentation cones for removing quickly settleable solids are at the end of the rearing area behind the wing shaped bars (wing shape of bars reduce its' hydrodynamic resistance). There were unused areas where the water was turning back to go to another rearing section at both ends of the system. That was the area where the filtering media were placed. For the first test, with lower amount of media (3.27 m<sup>3</sup>), they were placed at both ends of the system. In second test whole the free area was filled with filtration media at one end of the system (12.97 m<sup>3</sup>). Bioblok<sup>\*</sup> 100 (100 m<sup>2</sup>.m<sup>-3</sup> - Expo-Net<sup>®</sup> A/S Denmark) was the filtration media used in both cases. The composition of the system and positions of biological filtration is shown at the Fig. 1 (1a for first, 1b for second test).

The tests were performed in three phases and repeated with modification in two consecutive years (2012 and 2013). First phase was always measuring the water quality parameters for 7 days prior installing the filtration elements. Second phase was adding the biological filtration and measuring the changes in chemical parameters for another 10 and 17 days daily, for first and second year respectively, and then every few days (in the second year) and third phase was periodical controlling of biological filtration function every 14 days or month. In a first year of study we compared water physic-chemical parameters between two systems. The main differences were



1: Composition of the system and placement of biological filtering media at first and second test (pic. 1a and 1b resp.). Water flow is oriented clockwise. \*1 - inflow; 2 - outflow; 3 - airlift; 4 - airlift wall; 5 - wing shaped bars; 6 - sedimentation cones; 7 - Biobloks

that one was equipped with BF (F) and one not (NF) and system NF was stocked with 5 times more fish (weight). The amount of feed load into the system was similar and the inflow/outflow was the same. Both tests were performed in the same part of year so the temperature was similar for both tests. The water inflow was set to 8 l.s<sup>-1</sup> so the whole volume of water was exchanged 4.17 times a day. The stocking density was 106,500 pcs of 6,1 g average weight (652.5 kg-6.95 kg.m<sup>-3</sup>) fed maximally 1.8 % (cca 11 kg) and 103,000 pcs fish 30 g in average (3090 kg-34.33 kg.m-3) fed max. 0.4 % of fish weight (cca 12 kg) in systems F and NF respectively for the first test. There were 37,300 pcs fish with average weight of 8.5 g (317.05 kg-3.53 kg.m<sup>-3</sup>) in second test. Rainbow trout (Oncorhynchus mykiss; Walbaum, 1792) was used in both years. Stocking densities were according to farmers needs and couldn't be changed. Feeding was adjusted according to actual water temperature, oxygen saturation and fish mortality in the systems. Fish were fed twice a day at 8 a.m. and 14 o'clock. Biomar Inicio Plus (1.1; 1.5 and 2.0 mm with 56/18; 54/21 and 52/32 - Protein/ Fat rating in % of feed weight) was used as feed in 2012 and 2013. The test duration was from  $12^{\text{th}}$ Jully to 18th September 2012 and from 16th Jully to 6<sup>th</sup> November 2013. Filtration media were installed in the afternoon 17th Jully 2012 and on morning 22<sup>nd</sup> Jully 2013. The filtering media weren't cleaned for whole test duration (18th Jully to 18th September 2012) for the first test and were cleaned once by pressurized water (at 67<sup>th</sup> day–25<sup>th</sup> September 2013) and then stayed in the system until January 2014 for second test. Water pH, temperature, O<sub>2</sub> content [mg.l<sup>-1</sup>] and saturation [%] were measured by multimeter HQ40d multi equipped with IntelliCAL™ LDO101 Rugged Luminescent/Optical Dissolved Oxygen (LDO) probe and IntelliCAL<sup>™</sup> PHC101 Rugged Gel Filled pH Electrode (Hach Company,

USA) twice a day during 1<sup>st</sup> and 2<sup>nd</sup> period and once per day ever since. The water analysis were made by spectrophotometer WTW PhotoLab Spectral (WTW, Germany) with use of semi micro methods according to standards for water analysis during first phase (APHA, 1998; Pitter, 2009). Measuring continued with field spectrophotometer PF-12 plus (Macherey-Nagel GmbH & Co.KG, Germany) with VISOCOLOR ECO test sets for chlorides, ammonium, nitrites and nitrates (Macherey-Nagel GmbH & Co.KG, Germany) every day during 2<sup>nd</sup> phase. Periodical measuring in 14 day or a month long intervals continued with spectrophotometer photoLAB UV-VIS 6600 (WTW, Germany) with the use of the same methods as in the 1<sup>st</sup> during 3<sup>rd</sup> phase. Samples were taken and physic-chemical parameters measured at 8:00 a.m. pre feeding.

#### RESULTS

The temperature during both tests was from 15.0-19.8 °C. When comparing systems F and NF the amounts of ammonia nitrogen (N-NH<sub>4</sub><sup>+</sup>) reached levels up to 0.7 and  $0.5 \pm 0.2$  for systems F and NF respectively, during the 1<sup>st</sup> test (2012). Then the N-NH<sub>4</sub><sup>+</sup> levels went down straight right after biological filter installation in system F and stayed the same in system NF. The nitrite nitrogen (N-NO<sub>2</sub><sup>-</sup>) concentrations raised in system F and stayed low in system NF as it is shown in graph (Fig. 2).

Nitrite levels in system F stayed more than 3 times higher than in system NF (0.276 : 0.092 and 0.227 : 0.045 mg.l<sup>-1</sup> at 20<sup>th</sup> August 2012 and 18<sup>th</sup> September 2012 in systems F and NF respectively) in the third phase of the test. Daily feed loading to the systems was similar so the nitrogen loading of the systems was comparable. The filtering media were removed from the system at 18<sup>th</sup> September 2012. The increase of nitrites in system F was permanent and increased the breeding costs because



2: Feed loading and changes in ammonia and nitrite nitrogen during second phase of the test 1.

the need of adding chlorides to avoid the toxicity of nitrites (Kroupová *et al.*, 2005; EIFAC, 1980) in case of test 1. The BF insufficiency for nitritation has shown after feed load increased ( $\geq 16$  kg–d<sup>-1</sup>). Persistent rise of N-NH<sub>4</sub><sup>+</sup> (0.52 mg.l<sup>-1</sup>) concentration in system led to stock reduction at the end of the test (18<sup>th</sup> September 2012). The N-NO<sub>2</sub><sup>-</sup> felt down to a levels similar to system NF (0.045 mg.l<sup>-1</sup>) and N-NH<sub>4</sub><sup>+</sup> raised to 0.71 mg.l<sup>-1</sup> until 20<sup>th</sup> September 2012 even after stock and feeding reduction.

The N-NH<sub>4</sub><sup>+</sup> and N-NO<sub>2</sub><sup>-</sup> levels were  $0.57 \pm 0.02$ and  $0.389 \pm 0.020$  mg,l<sup>-1</sup> before installation of BF during the second test. Ammonia levels felt down to 0.10 mg,l<sup>-1</sup> N-NH<sub>4</sub><sup>+</sup> in 21 days after BF installation. Nitrite concentration maximum culminated at 0.400 mg,l<sup>-1</sup> N-NO<sub>2</sub><sup>-</sup> from 26<sup>th</sup> to 28<sup>th</sup> day and then felt down to 0.150 mg,l<sup>-1</sup> N-NO<sub>2</sub><sup>-</sup> in next 8 days where it stayed for next 25 days (Graph a at Fig. 3). There was an increase of N-NO<sub>2</sub><sup>-</sup> concentration for 30 days After cleaning the filtering media and then N-NO<sub>2</sub><sup>-</sup> levels felt even to lower levels than before cleaning the filter (<0.100 mg.l<sup>-1</sup>). The maximum concentration of N-NO<sub>2</sub><sup>-</sup> was even higher after cleaning the BF media than after its first installation (graph b at Fig. 3). BF nitrification efficiency was from 0.00 to 60.00 % and from -122.22 to 7.14 % per pass for N-NH<sub>4</sub><sup>+</sup> and N-NO<sub>2</sub><sup>-</sup> respectively during the second test. Numbers are calculated from values measured before and after passage of water thru BF.

#### DISCUSSION

A rapid decrease of  $N-NH_4^+$  concentration followed by buildup of higher concentrations of  $N-NO_2^-$ , as it is common for biological filter startup (Bregnballe, 2010), during both tests came after installing BF to the system. The difference of length of the process was probably caused by lower amount



3: Graphs of changes in temperature [°C], feed loading [kg], oxygen content [mg.l<sup>-1</sup>] – right axis, N-NH<sub>4</sub><sup>+</sup> and N-NO<sub>2</sub><sup>-</sup> concentrations [mg,l<sup>-1</sup>] – left axis.

of bacteria in the system in second year before installing BF. The transparency of water was much lower (seen by eyes, not measured) so there were some nitrification bacteria already floating in flocks which colonized the BF surface much faster in a first year. The filter startup speed is influenced by physic chemical parameters of water and the amount of nitrifying bacteria present or added to the system (Bartrolí et al., 2011; Satoh et al., 2003). Ammonia concentration was the limiting factor for feeding the fish for the system NF. In system F the ammonia was effectively removed by the BF until the amount of the feed added reached 16 kg during the first test, then N-NH4<sup>+</sup> concentrations started to rise too (20<sup>th</sup> August 2012 – 0.35 mg.l<sup>-1</sup>). This shows the increase of system carrying capacity for feed loading by four fold. On the other hand the insufficiency of BF capacity caused increased costs of running the system because the need of continual addition of chlorides (EIFAC, 1980; Kroupová et al., 2005). In our case maintaining chloride number  $(mg.l^{-1} Cl^{-}.(mg.l^{-1} N-NO_2^{-})^{-1})$  at level supposed to be safe for fish (100 – Kroupová et al., 2005) means addition of 33 kg of salt (NaCl) to the system every day. The need of adding the chlorides and measuring them increased costs of farming and enlarged farmers' daily schedule. Insufficient filtration capacity would probably be even more evident if the water inflow was reduced to reduce the chlorides flushing from the system. The insufficient processing of nitrites by small biological filter was probably caused by a competition among nitritation and nitratation bacteria which are both

slow growing bacteria but nitratation bacteria grow 16% faster (Painter and Loveless, 1983). Nitritation bacteria probably colonized most of the filtration surface and didn't allowed a growth of nitratation bacteria. Although there was a massive growth of heterotrophic bacteria over nitrification bacteria which probably supported this imbalance. The filtering capacity in the second test was increased and did not reached its' limit for the whole duration of observation (22<sup>nd</sup> Jully to 6<sup>th</sup> December 2013). The only complication in BF function came after cleaning up the filter media by pressurized water. Filter media were extracted from the system, cleaned up and returned back. This probably flushed away most of the nitratation bacteria present on BF media surface. The mechanical effect of pressurized water can be compared to addition of mechanical sponge particles to disc filter media chamber to clean the cake on filtration discs membrane by Kimura et al. (2000). They recommend to return a part of cleaned up bacterial matter back to the filter to maintain its proper function. Addition of BF is highly environmentally valuable way to increase the food fish production which is relatively easily and cost friendly applicable to existing rearing facilities without major structural changes. Bioblok has proven itself as suitable media for this kind of application. This paper should have the impact to the use of biological filters in practice (on farms) and should lead people to not to use under dimensioned biological filters if they do not need more work and caring at their farms.

#### CONCLUSION

The main conclusion of this work is that under dimensioned BF added to rearing system can increase the capacity of the system for feed load but also increase the rearing costs or do harm to the fish if the level of  $N-NO_2^-$  is not controlled and chlorides are not added to prevent its toxicity. If the BF is dimensioned properly, which depends on planned feed amount added to the system daily, the system can work as simple RAS with increase of rearing intensity and/or reduction of water exchange needs. The system equipped with right amount of BF can consume at least 5 times less water or carry 5 times more fish/higher feed load. It is better to reduce the feed input to the system than install under dimensioned BF into it. The second conclusion is that cleaning of BF media should not be done by pressurized water but only with larger amounts of low pressure water which should not flush away the nitrification bacteria.

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