

## Filtration rates of the non-native Chinese mystery snail (*Bellamya chinensis*) and potential impacts on microbial communities

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### ABSTRACT

#### Filtration rates of the non-native Chinese mystery snail (*Bellamya chinensis*) and potential impacts on microbial communities

Invasive species in the phylum Mollusca, including gastropods and bivalves, have caused substantial impacts in freshwater ecosystems. The Chinese mystery snail, *Bellamya chinensis*, is a large viviparid snail native to Southeastern Asia and widely introduced throughout United States and parts of Canada and Europe. *B. chinensis* is a facultative filter-feeding detritivore that can both graze epiphytic diatoms using its radula and filter-feed its breathing water. Despite mounting concern associated with the expanding range and increasing abundance of *B. chinensis* in many parts of its invaded range, the potential ecological impacts of this non-native species remain largely unknown. Here, we used a series of laboratory experiments to assess filtration rates of *B. chinensis* and quantify its effects on microbial communities. According to both microcosm (24-hour, 4-L suspension) and mesocosm (5-day, 90-L suspension) experimental trials, *B. chinensis* exhibited an average filtration rate of 106-113 mL snail<sup>-1</sup>h<sup>-1</sup> (1.45 mL mg DW<sup>-1</sup>h<sup>-1</sup>) and an individual maximum of 471 mL snail<sup>-1</sup>h<sup>-1</sup> (6.15 mL mg DW<sup>-1</sup>h<sup>-1</sup>). These values are comparable to reported filtration rates for high-profile invasive, freshwater bivalves. Relationships between snail size and filtration rate relationship suggests that *B. chinensis* display an ontogenetic shift in feeding behavior from primarily radular grazing to increased filter-feeding at threshold size of approximately 44 mm shell height. Our experiments also revealed that high snail densities can result in small, significant shifts in bacterial community composition. These results suggest that *B. chinensis* may influence microbial communities either directly by using bacteria as a food source or indirectly by producing sufficiently large quantities of fecal and pseudo-fecal material to affect bacterial activity and growth. The overall ecological effects and importance of *B. chinensis* filtration behavior remain unclear, but our experimental results suggest that these impacts may be large and should be further investigated to better understand its potential role in coupling benthic and pelagic food webs in lake ecosystems.

**Key words:** Invasive species, chlorophyll-*a*, *Dreissena polymorpha*, *Corbicula fluminea*, *Potamopyrgus antipodarum*, *Pomacea canaliculata*.

### RESUMEN

#### Tasas de filtración de la especie introducida *Bellamya chinensis* y su potencial impacto en las comunidades microbianas

Las especies invasoras del phylum Mollusca, incluyendo los gasterópodos y bivalvos, han causado impactos importantes en los ecosistemas dulceacuícolas. *Bellamya chinensis*, es un vivíparo de gran tamaño, nativo del sureste de Asia y ampliamente introducido a lo largo de los Estados Unidos y parte de Canadá y Europa. *B. chinensis* es una especie detritívora-filtradora facultativa, que puede tanto ramonear diatomeas epifitas usando su rádula como filtrar el agua que respiran. A pesar de la creciente preocupación asociada al incremento en la abundancia y rango de distribución de *B. chinensis* en las regiones ya colonizadas, el potencial impacto ecológico de esta especie introducida permanece ampliamente desconocido. En este estudio, usamos series de experimentos de laboratorio para evaluar las tasas de filtración de *B. chinensis* y cuantificar su efecto en las comunidades microbianas. De acuerdo con los experimentos realizados tanto en los microcosmos (24-hour, 4-L suspensión) como en los mesocosmos (5-day, 90-L suspensión), *B. chinensis* mostró una tasa promedio de filtración de 106-113 mL caracol<sup>-1</sup>h<sup>-1</sup> (1.45 mL mg peso seco<sup>-1</sup>h<sup>-1</sup>) y un máximo por individuo de 471 mL caracol<sup>-1</sup>h<sup>-1</sup> (6.15 mL mg peso seco<sup>-1</sup>h<sup>-1</sup>). Estos valores son comparables a otros reportados para especies de bivalvos dulceacuícolas altamente invasivas. La relación entre el tamaño de los caracoles y las tasas de filtración sugieren que *B. chinensis* muestra un cambio ontogénico

en la manera de alimentarse, de ramoneo a una mayor alimentación por filtración, a partir de un umbral de tamaño de la concha de aproximadamente 44 mm de altura. Nuestros experimentos también revelan que altas densidades de caracoles generan pequeños cambios pero significativos en las comunidades microbianas. Estos resultados sugieren que *B. chinensis* afectaría las comunidades microbianas de forma directa usando las bacterias como fuente de alimentación o indirectamente al producir una cantidad de materia fecal o pseudo-fecal, suficiente para afectar la actividad y crecimiento bacteriano. El impacto ecológico global y el comportamiento como filtrador de *B. chinensis* aún no son claros, pero nuestros resultados experimentales sugieren que estos impactos pueden ser importantes y se deben investigar mejor para entender más su papel potencial en el acoplamiento de las redes tróficas bentónicas y pelágicas en los sistemas lacustres.

**Palabras clave:** Especies invasoras, clorofila-a, *Dreissena polymorpha*, *Corbicula fluminea*, *Potamopyrgus antipodarum*, *Pomacea canaliculata*.

## INTRODUCTION

Invasive species in the phylum Mollusca, including gastropods and bivalves, have caused substantial impacts in freshwater ecosystems, including alteration of water biogeochemistry, extirpation of native species, and modification of entire food webs (Strayer, 1999; Sousa *et al.* 2009). Notable examples range from the widespread invasion of zebra mussel (*Dreissena polymorpha*) and quagga mussel (*Dreissena bugensis*) throughout North America (Strayer, 2009; Higgins & Vander Zanden, 2010) to the numerous introductions of Asian clam (*Corbicula fluminea*: Hakenkamp *et al.*, 2001; Vaughn & Spooner, 2006), golden mussel (*Limnoperna fortunei*: Ricciardi, 1998; Boltovskoy *et al.*, 2009), New Zealand mudsnail (*Potamopyrgus antipodarum*: Hall *et al.*, 2003; Kerans *et al.*, 2005) and golden apple snail (*Pomacea canaliculata*: Carlsson *et al.*, 2004; Rawlings *et al.* 2007) in recent decades. In one such example, the invasion of golden apple snails in Southeast Asia drove wetlands from a macrophyte-dominated, clear water state to a turbid, phytoplankton-dominated state with concomitant shifts in the biological communities (Carlsson, *et al.* 2004). Although the ecosystem consequences of the aforementioned invaders have garnered significant scientific attention, we know very little of the ecology and potential ecological impacts of numerous other non-native mollusks (Strayer, 2012).

The Chinese mystery snail, *Bellamya* [= *Ci-pangopaludina*] *chinensis* (Gray, 1834), is a viviparid snail native to southeastern Asia and first documented in the United States over a century ago in Chinese markets of San Francisco (Wood, 1892). *Bellamya chinensis* (hereafter “*Bellamya*”) is the second largest snail in North America (approaching 70 mm maximum shell height), and its thick outer shell and a hard operculum flap covering the aperture (shell opening) provide a high degree of protection from predators and unfavorable environmental conditions. *Bellamya* was likely introduced to the United States multiple times through the aquarium trade and water gardening industry or for culinary purposes (Mackie, 1999), and it is now widely distributed in lakes and slow-moving rivers across North America. Recent evidence also suggests that *Bellamya* is highly resistant to desiccation leading to the potential for overland transport via boats (Havel, 2011). *Bellamya*’s invaded range includes at least 37 states in the United States, several provinces of southern Canada (Jokinen, 1982; Bury *et al.*, 2007), and parts of Europe (Soes *et al.*, 2011). During the 1960s, *Bellamya* were reportedly removed from the Laurentian Great Lakes by the metric ton (Mills *et al.*, 1993).

Despite mounting concern associated with the expanding range and increasing abundance of *Bellamya* in many parts of its invaded range (Bury *et al.*, 2007; Karatayev *et al.*, 2009; Solomon *et al.*, 2010; Chaine *et al.*, 2012), the

potential ecological impacts of this non-native species remain speculative. Johnson *et al.* (2009) found that the presence of *Bellamyia* caused substantial declines in the growth and abundance of two native snails in an experimental setting, reduced algal biomass, and increased the N:P ratio in the water column. Similarly, Clark (2009) found that *Bellamyia* caused decreased growth rates in a native *Physa* snail. Results from a field survey by Solomon *et al.* (2010) revealed no difference in snail assemblage structure associated with *Bellamyia* presence or abundance at the scale of an entire lake, although some native snail species tended not to occur at sites where *Bellamyia* was abundant. With the exception of these studies, little else is known about the potential ecological effects of this species, including its degree of reliance on benthic vs. pelagic resources and/or its effects on microbial communities. However, these impacts may be substantial because *Bellamyia* is unique in that it is a facultative filter-feeding detritivore that switches between grazing benthic and epiphytic diatoms using its radula and filter-feeding the water column (Plinski *et al.*, 1978; Dillon, 2000). Indeed, the popularity of using *Bellamyia* to clarify water in the pet aquarium and water garden trades attests to their efficiency as filter-feeders.

Bacteria are critical in freshwater ecosystems because they cycle essential nutrients and remove harmful toxins or chemicals from the water (Newton *et al.*, 2011). To date, little is known about impacts of species invasions on microbial communities (Ehrenfeld, 2010) and how these changes may alter nutrient cycles in invaded ecosystems (van der Putten *et al.*, 2007). *Bellamyia* may interact either directly with microbial populations by utilizing bacteria as a food source or indirectly by influencing the composition, activity, and growth of microbial communities via production of large quantities of fecal and pseudo-fecal material. Previous studies of invasive zebra mussel have shown direct effects on the abundance of major groups of microbes (e.g., Frischer *et al.*, 2000; Lavrentyev *et al.*, 2000; Lohner *et al.*, 2007) and a significant increase in the abundance of sediment-associated bacteria (Higgins *et al.*, 2010).

In the present study, we provide the first quantification of filtration rates by *Bellamyia* and investigate its potential effects on the diversity and composition of freshwater microbial communities. Given the vast diversity of microbial cells in the environment, the characterization of microbial community structure has been a challenge. Here, we used a ribosomal DNA fingerprinting technique (specifically, automated rRNA intergenic spacer analysis) to examine shifts in the whole bacterial community (including both rare and abundant taxa) in response to increasing densities of *Bellamyia* in an experimental setting.

## METHODS

### Experimental design

We evaluated the filter feeding capacity of *Bellamyia* using an indoor microcosm experiment during February 2009 in the School of Aquatic and Fishery Science's hatchery at the University of Washington. Glass microcosms (4-L volume) were filled with water directly from Lake Washington and experienced ambient photo-period (L:D = 12:12 hours) and water temperatures (16.8 °C ± 0.3 °C). Natural seston was kept suspended using an aeration (bubbling) device. To each of 72 microcosms we added either none (control,  $n = 18$ ) or one snail (collected from the wild) from each of three size ranges: 30-40 mm ( $n = 18$ ), 40-50 mm ( $n = 18$ ), 50-60 mm ( $n = 18$ ); measured according to shell height. At time zero (initial) and 24 hours (final) of the experiment we siphoned water samples from two inches below the surface of the water in the center of the microcosm for chlorophyll-*a* analysis. Ash-free dry weight (DW) of each snail was determined.

We assessed both filtration rates and the effects of *Bellamyia* on microbial community composition using an indoor mesocosm experiment during August 2009. Mesocosms (90-L volume, 0.18 m<sup>2</sup> area) were covered with 5 kg of coarse gravel (0.5-2.0 cm in diameter, 3 cm depth), filled with water directly from Lake Washington, and experienced ambient photo-period (L:D = 14:10 hours) and water temperatures (21.5 °C ± 0.5 °C).

Natural seston was kept suspended using an aeration (bubbling) device. Four pairs of ceramic tiles were placed on the bottom surface of each mesocosm in each cardinal direction (total of 8 tiles per mesocosm). After a five-day pre-experiment period, we added 0 (control), 2 (low) or 4 (high) snails to each of 6 mesocosms (replicates), representing a total of 18 mesocosms. These snail densities (11 and 22 ind m<sup>-2</sup>, respectively) are comparable to those reported in surveys of Wisconsin, Washington State and European lakes, which ranged from 1 to 38 ind m<sup>-2</sup> (Olden *et al.*, 2009; Solomon *et al.*, 2010; Sousa *et al.*, 2009). Snails measured 45-50 mm in shell height and were collected from the wild one-day prior to the start of the experiment. On day zero (initial) and day five (final) of the experiment we (1) collected water samples from two inches below the surface of the water in the center of the mesocosm for chlorophyll-*a* and microbial assemblage analysis, and (2) collected periphyton by scraping the top of one tile in each of the cardinal directions (total of four tiles per time period).

### Chlorophyll-*a* concentration and filtration rate

For both the microcosm and mesocosm experiments, chlorophyll *a* (mg m<sup>-3</sup>) was measured by filtering a 500 mL aliquot of water through a 47 mm Whatman GF/C filter paper and using a Turner Designs spectrophotometer following standardized protocols (<http://lter.limnology.wisc.edu/research/protocols>). A large number of studies have shown that filtration rates can be reliably measured using the clearance method and that consumption models based on natural seston produce more accurate predictions compared to data based on algal monocultures (reviewed in Riisgård, 2001). Filtration rates were estimated following Coughlan (1969) and many other studies by measuring the decline in chlorophyll-*a* over the experimental period:

$$FR = \frac{M}{n} \times \frac{(\ln C_{\text{initial}} - \ln C_{\text{final}}) - (\ln \bar{C}'_{\text{initial}} - \ln \bar{C}'_{\text{final}})}{t}$$

where *FR* is the filtration rate (volume cleared per snail per hour: mL snail<sup>-1</sup>h<sup>-1</sup>), *M* is the

volume of the water suspension (mL), *n* is the number of snails, *C*<sub>initial</sub> and *C*<sub>final</sub> are the initial and final concentrations in the snail treatment,  $\bar{C}'_{\text{initial}}$  and  $\bar{C}'_{\text{final}}$  are the mean initial and final concentrations in control treatments, and *t* is elapsed time (hours). In this formulation, the mean change in control concentrations (without snails) was subtracted from the change in experimental concentrations (with snails). Reductions of chlorophyll-*a* in controls would likely be attributable to zooplankton grazing and/or pigment degradation. Both experiments involved relatively long incubation times (24 and 120 hours), therefore small amounts of pseudofecal material were generated by the snails and could have been re-suspended in the water column. Thus, some particulate material may have been re-filtered, and therefore the measured filtration rates presented should be considered conservative estimates compared to a situation where pseudofeces have been removed during the course of the experiment.

### Bacterial community composition

Bacterial community composition was assessed using automated ribosomal intergenic spacer analysis (ARISA: Fisher & Triplett, 1999). ARISA generates fingerprints of the microbial community based on the length heterogeneity in the intergenic spacer region between the 16S and 23S rRNA genes, which varies among organisms. Although ARISA has similar limitations as other PCR-based fingerprinting approaches (Fisher & Triplett, 1999), it has been shown to give a robust, high-resolution view of bacterial assemblages in aquatic ecosystems (Brown *et al.*, 2005; Yannarell & Triplett, 2005) and can represent species-level taxonomic resolution (98-99 % sequence similarity; Brown *et al.*, 2005). A sample of 500 mL of water was filtered in duplicate onto a 0.22 μm filter (Supor-200, Pall Gelman, East Hills, NY), followed with 3 mL preservation buffer (10 mM Tris pH 8.0, 100 mM EDTA, 0.5 M NaCl) and frozen at -80° until analysis. DNA was extracted from replicate filters using the Qiagen DNeasy Blood and Tissue Mini-kit (Qiagen, Valencia, CA). The 16S-23S

intergenic region was amplified using the polymerase chain reaction (PCR) from the total extracted DNA using 6-FAM-labelled universal 1406-F primer (5'-TGYACACACCGCCCGT-3') and bacterial specific primer 23S-R (5'-GGGTT BCCCCATTCTRG-3') (Fisher & Triplett, 1999; Yannarell *et al.*, 2003). For each sample, four independent PCRs were performed, pooled, ethanol precipitated to remove unincorporated primers and run on a MegaBace 96 capillary sequencer along with ROX labeled size standards (50-1500 bp ladder, BioVentures, Inc). This sequencer is routinely used for fragment analysis and can resolve differences of 2 bp in fragments in the 300-400 bp range and differences of 10 bp for larger fragments (1000-1500 bp).

Fragment lengths were sized using DAX software (<http://www.dax.nl/dax.htm>) and a signal to noise cutoff was used to verify presence of peaks. Operational taxonomic units (OTUs) were generated by binning ARISA fragments into successively larger length bins based on their size and eliminating fragments that were <150 and >1300 bp (Brown *et al.*, 2005). Although OTUs are arbitrarily numbered groups of taxa and are not traceable to phylogenetic or functional groups, the ARISA approach provides a rapid and affordable way to assess changes in taxonomic composition of bacterial communities. We used both peak area and maximum peak area to estimate relative abundance of OTUs in our samples (Yannarell & Triplett, 2005), as well as examined OTU presence-absence.

### Data analysis

Results from the microcosm experiments were used to assess the relationship between snail size (shell height, mm) and filtration rate (mL snail<sup>-1</sup>h<sup>-1</sup>). Initial data exploration revealed that a linear model was not appropriate for the entire range of values and that a non-linear function was also insufficient. This prompted the comparison of three models: linear regression, quadratic regression and piecewise regression. Piecewise or segmented regression recognizes that different linear relationships may occur over different ranges of the independent variable (in this case,

snail size). Breakpoints are values on the x-axis where a change in the slope of the different linear relationships can be defined; these breaks may or may not be known before the analysis (Toms & Lesperance, 2003). Although this method is flexible enough to include segments (e.g., linear, polynomial) and breakpoints (e.g., sharp, smooth) of different forms, we estimated one sharp breakpoint and two linear, continuous functions.

Results from the mesocosm experiment were analyzed using univariate and multivariate statistics to assess the effects of *Bellamya* on algal biomass (water column and substrate) and to quantify impacts on microbial (OTU) community composition. Student's t-tests tested for differences in chlorophyll-*a* between initial and final conditions for the low ( $n = 2$ ) and high ( $n = 4$ ) snail density treatments (after controlling for changes in the control treatment).

Multivariate analyses of microbial community composition included Mantel's tests, analysis of multivariate similarity (ANOSIM) and non-metric multidimensional scaling (NMDS). We used Sørensen's similarity index (OTU presence-absence) and Bray-Curtis similarity index (OTU abundance according to area and peak height assignments) to summarize compositional similarity between treatment-replicate samples (3 treatment levels  $\times$  6 replicates = 18 observations). Sørensen index was used because it represents the complement to the Bray-Curtis index when using presence-absence data (Legendre & Legendre, 1998), thus allowing for a direct comparison of the multivariate results based on OTU presence-absence and abundance (peak height, maximum area). Similarity matrices were calculated separately for each for treatment (control, low, high) to allow for a comparison between the initial and final conditions.

Deep sequencing of microbial assemblages using 454 sequencing allows for the detection of extremely low abundance taxa. These extremely low abundance or unique taxa may influence the results from multivariate analyses and make data interpretation less clear (Rudi *et al.*, 2007; Gobet *et al.*, 2010). To assess how the occurrence of rare taxa might affect the results, Mantel tests were performed to assess the relationships between

dissimilarity matrices of the entire OTU dataset and two reduced datasets where OTUs that occurred in  $< 2\%$  or  $< 5\%$  of the samples were removed. Gastropods can often hosts their own unique microbial assemblages, therefore OTUs that were only present in the *Bellamya* treatments (i.e. were unique to snails) were removed from all datasets. We found statistically significant pairwise correlations between all datasets (Mantel  $R = 0.82-0.99$ , all  $P < 0.001$ ), indicating that the choice of OTU occurrence vs. abundance and the effect of rare OTUs did not heavily influence the results from the multivariate analysis. Consequently, we used the full dataset (not excluding “rare” OTU) containing OTU presence-absence.

We assessed the effects of *Bellamya* on bacterial community composition using the complementarily approaches of multivariate ordination (NMDS) and analyses of variance (ANOSIM). NMDS is an ordination method that preserves the rank ordered distances between observations in ordination space; here it was used to visualize differences in microbial composition between the initial and final time periods in each of the snail treatments (control, low, high). NMDS is an iterative approach that rearranges observations in ordination space to minimize a measure of disagreement (referred to as stress) between the compositional dissimilarities and the distance between points in the ordination diagram (Kruskal, 1964). We used a distance matrix based on Sørensen’s similarity index to ordinate the observations (replicates) in 2 dimensions with 100 random starts and tested the significance of the stress value with a Monte Carlo randomization test.

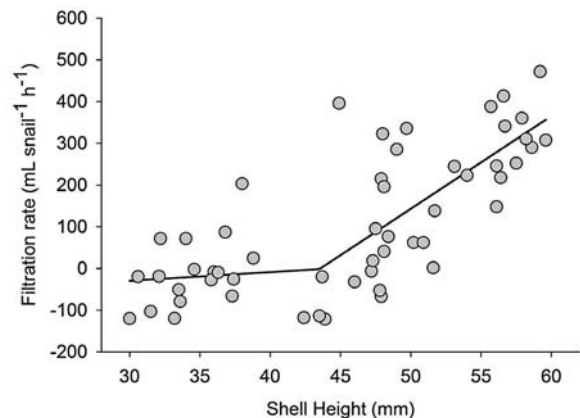
We used ANOSIM (Clarke, 1993), a nonparametric multivariate procedure analogous to analysis of variance, to test for differences in microbial community composition between the initial and final time periods in each of the snail treatments (control, low, high). ANOSIM tests predefined groups against random groups in ordination space by comparing the average of all rank similarities among observations within groups to the average of rank similarities among observations between groups (expressed as a  $R_{ANOSIM}$  statistic). We conducted 999 random permutations to assess the statistical significance of  $R$ .

All analyses were conducted in R version 2.15 (R Development Core Team 2012).

## RESULTS

### Filtration rates

The mean filtration rate in the 24-hour microcosm experiment (where *Bellamya* were examined individually) was  $106 \text{ mL snail}^{-1}\text{h}^{-1}$  (SD = 145) or  $1.45 \text{ mL mg DW}^{-1}\text{h}^{-1}$  (SD = 2.30); snails ranged from 30 to 60 mm in size. The individual maximum filtration rate was  $471 \text{ mL snail}^{-1}\text{h}^{-1}$  ( $6.15 \text{ mL mg DW}^{-1}\text{h}^{-1}$ ). The mean filtration rate, based on the 5-day mesocosm experiment where *Bellamya* were examined in groups of 2 or 4 individuals, was  $113 \text{ mL snail}^{-1}\text{h}^{-1}$  (SD = 158). Estimated filtration rates were over two times greater in the high- versus low-density treatment ( $159$  vs.  $66 \text{ mL snail}^{-1}\text{h}^{-1}$ ), suggesting that individual snails were filtering at a faster rate when in the presence of more conspecifics. Notably, we found very similar mean filtration rates for snails in the 45-50 mm size range based on the 24-hour



**Figure 1.** *Bellamya* filtration rates ( $\text{mL mussel}^{-1}\text{h}^{-1}$ ) as a function of snail shell height (mm) based on the microcosm experiment. Snails were held in isolation for 24 hours in a 4-L of water containing natural seston. Negative values reflect greater declines in chlorophyll-*a* concentrations in the control compared to the treatment microcosms. *Tasas de filtración de Bellamya* ( $\text{mL mejillón}^{-1}\text{h}^{-1}$ ) como función de la altura de la concha (mm) basadas en los experimentos en microcosmos. Los caracoles fueron aislados durante 24 horas en 4 litros de agua con seston natural. Valores negativos reflejan mayor reducción en la concentración de la clorofila-*a*, en el control que en los microcosmos tratados.

microcosm experiment ( $110 \pm 145 \text{ mL snail}^{-1}\text{h}^{-1}$ ) and 5-day mesocosm experiments ( $113 \pm 158 \text{ mL snail}^{-1}\text{h}^{-1}$ ) despite differences in initial chlorophyll-*a* concentrations and water temperatures.

*Bellamya* filtration rates increased with snail size (Fig. 1). Although statistical support for a linear (adj.  $R^2 = 0.52$ ,  $F_{1,52} = 56.7$ ,  $p < 0.001$ ) and quadratic relationship (adj.  $R^2 = 0.55$ ,  $F_{2,51} = 34.2$ ,  $p < 0.001$ ) was evident, the strongest model was represented by the piecewise regression (adj.  $R^2 = 0.56$ ,  $F_{3,50} = 34.2$ ,  $p < 0.001$ ). Based on the piecewise model there was strong support for an estimated breakpoint of  $43.5 \pm 4.1 \text{ mm}$  (mean  $\pm 1 \text{ SE}$ ) ( $t = 10.51$ ,  $p < 0.001$ ); snails less than the threshold size of 43.5 mm showed no evidence of a relationship with filtration rate (slope = 1.6,  $t = 0.20$ ,  $p = 0.984$ ), whereas snails exceeding the threshold size demonstrated a strong positive relationship with filtration rate (slope = 356.4,  $t = 9.60$ ,  $p < 0.001$ ).

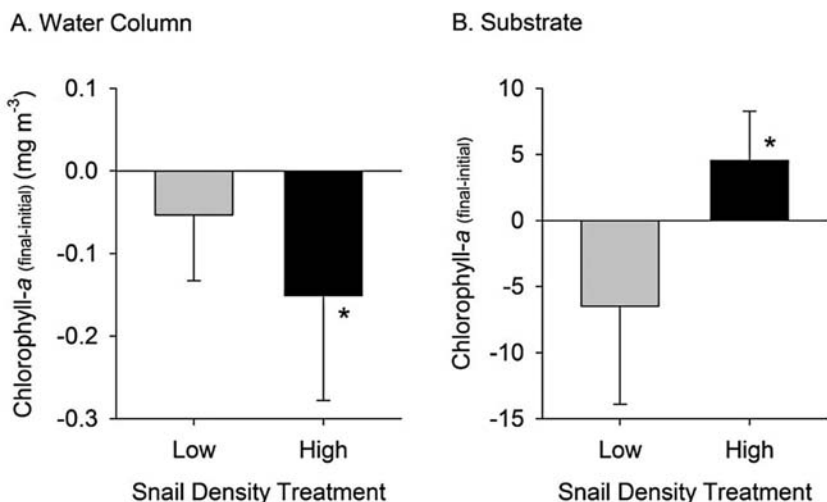
### Water column and benthic algal biomass (chlorophyll-*a*)

Water column chlorophyll-*a* decreased in the presence of *Bellamya* during the mesocosm

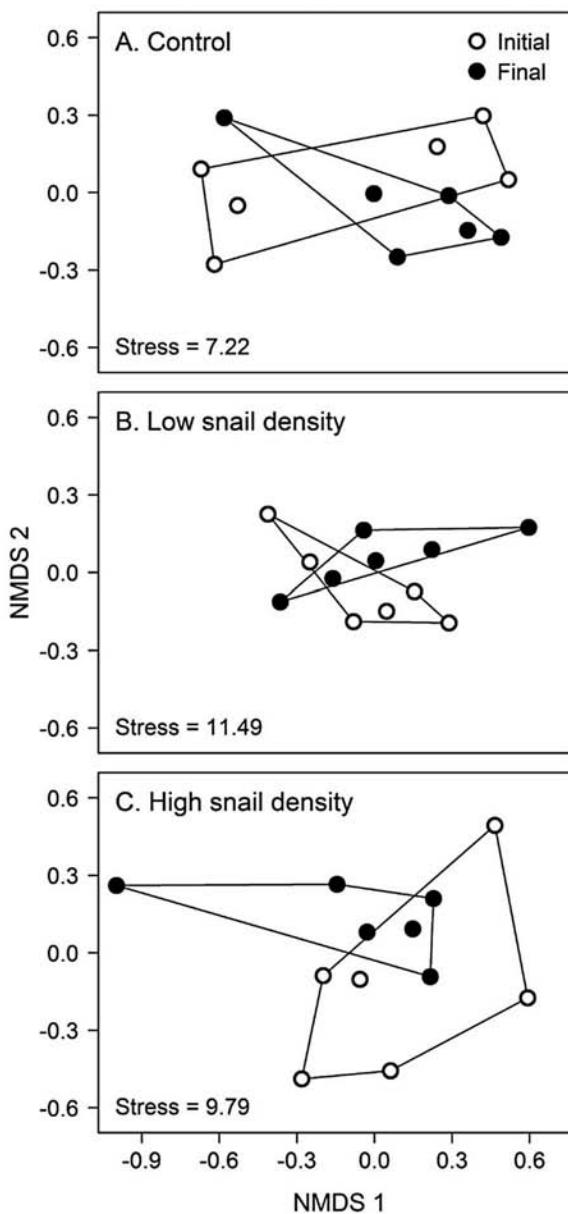
experiments, with a 54 % decrease in the low-density treatment ( $t = -1.64$ ,  $p = 0.162$ ) and a 155 % decrease in the high-density treatment ( $t = -2.92$ ,  $p = 0.033$ ) (Fig. 2A). Reductions in chlorophyll-*a* were three times greater in the high- compared to the low-density treatments, despite having only twice the number of snails (11 vs. 22 snails  $\text{m}^{-2}$ ). By contrast, benthic chlorophyll-*a* showed variable responses to the *Bellamya* treatments (Fig. 2B). The low-density treatment resulted in a 64 % reduction in chlorophyll-*a* ( $t = -2.14$ ,  $p = 0.085$ ), whereas the high-density treatment resulted in a 40 % increase in chlorophyll-*a* ( $t = -3.01$ ,  $p = 0.030$ ).

### Bacterial community composition

Bacterial community composition changed significantly over the course of the mesocosm experiment in the high-density treatment ( $R_{\text{ANOSIM}} = 0.132$ ,  $p = 0.049$ ) but showed little change in both the low-density treatment ( $R_{\text{ANOSIM}} = 0.147$ ,  $p = 0.111$ ) as well as the control ( $R_{\text{ANOSIM}} = 0.075$ ,  $p = 0.176$ ). NMDS ordination plots showed that differences in community composition were generally modest, but distinction between initial and final time peri-



**Figure 2.** Change in water column (A) and benthic (B) chlorophyll *a* ( $\text{mg m}^{-3}$ ) over the course of the mesocosm experiment for low (grey bar) and high (black bar) snail density treatments. \* indicates that mean is statistically different from zero based on a Student's *t*-test and  $p < 0.05$ . Cambios en la concentración de clorofila *a* ( $\text{mg m}^{-3}$ ) en la columna de agua (A) y en el bentos (B) durante el experimento en el mesocosmos, para tratamientos con bajas (barra gris) y altas (barra negra) densidades de caracoles. \* significa que la media es estadísticamente diferente de cero basándose en el *t*-test Student y  $p < 0.05$ .



**Figure 3.** NMDS ordinations summarizing patterns in microbial community composition in (A) control, (B) low and (C) high *Bellamyia* density treatments plots. *Patrones de ordenación NMDS, de la composición de la comunidad microbiana en los tratamientos con las diferentes densidades de Bellamyia; control (A), baja (B) y alta (C).*

ods was greater in the presence of *Bellamyia* (Fig. 3B, C) compared to the control (Fig. 3A). In addition, high-density snail treatments had reduced differences in community composition among replicates (Fig. 3C), as demonstrated by a higher average rank similarities in the final time

period (86 %) compared to the initial time period (61 %). Snail density did not have a significant impact on total bacterial abundance over the course of the experiment (results not shown).

## DISCUSSION

Chinese mystery snail, *Bellamyia chinensis*, is a versatile consumer that switches between grazing microalgae using its radula and filter-feeding its breathing water (Plinski *et al.*, 1977; Dillon, 2000). Our results showed that *Bellamyia* demonstrated high rates of water filtration, caused significant declines in pelagic algal biomass under experimental settings and impacted bacterial community composition.

According to both microcosm (24-hour) and mesocosm (5-day) experimental trials, *Bellamyia* exhibited an average filtration rate of 106–113 mL snail<sup>-1</sup>h<sup>-1</sup> (1.45 mL mg DW<sup>-1</sup>h<sup>-1</sup>) and an individual maximum of 471 mL snail<sup>-1</sup>h<sup>-1</sup> (6.15 mL mg DW<sup>-1</sup>h<sup>-1</sup>). A number of difficulties arise when attempting to compare the filtration rates of *Bellamyia* to those estimated for non-native bivalves in past studies. Important factors such as concentration of suspension, the nature of the suspension used, and the degree to which “refiltration” occurs within chambers differ from one experiment to the next. Despite these challenges, a qualitative comparison suggests that that *Bellamyia* filtration rates on natural seston are reasonably comparable to reported values for other invasive, freshwater and marine bivalves, including zebra mussel (*Dreissena polymorpha*), quagga mussel (*Dreissena bugensis*), Asian clam (*Corbicula fluminea*), golden mussel (*Limnoperna fortunei*) blue mussel (*Mytilus edulis*) (Table 1). Consequently, in certain environments, we expect that *Bellamyia* filtering-feeding behavior may shift primary production from pelagic to benthic zones.

Across the wide size spectrum of *Bellamyia* examined in our study we found that filtration rates were positively, and non-linearly, related to snail shell height. *Bellamyia* filtration rates were explained by a quadratic relationship with snail size, although we found slightly stronger



statistical support for a marked increase at a threshold size of 43.5 mm; below that size estimated filtration rates were near zero (and had no relationship with size), but above that size filtration rates increased with size. This provides the first evidence that *Bellamya* may display an ontogenetic shift in feeding behavior from primarily radular grazing to filter-feeding with increasing size (age). Our results also suggest that *Bellamya* may increase their filter-feeding behavior in the presence of higher densities of conspecifics. Two lines of evidence support this statement: (1) estimated filtration rates were over two times greater in the high- versus low-density treatment (159 vs. 66 mL snail<sup>-1</sup>h<sup>-1</sup>) and (2) greater proportional decrease in water column chlorophyll-*a* in high vs. low density treatment were observed. This shift from grazing benthic

microalgae to filter-feeding at higher densities may explain the opposing increase (high-density treatment) vs. decrease (low-density treatment) in benthic chlorophyll-*a*. This is likely caused, in part, by greater total snail excretion leading to elevated N:P in the water column (Johnson *et al.*, 2009) that promotes periphyton production.

Knowledge of how invasive species may affect bacterial diversity, abundance, and associated processes are important to understand potential impacts on ecosystem function. Previous studies have focused almost exclusively on invasive bivalves, specifically zebra and quagga mussel and have revealed shifts in benthic bacterial communities associated with invasions (Findlay *et al.*, 1998; Frischer *et al.*, 2000; Lavrentyev *et al.*, 2000; Viergutz *et al.*, 2007). For example, Lohner *et al.* (2007) observed an

**Table 1.** Comparison of feeding rates of *Bellamya chinensis* found in this study with those of other freshwater and marine invasive bivalves (adult estimates). *Comparación de las tasas de filtración de Bellamya chinensis encontradas en este estudio con las de otras especies invasoras de bivalvos dulceacuícolas y marinas (estimaciones en adultos).*

Species	Filtration Rate (mL ind <sup>-1</sup> h <sup>-1</sup> )	Filtration Rate (mL mg DW <sup>-1</sup> h <sup>-1</sup> )	Reference
<i>Bellamya chinensis</i>	106-113	1.5	this study
<i>Dreissena polymorpha</i>		16.4	Kondratev (1963)
		3.2	Micheev (1966)
		16.5	Kryger and Riisgård (1988)
	78-170	1.6-3.5	Reeders <i>et al.</i> (1989)
		4.3	Reeders and bij de Vaate (1990)
		1.9	Aldridge <i>et al.</i> (1995)
		16.2	Fanslow <i>et al.</i> (1995)
	375	9.1	Sprung (1995)
	40	4.1	Berg <i>et al.</i> (1996)
	125	2.1	
	114-133		Roditi <i>et al.</i> (1996)
	22-80	6.1-13.5	Lei <i>et al.</i> (1996)
	125-223	4.6-9.1	Horgan and Mills (1997)
110-225	3.1-6.9	Diggins (2001)	
60-170	3.8-10.7		
<i>Dreissena bugensis</i>	40-200	2.2-10.8	Diggins (2001)
	120-310	2.7-7.0	
<i>Corbicula fluminea</i>	347	20.5	Buttner and Heidinger (1981)
	567	1.9	Way <i>et al.</i> (1990)
	490	2.2	Silverman <i>et al.</i> (1997)
<i>Limnoperna fortunei</i>	19-133	11.9-24.5	Rückert <i>et al.</i> (2004)
	125-350	9.9-29.5	Sylvester <i>et al.</i> (2005)
	100-214	1.5-3.1	Cataldo <i>et al.</i> (2012)
<i>Mytilus edulis</i>	17-2767	1.1-11.3	Winter (1973)
		1.9	Bayne and Scullard (1977)
	281-799		Clausen and Riisgård (1996)

increase in bacterial density, activity, metabolic diversity and structure in zebra mussel clusters relative to bare sediment. Here we observed a shift in bacterial community composition and decreased community variability in the water column under high *Bellamy*a filtration rates, but no change in total abundance.

*Bellamy*a may interact directly with microbial populations by utilizing bacteria as a food source, resulting in a community shift when bacteria respond with different growth rates. Alternatively, *Bellamy*a-bacterial interactions may be indirect, mediated by the production of large quantities of fecal and pseudo-fecal material that differentially affect the composition, activity, and growth of bacterial taxa. Recent observations made for zebra mussel in the Hudson River, New York, suggest that there is increased total bacterial biomass and heterotrophic activity associated with zebra mussel colonies compared with surface sediments (Findlay *et al.*, 1998; Strayer, 1999). Invasive bivalves can directly affect the abundance of major groups of microbes through feeding and altering resource partitioning, but whether those affects are biased towards particular bacterial functional groups remains understudied. Frischer *et al.* (2000) reported that zebra mussels are likely to promote the enrichment of particular bacteria groups, including Gammaproteobacteria and Betaproteobacteria and negatively impact Deltaproteobacteria and Flavobacteria. Unfortunately, ARISA does not allow for the identification of particular bacterial taxa or groups. However, the reduced differences in bacterial community composition among replicates in the high-density *Bellamy*a treatments suggest that filtering differentially impacted bacterial taxa.

Changes in microbial abundance can have cascading ecosystem effects. Our laboratory experiments revealed that *Bellamy*a are capable of high filtration rates, thus adding to the small but growing body of research suggesting that *Bellamy*a may play an important role in freshwater ecosystems. Although we know very little about the feeding and nutrition of *Bellamy*a, limited studies from over three decades ago suggest that *Bellamy*a feeds primarily on peri-

phyton. Stomach content analysis indicated that diatoms were the largest portion of *Bellamy*a diet in the Ottawa River, and that radular feeding showed no selectivity among periphyton taxa (Stanczykowska *et al.*, 1972; Plinski *et al.*, 1978; Jokinen, 1982). More recent research examining carbon stable isotope ratios of *Bellamy*a collected from one Wisconsin lake suggests heavy reliance on benthic resources (Solomon *et al.*, 2010), although this has not been rigorously tested. Given the filter-feeding behavior of *Bellamy*a (this study) and its effects on N:P that define ecological stoichiometry and control algal growth (Johnson *et al.*, 2009), additional experimental and field efforts are needed to understand the biology and ecology of *Bellamy*a *chinesis*. In conclusion, even though *Bellamy*a are only facultative filter-feeders (compared to exclusive filter-feeders such as mussels), they have the potential to serve an important, yet unexplored role in coupling benthic and pelagic food webs in lake ecosystems (Vadeboncoeur *et al.*, 2002; Wagner *et al.* 2012); this topic requires more investigation.

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## REFERENCES

- ALDRIDGE, D. W., B. S. PAYNE & A. C. MILLER. 1995. Oxygen consumption, nitrogenous excretion, and filtration rates of *Dreissena polymorpha*

- at acclimation temperatures between 20 and 32 °C. *Canadian Journal of Fisheries and Aquatic Sciences*, 52: 1761–1767.
- BAYNE, B. L. & C. SCULLARD. 1977. Rates of nitrogen excretion by species of *Mytilus* (Bivalvia: Mollusca). *Journal of the Marine Biological Association of the United Kingdom*, 57: 355–369.
- BERG, D. J., S. W. FISHER & P. F. LANDRUM. 1996. Clearance and processing of algal particles by zebra mussels (*Dreissena polymorpha*). *Journal of Great Lakes Research*, 22: 779–788.
- BOLTOVSKOY, D., A. KARATAYEV, L. BUR-LAKOVA, D. CATALDO, V. KARATAYEV, F. SYLVESTER & A. MARINELARENA. 2009. Significant ecosystem-wide effects of the swiftly spreading invasive freshwater bivalve *Limnoperna fortunei*. *Hydrobiologia*, 636: 271–284.
- BROWN, M. V., M. S. SCHWALBACH, I. HEWSON & J. A. FUHRMAN. 2005. Coupling 16S-ITS rDNA clone libraries and automated ribosomal intergenic spacer analysis to show marine microbial diversity: development and application to a time series. *Environmental Microbiology*, 7: 1466–1479.
- BURY, J. A., B. E. SIETMAN & B. N. KARNES. 2007. Distribution of the non-native viviparid snails, *Bellamyia chinensis* and *Viviparus georgianus*, in Minnesota and the first record of *Bellamyia japonica* from Wisconsin. *Journal of Freshwater Ecology*, 22: 697–703.
- BUTTNER, J. & R. HEIDINGER. 1981. Rate of filtration in the Asiatic clam, *Corbicula fluminea*. *Transactions of the Illinois State Academy of Science*, 74: 13–17.
- CARLSSON, N. O. L., C. BRONMARK & L. A. HANSSON. 2004. Invading herbivory: The golden apple snail alters ecosystem functioning in Asian wetlands. *Ecology*, 85: 1575–1580.
- CATALDO, D., I. O' FARRELL, E. PAOLUCCI, F. SYLVESTER & D. BOLTOVSKOY. 2012. Impact of the invasive golden mussel (*Limnoperna fortunei*) on phytoplankton and nutrient cycling. *Aquatic Invasions*, 7: 91–100.
- CHAINED, N. M. & 12 OTHER AUTHORS. 2012. Population estimate of Chinese mystery snail (*Bellamyia chinensis*) in a Nebraska reservoir. *Bio Invasions Records*, 1: 283–287.
- CLARK, G. T. 2009. *Distribution, growth, and competitive impacts of the exotic Chinese mystery snail (Bellamyia chinensis) in the James River, southwest Missouri*. MS Thesis, Missouri State University, Missouri, U.S.
- CLARKE, K. R. 1993. Non-parametric multivariate analyses of changes in community structure. *Australian Journal of Ecology*, 18: 117–143.
- CLAUSEN, I. & H. U. RIISGARD. 1996. Growth, filtration and respiration in the mussel *Mytilus edulis*: No evidence for physiological regulation of the filter-pump to nutritional needs. *Marine Ecology Progress Series*, 141: 37–45.
- COUGHLAN, J. 1969. The estimation of filtering rates from the clearance of suspensions. *Marine Biology*, 2: 356–358.
- DIGGINS, T. P. 2001. A seasonal comparison of suspended sediment filtration by quagga (*Dreissena bugensis*) and zebra (*D. polymorpha*) mussels. *Journal of Great Lakes Research*, 27: 457–466.
- DILLON, R. T. 2000. *The Ecology of Freshwater Molluscs*, Cambridge, Cambridge University Press.
- EHRENFELD, J. G. 2010. Ecosystem consequences of biological invasions. *Annual Review of Ecology, Evolution, and Systematics*, 41: 59–80.
- FANSLAW, D. L., T. F. NALEPA & G. A. LANG. 1995. Filtration rates of the zebra mussel (*Dreissena polymorpha*) on natural seston from Saginaw Bay, Lake Huron. *Journal of Great Lakes Research*, 21: 489–500.
- FISHER, M. M. & E. W. TRIPLETT. 1999. Automated approach for ribosomal intergenic spacer analysis of microbial diversity and its application to freshwater bacterial communities. *Applied and Environmental Microbiology*, 65: 4630–4636.
- FRISCHER, M. E., S. A. NIERZWICKI-BAUER, R. H. PARSONS, K. VATHANODORN & K. R. WAITKUS. 2000. Interactions between zebra mussels (*Dreissena polymorpha*) and microbial communities. *Canadian Journal of Fisheries and Aquatic Sciences*, 57: 591–599.
- GOBET, A., C. QUINCE & A. RAMETTE. 2010. Multivariate cutoff level analysis (MultiCoLA) of large community data sets. *Nucleic Acids Research*, 38: e155.
- HAKENKAMP, C. C., S. G. RIBBLETT, M. A. PALMER, C. M. SWAN, J. W. REID & M. R. GOODISON. 2001. The impact of an introduced bivalve (*Corbicula fluminea*) on the benthos of a sandy stream. *Freshwater Biology*, 46: 491–501.
- HALL, R., J. L. TANK & M. F. DYBDAHL. 2003. Exotic snails dominate nitrogen and carbon cycling in a highly productive stream. *Frontiers in Ecology and the Environment*, 1: 407–411.

- HAVEL, J. E. 2011. Survival of the exotic Chinese mystery snail (*Cipangopaludina chinensis mal-leata*) during air exposure and implications for overland dispersal by boats. *Hydrobiologia*, 668: 195–202.
- HIGGINS, S. N., M. J. VANDER ZANDEN, L. N. JOPPA & Y. VADEBONCOEUR. 2011. The effect of dreissenid invasions on chlorophyll and the chlorophyll: total phosphorus ratio in north-temperate lakes. *Canadian Journal of Fisheries and Aquatic Sciences*, 68: 319–329.
- HORGAN, M. J. & E. L. MILLS. 1997. Clearance rates and filtering activity of zebra mussel (*Dreissena polymorpha*): implications for freshwater lakes. *Canadian Journal of Fisheries and Aquatic Sciences*, 54: 249–255.
- JOHNSON, P. T. J., J. D. OLDEN, C. T. SOLOMON & M. J. VANDER ZANDEN. 2009. Interactions among invaders: community and ecosystem effects of multiple invasive species in an experimental aquatic system. *Oecologia*, 159: 161–170.
- JOKINEN, E. H. 1982. *Cipangopaludina chinensis* (Gastropoda, Viviparidae) in North America; review and update. *Nautilus*, 96: 89–95.
- KARATAYEV, A. Y., L. E. BURLAKOVA, V. A. KARATAYEV & D. K. PADILLA. 2009. Introduction, distribution, spread, and impacts of exotic freshwater gastropods in Texas. *Hydrobiologia*, 619: 181–194.
- KERANS, B. L., M. E. DYBDAHL, M. M. GAN-GLOFF & J. E. JANNOT. 2005. *Potamopyrgus antipodarum*: distribution, density, and effects on native macroinvertebrate assemblages in the Greater Yellowstone Ecosystem. *Journal of the North American Benthological Society*, 24: 123–138.
- KONDRATEV, G. P. 1963. O nekotorye osobennost-jah filtracii u presnovodnyh molljuskov. *Nauchn. Dokl. Vyssh. Shk. Biol. Nauki.*, 1: 13–16.
- KRUSKAL, J. B. 1964. Nonmetric multidimensional scaling: a numerical method. *Psychometrika*, 29: 115–129.
- KRYGER, J. & H. U. RIISGÅRD. 1988. Filtration rate capacities of 6 species of European freshwater bivalves. *Oecologia*, 77: 34–38.
- LAVRENTYEV, P. J., W. S. GARDNER & L. Y. YANG. 2000. Effects of the zebra mussel on nitrogen dynamics and the microbial community at the sediment-water interface. *Aquatic Microbial Ecology*, 21: 187–194.
- LEGENDRE, P. & L. LEGENDRE 1998. *Numerical Ecology*, Amsterdam, The Netherlands, Elsevier Scientific.
- LEI, J., B. S. PAYNE & S. Y. WANG. 1996. Filtration dynamics of the zebra mussel, *Dreissena polymorpha*. *Canadian Journal of Fisheries and Aquatic Sciences*, 53: 29–37.
- LOHNER, R. N., SIGLER, V., MAYER, C. M. & C. BALOGH. 2007. A comparison of the benthic bacterial communities within and surrounding *Dreissena* clusters in lakes. *Microbial Ecology*, 54: 469–477.
- MACKIE, G. L. 1999. Introduction of molluscs through the import for live food. In: CLAUDI, R. & J. LEACH. (eds.) *Non-indigenous freshwater organisms: vectors biology and impacts*. Boca Raton: Lewis Publishers.
- MICHEEV, V. P. 1966. O skorosti fil tracii vody Drejssenoj. *Trl Inst. Biol. Vodokhran Akad. Nauk. SSSR*, 12: 134–138.
- MILLS, E. L., J. H. LEACH, J. T. CARLTON & C. L. SECOR. 1993. Exotic species in the Great Lakes—a history of biotic crises and anthropogenic introductions. *Journal of Great Lakes Research*, 19: 1–54.
- NEWTON, R. J., S. E. JONES, A. EILER, K. D. MCMAHON & S. BERTILSSON. 2011. A guide to the natural history of freshwater lake bacteria. *Microbiology and Molecular Biology Reviews*, 75: 14–49.
- OLDEN, J. D., E. R. LARSON & M. C. MIMS. 2009. Home-field advantage: native signal crayfish (*Pacifastacus leniusculus*) out consume newly introduced crayfishes for invasive Chinese mystery snail (*Bellamya chinensis*). *Aquatic Ecology*, 43: 1073–1084.
- PLINSKI, M., W. LAWACZ, A. M. STANCZYKOWSKA & E. MAGNIN. 1978. Etude quantitative et qualitative de la nourriture des *Viviparus mal-leatus* (Reeve) (Gastropoda, Prosobranchia) dans deux lacs de la region de Montreal. *Canadian Journal of Zoology*, 56: 272–279.
- RAWLINGS, T. A., K. A. HAYES, R. H. COWIE & T. M. COLLINS. 2007. The identity, distribution, and impacts of non-native apple snails in the continental United States. *BMC Evolutionary Biology*, 7: 97.
- REEDERS, H. H. & A. BIJ DE VAATE. 1990. Zebra mussels (*Dreissena polymorpha*): a new perspective for water quality management. *Hydrobiologia*, 200/201: 437–450.

- REEDERS, H. H., A. B. DEVAATE & F. J. SLIM. 1989. The filtration rate of *Dreissena polymorpha* (Bivalvia) in 3 Dutch Lakes with reference to biological water quality management. *Freshwater Biology*, 22: 133–141.
- RICCIARDI, A. 1998. Global range expansion of the Asian mussel *Limnoperna fortunei* (Mytilidae): Another fouling threat to freshwater systems. *Biofouling*, 13: 97–106.
- RIISGÅRD, H. U. 2001. On measurement of filtration rates in bivalves - the stony road to reliable data: review and interpretation. *Marine Ecology Progress Series*, 211: 275–291.
- RODITI, H. A., N. F. CARACO, J. J. COLE & D. L. STRAYER. 1996. Filtration of Hudson River water by the zebra mussel (*Dreissena polymorpha*). *Estuaries*, 19: 824–832.
- RÜCKERT, G., M. C., S. C. & M. E. ROLLA. 2004. Alimentação de *Limnoperna fortunei* (Dunker 1857): taxas de filtração com ênfase ao uso de Cyanobacteria. *Acta Scientiarum, Biological Sciences*, 26: 421–429.
- RUDI, K., M. ZIMONJA, P. TROSVIK & T. NAES. 2007. Use of multivariate statistics for 16S rRNA gene analysis of microbial communities. *International Journal of Food Microbiology*, 120: 95–99.
- SILVERMAN, H., S. J. NICHOLS, J. S. CHERRY, E. ACHBERGER, J. W. LYNN & T. H. DIETZ. 1997. Clearance of laboratory-cultured bacteria by freshwater bivalves: differences between lentic and lotic unionids. *Canadian Journal of Zoology*, 75: 1857–1866.
- SOES, D. M., G. D. MAJOR & S. M. A. KEULEN. 2011. *Bellamyia chinensis* (Gray, 1834) (Gastropoda: Viviparidae), a new alien snail species for the European fauna. *Aquatic Invasions*, 6: 97–102.
- SOLOMON, C. T., J. D. OLDEN, P. T. J. JOHNSON, R. T. DILLON & M. J. VANDER ZANDEN. 2010. Distribution and community-level effects of the Chinese mystery snail (*Bellamyia chinensis*) in northern Wisconsin lakes. *Biological Invasions*, 12: 1591–1605.
- SOUSA, R., J. L. GUTIERREZ & D. C. ALDRIDGE. 2009. Non-indigenous invasive bivalves as ecosystem engineers. *Biological Invasions*, 11: 2367–2385.
- SPRUNG, M. 1995. Physiological energetics of the zebra mussel *Dreissena polymorpha* in Lakes. 2. Food uptake and gross growth efficiency. *Hydrobiologia*, 304: 133–146.
- STANCZYKOWSKA, A. M., M. M. PLINSKI & E. MAGNIN. 1972. Study on 3 populations of *Viviparus malleatus* (Reeve) (Gastropoda, Prosobranchia) of region of Montreal. 2. Qualitative and quantitative study on nutrition. *Canadian Journal of Zoology*, 50: 1617–1624.
- STRAYER, D. L. 1999. Effects of alien species on freshwater mollusks in North America. *Journal of the North American Benthological Society*, 18: 74–98.
- STRAYER, D. L. 2012. Eight questions about invasions and ecosystem functioning. *Ecology Letters*, 15: 1199–1210.
- SYLVESTER, F., J. DORADO, D. BOLTOVSKOY, A. JUAREZ & D. CATALDO. 2005. Filtration rates of the invasive pest bivalve *Limnoperna fortunei* as a function of size and temperature. *Hydrobiologia*, 534: 71–80.
- TOMS, J. D. & M. L. LESPERANCE. 2003. Piecewise regression: a tool for identifying ecological thresholds. *Ecology*, 84: 2034–2041.
- VADEBONCOEUR, Y., M. J. VANDER ZANDEN & D. M. LODGE. 2002. Putting the lake back together: re-integrating benthic pathways into lake food web models. *BioScience*, 52: 44–54.
- VAN DER PUTTEN, W. H., J. N. KLIRONOMOS & D. A. WARDLE. 2007. Microbial ecology of biological invasions. *ISME Journal*, 1: 28–37.
- VAUGHN, C. C. & D. E. SPOONER. 2006. Scale-dependent associations between native freshwater mussels and invasive *Corbicula*. *Hydrobiologia*, 568: 331–339.
- VIERGUTZ, C., M. KATHOL, H. NORF, H. ARNDT & M. WEITERE. 2007. Control of microbial communities by the macrofauna: a sensitive interaction in the context of extreme summer temperatures? *Oecologia*, 151: 115–124.
- WAGNER, A., S. VOLKMAN & P. M. A. DETTINGER-KLEMM. 2012. Benthic–pelagic coupling in lake ecosystems: the key role of chironomid pupae as prey of pelagic fish. *Ecosphere*, 3(2): 14. <http://dx.doi.org/10.1890/ES11-00181.1>.
- WAY, C. M., D. J. HORNBAACH, C. A. MILLERWAY, B. S. PAYNE & A. C. MILLER. 1990. Dynamics of filter feeding in *Corbicula fluminea* (Bivalvia, Corbiculidae). *Canadian Journal of Zoology*, 68: 115–120.
- WINTER, J. E. 1973. The filtration rate of *Mytilus edulis* and its dependence on algal concentration, measured by continuous automatic recording apparatus. *Marine Biology*, 22: 317–328.

WOOD, W. M. 1892. *Paludina japonica* Mart. for sale in the San Francisco Chinese markets. *Nautilus*, 5: 114–115.

YANNARELL, A. C., A. D. KENT, G. H. LAUSTER, T. K. KRATZ & E. W. TRIPLETT. 2003. Temporal patterns in bacterial communities in three tem-

perate lakes of different trophic status. *Microbial Ecology*, 46: 391–405.

YANNARELL, A. C. & E. W. TRIPLETT. 2005. Geographic and environmental sources of variation in lake bacterial community composition. *Applied and Environmental Microbiology*, 71: 227–239.