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1 2 3 4 5 6 7 8 9	Correspondence Author: Arnie Eversole Dept. of Forestry and Natural Resources Clemson Univ. Clemson, SC 29634-0317 864-656-5328 fax 656-5332 aevrsl@clemson.edu
11 12 13 14 15 16	Effect of Temperature and Phytoplankton Concentration on Freshwater Mussel Filtration in the Partitioned Aquaculture System
18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41	Arnold G. Eversole and Kevin R. Stuart Department of Forestry and Natural Resources Clemson University Clemson, South Carolina 29634 USA and David E. Brune Department of Agriculture and Biological Engineering Clemson University Clemson, South Carolina 29634 USA

Abstract

The freshwater mussel Elliptio complanata was provided green algaldominated water from a Partitioned Aquaculture System (PAS) over a range of water temperatures (7-32 C) and suspended particulate organic carbon (POC) concentrations (1-32 mg POC L⁻¹) to determine filtration rates as mg POC kg⁻¹ wet tissue weight h⁻¹. Filtration rates increased with both increased water temperature and POC concentration. The predicted filtration rate (PFR) response to water temperature and POC concentrations is: PFR = 160.36 - 33.27T + 2.57T² – 0.06T³ + 0.02T*POC². Within the test conditions the predicted maximum filtration rate of 510 mg POC kg⁻¹ wet tissue h⁻¹ occurred at 26 C and 32 mg POC L⁻¹ and predicted minimum filtration rate of 25 mg POC kg⁻¹ wet tissue h⁻¹ at 10.5 C and 1 mg POC L⁻¹. A model to describe a mussel filtration rate response to PAS water conditions requires both water temperature and POC concentration data.

Key words: filtration rate, freshwater mussel, PAS, culture system

Introduction

The Partitioned Aquaculture System (PAS) separates a culture pond into four distinct components (paddlewheels, algal basin, filter feeder area and culture species area) linked by a homogeneous water velocity field (Brune et al. 2004). Paddlewheels provide water flow, nutrient mixing, and uniform sunlight exposure throughout the algal basin and the filter feeder and culture species areas.

67 Phytoplankton in the algal basin (95% of the PAS area) use inorganic fish 68 metabolic wastes (nutrients) to sustain high primary productivity levels. Feeding 69 rates and carrying capacity of the PAS is about 3-5 times that of conventional 70 channel catfish (Ictalurus punctatus) culture ponds as a result of sustained high 71 phytoplankton productivity. Filter-feeding organisms, by harvesting 72 phytoplankton, reduce cell age and stabilize a standing crop of faster growing phytoplankton cells at reduced respiration rates and increased oxygen production 73 74 per unit volume (Brune et al. 2004). Several filter-feeding species including Nile tilapia (Oreochromis niloticus), 75 76 silver carp (Hypophthalmichthys moltrix), and the native freshwater mussel 77 (Elliptio complanata) have been used in PAS production trials. Each species has 78 attributes and drawbacks in their use in a PAS production model. For example, 79 both Nile tilapia and silver carp effectively control cyanobacteria in PAS waters 80 (Turker et al. 2003b, Mueller et al. 2004); however, both are also non-native 81 species which may be either prohibited by law or require time-consuming use 82 permits and expensive precautions for use in aquaculture. Nile tilapia, a tropical 83 and a thermophilic species, filtration rates in a cool-water regime (17-23 C) were 84 significantly lower than in the 26-32 C warm-water regime (Turker et al. 2003c). 85 Tilapias cease feeding below 16 C and mortality occurs below 13 C (Chervenski 86 1982, Ross 1999). A catfish feed-rate protocol used in PAS production trials, 87 based on Q₁₀, provided for feeding to continue at water temperatures below 10 C 88 (Elvidge 1998). Although silver carp survives and may filter at temperatures

lower than tilapia, we were reluctant to use silver carp because of permitting and

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supply difficulties and the dangers associated with an escaped over-wintering species. Elliptio complanata also filtered phytoplankton from PAS waters at water temperatures lower than Nile tilapia (Stuart et al. 1999). Use of a native filter feeding organisms such as freshwater mussels would avoid many of the potential problems associated with the use of either tilapia or silver carp in the PAS.

Nile tilapia, silver carp and E, complanata filtration rates increase with

Nile tilapia, silver carp and <u>E. complanata</u> filtration rates increase with increasing particulate organic carbon (POC) levels in PAS water (Stuart et al. 2001, Turker et al. 2003a, b). Within a restricted water temperature range (26-30 C), Nile tilapia and silver carp filtration rates both reach asymptotic maxima at POC levels which are different for green-algal and cyanobacterial dominated PAS waters (Turker et al. 2003b). At a lower temperature range (23-27 C), <u>E. complanata</u> filtration rate also increases to a maximum in green-algal dominated water with increasing POC; however, a maximum filtration rate was not achieved by Stuart et al. 2001 in the experiment using cyanobacterial-dominated PAS water.

Since the classic papers of Walz (1978) and Winter (1978) we have known that bivalve filtration was a function of phytoplankton (seston) concentration and water temperature. Very few studies have investigated bivalve filtration rates simultaneously over a range of water temperatures and phytoplankton concentrations. One of these was Lui's (1985) study of the freshwater pearl mussel (Anodonta woodiana) where filtration rate increased with suspension concentration and peaked at an intermediate water temperature.

Unfortunately only three water temperatures and an inorganic suspension

concentration were tested. The purpose of this study was to determine <u>E.</u>

complanata filtration rates over a range of POC levels and water temperatures

that may be encountered during a typical catfish production schedule in the PAS.

Filtration rate, which is sometimes referred to as uptake rate, in this study is

defined as the POC mass removed from the water column per unit of mussel

mass per time.

Materials and Methods

Filter-feeding experiments were conducted using algal-rich water from PAS units at Calhoun Field Station, Clemson University, SC, USA. Mussels collected from Big Garvin Creek, SC, were held in PAS culture units between filtration rate experiments. Test mussels were stocked at the same biomass (1.22 kg wet tissue weight per tank) in eight 127-L continuous stirred tank reactors (CSTR) while a ninth CSTR without mussels was used as a control. The tissue biomass of <u>E. complanata</u> was estimated from the relationship: tissue wet weight (g) = 1.8806 + 0.2626 whole animal weight (g); r²=0.94; n=110 (Starkey 1999).

Each CSTR received PAS water for 24 h before the addition of mussels. Individual timers and solenoid valves provided an intermittent addition of water at 0.07, 0.1, 0.2, 0.6, 1.0, 1.5, 2.0, 2.5, and 3.0 L min⁻¹ to CSTRs for 72-h experimental periods. Water was discharged through a standpipe and an

airstone helped maintain a mixed water column. Mussels were held off the

bottom (approx. 8 cm) by screening to avoid suspension of feces and pseudofeces. After each experiment mussels were returned to the PAS, and the CSTRs and water delivery pipes were thoroughly cleaned.

Water temperatures were recorded at 2-4 h intervals from 0700 to 2300 h during the 72-h experimental periods. Dissolved oxygen, pH and nitrite-nitrogen were measured once every 24 h during the experiments. Dissolved oxygen and temperature values were measured with an YSI polargraphic oxygen meter (model 58, YSI Inc. Yellow Springs, OH). Nitrite-nitrogen was measured using a spectrophotometer (APHA 1989) and pH values with a Hach kit (model FF-1A, HACH Company, Loveland, CO).

A water sample taken from a representative CSTR before stocking mussels was centrifuged at 15,000 rpm for 15 min and decanted. The pellet representing the suspended matter was then resuspended in a known volume of water. Aliquots (n=5) of the sample were rediluted and the particulate organic carbon (POC) levels were determined with a Rosemount Dohrman total organic carbon analyzer (model DC-190, Rosemount Dohrman, Cincinnati, OH). Optical transmission values of each aliquot were determined at 750 nm with the spectrophotometer (APHA 1989). A standard curve of aliquot POC and transmittance for each experiment was determined by regression analysis. The coefficient of determination (r²) for these linear regression models (n=13) ranged from 0.93 to 0.97. Water samples for subsequent POC values were taken at timed intervals from the incoming and outgoing (mussel filtered) water. The POC values for these samples were determined from the initial sample transmittance

values using the specific standard curve for each filter-feeding experiment. The net change between the incoming and the outgoing water in the control (CSTR without mussels) represented incidental settlement and was used to correct filtration rates for each experiment. Filtration rates used in the analysis were restricted to those samples after the incoming water replaced twice the volume of CSTR. Mussel filtration rate (FR as mg POC kg wet tissue⁻¹ h⁻¹) was calculated as:

 $FR = (POC_i - POC_o) \ x \ flow \ rate/mussel \ biomass$ where POC_i is the suspended particulate organic carbon in incoming water (mg $POC\ L^{-1}$), POC_o is the suspended particulate carbon in outgoing water, flow rate is L min⁻¹ and mussel biomass is kg wet tissue.

Algal cells in water samples from PAS units were counted and if the number of green-algal cells exceeded 60% of total cell abundance in duplicate hemocytometer counts, the water was classified as green-algal dominated for the experiments. Filter experiments using water dominated by green algae were replicated over a range of ambient water temperatures from May to January. Controlled flow rates and mussel filtering activity provided a set of POC concentrations for the experimental temperature range.

Analysis of variance (ANOVA) using the general linear model was used to detect differences in water quality values between incoming and outgoing water from the control (CSTR without mussels). Multiple regressions were run to obtain a response of the mussels (i.e., filtration rate) to water temperature and

POC levels. The alpha level was set at 0.05 and SAS was used for statistical procedures.

RESULTS

Water quality parameters were similar among the 13 experimental trials. Mean (range) dissolved oxygen, pH and nitrite-nitrogen were 7.55 mg L⁻¹ (5.2-10.9), 7.8 (6.5-8.5) and 0.07 mg L⁻¹ (0.01-0.35), respectively. <u>Scenedesmus, Ankistrodesmus</u> and <u>Planktospheria</u> were the most abundant taxa in the greenalgal experimental water. Pennate diatoms were observed in each of the experimental waters whereas the cyannobacteria, <u>Microcystis</u> and <u>Merismopedia</u>, were observed in three of the experimental trials and in only limited numbers.

Mean (±SE) POC levels of the incoming and outgoing water in the control

Mean (\pm SE) POC levels of the incoming and outgoing water in the control CSTR (without mussels) were 18.53 ± 0.34 mg POC L⁻¹ and 17.01 ± 0.32 mg POC L⁻¹, respectively. The difference between incoming and outgoing water POC concentrations in the control was not significant, indicating phytoplankton suspensions were not affected by sedimentation or wall attachment.

Filtration estimates (n=1046), 42 outliers removed when mussels experienced unusual conditions (e.g., algal bloom crash), are plotted in Figure 1a. High POC levels (> 20 mg POC L⁻¹) were not experienced at low water temperatures (<15 C) in the PAS and because of water delivery system limitations at low flow rates (< 0.07 L min⁻¹), POC levels < 10 mg POC L⁻¹ were not observed at high water temperatures. The highest individual filtration rates

were observed at 26-28 C and 25-32 mg POC L⁻¹, and the lowest individual filtration rates were at 10 C and 7-10 mg POC L⁻¹.

Figure 1b represents the mussel's predicted filtration rate (PFR) in water temperatures from 7 to 32 C and POC levels from 1 to 32 mg POC L-1. The response surface relationship is:

PFR = $160.36 - 33.27T + 2.57T^2 - 0.06T^3 + 0.02T * POC^2$; $r^2 = 0.69$. Water temperature was the most important variable explaining the response surface. The rate of change in the mussel filtration rate in response to increased POC concentration was slower at low water temperatures (e.g., <15 C) than at water temperatures > 20 C. Predicted maximum filtration rates occurred at progressively higher water temperatures with increased POC concentrations. Within the range of experimental conditions experienced, the predicted maximum filtration rate of 510 mg POC kg⁻¹ wet tissue h⁻¹ was at 26 C and 32 mg POC L⁻¹, and the predicted minimum filtration rate was 25 mg POC kg⁻¹ wet tissue h⁻¹ at

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10.5 C and 1 mg POC L⁻¹. The predicted filtration rates at low water

temperature/high POC levels and high temperature/low POC need to be viewed

with some caution because of the limited data in these sections of the response

surface.

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Discussion

Several factors in the water column influence filtration rates in freshwater mussels including water temperature and phytoplankton (particle) size, composition, and concentration (Wagner 1976, Walz 1978, Lui 1985, Sprung and

229	Rose 1988, Helfrich et al. 1995, Vanderploegetal et al. 1995, Lei et al. 1996).
230	The test water phytoplankton was dominated by <u>Scenedesmus</u> and
231	Ankistrodesmus, both within the size range of phytoplankton effectively filtered by
232	E. complanata (Paterson 1986, Stuart et al. 2001). Over a limited temperature
233	range (23-27 C), E. complanata filtration rate increased to 511 mg POC kg ⁻¹ wet
234	tissue h ⁻¹ as POC levels increased from 10 to 28 mg POC L ⁻¹ (Stuart et al. 2001).
235	Similar increases in filtration rate, defined as the mass of phytoplankton (seston)
236	removed from the water column per time, have been observed in other
237	freshwater bivalves (Walz 1978, Lui 1985, Hornbach et al. 1984, Sprung and
238	Rose 1988, Lei et al. 1996). In these cases, the bivalves responded to increased
239	phytoplankton concentration by increasing pumping rate the volume of water
240	filtered (e.g., ml h ⁻¹) up to a concentration with further phytoplankton increases
241	resulting in a decrease in the volume of water filtered and the eventual
242	stabilization of filtration rate (e.g., Lei et al.1996). A reduction in the volume of
243	water filtered at high phytoplankton concentrations would represent an energy-
244	saving measure for the same food reward per unit of energy expended. Although
245	the regulatory mechanism was not determined, it is clear that E. complanata
246	filtration rate responded to the quantity of POC in the PAS water.
247	Temperature influences physiological activity and consequently, also the
248	filtration rate of freshwater mussels. Filtration and pumping rates of freshwater
249	mussels have been observed to increase with increasing temperature to a
250	maximum, and with further temperature increases a decrease in these rates
251	(Wagner 1976, Lui 1985, Lei et al. 1996). Elliptio complanata exhibited the same

pattern of increasing filtration rate to a peak followed with a decrease in filtration rate as temperature increased from 7 to 32 C. Filtration rates estimated over a range of ambient water temperatures should represent more accurately the temperature optimum for <u>E. complanata</u> filtration than data collected from mussels acclimated to different laboratory temperatures (e.g., Wagner 1976).

Water temperature and POC concentration variables interacted to influence filtration rate of <u>E. complanata</u>. The rate <u>E. complanata</u> increased filtration with POC concentration levels was faster at water temperatures near the optimum than under suboptimum temperatures, including low and high water temperatures. Conversely, as the POC levels increased, maximum filtration rates occurred at progressively higher water temperatures. A model to describe <u>E. complanata</u> filtration rate responses to the prevailing PAS water conditions would require both temperature and POC concentration data.

Rarely do high POC levels occur at low water temperatures or low POC levels occur at high temperatures in PAS waters during a production season (Brune et al. 2004). However unexpected low POC concentrations (20 mg POC L⁻¹) were observed at high water temperatures (25 C) in the PAS unit following a sharp decrease in the algal standing crop biomass during the week of June 12, 2000 (e.g., water temperature and POC that were 28 C and 32 mg POC L⁻¹ before the decrease were 25 C and 20 mg POC L⁻¹ after). The predicted filtration rate of *E. complanata* was 500 mg POC kg⁻¹ wet tissue h⁻¹ prior to the decrease in POC and more than doubles the 197 mg POC kg⁻¹ wet tissue h⁻¹ expected after the POC decrease.

275 Freshwater mussels offer an alternative to the use of tilapia as a filter 276 feeder in culture operations. Mussels operate over a wider range of temperatures 277 than tilapia, over winter in culture ponds and avoid the expense of a wintering 278 tilapia facility. Both green algae and cyanobacteria are filtered by mussels but not at the same rates reported for tilapia and silver carp (Stuart et al. 2001, 279 280 Turker et al. 2003a, b). Mussels however appear more effective with the smaller-281 sized phytoplankton taxa than either tilapia or silver carp and should provide a 282 nice compliment to the filtering capabilities of either of these fish taxa. 283 284 Acknowledgments 285 We tank Ron Gantt for conducting carbon analysis, Dr. Larry Grimes with 286 statistical help, Teresa Wilson and Heather Irwin with graphics, and Scott Davis 287 for help with algae identification. This study was funded by the SC Aquaculture 288 Research Initiative and USDA (NRI and Rural America programs). This is 289 technical contribution number xxxx of the SC Agricultural Experiment Station, 290 Clemson, SC, USA. 291 292 293 294 295 296 References 297 298 American Public Health Association, APHA, 1989. Standard methods for the

examination of water and wastewater. American Water Works Association

299

300	and Water Pollution Control Federation. Washington, D. C., USA. 1584
301	pp.
302	Brune, D. E., Schwartz, G., Collier, J. A., Schwedler, T. E., Eversole, A. G., 2004
303	Partitioned Aquaculture System. In: Tucker, C. S., Hargreaves, J. A.
304	(Eds.). Biology and Culture of the Channel Catfish. Elsevier Science,
305	Amsterdam, Netherlands. pp.561-584.
306	Chervinski, J., 1982. Environmental physiology of tilapias. In: Pullin R. S. V.,
307	Lowe-McConnell R. H. (Eds.). The Biology and Culture of Tilapias.
308	ICLARM Conference Proceedings, Manila, Philippines, Volume 7, pp. 119
309	128.
310	Elvidge, R. D., 1998. Production of channel catfish <u>Ictalurus punctatus</u> in the
311	Partitioned Aquaculture System with stocking rates and co-culture of Nile
312	tilapia Orechromis nilotica. Clemson University MS thesis, Clemson, South
313	Carolia, USA.
314	Helfrich, L.A., Weigmann, D.L., M. Zimmerman, M., 1995. Control of suspended
315	solids and phytoplankton with fishes and a mussel. Water Resources Bull
316	31, 307-315.
317	Hornbach, D.J., Burky, A.J., Way, C.M., Wissing, T.E., 1984. Effects of
318	particle concentration and season on the filtration rates of the freshwater
319	clam, <u>Sphaerium striatinum</u> Lamarck (Bivalvia: Pisidiidae). Hydrobiologia
320	108, 83-96.
321	Lei, J., Payne, B.S., Wang, S.Y., 1996. Filtration dynamics of the zebra
322	mussel, Dreissena polymorpha, Can. J. Fish, Aguat, Sci. 53, 29-37.

323	Lui, F.G., 1985. Studies on the feeding and filtration rate of freshwater pearl
324	mussel, Anodonta woodiana (in Chinese, with English abstract). Bull.
325	Malacol., Republic of China 11, 25-34.
326	Mueller, C.R., Eversole, A.G., Brune, D.E., 2004. Effects of silver carp
327	Hypophthalmichthys moltrix and freshwater mussel Elliptio complanata
328	filtration on the phytoplankton community of the Partitioned Aquaculture
329	System units. J. World Aquacult. Soc. 35, 372-382.
330	Paterson, C.G., 1986. Particle-size selectivity in the freshwater bivalve Elliptio
331	complanata (Lightfoot). Veliger 29, 235-237.
332	Ross, L.G., 1999. Environmental physiology and energetics. In: Beveridge,
333	M.C.M., McAndrew, B.J. (Eds.). Tilapias: Biology and Exploitation. Fish
334	Ser., Vol. 25. Kluwer Academic Publishers, Boston. pp.89-128.
335	Sprung, M. Rose, U., 1988. Influence of food size and food quantity on the
336	feeding of the mussel <u>Dreissina polymorpha</u> . Oecologia 77, 526-532.
337	Starkey, R.W., 1999. Oxygen mass balance of the Partitioned Aquaculture
338	System. M.Sc. thesis. Clemson University, Clemson, South Carolina,
339	USA.
340	Stuart, K.R., Eversole, A. G., Brune, D.E., 1999. Effect of flow rate and
341	temperature on the algal uptake by freshwater mussels. Proceed.
342	Freshwater Mollusk Conservation Soc. 1, 219-224.
343	Stuart, R. K., Eversole, A. G., Brune, D. E., 2001. Filtration of green algae and
344	cyanobacteria by freshwater mussels in the Partitioned Aquaculture
345	System. J. World Aquacult. Soc. 32, 105 -111.

346	Turker, H., Eversole, A.G., Brune, D.E., 2003a. Filtration of green algae and
347	cyanobacteria by Nile tilapia, Oreochromis niloticus, in the Partitioned
348	Aquacluture System. Aquaculture 215, 93-101.
349	Turker, H., Eversole, A.G., Brune, D.E., 2003b. Comparative Nile tilapia and
350	silver carp filtration rates of Partitioned Aquaculture System
351	phytoplankton. Aquaculture 220, 449-457.
352	Turker, H., Eversole, A.G., Brune, D.E., 2003c. Effect of temperature and
353	phytoplankton concentration on Nile tilapia Oreochromis niloticus (L.)
354	filtration rate. Aquacult. Res. 34:453-459.
355	Vanderploegetal, H.A., Leiberg, J.R., Nalepa, T.F. 1995. From picoplankton to
356	microplankton: temperature-driven filtration by the unionid bivalve
357	Lampsilis radiata siliquoidea in Lake St. Clair. Can. J. Fish Aquat. Sci 52,
358	63-74.
359	Wagner, R. E., 1976. The effects of size and temperature on the filtration rate of
360	the freshwater mussel. Bios 47: 168-178.
361	Walz, N. 1978. The energy balance of the freshwater mussel <u>Dreissena</u>
362	polymorpha Pallas in laboratory experiments and in Lake Constance.
363	Hydorbiologia 55, 83-105.
364	Winter, T.E. 1978. A review of the knowledge of suspension-feeding in
365	lamellibranchiate bivalves, with special reference to artificial aquaculture
366	systems. Aquaculture 13, 1-33.

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368	List of Figures
369	Figure 1. Filtration rate of Elliptio complanata in terms of mg POC kg ⁻¹ wet
370	tissue h ⁻¹ versus temperature (T, C) and suspended particulate organic carbon (
371	POC, mg POC L ⁻¹) from 7 to 32 C and 1 to 32 mg POC L ⁻¹ . The upper panel (a)
372	represents the individual observations (n=1046) and the lower panel (b) is the
373	response surface described by the predicted filtration rate (PFR) equation: PFR =
374	$160.36 - 33.27T + 2.57 T^2 - 0.07 T^3 + 0.02T*POC^2$.

