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ABSTRACT
Total suspended solids control was evaluated in a channel catfish (*Ictalurus punctatus*) biofloc technology production system. Settling chamber flow rates were 0.9 (LO) or 2.9 (HI) L/min to reduce total suspended solids to 300 mg/L; solids were not removed from control tanks. Channel catfish yields (7.6–8.7 kg/m^3) were not affected significantly, but control fish were skewed toward smaller size classes. Control treatment channel catfish tolerated 1,410 mg/L total suspended solids without adverse effects. LO- and HI-treatment fillet geosmin concentrations were high enough to be designated as off-flavor. Water quality results suggested that nitrification was affected by solids removal.

KEYWORDS
Biofloc technology; channel catfish; total suspended solids; geosmin; MIB

Introduction

High daily feeding rate in response to high stocking rate is one characteristic of the biofloc technology (BFT) production system. In the BFT system, mean daily feed rate ranges from 78 to 88 g/m^3 for white shrimp (*Litopenaeus vannamei*) (Gaona et al. 2017; Ray, Dillon, and Lotz 2011; Ray et al. 2010; Schveitzer et al. 2013), from 58 to 88 g/m^3 for channel catfish (*Ictalurus punctatus*) (Green and Schrader 2015; Green, Schrader, and Perschbacher 2014; Schrader, Green, and Perschbacher 2011), and from 150 to 250 g/m^3 for tilapia (*Oreochromis* spp.) (Azim and Little 2008; Milstein et al. 2001). In contrast, mean daily feeding rate in intensively managed earthen ponds rarely exceeds 100 kg/ha (approximately 7–11 g/m^3 depending upon mean pond depth). Concomitant with the high feeding rate is an increase in total suspended solids (TSS) in the BFT system. In the absence of solids removal, TSS and feed input, nitrate-nitrogen, or soluble reactive phosphorus are positively correlated (Green and Schrader 2015; Green, Schrader, and Perschbacher 2014). While

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removing solids from a shrimp BFT system affected significantly concentrations of water quality variables (Ray, Dillon, and Lotz 2011; Ray et al. 2010), the effect of solids removal on water quality in a catfish BFT system remains to be investigated.

Increased TSS concentration can be detrimental to culture animal performance. Weekly means of daily feed consumption by channel catfish being grown to stocker size in an outdoor BFT system ceased to increase once TSS concentration exceeded 560–850 mg/L (Green, Schrader, and Perschbacher 2014). These authors reported some mortality and abnormal swimming behavior once TSS exceeded 2,000 mg/L. Reducing TSS to 300 mg/L or less using settling chambers to remove solids improves shrimp growth and yield (Gaona et al. 2017; Ray, Dillon, and Lotz 2011; Ray et al. 2010; Schveitzer et al. 2013; Vinatea et al. 2010). Use of settling chambers in the channel catfish BFT system has not been tested but is warranted given the reduced feed consumption (and presumably, reduced growth) observed at high TSS concentrations (Green, Schrader, and Perschbacher 2014).

The microbial off-flavor compounds geosmin and 2-methylisoborneol (MIB) can accumulate in fish flesh and impart "earthy" and "musty" off-flavors respectively. Episodes of earthy and musty off-flavors can render fish unmarketable and has been estimated to cost U.S. catfish producers $10–60 million annually (Tucker 2000). Geosmin and MIB have been detected in the fillets of channel catfish reared in the outdoor BFT system without solids control, but the episodes and intensities of geosmin- and MIB-induced off-flavors are substantially lower compared to catfish grown in earthen ponds (Green and Schrader 2015; Green, Schrader, and Perschbacher 2014; Schrader, Green, and Perschbacher 2011). Actinomycetes in the genera *Norcardia* and *Streptomyces* are present in the biosolids found in multiple components of freshwater RAS systems and are attributed as the main sources of geosmin and MIB (Guttman and van Rijn 2008; Schrader and Summerfelt 2010). Biofloc particles in the catfish BFT system may provide a good substrate for actinomycete colonization given the positive correlations detected between geosmin or MIB and TSS in tank water (Green, Schrader, and Perschbacher 2014). However, there are no data on the effects of solids removal on geosmin and MIB concentrations in fillets from catfish grown in the BFT system.

The objective of this study was to evaluate the effect of three solids management protocols in an outdoor photoautotrophic-chemoautotrophic (mixotrophic) BFT production system on channel catfish production, the incidence and intensity of geosmin and MIB off-flavor in catfish fillets, and on water quality.

Materials and methods

This experiment to determine the effect of solids management was carried out in nine 15.7-m^3 wood-framed, high-density polyethylene-lined tanks (described in detail by Green, Schrader, and Perschbacher 2014) located outdoors at the United States Department of Agriculture (USDA), Agricultural Research Service (ARS), Harry K. Dupree Stuttgart National Aquaculture Research Center (SNARC), Stuttgart, Arkansas, USA. During the 1–16 October 2013 U.S. government shutdown (days 165 to 180), the one essential employee (McEntire) authorized to work cared for all fish at SNARC. For this experiment, he ensured fish well-being, fed fish, drained settling chambers every two days, and maintained tank water levels as needed. All other experiment activities were suspended.

Each tank was equipped with a 117-L (operating volume) conical bottom settling chamber. Water was moved from the tank to the settling chamber by airlift pump; air injection depth controlled the flow rate. Two flow rates (0.9 and 2.9 L/min; LO and HI respectively) were the treatments tested; hydraulic retention time was 11.8 and 3.7 d respectively for the tanks and 127 and 40 min respectively for the settling chambers. Solids were not removed from control tanks. Three tanks were assigned randomly to each treatment. Settling chamber operation began on study day 52, once TSS concentration approximated 400 mg/L (Green, Schrader, and Perschbacher 2014), and continued uninterrupted through the end of the experiment. Settled solids were drained from chambers on average every 3 d, and the solids removed were quantified (as dry matter) based on sludge TSS. The mean quantity of sludge discarded from each chamber every 2 days during the government shutdown was assumed to be the mean quantity removed on days 154, 158, 161, and 185. Solids retention time (SRT; days) was calculated each time solids were discharged as: SRT = (V * TSS) / S_d, where V = system volume (m^3), tank TSS (g/m^3), and S_d = solids discharged (g/d) (Tchobanoglous, Burton, and Stensel 2003).

Tanks were filled on 8 April 2013 with groundwater (total alkalinity = 226.4 mg/L as $CaCO_3$) and 2.3 m^3 of water from a SNARC pond containing an algal bloom to enhance the development of the algal bloom in order to help control ammonia levels initially. During the next 26 d each tank was dosed with a total of 0.5 kg 9–27-0 (N-P_2O_5-K_2O) and 0.4 kg 46–0-0 fertilizer, and 4.5 kg dried molasses and beet pulp (Sweet45; Westway Feed Products, New Orleans, Louisiana, USA), and treated with 2.5 kg $CaCl_2$ to ensure chloride concentration exceeded 100 mg/L. Feed-grade sodium bicarbonate (Church & Dwight, Co., Princeton, New Jersey, USA) was added to each tank as needed to maintain pH and total alkalinity following Loyless and Malone (1997). Water (ca. 0.9 m^3/d) was added to each tank on 27 occasions to replace losses from evaporation and draining of the settling chamber.

Channel catfish (2012 year class; 46.9 ± 12.4 g/fish) were stocked into tanks on 19 April 2013 at 15.0 fish/m^3 (0.74 ± 0.03 kg/m^3). Animal care and experimental protocol were approved by the SNARC Institutional Animal Care and Use Committee and conformed to ARS Policies and Procedures 130.4 and 635.1.

Fish in each tank were fed as much 32% protein feed (Delta Western Feed Mill, Indianola, Mississippi USA) as they could consume in 10 min (apparent satiation) and the quantity recorded. On days 63, 98, and 127 at least 20% of each tank's fish population was captured by seine net and weighed in bulk as two to three lots of at least 20 fish each, and fish were returned alive to their tank. Fish were harvested from all tanks on days 209–210. At harvest, a minimum of 75 fish/tank were weighed individually (to the nearest 0.1 g), and the remainder were counted and weighed in bulk (to the nearest 0.1 kg). The total fish biomass harvested per tank was the gross fish yield (GFY, kg/m^3), and the net fish yield (NFY, kg/m^3) was calculated as the final fish biomass minus the initial fish biomass. Individually weighed fish were assigned to size classes—kg/fish (size range): submarketable (< 0.34 kg/fish), out-of-size (0.34 to 0.45 kg/fish), 0.45 (0.45 to 0.57 kg/fish), 0.57 (0.57 to 0.68 kg/fish), 0.68 (0.68 to 0.79 kg/fish), 0.79 (0.79 to 0.91 kg/fish), 0.91 (0.91 to 1.02 kg/fish), and 1.02 (≥ 1.02 kg/fish). Feed conversion ratio (FCR) was calculated for each tank as the total quantity of feed fed on a dry matter basis divided by the net (wet) weight of fish harvested.

Five catfish were selected at random from each tank at harvest, euthanized immediately by cranial percussion, and filleted. Catfish fillets (one fillet/fish) were placed in individual plastic bags, vacuum sealed, and immediately frozen until overnight shipment to the USDA-ARS Natural Products Utilization Research Unit, University, Mississippi, USA, for analysis of geosmin and MIB levels. Fish fillets were maintained frozen until processed according to Lloyd and Grimm (1999) to obtain microwave distillates that were analyzed using the procedures of Lloyd et al. (1998) and as modified by Schrader et al. (2003) to quantify geosmin and MIB by solid phase microextraction and gas chromatography-mass spectrometry. Catfish samples were collected only at the end of the study to permit bioaccumulation of geosmin and/or MIB throughout the grow-out period and to ensure that adequate fillet material was available for analysis (>20 g/fillet).

Water samples were collected from each tank between 0700 and 0800 h and analyzed beginning 2 d before stocking fish and continuing at weekly intervals through the end of the experiment. Total ammonia-nitrogen (NH_4-N) was analyzed fluorometrically using the o-phthaldialdehyde method in a flow injection system (Genfa and Dasgupta 1989). Nitrite-nitrogen (NO_2-N, diazotization), nitrate-nitrogen (NO_3-N, cadmium reduction), and soluble reactive phosphorus (PO4-P; ascorbic acid method) were analyzed using flow injection analysis according to manufacturer instructions (FIAlab 2500; FIAlab Instruments, Bellevue, Washington, USA). Total alkalinity, settleable solids,

total suspended solids (TSS), volatile suspended solids (VSS), and settleable solids (SS) were measured using the methods of Eaton et al. (2005). Chlorophyll *a* was extracted in 2:1 chloroform:methanol from phytoplankton (for this study, "phytoplankton" includes planktonic algae and cyanobacteria as well as biofloc-associated algae) previously filtered from water samples by using a 0.45-µm pore size glass fiber filter, and the chlorophyll *a* concentration in the extract was determined by spectroscopy (Lloyd and Tucker 1988). Sample pH was measured electrometrically (Accumet AB15; Fisher Scientific, Waltham, Massachusetts, USA).

Accumulation of NO_3-N was used as a proxy for the quantity of NH_4-N oxidized per gram of TSS, an indicator of the solids-specific nitrification rate (SSNR; g NH_4-N/g TSS/d), and was estimated weekly using the change in NO_3-N concentration as follows: SSNR = ([NO_3-N_{T2} – NO_3-N_{T1}] / [T2 – T1])/ TSS, where T1 and T2 denote times 1 and 2 respectively in days (Water Environment Federation 1996).

Settling chamber effluent returning to each tank was sampled for TSS weekly beginning day 75 and continuing at 7–14-d intervals. Tank water TSS was assumed to represent settling chamber influent. The percent reduction in TSS was calculated based on settling chamber influent and effluent TSS.

Dissolved oxygen (DO) concentration and water temperature in each tank were monitored continuously by a galvanic oxygen sensor (Type III, Oxyguard, Birkerød, Denmark) and a thermistor (Model 109, Campbell Scientific, Logan, Utah, USA) connected to a datalogger (Model CR206 or CR1000, Campbell Scientific, Logan, Utah, USA).

After confirming homogeneity of variance and normality, fish production and water quality data were analyzed by mixed model analysis of variance (ANOVA) using PROC MIXED and regression analysis (PROC REG) in SAS version 9.4 (SAS Institute, Inc., Cary, North Carolina, USA). Regression line slopes determined for each tank were compared using PROC MIXED. The CONTRAST statement was used to compare treatment means in PROC MIXED. Settling chamber flow rate was the fixed effect and tank the random effect for the MIXED procedure. Percent data were log transformed prior to data analysis (Sokal and Rohlf 1995). To determine if there were associations between treatment and fish in each market size category, market size distributions were analyzed by PROC FREQ in SAS version 9.4 to produce Chi-square and likelihood ratios. Water quality data were divided into two periods for statistical analyses: (1) presettling chamber operation (days 1 to 51) and (2) settling chamber operation (days 52 to harvest). The PROC CORR procedure was used to calculate Spearman correlation coefficients within treatment for weekly NH_4-N and chlorophyll a concentrations and weekly averages of daily feed input. Differences among treatments were declared significant at $P \leq 0.05$.

Results

Daily feed consumption (as % of biomass) in all treatments decreased linearly as individual weight increased, and regression line slopes did not differ significantly ($P > 0.05$) among treatments. Daily feed consumption averaged 3.0% and 0.9% of biomass at the beginning and end of the experiment respectively. Sustained, high daily feed consumption (peak feed) occurred from day 51 to 148, did not differ significantly among treatments ($P = 0.240$), and averaged 99.3, 105.7, and 111.0 g/m^3 for the control, LO, and HI treatments respectively. Total feed consumption and FCR did not differ significantly among treatments (Table 1).

Mean individual fish weight at harvest ranged from 541.7 to 591.6 g/fish and did not differ significantly among treatments ($P = 0.384$; Table 1). Fish growth during the experiment was linear in all treatments, and analysis of growth curve slopes (data not shown) did not reveal significant treatment differences ($P = 0.362$).

Gross and net fish yields did not differ significantly among treatments (Table 1). Survival also did not differ significantly among treatments. An analysis of covariance, performed because LO treatment survival was 6.5% lower than the remaining treatments, indicated survival was not a covariate ($P = 0.430$). No significant treatment differences were detected for fillet geosmin ($P = 0.251$) or MIB ($P = 0.329$) concentrations (Table 1).

Distributions of harvested fish size differed significantly among treatments (Figure 1; $P < 0.001$). The fish population in the control treatment was skewed toward the smaller size classes, and frequency analysis results indicated a greater than expected number of fish in the submarketable, out-of-size, and 0.45 kg/fish size classes, and fewer than expected numbers in each of the larger size classes. Numbers of fish in each size class in the LO treatment approximated the expected numbers for all size classes. In the HI treatment, there were

Table 1. Least squares mean gross fish yield (GFY), net fish yield (NFY), total feed fed, individual weight, survival, feed conversion ratio (FCR), fillet geosmin (GSM), and 2-methylisoborneol (MIB) concentrations for channel catfish reared for 210 days in a biofloc technology production system subjected to two levels of settling chamber influent flow rate: LO = 0.9 L/min, HI = 2.9 L/min. Solids were not removed from control tanks.

	Treatment			Pooled	
Variable	Control	LO	HI	SE	*Pr > F**
GFY (kg/m^3)	7.7	7.6	8.7	0.4	0.119
NFY (kg/m^3)	6.9	6.9	8.0	0.4	0.124
Total Feed (kg/m^3)	13.7	14.4	15.4	0.5	0.147
Individual Weight (g/fish)	541.7	579.5	591.6	24.5	0.384
Survival (%)	96.7	90.4	97.2	0.4	0.281
FCR	1.7	1.6	1.6	0.1	0.443
Fillet GSM (ng/kg)	13	2,151	1,288	812	0.251
Fillet MIB (ng/kg)	19	50	32	13	0.329

*Probability value for the *F* statistic.

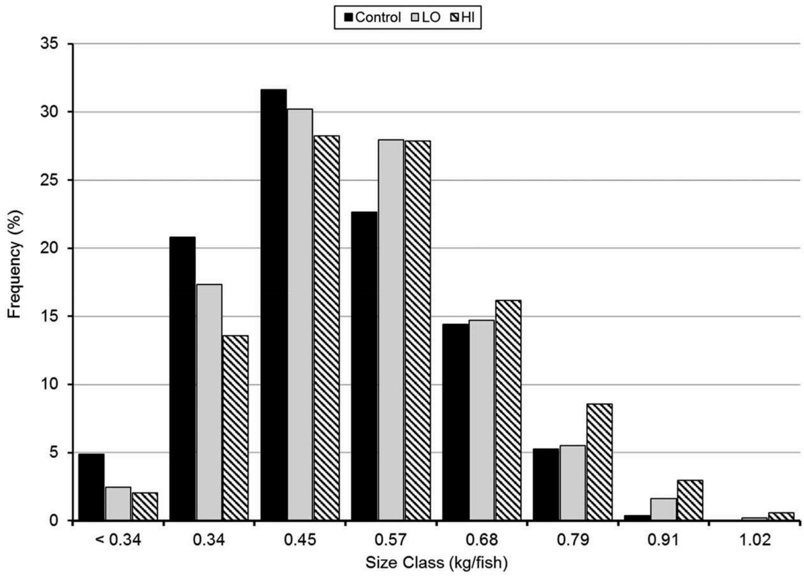

Figure 1. Size distribution of channel catfish harvested after 210 days from the biofloc technology production system control, LO, and HI treatments. Fish in the < 0.34 and 0.34 kg/fish size classes are considered submarketable and out-of-size respectively. Settling chamber flow rates were 0.9 L/min (LO) and 2.9 L/min (HI). Solids were not removed from control tanks.

fewer than expected submarketable and out-of-size fish, expected numbers of fish in the 0.45, 0.57, and 0.68 kg/fish size classes, and greater than expected numbers in the larger size classes.

Minimum and mean DO were 68% and 87% of saturation (5.2 and 7.4 mg/L) respectively and did not differ significantly ($P = 0.573$) among treatments. Mean water temperature did not differ among treatments over the entire experiment ($P = 0.594$; 24.4°C). No water quality treatment differences ($P > 0.05$) were detected during period 1 (days 1–51). Significant treatment differences were detected in mean water quality variable concentrations during period 2 (Table 2). Control treatment means generally were significantly greater than one or both exchange treatments. However, no significant treatment differences were detected for NH_4-N, chlorophyll *a*, and pH.

Mean chlorophyll *a* concentration in all treatments peaked at 2,100 to 2,500 mg/m^3 within the first month and by the end of the second month had declined and stabilized at 500 to 1,000 mg/m^3 during period 2 (Figure 2, top). Increases in mean TSS concentration were similar among treatments during period 1, and TSS concentrations stabilized between 200–400 mg/L in the LO and HI treatments during period 2 but continued to accumulate in the control treatment (Figure 2, middle). Increase in mean NO_3-N concentrations was similar among treatments until day 75, after which the curves diverged (Figure 2, bottom).

Table 2. Water quality least squares means during periods 1 (stock to day 51) and 2 (day 52 to harvest). The channel catfish biofloc technology production system was subjected to two levels of settling chamber influent flow rate: LO = 0.9 L/min, HI = 2.9 L/min. Solids were not removed from control tanks.

	Period 1					Period 2				
	Treatment					Treatment				
Variable*	Control	LO	HI	SE	*Pr > F*	Control	LO	HI	SE	*Pr > F*
NH_4-N	0.38	0.29	0.41	0.09	0.612	0.03	0.05	0.05	0.01	0.104
NO_2-N	1.02	0.84	1.60	0.31	0.264	0.08 c	0.18 b	0.33 a	0.03	0.003
NO_3-N	6.19	6.29	7.21	0.58	0.441	85.9 a	61.4 b	73.4 ab	5.5	0.021
PO_4-P	1.28	1.48	1.75	0.14	0.140	24.8 a	14.7 b	18.2 ab	1.9	0.029
Chl *a*	1,618	1,714	1,357	129	0.209	706	679	638	56	0.705
T Alk	198.5	196.9	182.6	8.8	0.430	103.2 a	98.0 a	78.8 b	3.6	0.007
pH	8.49	8.48	8.36	0.05	0.281	7.51	7.68	7.52	0.04	0.054
SS	14	14	16	2	0.719	65 a	17 b	14 b	1	< 0.001
TSS	242.8	247.5	259.3	12.8	0.661	829.4 a	306.3 b	305.0 b	29.3	< 0.001
VSS	213.1	217.2	218.4	10.5	0.933	672.2 a	268.6 b	263.1 b	21.2	< 0.001

*Total ammonia-nitrogen mg/L NH_4-N, nitrite-nitrogen mg/L NO_2-N, nitrate-nitrogen mg/L NO_3-N, soluble reactive phosphorus mg/L PO_4-P, chlorophyll *a* (Chl *a*) mg/m^3, total alkalinity (T Alk) mg/L as $CaCO_3$, settleable solids (SS) mL/L, total suspended solids (TSS) mg/L, volatile suspended solids (VSS) mg/L.

Regression line slopes for NO_3-N concentration over time during period 2 differed significantly among treatments. Control treatment mean slope (0.842 ± 0.069; ± standard error, SE) was significantly greater ($P < 0.010$) than for the LO treatment (0.477 ± 0.069) but not different ($P = 0.054$) from the HI treatment (0.608 ± 0.069). And HI and LO treatment mean slopes did not differ significantly ($P = 0.226$). The HI treatment mean slope was 28% smaller than the control and 22% larger than the LO treatment means. Estimated solids-specific nitrification rate during period 2 did not differ significantly ($P = 0.180$) among treatments and averaged 0.0012, 0.0012, and 0.0017 g NH_4-N/g TSS/d for the control, LO, and HI treatment respectively.

Sodium bicarbonate additions to tanks varied throughout the experiment. Mean total sodium bicarbonate addition to control treatment tanks was 630 g/m^3, significantly greater than the 246 g/m^3 ($P = 0.003$) or 405 g/m^3 ($P = 0.031$) added to LO or HI treatment tanks respectively. Additions did not differ significantly ($P = 0.096$) between the LO and HI treatments. Total alkalinity did not differ significantly among treatments during period 1 but was significantly greater in the control and LO treatments during period 2 (Table 2).

Settling chambers reduced TSS on average by 80% and 63% ($P = 0.050$) for the LO and HI treatments respectively. On a dry matter basis solids removed as sludge from settling chambers averaged 36 kg and 39 kg ($P = 0.221$) for the LO and HI treatments respectively. The total volume of solids removed as sludge from settling chambers averaged 5,793 L and 5,464 L ($P = 0.173$) for the LO and HI treatments respectively. The quantity of solids discharged as a percent of feed input did not differ significantly ($P = 0.260$) and averaged

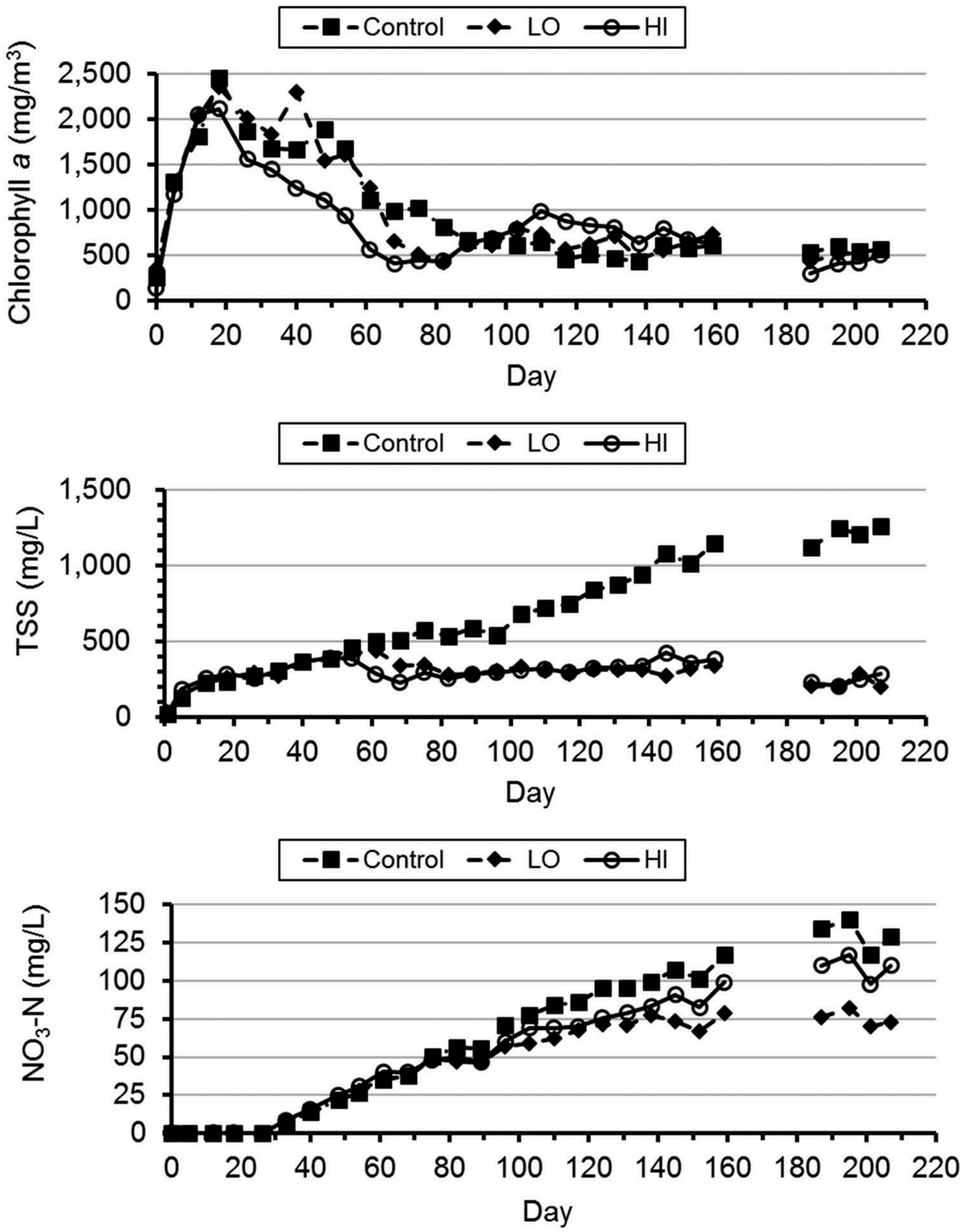

Figure 2. Mean weekly chlorophyll *a* (top panel), total suspended solids (middle panel), and nitrate-nitrogen (bottom panel) concentrations in a channel catfish biofloc technology production system subjected to two levels of settling chamber influent flow rate: LO = 0.9 L/min, HI = 2.9 L/min. Solids were not removed from control tanks. Settling chambers were activated on day 52. The gap in data corresponds to the 1–16 October 2013 shutdown of the USA government.

15.6% and 16.1% for the LO and HI treatments respectively. Mean SRT was 11.0 d for the LO treatment, not significantly different (P = 0.051) from the HI treatment (8.4 d).

Discussion

When intensifying channel catfish production, biological processes occurring in the outdoor photoautotrophic-chemoautotrophic (mixotrophic) BFT system function to maintain water quality, primarily low NH_4-N and NO_2-N concentrations, in response to high feeding rates. Channel catfish likely derive minimal nutrition from the biofloc and provision of increasing quantities of formulated feed are necessary to sustain rapid growth (Bonneau et al. 1972; Lilyestrom, Romaire, and Aharon 1987). Metabolism of feed by fish drives the increase in solids (living and dead particulate organic matter) concentrations as noted in the control treatment and by Green, Schrader, and Perschbacher (2014). Unmanaged, TSS concentrations in an earlier BFT study increased to levels that appeared to impact channel catfish negatively, with unexplained mortality recorded at TSS > 2,000 mg/L and abnormal swimming behavior observed when TSS exceeded about 1,600 mg/L (Green, Schrader, and Perschbacher 2014). The absence of significant differences among treatments means in the present experiment for daily and total feed consumption, growth curve slopes, and production variables indicated that TSS concentration, which reached a maximum of 1,200 to 1,410 mg/L in control treatment tanks, did not affect fish adversely. Stocker-size catfish, which were grown by Green, Schrader, and Perschbacher (2014), may be more susceptible to high TSS concentrations than the food-size fish produced in this study.

Production variables were similar to results of previous experiments in the channel catfish BFT system (Green and Schrader 2015; Green, Schrader, and Perschbacher 2014; Schrader, Green, and Perschbacher 2011). Differential effects of TSS concentration on production variables are reported in other published studies. In a 21-d study, Poli, Schveitzer, and Nuñer (2015) reported that survival and mean final weight of larval South American catfish (*Rhamdia quelen*) reared for 21 days in BFT systems maintained at 157.0 to 908.7 mg/L TSS did not differ significantly. Shrimp growth in BFT culture was better when TSS concentration was low (less than about 300 mg/L) (Gaona et al. 2017; Ray, Dillon, and Lotz 2011; Ray et al. 2010; Schveitzer et al. 2013; Vinatea et al. 2010), whereas survival was (Gaona et al. 2017; Schveitzer et al. 2013) or was not (Ray, Dillon, and Lotz 2011; Ray et al. 2010) affected by TSS concentration.

Despite the absence among treatments of significant differences in production variables, fish size distributions at harvest did vary significantly. Fish from the control treatment were skewed toward the smaller size classes, which can affect marketability (Wiese et al. 2006). Control treatment fish consumed 5% to 11% less feed than fish in the exchange treatments, which likely affected growth, and the cause of which is not known. Additional research on how TSS affects catfish gill morphology and hematology would

be useful. Compared to the control treatment, GFY in the LO and HI treatments was 9%–12% higher for size classes ≥ 0.45 kg/fish and 23%–48% higher for size classes ≥ 0.57 kg/fish. Thus, more fish in the control treatment must be carried over to the next growing season, which increases risk to the farmer. While these differences were not significant, they will affect farm revenue.

Geosmin concentrations detected in fillet samples from the control treatment did not exceed previously reported threshold sensory detection limits of trained catfish processing plant flavor testers for geosmin (250–500 ng/kg), but did for the LO and HI treatments (Grimm, Lloyd, and Zimba 2004). The absence of a significant difference among treatments for fillet geosmin concentration resulted from the high among tank variability for the LO and HI treatments. Fillet concentrations of MIB did not exceed the 100–200 ng/kg sensory detection limit (Grimm, Lloyd, and Zimba 2004) in any treatment. Thus, fish from the LO and HI treatments likely would be classified as having an objectionable "earthy" off-flavor. The present results contrast with previous observations that in the outdoor BFT system the frequency and intensity of geosmin- and MIB-induced off-flavor in channel catfish is substantially lower than for earthen pond catfish culture (Green and Schrader 2015; Green, Schrader, and Perschbacher 2014; Schrader and Dennis 2005; Schrader, Green, and Perschbacher 2011). Off-flavor-producing cyanobacteria and actinomycetes are reported to occur episodically in the catfish BFT tanks, but their presence has not been correlated with high aqueous geosmin or MIB concentrations (Green, Schrader, and Perschbacher 2014; Schrader, Green, and Perschbacher 2011). However, the large variability of mean geosmin concentrations in the catfish flesh among the tanks within the solids removal treatments in the present study likely resulted from the inconsistent presence and/or abundance of geosmin-producing microorganisms in the BFT tanks. Further research to identify the source(s) of geosmin and MIB in the outdoor BFT system is needed.

Solids concentrations were reduced to around 300 mg/L in both solids removal treatments. The significantly lower slope of NO_3-N accumulation in the LO compared to control treatment during period 2 suggests that nitrification decreased because of solids removal. Metabolic output of NH_4-N by fish in all treatments likely was similar during period 2 because of similar feed input. The significantly lower quantities of sodium bicarbonate required to maintain pH and alkalinity and the slower accumulation of NO_3-N in the solids removal treatments support the decreased nitrification hypothesis. Additionally, sludge retention times in exchange treatments were within the typical SRT of 3 to 18 d recommended for complete nitrification (Tchobanoglous, Burton, and Stensel 2003). Others also report decreased nitrification in shrimp BFT systems in response to solids removal. Nitrification rate inferred from inhibited dark bottle respiration was significantly lower in the 200 mg/L TSS treatment compared to

the 400–600 and 800–1,000 mg/L TSS treatments (Schveitzer et al. 2013). And reducing TSS concentration from 782 to 459 mg/L significantly reduced nitrification rate as indicated by significantly different slopes for NO_3-N concentration over time (Ray et al. 2010). Nitrification occurred when TSS concentration was maintained at about 300 mg/L but was not detected at about 200 mg/L TSS (Ray et al. 2012).

On the other hand, the observed decreased nitrification could be the result of dilution caused by addition of replacement water following draining of settling chambers. But since similar volumes of solids were drained from LO and HI treatment settling chambers, there should be no differential response because of dilution. Similarly, there is a danger with excessive solids removal of washing out the nitrifying bacteria from the system, thereby reducing nitrification and allowing heterotrophs to proliferate (Ebeling, Timmons, and Bisogni 2006; U.S. EPA 1993).

The fate of unaccounted for excreted nitrogen (N) and phosphorus (P) in the solids removal treatments compared to the control is unclear given the absence of significant treatment differences for total feed fed, FCR, and production variables. Unlike earthen ponds where the accumulation of organic N (Gross, Boyd, and Wood 2000) and PO_4-P adsorption (Masuda and Boyd 1994) occur in pond sediments, neither process occurs in outdoor, HDPE-lined BFT tanks because they do not contain soil and similar sediments. Similar chlorophyll *a* concentrations among treatments in the present experiment indicated that differential N and P uptake by phytoplankton likely would not elucidate the fate of unaccounted N and P. Ammonia-nitrogen can be lost by volatilization, and 3.8%, on average, of NH_4-N concentration can be volatilized at pH > 8.3 (Gross, Boyd, and Wood 1999). However, NH_4-N volatilization in the present experiment likely was minimal because of low NH_4-N concentration and pH in the mid-7 range. Denitrification in earthen ponds occurs in the sediments and can account for 17.4% of N losses in channel catfish ponds (Gross, Boyd, and Wood 2000; Hargreaves 1998). Denitrification also is reported to occur in anaerobic microenvironments in the water column of earthen ponds (Avnimelech and Zohar 1986). While anaerobic microenvironments likely did not occur in the tank water column during the present experiment because of the continuous aeration, denitrification, which was not measured, possibly occurred in the settling chamber. Evidence of denitrification was detected for settling chambers used to manage TSS levels in a shrimp brackish water BFT system (Ray, Dillon, and Lotz 2011). Both denitrification and dissimilatory nitrate reduction to ammonium were detected in laboratory reactors that contained biofloc particles from a tilapia BFT system (Chutivisut, Pungrasmi, and Powtongsook 2014). Although not quantified in the present experiment, N and P are lost from the system when sludge is drained from settling chambers. Nitrogen discharged as sludge from a shrimp BFT system

ranged from 28% to 46% of total nitrogen discharged from the system (Arantes et al. 2017). Clearly, development of N and P budgets for the outdoor, mixotrophic BFT system would elucidate the fate of added nutrients.

In summary, both settling chamber flow rates effectively maintained TSS concentrations around 300 mg/L. Channel catfish performance was not affected when exposed to TSS concentration as high as 1,410 mg/L and exhibited no apparent adverse effects. But fish size distribution was skewed toward larger sizes when solids were removed. Water quality results suggested that nitrification was reduced in response to solids removal, especially in the LO treatment. Fish fillets from the solids removal treatments contained enough geosmin to likely be classified as having an objectionable "earthy" off-flavor. Research is needed to identify the microbial source of geosmin and MIB in the outdoor catfish BFT system before the development of mitigation strategies can commence. Additionally, it appears important to maintain a TSS concentration that ensures rapid nitrification while any TSS reduction is unlikely to significantly affect the intensity of episodes of microbial-related off-flavor.

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Disclosure statement

No potential conflict of interest was reported by the authors.

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