

Assimilation of particulate organic matter, particulate organic nitrogen, and particulate organic phosphorus from filtered green microalgae (*Tetraselmis* sp.) by green mussel (*Perna viridis*)

Somsak Pattananantapan^{1*}, Wanna Musig² and Yont Musig¹

ABSTRACT

Percentage of organic matter, nitrogen and phosphorus from filtered *Tetraselmis* sp. assimilated by green mussel (*Perna viridis*) were estimated from total amount of organic matter and nutrients in filtered microalgae and the amount of organic matter and nutrients remaining in feces and pseudofeces of the bivalves. According to this study, 37.8% of filtered particulate organic matter, 54.0% of filtered particulate organic nitrogen, and 77.7% of filtered particulate organic phosphorus were assimilated by green mussel. Feces and pseudofeces of the bivalve which were rejected or excreted back into the water before settling down to the bottom contained 62.2% of particulate organic matter, 46.0% of particulate nitrogen, and 22.3% of particulate organic phosphorus. The results of this study indicated the possibility of using green mussels to remove particulate organic matter, particulate organic nitrogen and particulate organic phosphorus from surrounding water by assimilating some portion of them into its biomass while transferring the other portion of unassimilated ingested particulate organic matter and nutrients together with uningested filtered particulate organic matter and nutrients to the sediment.

INTRODUCTION

Suspension-feeding bivalves can clear seston particles greater than 3- μ diameter from water column with high efficiency (Bayne and Newell, 1983; Bayne and Hawkins, 1992). Captured particles are sorted on pallial organs prior to ingestion, with less nutritious and excess particles being immediately rejected as pseudofeces. Ingested material is digested and the remains are excreted as feces. Bivalves digest and

assimilate different sources of particulate organic matter with varying efficiencies (Bayne and Newell, 1983; Kreeger and Newell, 2001). Some parts of undigested and/or unassimilated organic matter and nutrients are excreted as feces. A substantial amount of particulate organic matter is also rejected as pseudofeces. This amount of uningested and unassimilated organic matter and nutrients are eventually transferred to the sediment surface through the sedimentation of feces and pseudofeces particles. Hence,

¹Department of Aquaculture, Faculty of Fisheries, Kasetsart University, Bangkok 10900, Thailand

²Department of Biology, Faculty of science, Ramkhamhaeng University, Bangkok 10240, Thailand

*Corresponding author, e-mail: bb_ku53@hotmail.com

suspension-feeding bivalves have an important role in organic matter and nutrient recycling in aquatic ecosystems. Filtering of particulate organic matter by bivalve molluscs helps lessen the problems of eutrophication and algal blooms in water bodies. However, their ability in cleaning up eutrophic waters depends directly on their ability to filter and assimilate particulate organic matter and particulate nutrients. In this study, the assimilation of filtered particulate organic matter, filtered particulate nitrogen and filter particulate phosphorus from filtered green microalgae (*Tetraselmis* sp.) by green mussel (*Perna viridis*) was investigated in order to evaluate the green mussel's ability to remove particulate organic matter and particulate nutrients from surrounding waters.

MATERIALS AND METHODS

The study was done in 1000-ml glass aquaria filled with 500 ml seawater. Green mussel used for the experiment were collected from the green mussel farm of the Sriracha Fisheries Research Center in Chonburi province. The bivalves were transported to the laboratory of the Faculty of Fisheries, Kasetsart University, Bangkok, and acclimated in a fiber-glass tank for 1 week prior to the experiment. Feeding was stopped 24 h before the experiment. The size of the green mussels used for this experiment was between 5.3 to 6.6 cm in length and 8.42 to 15.71 g fresh total weight (including shell). Six aquaria were set for the experiment, with three bivalves in each aquarium. The green mussels in each experimental unit were fed with the same

amount of green microalgae, *Tetraselmis* sp. After 5 h, the bivalves were removed from the experimental aquaria and placed in another set of aquaria with clean seawater. Water samples were taken from each aquarium to analyze for the remaining amount of the algae. Feces and pseudofeces were also collected from each aquarium after the bivalves were removed and were collected again from the second set of aquaria 12 h after the green mussels were transferred into them. Samples were pooled in pair between aquaria 1 and 2, 3 and 4, and 5 and 6, to obtain three pool samples before analyzing. Distilled water was added to feces and pseudofeces samples and sample volume was adjusted to 150 ml. Then the samples were gently mixed by homogenizer and divided into three equal parts for the analysis of particulate organic matter, particulate nitrogen, and particulate phosphorus.

Dried weights of algae, feces and pseudofeces were measured by filtering the samples through G/FC glass fiber filter, drying them in an oven at 103 - 105°C overnight, and weighing (APHA *et al.*, 2005). Then the samples were transferred to a muffle furnace and ignited at 450°C for 4 h, cooled in dessicator and weighed to obtain organic matter fraction which is equal to the weight lost after 4 h in the muffle furnace. Particulate nitrogen was analyzed using Kjeltec 1035 analyzer unit, Tecator: Digestion block, Foss, Model 2520. Particulate phosphorus was analyzed spectrophotometrically after percholic acid digestion.

Assimilation rates of particulate organic matter, particulate nitrogen, and particulate organic phosphorus from filtered

Tetraselmis sp. by green mussel were calculated as follows:

Assimilation rate (%) =

$$\frac{\text{total amount in filtered } \textit{Tetraselmis} \text{ sp.} - \text{amount in feces and psuedofeces}}{\text{total amount in filtered } \textit{Tetraselmis} \text{ sp.}} \times 100$$

RESULTS AND DISCUSSION

The amount of particulate organic matter added in the form of green microalgae,

Tetraselmis sp., into each experimental aquarium containing green mussel was 31.50 mg. Within 5 h of the experimental period, the green mussel had filtered all added particulate organic matter from the water column. From 31.50 mg filtered particulate organic matter, 19.60 mg or 62.2% were rejected as psuedofeces and defecated as feces. Percentage of filtered particulate organic matter assimilated by green mussels was 37.8%. Assimilation rate per biomass of filtered particulate organic matter by green mussel was 0.39 mg/g total wet weight (including shell) or 21.78 mg/g dry weight (excluding shell) (Table 1).

Table 1. Assimilation of particulate organic matter (POM), particulate organic nitrogen (PON), and particulate organic phosphorus (POP) from filtered *Tetraselmis* sp. by green mussel (mean \pm S.D.)

	POM	PON	POP
Total weight of green mussel (g):			
wet weight including shell	80.41 \pm 3.34	80.41 \pm 3.34	80.41 \pm 3.34
dry weight without shell	1.45 \pm 0.02	1.45 \pm 0.02	1.45 \pm 0.02
Initial amount in given feed (mg)	31.50	4.331	0.695
Amount remaining in water after 5 h filtration (mg)	0	0	0
Percent removal from water	100	100	100
Amount filtered from water by green mussel (mg):	31.50	4.331	0.695
mg/g wet weight including shell	0.39 \pm 0.2	0.054 \pm 0.002	0.009 \pm 0.0004
mg/g dry weight excluding shell	21.78 \pm 0.23	2.994 \pm 0.032	0.480 \pm 0.0051
Rejected as psuedofeces and defecated as feces:			
mg	19.60 \pm 2.42	1.992 \pm 0.331	0.155 \pm 0.0141
%	62.2 \pm 7.7	46.0 \pm 7.7	22.3 \pm 2.01
Assimilated by green mussel (%)	37.8 \pm 7.7	54.0 \pm 7.7	77.7 \pm 2.01

Nitrogen content of added *Tetraselmis* sp. was 4.331 mg. Of the added 4.331 mg particulate organic nitrogen filtered by green mussels, 1.992 mg or 46.0% of filtered particulate organic nitrogen was rejected as pseudofeces and defecated as feces. Percentage of filtered particulate organic nitrogen assimilated by green mussel was 54.0%. Assimilation rate per biomass of filtered particulate nitrogen by green mussel was 0.054 mg/g of total wet weight (including shell) or 2.994 mg/g dry weight (excluding shell) (Table 1).

Phosphorus content of added *Tetraselmis* sp. was 0.969 mg. Of the added 0.969 mg particulate organic phosphorus filtered by green mussels, 0.155 mg or 22.3% of filtered particulate organic phosphorus was rejected as pseudofeces and defecated as feces. Percentage of filtered particulate organic phosphorus assimilated by green mussel was 77.7%. Assimilation rate per biomass of filtered particulate organic phosphorus by green mussel was 0.009 mg/g total wet weight (including shell) or 0.480 mg/g dry weight (excluding shell) (Table 1).

Results from this experiment indicated that green mussels are very effective in clearing seston particles. All *Tetraselmis* sp. added into the experimental aquaria were filtered out from the water column within 5 h. Utilization rate of filtered particulate organic matter at the rate of 37.8% indicated that a bigger portion (62.2%) of organic matter removed from the water column via green mussel filtration was rejected as pseudofeces and/or defecated as feces. In bivalve mollusks, captured particles are sorted on pallial organs prior

to ingestion, with less nutritious and excess particles being immediately rejected as pseudofeces (Newell and Jordan, 1983; Newell and Langdon, 1996; Ward *et al.*, 1997). Ingested material is subjected to extracellular and intracellular digestion, and the remains are defecated. According to Bayne and Newell (1983) and Kreeger and Newell (2001), bivalves digest and assimilate different sources of particulate organic matter with efficiencies that can vary from 20 to 90%, which indicated that bivalves can assimilate between 20 and 90% of ingested particulate organic matter. In this experiment, assimilation efficiency of particulate organic matter by green mussel was 37.8% or higher depending on how much filtered microalgae was ingested. If all filtered microalgae were ingested, assimilation efficiency will be 37.8%. Normally some part of filtered particulate matter will be rejected out as pseudofeces which will result in higher value of assimilation efficiency. According to Newell and Jordan (1983) and Kaspar *et al.* (1985), 50% of particulate organic nitrogen cleared from the water column by eastern oyster (*Crassostrea virginica*) was assimilated, and the remainder was voided in biodeposits leading to the increase in sediment nutrient content.

Assimilation rates of particulate organic nitrogen and particulate organic phosphorus in filtered *Tetraselmis* sp. by green mussel in this experiment were 54.0 and 77.7%, respectively. High assimilation rates of particulate organic nitrogen and particulate organic phosphorus by green mussel indicated their potential as a zooremediators for the remediation of eutrophic water with high content of particulate organic nutrients

and phytoplankton. Dame and Dankers (1988) reported that mussel bed of *Mytilus edulis* filtered out suspended solids and significantly reduced chlorophyll *a* and organic carbon in overlying water.

However, 46% of filtered particulate organic nitrogen and 22.3% of filtered particulate organic phosphorus were left in the forms of feces and pseudofeces. In natural habitats, these biodeposits tend to fall down to sediment surface. Feces and pseudofeces are voided from bivalves as mucus bound aggregates which have a faster sinking velocity about 40 times that of nonaggregated particles (Kautsky and Evan, 1987; Widdows, *et al.*, 1998). In locations where bottom water currents are below the critical erosional bottom shear stress (Newell *et al.*, 2005), the biodeposits undergo a de-watering process and gradually become incorporated into the sediments (Haven and Morales-Alamo, 1966, 1968; Kaspar *et al.*, 1985; Jaramillo *et al.*, 1992; Widdow *et al.*, 1998) leading to the increase in sediment nutrient content (Kaspar *et al.*, 1985; Kautsky and Evan, 1987; Deslous-Paoli *et al.*, 1992). Resuspension of biodeposits from intertidal or shallow-water bivalve population is more likely than those from bivalves living in either deeper water or grown in suspended aquaculture systems, where the underlying sediments are isolated from frequent disturbance by wave action (Dame *et al.*, 1991a).

The ability of green mussels to clear off all added *Tetraselmis* sp. within 5 h indicated their ability to exert control on phytoplankton population in eutrophic water. A number of studies have provided

strong evidences that natural population of suspension-feeding bivalves can exert top-down control of phytoplankton in coastal water (Jordan and Valiela, 1982; Asmus and Asmus, 1991; Dame *et al.*, 1991).

CONCLUSION

Green mussels are very effective in filtering suspended solids. All added particulate organic matter in the form of green microalgae, *Tetraselmis* sp, were filtered out from water within 5 h. Organic matter, nitrogen, and phosphorus from filtered particulate matter were assimilated at the rates of 37.8% for filtered particulate organic matter, 54.0% for filtered particulate organic nitrogen, and 77.7% for filtered particulate organic phosphorus. Results from this study indicated the ability of green mussels to remediate particulate organic matter rich water by filtering, ingesting and assimilating some portion of particulate organic matter and nutrients into their biomass while transferring the other portion of unassimilated ingested particulate organic matter and nutrients together with uningested filtered particulate organic matter and nutrients to the bottom sediment. However, more studies are needed to confirm its assimilation efficiency on other species of microalgae and other particulate organic matters found in eutrophic waters.

ACKNOWLEDGEMENT

This research is a part of a research project funded by The National Research Council of Thailand.

LITERATURE CITED

- APHA, AWWA and WEF. 2005. **Standard Methods for the Examination of Water and Wastewater 21th ed.** APHA, Washington, D.C.
- Asmus, R.M. and H. Asmus. 1991. Mussel beds, limiting or promoting phytoplankton. **J. Exp. Mar. Biol. Ecol.** 148:215-232.
- Bayne, B.L. and R.C. Newell. 1983. Physiological energetic of marine mollusca. In: A.S.M. Saleudin and K.M. Wibur, editors. **The Mollusca**, vol 4. New York: Academic Press. pp. 407-515.
- Bayne, B.L. and A.J.S. Hawkins. 1992. **Ecological and physiological aspects of herbivory in benthic suspension-feeding molluscs.** In: D.M. Jphn, S.J. Hawkins and J.H. price, editors. Plant-animal interactions in the marine benthos. Systematics Association Special Volume No. 46, Oxford: Clarendon Press. pp 265-288.
- Dame, R.F. and N. Dankers. 1988. Uptake and release of materials by Wadden Sea mussel bed. **J.Exp.Mar.Biol. Ecol.** 118:207-216.
- Dame, R.F., N. Dankers and T.C. Prins. 1991. The influence of mussel beds on nutrients in the Wadden Sea and eastern Scheldt estuaries. **Estuaries**, 14:130-138.
- Deslous-Paoli, J.M., A.M. Lannou and P. Geairon. 1992. Effects of the feeding behavior of *Crassostrea gigas* (Bivalve mollusk) on bioedimentation of natural particulate matter. **Hydrobiologia** 231:85-91.
- Haven, D.S. and R. Morales-Alamo. 1966. Aspects of biodeposition by oysters and other invertebrate filter feeders. **Limnol. Oceanogr.** 11:487-498.
- Haven, D.S. and R. Morales-Alamo. 1968. Occurrence and transport of faecal pellets in suspension in a tidal estuary. **Sediment. Geol.** 2:141-151.
- Jaramillo, E., C. Bertran and A. Bravo. 1992. Mussel biodeposition in an estuary in southern Chile. **Mar. Ecol. Prog. Ser.** 82:85-94.
- Jordan, T.E. and I. Valiela. 1982. A nitrogen budget of the ribbed mussel, *Geukensia demissa*, and its significant in nitrogen flow in a New England salt marsh. **Limnol. Oceanogr.** 27:75-90.
- Kaspar, H.F., P.A. Gillespie and I.C. Boyer. 1985. Effects of mussel aquaculture on the nitrogen cycle and benthic communities in Kenepuru Sound, Marlborough Sounds, New Zealand. **Mar. Bol.** 85:127-136.
- Kautsky, N. and S. Evan. 1987. Role of biodeposition by *Mytilus edulis* in the circulation of matter and nutrients in a Baltic coastal ecosystem. **Mar. Ecol. Prog. Ser.** 38:201-212.
- Kreeger, D.A. and R.I.E. Newell. 2001. Seasonal utilization of different seston carbon sources by the ribbed mussel, *Geukensia demissa* (Dillwyn) in a mid- Atlantic salt marsh. **J.Exp. Mar. Biol.Ecol.** 260:71-91.
- Newell, R.I.E. and S.J. Jordan. 1983. Preferential ingestion of organic material by the American oyster, *Crassostrea virginica*. **Mar. Ecol. Prog. Ser.** 13:47-53.

- Newell, R.I.E. and C.J. Langdon. 1996. **Mechanisms and Physiology of Larval and Adult Feeding.** In: V.S. Kennedy, R.I.E. Newell and A. Eble, editors. *The Eastern Oyster, Crassostrea virginica*. College Park, M.D.: Maryland Sea Grant Publication. Pp. 185-230.
- Newell, R.I.E., T.R. Fischer and R.R. Holyoke. 2005. **Influence of eastern oysters on N and P regeneration in Chesapeake Bay, USA.** In: R. Dame and S. Olenin, editors. *The comparative roles of suspension-feeders in ecosystem.* NATO Science Series IV-Earth and Environmental Sciences. Dordrech: Kluwer.
- Ward, J.E., J.S. Levinton and S.E. Shumway. 1997. Site of particle selection in bivalve mollusk. **Nature** 390:131-132.
- Widdows, J., M.D. Brinsley and P.N. Salkeld. 1998. Use of annular flume to determine the influence of current velocity and bivalves on material flux at the sediment-water interface. **Estuaries.** 21:552-559.