Impact of Green Mussel (*Perna Viridis*) Raft-Culture on Benthic Environment in Sriracha Coastal Water, Thailand

Tritep Vichkovitten, Alongot Intarachart, and Kanokwan Khaodon

Abstract— *Field studies were carried out to determine and compare the impact of organic loads due to the biodeposition of a mussel raft culture farm on sediment in Thai coastal waters. The sediment and water environmental parameters were examined in January, April, July and October 2006 at four stations. Three stations were located in the mussel farms that varied in terms of operation duration: long period of operation (>6 yr), intermediate period of operation (3-5 yr) and newly established (1-2 yr) while the forth was designed as a reference station located about 1 km away from a mussel farm. The results indicated differences in sediment organic matter (OM) content between mussel raft culture stations compared to reference station, with much higher OM found in the prolonged culture duration. The amount of sediment sulfide concentrations at the mussel cultured site were higher than at the reference site indicated greater anaerobic metabolism. The porewater pools of phosphate were generally low (<20 µM) throughout the examined depths at all studied sites; however, higher concentrations were observed at the reference site compared to raft mussel cultured sites. Like phosphate, mussel culture duration played a role on ammonium concentrations in porewater (200-600 µM), as more abundant pools were measured at the longer duration period. Despite being two orders of magnitude lower than* NH_4^+ *in the porewater, the concentrations of nitrite+nitrate* ($NO_2^-+NO_3^-$) *were quite constant throughout the examined depth and did not show a significant site difference. Seasonal variations of sediment sulfide and regernarated nutrients were less pronounced between raft mussel cultured sites, however, the highest pools were observed in summer.*

Keywords— **Biodeposition, green mussel, *perna viridis*, sediment, sulfide.**

1. INTRODUCTION

Shellfish aquaculture is growing rapidly in both culture areas and production throughout the world [1]. Shellfish are suspended feeders which, compared to other organisms, provide the advantage of requriring no artificial feed. The growth of shellfish is supported by natural food sources including phytoplankton and other suspended organic materials. Therefore, shellfish farming are typically located in productive estuaries and coastal waters. After feeding on suspended particles from the water column, shellfish produce biodeposition of faeces and pseudofaeces as waste products, which can accelerate sedimentation on the seabed. The accumulation of biodeposits potentially alters the physical, chemical and biological characteristics of the benthic environments underneath the culture areas [2]-[4]. In addition, shell debris is involved in the alteration of surface sediment characteristics. However, the influence of shellfish farming on environmental quality is thought to be less severe than the impact of finfish culture, which is greater because the accumulation of biodeposits is further increased by the supply of organic matter from both consumed food and uneaten food [5]-[6]. In general, the mineralization processes of organic rich sediment increases sediment oxygen demand and supplies regenerated nutrients to the overlying water. Oxygen, which is the primary electron acceptor in the decomposition processes of organic matter, has been limited to only a thin layer on surface sediment where oxic respiration takes place. In the anoxic zone, where oxygen is absent, sulfate reduction is the most important mineralization pathway [7], generating the end product of hydrogen sulfide (H_2S), which can be toxic to benthic organisms.

Shellfish farming impacts the marine environments in a variety of ways, ranging from minimal to significant effects [8]-[10]. A wide variety of negative effects have generally been reported for shellfish farming since biodeposition can induce severe organic matter accumulation leading to development of anoxic conditions in the surface sediments during the decomposition processes [3]-[4], [7]-[10]. The remineralization of biodeposits can enhance nitrogen regeneration rates and alter denitrification [2], [8]-[10] leading to acceleration of nutrient turnover and redistribution of nutrients to the overlying water. Furthermore, burial of biodeposits may lead to the removal of nutrients from the water column [2], [9], [11]. This organic enrichment could cause dramatic changes in benthic biodiversity and community structure [4], [8], [12]. The impact of biodeposits on benthic environments may be even greater in areas where currents are not strong enough to transport these materials [13], and thus reduce the oxygenated layer in sediment and the bottom oxygen in the overlying water may be depleted.

In Thailand, shellfish production almost doubled from 74,000 ton in 1988 to 140,000 ton in 2000 [14] and accelerated to reach 297,000 ton in 2007 [15], with green mussel comprising the majority of cultured species.

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Green mussel (*Perna viridis*), which is widely distributed in the Indo-Pacific region, is recognized as a commercially valuable seafood with high culture potential in many coastal areas in the region [14], [16]. Mussel culture activities have been practiced in Thailand for more than 100 yr with the traditional culture method using bamboo poles as the mussel settlement substrates. Since 1994, raft culture technique has been developed and become popular on the east coast especially, in the Chonburi province. The advantage of raft culture compared to the bamboo pole technique is that cultured mussels are kept totally submerged all the time to avoid dehydration due to air exposure during low tide, therefore enhancing the growth of the mussels. Furthermore, materials used for raft farm construction, such as rope and buoys, can be reused with the next crop, but bamboo poles cannot. Since raft mussel culture was established in this area more than two decades ago, the number of farms has expanded dramatically. This growth has aroused rising concern over the potential impact of raft mussel culture on the marine environment. However, the environmental alterations in the coastal area of Thailand that can be caused by this kind of aquaculture activity have yet to be assessed.

The present study aimed to investigate the impact of raft mussel culture on the benthic environment of three mussel farms at different sites with varying periods of operation since farm establishment (1-2 yr, 3-5 yr and >6 yr). The sediment qualities underneath the mussel raft were examined for both spatial and temporal effects.

2. MATERIALS AND METHODS

2.1 Studies sites

The study was carried out in Sriracha Bay of Chonburi province, which is located in the inner Gulf of Thailand (Fig. 1). Sriracha Bay is open and generally exposed to wind and wave action, resulting in less accumulation of natural sedimentation. This area is influenced by large amounts of freshwater from the Bangpakong River, particularly in the rainy season, which makes it suitable as a coastal aquaculture ground for a number of species including oyster (*Crasostrea* sp.) and green mussel (*Perna viridis*).

Raft mussel culture has been practiced in Sriracha Bay for more than two decades and has become a more popular mussel culture method than the old fashioned bamboo pole method because many parts of the bay have a hard subsurface sediment below 30 cm depth, which makes it difficult to hold the 6-8 m long bamboo poles in position against the water current. Green mussel culture can be operated in this area all year round, but the two most suitable times for spat collection are November to February and May to July [17]. Green mussels can be harvested six months after spat settlement. The operating size of a raft culture varies from 10 m x 10 m to 40 m x 40 m depending on areas and operating budgets.

Four sites were chosen on the basis of mussel culture duration since farm establishment, which expected to create the different biogeochemical conditions on surface sediment beneath the mussel raft. All sampling stations were located in the center of the mussel culture rafts (40 m x 40 m in size). Station M1 (0708273UTM1458014) represented an area in which farming has been practiced for more than six years; station M2 (0708840UTM 1458646) for three to five years; and station M3 (0708433 UTM1458646) for one to two years. The reference station (R) was located about one kilometer away from raft mussel culture operation (0770862UTM1459797). Field collections were conducted four times in 2006, January, April, July and October to investigate station differences and seasonal variations of raft mussel culture on sediment and porewater quality.

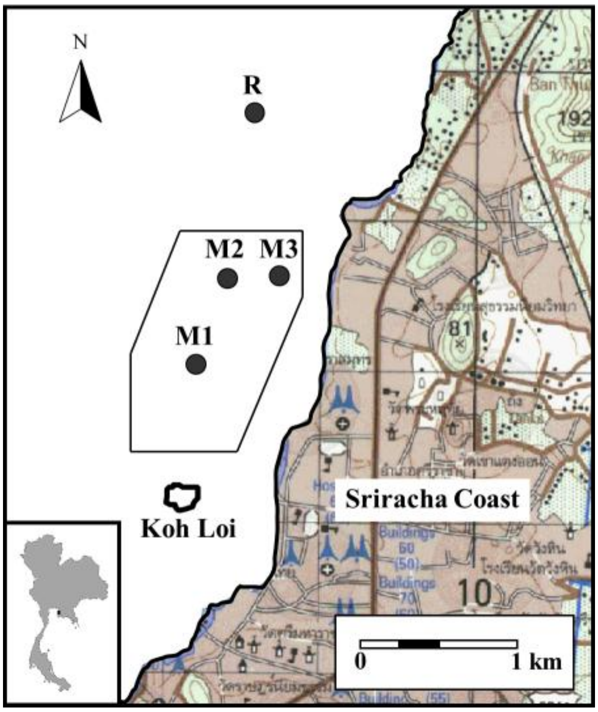

Fig. 1. Sampling stations in Sriracha Bay of Chonburi Province located on the east coast in the inner Gulf of Thailand.

2.2 Sediment and porewater quality measurement

The general conditions of the sediment underneath the mussel raft, including shell and shell fragments, were visually investigated prior to the core sampling. Sediment samples were retrieved from each sampling station in triplicate plexiglass hand corers (i.d. 8 cm) by divers and kept in coolers until delivered to the laboratory. Each sediment core was then sliced into 1-cm sections from the surface down to 5 cm depth. The sediment at each depth was kept in a zip-lock bag and mixed well prior to the analyses. Each section was divided into two portions, one used for water content, organic matter and acid volatile sulfide determination, and the other for sediment porewater extraction. Water content was measured after the weighed sediment was dried at 105 ºC for 24 h. Organic matter content in the sediment was measured as loss on ignition (LOI) after drying at 550 ºC for 6 h. The acid volatile sulfide concentrations in the sediment were determined by acidifying sediment samples with sulfuric acid and collecting the discharged H_2S in a dosimeter tube with an H_2S-absorbent column (Gastec, Japan), which measures the amount of H_2S released. Briefly, a 1 to 2-g wet sediment was weighed into a sulfide reactor column. One end of the detector tube was connected to the sulfide reactor column via a silicone tube and the other end was connected to the suction pump. Then 2 ml of 18N H_2SO_4 was added into the reactor column, where the solid phase sulfide in the sediments was converted to H_2S. The H_2S gas was pulled in through the detector tube by a vacuum pump within 2 min of sampling time. The read value from the detector tube was then converted to a sediment dry weight basis. Porewater was extracted from the sediment by centrifugation at 5000 rpm for 30 min with a refrigerated centrifuge (Universal 32R, Hettich). Supernatant water was removed from the centrifuge tube, filtered through Whatman GF/F filters and frozen for later determination of NH_4^+, NO_2^-+NO_3^- and PO_4^{3-} concentrations by Auto Analyzer (SAN^{plus}, Skalar).

During the field sediment sampling collection, bottom water (1 m above the sediment surface) was examined for temperature, pH and salinity using a Multi-Parameter Sensor (YSI 650 MDS). Water sample was also collected for measurement of NH_4^+, NO_2^-+NO_3^- and PO_4^{3-} concentrations after being filtered through Whatman GF/F filters.

2.3 Statistical analysis

Spatial and temporal fluctuations in sediment characteristics including water content, organic matter content, acid volatile sulfide, ammonium, nitrite+nitrate and phosphate were assessed by analysis of variance (ANOVA) with space (station) and time (season) as sources of variations. Four sampling stations, consisting of M1, M2, M3 and the reference station (R), were considered as the spatial factor whereas four sampling periods, January, April, July and October 2006, represented the seasonal factor. Interaction effects between station and season were also taken into account. When a significant difference ($p<0.05$) for the main effect was observed, the means were analyzed by a Tukey multiple comparison test. All statistical analyses were carried out using MINITAB statistical software version 14 (Minitab Inc., USA).

3. RESULTS

The sediment beneath mussel rafts had a finer texture of mud and silt with a higher degree of shells and shell fragments accumulation following culture duration compared to the reference station.

Water content in the sediment at all stations reached the highest values in the upper most layers and showed a decreasing trend with depth (Fig. 2). The sediment at stations M1 and M2 was poorly compacted and had a very high water content (56-63%) in the surface layer and decreased to 28-45% in the deepest layer, whereas sediment water content from station M3 was closely related to that of reference station with 29-36% in the topmost layer and 22-27% at depth. There were statistically significant difference in sediment water content among locations ($p<0.01$) but not across the sampling times ($p=0.023$).

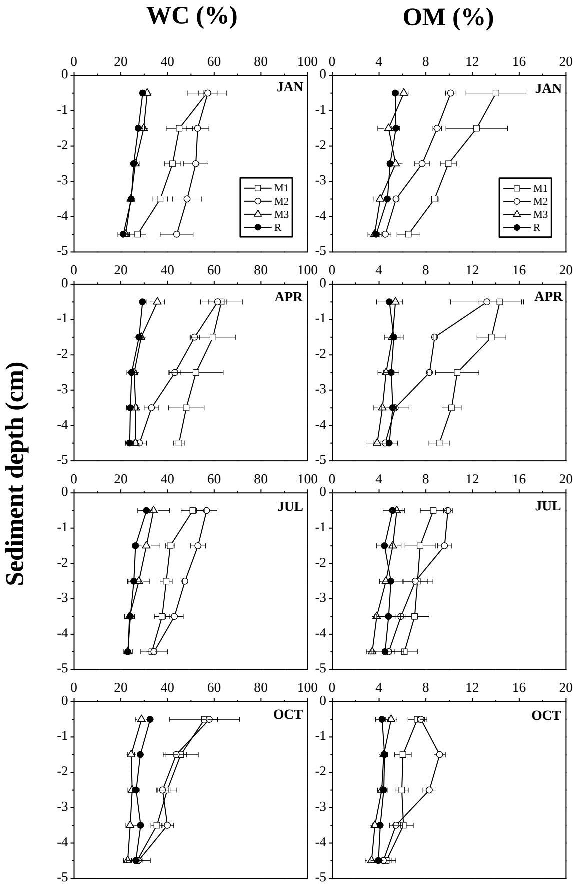

Fig. 2. Seasonal variations of sediment water content (WC) and organic matter content (OM) at mussel raft-cultured stations (M1, M2 and M3) compared to reference station (R).

There were marked differences in sediment organic matter (OM) content between mussel raft culture stations and the reference station, with much higher OM found in the prolonged culture duration (Fig. 2). OM in sediment also varied significantly among locations ($p<0.01$) and across the sampling times ($p<0.01$). High OM was found in the surface sediment from both M1 and M2 stations (7.3-14.3%), especially in January and April (10.1-14.3%) then decreased sharply with depth to reach the value between 4.4 to 4.8%. Station M3 and the reference station had quite homogeneous depth profiles with OM values ranging from 5.5% in the topmost layer to 3.8% at depth.

The acid volatile sulfide (AVS) pools were remarkably higher at mussel cultured sites compared to the reference site ($p<0.01$). The surface layer AVS pools at reference station were low (0.02-0.08 mg g DW^{-1}) and exhibited increasing trend throughout the examined depth (0.05-0.18 mg g DW^{-1}). In contrast, the subsurface maxima at 1-3 cm were generally pronounced (up to 0.75 mg g DW^{-1}) in sediment from all mussel cultured stations. It is noticed that stations M1 and M2 had a subsurface peak at the 1-2 cm layer whereas station M3 had a deeper subsurface peak at the 2-3 cm depth horizon (Fig. 3).

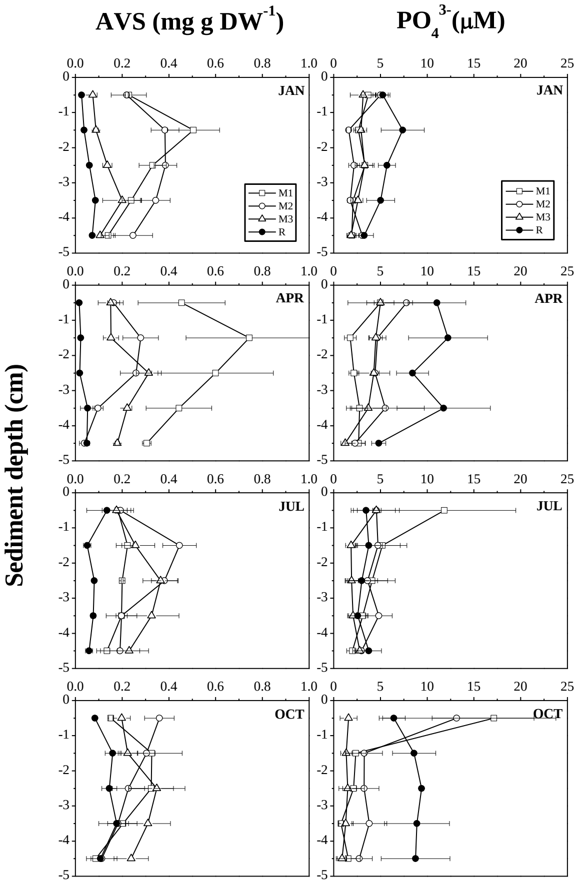

Fig. 3. Seasonal variations of vertical profiles of sediment acid volatile sulfide (AVS) and porewater phosphate concentrations (PO_4^{3-}) between mussel raft-cultured stations (M1, M2 and M3) and reference station (R).

The influence of raft-cultured farming duration was clearly seen, especially in January and April when the longer duration of farming (M1) exhibited a higher AVS pools in the sediment than the shorter durations of M2 and M3. The difference in AVS pools was compatible with the evidence of a black layer in the sediment at station M1, which was present a few millimeters beneath the sediment surface with the presence of a strong H_2S odor. Statistical analysis indicated no significant differences (p=0.528) among AVS pools at all stations; however, the greatest seasonal variability was apparent at station M1 (p<0.01) where the highest pool was observed in April and the lowest pool in October.

The porewater pools of phosphate (PO_4^{3-}) were generally low (<20 µM) in all examined depths at all sites (Fig. 3); however, higher concentrations were observed at the reference site compared to the raft mussel cultured sites. The phosphate concentrations at stations M1, M2 and M3 were high in the surface layer (up to 18 µM in 0-1 cm) and decreased to about 2 µM in the deeper layers. This was not the case for the phosphate pools at station R, where subsurface maxima were reached at 1-2 cm depth (up to 13 µM).

Seasonal variations were less pronounced among the raft mussel cultured sites than at the reference site, where the highest porewater pools of phosphate were found in April and the lowest pools in July.

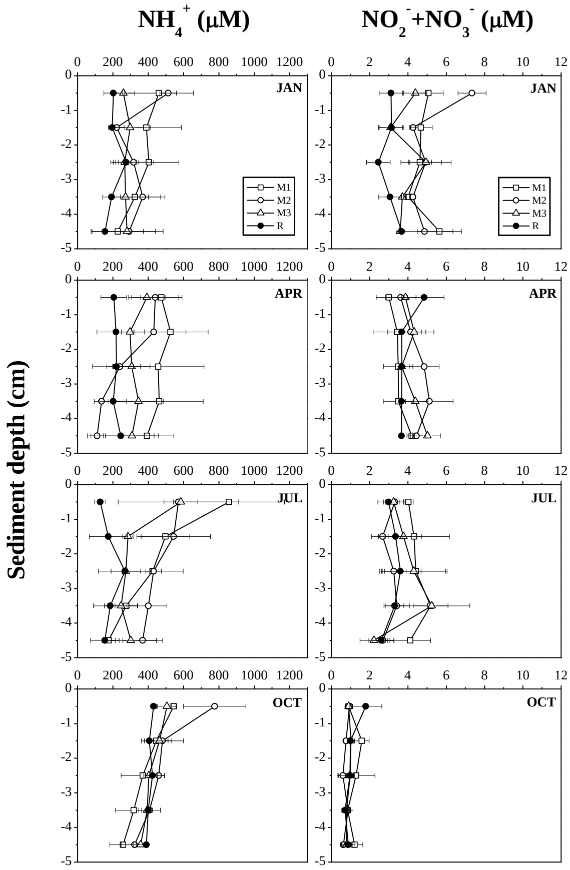

Fig. 4. Seasonal variations of vertical profiles of ammonium (NH_4^+) concentrations and nitrite+nitrate (NO_2^- +NO_3^-) concentrations between mussel raft-cultured stations (M1, M2 and M3) and reference station (R).

High ammonium (NH_4^+) pools (200-600 µM) were observed in porewater from all stations. Like phosphate, mussel culture duration played a role in ammonium concentrations in porewater, as more abundant pools were measured for the longer (M1) than for the shorter (M2 and M3) culture periods, while the reference station (R) contained the lowest pools (Fig. 4). The highest ammonium concentrations at stations M1, M2 and M3 were in the surface layer and showed a decreasing trend with depth. The depth profiles of ammonium concentrations at the reference station were devided into two categories; homogeneous gradient with depth found in April (200-250 µM) and October (390-430 µM), and presence of subsurface maxima detected in January (280 µM) and July (270 µM). Despite being two orders of magnitude lower than NH_4^+ in the porewater, the concentrations of nitrite+nitrate (NO_2^-+NO_3^-) were quite constant throughout the examined depths (p=0.701) and did not show a significant site difference (p=0.054). But seasonal influence on NO_2^-+NO_3^- concentrations was statistically significant, especially in October, when very low concentrations (0.60-1.80 µM) were detected among the four sampling stations (p<0.01).

Table 1. Bottom water conditions among four sampling stations located at raft mussel cultured sites and the reference site from January to October 2006.

Sampling period	Station	Parameters					
		Temp (ºC)	pH	Sal (‰)	PO_4^{3-} (μM)	NH_4^+ (μM)	NO_2+NO_3^- (μM)
January	M1	28.0	7.96	33.9	0.12	0.17	0.81
	M2	28.1	7.47	32.8	0.09	0.26	0.31
	M3	28.2	7.62	33.6	0.09	0.24	0.62
	R	28.1	7.84	34.2	0.20	0.24	0.33
April	M1	31.6	7.87	31.8	0.14	1.43	0.50
	M2	31.5	7.73	31.6	0.19	1.02	0.59
	M3	31.4	7.75	31.3	0.15	0.76	0.59
	R	31.4	7.80	31.1	0.33	1.07	0.29
July	M1	29.3	7.97	23.6	0.13	1.67	0.52
	M2	29.3	7.98	23.4	0.15	0.64	0.31
	M3	29.3	7.98	23.4	0.13	0.62	0.26
	R	29.3	7.98	23.5	0.12	0.24	0.31
October	M1	30.3	8.05	30.6	0.30	0.17	0.45
	M2	30.3	8.06	30.7	0.12	0.07	0.90
	M3	30.4	8.06	30.7	0.14	0.14	0.59
	R	30.0	8.06	30.3	0.26	0.17	0.43

Bottom water quality exhibited a similar pattern among the four sampling stations (Table 1). At all stations, temperature was found to vary between a minimum of 28.0 ºC in January and a maximum of 31.6 ºC in April with intermediate temperatures in July and October. The pH values varied from 7.47 to 8.06 and showed a slightly increasing trend from January to October at all stations. High salinity (32.8-34.2 ppt) was pronounced in January then decreased to 23.4-23.6 ppt in July before increasing to 30.3-30.7 ppt in October. Nutrients in bottom water were generally low and quite homogeneous at all sampling stations. Phosphate concentrations fluctuated within a narrow range of 0.09-0.33 µM with no significant difference observed among the sampling periods. Ammonium concentrations varied from 0.07 to 1.43 µM with high values recorded in April and the lowest values in January and October. Nitrite+nitrate values varied from 0.26 to 0.90 µM and exhibited a small seasonal change.

4. DISCUSSION

The present study indicates that raft-mussel culture greatly influenced the characteristics and biogeochemistry of the underlying sediments. Fine texture with high organic matter content in the sediment at mussel culture stations reflects the greater deposition of organic rich fecae and pseudofecae generated from mussel culture activities, which then accumulate within the sediments under the mussel raft and are stabilized by the matrix of shells and shell fragments. Similar accumulations of organic rich sediments have previously been reported in other environments for shellfish aquaculture [2], [6], [9]-[10], [13]. Such high organic matter inputs to the mussel-farmed sediments would be expected to fuel benthic metabolism. Sediment AVS pools at the mussel culture stations were up to 30 times higher than those at the reference station. AVS is composed of free sulfide and iron monosulfide, which forms when sulfide precipitates with porewater ferrous iron (Fe^{2+}), and can be considered to represent the initial product of sulfate reduction, the dominant anaerobic respiration process in marine sediments [7]. Therefore, the much greater pools of AVS in sediment below the mussel raft directly demonstrates that anaerobic metabolism products were sequestered at the mussel culture stations larger than at the reference station.

Although dissolved phosphate concentrations in organic rich sediment contribute to a minor fraction (~1%) of the sedimentary phosphorus pools [7], the profiles may provide information on phosphate remineralization. The porewater phosphate pools and depth profiles were similar among the mussel raft culture stations and differed markedly from those at the reference station. The reaction that controls release of phosphate to the porewater is a highly complex phenomenon involving interrelated chemical, biological and physical processes. It is known that the phosphate regeneration rate during organic matter mineralization as well as the sediment capacity to bind and retain inorganic phosphate are major reactions contribute to phosphate pools [10]-[11]. High phosphate pools in surface sediment at mussel culture stations are possibly attributable to reoxidation of iron oxides. Sulfide is a strong competitor for oxidized iron [7]. Therefore, the presence of hydrogen sulfide is resulted in the reduction of sediment sorption capacity as ferrous iron can directly precipitate sulfide to generate FeS, and this can further react with elemental sulfur or polysulfides to form pyrite (FeS_2). However, the observed low phosphate concentrations from all mussel culture stations suggest that sediment underneath the mussel rafts had higher buffering capacity compared to the reference station. Sundby et al. [18] pointed out that sorption equilibria can continue to buffer the concentration of phosphate in porewater as long as the buffering capacity is not exceeded. A clear picture of the processes of the phosphate release and uptake in sediment emerged when the ratio between dissolved ammonium and phosphate (NH_4^+/PO_4^{3-}) had been determined [19]-[20]. The variable NH_4^+/PO_4^{3-} ratios recorded in the present study suggest that reactions to phosphorus removal or addition take place. The higher NH_4^+/PO_4^{3-} ratio found at the mussel culture stations compared to the reference station are in accordance with the steady low phosphate concentration depth profiles, suggesting that a reaction to phosphorus removal occurs. The lower NH_4^+/PO_4^{3-} ratio found at the reference station was probably a consequence of background degradation of organic matter originating from phytoplankton depositions in this areas. High porewater phosphate concentrations at the reference station could be attributed to low buffering capacity leading to accumulation of phosphate in porewater. This is consistent with low AVS pools, where oxidized iron complex with phosphate is reoxidized and releases phosphate into porewater.

Ammonium pools in porewater were higher at mussel culture stations than at the reference station, and duration of farm operation had significant effects on the pools which expressed in a decreasing order as M1>M2>M3>R, suggesting that raft mussel culture influenced ammonium regeneration in porewater. Ammonium is generally the end product of both aerobic and anaerobic mineralization of organic matter. The greater ammonium concentrations in porewater at mussel culture stations can be resulted from a mineralization of organic matter addition to the sediment [2]-[3]. Increased ammonium effluxes are characteristic of enriched sediment and have been reported as the effects of shellfish cultivation on the underlying sediments in many areas [2]-[3], [8]-[10]. High concentrations found in surface sediments suggest that ammonium migrates upward from the subsurface to the surface layer and is probably released out to the water column, which is in accordance with lower pools of ammonium in the water column. The strongest ammonium release from the sediment usually occurs under anoxic conditions. Higher pools of AVS found in sediment underneath the mussel rafts compared to the reference station indicated greater anaerobic decomposition of organic matter in these sediments. Low ammonium pools at the reference station could therefore be attributed to low organic matter substrate for bacterial mineralization processes as indicated by low AVS pools in the sediment. Although station M3 had an amount of organic matter content similar to that of the reference station, the ammonium pool was greater. This phenomenon could be explained by differences in the quality of deposited material at the two stations in that biodeposits from mussel raft contained a great deal of labile and easy degradable organic matter in fecae and pseudofecae compared to the reference station. Different organic inputs combined with different nitrogen mineralization processes occurring in sediments contributed to marked difference in concentration profiles of ammonium between sediments at mussel culture stations and those at the reference station. Nitrogen cycling in sediments involves a variety of pathways where aerobic nitrification could be observed in the oxic conditions, whereas anaerobic nitrogen mineralization processes such as denitrification and dissimilatory nitrate reduction to ammonium are favorable in reduced conditions [2], [9]. In addition, Gilbert et al. [2] suggested that dissimilatory nitrate reduction to ammonium processes contributed to a majority end product of dissimilatory nitrate reduction compared to nitrogen gas. Therefore, most of the nitrate was reduced to ammonium and recycled in sediment or effluxed to the overlying water, and this was consistent with the low pools of nitrite+nitrate observed in sediments at all stations.

Among the environmental conditions, temperature has been recognized as one of the most important factor influencing the organic matter decomposition and nutrient mineralization in coastal ecosystems. The large differences in temperature with seasonal variation, especially in temperate regions, might play a major role in the variation of benthic metabolism [9], but the effects of temperature could be less pronounced in tropical regions. The effects of seasonal variation on nutrient concentrations in the water column was clearly seen in April, when the dry period along with high temperatures led to considerable increase of ammonium and phosphate productions. The ammonium release from sediments after the anoxic period could have induced an increase in ammonium concentration in the water column, which would probably have enhanced pelagic nitrification and thus increased nitrite+nitrate concentrations. However, during the wet season in July, freshwater inputs seemed to have less influence on ammonium and nitrite+nitrate in the water column. The low concentrations of ammonium in the water column in January and October suggested that slow release of ammonium flux from sediment in concert with the nitrification processes led to increased nitrite+nitrate in the water column.

5. CONCLUSION

Raft mussel culture plays a significant role on alteration of sediment characteristics and nutrient mineralization in coastal ecosystems. In the mussel station sediment enrichment of organic compounds and the consequent of benthic environmental characteristics modification determined as an increase in reduced compounds and regenerated nutrients to the overlying water suggesting that long established farm potentially cause the eutrofication in the costal areas. With increasing culture activity, therefore, continued environmental monitoring is important to ensure that the overall health of the ecosystem is maintained.

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