

## Significant factors affecting the economic sustainability of closed aquaponic systems.

### Part II: fish and plant growth

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**Abstract.** Two identical closed ebb-flow substrate aquaponic systems for warm-water fish were tested for fish and plant productivity. Each system contained 3.7 m<sup>3</sup> water, and the relationship of the water volume in the aquaculture tank to the settling basin (sedimenter, clarifier), the biofilter and the hydroponic units was 2.25:1:0.075:0.6 (fish tank:hydroponic unit = 3.75). The comparative batch cultivation of African catfish (*Clarias gariepinus*) and Nile tilapia (*Oreochromis niloticus*) at the fish feed input level of 200 g per day resulted in non-significant total growth of 13 kg. The feed conversion ratios (FCR) were 1.0, better values compared with earlier tested closed aquaponic systems. The specific growth ratio was not significantly different ( $p < 0.05$ ) between *C. gariepinus* ( $0.65\% \text{ d}^{-1} \pm 0.25$ ) and *O. niloticus* ( $0.71\% \text{ d}^{-1} \pm 0.19$ ). Combined dissolved oxygen (DO) levels in the fish tank, clarifier and sump were significantly different between the two species, with  $6.95 \text{ mg L}^{-1} (\pm 0.50)$  in *C. gariepinus* and  $4.73 \text{ mg L}^{-1} (\pm 1.05)$  in *O. niloticus*. Significantly better growth was observed for plants in combination with the Nile tilapia, with  $55.89 \text{ g} (\pm 49.77)$  for lettuce and  $168.27 \text{ g} (\pm 350.88)$  for cucumber fruits. This is referred to a slightly different light regime and a different fish species cultivation. The available plant nutrients (nitrite, nitrate), as well as the suspended particle load, in both identical systems, also differed, with consequences for system maintenance and substrate biofilter activity.

**Key Words:** African catfish, aquaponics, biomass weight gain, *Clarias gariepinus*, ebb and flow system, fish to plant combination, *Tilapia*.

**Zusammenfassung.** Zwei identische Ebbe-und Flut Aquaponiksysteme wurden hinsichtlich der Fisch und Pflanzenproduktion verglichen. Die Aquaponiksysteme wurden im geschlossenen Süßwasserkreislauf betrieben. Die Hydroponikeinheit des Aquaponiksystems bestand aus Kiessubstrat. Jedes System besaß ein Wasservolumen von 3,7 m<sup>3</sup>, mit einem Verhältnis des Wasservolumens im Aquakulturbehälter zum Sedimenter, dem Biofilter und der Hydroponikeinheit von 2,25:1:0,075:0,6 (Fischbehälter:Hydroponik Einheit = 3,75). Das Wachstum des Afrikanischen Raubwelses (*Clarias gariepinus*) und des Nil Tilapia (*Oreochromis niloticus*) jeweils einer Altersgruppe zeigte bei einer konstanten Futtergabe von 200 g pro Tag einen nicht signifikanten Biomassezuwachs von 13 kg. Die Fütterungskoeffizienten waren 1,0 und zeigten bessere Werte im Vergleich zu früheren Studien bei geschlossenen Aquaponiksystemen. Die spezifische Wachstumsrate (SWR) war nicht signifikant ( $p < 0,05$ ) zwischen *C. gariepinus* ( $0,65\% \text{ d}^{-1} \pm 0,25$ ) und *O. niloticus* ( $0,71\% \text{ d}^{-1} \pm 0,19$ ). Die kombinierten Sauerstoffwerte im Fischbecken, Sedimenter und Pumpensumpf unterschieden sich signifikant zwischen den beiden Arten, mit  $6,95 \text{ mg L}^{-1} (\pm 0,50)$  bei *C. gariepinus* und  $4.73 \text{ mg L}^{-1} (\pm 1,05)$  bei *O. niloticus*. Ein signifikant besseres Pflanzenwachstum wurde in Kombination mit dem Nil Tilapia erreicht, bei Kopfsalat mit  $55,89 \text{ g} (\pm 49,77)$  und Gurkenfrüchten mit  $168,27 \text{ g} (\pm 350,88)$ . Ursachen sind in einem gering differierendem Lichtregime und den verschiedenen Fischarten zu sehen. Die verfügbaren Pflanzennährstoffe (Nitrit, Nitrat) sowie gelöste Partikel (Trübung) differierten signifikant zwischen den beiden Systemen, mit Konsequenzen hinsichtlich der Nährstoffmenge im System und der Biofilteraktivität.

**Schlüsselworte:** Afrikanischer Wels, Aquaponik, Biomassezuwachs, *Clarias gariepinus*, Ebbe- und Flut System, Kombination Fisch und Pflanze, *Tilapia*.

**Introduction.** Aquaponics combines the production of fish and plants in a soilless environment, benefiting the performance and growth rates of the cultivated species. Previous studies demonstrated a 12.5% better growth rate of Nile tilapia (*Oreochromis niloticus*) in an aquaponics system (Rakocy 1989a), compared with the cultivation of animals and plants separately (Savidov 2005b). However, combined fish and plant cultivation occurs in a variety of different systems, from, for example, outdoor fish and

rice cultivation and plant units for wastewater treatment to modern closed recirculation systems with various fish-plant-substrate combinations (Knaus 2012).

Aquaculture as a business requires a stable run of the cultivation system, maintaining all environmental factors under control. This is more difficult under closed aquaponic conditions, where stocking density, feed input and harvesting directly influence fish, as well as plant growth. With a high number of variables in system design, such as the type of aquaponics, component ratios and system alterations (Palm et al 2014), the systems studied up to now often differ, making comparative conclusions difficult. Lennard & Leonard (2006) compared a gravel substrate system with a floating raft system and a NFT (nutrient film technique) system, achieving a significantly better biomass gain and yield of Murray cod (*Maccullochella peelii peelii*), combined with Green Oak lettuce (*Lactuca sativa*), in the gravel system. However, the environmental parameters did not show significant differences between the three tested systems. Kotzen & Appelbaum (2010) compared a floating raft and later a gravel based brackish water system with a fresh water floating raft system in a desert environment. The water quality parameters differed significantly between the systems with higher electric conductivity (EC) levels in the brackish system without huge negative effects on fish (*Tilapia* sp.) and plant growth. Most of the plant species (aubergine - *Solanum melongena*, celery - *Apium graveolens*, chard - *Beta vulgaris* 'cicla', chilipepper - *Capsicum annuum*, chives - *Allium schoenoprasum*, kohlrabi - *Brassica oleracea*, parsley - *Petroselinum* sp., mint - *Mentha* sp.) grew successfully. Most studies so far have examined stand-alone systems, such as Rakocy's UVI-Raft system (Rakocy et al 2006; Savidov 2005a; Al-Hafedh et al 2008), a light expanded clay aggregate system from Graber & Junge (2009) and the high mechanised, nearly emission-free NFT system of Kloas et al (2011). Our system at University of Rostock was built as a low-tech gravel substrate ebb and flow aquaponic system for warm-water fish in a freshwater environment. The system used no additional fertilizers, had a low water in and output and an optimal fish feed input of 200 g feed per day under the cultivation of *O. niloticus* fry and different plants. Control of the chemo-physical parameters of dissolved oxygen, salinity and conductivity demonstrated a stable run of the system (Palm et al 2014) after a run-in phase, an exponential phase (until the maximum capacity of the system was reached), and a steady state that would allow constant fish and plant production. In addition to the system design that directly influenced the chemo-physical characteristics during the production cycle, other factors, such as feed design, fish welfare, parasites and pathogen control, as well as fish and plant choice in combination with cultivation practices (batch, staggered, intercropping, polyculture) and product quality, were discussed as being essential to achieve economic sustainability.

Palm et al (2014) described the effects of the system design and chemo-physical parameters on the stability of a low-tech warm-water aquaponic system. The present study describes the performance of the above system under the comparative cultivation of African catfish - *Clarias gariepinus* and Nile tilapia - *Oreochromis niloticus*, and in combination with lettuce (*Lactuca sativa*), cucumber (*Cucumis sativus*), tomato (*Solanum lycopersicum*) and basil (*Ocimum basilicum*), under the optimal feeding regime. The effects of the different fish species on the system performance, with the cultivated fish being the only viable factor in the system, are discussed.

## Material and Method

**Experimental design and data collection.** The tested facility is characterised as a low-tech closed ebb-flow substrate aquaponic system (total fixed costs for each cycle: 2,562.38 €) (Table 1), combining warm-water fish with a gravel bed plant cultivation unit. It consisted of two separate, identical aquaponic subsystems (cycle I and cycle II), built in a temperate glasshouse on a surface area of appr. 50.00 m<sup>2</sup> (Palm et al 2014) (Figure 1).

Table 1

Fixed costs of components for each low tech gravel ebb-and flow aquaponic cycle

<i>Component</i>	<i>Costs (€)</i>
Fish tank (fibre glass)	839.09
Clarifier	450.46
Wooden plant boxes with sump	393.22
Gravel substrate	252.84
Trickling filter	106.97
Biocarrier KNS	50.00
Pump UP 40	146.80
Heater 3 KW	149.00
Automatic feeder	139.00
Low level air pump	35.00
Sum total	2,562.38



Figure 1. Aquaponic facility with two identical recirculating subsystems (cycle I and II) at the University of Rostock: a. fish tank connected to a clarifier, b. plant boxes with water inflow, c. tomato plants, d. tomato plants under comparative soil cultivation (also see Palm et al 2014).

The water volume was 3.7 m<sup>3</sup> in each recirculation system, consisting of a single glass fibre fish tank (3.90 m<sup>3</sup>, 2.05 x 2.05 x 0.93 m, AquaLogistik, Möhnesee-Wippringsen, Germany) filled with appr. 1,800 L, a clarifier (1.00 m<sup>3</sup>, IBC) filled with nearly 800 L, four wooden plant boxes (4 x 2.00 m<sup>2</sup>) with a sump (0.61 m<sup>3</sup>), a trickling filter (200 L) filled with Biocarrier KNS (60 L) and a single pump (UP 40, 3.000 L h<sup>-1</sup>, AquaLogistik, Möhnesee-Wippringsen, Germany) with a flow-type heater (3.00 KW) with automatic control. The hydroponic area was equipped with overhead light (RZB Light Stream Flat-Type Maxi, Osram Powerstar HQI-T 400 WIN, Germany) for illumination at night. The plant boxes (1.00 x 2.00 x 0.30 m) were laid out with polyethylene foil (3 mm) and filled with gravel (2,000 kg in each cycle) as a substrate, with a maximum water level of 20 cm (120.00 L). The plant boxes were equipped with a water siphon (bell pipe) that

allowed one maximum water level within one hour (ebb and flow system, 24 times per day). The filtered amount of water through the plant boxes was set for 11,520 L per day, passing through the hydroponic unit 3.1 times in 24 hours. The relationship of the water volumes in the aquaculture unit (1,800 L), the sedimenter (800 L), the biofilter (60 L volume biocarrier), and the hydroponic unit (480 L) was 2.25:1:0.075:0.6 (fish tank:hydroponic unit = 3.75).

The entire experiment was carried out from the 03.12.2012 until 04.03.2013. The fish species (*C. gariepinus* and *O. niloticus*) were cultivated in each subsystem (cycle I and II), applying the same plant species, with an acclimatisation phase of 30 days at the beginning, an experimental time of 53 days (83 days) and a de-acclimatisation phase of 9 days (overall 92 days) at the end of the experiment. The measurements of chemophysical water parameters of temperature [ $^{\circ}\text{C}$ ], oxygen [ $\text{mg L}^{-1}$ ], oxygen [%], salinity [‰], conductivity [ $\mu\text{S cm}^{-1}$ ], pH, and redox potential [mv] were taken twice a week by using an HQ40D multimeter (Hach Lange GmbH, Germany). Additionally, the nutrient parameters TAN-N [ $\text{mg L}^{-1}$ ],  $\text{NH}_3\text{-N}$  [ $\text{mg L}^{-1}$ ],  $\text{NO}_2\text{-N}$  [ $\text{mg L}^{-1}$ ],  $\text{NO}_3\text{-N}$  [ $\text{mg L}^{-1}$ ], phosphorus [ $\text{mg L}^{-1}$ ] and suspended particles [ $\text{mg L}^{-1}$ ] were measured by using the spectral photometer DR-3900 (Hach Lange GmbH, Germany). PAR (Photosynthetically Active Radiation) [ $\mu\text{m m}^{-2} \text{s}^{-1}$ ] was taken by using a LI-190SA Quantum Sensor (LI-COR).

**Fish and plant species.** The fish tank of cycle I was stocked with mixed sex *C. gariepinus*, 480.23 g initial fish weight ( $\pm 75.68$ ) and an initial biomass of 16,808 g, obtained from a local fish farm (PAL GmbH, Abtshagen, Germany). At the same time, fish tank of cycle II was stocked with mixed sex *O. niloticus*, 173.51 g ( $\pm 14.64$ ) initial fish weight and 14,054 g total initial biomass. All fish were fed by automatic feeders with an E-2P Stella (Skretting), with an average feed input, during the acclimatisation phase combined with the experimental phase, of 165.36 g ( $\pm 74.37$ ) (total of 83 days), and a constant feed supply of 200 g per day during the 53 days of the experimental phase in cycle I and cycle II.

For plant cultivation, species of lettuce (*Lactuca sativa*), tomato (*Solanum lycopersicum*), cucumber (*Cucumis sativus*) and basil (*Ocimum basilicum*), at a minimum distance of 0.25-0.30 m, were used in the same number and position in both cycles (I and II). Plant seeds originated from N.L. Chrestensen Erfurter Samen- und Pflanzenzucht GmbH (Erfurt, Germany) and Gartenland GmbH (Aschersleben, Germany).

**Statistical analyses.** Tests were performed in order to identify possible effects caused by the fish and plant choice between the two cycles (duplicate groups). All data from the fish tank, clarifier and sump were combined, calculating the mean for each data set and cycle. Values were compared between cycle I and cycle II, using the Shapiro-Wilk test, followed by the *t*-test and Levene statistic, in the case of normal distribution. Otherwise, the Mann-and-Whitney test was performed to determine significant differences at the  $p < 0.05$  level. All data were analysed by Microsoft Excel (2010) and the SPSS 20.0 statistical software package (IBM).

## Results

**Water parameters.** Water parameters showed significant ( $p < 0.05$ ) differences in the oxygen levels between the two systems (Figure 2), with  $6.95 \text{ mg L}^{-1}$  ( $\pm 0.50$ ) in cycle I (*C. gariepinus*) and  $4.73 \text{ mg L}^{-1}$  ( $\pm 1.05$ ) in cycle II (*O. niloticus*), and an oxygen saturation of  $81.68 \%$  ( $\pm 5.90$ ) in cycle I and  $56.02 \%$  ( $\pm 12.92$ ) in cycle II (Table 2).

Nitrite concentrations differed significantly between cycle I and II, with  $0.09 \text{ mg L}^{-1}$  ( $\pm 0.05$ ) in cycle I and  $0.21 \text{ mg L}^{-1}$  ( $\pm 0.15$ ) in cycle II, as did nitrate concentrations, with  $40.54 \text{ mg L}^{-1}$  ( $\pm 10.97$ ) in cycle I and  $31.06 \text{ mg L}^{-1}$  ( $\pm 5.17$ ) in cycle II. The phosphate concentrations were not significantly different between the cycles, but during the run of the experiment, a decreasing trend was observed (Figure 3). Suspended particles were significantly different, with  $9.77 \text{ mg L}^{-1}$  ( $\pm 4.13$ ) in cycle I and  $3.20 \text{ mg L}^{-1}$  ( $\pm 1.71$ ) in cycle II.

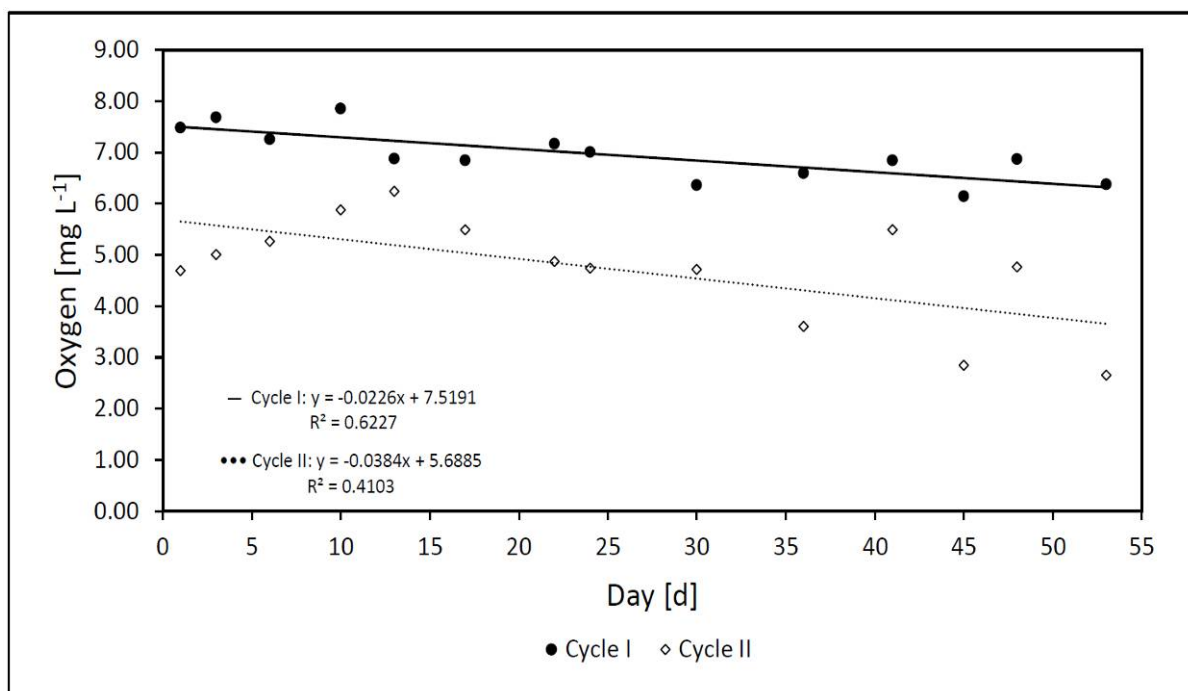


Figure 2. Oxygen [mg L<sup>-1</sup>] distribution with *C. gariepinus* (cycle I) and *O. niloticus* (cycle II).

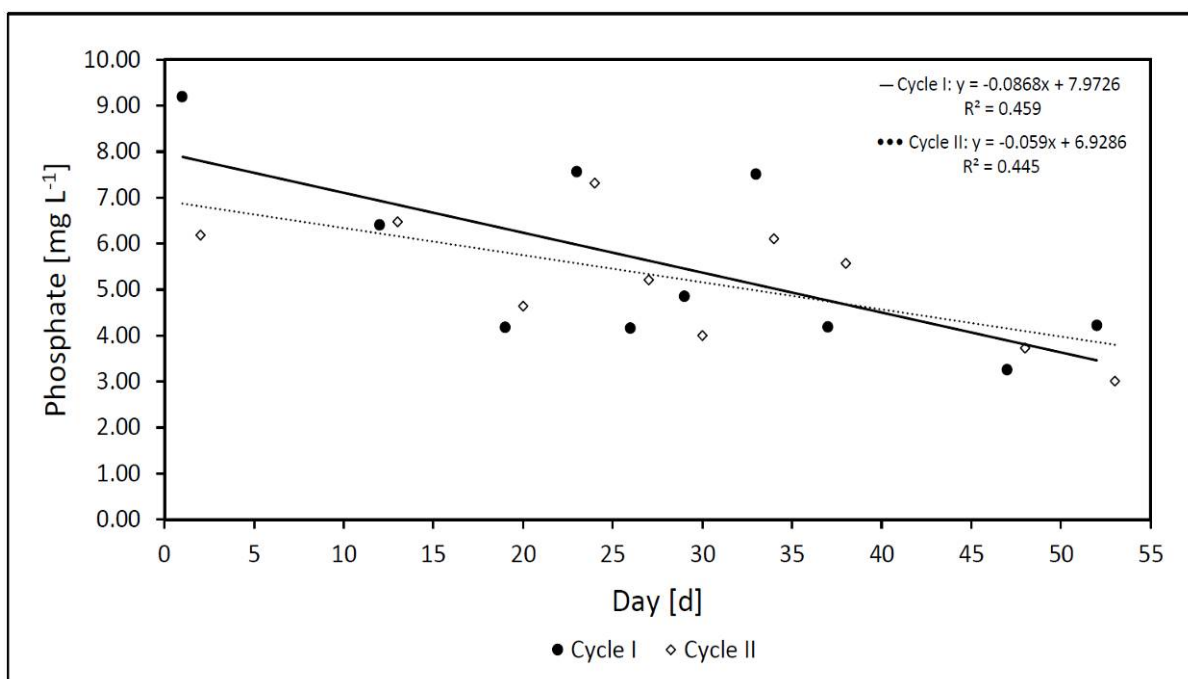


Figure 3. Phosphate [mg L<sup>-1</sup>] distribution with *C. gariepinus* (cycle I) and *O. niloticus* (cycle II).



Table 2

Chemo-physical water parameters of *C. gariepinus* (cycle I) and *O. niloticus* (cycle II)

Parameter	Cycle I	Cycle II
	Mean $\pm$ SD	Mean $\pm$ SD
Temperature [°C]	23.46 <sup>a</sup> $\pm$ 1.31	23.84 <sup>a</sup> $\pm$ 1.39
Oxygen [mg L <sup>-1</sup> ]	6.95 <sup>a</sup> $\pm$ 0.50	4.73 <sup>b</sup> $\pm$ 1.05
Oxygen saturation [%]	81.68 <sup>a</sup> $\pm$ 5.90	56.02 <sup>b</sup> $\pm$ 12.92
Salinity [‰]	0.64 <sup>a</sup> $\pm$ 0.05	0.62 <sup>a</sup> $\pm$ 0.04
Redox potential [mV]	107.69 <sup>a</sup> $\pm$ 14.27	107.45 <sup>a</sup> $\pm$ 13.00
Conductivity [ $\mu$ S cm <sup>-1</sup> ]	1,245.45 <sup>a</sup> $\pm$ 106.95	1,215.93 <sup>a</sup> $\pm$ 63.73
pH	7.64 <sup>a</sup> $\pm$ 0.18	7.59 <sup>a</sup> $\pm$ 0.10
NH <sub>3</sub> -N [mg L <sup>-1</sup> ]	0.16 <sup>a</sup> $\pm$ 0.13	0.16 <sup>a</sup> $\pm$ 0.11
NO <sub>2</sub> -N [mg L <sup>-1</sup> ]	0.09 <sup>a</sup> $\pm$ 0.05	0.21 <sup>b</sup> $\pm$ 0.15
NO <sub>3</sub> -N [mg L <sup>-1</sup> ]	40.54 <sup>a</sup> $\pm$ 10.97	31.06 <sup>b</sup> $\pm$ 5.17
Phosphate [mg L <sup>-1</sup> ]	5.55 <sup>a</sup> $\pm$ 1.97	5.22 <sup>a</sup> $\pm$ 1.36
Suspended particles [mg L <sup>-1</sup> ]	9.77 <sup>a</sup> $\pm$ 4.13	3.20 <sup>b</sup> $\pm$ 1.71

Means ( $\pm$  SD), different letters in groups showing significant differences ( $p < 0.05$ ).

**Fish growth parameters.** The biomass weight gain of *C. gariepinus* and *O. niloticus* was nearly the same, with 13,774.00 g (cycle I) and 13,294.00 g (cycle II, Table 3). The same was found with the feed conversion ratio (FCR), 1.00 for *C. gariepinus* and 1.03 for *O. niloticus*, with an overall amount of 13,725.00 g of feed over 83 days and 200 g d<sup>-1</sup> over 53 experimental days in cycle I and II, respectively. No mortality was observed. The individual growth parameters (Table 4) were different ( $p < 0.05$ ) between *C. gariepinus* and *O. niloticus*, with an initial body weight of 480.23 g ( $\pm 75.68$ ) and 173.51 g ( $\pm 14.64$ ), a final body weight of 873.77 g ( $\pm 165.00$ ) and 337.63 g ( $\pm 54.04$ ), and a weight gain of 393.54 g ( $\pm 171.21$ ) and 164.01 g ( $\pm 55.25$ ), respectively. The specific growth ratios (SGR) were not significantly different in *C. gariepinus* (0.65% d<sup>-1</sup>  $\pm 0.25$ ) and *O. niloticus* (0.71% d<sup>-1</sup>  $\pm 0.19$ ). The fish gross biomass of both species was very similar (Figure 4), but the individual weight gain differed significantly between *C. gariepinus* and *O. niloticus*. The curves followed  $y = 4.1019x + 442.34$  ( $R^2 = 0.86$ ) for *C. gariepinus* and  $y = 1.6612x + 150.25$  ( $R^2 = 0.71$ ) for *O. niloticus*.

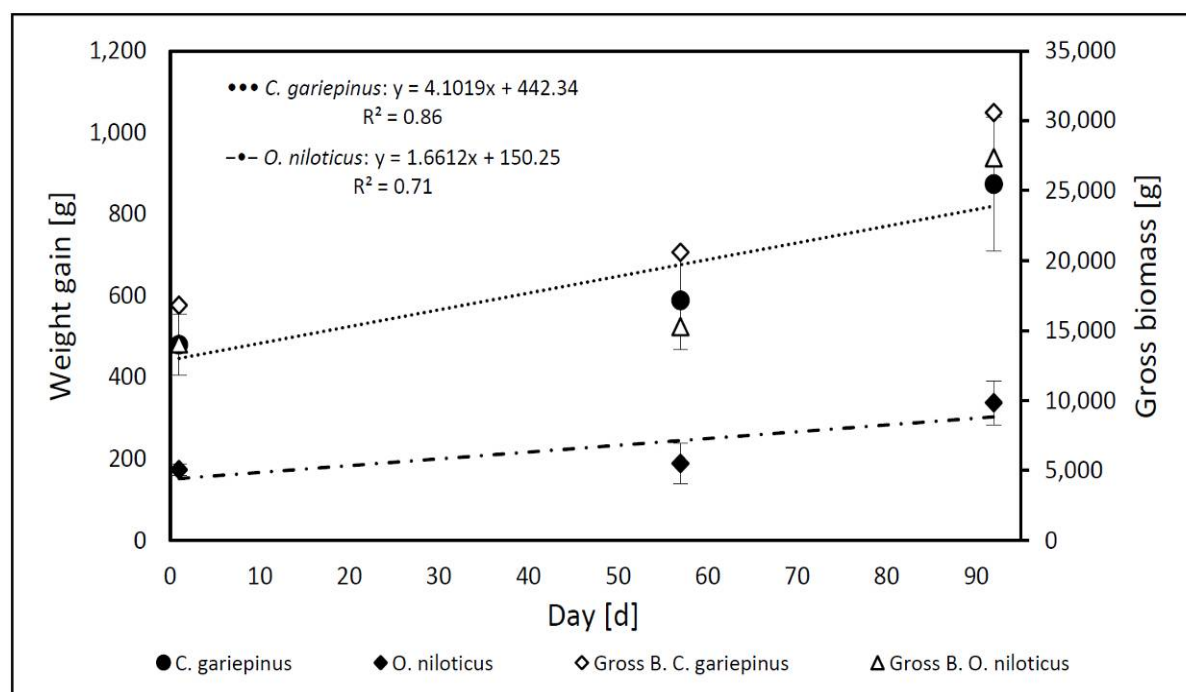


Figure 4. Individual weight gain [g] of *C. gariepinus* and *O. niloticus* (Mean  $\pm$  SD) and gross biomass (Gross B.) [g] after 92 days.

Table 3

Fish biomass parameters (total values) of *C. gariepinus* (Cycle I) and *O. niloticus* (Cycle II)

Parameter	Cycle I	Cycle II
Fish species	<i>C. gariepinus</i>	<i>O. niloticus</i>
Initial biomass [g]	16,808.00	14,054.00
Final biomass [g]	30,582.00	27,348.00
Initial stocking density [kg m <sup>-3</sup> ]	6.72	5.62
Final stocking density [kg m <sup>-3</sup> ]	12.23	10.94
Biomass weight gain [g] <sup>1</sup>	13,774.00	13,294.00
FCR <sup>2</sup>	1.00	1.03
Mortality [%]	0.00	0.00

<sup>1</sup> Biomass weight gain [g] calculated as difference from final biomass [g] and initial biomass [g];

<sup>2</sup> Feed conversion ratio (FCR) calculated as feed assignment [g] and biomass weight gain [g]<sup>-1</sup>.

Table 4

Individual fish growth parameters of *C. gariepinus* (Cycle I) and *O. niloticus* (Cycle II)

Parameter	Cycle I	Cycle II
	Mean ± SD	Mean ± SD
Initial body weight [g]	480.23 <sup>a</sup> ±75.68	173.51 <sup>b</sup> ±14.64
Final body weight [g]	873.77 <sup>a</sup> ±165.00	337.63 <sup>b</sup> ±54.04
Weight gain [g]	393.54 <sup>a</sup> ±171.21	164.01 <sup>b</sup> ±55.25
SGR <sup>1</sup> [% day <sup>-1</sup> ]	0.65 <sup>a</sup> ±0.25	0.71 <sup>a</sup> ±0.19

Means (± SD), different letters in groups showing significant differences (p < 0.05). <sup>1</sup> SGR = specific growth ratio [% d<sup>-1</sup>] = (ln W<sub>t</sub> - ln W<sub>0</sub>) × 100 days<sup>-1</sup>, with 83 days (acclimatisation phase of 30 days and 53 experimental days).

**Plant growth parameters.** The highest growth was found for tomato and cucumber in both cycles (Figures 5, 6), followed by basil and lettuce. In general, all plants showed better growth performances in cycle II in combination with *O. niloticus* (Table 5).

Table 5

Plant growing parameters [g] of lettuce, cucumber, tomato and basil of cycle I (*C. gariepinus*) and cycle II (*O. niloticus*). PAR = Photosynthetically Active Radiation [μmol m<sup>2</sup> s<sup>-1</sup>]

Parameter	Plant species	Cycle I Mean ± SD	Cycle II Mean ± SD
Plant weight [g]	lettuce	28.89 <sup>a</sup> ±48.07	55.89 <sup>b</sup> ±49.77
Fruit weight [g]		-	-
Plant gross biomass [g] <sup>1</sup>		28.89 <sup>a</sup> ±48.07	55.89 <sup>b</sup> ±49.77
Biomass [g plant <sup>-1</sup> ] <sup>2</sup>		1.61	3.11
PAR [μmol m <sup>2</sup> s <sup>-1</sup> ]		17.17 <sup>a</sup> ±31.76	4.28 <sup>a</sup> ±6.65
Plant weight [g]	cucumber	280.93 <sup>a</sup> ±342.28	217.07 <sup>a</sup> ±276.45
Fruit weight [g]		44.67 <sup>a</sup> ±134.41	168.27 <sup>b</sup> ±350.88
Plant gross biomass [g] <sup>1</sup>		325.60 <sup>a</sup> ±374.86	385.33 <sup>a</sup> ±400.00
Biomass [g plant <sup>-1</sup> ] <sup>2</sup>		21.71	25.69
PAR [μmol m <sup>2</sup> s <sup>-1</sup> ]		39.07 <sup>a</sup> ±37.92	142.40 <sup>b</sup> ±31.70
Plant weight [g]	tomato	681.67 <sup>a</sup> ±481.73	733.67 <sup>a</sup> ±301.20
Fruit weight [g]		0.33 <sup>a</sup> ±1.29	0.47 <sup>a</sup> ±1.36
Plant gross biomass [g] <sup>1</sup>		682.00 <sup>a</sup> ±482.45	734.13 <sup>a</sup> ±301.42
Biomass [g plant <sup>-1</sup> ] <sup>2</sup>		45.47	48.94
PAR [μmol m <sup>2</sup> s <sup>-1</sup> ]		50.73 <sup>a</sup> ±53.22	63.87 <sup>a</sup> ±24.35
Plant weight [g]	basil	117.94 <sup>a</sup> ±92.30	159.00 <sup>a</sup> ±96.91
Fruit weight [g]		-	-
Plant gross biomass [g] <sup>1</sup>		117.94 <sup>a</sup> ±92.30	159.00 <sup>a</sup> ±96.91
Biomass [g plant <sup>-1</sup> ] <sup>2</sup>		6.55	8.83
PAR [μmol m <sup>2</sup> s <sup>-1</sup> ]		23.33 <sup>a</sup> ±42.32	50.17 <sup>b</sup> ±49.62

Means (± SD), different letters in groups showing significant differences (p < 0.05). <sup>1</sup> Plant gross biomass [g] calculated with plant weight [g] and fruit weight [g]. <sup>2</sup> Biomass [g plant<sup>-1</sup>] calculated from plant gross biomass and N (lettuce: N = 18, cucumber: N = 15, tomato: N = 15, basil: N = 18).

Significant differences were seen in lettuce (plant weight and plant gross biomass:  $28.89 \text{ g} \pm 48.07$ , cycle I;  $55.89 \pm 49.77$ , cycle II) and the fruit weight of cucumber ( $44.67 \text{ g} \pm 134.41$ , cycle I;  $168.27 \text{ g} \pm 350.88$ , cycle II). The growth per single tomato plant was less than recorded during the exponential phase and the steady phase in the former experiment (Palm et al 2014) with  $45.47 \text{ g plant}^{-1}$  in cycle I and  $48.94 \text{ g plant}^{-1}$  in cycle II (Table 6).

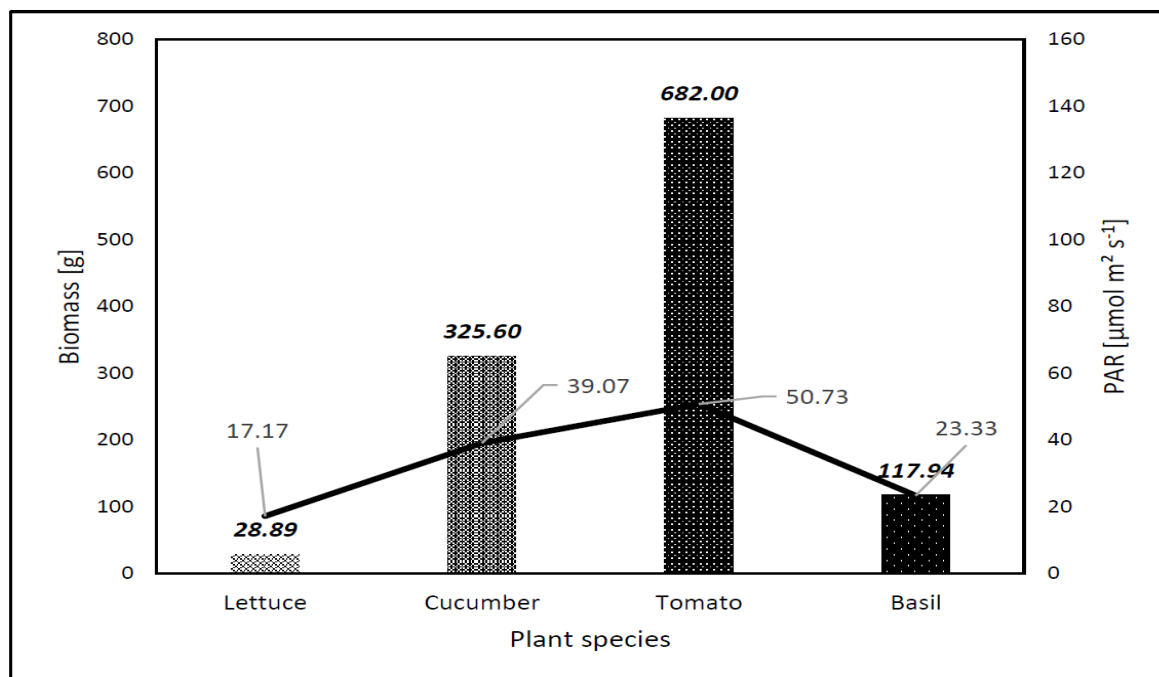


Figure 5. Cycle I (*C. gariepinus*), gross biomass of lettuce, cucumber, tomato, basil [g] and light distribution of Photosynthetically Active Radiation (PAR) [ $\mu\text{mol m}^{-2} \text{s}^{-1}$ ].

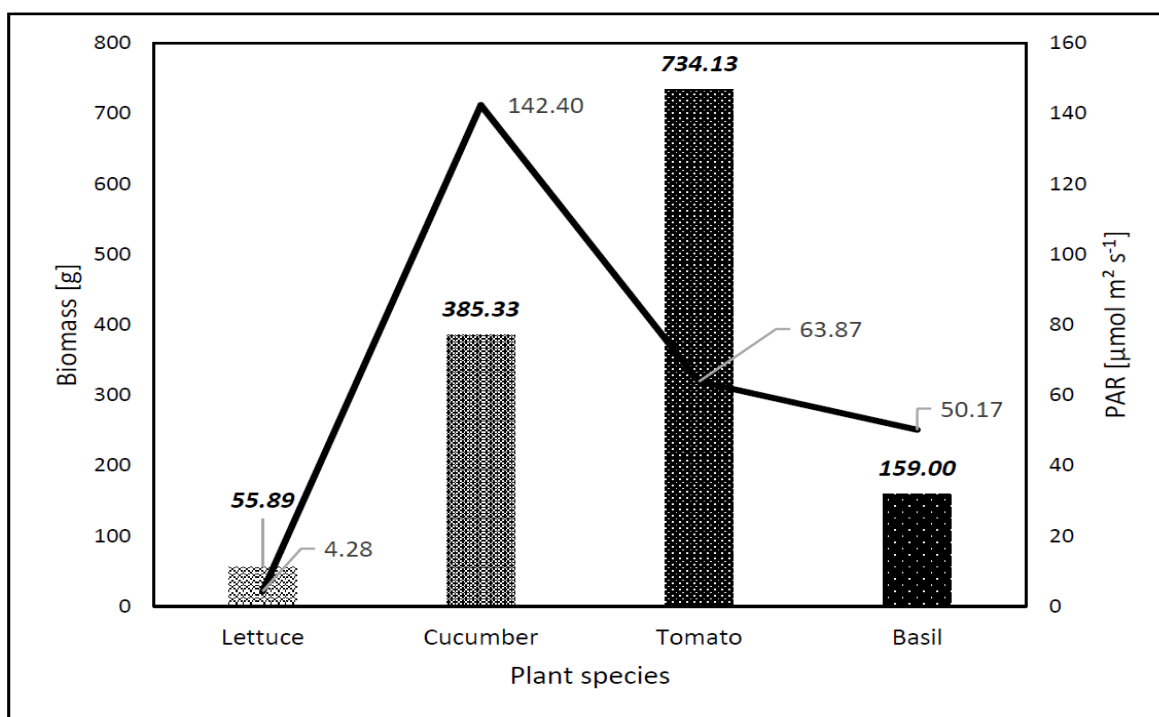


Figure 6. Cycle II (*O. niloticus*), gross biomass of lettuce, cucumber, tomato, basil [g] and light distribution of Photosynthetically Active Radiation (PAR) [ $\mu\text{mol m}^{-2} \text{s}^{-1}$ ].



The photosynthetic active radiation light (PAR) was significantly different in cucumber ( $39.07 \mu\text{mol m}^2\text{s}^{-1} \pm 37.92$ , cycle I;  $142.40 \mu\text{mol m}^2\text{s}^{-1} \pm 31.70$ , cycle II) and basil ( $23.33 \mu\text{mol m}^2\text{s}^{-1} \pm 42.32$ , cycle I;  $50.17 \mu\text{mol m}^2\text{s}^{-1} \pm 49.62$ , cycle II). The highest values were shown in cycle II for cucumber, with  $142.40 \mu\text{mol m}^2\text{s}^{-1}$ , followed by tomato, with  $63.87 \mu\text{mol m}^2\text{s}^{-1}$ , and basil, with  $50.17 \mu\text{mol m}^2\text{s}^{-1}$ . In cycle I, the highest amount of PAR was recorded for tomato with  $50.37 \mu\text{mol m}^2\text{s}^{-1}$ .

Table 6  
Plant growth of tomato in cycle I (*C. gariepinus*) and cycle II (*O. niloticus*) compared with the three subexperiments (SEI, II, III) of Palm et al (2014)

Experimental trials	SE I	SE II	SE III	Cycle I	Cycle II
Gross biomass [g]	158.70	7,893.90	11,973.50	682.00	734.13
Biomass [g plant <sup>-1</sup> ]	2.65	131.57	199.56	45.47	48.94

## Discussion

**Water parameters.** The chemo-physical water parameters for growing *O. niloticus* and *C. gariepinus* were optimal (Pullin & Lowe-McConnell 1982) during the entire run of the experiment. The temperature was in its optimal range and did not distinctly differ between the cycles. The dissolved oxygen (DO) levels, however, differed significantly ( $p < 0.05$ ), with  $6.95 \text{ mg L}^{-1} (\pm 0.50)$  for *C. gariepinus* and  $4.73 \text{ mg L}^{-1} (\pm 1.05)$  for *O. niloticus*, or 32% less for the *O. niloticus*, compared with the *C. gariepinus*. *O. niloticus* have been reported to be very tolerant of reduced levels of dissolved oxygen, and a short-term DO limit of 0.1 ppm was the minimum requirement for *O. niloticus* (Pullin & Lowe-McConnell 1982). Oxygen saturation levels below 32% and 25%, however, have a growth-limiting effect for *Sarotherodon mossambicus* (Pullin & Lowe-McConnell 1982). The observed minimum oxygen levels for *O. niloticus* did not reach less than  $4.73 \text{ mg L}^{-1}$  and 56.02% (cycle II) in the present study. Similarly, *C. gariepinus* is highly tolerant of low oxygen concentrations in aquaculture due to their special air-breathing organ (Oellermann 1996). Consequently, with oxygen saturation levels of 81.68% and  $6.95 \text{ mg L}^{-1}$  for *C. gariepinus*, oxygen levels met the requirements for both studied fish species. It is interesting to note that under identical cultivation conditions, the difference in DO levels between the cycles was  $2.22 \text{ mg L}^{-1}$  (32%).

During the steady state condition and a stable fish feed input of 200 g, no negative effects of salinity, conductivity and pH were observed. The  $\text{NH}_3\text{-N}$  concentration of  $0.16 \text{ mg L}^{-1} (\pm 0.13)$  for *C. gariepinus* was still two times smaller than that recommended by Schram et al (2010), with levels of  $0.34 \text{ mg NH}_3\text{-N mg L}^{-1}$  and no negative effects on *C. gariepinus* growth. Significant differences were found for nitrite and nitrate concentrations between both cycles (*C. gariepinus*: nitrite  $0.09 \text{ mg L}^{-1} \pm 0.05$ , nitrate  $40.54 \text{ mg L}^{-1} \pm 10.97$ ; *O. niloticus*: nitrite  $0.21 \text{ mg L}^{-1} \pm 0.15$ , nitrate  $31.06 \text{ mg L}^{-1} \pm 5.17$ ). For *C. gariepinus*, Roques et al (2013) advised a maximum water nitrite level of  $0.6 \text{ mg L}^{-1}$ , and Schram et al (2012) recorded an upper level for nitrate of  $140 \text{ mg L}^{-1}$ . For *O. niloticus*, El-Sherif & El-Feky (2008) found a lethal level of un-ionized ammonia ( $\text{NH}_3\text{-N}$ , 48 hr-LC50) of  $7.1 \text{ mg L}^{-1}$ , whereas fish begin to die already at ammonia concentrations around  $2 \text{ mg L}^{-1}$  (Rakocy 1989b). Nitrite is tolerated below  $2.1 \text{ mg L}^{-1}$  (Lim & Webster 2006) and nitrate up to 300-400  $\text{mg L}^{-1}$  (DeLong et al 2009).

A stable fish feed amount of 200 g per day resulted in decreasing phosphate levels in both cycles (Figure 3), following an increasing plant biomass during the run of the experiment. We can categorize our ebb and flow aquaponic system as a low-nutrient and phosphate-limited system, directly depending on the level of fish feed input. The limited fish feed resulted in low nutrient availability in comparison with a classical separate hydroponic culture, only controlled by the respective fish digestion physiology.

**Fish growth.** All water parameters enabled an optimal animal welfare and growth performance of both fish species, resulting in no mortality. In general, the fish growth is correlated to the protein content [%] of the feed and the feed composition. Protein

requirement in fish is reduced with age. Abdel-Tawwab et al (2010) recorded the optimum growth performance of tilapia fry (0.4-0.5 g) at 45% crude protein, and a reduced requirement of 35% crude protein for the fingerlings (17-22 g) and advanced juveniles (37-43 g). Ali & Jauncey (2004b) showed better growth rates and feed efficiency for *C. gariepinus* fry (13.45 g  $\pm$  0.05 g) at protein levels of 35% and 40%. Similarly, the carbohydrate content ranged between 27-38% and the lipid content from 11-16% for small fish (12.32 g, Ali & Jauncey 2004a). The fish feed crude components, in the present study, contained 47% protein and 14% lipid, and was nearly at optimum levels for the cultured species.

The FCRs were close to optimal conditions for both fish species in batch cultivation, *C. gariepinus* (FCR = 1.00) and *O. niloticus* (1.03). Better feed conversion ratios of 0.83 and 0.93 with *O. niloticus* fry (0.50 g) in the same system were found with smaller fish, an increasing feed input and a longer experimental time interval (Palm et al 2014). Earlier experiments on closed floating raft aquaponic systems showed much lower feed conversion ratios, with 1.7 (initial weight 79.2 g) for younger monosex *O. niloticus* and 1.8 (initial weight 58.8 g) for Red tilapia in the UVI floating raft system (Rakocy et al 2006). Al-Hafedh et al (2008), adopting the Rakocy-UVI system, also reported a lower FCR of 1.4 for staggered mixed sex *O. niloticus* populations (initial weights of 42.5 g, 74.8 g, 138 g, 248 g), and 1.3 for all male *O. niloticus* in a total of 8 harvests and a year round production of leaf lettuce. Savidov (2005b) recorded an FCR of 1.3 with the staggered production of initially 100 g *O. niloticus* juveniles and different plants like cucumber, tomato or basil, also in an UVI aquaponic system. An experimental raft aquaponic system with an ordinary clarifier, *O. niloticus* adults (376.0 g) and water spinach (*Ipomoea aquatica*) had an FCR of 3.8, compared with the same system connected to a swirl separator, with an insignificant FCR of 2.5 ( $p < 0.05$  *t*-test, Danaher et al 2013). Consequently, our system performed much better than recorded in earlier studies on closed aquaponic systems.

Chowdhury (2011) reported an optimal feeding regime for *O. niloticus* with 3% of the biomass for 80-115 g and larger fish, and 1.2% for fish over 260 g. A feed ratio of 200 g per day in the present study equals 1.42% of the biomass for fish with an initial weight of 173.51 g and is close to the optimum for *O. niloticus*. *C. gariepinus*, with an initial weight of 480.23 g ( $\pm$  75.68) were fed with 1.19% of the initial biomass per day. Rueda (2004) reported a better feed conversion ratio of 0.73 for *C. gariepinus* (initial body weight: 55.00 g; final body weight: 432.1 g) held under different light regimes for six weeks in aquaria of 120 L. *C. gariepinus* tends to have a better growth performance, particularly under non-continuous illumination (12 h darkness, 12 h light), seen in better specific growth ratios (4.7-4.9%  $d^{-1}$ ). Also, *C. gariepinus* juveniles (83.0-198.0 g) showed better feed conversion ratios of 0.8-0.9 in homogenous, as well as in heterogeneous, groups (Martins et al 2005). However, it must be kept in mind that our fish were not fed *ad libitum*, thus limiting the growth performance of the *C. gariepinus* compared with earlier studies.

The specific growth ratios (SGR) of the fish in the present study were not significantly different with 0.65%  $d^{-1}$  in *C. gariepinus* and 0.71%  $d^{-1}$  in *O. niloticus* (4.74 g and 1.97 g  $d^{-1}$  respectively). In the same system, younger fish showed better daily growth rates with insignificant SGR's of 3.04%  $d^{-1}$  and 2.98%  $d^{-1}$  in *O. niloticus* postlarvae (0.50 g initial body weight, Palm et al 2014). A similar result was reported by Rakocy et al (2006) for other closed floating raft aquaponic systems, with 4.4 g  $d^{-1}$  in *O. niloticus* (79.2 g initial weight) and 2.7 g  $d^{-1}$  in Red tilapia (58.8 g initial weight). Better SGRs of 1 and 1.1%  $d^{-1}$  were found by Al-Hafedh et al (2008) for mixed sex and all-male *O. niloticus* in a staggered round year production (8 harvests). For only seven harvests, the average growth rates were 1.2 g fish $^{-1}$   $d^{-1}$  for mixed sex *O. niloticus* and 1.5 g fish $^{-1}$   $d^{-1}$  for all male population. Advanced juveniles (37-43 g) performed similar to our preadult *O. niloticus*, with an SGR of 0.71%  $d^{-1}$  (initial weight: 173.51 g) and 0.6%  $d^{-1}$  respectively (Abdel-Tawwab et al 2010). Adult *C. gariepinus* of 432.1 g final body weight (six weeks experimental time in aquaria) demonstrated a higher specific growth ratio of 4.8%  $d^{-1}$  (Rueda 2004), seven times higher than in the present study (with 0.65%  $d^{-1}$  and a final body weight of 873.77 g). This can be explained by the age difference and

growth conditions resulting from the limited feed input (200 g), combined with the steady growth of the fish.

Comparing the growth performances of both fish, *C. gariepinus* showed significantly greater individual weight gain (393.54 g  $\pm$  171.21), compared with *O. niloticus* (164.01 g  $\pm$  55.25, Figure 4), but with, however, nearly the same biomass weight gain of 13,774.00 g for the *C. gariepinus* and 13,294.00 g for *O. niloticus* (Table 3). Because the feed intake for both fish species was the same, they converted all given feed into biomass, with the stable feed amount of 200 g d<sup>-1</sup> restricting the growth, independent of the cultured species. The observed differences in growth performance can be explained by the different fish species physiology, the initial biomass and the size of the fish, combined with the restricted feed amount. An earlier investigation of the present aquaponic system found that the optimum feed input level is 200 g per day for Nile tilapia (Palm et al 2014). A fish feed overload (> 200 g) resulted in malperformance of the system, and a growth depression from decreasing oxygen levels. During the present study, oxygen values were more stable in cycle I ( $y = -0.0226x + 7.5191$ ,  $R^2 = 0.6227$ ) and cycle II ( $y = -0.0384x + 5.6885$ ,  $R^2 = 0.4103$ , Figure 2) but with a clear negative linear correlation. A higher fish feed amount would decrease the oxygen levels to more critical values for the fish, with the risk of growth stagnation.

**Plant growth.** The plants performed differently during the run of the experiment, with better weight gain (plant gross biomass) in the cultivation of *O. niloticus* (cycle II, Figure 6) in contrast to *C. gariepinus* (Figure 5). Significant differences in plant growth occurred in lettuce gross biomass (*C. gariepinus*: 28.89 g  $\pm$  48.07 and *O. niloticus*: 55.89 g  $\pm$  49.77) and cucumber fruit weight (*C. gariepinus*: 44.67 g  $\pm$  134.41 and *O. niloticus*: 168.27 g  $\pm$  350.88). Tomato and basil growth were not significantly different between the cycles, but also had a positive trend in the cycle with *O. niloticus*. It is important to note that the growth of tomato was lower in both cycles compared with an earlier experiment with *O. niloticus* (Table 6) (Palm et al 2014). Reasons can be seen in a different cultivation period and light regime (March-August vs December-March in the present study), an increasing feed input per day in each sub-experiment (SE) with 24.85 g in SE I, 131.12g in SE II and 221.72 g in SE III compared with a steady feed input of 200 g d<sup>-1</sup>. Also in the former investigation, a possible accumulation of plant nutrient matters over the entire experimental time of 160 days could have taken place (although plants were seeded newly in each SE) compared with a shorter run of the experiment in the present study.

Our aquaponic systems were built in a glasshouse in an east to west orientation, with the hydroponic systems arranged, in parallel, south to north. The southern aquaponic unit (cycle II) with *O. niloticus* was slightly more exposed to light during daytime, which might have had an effect on plant growth. The exposure to photosynthetic active radiation light (PAR) during night was also significantly different for cucumber and basil, and in total values, higher in cycle II than in cycle I. The light illumination might be a reason for the observed differences of the plant growth between the cycles, though an expected better growth of cucumber gross biomass under the better light regime in cycle II (*O. niloticus*) did not occur. However, significant differences were recorded for the fruit weight of cucumber, with more biomass in cycle II with the *O. niloticus*. Other reasons for the observed effects might be the nutrient concentration that differed between the cycles. Ammonia levels (NH<sub>3</sub>-N) were the same for cycle I and cycle II (0.16 mg L<sup>-1</sup>), but differed significantly for nitrite (*C. gariepinus*: 0.09 mg L<sup>-1</sup>  $\pm$  0.05) in contrast to the 2.3 times higher values for *O. niloticus* (0.21 mg L<sup>-1</sup>  $\pm$  0.15). This suggests reduced oxidation of nitrite in cycle II, and a possible disparity of bacteria activity. Nitrite accumulation as a consequence of e.g. *Nitrobacter* inhibition is a known phenomenon in biofilters (Van Rijn 1996), caused by oxygen limitation through accumulation of organic matter. Other factors for nitrite accumulation are suboptimal pH, substrate and product inhibition or light intensity (Van Rijn 1996). pH values were insignificant and at optimum levels for nitrification (cycle I: 7.64  $\pm$  0.18 and cycle II: 7.59  $\pm$  0.10). Organic matter was accumulated in cycle I with the *C. gariepinus*, with three fold more suspended particles (9.77 mg L<sup>-1</sup>) in contrast to the *O. niloticus* cycle. According to

our data, higher oxygen levels, better nitrification and nitrate availability would support plant growth in cycle I with *C. gariepinus*. Less organic load and slightly higher light availability during day and night caused by the system orientation in the glasshouse would be beneficial for the *O. niloticus* system. The real importance of these adverse effects cannot be decided at present, including possible effects of a different fish physiology and feed digestion. More studies are needed to explain the plant performance in such closed aquaponic systems.

**Conclusions.** The present study describes a balanced low-tech ebb and flow aquaponic system, limiting the optimal fish feed input level to 200 g per day for the cultivation of warm- and freshwater fish. The water parameters, within an optimal range for both fish species, resulted in good animal welfare under batch cultivation. Fish growth was close to the optimum level for *O. niloticus*, with better FCR (*C. gariepinus* 1.00 and *O. niloticus* 1.03, no mortality) values compared with earlier tested closed aquaponic systems. In contrast, *C. gariepinus* had a lower performance due to the limited feed input. The effect of constant feed input resulted in nearly the same biomass weight gain of 13 kg, demonstrating that all given feed was converted into biomass, independent of the cultured fish species. Plant growth was better under cultivation of *O. niloticus*, possibly an effect of light orientation and a different fish physiology. A different total oxygen level in both systems (32%) demonstrates different oxygen consumption, an effect of the cultivated fish species with relevance for the system performance. The possible impact of the fish physiology on the observed water parameters, the availability of plant nutrients in the system and the general performance of the combined fish and plant cultivation must be kept in mind in order to achieve economic sustainability in closed aquaponics.

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