Variability in Recruitment of Non-native mussel *Mytilopsis adamsi* Morrison, 1946 in Haad-kaew Lagoon, Songkhla Province

Kringpaka Wangkulangkul

A Thesis Submitted in Partial Fulfillment of the Requirements for the Degree of Master of Science in Ecology (International Program)

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Author	Miss Kringpaka Wangkulangkul
Major Program	Ecology (International Program)

Major Advisor:	**Examining Committee:**
……………………………………….…	………………………………..…Chairperson
(Asst. Prof. Dr. Vachira Lheknim)	(Asst. Prof. Dr. Pitiwong Tantichodok)
	……………………………………………
	(Asst. Prof. Dr. Vachira Lheknim)
	……………………………………………
	(Assoc. Prof. Dr. Sommai Chaiyvareesajja)

The Graduate School, Prince of Songkla University, has approved this thesis as partial fulfillment of the requirements for the Master of Science Degree in Ecology (International Program)

……………………………………….…..

(Assoc. Prof. Dr. Krerkchai Thongnoo)

Dean of Graduate School

ชื่อวิทยานิพนธ์	ความผันแปรของการเข้าสู่พื้นที่ของหอยสองฝาต่างถิ่น *Mytilopsis adamsi* Morrison, 1946 บริเวณหาดแก้วลากูน จังหวัดสงขลา
ผู้เขียน	นางสาวกริ่งผกา วังกุลางกูร
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ปีการศึกษา	2552

บทคัดย่อ

ในลุ่มน้ำทะเลสาบสงขลา พบหอยสองฝาต่างถิ่น*Mytilopsis adamsi* Morrison, 1946 (Bivalvia: Dreissenidae) เป็นครั้งแรกที่หาดแก้วลากูน จังหวัดสงขลา การศึกษาครั้งนี้มีวัตถุประสงค์เพื่อทราบสภาวะที่เหมาะสมต่อการดำรงชีวิตและสืบพันธุ์รวมทั้งระยะเวลาที่มีการสืบพันธุ์ของ *M. adamsi* ซึ่งทำให้เข้าใจถึงการรักษาสมดุลในประชากร และพลวัตรของประชากร โดยการศึกษาความผันแปรเชิงสถานที่และกาลเวลาของการเข้าสู่พื้นที่และความสัมพันธ์ระหว่างการเข้าสู่พื้นที่และปัจจัยสิ่งแวดล้อมในหาดแก้วลากูน สุ่มตัวอย่างลูกหอยทุกเดือนโดยใช้แผ่นล่อลูกหอยแช่น้ำเป็นเวลา 1 เดือน หลังจากนั้นคำนวณความหนาแน่นของการเข้าสู่พื้นที่จากจำนวนลูกหอยที่ลงเกาะบนแผ่นล่อ วัดความเค็ม อุณหภูมิของน้ำ ค่าpH ค่าออกซิเจนละลายน้ำ สุ่มตัวอย่างหาค่าความหนาแน่นของลูกหอยในระยะแพลงก์ตอน แพลงก์ตอนพืชและสัตว์ ทุกๆเดือน เป็นระยะเวลา 1 ปีตั้งแต่ พฤษภาคม 2550 ถึง เมษายน 2551 โดยแบ่งออกเป็น 3 ฤดูกาล พื้นที่ศึกษาแบ่งออกเป็น 2 ลากูนย่อย คือ ลากูนกึ่งปิด และลากูนเปิด

จากการศึกษาพบว่าการเข้าสู่พื้นที่ในฤดูกาลและลากูนที่ต่างกันไม่มีความแตกต่างกันอย่างมีนัยสำคัญ แต่ความแปรผันทั้งสองลักษณะนี้กลับสังเกตได้ชัดเจน พบความหนาแน่นของลูกหอยที่เข้าสู่พื้นที่มากที่สุดในลากูนกึ่งปิด และในฤดูลมตะวันออกเฉียงใต้ การเข้าสู่พื้นที่มีมากใน 2 ช่วง คือ ระหว่างเดือนพฤษภาคมถึงเดือนสิงหาคม 2550 ในฤดูมรสุมตะวันตกเฉียงใต้ ซึ่งพบลูกหอยน้อยกว่า และระหว่างเดือนพฤศจิกายน 2550 ถึงเดือนมีนาคม 2551 ซึ่งเป็นช่วงต่อระหว่างฤดูมรสุมตะวันออกเฉียงเหนือและฤดูลมตะวันออกเฉียงใต้ ความหนาแน่นของลูกหอยมีความสัมพันธ์เชิงลบต่อความเค็ม แต่มีความสัมพันธ์เชิงบวกต่อความหนาแน่นของ สาหร่ายสีเขียวแกมน้ำเงิน สาหร่ายเซลล์เดียวที่มีแฟลกเจลลา และ ไดอะตอม

การลดลงของความเค็มในช่วงกลางฤดูมรสุมตะวันออกเฉียงเหนือและค่าความเค็มที่ต่ำในฤดูลมตะวันออกเฉียงใต้ น่าจะโน้มนำให้เกิดการเข้าสู่พื้นที่มากในช่วงรอยต่อฤดู พบความหนาแน่นของแพลงก์ตอนพืชสูงในช่วงฤดูมรสุมตะวันตกเฉียงใต้ ซึ่งน่าจะทำให้เกิดการสืบพันธุ์และการเข้าสู่พื้นที่มากเนื่องจากมีอาหารสำหรับหอยในการสืบพันธุ์อย่างพอเพียง

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Author Miss Kringpaka Wangkulangkul

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ABSTRACT

Mytilopsis adamsi Morrison, 1946 (Bivalvia: Dreissenidae) has been found for the first time in Songkhla Lake Basin as an introduced species in Haad-kaew Lagoon, Songkhla. This present study aims to gain the knowledge of *M. adamsi* favorite environmental characteristics for survival and reproduction, and the period of its reproductive activity which guide us to understand the population regulation and dynamics of the species. The investigation was done in Haad-kaew Lagoon on spatial and temporal variability of recruitment, and environmental parameters that potentially have an effect on the success of recruitment were determined. The densities of recruits were obtained from the number of mussel settle on the collecting panels that immersed in the water for a month. Salinity, water temperature, pH, dissolved oxygen, abundance of larvae and dominant phytoplankton and zooplankton were monitored. The study is comprised of 3 seasons in one year, since May 2007 to April 2008. The study sites include seasonally-closed lagoon and open lagoon of Haad-kaew Lagoon.

Although the effects of season and habitat on *M. adamsi* recruitment were not significant, the seasonal and habitat variability of recruitment in Haad-kaew Lagoon was found noticeably. The density of recruits was highest in seasonally-closed lagoon and in the south-east predominant wind season. There were two peaks of intense recruitment. A minor peak was from May to August 2007 in the south-west monsoon season, and a major pulse was from November 2007-March 2008 of the intermediate between the north-east monsoon and the south-east predominant wind. The density of *M. adamsi* recruits was negatively related to salinity, but was positively related to the density of cyanophyte, phytoflagellate, and diatom. The

decrease of salinity in the mid north-east monsoon and low salinity in the south-east predominant wind possibly caused the major peak of recruitment found. The minor peak of recruitment in south-west monsoon was potentially due to the high abundance of *M. adamsi*'s food, phytoplankton, which provided the proper condition for the reproduction.

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CHAPTER 1

General introduction & Insight

Biological invasions, or the establishments of species beyond their historical range, have been identified as one of the major threats to the maintenance of biodiversity and ecosystem (Bax *et al*., 2001). Coastal marine habitats are among the most heavily invaded systems on earth (Grosholdz, 2002). Introductions may be accidental or intentional, and the four main sources of introduction to the coasts are via aquaculture, in ballast water, attachment to ships, and when canals create new connections between oceans (Branch and Steffani, 2004). There were a large number of non-indigenous species occupied coastal estuarine and marine habitats. The impact of such species possibly occurred not only on single native species, but also extended to an entire community in both negative and positive ways (Lambert, 1992). Some species can also create new habitat or alter physical condition as the role of ecosystem engineers (Sax *et al.*, 2005). The severe economic impact of these species is also evident; in USA costs of invasion problems have been estimated to range from millions to billions of dollars annually (Sakai *et al*., 2001). Thailand is a country with very long coastline, ca 1,500 miles, associated with diverse productive coastal ecosystems and heavily used for aquaculture, tourism, and transportation. These coastal habitats were also susceptible for any alien species invaded to Thai's territory.

Among the bivalves, mussels have frequently been reported as alien invasive species, both mytilids and dreissenids, since they have high fecundity, rapid growth, a short life span, and good dispersal ability via larval dispersion. For example, a mytilid, Asian date mussel *Musculista senhousia* (Benson in Cantor, 1842), is native to north western Pacific and have been invaded North America, Europe, Australia and New Zealand (NIMPIS, 2002b). The zebra mussel *Dreissena polymorpha* (Pallas, 1771) is one of the most significant invasive species with the highly publicized impact (Nalepa and Schloesser, 1992). This species has spread over most of Western Europe and Eastern North America, and continues to expand its range (Strayer and Malcom, 2006). The invasive mussels are considered to be highly opportunistic and prolific.

They can survive under extreme environmental conditions, including wide variations in salinity, polluted water with low oxygen or high heavy metal and hydrocarbon concentrations (Raju *et al*., 1975; NIMPIS, 2002b; Rajagopal *et al*., 2003; Salgado-Barragán and Toledano-Granados, 2006).

Mytilopsis: an invader of the Indo-Pacific

Prior to 1929, *Mytilopsis* sp. (Mollusca: Bivalvia: Dreissenidae) had been reported from Fiji and described as *Mytilopsis allyneana* by Hertlein and Hanna (1949). Morton (1980) pointed out that the Indo-Pacific specimens named as *M. allyneana* were considered to be a junior synonym of the western Atlantic *Mytilopsis sallei* (Recluz, 1849) that crossed the Panama Canal in the 1960's. Later Marelli and Gray (1985) made a critical analysis of the Indo-Pacific specimens and suggested that *M. allyneana* is a synonym of *Mytilopsis adamsi* Morrison, 1946, the eastern Pacific species, and considered *M. sallei* as being a distinct species. They also proposed that the species was possibly transported to the Indo-Pacific area during the 19th century on British cargo ships that operated between Central America to Fiji and India, prior to the opening of the Panama Canal. GISP (2004) suggested that *M. adamsi* should be the valid name, while *M. sallei* is its junior synonym. In addition they proposed that *M. adamsi* is native to the tropical and subtropical western Atlantic (from the Gulf of Mexico to Colombia), but possibly transited through the Panama Canal that opened for shipping traffic in 1914. Most publications in the Indo-Pacific region referred to this species as *M. sallei*, however Salgado-Barragán and Toledano-Granados (2006) and Marelli (2007, personal communication) state that all Asian reports of *M. sallei* are likely to be *M. adamsi*. This dreissenid species has been reported to have invaded many Indo-Pacific regions including Fiji (prior to 1950), Hong Kong (early 1980s), India (ca. 1967), Indonesia, Japan, the Philippines, Singapore, Malaysia, Taiwan (1970s), Australia and Thailand (GISP, 2004; Marelli and Gray, 1985; Morton, 1989; Willan *et al*., 2000; Tan and Morton, 2006, Wangkulangkul and Lheknim, 2008).

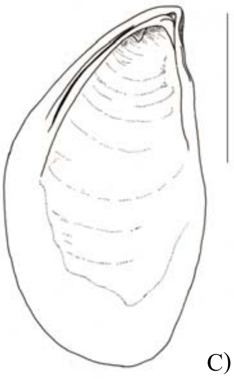
Shell Morphology

Mytilopsis adamsi is a small, finger-nail sized mussel, growing to an average length of 25mm (Figure 1.1). The shell is mytiliform with byssal threads, the exterior bears fine concentric lines. The periostracum is thin, appears white, cream and medium brown colored, and is generally eroded anteriorly. Small individuals may have a light and dark zigzag pattern. The shell interior is white to bluish-grey and is a mottled grey-black in the central portion. The shell is an inequivalve, and the right valve overlaps the left. The left valve is slightly larger postero-ventrally. The ventral margin is relatively straight, less indented and wide. The posterior muscle scar is difficult to detect. Anterior adductor muscles are attached to a shelf-like septum, which in this species is relatively flat, being nearly on the same plane as the hinge plate. The anterior retractor muscles are attached to an apophysis, located laterally to the septum near the dorsal shell margin. The apophysis is well developed, but the shape is varied. In larger specimens, the angle point into the shell cavity is about equal to or less than 90°, while smaller specimens have a broader angle.

The maximum length was reported to be 31 mm from Visakhaptnam harbor (Morton, 1981) and 30 mm (Ganapathi *et al.*, 1971) in India. The maximum width was reported to be 9.68 mm and the maximum height was 12.58 mm (Marelli and Gray, 1983).

A)

B)



C)

Figure 1.1 *Mytilopsis adamsi* Morrison, 1946

A) Right and left valve of *M. adamsi*, scale bar = 1 cm

B) Dense colony collected from Haad-kaew Lagoon, Songkhla in May 2006

C) Interior view of the left valve of *M. adamsi*, Scale bar = 1 cm

Reproduction, growth and life span

Karande and Menon (1975) reported that *Mytilopsis sallei* is an ambisexual bivalve, capable of functioning as a male, female or hermaphrodite. Morton (1989) suggested that it is dioecious and the majority of the population is probably semelparous. According to many studies, *M. adamsi* is an opportunistic r-strategist, having high, rapid growth and fast maturity rate (Ganapathi *et al*., 1971; Kalyanasundaram, 1975; Raju *et al*., 1975; Rao *et al*., 1975; Morton, 1989). Morton (1989) also reported that the minimum length at the first maturity of this species in Hong Kong was 8 mm and individuals are mature within one year. It matures within a month, at 8-10 mm shell length (Karande and Menon, 1975; Morton, 1989). Ten thousands of eggs can be released into the water column where fertilization takes place (NIMPIS, 2002a). The pelagic larval duration is short and settlement occurs within a few days (~4 days, Kalyanasundaram, 1975). A maximum size is reached within six months, and mussels live for about 12-13 months in India (Rao *et al.*, 1974) and up to 20 months in Hong Kong (Morton, 1989).

Feeding

Mytilopsis adamsi is a suspension feeder. The adult feeds on zooplankton, phytoplankton and other suspended particulate organic matter, but larva can not consume large zooplankton (NIMPIS, 2002a).

Habitat

Mytilopsis adamsi is gregarious in nature and attaches to any underwater surface. It has been found in clusters and is rarely seen as a single individual. In its native habitat, *M. adamsi* is a colonial surface dweller of sheltered waters, for example, shallow coastal lagoons (NIMPIS, 2002a). It possessed broad temperature and salinity tolerances (Raju *et al*., 1975; Bax *et al*., 2001). In introduced habitats, it is found in intertidal and shallow water, at an optimal range of temperatures from 10 to 35 °C and salinities of 0 to 27 ppt (NIMPIS, 2002a). In the laboratory, it survived at 5-40 °C and 0.083-50 ppt (Rao *et al*., 1975). It has not been found in any water deeper than a few meters.

Effects of invasion

Mytilopsis adamsi is a massive fouling organism, able to attach to every submerged substrate, such as wharves, marinas and fishing gears, (NIMPIS, 2002a). In its infected habitats, it forms dense monospecific groups, which possibly exclude most other species, and can lead to a substantial reduction in biodiversity (NIMPIS, 2002a). In contrast, many publications stated that in the presence of dreissenids mussel colony, the density of deposit-feeding and carnivorous macroinvertebrates and taxonomic richness increases (Padilla *et al*., 1996; Horvath *et al*., 1999; Ricciardi, 2003), since the mussel beds enhanced spatial heterogeneity and biodeposits. In addition demersal fish can gain more food and probably cause an increase in density (Nalepa and Schloesser, 1992). The introductions of mussels usually alter the composition of planktonic community because the mussels are suspension feeders consuming mainly phytoplankton. Most of the studies working with *Dreissena polymorpha* (the freshwater species) (Padilla *et al*., 1996; Bastviken *et* *al*., 1998; Jack and Thorp, 2000), but there have been no available data for *M. adamsi*. However the effect of mussels on the plankton is expected to be similar.

Recruitment in mussel

Seed and Suchanek (1992) defined recruitment of mussel as the process of successful colonization, which reflects the settlement rate and post-settlement mortality. The act of settlement in bivalves involves descent from the plankton to the substrate, including a sequence of swimming, crawling behavior to find a suitable substratum, and metamorphosis (Gosling, 2003). Within this process, settlement is considered to be a critical phase in the life history of a bivalve because the ability to move is then lost, and there is re-organization of the body, so mass mortality usually occurs. Post-settlement mortality is often very important. Settlement is generally difficult to measure in the field. McQuaid and Lindsay (2007) also found that recruitment usually has a positive correlation with the adult density and recruitment of post-settlement larvae influences the population structure of mussels more directly than does larval settlement. It is therefore probably a more responsible choice to use the recruitment rate as the main population parameter to predict the abundance of mussels that enter the population.

There are a number of factors that influence the recruitment and reproductive activities of mussels. The location and period of time have frequently been responsible for variations in these activities. Porri *et al*. (2006b) examined the effect of neap tide on the recruitment of *Perna perna* in South Africa and found that the recruitment intensity differed over the year; the phase of the moon appeared to have an effect on settler abundance, but only when and where densities were high. Tumanda *et al*. (1997) who studied recruitment of *Modiolus metcalfei* in the Philippines; found that the recruitment pattern appeared to be unimodal, with the peak occurring during the months of May-July. The two periods of reproductive activity per year in *M. sallei* (Recluz, 1849) were reported in Hong Kong (Morton, 1989). Tan and Morton (2006) surveyed the distribution and density of *M. sallei* in tidal monsoon drains and estuaries in Singapore and Johor Bahru (Malaysia), and concluded that recruitment patterns of *M. sallei* were different at the different locations without the explanation.

Some publications have reported that temperature and salinity were related to *M. adamsi* reproductive activity and survival (Kalyanasundaram, 1975; Raju *et al*., 1975; Rao *et al*., 1975; Morton, 1989). Spawning appears to be triggered by changes in salinity (NIMPIS, 2002a). Puyana (1995) found the spawning activity of *M. adamsi* was stimulated by a rapid drop of salinity associated with seasonal freshwater outflow. The surviving *M. adamsi* were found in a range of dissolved oxygen and pH values in India and Hong Kong (NIMPIS, 2002a). In addition the abundance of larva is usually correlated with recruitment in many mussels (Martel *et al*., 1994; Gosling, 2003; McQuaid and Lindsay, 2007). Food supplies have also been reported to have an influence on bivalve larval growth and mortality (His *et al*., 1989; Horvath and Lamberti, 1999; Gosling, 2003; Bos *et al*., 2006), and this can affect subsequent recruitment.

Current status of *Mytilopsis adamsi* Morrison, 1946 in Thailand

Recently, an invasive false mussel *Mytilopsis adamsi* has been reported to establish in Pak Phanang estuary, Haad-kaew Lagoon and Thale Sap Songkhla of the Songkhla Lake Basin and occurred on the coast in Pattani province, all are on the coast of the lower south of the Gulf of Thailand (Swennen *et al*., 2001; Wangkulangkul and Lheknim, 2008).

The Pak Phanang estuary is a part of the Pak Phanang River system (Figure 1.2). The estuary is occupied by a mangrove forest, several tidal creeks, and an extensive mud flat. In the rivers the salinity generally decreases with distance from the mouth. Salinity barrages were constructed in the lower reach of the river, which divided the river into 2 regions in the dry season. The rivers above the barrages have become temporally freshwater while the salinity below the barrage is brackish or seawater. *M. adamsi* were found occupied the whole brackish area of the Pak Phanang estuary (personal observation; Wangkulangkul and Lheknim, 2008). The personal interviewing with local people indicated that *M. adamsi* have been found in the Pak Phanang estuary for decade.

Songkhla Lake Basin consists of a large lagoon system, several rivers, and a small coastal lagoon, Haad-kaew Lagoon (Figure 1.2). The mouth of the lagoon system opens into the sea at the south part and the salinity increase southward while the water in the upper part is fresh. In the lagoon system, the mussel has colonized at the mouth of two canals that open into Thale Sap Songkhla (the lowest part), and both populations found live in the brackish water. *M. adamsi* population has occupied the entire area of Haad-kaew Lagoon. It has formed dense clusters on sea bottom, attached pneumatophores of mangrove trees and usually densely attach the fishing and aquaculture gears used in Haad-kaew Lagoon which cause economic problems because the mussel colonies clog the gear rapidly and reduce the efficiency of the gears.

When location, environmental settling and biological background of *M. adamsi* elsewhere are considered (NIMPIS, 2002a), it is possible that this species has a high potential to invade and colonize the whole part of the Songkhla Lake Basin, where more than 0.5 million people depend their livelihood on cage aquaculture and harvest aquatic resources. According to Sakai *et al*. (2001), who reviewed the population biology of invasive species and proposed classification steps in the invasion process, the current situations of *M. adamsi* in Haad-kaew Lagoon possibly matched up with the spread step, which it can reproduce and enhance its abundance rapidly. This suggested that the aquatic biodiversity and economy of local people in the Songkhla Lake Basin are possibly at the highly potential risk.

Although the biology of *M. adamsi* has been studied and reported in India and Hong Kong (Ganapathi *et al*., 1971; Kalyanasundaram, 1975; Raju *et al*., 1975; Rao *et al*., 1975; Morton, 1989), there is a big gap of knowledge of *M. adamsi* biology in Thailand and even the South East Asia that should be fulfilled. Simberloff (2003) and Sax *et al*. (2005) found that application of background information elsewhere for invasive species management could not solve the problems because the invasive populations are like the founders that genetically and ecologically differ from their native populations and also the invasive populations in other area.

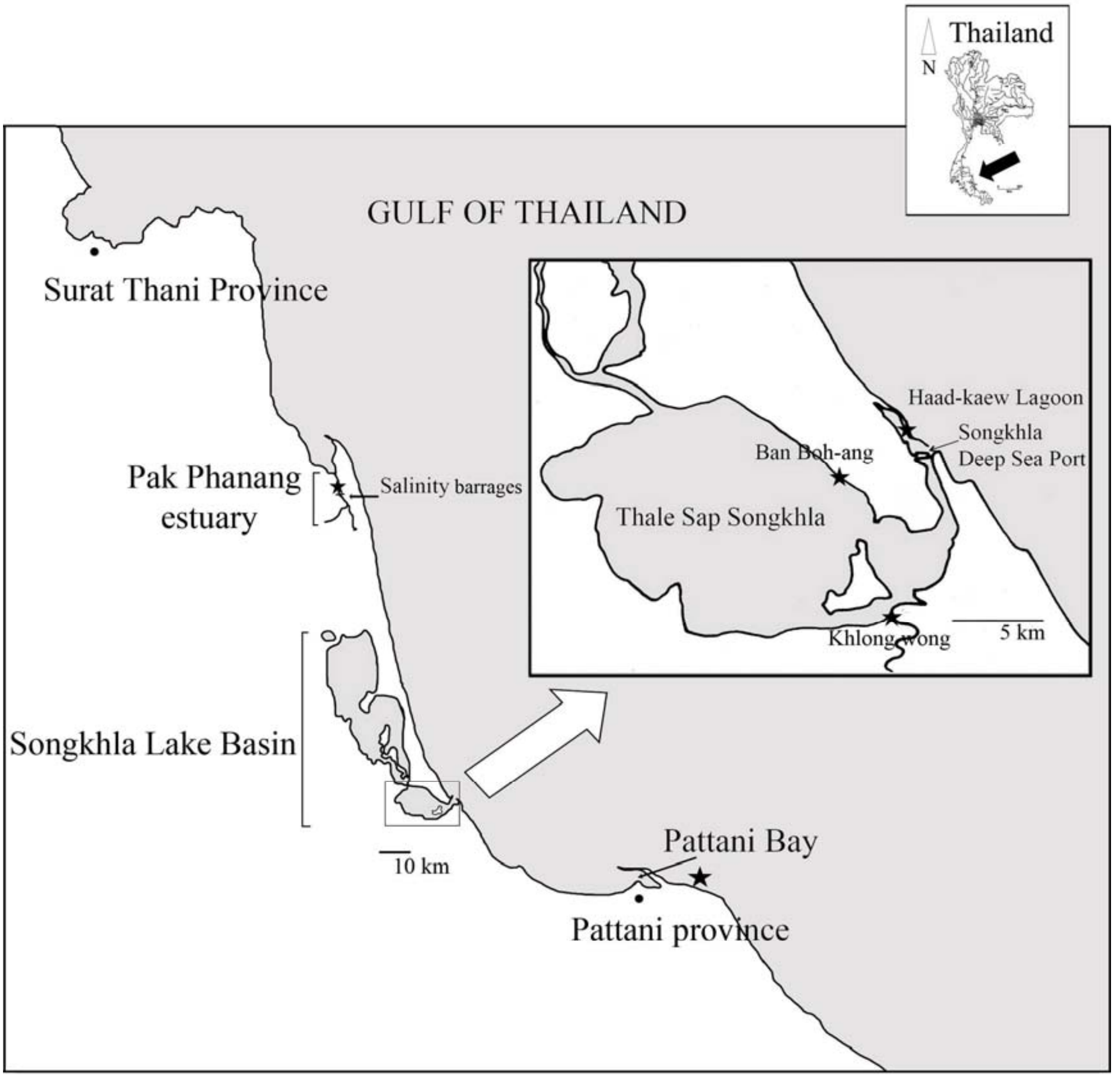


Figure 1.2 The lower Gulf of Thailand showing the observation sites of *Mytilopsis adamsi* Morrison, 1946 (indicated by ★). Inset: the Thale Sap Songkhla and Haad-kaew Lagoon.

Concepts and Aims

In order to control and manage *M. adamsi* invasion in Thailand, the understanding of its basic biology, for example, the spreading capability, adaptations of its life history to this area, is needed. This present study aims to gain the knowledge of the suitable environmental characteristics for *M. adamsi* survival and reproduction and the period of its reproductive activity in their natural habitat which guide us to understand the population regulation and dynamics of the species. The information obtained could be used to predict what the area having high risk to be invaded in the future and when the species have high potential to spread. For this basic study of the species, recruitment was used as a key population parameter representing the magnitude of reproductive activity, because it reveals the success of *M. adamsi* reproduction (Seed and Suchanek, 1992). Besides, the factors affecting the condition of adult and larvae are consequently influence on the recruitment. The recruitment usually has a positive correlation with adult density and is influenced by the population structure of the mussels (Porri *et al*., 2006a), so it can be used to indicate the population dynamic more reliable than the other parameters involving in the reproductive activity such as fecundity, number of larvae and settlement. These parameters are hardly linked to the future adult population since the mortality usually occurs (Porri, 2003). However these parameters and the process of the reproduction are essential and further studies are required.

Organisms have to face with many factors acting together in the habitat. The environmental conditions are able to alter the mussel physiological response, such as growth rate, clearance rate and reproductive output (Kalyanasundaram, 1975; Rao *et al*., 1975; His *et al*., 1989; Gosling, 2003). Recently, there was no available study on physical and biological factors that may affect the recruitment of *M. adamsi* in natural habitat. Raju *et al*. (1975), Rao *et al*. (1975), Kalyanasundaram (1975), Morton (1989) and NIMPIS (2002a) reported that temperature and salinity have an effect on *M. adamsi* reproductive activity and survival. The abundance of larvae (Martel *et al*., 1994; Gosling, 2003; McQuaid and Lindsay, 2007) and food supplies (His *et al*., 1989; Horvath and Lamberti, 1999; Gosling, 2003; Bos *et al*., 2006) were reported to be correlated with recruitment, larval growth, and mortality in some mussels, but there has been no explanation for *M. adamsi*.

Wangkulangkul and Lheknim (2008) proposed that *M. adamsi* was possibly transported to Haad-kaew Lagoon via international commercial cargo ships that anchored at the Songkhla Deep Sea Port. Haad-kaew Lagoon population was believed to be the source population for the Songkhla Lake Basin as it was the nearest infected site from the port. In general, the population characteristics of the species inhabiting in the estuary are spatially and temporarily varied due to the variation in both scale of physical and biological factors (Chícharo and Chícharo, 2001; Pombo *et al*., 2005; Chuwen *et al*., 2009). In Haad-kaew Lagoon, some physical and biological conditions dramatically vary which are possibly due to season and the variation between locations, for example, the phenomena of phytoplankton blooms were occasionally observed, and the salinity was found change rapidly after the rainstorm. The knowledge of the influence of the change in these environmental characteristics on *M. adamsi* recruitment in this area has never been investigated.

To achieve the aim of the study, the spatial and temporal design was used to study the variability in *M. adamsi* recruitment and some environmental parameters which may have influenced the recruitment in Haad-kaew Lagoon. Correlations between these certain physical and biological parameters and the recruitment were also conducted.

Research questions

1. Are there spatial and temporal variability pattern of *Mytilopsis adamsi* recruitment and some environmental parameters; and how?
2. How are likely environmental parameters affected on the recruitment of *M. adamsi* in Haad-kaew Lagoon?

Structure of the thesis

This thesis is divided into 5 chapters. Chapter 1 presents the general introduction, the background information of *Mytilopsis adamsi* and highlighting the important of the study and main research questions. Chapter 2 describes the detail of the study sites, pilot study to determine for optimal collecting panels, general experimental designs, and data collection. Chapter 3 examines the possible correlations between environmental factors and the recruitment of *M. adamsi*. Chapter 4 investigates the spatial and temporal variability in recruitment of *M. adamsi* and some environmental characteristics. Finally, the knowledge gain from chapter 3 and 4 is used to discuss for the further species control and management in chapter 5.

CHAPTER 2

Study sites, Pilot study,
General experimental designs and data collection

Study area

Haad-kaew Lagoon is a relatively small shallow coastal lagoon, oriented parallel to the coast (3 km^2, 7°14´14.35˝N, 100°33´47.04˝E, Figure 2.1), and located in Singhanakhon District, Songkhla Province in the lower south of the Gulf of Thailand. The lagoon is a part of the Songkhla Lake Basin which consists of the largest lagoonal system in Thailand and several rivers that running into it. Haad-kaew Lagoon is adjacent to the mouth of the Thale Sap Songkhla, the lowest part of the lagoonal system. It has been separated from the sea by a relatively narrow sand dune. The lagoon consists of a seasonally-closed lagoon and an open lagoon, separated by sand barrier which is the result of a strong seawater surge during the Northeast monsoon since early 2006. Sand was carried toward the shoreline and filled up some parts of the lagoon. The microhabitat in Haad-kaew Lagoon morphologically varied, as the physical condition in each sub-lagoon are heterogeneous (personal observation; Wangkulangkul and Lheknim, 2008). The seasonally-closed lagoon is affected by seawater surge and seasonal freshwater runoff. The positions which the surge and freshwater run into the lagoon are different depend on season. The open lagoon is significantly influenced by tide and by seasonal freshwater runoff, so the salinity gradually increases mouthward. The seasonality of physical conditions in Haad-kaew Lagoon possibly promotes the seasonal variability in biological factors influencing the *Mytilopsis adamsi* population characteristics or has the effect on the mussel directly.

Tide in the Gulf of Thailand is semidiurnal having two high waters and two low waters each day. During the study period (May 2007-April 2008), heights of water ranged from 0.48 m to 1.49 m in December and August 2007 respectively (Figure 2.2; Hydrographic Department, 2008-personal communication). Climatic variables were obtained from East Coast of Southern Thailand Meteorological Centre, located approximately 25 km apart from the study site via personal communication (10/08). Total amount of annual rainfall of Songkhla Province is 1,974 mm. The highest amount of rainfall is in October 2007 (351.6 mm) and the lowest amount of rainfall is in March 2008 (36.7 mm). The average of mean air temperature of the whole year is 28.3 °C. The average relative humidity of every month is 79.7% (Figure 2.2). This area is influenced by tropical monsoons. There are 3 seasons in a year according to seasonal wind direction: an intermediate rainy period influenced by strong south-west (SW) monsoon (May-September), a rainy period influenced by north-east (NE) monsoon (October-December) and a relatively dry influenced by south-east (SE) predominant wind period (January-April) (Evenson, 1983; Thai Marine Meteorological Center, 2006).

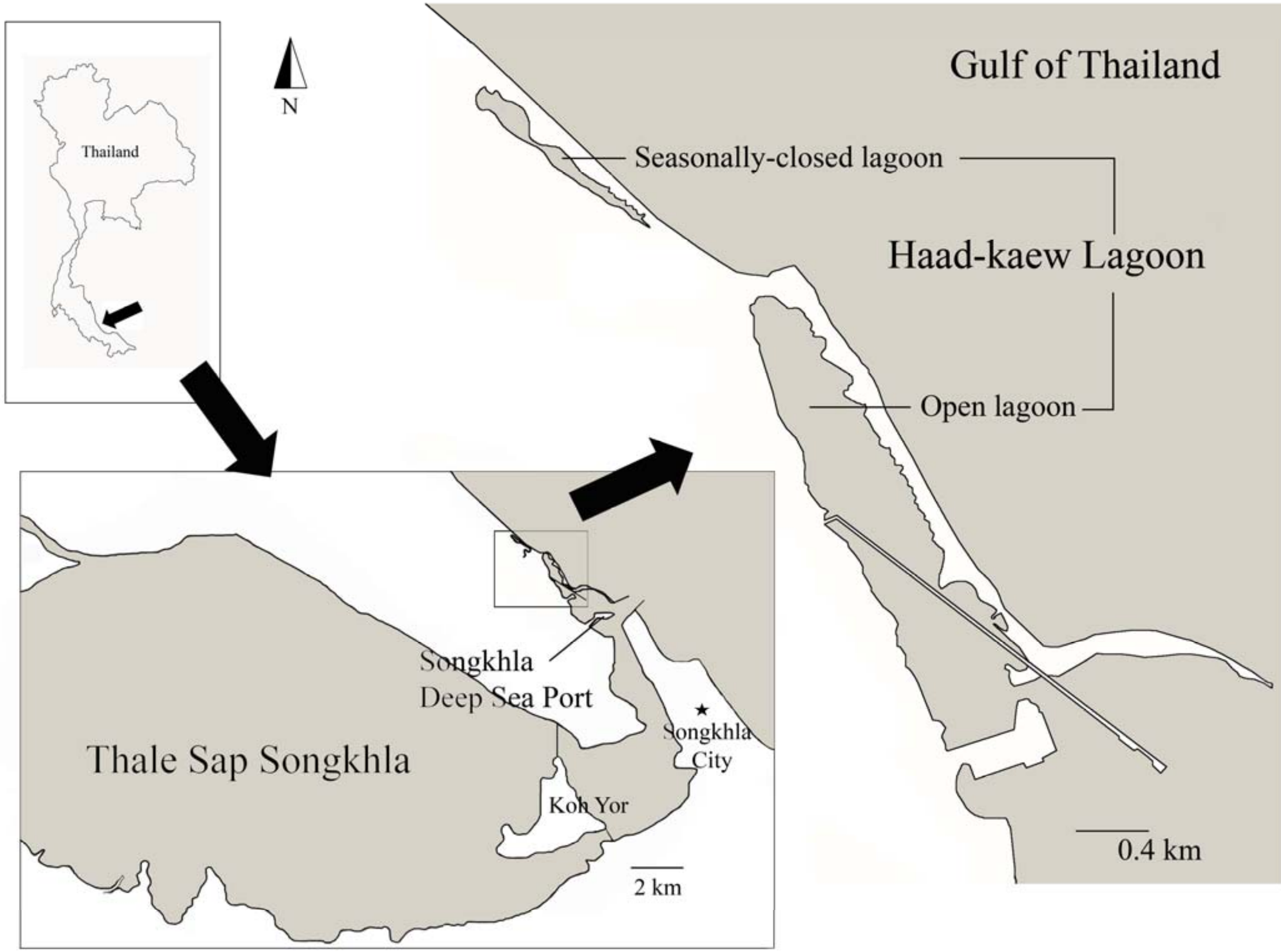


Figure 2.1 Haad-kaew Lagoon showing the seasonally-closed lagoon in the upper part and the open lagoon in the lower part. Inset: the lower part of Songkhla Lake Basin showing the Thale Sap Songkhla, Haad-kaew Lagoon, Songkhla Deep Sea Port, and Songkhla City.

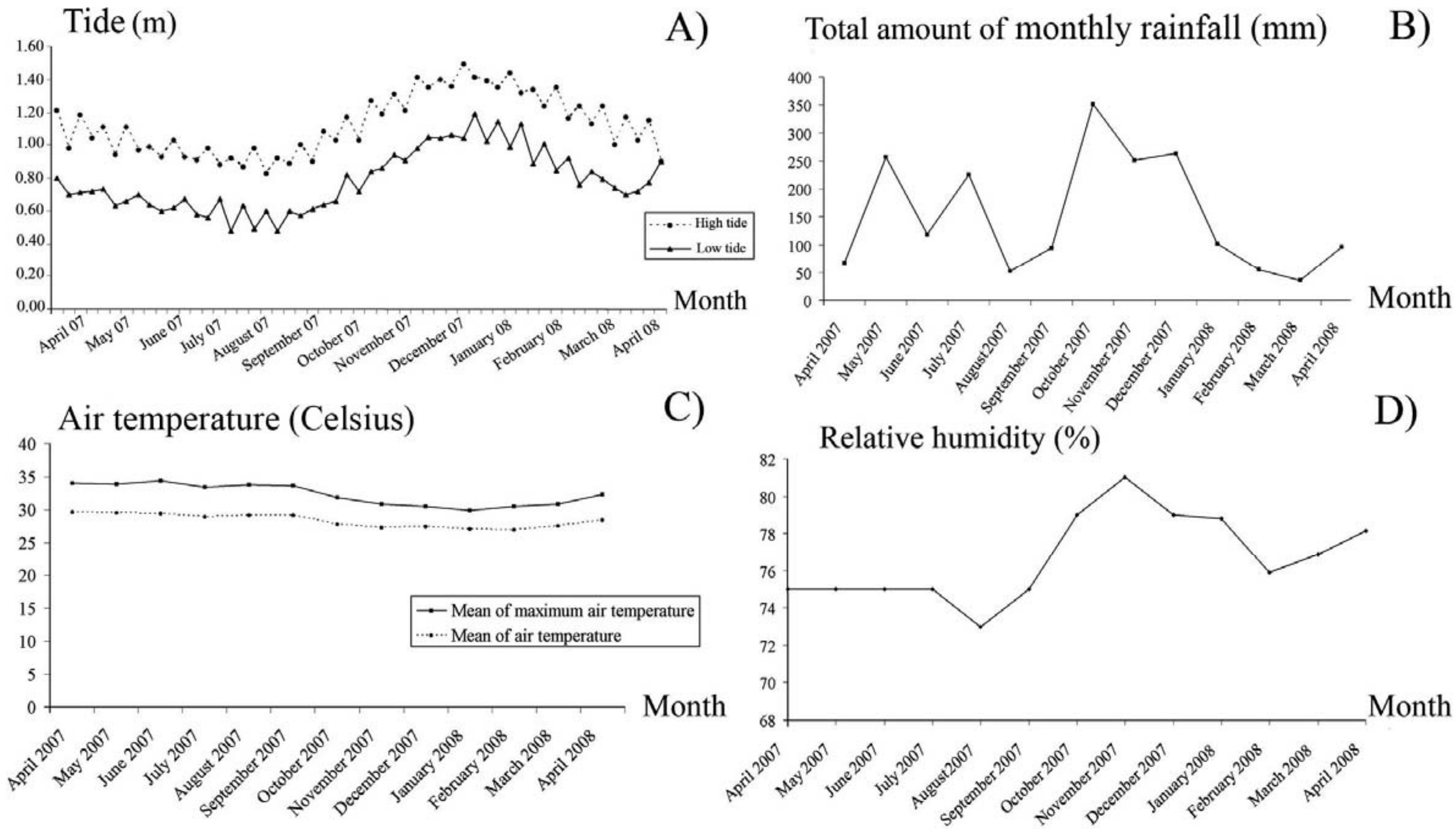


Figure 2.2 Tide (A), total amount of monthly rainfall (B), monthly air temperature (C) and relative humidity (D) in Songkhla province from April 2007 to April 2008. Each month includes 2 neap tides and 2 spring tides except in December 2007 include 3 neap tides (The type of tides-neap and spring- was determined by considering phase of moon).

Study sites

The study area included both parts of the lagoon which are physically and biologically different (personal observation).

Seasonally-closed lagoon (Figure 2.3):

During the northeast monsoon season the strong marine surge and fresh water runoff usually removes the sand from some part of the sand dune, which creates one or several small creeks in this lagoon. In that period the lagoon is recognized to be “open”. Otherwise, in the other season, sand deposition fills the creeks and the lagoon is closed. The water level and physio-chemical conditions of this part is not influenced by tide, but from seawater surge especially in the period of the northeast monsoon and seasonal freshwater runoff. The depth of lagoon ranges from 0.5 -1.5 m. It has the average width and length of 50 m and 700 m respectively. The salinity in this part more fluctuates than another and usually increases from the northwest to the southeast end. Sea bottom is muddy-sand. There are several patches of mangrove forest along the shore of the lagoon providing a lot of roots or stems for a large number of *Mytilopsis adamsi* to colonize. In addition, large colonies of *M. adamsi* are found on piers, rocks, and on the bottom by covering more than half of the whole shallow area. Some mangrove animals were also found but in relatively small number such as grapsid crabs and snails. There is a hotel located near the shore, which usually discharge sewage into the lagoon. The phenomena of planktonic bloom temporarily observed reveal the high nutrient concentration in this lagoon.

Open lagoon (Figure 2.4 and 2.5):

The condition of this part is significantly influenced by tide and by seasonal freshwater runoff. The depth ranges from 1-3 m. It has the average width and length of 200 m and 1,400 m respectively. The salinity slightly fluctuates and increases from a polyhaline (30-18 ppt) interior to the marine exterior (Wangkulangkul and Lheknim, 2008). Sea bottom is muddy-sand. A small village located near the shore and regularly discharges the domestic waste and sewage into the lagoon. At one end the lagoon opens into the mouth of the Songkhla lagoon system near to the Songkhla Deep Sea Port (Figure 2.1). From observation I found *M. adamsi* colonized only on the vertical submerged materials for instance poles, nets, or roots, which have high abundance in the interior area. The other mussels, *Perna viridis* (Linnaeus, 1758) and *Musculista senhousia* (Benson, 1842), were found colonized together with *M. adamsi* in the marine exterior.

Figure 2.3 Seasonally-closed lagoon of Haad-kaew Lagoon. The photo is taken by Suebpong Sa-nguansil on August 20, 2008.

Figure 2.4 Outer part of the open lagoon of Haad-kaew Lagoon. The photo is taken by Kringpaka Wangkulangkul on August 20, 2008.

Figure 2.5 Inner part of open lagoon of Haad-kaew Lagoon. The photo is taken by Suebpong Sa-nguansil on August 20, 2008

Pilot study: Determination for optimal collecting panels.

Introduction

Recruitment of mussels in the field can be quantified by counting number of individuals that have survived for a certain period after settlement (Molares and Fuentes, 1995; Dobretsov and Wahl, 2001; Porri *et al*., 2006b; Smith *et al*., 2009). In the experiment, the mussels were allowed to settle on natural filamentous substrata or on artificial substrates. The latter have the advantage over the biotic material in terms of constant surface area and textural composition. The artificial materials also provide the accurate number of settlers settling on the substrate in the fixed time interval (Gosling, 2003; Porri, 2003).

Bayne (1964) proposed the theory of primary and secondary settlements: two different size classes of larvae settle on the different substrata: smaller individuals primarily settle on filamentous material, and larger ones secondarily settle on adult mussels beds (Buchanan and Babcock, 1997). There are evidences supporting this theory that mussels preferentially settle on filamentous substrate (Alfaro and Jeffs, 2002; Gosling, 2003; Rorem *et al*., 2006) but Porri (2003) found different sizes of mussel on the same collectors, and Lasiak and Barnard (1995) found that type of substratum had no effect on the settlement. This theory must be proven for application to quantify *Mytilopsis adamsi* recruitment in Haad-kaew Lagoon.

The objective of this pilot study was to find the most effective *M. adamsi* collector. The type of collectors which provided the highest density of mussel attaching on would be used in the further experiment.

Materials and method

Several types of artificial panels of different substrates were employed. They were commercial scouring pads (Scotch-Brite™), Polyethylene rope frames, smooth cement tiles, plastic and rubber wood pads. In the experiment, 10x10 cm^2 of each collector was used. There were 4 stations across the lagoon, located near the bridge in the seasonally-closed lagoon. At each station all types of panels were deployed with 2 replications. Prior to trial, new pads were soaked in the brackish water for one week, to remove soluble substances that might inhibit settlement and allow biofilms to develop. The panels were tied with fishing lines and randomly hung on the plastic circle frame. The plastic frames were tied to a fishing buoy at the topside and tied to a concrete brick as anchor with nylon rope, which held the frame in mid-water depth (Figure 2.6). Settlement panels were collected and replaced at every 2-day in all 4 stations, from January 26 to February 13, 2007. After the panels were collected, they were taken to the laboratory for post-larval rearing (until the mussels were visible under stereomicroscope).

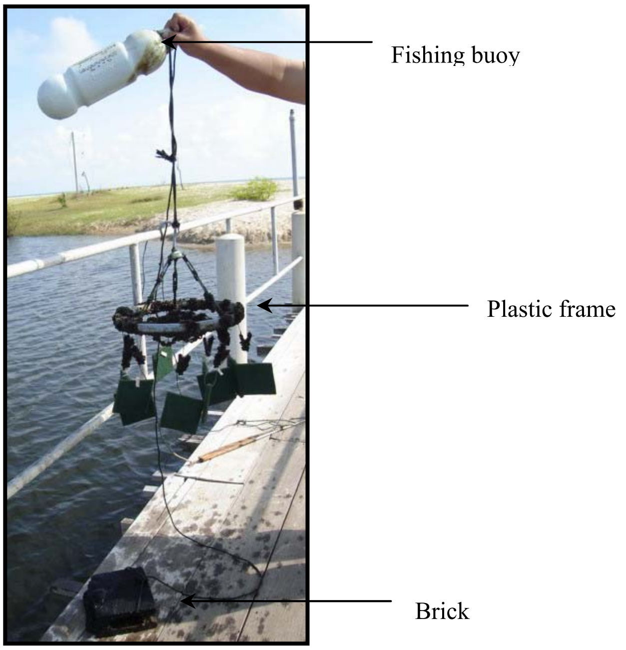


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Figure 2.6 Arrangement of settlement panels deployment

In laboratory, the panels were tied to the fishing lines that were fastened in plastic boxes, filled with filtered aerated brackish water from Haad-kaew Lagoon. Each type of panels was kept separately. In order to avoid the contamination of larvae of *M. adamsi* from Haad-kaew Lagoon, living plankton was collected from Koh Yor for larval rearing in laboratory. After 7 days, the larvae grew up to be visible clearly under stereomicroscope, and then the number of all individuals attaching on the panels were counted. The surface area of each side of each panel was divided into 4 equal quadrats, and two quadrats were randomly sampled for counting. The density of the recruitment (individual/ 100 cm^2) was obtained from both sides of the panel.

The one-way ANOVA was employed to investigate the influence of type of collectors on density of recruits by using Statistix for Window version 1.0 (Analytical Software, 1996). The obviously different size of mussel found on the scouring pads were randomly sampled and measured for shell length, closed to 0.1 mm, to prove whether they were from the different cohorts. All sampled shell lengths were used to build up a length-frequency distribution histogram and analyzed for any independent cohort, using FISAT II version 1.2.2 (FAO, 2005).

Results and discussion

There was a significant difference between recruitment densities on 5 types of collecting panel ($F_{4, 330} = 11.03$, $P < 0.05$, Figure 2.7). Tukey's test suggested that there was insignificant difference between density on scouring pads and rope frames. However mean density of mussel recruitment on scouring pads was the highest (Figure 2.7). Therefore, scouring pad was the most favorable substrate for *M. adamsi* settlement.

The length-frequency distribution histogram indicated two distinct groups of *M. adamsi* shell length (Figure 2.8), suggesting that there were at least 2 cohorts of mussel recruited on scouring pads. The smaller cohort likely referred to the larvae primarily settled on the pads in the 2-day period of immersion while larger cohorts (>4 mm) indicated reattachment. If the theory of primary and secondary settlement was true for *M. adamsi* in Haad-kaew lagoon, the large individuals would not found on the scouring pads. Besides, under the same theory, if the scouring pads were used in the next experiment (chapter 3 and 4), they would be immersed in the water for approximately one month, consequently there would be high probability that the large mussel dislodge from the scouring pads and secondarily settle on the hard substrate and the number of recruits obtained after collection would be underestimated. However, the result from this pilot study suggested that the type of collector did not influence on the age of mussel attaching on the collector. Eventually the scouring pads were used in the next experiment and the number of mussel found on the pads after 1-month immersion were assumed to be the number of mussel recruits in that period by ignoring the probability of dislodgement.

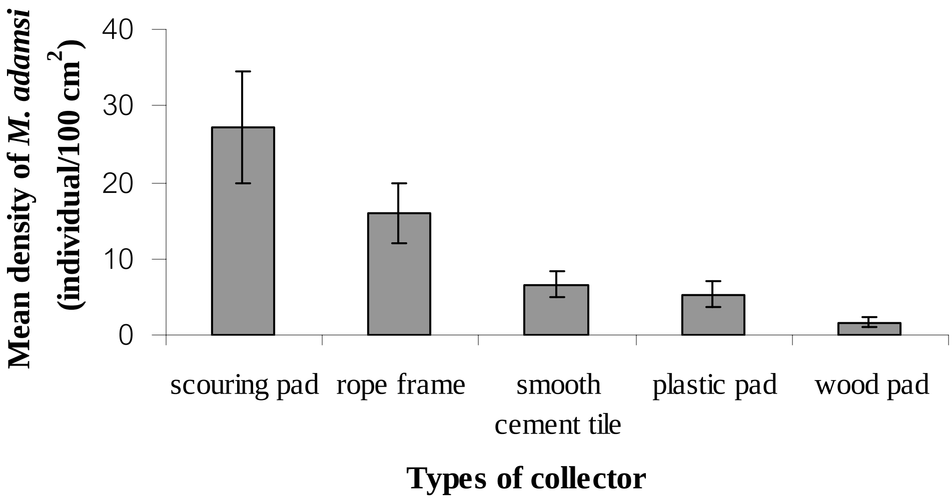


Figure 2.7 Mean density of *Mytilopsis adamsi* (individuals/100 cm^2) ± S.E. attaching on 5 types of collecting panel.

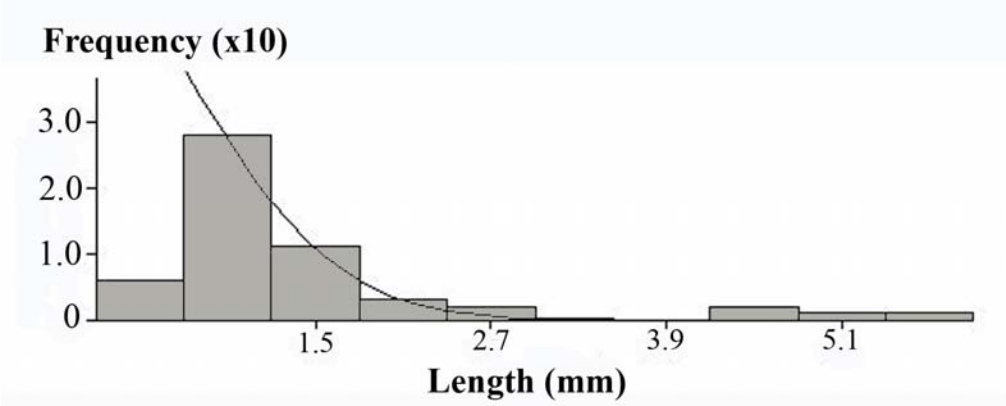


Figure 2.8 Length-frequency distribution of *Mytilopsis adamsi* (n = 53) randomly sampled from mussels recruited on scouring pads on February 27, 2007 and March 1, 2007

General experimental design

Spatial variations:

The seasonally-closed lagoon and the open lagoon are the major habitats found within the Haad-kaew Lagoon (Figure 2.1). In order to eliminate confounding factors, the sampling design incorporated four spatial levels: scouring pads, stations, zones and habitats. According to the area of the lagoon, the open lagoon is relatively larger than the seasonally-closed lagoon; thus three and two replicated zones were allocated to each habitat respectively. Within each zone, 4 stations were randomly established, 100-500 m apart. Three replicate hauls (scouring pad) were taken at each station. In this design, zones and stations within each lagoon were defined as being fixed, as the same zones and the same stations were visited on all occasions.

Temporal variation:

A hierarchical design for temporal variations of abundance of the recruits was classified into two levels, seasons and months. There are 3 seasons in this study, which were named as south-west (SW) monsoon, north-east (NE) monsoon and south-east (SE) predominant wind period according to the monsoon wind (See study area). The study was conducted between May 2007 and April 2008 with a monthly sampling. The replicate months for the season were designed according to its season, which aims to provide greater generality to the seasons and habitat results (Table 2.1).

Table 2.1 Date of collection. SW = south-west monsoon, NE = north- east monsoon and SE = south-east predominant wind period.

Season	Month	Date of collection	Season	Month	Date of collection
SW	1	May 23, 2007	NE	2	November 17, 2007
SW	2	June 21, 2007	NE	3	December 15, 2007
SW	3	July 20, 2007	SE	1	January 12, 2008
SW	4	August 20,2007	SE	2	February 12, 2008
SW	5	September 20, 2007	SE	3	March 12, 2008
NE	1	October 21, 2007	SE	4	April 14, 2008

Data collection

The study area included both seasonally-closed and open lagoons, which were divided into 2 and 3 zones respectively. In each zone, 4 stations were randomly designated. During the study, the following factors were sampled every month. At each station, 3 scouring pads (10 x 10 cm^2) were immersed in water, which allowed mussel larvae to attach. The pads were arranged in the same manner as in the pilot study. The pads were soaked in brackish water a week before, to remove soluble substances and allow biofilms to develop naturally. The pads were tied randomly to the floating plastic frame, approximately 80 cm below the surface, and were incubated for one lunar cycle (~30 days). After 30 day sampling period, the pads were transferred to the laboratory and frozen until counting was done. The density of the recruitment on each scouring pad (individuals/ 100 cm^2) was obtained from both sides using a dissecting microscope.

Salinity, water temperature, pH and dissolved oxygen were measured every 14 days during the bi-monthly peak of neap tides. A Refracto-salinometer was used to measure salinity, and a thermometer for water temperature, a hand held Hanna pH meter and a JENWAY DO Meter Model 9200 Dissolved Oxygen System for pH and dissolved oxygen. The plankton abundances were also assessed in each zone on the same day as scouring pads were collected. Four samples were taken from each zone using vertical haul plankton net (22-µm mesh, 30 cm. diameter opening). Plankton samples were preserved in 10: 90, formalin: brackish water.

Zooplankton and phytoplankton samples were identified and counted under a compound microscope using Sedgewick rafter counting cell (with 100x magnification) and haemacytometer blood counting chamber (with 400x magnification), respectively. The planktons were identified to the lowest taxonomic level possible. The planktons were taxonomically sorted out using external morphology. Phytoflagellates were defined as any small single-cell phytoplankton that could not be properly identified and which usually had one or more flagella and absence of a shell. Copepods and nauplius larvae were grouped as planktonic crustaceans. There were six groups of phytoplankton: chlorophytes, diatoms, phytoflagellates, dinoflagellates, cyanophytes (the group's name of filamentous and small colony cyanophytes), and large colony cyanophytes and 3 groups of zooplankton: rotifers, planktonic crustaceans, and D-shaped larvae. In the seasonally-closed lagoon, *Mytilopsis adamsi* dominate this lagoon, that there is just a relatively very small number of other bivalves found (personal observation). Thus the D-shaped larvae found in this lagoon were assumed to be the larvae of *M. adamsi*. In the open lagoon, the morphology of several different shape and size larvae were compared, and then the larvae that resemble to the larvae from seasonally-closed lagoon were presumed to be the larvae of *M. adamsi*. The number of D-shaped larvae which were possibly the larvae of *M. adamsi* was used in this study. The numbers of all planktons were calculated to be the density (individual/L).

CHAPTER 3

Environmental factors influencing recruitment of invasive mussel *Mytilopsis adamsi* Morrison, 1946 in Haad-kaew Lagoon, in the lower south of the Gulf of Thailand

INTRODUCTION

Over the past decades, dreissenid mussels have frequently been recognized as alien invasive species. A species found in brackish waters *Mytilopsis adamsi* Morrison, 1946, native to the eastern Pacific, has spread across the Indo-Pacific region, to countries such as Fiji, India, Malaysia, Singapore, and Australia (Ganapathi *et al*., 1971; Marelli and Gray, 1985; Willan *et al*., 2000; Tan and Morton, 2006). Most reports have referred to this species as *Mytilopsis sallei*, however Salgado-Barragán and Toledano-Granado (2006) and Marelli (2007, personal communication) state that all Asian reports of *M. sallei* are likely to be *M. adamsi*. In Thailand, there have been records of *M. adamsi* from Pattani Province, Pak Phanang estuary, Thale Sap Songkhla of the Songkhla Lake Basin and Haad-kaew Lagoon (Swennen *et al*., 2001; Wangkulangkul and Lheknim, 2008).

Preliminary observations from Haad-kaew Lagoon indicated that *M. adamsi* has formed dense clusters covering more than half of the bottom in the shallow zone. It also found in dense patches attached to pneumatophores of mangroves, floating cages and bridges. Therefore, *M. adamsi* may be in its spreading stage according to the generalized steps in the invasion process proposed by Sakai *et al*. (2001). Basic biological knowledge of this species in their new environment is required, in order to devise appropriate management interventions to control this species. Especially in view of the findings that environmental conditions are able to alter mussel physiological responses, such as growth rate, respiration rate, clearance rate, assimilation efficiency, mortality, excretion and reproductive output. (Kalyanasundaram, 1975; Raju *et al*., 1975; His *et al*., 1989; Gosling, 2003; Bos *et al*., 2006).

Seed and Suchanek (1992) defined recruitment of mussels as the process of successful colonization that reflects the settlement rate and post-settlement mortality. Currently, there is no available information on the physical and biological parameters that may influence recruitment of *M. adamsi* in their natural habitat. Raju *et al*. (1975), Rao *et al*. (1975), Kalyanasundaram (1975), Morton (1989) and NIMPIS (2002a) reported temperature and salinity had an effect on the reproductive activity and survival of *M. sallei* in India and Hong Kong. In addition, the abundance of larvae (Martel *et al*., 1994; Gosling, 2003; McQuaid and Lindsay, 2007) and food supply (His *et al*., 1989; Horvath and Lamberti, 1999; Gosling, 2003; Bos *et al*., 2006) were usually correlated with recruitment, larval growth and mortality in mussels.

The present study aims to provide background knowledge on the biology of *M. adamsi* in Thailand, by exploring potential physical and biological characteristics that are likely to control the abundance of *M. adamsi*. To achieve this, I used recruitment as a key population parameter, since it not only reveals the reproductive success, but can also be a tool to understand the population dynamics of the species. The density of *M. adamsi* recruits and environmental parameters such as salinity, water temperature, pH, dissolved oxygen, density of phytoplankton, zooplankton, and D-shaped larvae in Haad-kaew Lagoon were measured. The potential correlations between these parameters and the density of *M. adamsi* recruits were investigated.

METHODS

To achieve the aims of the study, multi-stage sampling was used to collect the data. The data used to examine the correlation between recruitment of *Mytilopsis adamsi* and environmental characteristics were obtained from the method given in chapter 2. So, only the details of the statistical analysis were described here.

Data Analysis

To understand the relationship between the density of *M. adamsi* recruits and environmental factors, Principal Component Analysis (PCA), Linear Discriminant Analysis, Pearson correlations and multivariate regressions analysis were used. The environmental factors consist of the measured physical parameters and biological parameters. The physical parameters were salinity, water temperature, pH, and dissolved oxygen. The biological parameters were density of *M. adamsi* recruits, chlorophytes, diatoms, phytoflagellates, dinoflagellates, large colony cyanophytes, rotifers, D-shaped larvae and planktonic crustaceans.

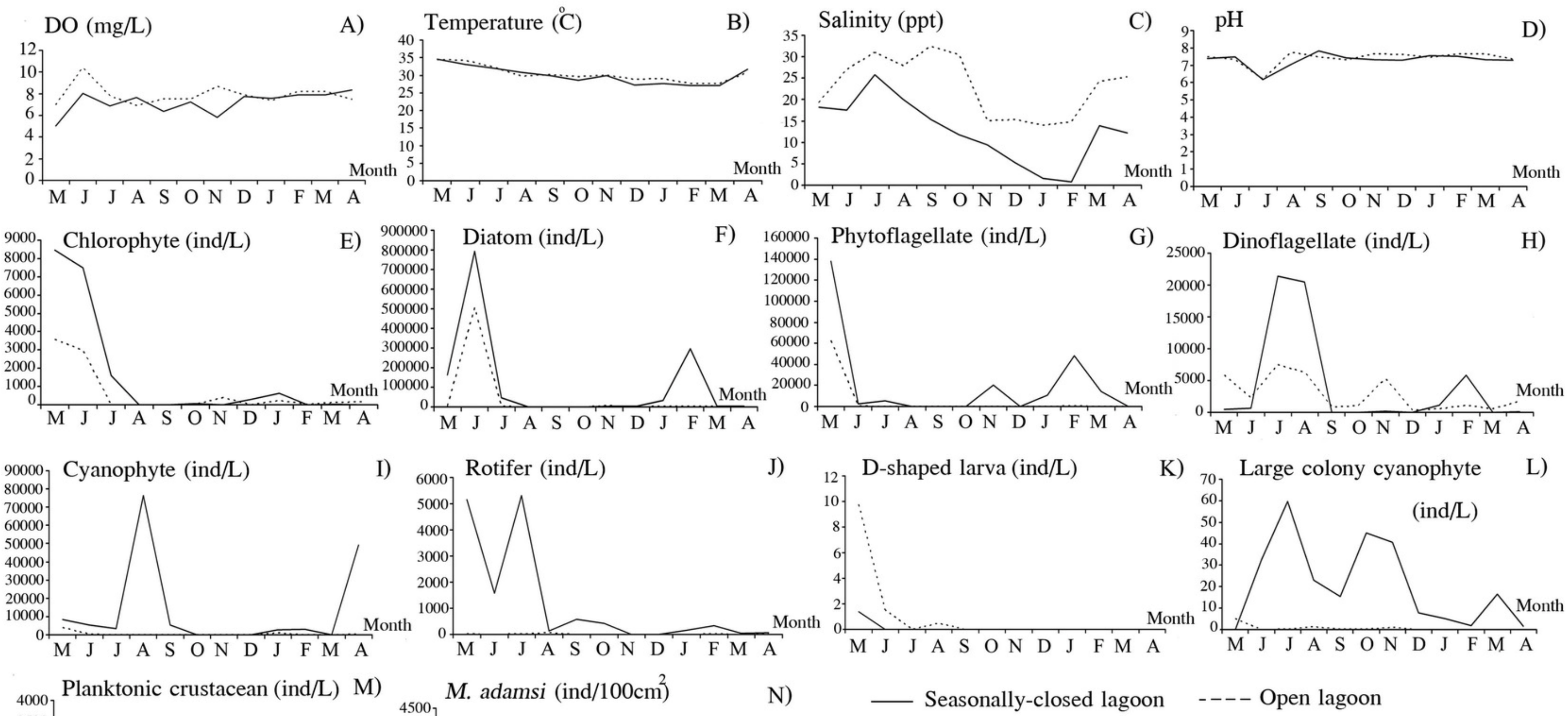
Prior to the analyses, data of the organisms density were log (x+1) transformed. Initially PCA, on covariances, was performed to picture the structure of the data. Using Linear Discriminant Analysis I verified the relationship between all biological and physiological variables using the season as an independent category (from the PCA results seasonal variations to be an important determinant of *M. adamsi* recruitment and other variables was found). The results of Discriminant analysis are presented as canonical point plots and multivariate means (labeled by circle and biplot rays). I used stepwise multivariate regression analysis to ascertain if the physical and biological parameters could predict the density of *M. adamsi* recruits. Since the results from the Linear Discriminant Analysis confirmed seasonal trends (from the PCA results seasonal variations were the important factor), the multivariate regression that was later performed was analyzed separately on a seasonal basis. Using a t-test, I then investigated whether there was significant variance contributed by each factor to the multivariate regression equation within the season. The factor, D-shaped larvae, was removed from the regression analysis in the NE monsoon and SE predominant wind period, while the temperature was removed from the NE monsoon, because they were too highly correlated with other factors. The Pearson correlation coefficients between the density of *M. adamsi* recruits and selected factors from the multivariate regression analysis were calculated (See Table 3.4). Statistical analyses were performed with the JMP 8 -trial package (SAS, 2008) and Statistix for Window version 1.0 (Analytical Software, 1996). $P<0.05$ was accepted as the level of significance.

RESULTS

Physical and biological parameters and *Mytilopsis adamsi* recruitment

The magnitude of all parameters fluctuated over the study period, except for water temperature and pH (Figures 3.1A-N), which were higher in the SW monsoon season, from May to September 2007. Water temperature and pH ranged from 18.7 to 36.7 C, and 6.1 to 7.9 respectively. Dissolved oxygen varied between 2.9 and 13.5 mg/L. Salinity varied from fresh water to saline water ranging from 0 ppt to 33.5 ppt. All biological parameters had their lowest density of 0 individual/L. The highest densities of the organisms were present in the following ascending order: diatoms (4,160,663 individuals/L), cyanophytes (466,430 individuals/L), phytoflagellates (442,266 individuals/L), dinoflagellates (113,176 individuals/L), chlorophytes (64,927 individuals/L), rotifers (12,921 individuals/L), planktonic crustaceans (7,026 individuals/L), large colony cyanophytes (485 individuals/L), and D-shaped larvae (69 individuals/L). Diatoms and cyanophytes were the dominant phytoplankton in this study. The density of *Mytilopsis adamsi* recruits ranged from 0 to 5,690 individuals per 100 cm^2. The highest density was found during the SE predominant wind period in January 2008.

The dominant genus of diatoms was *Chaetoceros*, which potentially contributed to the peak of diatoms density in the SW monsoon season (Figure 3.1F). The major components of phytoflagellates were either cryptophytes or crysophytes. The dominant cyanophytes were *Oscillatoria* spp. *Gymnodinium* spp. was the main component of the dinoflagellates. The highest abundance among the chlorophytes was *Closterium* spp. The large colony cyanophytes was mainly *Microcystis* sp. *Brachionus* spp. was the major constituent of the rotifers. Among the planktonic crustaceans, calanoid and cyclopoid copepods, and nauplius larvae were the main components.



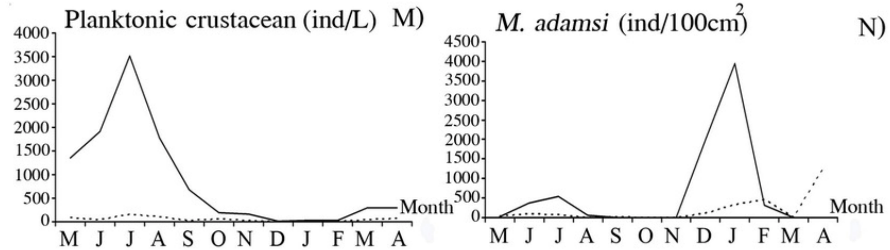


Figure 3.1 Monthly variations of means of physical variables and densities of biological variables collecting from Haad-kaew Lagoon during May 2007 to April 2008. SW monsoon season was from May-September 2007; NE monsoon season was from October-December 2007; SE predominant wind period was from January-April 2008

Relationship between *Mytilopsis adamsi* recruits and physical and biological factors

Principal Component Analysis

Principal Component Analysis yielded 14 principle components. The first two principal components accounted for 85.45% of the variation in the sample and had eigenvalues = 77.88 and 9.05 respectively (Table 3.1). The first principal component was mainly related to salinity which had a relatively high positive score on this axis (0.98). Water temperatures, density of phytoflagellates, cyanophytes and planktonic crustaceans had high positive scores and were related to the second principal component (0.59, 0.46, 0.35, and 0.35 respectively).

The score plot of the first and second principal components showed a seasonal trend according to salinity (Figure 3.2). The PC 1 highlighted a gradient of salinity, extremely high on the right end and low on the left. The pattern depicted in Figure 3.2 indicates that parameters during the SW monsoon were relatively different from the other seasons whilst data from the NE monsoon and the SE predominant wind period were not clearly isolated. Habitats ambiguously influenced the parameters as the data from each lagoon overlapped. During the SW and NE monsoons, the data from the seasonally-closed lagoon and the open lagoon intersected; otherwise during the SE predominant wind period, the data from both lagoons were reasonably separated. This indicates that the season seem to be a moderator that has an effect on the relationship between *M. adamsi* recruits and selected physical and biological parameters.

Table 3.1 Eigenvectors, eigenvalues and explained variation (%) on the Principal Component Axes of physical and biological variables collected from Haad-kaew Lagoon during May 2007 to April 2008

	Component 1	Component 2
Eigenvectors		
Dissolved oxygen (DO)	0.01176	-0.06026
Water temperature (Tmp)	0.12234	0.59810
Salinity (Sal)	0.98469	-0.03653
pH	-0.01589	-0.06973
Mytilopsis adamsi recruits (ADS)	-0.03726	0.03307
Chlorophytes (CHL)	-0.01060	0.30839
Diatoms (DIA)	-0.03580	0.13756
Phytoflagellates(PHF)	-0.05325	0.46055
Large colony cyanophytes (CYC)	-0.00616	0.02164
Dinoflagellates (DNF)	0.05932	0.13646
Cyanophytes (CYA)	-0.05757	0.34614
Rotifers (ROT)	-0.02841	0.21354
D-shaped larvae (DLA)	0.00176	0.03997
Planktonic crustaceans (PNC)	0.04148	0.34515
Eigenvalues	77.8841	9.0489
Explained variation (%)	76.56	8.90

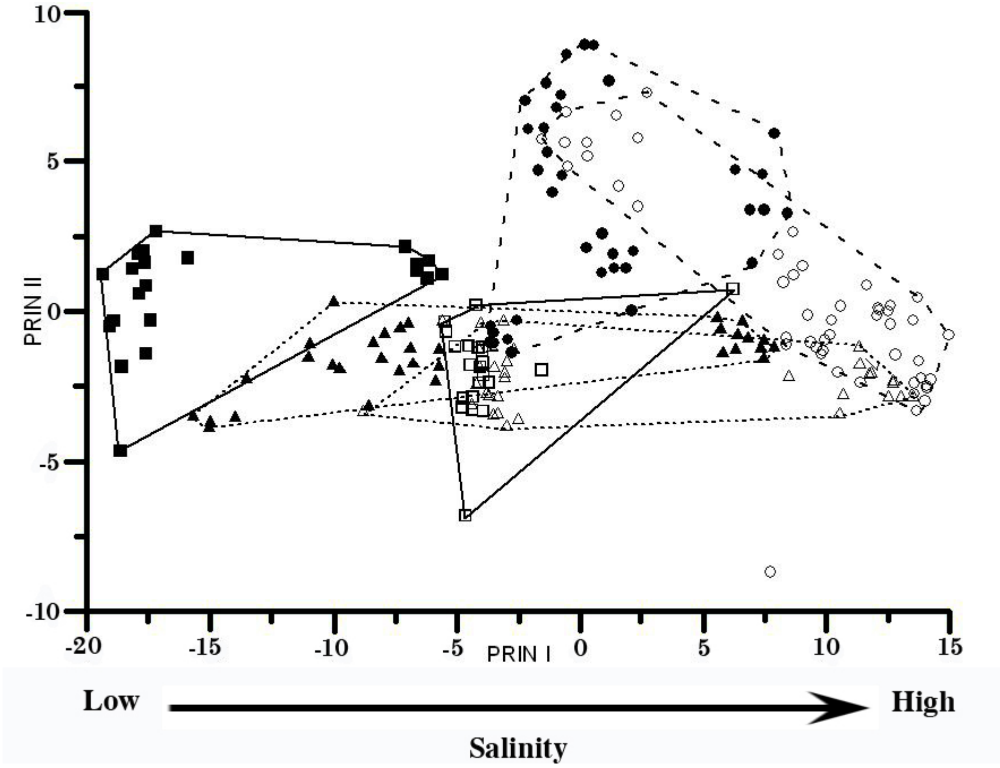


Figure 3.2 Score plot from the Principal Component Analysis of physical and biological variables collecting from Haad-kaew Lagoon during May 2007 to April 2008. Circle marker = SW monsoon; triangle = NE monsoon; square = SE predominant wind period. Clear form = open lagoon; solid form = seasonally-closed lagoon. One outlier from the SW monsoon was ignored. Percentage variances explain for PRIN I and II are 76.56 and 8.90 respectively.

Canonical Discriminant Analysis

Linear Discriminant Analysis showed that only the first canonical axis had eigenvalues >1 (2.10), while the second canonical axis had eigenvalue of 0.77 (Table 3.2). The first axis was mainly related to the density of the D-shaped larvae which had the highest positive score (1.10), while the second and third variables were the density of rotifers (0.49) and the large colony cyanophytes (-0.48). The second axis was also related to the density of the D-shaped larvae and this had a high negative score (-1.30), while the second and third variables were the density of the large colony cyanophytes (-0.58) and the *M. adamsi* (0.55) recruits. The canonical plot (Figure 3.3) of the variable score on the first and second canonical axes indicated that salinity had a larger effect on the organisms when compared to other physical factors. The abundance of dinoflagellates, phytoflagellates, chlorophytes, rotifers, D-shaped larvae and planktonic crustaceans were high during the SW monsoon. The density of the *M. adamsi* recruits was higher during the SE predominant wind period, at low salinity and water temperature. Figure 3.3 also indicated that *M. adamsi* recruits were likely to coexist with cyanophytes, dinoflagellates, and phytoflagellates, but not with the density of the D-shaped larvae. The D-shaped larvae were closely related to chlorophytes and planktonic crustaceans. Phytoflagellates, dinoflagellates and pH were positively related. Large colony cyanophytes and diatoms were distinct from the others, which were high during the NE monsoon period.

Table 3.2 Eigenvectors, eigenvalues and explained variation (%) on the canonical axes of the physical and biological variables collected from Haad-kaew Lagoon during May 2007 to April 2008.

	Canonical 1	Canonical 2
Eigenvectors		
Dissolved oxygen (DO)	-0.06493	-0.054319
Water temperature (Tmp)	0.1288099	0.0379122
Salinity (Sal)	0.0865815	0.0218425
pH	0.0454258	0.1335248
Mytilopsis adamsi recruitment (ADS)	-0.070274	0.5540955
Chlorophytes (CHL)	0.2482496	-0.330281
Diatoms (DIA)	-0.364941	-0.132181
Phytoflagellates (PHF)	0.0644679	0.2152286
Cyanophytes (large colony; CYC)	-0.479511	-0.582591
Dinoflagellates (DNF)	0.1068548	0.0870736
Cyanophytes (CYA)	-0.076819	0.5412032
Rotifers (ROT)	0.4927011	0.135146
D-shaped larvae (DLA)	1.0967424	-1.298728
Planktonic crustaceans (PNC)	0.278251	-0.129675
Eigenvalues	2.09781076	0.76913171
Explained variation (%)	73.1724	26.8276

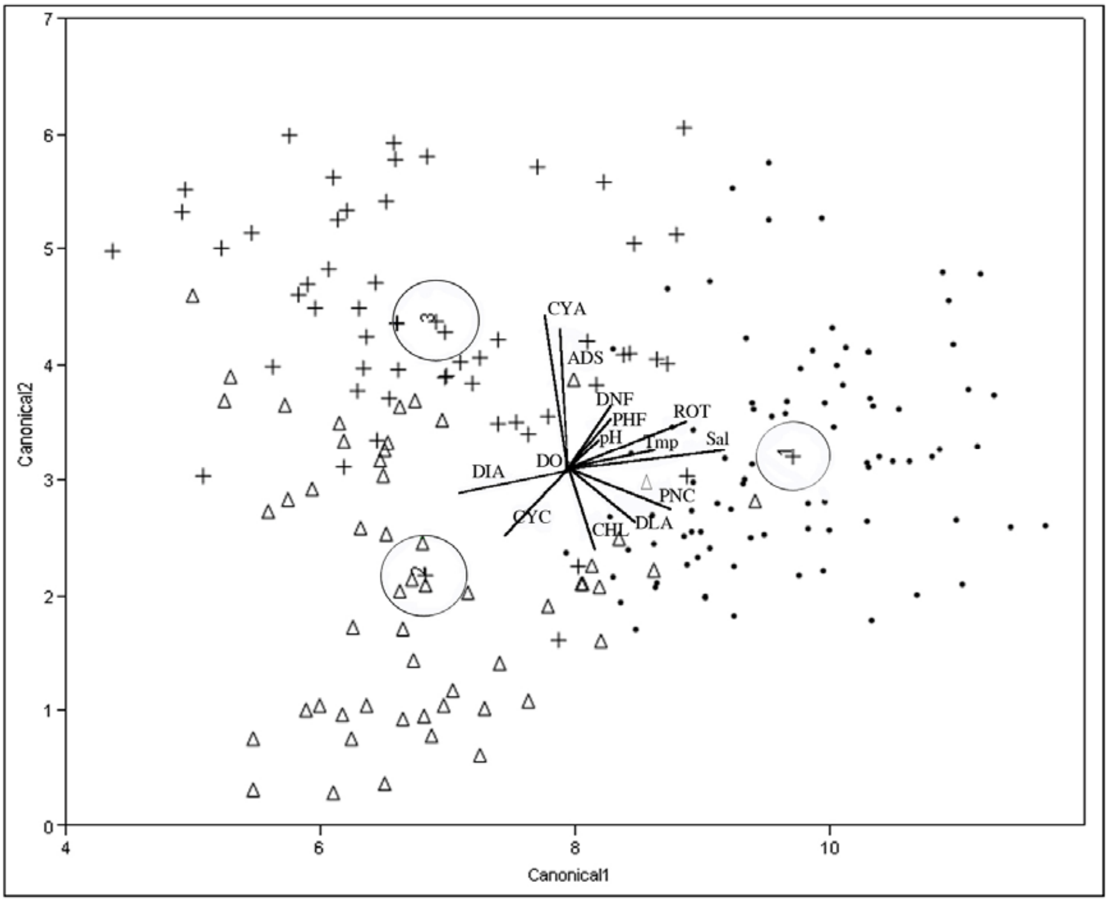


Figure 3.3 Canonical plot of a Linear Discriminant analysis of the physical and biological variables by season collected from Haad-kaew Lagoon during May 2007 to April 2008. The names of the variables have been abbreviated in the plot: DO = Dissolved oxygen, Tmp = Water temperature, Sal = Salinity, ADS = *Mytilopsis adamsi* recruits, CHL = Chlorophytes, DIA = Diatoms, PHF = Phytoflagellates, CYC = large colony Cyanophytes, DNF = Dinoflagellates, CYA = Cyanophytes, ROT = Rotifers, DLA = D-shaped larvae, PNC = Planktonic crustaceans. Numbers in the circles and signs refer to seasons; 1 and dot = SW monsoon (May-September 2007), 2 and triangle = NE monsoon (October-December 2007), 3 and plus = SE predominant wind period (January-April 2008).

Regression and Correlation Analysis

The biological and physical parameters influence on *Mytilopsis adamsi* recruitment varied from season to season (Table 3.3). During the SW monsoon season, the density of *M. adamsi* recruits was strongly correlated with DO, pH, phytoflagellates, dinoflagellates, and planktonic crustaceans (r^2 = 0.4917, n = 90, $p <$ 0.0001, Table 3.3). During the NE monsoon season however, pH, salinity, planktonic crustaceans, large colony cyanophytes and rotifers were strong predictors of the *M. adamsi* recruitment density (r^2 = 0.6864, n = 56, $p <$ 0.0001, Table 3.3). During the SE predominant wind period, the *M. adamsi* recruitment density was correlated with temperature, salinity, planktonic crustaceans, large colony cyanophytes, diatoms and rotifers (r^2 = 0.5667, n = 53, p < 0.0001, Table 3.3).

Further analysis using Pearson correlation (Table 3.4), indicated that in the SW monsoon season, dissolved oxygen, phytoflagellates, dinoflagellates, and planktonic crustaceans were positively correlated to the density of *M. adamsi* recruits but not with pH. Dinoflagellates had the highest coefficient (0.3812). During the NE monsoon season, salinity, pH and rotifers showed insignificant correlations, while the large colony cyanophytes and planktonic crustaceans showed significant negative correlation. Planktonic crustaceans had the highest negative coefficient (-0.5971) during this season. During the SE predominant wind period, water temperature, salinity, large colony cyanophytes and planktonic crustaceans had negative values while diatoms and rotifers had positive values. Interestingly, density of planktonic crustaceans was positively correlated to mussel recruitment in the SW monsoon season but negatively correlated in the NE monsoon season and the SE predominant wind period.

Table 3.3 Multivariate regression between the density of *Mytilopsis adamsi* recruits and environmental factors collected from Haad-kaew Lagoon in 3 seasons during May 2007 to April 2008. The SW monsoon season was from May-September 2007. The NE monsoon season was from October-December 2007. The SE predominant wind period was from January-April 2008. An asterisk indicates significant correlations (P<0.05).

Predictor variables	Coefficients	SD	Student's t	P
SW monsoon season				
Constant	-0.4498	0.54909	-0.82	0.4150
DO	0.1815	0.04300	4.22	0.0001*
pH	-0.1416	0.04825	-2.93	0.0043*
Dinoflagellates	0.1424	0.03854	3.69	0.0004*
Phytoflagellates	0.1135	0.03140	3.62	0.0005*
Planktonic crustaceans	0.2061	0.04105	5.02	<0.0001*
R-squared	0.4917			
NE monsoon season				
Constant	27.2002	4.68102	5.81	<0.0001*
pH	-3.3169	0.61692	-5.38	<0.0001*
Salinity	-0.0423	0.01498	-2.82	0.0068*
Large colony cyanophytes	-0.9516	0.2551	-3.73	0.0005*
Rotifers	0.5055	0.11451	4.41	0.0001*
Planktonic crustaceans	-0.3759	0.12723	-2.95	0.0047*
R-Squared	0.6864			
SE predominant wind period				
Constant	7.9561	2.25440	3.53	<0.0001*
Temperature	-0.2383	0.08634	-2.76	0.0082*
Salinity	0.0794	0.02504	3.17	0.0027*
Diatoms	-1.29E-06	5.89E-07	-2.19	0.0339*
Large colony cyanophytes	-2.0216	0.94195	-2.15	0.0371*
Rotifers	1.0786	0.24111	4.47	<0.0001*
Planktonic crustaceans	-0.5126	0.13381	-3.83	0.0004*
R-squared	0.5667			

Source	DF	SS	MS	F	P
SW monsoon season					
Regression	5	29.5013	5.90025	16.44	<0.0001*
Residual	85	30.5015	0.35884		
Total	90	60.0027			
NE monsoon season					
Regression	5	49.7767	9.95535	22.33	<0.0001*
Residual	51	22.7417	0.44592		
Total	56	72.5184			
SE predominant wind period					
Regression	6	38.199	6.36651	10.24	<0.0001*
Residual	47	29.2126	0.62154		
Total	53	67.4116			

Table 3.4 Pearson correlation coefficients between the density of *Mytilopsis adamsi* recruits and environmental factors collected from Haad-kaew Lagoon in 3 seasons during May 2007 to April 2008. The SW monsoon season was from May-September 2007. The NE monsoon season was from October-December 2007. The SE predominant wind period was from January-April 2008. An asterisk indicates significant correlations (P<0.05). Different superscripts show a significant different simple linear regression between seasons.

Environmental factors	Pearson correlation coefficients
SW monsoon season	
Dissolved oxygen	0.2240*
pH	-0.2735*^{a}
Phytoflagellates	0.3020*
Dinoflagellates	0.3812*
Planktonic crustaceans	0.3776*^{a}
NE monsoon season	
Salinity	-0.2458^{a}
pH	-0.0747^{b}
Large colony cyanophytes	-0.4694*^{a}
Rotifers	0.1304 ^{a}
Planktonic crustaceans	-0.5971*^{b}
SE predominant wind period	
Water temperature	-0.5656*
Salinity	-0.3492*^{b}
Diatoms	0.3680*
Large colony cyanophytes	-0.2638*^{b}
Rotifers	0.1851^{b}
Planktonic crustaceans	-0.5325*^{b}

DISCUSSION

The answer to the question "What are the potential physical and biological parameters that are likely to control the recruitment of *Mytilopsis adamsi* abundance in Haad-kaew Lagoon?" is relatively complex. The following section deals with the contribution of the physical and biological parameters to the variations in *M. adamsi* recruitment. It is of interest, that the variability in the *M. adamsi* recruitment associated with the physical and biological parameters in the present study is likely to be seasonal (Figure 3.2 and 3.3).

In the present study, there were two unequally strong recruitment pulses of *M. adamsi*, a minor pulse between May and August 2007 in the SW monsoon season, and a major pulse from November 2007 to March 2008 in the time between the NE monsoon and the SE predominant wind period pulses. Morton (1989) reported the congruent pattern of *M. sallei* recruitments in Hong Kong that also has two recruitment periods per year, at an intermediate time between spring and summer and between autumn and winter. Tuaycharden *et al*. (1988) reported that *Perna viridis* in upper Gulf of Thailand had two spawning seasons from May to August and from November to February, which almost overlapped recruitment seasons of *M. adamsi* in this study. The marked seasonality in the total abundance of *M. adamsi* recruits in the present study also closely parallels the trends of spawning in the fishes *Stolephorus spp*. suggested by Tiews *et al*. (1975), that exhibited a peak during the northeast monsoon season (October to March) and a period of little or no spawning activity from April to July. Two pulses of reproductive activity have also been found in many tropical juvenile fishes (Pauly and Navaluna, 1983) and the sunset elongate clam *Gari elongata* in the Philippines (del Norte-Campos, 2004). Pauly and Navaluna (1983) analysed the pattern of juvenile fishes recruited to coastal zones of the Philippines, and found that recruitment showed two asymmetric pulses. They also concluded that there was a very close relationship between spawning/recruitment patterns and the monsoon winds. In the present study, I found that the density of *M. adamsi* recruits partly matched with changes in salinity and diatoms, phytoflagellates and dinoflagellates abundances at certain times of the year (Figures 3.1C, 3.1F, 3.1G, and 3.1H). This indicates that seasonal cycles in the recruitment of *M. adamsi* in Haad-kaew Lagoon are the result of physical and biological factors that are likely to affect the settlement and mortality of the recruits.

Dissolved oxygen is shown to be positively correlated with *M. adamsi* recruitment in the SW monsoon season (Table 3.4). There was no explanation for the relationship between DO and *M. adamsi* recruitment, however, NIMPIS (2002a) reported that *M. adamsi* occurred in DO conditions ranging from 0 - 7.76 mg/L, indicating that *M. adamsi* can tolerate a low DO. In this study most DO values are relatively stable and fluctuate around 7 mg/L, which is a usual circumstance (Figure 3.1).

In contrast with DO, salinity in Haad-kaew Lagoon varied from freshwater to saline water i.e. from nil to 33.5 ppt. In the seasonally-closed lagoon, it gradually declined from the mid SW monsoon period towards nil salinity at the mid SE predominant wind period while in the open lagoon it dropped from its normal range. In this time *M. adamsi* recruitment significantly increased simultaneously (Figure 3.1N). Raju *et al*. (1975) showed that development of fertilized eggs of *M. sallei* was observed in salinity that ranged from freshwater to below 25 ppt In addition, Kalyanasundaram (1975) and NIMPIS (2002a) mentioned that spawning of *M. adamsi* appears to be triggered by a change in salinity. The high *M. adamsi* recruitment in the NE monsoon season and the SE predominant wind period period may be driven by low salinity and the sharp increase of recruitment was induced by the decrease of salinity. This phenomenon was also observed in other brackish dreissenid, *Mytilopsis leucophaeata* in Europe (Verween, 2006), and *Mytilopsis sallei* in the Colombian Caribbean (Puyana, 1995). Panichsook *et al*. (1985) studied the biology of *Perna viridis*, a mussel that have the same niches with *M. adamsi*, in Nakhorn Si Thammarat and reported that the ripe egg were mostly found in the relatively low salinity. The mussels may be stressed in the low salinity, thus they spawn to produce their offspring before they face with the vital condition. On other sides, the larvae may better survive in the low salinity.

Water temperature was negatively correlated with *M. adamsi* recruits for the SE predominant wind period, while a negative correlation with pH was found in the SW monsoon seasons (Table 3.4). The decrease of temperature may favor spawning of *M. adamsi* at the end of the SW monsoon approaching the NE monsoon season when the water temperature dropped below 32 °C, as found by Raju *et al*. (1975). Further, Kalyanasundaram (1975) reported some evidence regarding temperature influence spawning in *M. adamsi*; that there was no spawning at 32-40 °C over 48 h in the laboratory.

The average pH in this study was fairly constant (Figure 3.1D) and appeared to be suitable for *M. adamsi* as compared to the study in Hong Kong (Morton, 1989). In this study there was no strong evidence to indicate the influence of pH on recruitment.

Bivalves are generally considered to be keystone herbivores in many estuarine ecosystems. However, bivalves can also feed upon zooplankton (Prins and Escaravage, 2005). In this study, rotifers did not display any correlation with *M. adamsi* recruitment while planktonic crustaceans showed positive correlation only in the SW monsoon. In the NE monsoon and the SE predominant wind period planktonic crustaceans exhibited a negative correlation with *M. adamsi*. There was no information on rotifers and planktonic crustaceans in the diet of *M. adamsi*, while rotifers were reported to be generally more vulnerable to predation by zebra mussels *D. polymorpha*, the fresh water dreissenid, than planktonic crustaceans due to their smaller size and lower motility (MacIsaac, *et al*., 1991; MacIsaac, *et al*., 1995). Mesocosm experiments conducted by Prins and Escaravage (2005) reported that copepods were strongly reduced in the presence of mussels, *Mytilus edulis*. *M. adamsi* is presumed to have a similar niche with *D. polymorpha*, according to their evolution, and rotifers and planktonic crustaceans are therefore probably considered to be an important food source that influences *M. adamsi* recruitment. The high abundance of *M. adamsi* recruitment co-exists with the low abundance of planktonic crustaceans during the NE monsoon and the SE predominant wind period possibly is the result of the competition for food between larvae and this zooplankton or mussel feed abundantly on this zooplankton.

M. adamsi recruits showed a positive correlation with dinoflagellates and phytoflagellates during the SW monsoon while showing a negative correlation with the large colony cyanophytes during NE monsoon and the SE predominant wind period. It is interesting to note that *M. adamsi* recruitment overlapped with the peak in diatoms, phytoflagellates, and dinoflagellates abundances (Figures 3.1F, 3.1G, 3.1H, and 3.1N). Phytoplanktons were generally considered to be the principal food for both the adult and larva of bivalves. Naddafi *et al*. (2007) demonstrated selective feeding in freshwater zebra mussel *Dreissena polymorpha* and found that they preferred to feed on cryptophytes, while also selecting some diatoms, chrysophyte and dinoflagellates, according to their size and reserves of fatty acids that are known to have an effect on the growth, mortality, egg quality and recruitment success of bivalves (Vanderploeg *et al*., 1996; Wacker and von Elert, 2004). Trottet *et al*. (2008) reported that dinoflagellates and diatoms were the highest plankton retained by *Mytilus edulis* L. in the mesocosm experiment in marine lagoon of Magdalen Islands, Canada. This suggests that the spawning events of *M. adamsi* are synchronized with phytoplankton bloom, which probably provides food that will increase the survival of larvae settling and larval growth that consequently reduces the risk of predation indirectly (see Ram *et al*., 1996; Tyler-Walters, 2008). However the physiological mechanisms underlying this phenomenon in this species have not been investigated.

Planktonic D-shaped larvae in the present study did not contribute to predicting the density of *M. adamsi* recruits and did not show a positive correlation with recruitment during any season. The abundance of the larvae was highest during the SW monsoon season when there was a minor peak in recruitment. During the NE monsoon season and the SE predominant wind period no larvae were found, despite the fact that there was considerable recruitment in both seasons. A relationship between recruitment and their planktonic larvae has been exhibited in many studies (Bayne, 1964; Wallace, 1985; Buchanan and Babcock, 1997; Hughes *et al*., 2000). McQuaid and Lawrie (2005) reviewed many studies and proposed that this feature is not universal for mussel populations. The cause of this inconsistency may be due to the larval dispersion, mortality, or to a prolongation of the larval period. Dispersion is not likely to play a key role, as Haad-kaew Lagoon is a semi-enclosed habitat. The mortality may be caused by predation or larvae confronting a lethal environment. There is still a lack of information to clarify any hypothesis, and further studies should be undertaken to determine whether larval mortality is a significant factor. The length of larval life in bivalves varies, depending on environmental factors and the availability of substratum (Gosling, 2003; Porri, 2003). For example, the duration of larvae in *D. polymorpha* can vary from 6 to 52 weeks (Sprung, 1993), while in

Mytilus edulis larval settling times are 1-6 months (Tyler-Walters, 2008). Larval duration in *M. adamsi* was found to be comparatively short and settlement usually occurs within a few days after spawning (~4 days in Kalyanasundaram, 1975; NIMPIS, 2002a). The ability of *M. adamsi* larvae to delay their metamorphosis remains unknown. However if most larvae in the SW monsoon prolonged their planktonic stage and settled in the period of peak recruitment (i.e. November 07, Figure 3.1K), the larval sample would be found in the early NE monsoon (i.e. October 07, Figure 3.1K). Possibly, most of the larvae in the SW monsoon died before recruitment. In the NE and SE monsoon the larvae were likely to settle prior to the plankton sampling, and thus larvae were not found.

In this investigation, the abundance of *M. adamsi* was related to some physical and biological parameters, mainly salinity and food supply. The species appears to prefer low salinity and a high abundance of some groups of phytoplankton: phytoflagellates, dinoflagellates, and diatoms. The results support field observations suggest that the preferred habitat of *M. adamsi* was a shallow coastal lagoon or a brackish shelter habitat that invariably have an appropriate salinity and high phytoplankton abundance.

CHAPTER 4

Spatial and temporal variability in recruitment of the invasive mussel *Mytilopsis adamsi* Morrison, 1946 and some environmental characteristics in Haad-kaew Lagoon, in the lower south of the Gulf of Thailand

INTRODUCTION

An invasive mussel *Mytilopsis adamsi* Morrison, 1946 (Bivalvia: Dreissenidae) has been established in Haad-kaew Lagoon, a small shallow estuarine lagoon located at the coast of the lower south of the Gulf of Thailand. The species has occupied the whole lagoon with its dense colony, and may be in its spreading stage according to the generalized step of the invasion process proposed by Sakai *et al*., 2001. In order to establish suitable management practices for this species in this area, its basic biological information is needed.

There have been many studies that deal with spatial and temporal variations in populations characteristics of mytilid mussels dwelling in wave-exposed intertidal habitats (Hunt and Scheibling, 1998; Porri *et al*., 2006a; Porri *et al*., 2006b; Smith *et al*., 2009). These have been recognized as "open" mussel populations (Porri, 2003). In contrast *M. adamsi* is distinct. It inhibits in the sheltered shallow coastal lagoon (NIMPIS, 2002a), thus the population is somewhat "close", and the factors influencing its biology are different. Some coastal lagoons are among the most productive aquatic ecosystems being rich in phytoplankton and macrophytes. The accumulation of organic matter usually leads to oxygen depletion and sulfate reduction which causes the death of the macrofauna (Gattuso, 2007). Water temperature, tidal amplitude, and salinity are also the important factors that affect the biology of the organisms in the coastal lagoons (Chícharo and Chícharo, 2001; Basset *et al*., 2006; Wilkoňska and Kapusta, 2007; Chuwen *et al*., 2009)

The reproductive activities in a population establish the regulation and dynamics of the population. Some organisms reproduce continuously all year round while some have the rhythmic reproduction cycles. Karande and Menon (1975) and Morton (1989) have reported two periods of reproductive activity for *M. sallei* per year. Other dreissenids have shown pulses of reproduction*: Mytilopsis leucophaeata* in Europe (Verween *et al*., 2005), and *Mytilopsis sallei* in the Colombian Caribbean (Puyana, 1995). Knowledge of the time of reproduction is useful for invasive species control such as it introduces the possibility of using chemical treatment during the mussel's reproductive period (Verween, 2006). In this present study recruitment was used to determine the mussel's reproductive season, because it is the stage when mussels have survived from their post-settlement mortality and have entered the adult population, to demonstrate the success of *M. adamsi* reproduction. There were several factors that potentially influenced on the mussel recruitment including temperature, salinity, food supplies and abundance of larvae. Raju *et al*. (1975), Rao *et al*. (1975), Kalyanasundaram (1975) have demonstrated that temperature and salinity were the two major key parameters determining the success of reproduction and survival of *M. sallei* in India, and this was similar to the report by Morton (1989) in Hong Kong. Moreover, recruitment, larval growth, and mortality in mussels were usually correlated with the abundance of larvae (Martel *et al*., 1994; Gosling, 2003; McQuaid and Lawrie, 2005) and food supplies (His *et al*., 1989; Horvath and Lamberti, 1999; Gosling, 2003; Bos *et al*., 2006).

The high variability of the habitat characteristics in Haad-kaew Lagoon make it a suitable study site for investigating what are the preferred conditions for *M. adamsi* recruitment. The microhabitat in Haad-kaew Lagoon varies morphologically and the physical conditions are heterogeneous (personal observation; Wangkulangkul and Lheknim, 2008). The seasonality of the physical conditions in Haad-kaew Lagoon possibly promotes the seasonal variability in biological factors, such as the amount of phytoplankton that will influence the *M. adamsi* population characteristics or will have an effect on the mussel directly (See also chapter 3).

The objective of this study was to understand the population biology of *M. adamsi* and provide the background knowledge for the further studies and the management of the population. This research was conducted to gain the knowledge of the period(s) of its reproductive activity and the environmental condition affecting its reproduction, by investigating the spatial and temporal variations in recruitment of *M. adamsi* and some environmental characteristic, such as salinity, water temperature, pH, dissolved oxygen, density of phytoplankton, zooplankton, and the D-shaped larvae in Haad-kaew Lagoon.

METHOD

To achieve the aims of the study, multi-stage sampling was used to collect the data. The data used to examine the spatial and temporal variations in recruitment of mussel and the environmental characteristics were obtained from the methods given in chapter 2. So, only the details of the statistical analysis descriptions are provided here.

Data Analysis

The Mixed Model ANOVA (two-way ANOVA) was employed to determine the spatial and temporal variability of the density of *M. adamsi* recruits and environmental factors during the study period. The two main independent variables were habitat and season. There were two levels of habitats in this study, the seasonally-closed and the open lagoons and three levels of seasons; the SW monsoon from May-September 2007; the NE monsoon from October-December 2007; and the SE predominant wind period from January-April 2008. The month was a random factor nested within each season. In this study, the word “month” was used for the small scale temporal factors although soaking periods were not concordant with the real month. The spatial factors treated as fixed were habitat and zone. The temporal factor treated as being fixed was the season while the month was treated as being random. The zone was nested in the habitat; the factor month was nested in the season, while the station was a random effect, nested in all the above spatial and temporal factors. The density of recruits and the environmental factors were treated as dependent variables. The normality of dependent variables was examined using the Shapiro-Wilk normality test (Shapiro and Wilk, 1965). The homogeneity of variances was tested using the Levene's test (Levene, 1960). The density data of mussel recruits were transformed to logarithms (density+0.5) while other biological variables were log (density+1) transformed in the case that dependent data didn't have normality or homogeneity of variance. In some cases, transformation did not produce homogeneous variances, but nevertheless ANOVA was used, because it is fairly robust to departures from homogeneity when sample sizes are equal (Underwood, 1997). A setting of $\alpha = 0.01$ was used in such cases to compensate for the increased likelihood of Type I error. From preliminary analysis, Levene's test showed that all biological variables exhibited unequal variances, so $\alpha = 0.01$ was applied for these variables. All physical variables presented equal variances. In the case, where two-way interactions between lagoons and seasons occurred, the significant differences among levels of lagoon and season were tested separately with one-way nested ANOVA and Tukey's test for a posteriori comparison of the means. In the one-way ANOVA, zone was nested in habitat and month was nested in season. Season, habitat, and zone were treated as being fixed while month was treated as being random. All statistical analyses were performed on JMP® 8.0 (trial version; SAS, 2008).

RESULTS

Spatial and temporal variability in the density of *Mytilopsis adamsi* recruitment

During the sampling period, the density of *Mytilopsis adamsi* ranged from 0 to 5,690 mussels per 100 cm^2. The highest density was found in the December 2007 (NE monsoon). There were two pulses of recruitment of *M. adamsi* in the study period. A minor pulse was from May to August 2007 in the SW monsoon season, and a major pulse was from November 2007-March 2008, mid way between the NE monsoon and the SE predominant wind period (Figure 3.1N, chapter 3). There was a two-way interaction between factor habitats and seasons on the density of the recruits ($F_{2, 447} = 36.647$, $p < 0.0001$, Table 4.1). In the SW monsoon the density of mussel recruits in the seasonally-closed lagoon was higher than in the open lagoon; while in the NE monsoon and the SE predominant wind period the density in the open lagoon was higher (Figure 4.1). Habitat ($F_{1, 645} = 0.017$, $p = 0.904$, Table 4.2) and season ($F_{2, 638} = 0.753$, $p = 0.498$, Table 4.2) had no influence on the density of the *M. adamsi* recruits.

Table 4.1 Mixed Model ANOVA of the density of *Mytilopsis adamsi* recruits collected from Haad-kaew Lagoon during May 2007 to April 2008. Asterisk (**) indicated the significant *F* ratio, considered at the 0.01 level.

Sources	df	MS	F	p
Habitat	1	3.797	0.212	0.676
Season	2	19.222	0.319	0.734
Habitat*Season	2	14.288	36.647	<0.0001**
Zone(Habitat)	3	17.915	45.949	<0.0001**
Month(Season)	9	60.146	154.265	<0.0001**
Station(H,S,Z,M)	169	0.309	0.792	0.962
Error	447	0.390		

Table 4.2 One-way nested ANOVA of the density of *Mytilopsis adamsi* recruits collected from Haad-kaew Lagoon during May 2007 to April 2008. Asterisk (**) indicated the significant *F* ratio, considered at the 0.01 level.

Sources	df	MS	F	p	Tukey's test
Habitat	1	0.414	0.017	0.904	No test
Zone(Habitat)	3	24.143	16.149	<0.0001**	
Error	645	1.495			
Season	2	44.672	0.753	0.498	No test
Month(Season)	9	59.312	96.207	<0.0001**	
Error	638	0.616			

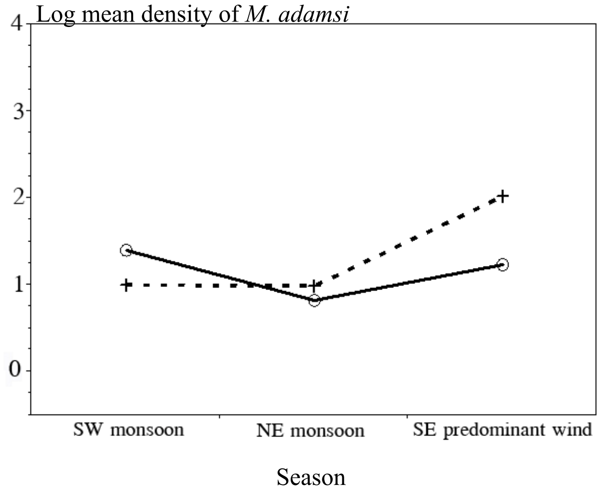


Figure 4.1 Log (means) plot between season and habitat of the density of *Mytilopsis adamsi* recruits collected from Haad-kaew Lagoon during May 2007 to April 2008. Solid line = seasonally-closed lagoon; broken line = open lagoon.

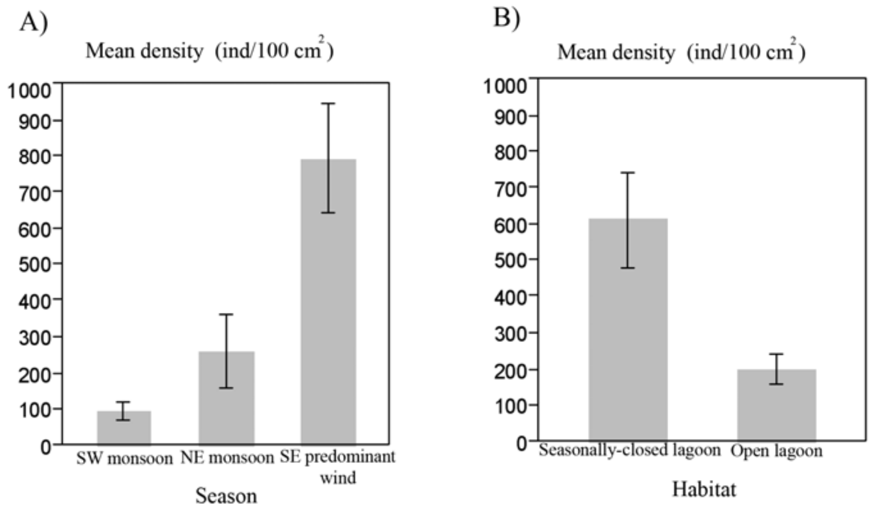


Figure 4.2 A) Seasonal and B) habitat variation of the density of *Mytilopsis adamsi* recruits (mean ± SE) collecting from Haad-kaew Lagoon during May 2007 to April 2008.

Habitat and seasonal variability in the environmental characteristics in Haad-kaew Lagoon

The ranges of environmental characteristics have been given in chapter 3. There were two-way interactions between factor habitat and season for dissolved oxygen ($F_{2, 22}$ = 4.613, p = 0.021, Table 4.3), salinity ($F_{2, 26}$ = 8.654, p = 0.001, Table 4.3), phytoflagellates ($F_{2, 26}$ = 6.499, p = 0.005, Table 4.3), rotifers ($F_{2, 26}$ = 7.948, p = 0.002, Table 4.3), D-shaped larvae ($F_{2, 26}$ = 7.97, p = 0.001, Table 4.3) and planktonic crustaceans ($F_{2, 26}$ = 13.628, p <0.0001, Table 4.3). The value of the dissolved oxygen was higher in the open lagoon except in the SE predominant wind period (Figure 4.3A). Salinity in the open lagoon was higher than in the seasonally-closed lagoon in every season, which it's highest value in the SW monsoon and lowest in the SE monsoon (Figure 4.3B). In the SW monsoon and SE predominant wind period, the Least Square means of the phytoflagellates density in the seasonally-closed lagoon were higher than in the open lagoon, which is contrary to that in the NE monsoon. Moreover, in the seasonally-closed lagoon Least Square means, in SE predominant wind period were highest and in the NE monsoon were the lowest, while in the open lagoon the Least Square means in the SW monsoon were highest (Figure 4.3C). The Least Square means of rotifers density in the seasonally-closed lagoon were higher than in the open lagoon, and were higher in the SW monsoon than in the SE predominant wind period and the NE monsoon respectively (Figure 4.3D). In the NE monsoon and the SE predominant wind period, Least Square means of the D-shaped larvae density were higher in the seasonally-closed lagoon than in the open lagoon, which is contrary to the situation in the SW monsoon (Figure 4.3E). Least Square means of the density of planktonic crustaceans were higher in the seasonally-closed lagoon than in the open lagoon, and Least Square means in the SW monsoon were higher than in other seasons (Figure 4.3F).

For variables when the two-way interactions were not found, there were no significant differences between seasons (Table 4.3) and the differences between lagoons were significant for the cyanophytes ($F_{1, 26}$ = 54.405, p = 0.005, Table 4.3) and the large colony cyanophytes ($F_{1, 26}$ = 59.944, p = 0.004, Table 4.3). The density of cyanophytes was higher in the seasonally-closed lagoon as compared to the open lagoon (Tukey's test in Table 4.4 and Figure 4.5). The densities of the large colony cyanophytes were higher in the seasonally-closed lagoon (Figure 4.5). In the case of the large colony cyanophytes the results from the Mixed Model ANOVA showed a significant effect of habitat on the large colony cyanophytes while one-way analysis revealed no significant effects. However the final result relied on the Mixed Model analysis.

One-way ANOVA was performed to examine the significant differences among seasons and habitats for environmental characteristics that presented two-way interactions between lagoons and seasons in the Mixed Model analysis. The environmental characteristics showing a significant effect of habitat that produce a higher abundance in the seasonally-closed lagoon were phytoflagellates ($F_{1, 226}$ = 49.121, p = 0.006, Table 4.4), rotifers ($F_{1, 226}$ = 54.487, p = 0.005, Table 4.4) and planktonic crustaceans ($F_{1, 226}$ = 90.050, p = 0.002, Table 4.4) while dissolved oxygen ($F_{1, 214}$ = 12.580, p = 0.038, Table 4.4), salinity ($F_{1, 235}$ = 342.079, p = 0.0003, Table 4.4) and D-shaped larvae ($F_{1, 224}$ = 86.032, p = 0.003, Table 4.4) in the open lagoon were higher (See also Figure 4.5). The season had an influence on salinity ($F_{2, 228}$ = 4.394, p = 0.047, Table 4.4), as in the SW monsoon it was significantly higher when compared to that in the NE monsoon and the SE predominant wind period. Salinity in the NE monsoon and SE predominant wind period were not significantly different.

Table 4.3 F-ratios and probabilities from the Mixed Model ANOVA of environmental characteristics collecting from Haad-kaew Lagoon during May 2007 to April 2008. The significant level of *F*-values were determined separately due to the homogeneity of variance of the data; the 0.05 (*) level for physical characteristics and the 0.01(**) level for the biological characteristics.

Sources	Dissolved oxygen (mg/L)				Water temperature (˚C)				Salinity (ppt)			
	df	MS	F	p	df	MS	F	p	df	MS	F	p
Habitat	1	9.021	27.848	0.010*	1	7.937	4.405	0.127	1	6453.360	462.885	0.0002*
Season	2	3.488	0.316	0.738	2	220.261	3.704	0.073	2	2190.940	4.668	0.055
Habitat*Season	2	8.652	4.613	0.021*	2	1.329	0.950	0.402	2	212.716	8.654	0.001*
Zone(Habitat)	3	0.324	0.173	0.914	3	1.802	1.288	0.303	3	13.942	0.567	0.641
Month(Season)	8	11.048	5.891	0.0004*	8	59.466	42.509	<0.0001*	9	538.546	21.911	<0.0001*
Station(H,S,Z,M)	164	0.274	0.146	1.000	164	1.209	0.864	0.707	180	1.197	1.000	1.000
Error	22	1.875			22	1.399			26	24.579		

Sources	pH				Chlorophytes (ind/L)				Cyanophytes (ind/L)			
	df	MS	F	p	df	MS	F	p	df	MS	F	p
Habitat	1	0.256	0.417	0.565	1	0.939	0.663	0.475	1	40.885	54.405	0.005**
Season	2	2.088	0.660	0.540	2	16.297	0.973	0.414	2	36.127	1.668	0.242
Habitat*Season	2	0.206	0.349	0.709	2	3.890	1.547	0.232	2	3.403	2.659	0.089
Zone(Habitat)	3	0.613	1.039	0.392	3	1.417	0.566	0.642	3	0.751	0.587	0.628
Month(Season)	9	3.162	5.357	0.0004*	9	16.753	6.697	<0.0001**	9	21.660	16.922	<0.0001**
Station(H,S,Z,M)	178	0.616	1.044	0.472	170	0.777	0.310	1.000	171	0.910	0.711	0.897
Error	26				26	2.502			26	1.280		

Sources	Large colony cyanophytes (ind/L)				Diatoms (ind/L)				D-shaped larvae (ind/L)			
	df	MS	F	p	df	MS	F	p	df	MS	F	p
Habitat	1	6.452	59.944	0.004**	1	0.345	0.316	0.613	1	0.021	2.793	0.193
Season	2	3.070	3.280	0.085	2	3.991	0.510	0.617	2	0.392	0.769	0.491
Habitat*Season	2	2.873	5.452	0.011	2	5.863	4.255	0.025	2	0.492	7.97	0.001**
Zone(Habitat)	3	0.108	0.204	0.892	3	1.092	0.795	0.509	3	0.007	0.124	0.945
Month(Season)	9	0.936	1.776	0.122	9	7.823	5.678	0.0002**	9	0.510	8.256	<0.0001**
Station(H,S,Z,M)	171	0.075	0.142	1.000	171	0.764	0.555	0.986	169	0.012	0.195	1.000
Error	26	0.527			26	1.378			26	0.062		

Table 4.3 (continued)

Sources	Dinoflagellates (ind/L)				Phytoflagellates (ind/L)				Rotifers (ind/L)			
	df	MS	F	p	df	MS	F	p	df	MS	F	p
Habitat	1	75.698	11.799	0.041	1	18.885	12.594	0.038	1	60.458	48.433	0.006**
Season	2	29.036	1.570	0.260	2	32.782	0.844	0.461	2	19.990	6.541	0.017
Habitat*Season	2	5.079	1.213	0.314	2	18.001	6.499	0.005**	2	7.683	7.948	0.002**
Zone(Habitat)	3	6.416	1.532	0.230	3	1.500	0.541	0.658	3	1.248	1.291	0.298
Month(Season)	9	18.49	4.414	0.001**	9	38.826	14.017	<0.0001**	9	3.056	3.161	0.0103
Station(H,S,Z,M)	171	0.918	0.219	1.000	171	0.773	0.279	1.000	171	0.180	0.186	1.000
Error	26	4.19			26	2.770			26	0.967		

Sources	Planktonic crustaceans (ind/L)			
	df	MS	F	p
Habitat	1	109.339	97.606	0.002**
Season	2	86.426	4.973	0.035
Habitat*Season	2	15.192	13.628	<0.0001**
Zone(Habitat)	3	1.120	1.005	0.406
Month(Season)	9	17.378	15.589	<0.0001**
Station(H,S,Z,M)	171	0.329	0.295	1.000
Error	26	1.115		

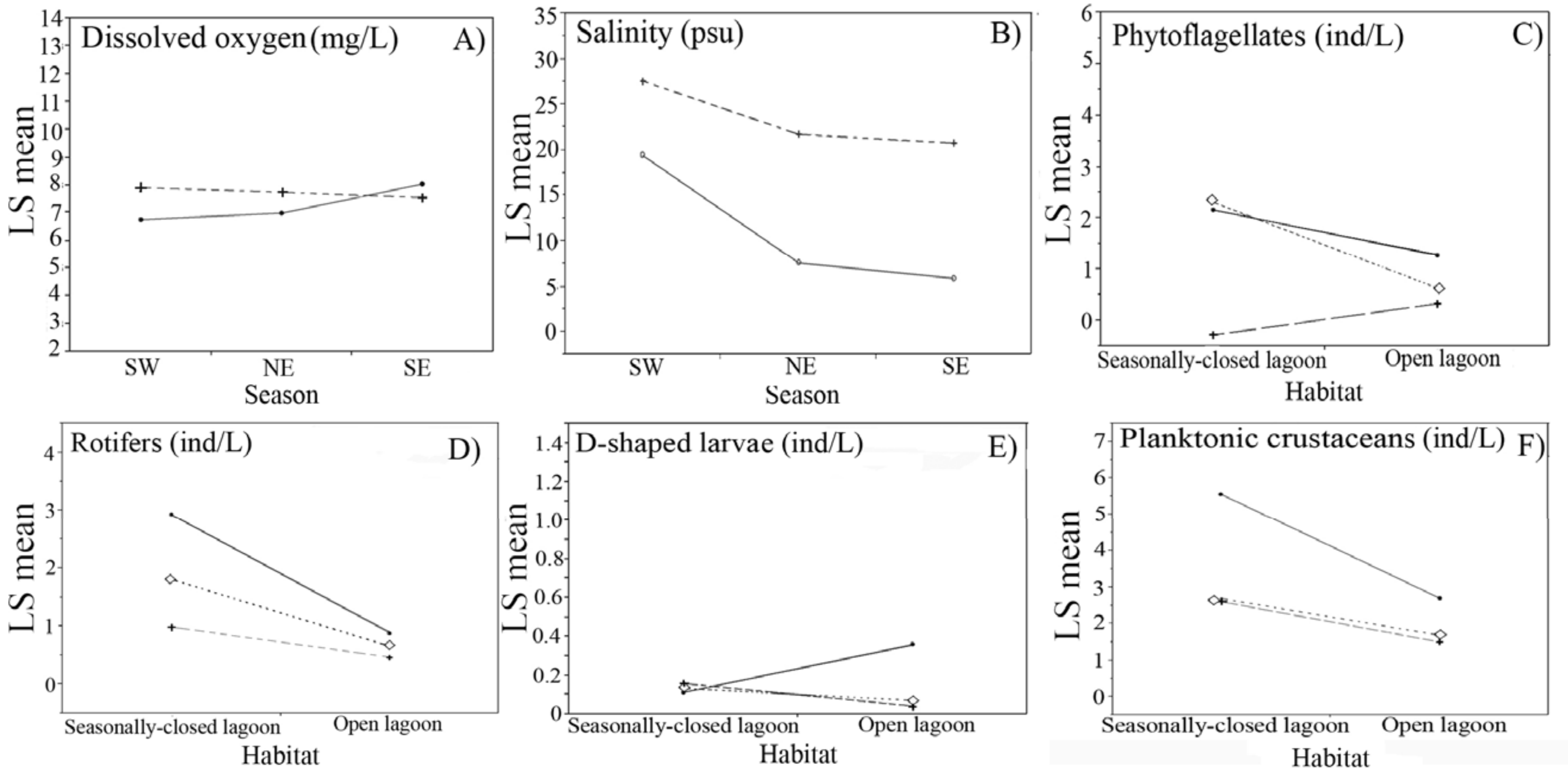


Figure 4.3 Least Square mean plot for seasonal and habitat variation on environmental characteristics collecting from Haad-kaew Lagoon during May 2007 to April 2008. In Figure A-B, solid lines refer to seasonally-closed lagoon; broken lines refer to open lagoon. In Figure C-F, solid lines refer to the SW monsoon (SW); dashed lines refer to the NE monsoon (NE); dot lines refer to the SE predominant wind period (SE).

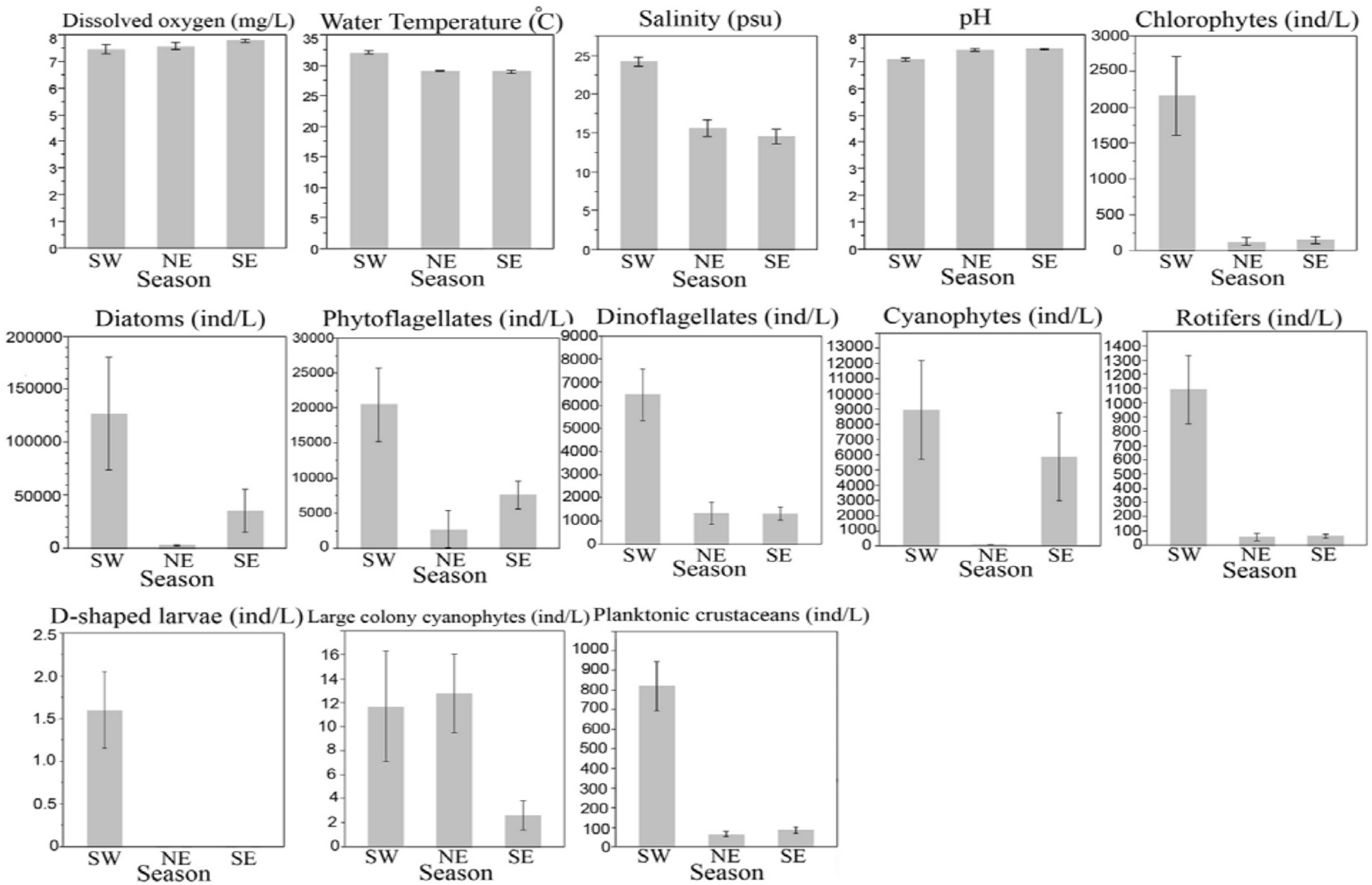


Figure 4.4 Seasonal variations in environmental characteristics (mean ± S.E.) collected from Haad-kaew Lagoon during May 2007 to April 2008. The SW monsoon (SW) was from May-September 2007; the NE monsoon (NE) was from October-December 2007; the SE predominant wind period (SE) was from January-April 2008

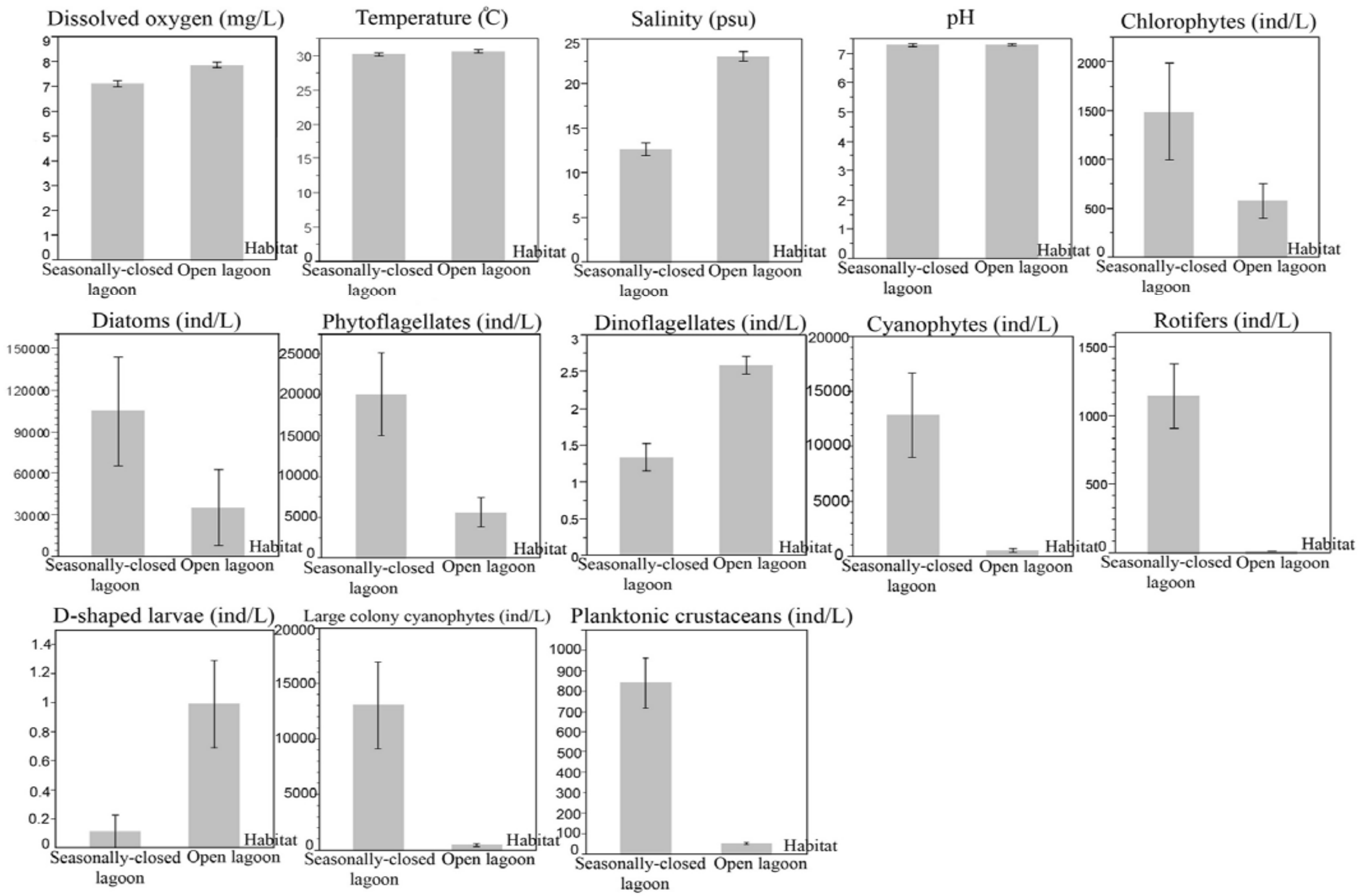


Figure 4.5 Habitat variations in environmental characteristics (mean ± SE) collected from Haad-kaew Lagoon during May 2007 to April 2008. Habitat 1 = seasonally- closed lagoon; Habitat 2 = open lagoon

Table 4.4 MS estimates and F-ratios from one-way ANOVA for the environmental characteristics collected from Haad-kaew Lagoon during May 2007 to April 2008. The significant level of *F*-values were determined separately due to the homogeneity of variance of the data; the 0.05 (*) level for physical characteristics and the 0.01(**) level for biological characteristics. Habitat: 1 = seasonally-closed lagoon, 2 = open lagoon. Season: 1 = SW monsoon, 2 = NE monsoon, 3 = SE predominant wind period. Different superscripts show the significantly different means.

Sources	Dissolved oxygen (mg/L)					Water temperature (˚C)				
	df	MS	F	p	Tukey's test	df	MS	F	p	Tukey's test
Habitat	1	28.278	12.580	0.038*	$2^{a} > 1^{b}$	1	10.489	4.260	0.131	No test
Zone(Habitat)	3	2.248	1.612	0.186		3	2.462	0.407	0.752	
Error	214	1.394				214	6.054			
Season	2	1.844	0.122	0.887	No test	2	261.870	4.181	0.057	No test
Month(Season)	8	15.092	15.017	<0.0001*		8	62.632	44.769	<0.0001*	
Error	208	1.005				208	1.399			

Sources	Salinity (ppt)					pH				
	df	MS	F	p	Tukey's test	df	MS	F	p	Tukey's test
Habitat	1	6278.365	342.079	0.0003*	$2^{a} > 1^{b}$	1	0.629	0.971	0.397	No test
Zone(Habitat)	3	18.353	0.367	0.779		3	0.647	0.904	0.442	
Error	235	49.930				233	0.716			
Season	2	2490.790	4.394	0.047*	$1^{a} > 2^{b}, 3^{b}$	2	1.909	0.580	0.580	No test
Month(Season)	9	566.822	16.189	<0.0001*		9	3.292	5.469	<0.0001*	
Error	228	35.012				226	0.602			

Table 4.4 (continued)

Sources	Chlorophytes (ind/L)					Cyanophytes (ind/L)				
	df	MS	F	p	Tukey's test	df	MS	F	p	Tukey's test
Habitat	1	11.193	7.958	0.667	No test	1	63.730	69.340	0.004**	1[a] > 2[b]
Zone(Habitat)	3	1.406	0.709	0.551		3	0.919	0.347	0.794	
Error	225	1.983				226	2.649			
Season	2	17.320	0.880	0.448	No test	2	36.509	1.446	0.285	No test
Month(Season)	9	19.680	17.047	<0.0001**		9	25.238	15.117	<0.0001**	
Error	218	1.154				219	1.669			

Sources	Large colony cyanophytes (ind/L)					Diatoms (ind/L)				
	df	MS	F	p	Tukey's test	df	MS	F	p	Tukey's test
Habitat	1	6.584	25.423	0.015	No test	1	13.616	28.397	0.013	No test
Zone(Habitat)	3	0.259	1.148	0.331		3	0.479	0.358	0.786	
Error	226	0.226				226	1.341			
Season	2	2.804	2.834	0.111	No test	2	4.683	0.470	0.640	No test
Month(Season)	9	0.989	4.946	<0.0001**		9	9.969	9.947	<0.0001**	
Error	219	0.200				219	1.002			

Sources	D-shaped larvae (ind/L)					Dinoflagellates (ind/L)				
	df	MS	F	p	Tukey's test	df	MS	F	p	Tukey's test
Habitat	1	0.459	86.032	0.003**	2[a] >1[b]	1	87.327	14.578	0.032	No test
Zone(Habitat)	3	0.005	0.093	0.958		3	5.990	2.424	0.065	
Error	224	0.057				226	2.471			
Season	2	0.673	1.039	0.393	No test	2	31.353	1.539	0.266	No test
Month(Season)	9	0.648	23.559	<0.0001**		9	2.367	10.807	<0.0001**	
Error	217	0.027				219	1.885			

Table 4.4 (continued)

Sources	Phytoflagellates (ind/L)					Rotifers (ind/L)				
	df	MS	F	p	Tukey's test	df	MS	F	p	Tukey's test
Habitat	1	60.693	49.121	0.006**	$1^{a} > 2^{b}$	1	117.484	54.487	0.005**	$1^{a} > 2^{b}$
Zone(Habitat)	3	1.236	0.369	0.779		3	2.156	2.889	0.036	
Error	226	3.351				226	0.746			
Season	2	33.436	0.772	0.490	No test	2	22.044	7.255	0.013	No test
Month(Season)	9	43.285	26.146	<0.0001**		9	3.038	3.036	0.002**	
Error	219	1.655				219	1.001			

Sources	Planktonic crustaceans (ind/L)				
	df	MS	F	p	Tukey's test
Habitat	1	184.562	90.050	0.002**	$1^{a} > 2^{b}$
Zone(Habitat)	3	2.050	0.915	0.436	
Error	226	2.239			
Season	2	96.749	5.356	0.029	No test
Month(Season)	9	18.064	11.704	<0.0001**	
Error	219	1.543			

DISCUSSION

Assessing spatial and temporal variability has been the general approach to understand community organization and population dynamics in aquatic organisms (Porri *et al*., 2006a). Here the effect of both scales on the recruitment of *Mytilopsis adamsi* and other environmental variables in Haad-kaew Lagoon were investigated. Although the effects of season and lagoon on *M. adamsi* recruitment were not significant (Table 4.2), the seasonal and spatial variability of recruitment in Haad-kaew Lagoon was found noticeably. The mean density (±SE) of mussel recruits in different seasons and lagoons (Figure 4.2) indicated that the density of recruits was highest in the seasonally-closed lagoon and in the SE predominant wind period season. It was found to be lowest in the SW monsoon. The effects of season and habitat on mussel recruitment were we can assume masked by the high variability of recruitment among the nested factors, month and zone (Table 4.2). Dissolved oxygen, pH, temperature, chlorophytes, diatoms, dinoflagellates, phytoflagellates, cyanophytes, rotifers, D-shaped larvae and planktonic crustaceans were also not significantly influenced by season while the nested factors, month, was significant (Table 4.3 and 4.4). Especially for the biological variables, the differences among seasons were obvious (Figure 4.4). I suggested that the high heterogeneity of variance among month nested within a season highlights the non significance of the factor season in these variables. Moreover, in the case of the seasonal factor, the unit of season allocated in this study possibly did not reflect the real natural season. However, when considering the amount of rainfall that is used to determine the unit of the season in this study period, it shows a pattern that is consistently determined by the season (Figure 2.2B in chapter 2) in that the amount of rainfall was highest in the NE monsoon (October 2007-December 2007) and lowest in the SE predominant wind period. Nevertheless there is another possibility that seasonal factors, such as rainfall and sea water flushing, may not affect the recruitment simultaneously. The climatic environment may not correspond to the hydrological environment. For example, the amount of rainfall was highest in October 2007 but the salinity that was suggested to be the main factor influencing the recruitment, was lowest in February 2008.

In Haad-kaew Lagoon, the effect from the sea and seasonal freshwater inflow possibly cause the variations in most of the lagoon characteristics, such as salinity, dissolved oxygen, pH, turbidity (personal observation). The variation of seawater flushing and freshwater inflow resulted mainly in a spatial and temporal variability of salinity and this was significant. Salinity in the open lagoon was influenced by tide so it was higher than in the seasonally-closed lagoon. Although the tide was relatively high from October 2007, the salinity declined continuously, which was possibly caused by the rainfall. The rapid drop in salinity in November 2007, especially in the open lagoon, was driven by the heavy rainfall and run-off since October 2007 (Figure 3.1C in chapter 3 and Figure 2.2B in chapter 2). The effect of the tide and wave driven by the wind possibly caused higher values of dissolved oxygen found in the open lagoon. Temperature and pH were not different among season and between lagoons. The all year round mean air temperature fluctuated in a small range of about 2-3 Celsius (Figure 2.2C in chapter 2), resulting in a small change of water temperature in the lagoons. The pH in both lagoons was reasonably constant throughout the year with an average of 7.

Most plankton had a high abundance in the seasonally-closed lagoon, this possibly due to the high nutrient concentration. The evidence for nutrient richness is supported by the statement that a “phytoplankton bloom” has been frequently observed. In tropical zone, mangrove communities surrounding coastal lagoons provide a high quality of organic matter to the aquatic system through litter fall. The organic matter becomes a source of nutrients that may affect the composition and productivity of the phytoplankton communities (Ake´-Castillo and Va´zquez, 2008). Actually, there are several patches of mangrove forest on the shore of the seasonally-closed lagoon that potentially could supply the nutrients to the lagoon. Besides, the other source of nutrient may be from the sewage discharged by the hotel located nearby and the nutrient cycle in the lagoon.

Although bivalves are generally regarded as herbivores, some recent studies reported that filter-feeding mussel also feed on zooplankton (Davenport *et al*., 2000; Robinson *et al*., 2002; Lehane and Davenport, 2002; Lehane and Davenport, 2006). The role of zooplankton in mussel biology is not only being as a food source for adult mussel but it also competes with mussel larvae for foods. The density of rotifers and planktonic crustaceans was high in the SW monsoon and found relatively low in the NE monsoon and SE predominant wind period when the mussels abundantly recruited (Figure 4.2 and 4.4). The density of planktonic crustaceans was also negatively correlated with the *M. adamsi* recruitment in the last two seasons (Table 3.4 in chapter 3). This indicated that the competition between this zooplankton and *M. adamsi* larva for food or the predation by mussel adult was a cause of the low density of the zooplankton.

The abundance of D-shaped larvae found in the open lagoon was higher than in the seasonally-closed lagoon. This may be the result of the fluctuation in salinity that induced the mussel to spawn and produce more offspring, similar to that in Puyana (1995). However there was no coupling between larvae and recruitment in this present study (see chapter 3).

The recruitment of *M. adamsi* exhibited a negative correlation with salinity (see chapter 3). The decrease of salinity to below 10 ppt in the mid NE monsoon accompanied with the low salinity in the SE predominant wind period possibly caused the major peak of recruitment in this season. Despite the finding that salinity was high in the SW monsoon, a minor peak of recruitment was found. The high abundance of its food, phytoplankton and zooplankton, in the SW monsoon probably provided a suitable condition for *M. adamsi* to increase its reproductive activity. Phytoplankton is generally the main food source of filter-feeding bivalves (Gosling, 2003). Diatoms, phytoflagellates and dinoflagellates were positively correlated with the density of the *M. adamsi* recruits (see chapter 3). The presence of a plentiful source of phytoplankton may cause a minor peak of recruitment of *M. adamsi*. Phytoplankton can act as a chemical cue for mussel spawning; such as with *D. polymorpha* that can be activated to spawn by an extract from marine algae (Ram *et al*., 1995). Moreover, spawning synchronized with phytoplankton bloom may increase the survival of larvae to their settling stage by increasing the larval growth rate and consequently reducing the risk of facing with predators. This consequence were observed and clearly interpreted in the case of the zebra mussel (Ram *et al*., 1996; Tyler-Walters, 2008). However such physiological consequences have not been investigated in *M. adamsi*.

The two unequal recruitment pulses of *M. adamsi* were potentially related to the monsoon wind (Figure 3.1N in chapter 3). Because the factors, especially salinity, that influence *M. adamsi* fluctuate due to the rainfall and seawater surges that are markedly associated with the monsoons. Two periods of reproductive activity of *M. sallei* per year were reported by Karande and Menon (1975) in India, and Morton (1989) in Hong Kong. However they did not demonstrate any relationship between the reproduction phases and the monsoon. The seasonality of the *M. adamsi* recruitment in the present study is similar to the trends of spawning in the fishs *Stolephorus spp*. as suggested by Tiews *et al*. (1975). In their study the fish exhibited a peak during the northeast monsoon season (October to March) followed by a period of little or no spawning activity from April to July. Pauly and Navaluna (1983) analysed the pattern of juvenile fishes recruited to coastal zones of the Philippines and found the two asymmetric pulses of recruitment in many fishes. They also concluded that there was a very close relationship between the spawning/recruitment patterns and the monsoon winds, and mentioned that the pattern was similar to other areas of the tropical Indo-Pacific.

CHAPTER 5

General discussion

The main purpose of this study is to gain the basic knowledge of *Mytilopsis adamsi* biology. In this study I found that the recruitment of *M. adamsi* was correlated with the density of some phytoplankton which potentially preferred by the mussel and the relatively low salinity. There are two pulses of recruitment, which potentially influenced by the monsoon. Sudden drop of salinity at the end of the northeast monsoon season was considered as a trigger to induce spawning, subsequently caused the majority of recruitment. The minor peak occurred during the mid of southwest monsoon season while most of plankton had high density. The information from the field observation and data collecting suggested that the preferred habitat of *M. adamsi* is shallow coastal lagoons or brackish shelter habitats which have variability of salinity and rich in phytoplankton. Information gains from this present study is hopefully contributed for using as basic guideline to predict the risk areas and aquaculture practices in Haad-kaew Lagoon, Thale Sap Songkhla and probably in the lower south of the Gulf of Thailand.

Once the invasive species has been introduced and established in any area, there are 2 approaches that are crucial for species management - the prevention of the further expansion and the species control. In order to inhibit the spreading out of such invasive species, we need to know the distribution strategies of the species and the environmental conditions which influence on the success of species colonization (Trowbridge, 1999; Sakai *et al*., 2001). In the invasive mussel control, biocide and biological control are employed in many countries (Nalepa and Schloesser, 1992; GISP, 2004; Verween, 2006; Molloy and Mayer, 2007). The susceptibility of mussel for biocide or their enemies is varied due to their stages of life and physiological conditions (Verween, 2006). Knowledge on population biology related to reproductive activity, population regulation and dynamics of the species is usually utilized to predict the area under risk to be invaded and to design the prevention method. Moreover, it will be background information for further species control and management.

Vulnerability of *Mytilopsis adamsi* in the lower Gulf of Thailand

The most preferable habitats of *Mytilopsis adamsi* found in Thale Sap Songkhla of the Songkhla Lake Basin, Pak Phanang estuary and the coast of Panarae in Pattani Province were shallow-brackish sheltered habitats, with the depth of the water less than 2 m. All above localities were subjected to salinity alteration due to freshwater run off with salinity ranged from 0 to 28 ppt In most observations *M. adamsi* were found associated with turbid waters resulted from suspended planktons and possibly nutrient-rich medium. Apparently, *M. adamsi* were common found attached on mangrove stems, roots, ropes and frames of fishing gears and other hard substratum such as poles of the piers, and even fouling on the hard parts of the native bivalves in the areas.

The complexity of the shore lines of the lower Gulf of Thailand give rise to a diversified habitats appropriate to colonization and establishment of *M. adamsi*. For example, embayments range from relatively smaller size as Pattani Bay to extremely large as Ban Don Bay in Surat Thani Province fed by tidal creeks and small streams, these are shallow-sheltered natural environments suit for the mussels. Some parts are bed rocks, such as along the area in the vicinity of Khanom (in Nakhon Si Thammarat Province) and Don Sak (in Surat Thani Province) are likely to provide favorable habitats for byssal threads attachment.

The complication of Thale Sap Songkhla, Thale Sap and Thale Luang of the Songkhla Lake system are also likely to offer a variety of environments and habitats for colonization and establishment of *M. adamsi*. The whole brackish waters of the Songkhla Lake system are likely to be the susceptible habitats for *M. adamsi*. Surprisingly, *M. adamsi* were only observed in the lower part and some tidal creeks of the Thale Sap Songkhla while they were absent from the rest of the Songkhla Lake system. In Haad-kaew Lagoon the colony of *M. adamsi* were found not only on the hard substrate, but also on the mud bottom of the soft substratum, suggesting that the species can also form the mussel beds in the lagoon system.

The water in Pak Phanang estuary is controlled by many salinity barrages which were closed in the dry season to prevent seawater inflow. When the barrages are opened the saline water can flow up to the upper part of the rivers and the water will become brackish, which is suitable for *M. adamsi* to live. The mussels larvae will also be carried to the upper part and colonize there. Although the *M. adamsi* populations were found for decade in the brackish area, up to now, the colonies of *M. adamsi* have not been found in the upper area or the other coastal habitat nearby. Because brackish water is like the isolated island surrounded by a barrier of sea or freshwater, it is unlikely for the mussel to cross these natural barriers by means of natural distribution especially in the larval stage. The mussels are possibly unable to establish in the pure freshwater or saline water, so that they are not found in the sea and the upper part of the basin.

Ecological and economical impacts of *Mytilopsis adamsi*

Ward and Ricciardi (2007) suggested that the interactions between introduced byssally attached mussels and benthic macroinvertebrates are universal, by that the introduction of these mussels was generally associated with increased macroinvertebrate density and taxonomic richness. In Haad-kaew Lagoon the colony of *Mytilopsis adamsi* were apparently found not only on the hard substrate, but also on the soft mud, suggesting that the species is able to form the mussel beds in the lagoon system. It is likely that if the mussels form the *M. adamsi* beds either in the Thale Sap Songkhla or the Pak Phanang estuary, this might promote high diversity of many macrobenthos such as polychaetes and amphipods and other benthic faunas. This is possibly a good role of this invasive *M. adamsi.*

However, in the permanently-opened part of Haad-kaew Lagoon, there are some native sessile bivalves, *Musculista senhousia*, *Perna viridis,* and *Crassostrea* spp., share the space with *M. adamsi*. The above natives are economically important, local fishermen mentioned that they are suffered from *M. adamsi* overgrowth and competition for the space with these natives. From observations, suspended ropes for growing the green lipped mussels, *P. viridis*, are attached by *M. adamsi* nearly half of their surface. *M. adamsi* also attach on the shell of other bivalves that the other suffocate from lack of water inflow. To remove and clean up *M. adamsi* from the ropes and fishing gears is costly and time consuming.

The Ban Don Bay in Surat Thani province is also one of the important sites for fisheries and aquaculture. The bivalve culture in the bay is well-known especially oysters *Crassostrea* spp., green lipped mussels *Perna viridis* and blood cockle *Anadara granosa*, which the yield cost more than 60 million baht annually (Surat Thani Provincial Fisheries Office, 2009). Supposing that the *M. adamsi* invade this area, it will have an effect on the production of the oysters and green lipped mussels by its fouling ability and compete for the space with cockle bed. Then, fisheries and aquaculture activities will also be damaged that we cannot estimate the value of this chaos.

The mussels are generally suspension feeders consuming mainly phytoplankton. The introductions of mussels usually alter the composition of planktonic community. Most researches working with freshwater zebra mussel *Dreissena polymorpha* (Padilla *et al*., 1996; Bastviken *et al*., 1998; Jack and Thorp, 2000), but there have been no available data for *M. adamsi*. The role of mussels on the planktonic community is possibly the same but whether it cause subsequent problem on aquatic food web or not probably depending on the native planktonic composition.

Implication to prevent *Mytilopsis adamsi* translocation and expansion

The mussel has two strategies to distribute naturally by means of adult dislodgement and the transport of planktonic larvae (Gosling, 2003; Karatayev *et al*., 2003). The larvae can disperse with a great distance in the water because of their tiny size, suitable morphology and adaptive biology. Because the duration of *Mytilopsis adamsi* planktonic stage was reported to be short, the natural distribution from other invaded countries is unlikely, so the possible modes of transport, or vectors, of *M. adamsi* are carrying by the vessels. The adult of *M. adamsi* are able to attach to hull and both larvae and adult may transfer alive in the ballast water. Many international organizations adopted the guidelines for monitoring and control of marine pests, these guidelines require ships to be inspected certain parts of the vessel under the designated protocols to prevent translocation of invasive species (GISP, 2004). There are several methods for treating the suspected species. The anti-fouling paint could reduce settlement of organisms on the hull. Before the ships disembark at the port, the hulls should be treated with chemical biocide. All ballast water exchange should be conducted far away in a distance from nearest land and in the fixed depth (GISP, 2004). These strategies are effective in many countries and should be enforced for routine practices in all international and regional ports in Thailand.

In case of local scales, for example the lower south of the Gulf of Thailand, the anthropogenic-induced translocations of *M. adamsi* from the recent infected areas are likely, since these areas are important in fisheries and aquaculture. The stationary fishing gears and boats provide excellent habitats for settlement and good vectors for *M. adamsi* to extend its distributional range. In order to prevent the boats or the gears from mussel attachment, after used in the infected area they should be dried if possible. The boats may be lift up from water and expose to the air until it is dry. The chemical treatment may be employed in urgent case under authority supervision. The issue that local people used *M. adamsi* as animal feeds, can be solved by publicizing the impacts of invasive species and proper management approaches. The translocation of the species should be avoided or conducted cautiously.

Implication for control of *Mytilopsis adamsi* in the lower Gulf of Thailand

Control aims to avoid undesirable economic and ecological impacts by reducing the abundance of the introduced populations to harmless levels. The control typically offers more practical, less damaging and (in the short-term at least) more affordable options than eradication. Moreover only the relatively small-size target population can be practically and successfully eradicated (GISP, 2004).

The uses of biocide and biological control are employed to reduce the abundance of mussel populations in many countries. Chlorine and chlorine dioxide are frequently used to eliminate *Dreissena polymorpha* from water-based infrastructure in the US (Boelman *et al*., 1997). Sodium hypochlorite and copper sulphate were used in Darwin Harbour estuary, Australia to remove *Mytilopsis* sp. (Bax *et al*., 2001). Up to now the most effective biological control for invasive mussel was reported by treating with bacteria toxins in *Dreissena* spp. Molloy and

Mayer (2007) reported the use of bacterial toxins derived from the naturally-occurring soil bacterium *Pseudomonas flourescens* strain CL0145A to control *Dreissena* spp. in the US. The predation by diving duck has been demonstrated to reduce biomass of *D. polymorpha* but had little impact on mussel population (Hamilton *et al*., 1994).

Most of biocides are used for killing the adult of mussels (Nalepa and Schloesser, 1992; GISP, 2004; Verween, 2006). However the adults have their defensive strategy against the use of biocide, such as closing their valves, while the larval phase is possibly the most vulnerable to change in external environment and also less resistant to the biocide (Claudi and Evans, 1993). Bayne *et al*. (1976) suggested the adult tend to be weaker after spawning when they have little energy reserves, so the most effectively use of biocide to control the mussels should be employed during the peak of reproductive activity. Then the knowledge of bivalve's life history and reproductive period are the most important facts for controlling mussel invasion.

In *Mytilopsis adamsi*, I suggested that the timing of recruitment is closed to the spawning period since the larval phase was reported to be short (Kalyanasundaram, 1975). The reproductive activity of *M. adamsi* in this area exhibits high strength at the end of the year, which related to monsoon wind, rain fall, and salinity decrease. The high density of recruits revealed the high density of the further adult population. There are many density-dependent factors that influence on the population, for example competition and pest. The vulnerableness of the adult in this situation to epidemic is helpful to adopt biocontrol. However there is still the shortage of information for *M. adamsi*'s pest anywhere.

In case of mussel population in the Thale Sap Songkhla and the Pak Phanang estuary, the eradication is unlikely to be done effectively. Haad-kaew Lagoon is a relatively smaller and “close” lagoon, so the eradication may be conducted successfully. Nevertheless the benefit from elimination of the mussel from Haad-kaew Lagoon is possibly not worthwhile because many farms located near shore have to be affected and most of diverse fauna in this lagoon will possibly disappear.

The further researches

Presently, we have known some of the environmental characteristics of the habitats which appropriate for *Mytilopsis adamsi* recruitment and the time of high recruitment in this area. Although there was the information elsewhere regarding *M. adamsi* biology, there has been the gap of knowledge in Thailand. The characteristics of each population may differ because the introduced populations are like the founder which has different genetic traits. Moreover the interaction between *M. adamsi* and ecosystem has never been investigated. In order to gain more knowledge for *M. adamsi* management in Thailand, the further research topics which should be done are given here in form of questions.

- What are the reproductive characteristics of *M. adamsi* in the lower Gulf of Thailand? When does the mussel spawn? The thorough researches on reproductive strategies of the mussel should be conducted to ensure and add up the information gained from this recent study.
- How much *M. adamsi* influence on the native community? How are the interaction between *M. adamsi* and the native sessile organisms and planktonic community? Does the presence of *M. adamsi* cause the economic problems, and how? How much the economic value that the invasion cost? Is the control worthwhile?
- What is the appropriate control strategy? What is/are the effective biocide and pest which will be adopted and how much for each infected areas? How the biocide and pest influence on the native community and ecosystem?
- What are the consequences of the control? The monitoring should be done continuously after the treatment.

Furthermore there is still the taxonomic ambiguity in the family Dreissenidae. Up to now the identity of the species that have invaded and spread in the Indo-Pacific region is uncertain. The revision of this taxon is urgently needed because the identification of the species is the first step to know whether that species is the introduced species and subsequently the proper management can be performed.

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Appendix

Publication

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Special issue "Invasive Aquatic Molluscs – ICAIS 2007 Conference Papers and Additional Records"
Frances E. Lucy and Thaddeus K. Graczyk (Guest Editors)

Research article

The occurrence of an invasive alien mussel *Mytilopsis adamsi* Morrison, 1946 (Bivalvia: Dreissenidae) in estuaries and lagoons of the lower south of the Gulf of Thailand with comments on their establishment

Kringpaka Wangkulangkul and Vachira Lheknim*
Department of Biology, Faculty of Science, Prince of Songkla University, Songkhla 90112, Thailand
**Corresponding author*
E-mail: vachira.l@psu.ac.th

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Abstract

The invasive false mussel, *Mytilopsis adamsi* Morrison, 1946 (Bivalvia: Dreissenidae), is a brackish water bivalve, native to tropical West Pacific coast of central America. The species has now become established in East Asian, South Asian and Southeast Asian countries. Species spread has been especially rapid with its distribution now including the lower part of the Gulf of Thailand. This is the first report of the establishment of this species in the lower part of the Gulf of Thailand, in Haad-kaew Lagoon and Thale Sap Songkhla, in Songkhla province, and the Pak Phanang Estuary in Nakhon Si Thammarat province, south Thailand. Descriptions of its morphology are consistent with previous descriptions from other areas as *M. adamsi*. Based on the available evidence, it is postulated that the species was transported to the areas between the year 1990 and 2000 via international commercial cargo ships. These findings indicate that the spread of *M. adamsi* is still in progress and that this invasive mussel continues to colonize the Songkhla Lagoon System.

Key words: establishment, *Mytilopsis*, Pak Phanang Estuary, south Thailand, Thale Sap Songkhla, Songkhla Lagoon System

Introduction

During a study of temporal and spatial variations of intertidal gastropods in the Haad-kaew Lagoon, Thailand between 2001 and 2003, voucher specimens of both the collected gastropods and bivalves were deposited in the Prince of Songkla University Zoological Collection. In 2006, Markus Huber, a conchologist from the Zoological Museum of the University of Zurich, Switzerland, visited and studied the bivalve specimens deposited in our collection and recognized that one of the unidentified mussels was a member of the genus *Mytilopsis* (Mollusca: Bivalvia: Dreissenidae), an alien invasive bivalve that had not been previously reported in Thailand. After comparison with other materials and type photos it was determined that the mussel was *Mytilopsis adamsi* Morrison, 1946. This was confirmed by Dan Marelli (pers. com. 12/07). *M. adamsi* was originally described from Panama Bay, Pearl Islands. It is a brackish species that invaded the Indo-Pacific Ocean during the 19th century and has reached Fiji (described as *M. allyneana* Hertlein and Hanna 1949), India, Malaysia, Singapore, Taiwan, and Australia (Marelli and Gray 1985; Morton 1989; Willan et al. 2000; Tan and Morton 2006). Whereas most authors report this species as *M. sallei*, Marelli and Gray (1985) synonymized *M. allyneana* with *adamsi* and considered *M. sallei* as distinct. It is quite likely however that all Asian reports of *M. sallei* are instead referable as *M. adamsi* (Dan Marelli, pers. com. 12/07).

Recently, *M. adamsi* has been observed in the Haad-kaew Lagoon and Thale Sap Songkhla in Songkhla province, and it has also presented in the Pak Phanang Estuary in Nakhon Si Thammarat province. As these mussels have frequently been reported to be alien invasive species (Kalyanasundaram 1975; Rao et al. 1989; Nalepa and Schloesser 1992; Branch and Steffani 2004; Bownes and McQuaid 2006; Tan and Morton 2006), the present report substantially expands their known range and also suggests an important impact in the local community they invade. The purpose of this paper is to report the occurrence of *M. adamsi* in the lower part of the Gulf of Thailand and to provide in addition evidence of the establishment of *M. adamsi* at these locations.

Material and Methods

Study sites

The Haad-kaew Lagoon is a relatively small shallow coastal lagoon, (3 km^2, 7°14'14.35"N, 100°33'47.04"E), adjacent to the Songkhla Lagoon System, in Songkhla Province (Figure 1). At present, the area can be divided into two parts, according to its morphology, a seasonally-

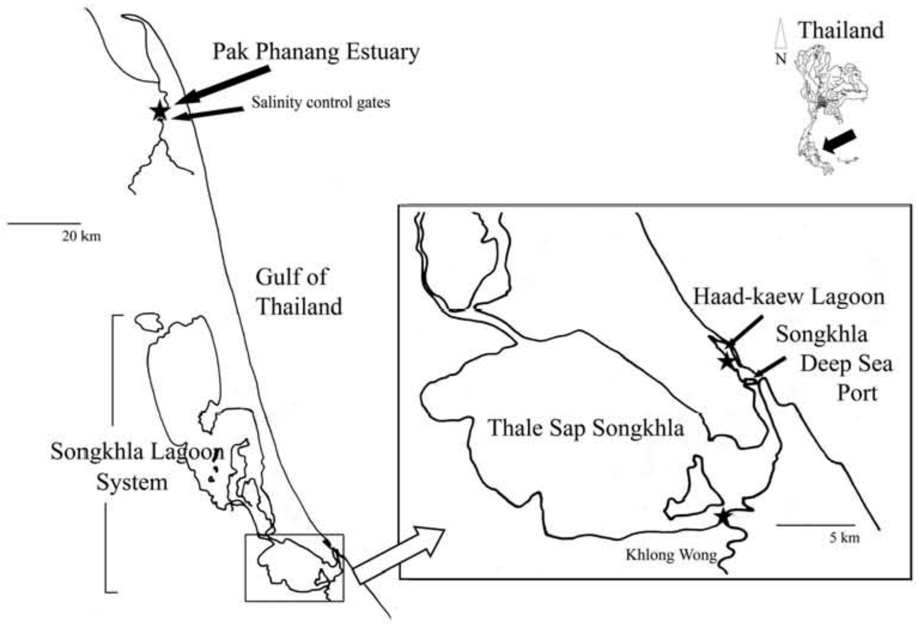

Figure 1. Part of the lower Gulf of Thailand in south Thailand showing the location of the Songkhla Lagoon System and Pak Phanang Estuary and sites of a sampling (indicated by★). Inset: the Thale Sap Songkhla and Haad-kaew Lagoon, showing the Songkhla Deep Sea Port and sampling sites of *Mytilopsis adamsi* Morrison, 1946, in this report (indicated by ★).

closed and an open lagoon. The system has an average mixed tidal amplitude of 0.8 m, an average depth of 2.0 m, with a substratum of muddy sand. Environmental parameters of the lagoon, especially salinity, change according to the season and distance from the open end (Lheknim and Leelawathanagoon 2004). In general, the salinity gradient changes from a polyhaline (18-30 psu) interior to the marine exterior. At one end the lagoon opens into the mouth of the Songkhla lagoon system near to where the Songkhla Deep Sea Port has been constructed.

Thale Sap Songkhla, (176 km^2, 7°10'34.40"N, 100°34'00"E), is the lowermost part of the Songkhla Lagoon System (Figure 1). The lower southeastern end of the Thale Sap Songkhla is connected to the lower part of the Gulf of Thailand by a narrow channel, allowing tides to inundate this lower region. In general, the salinity of the water in the Thale Sap Songkhla ranges from brackish to seawater depending on the season, with a mean water depth of 1.2 m in a relatively dry season, but the water level rises by about one meter during the northeast monsoon season (November-January). The bottom is muddy sand with debris. The Thale Sap Songkhla is known for the natural catches, and is also intensively utilized for the aquaculture.

The Pak Phanang Estuary (126 km^2, 8°23' 58.10"N, 100°08'54.75"E), is a part of the Pak Phanang River system and located in Nakhon Si Thammarat Province, southeast Thailand (Figure 1). The system has an average depth of 2.0 m. The substratum is muddy sand with debris. The eastern half of the estuary is occupied by a mangrove forest, several tidal creeks and an extensive mud flat (about 1–3 km wide) that emerges at low tide. The salinity in this river usually decreases with distance from the mouth of the river. Salinity control gates were constructed in the lower reach of the river in October 2003, to regulate the water level and salinity. As a result the region of the river above the control gates has become permanently freshwater while the salinity below the control gates is permanently brackish or seawater all the year round. The Pak Phanang Estuary is also well-known for its natural production of fish, shrimp and crabs.

Sampling of mussels and data analysis

The identification of mussels was based on characteristics presented in Marelli and Gray (1985). Haad-kaew Lagoon, especially the sesonally-closed sector, was visited several times for settlement observations and the mussel specimens were taken from this sector to the laboratory twice, in July 8th, 2006 and June 9th, 2007. Mussel sampling was conducted in the Pak Phanang Estuary adjacent to the salinity control gates and at Thale Sap Songkhla near to the mouth of Khlong Wong on March 31st, 2007 and June 9th, 2007, respectively. During each visit, samples were collected using a 15x15 cm^2 quadrat randomly placed on the substratum about 0.5 m in depth and all mussels from the quadrat were removed. During sampling, salinities at these sites were recorded by using a hand-held refractometer (except on July 2006 in Haad-kaew Lagoon). In the laboratory, shell lengths of the right valve of all the collected specimens were measured to the nearest 0.1 millimeters by using calipers. A shell length frequency distribution of *M. adamsi* from each sampling site was separately constructed and cohort analysis was made by means of Bhattacharya's method, available in FISAT II (FAO 2005), and used to distinguish existing cohorts at each location.

Results and Discussion

The salinities from the Haad-kaew Lagoon, Thale Sap Songkhla and Pak Phanang River (area around the water gate) taken at the times of sampling were 20, 6 and 31 psu, respectively. *M. adamsi* bivalves were observed on the substratum of the whole seasonally-closed portion of the Haad-kaew Lagoon as well as some attached on boat piers and other hard substrata, while densely populated colonies occurred on aquaculture cages in the permanent-ly open lagoon (Figure 2). In Thale Sap Songkhla, colonies of *M. adamsi* were byssally attached to mangrove root systems. In Pak Phanang Estuary, a densely populated colony of *M. adamsi* was found attached to the cement of the water control gate at a depth of 0.5 m. The results from these preliminary observations have indicated that *M. adamsi* is able to survive under brackish water conditions and in a wide range of salinities from 6 to 31 psu, as an euryhaline species. It is interesting to note that *M. adamsi* has an ability to byssally colonize many surfaces, ranging from muddy sand substratum to other submerged materials, such as shells of other molluscs, plastic bags, buoy lines, fish cages and fishing gears. Attachment at many of these sites can rapidly cause changes in local communities, because this mussel generally produces a dense monoculture that excludes other sessile organisms. The mussel subsequently provides a new habitat for associated fauna, such as amphipods and polychaetes to colonize, and probably results in a significant change in the community structure in the future as found in dreissenids and mytilids (Nalepa and Schloesser 1992; Crooks and Khim 1999; People 2006; Borthagaray and Carranza 2007).

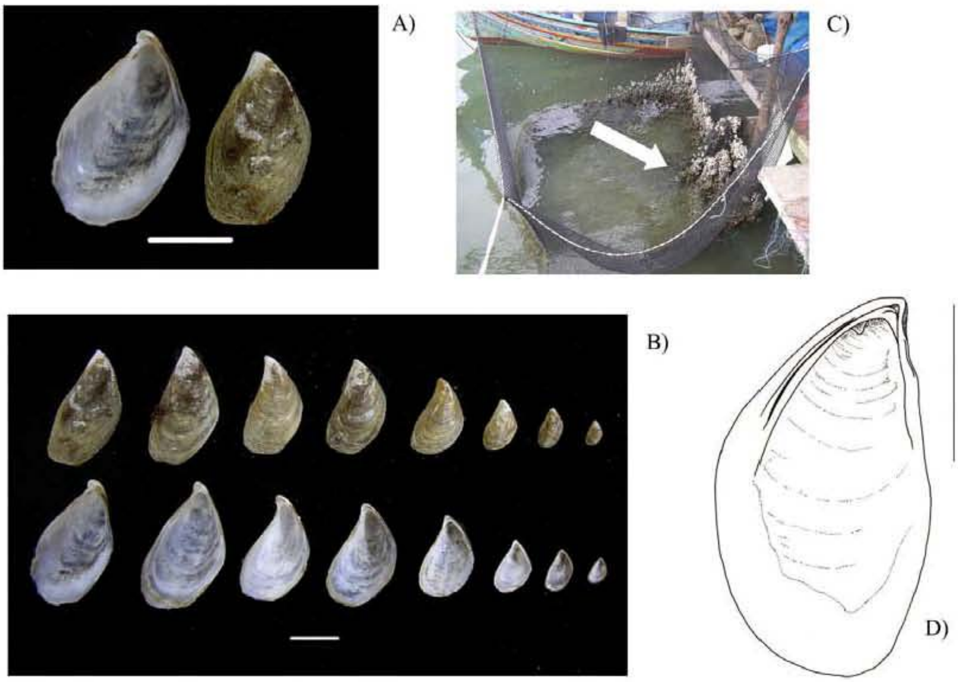

Figure 2. External morphology of *Mytilopsis adamsi* Morrison, 1946 scale bar 1 cm.
A) Right and left valve of *M. adamsi*;
B) Variability of shell shape of *M. adamsi*;
C) A dense colony of *M. adamsi* on the fish cages in Haad-kaew Lagoon, and
D) Interior view of the left valve of *M. adamsi*
(Photographs by Kringpaka Wangkulangkul)

Establishment and recruitment of M. adamsi

The histograms of the length-frequency distribution for all samples from all sampling locations, at Haad-kaew Lagoon, Thale Sap Songkhla and Pak Phanang River (Figures 3A, 3B, 3C and 3D), indicate multiple consecutive cohorts. The population size structure for July 2006 and June 2007 in the temporarily closed portion of Haad-kaew Lagoon (Figures 3A and 3B), shows a similar population structure, indicating that *M. adamsi* is able to establish self-sustaining populations with a tendency to reproduce asynchronously in a continuous spawning season. Interestingly, the recruitment cohorts from the seasonally-closed portion of Haad-kaew Lagoon and the Thale Sap Songkhla reveal a series of regular successive cohorts (Figures 3A, 3B and 3C respectively), while large size class and low successive recruitment is displayed at the mouth of Pak Phanang River (Figure 3D). Such patterns of recruitments may result from only small gradual changes in brackish water conditions within the seasonally-closed portion of Haad-kaew Lagoon and the Thale Sap Songkhla as a result of reduced tidal influence, while the strong tidal effects of seawater with a relatively low freshwater supply were observed at the mouth of Pak Phanang River. Previously, NIMPIS (2002) has suggested that spawning of *M. sallei* may be triggered by changes in salinity. The development of fertilized eggs were predominant in a salinity below 25 psu, allowing for the development of the trochophore and veliger larvae while advanced stages were not observed in salinities higher than

25 psu (Raju et al. 1975). The population structure from the Haad-kaew Lagoon and the Thale Sap Songkhla (Figures 3A, 3b and 3C respectively), reveal that the majority of recruitments seemed to have occurred before the sampling dates i.e., before June 2006 and July 2007 (this phenomena is being investigated in the field, unpublished data showed that the peak of recruitment occurred in February). They probably followed by the time of the peak salinity, usually around August and September at the eastern coast of south Thailand, which may prohibit fertilization of *M. adamsi*. Likewise, the population structure from the mouth of Pak Phanang River with very few specimens smaller than 15.3 mm implies that the higher salinity on the sampling period, (30 psu on the 31st March 2007), reduced the survival of the smaller specimens or inhibited spawning.

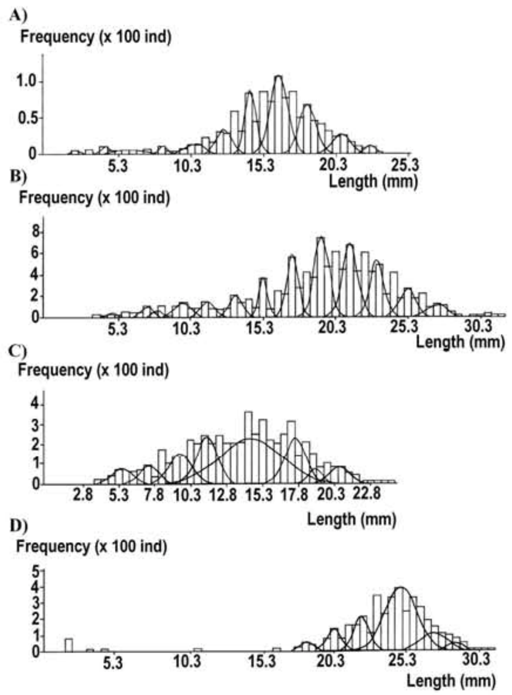

Figure 3. Length frequency distribution histogram of *Mytilopsis adamsi* Morrison, 1946. A) - specimens from Haad-kaew Lagoon in south Thailand on the July 8th, 2006 (n= 1185); B) - specimens from Haad-kaew Lagoon in south Thailand on the June 9th, 2007 (n=1104); C) - specimens obtained from the Thale Sap Songkhla (mouth of Khlong Wong) in south Thailand on the June 9th, 2007 (n=545) and D) - specimens collected from Pak Phanang River (salinity control gates) in south Thailand on the March 31st, 2007 (n= 405).

Hypothetical sources in south Thailand

The incidence of the mussel *M. adamsi* in south Thailand was probably the result of the accidental introduction of the species by international ships traveling to Hong Kong and Singapore between the 1980s and 1990s (Tan and Morton 2006), then subsequently to ports in the Gulf of Thailand. In an early study of Marine Lamellibranchiata in the Gulf of Thailand (Lynge 1909) and later in the Fauna of Thailand (Suvatti 1967), there is no mention *M. adamsi*. Recently, Swennen et al. (2001) reported the mussel at Panarae, Pattani Province, approximately 100 km south of Songkhla and identified it as *Mytilopsis adamsi* Morrison. This indicates that *M. adamsi* was possibly transported to Haad-kaew Lagoon and Thale Sap Songkhla between the years 1990 and 2000 via international commercial cargo ships operating between the Songkhla Deepwater Sea Port (see also Figure 1) and other regional ports of Hong Kong and Singapore. It is probable that the same method of introduction occurred in the Pak Phanang River, as there are several local shipyards and dock facilities located along the river. In conclusion, the most likely routes of introduction *M. adamsi* to these areas are believed to be from larvae released in discharged ballast water or adults attached to a boat or ship's hull, rather than to a natural dispersal of the species.

Conclusion

Although the presence of *M. adamsi* has been recorded in the south of Thailand since 2001 (Swennen et al. 2001), this is the first report of its widespread distribution and abundance in different habitats. It presently well established in the Haad-kaew Lagoon and continues into the lower part of the Songkhla Lagoon System and possibly also in the Pak Phanang Estuary. There are several passive dispersal routes upstream in the above mentioned systems, for example, by transportation on fishing and recreation boats and may be even by waterfowl. Further research on biology, potential impacts and control of this species is required for neighbouring coastal habitats, particularly in the Thale Sap and Thale Sap Songkhla of the Songkhla Lagoon System, due to its potential negative ecosystem impacts, especially in terms of economic damage to local communities.

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VITAE

Name Miss Kringpaka Wangkulangkul

Student ID 4910220012

Education al Attainment

Degree	Name of Institution	Year of Graduation
B. Sc. (Biology)	Prince of Songkla University	2006

Scholarship Awards during Enrolment

- TRF/BRT Special program for Biodiversity Research and training grant BRT T_351007
- Research assistantship from Faculty of Science Prince of Songkla University 2009
- Association for Tropical Biology & Conservation, Asia-Pacific Chapter (ATBC) supports for participating in the conference of the Association for Tropical Biology & Conservation, Asia-Pacific Chapter hosted by King Mongkut's University of Technology Thonburi in the title "Assessing and Restoring Biodiversity in a Human Dominated Landscape" at Imperial Mae Ping Hotel, Chiang Mai, Thailand on 12 to 15 February 2009

List of Publication and Proceedings

- Wangkulangkul, K. and Lheknim, V. 2008. The occurrence of an invasive alien mussel *Mytilopsis adamsi* Morrison, 1946 (Bivalvia: Dreissenidae) in estuaries and lagoons of the lower south of the Gulf of Thailand with comments on their establishment. Aquatic Invasions. Special Issue: “Invasive Aquatic Molluscs – ICAIS 2007 Conference Papers and Additional Records 3(3): 325-330.
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- Wangkulangkul, K. and Lheknim, V. Spatial and temporal variability in recruitment of the invasive mussel *Mytilopsis adamsi* Morrison, 1946 and some environmental characteristics in Haad-kaew Lagoon, in the lower south of the Gulf of Thailand. (In preparation)