REPRODUCTIVE PATTERN AND GUT CONTENT ANALYSIS OF ASIATIC HARD CLAM *Meretrix meretrix* (Linnaeus, 1758) IN MARUDU BAY, MALAYSIA

VIENNA ANASTASIA ADMODISASTRO

BORNEO MARINE RESEARCH INSTITUTE
UNIVERSITI MALAYSIA SABAH
2023

REPRODUCTIVE PATTERN AND GUT CONTENT ANALYSIS OF ASIATIC HARD CLAM *Meretrix meretrix* (Linnaeus, 1758) IN MARUDU BAY, MALAYSIA

VIENNA ANASTASIA ADMODISASTRO

THESIS SUBMITTED IN FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF SCIENCE

BORNEO MARINE RESEARCH INSTITUTE
UNIVERSITI MALAYSIA SABAH
2023

UNIVERSITI MALAYSIA SABAH

BORANG PENGESAHAN STATUS TESIS

JUDUL : **REPRODUCTIVE PATTERN AND GUT CONTENT ANALYSIS OF ASIATIC HARD CLAM *MERETRIX MERETRIX* (LINNAEUS, 1758) IN MARUDU BAY, MALAYSIA**

IJAZAH : **SARJANA SAINS**

BIDANG : **SAINS MARIN**

Saya **VIENNA ANASTASIA ADMODISASTRO**, Sesi **2019-2023**, mengaku membenarkan tesis Sarjana ini disimpan di Perpustakaan Universiti Malaysia Sabah dengan syarat-syarat kegunaan seperti berikut:-

1. Tesis ini adalah hak milik Universiti Malaysia Sabah
2. Perpustakaan Universiti Malaysia Sabah dibenarkan membuat salinan untuk tujuan pengajian sahaja.
3. Perpustakaan dibenarkan membuat salinan tesis ini sebagai bahan pertukaran antara institusi pengajian tinggi.
4. Sila tandakan (/):

☐	SULIT	(Mengandungi maklumat yang berdarjah keselamatan atau kepentingan Malaysia seperti yang termaktub di dalam AKTA RAHSIA 1972)
☐	TERHAD	(Mengandungi maklumat TERHAD yang telah ditentukan oleh organisasi/badan di mana penyelidikan dijalankan)
☐	TIDAK TERHAD	

Disahkan Oleh,

ANITA BINTI ARSAD
PUSTAKAWAN KANAN
UNIVERSITI MALAYSIA SABAH

VIENNA ANASTASIA ADMODISASTRO
MY1821017T

(Tandatangan Pustakawan)

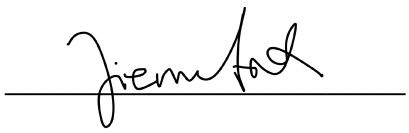
Tarikh : 14 Jun 2023

(Prof. Madya Dr. Julian Ransangan)
Penyelia Utama

DECLARATION

I hereby declare that the material in this thesis is my own except for quotations, excerpts, equations, summaries, and references, which have been duly acknowledged.

14 February 2023



Vienna Anastasia Admodisastro
MY1821017T

CERTIFICATION

NAME	:	**VIENNA ANASTASIA ADMODISASTRO**
MATRIC NUM.	:	**MY1821017T**
TITLE	:	**REPRODUCTIVE PATTERN AND GUT CONTENT ANALYSIS OF ASIATIC HARD CLAM *Meretrix meretrix* (Linnaeus, 1758) IN MARUDU BAY, MALAYSIA**
DEGREE	:	**MASTER OF SCIENCE**
FIELD	:	**MARINE SCIENCE**
VIVA DATE	:	**14 FEBRUARY 2023**

CERTIFIED BY;

Signature

SUPERVISOR
Assoc. Prof. Dr. Julian Ransangan

ACKNOWLEDGEMENT

Praises to Allah, the Almighty, for His countless blessings throughout my study tenure to complete this piece of important research work successfully. I would like to express my deep and sincere gratitude to my research supervisor, Associate Professor Dr. Julian Ransangan for giving me this opportunity and providing invaluable guidance throughout this research. It was a great privilege and honor to work and study under his guidance. I would also like to thank him sincerely for his motivation, support, friendship and empathy. I am very thankful to my family for their love and prayers throughout my study. I am extremely grateful to my mother Mainah Macartney and my sister Novia Indriaty for their endless love, support, motivation, and patience. Thank you for always believing in me. A special thanks to Prof. Dr. Aileen Tan for her inspiration and motivation. Thank you also to Gan Sze Hoon, Vanessa Rose Marie Stephen, Marina William, Christie Tahis, Aisah Dasuki, Nina Ho, Abby Zainal, Alexander Ho, Eva Francisco, and Leonard Lai for their constant encouragement and support. I would like to express my gratitude to my research colleagues and friends, Joanna W. Doinsing, Syahneiza Zahurin, Laditah Duisan, Mok Waiyee, and Nabilah Sieha for making this journey smoother and memorable. And to Ketot, Hitam, Blackie and Whitey for their affection and warmth. Last but not least, thanks to the support staff of the Borneo Marine Research Institute; Jeffry Molius, Yusdi Ismail, Veronica Ginus, Ronald Phan, Syafikah Usop, Herman@ Mohd Nazrie Bin Musanna, and Mail Kahar for their assistance. Finally, I would also like to thank the Ministry of Higher Education Malaysia for approving the Fundamental Research Grant Scheme (FRGS/1/2017/WAB01/UMS/02/4) to financially support this study.

Vienna Anastasia Admodisastro
14 February 2023

ABSTRACT

Asiatic hard clam, *Meretrix meretrix* is the indigenous bivalve species found in Sabah coastal areas and is an important species for artisanal fisheries, especially in Marudu Bay, Sabah. The present study was carried out to understand some aspects of reproductive biology such as sex ratio, stages of gametogenesis, temporal variation in the condition and gonad index of the clam, and its relationship to the environmental parameters of its habitat. This study was also conducted to determine the main diet of the clam and the composition of food particles in the clam's gut in comparison to the food particles found in their habitat and the environmental parameters that could affect and influence the clam's feeding selection. Sampling was carried out once a month for 10 months (May 2019 to February 2020) in Marudu Bay. Clam samples, environmental parameters, and water samples (for seston, chlorophyll-*a*, water quality and nutrients analyses, phytoplankton, and zooplankton) were collected at the sampling areas during the study period. Clam samples were collected using clam fishing gear called the 'kerek' from the substrate during low tide. A total of 750 clam samples were used for condition index, histological, and gut content analyses. Results showed that the clam is dioecious and its population in the bay was slightly skewed toward female clams (1.058:1) with the occurrence of hermaphrodite at an extremely low frequency (0.4%). The clam was noted to spawn throughout the year with two spawning peaks (June to July and November to December). There was a significant difference ($p<0.05$) in the condition index between the months. However, there was no significant difference ($p>0.05$) observed in the gonad index between the months and between the male and female gonad index. The condition index of the clam in the bay fell into the moderate fatness category and showed correlations with various environmental variables while the gonad index only showed a correlation with total rainfall. As for the gut content analysis, a total of 1475 food particles were found in the gut of the clams. Phytoplankton and zooplankton represented about 80.9% and 19.1% of the total food particles found in the gut of the clam, respectively. Meanwhile, diatoms were the most common food particles ingested by the clams with 95.81% and dinoflagellates only contributed about 4.19% of the total phytoplankton ingested by the clams. The five most dominant phytoplankton genera found in the gut of the clam throughout the study period are *Nitzschia* (24.71%), *Coscinodiscus* (23.38%), *Cyclotella* (13.70%), *Pleurosigma* (12.68%) and *Navicula* (8.95%). The dendrogram of similarity showed that the phytoplankton compositions of the water sample and in the gut were grouped apart. The result of One-way ANOSIM analysis comparing phytoplankton composition in gut and water samples indicated significant differences for all the months with an overall average R: 0.717, $p<0.001$. In general, the similarity between phytoplankton in the water sample and the gut was less than 50% and two clusters can be depicted. Cluster 1 had a similarity of less than 40% and Cluster 2 had a similarity between 40-50%. Cluster 1 (September 2019) showed significant differences between phytoplankton in the water sample and in the gut with R: 0.924, $p<0.008$. Meanwhile, all months except September 2019 were grouped in Cluster 2 with R: 0.788, $p<0.008$. The result from PERMANOVA following distance based linear modeling (DistLM) demonstrated that phytoplankton cell density, phytoplankton diversity, chlorophyll-*a*, and salinity were significantly ($p<0.05$) influencing the selective feeding behavior of the clam. The clam

population in Marudu Bay is balanced with a reproductive pattern that is active all year round and has short resting and spent stages like most tropical bivalves due to the high annual temperature and constant food supply throughout the year. The clam showed obvious selective feeding behavior where more benthic diatoms were selected as food due to the nutritional content, quantity, and availability of benthic diatoms in the clam's natural habitat. Although various environmental parameters influence the clam in various aspects, it was discovered that salinity is an important parameter influencing not only the condition index and gonad index but also the clam's selective feeding behavior. Drastic changes in salinity affect the condition and trigger the spawning of the clam. Meanwhile, at certain salinities, the feeding physiology of the clam, such as filtration and ingestion rate, reaches an optimum range and the clams actively filter food from the water column, making their selective feeding behavior more efficient.

ABSTRAK

POLA REPRODUKTIF DAN ANALISIS KANDUNGAN PERUT KERANG ASIATIK Meretrix meretrix (Linnaeus, 1758) DI TELUK MARUDU, MALAYSIA

Meretrix meretrix ialah spesies bivalvia yang terdapat di kawasan persisiran pantai Sabah dan merupakan spesies penting dalam perikanan kecil-kecilan terutamanya di Teluk Marudu, Sabah. Kajian ini dijalankan untuk memahami beberapa aspek biologi pembiakan kerang seperti nisbah jantina, peringkat gametogenesis, variasi temporal indeks kondisi dan gonad kerang, serta korelasinya dengan parameter persekitaran habitatnya. Kajian ini juga dijalankan untuk menentukan diet utama kerang dan komposisi partikel makanan dalam perut kerang berbanding dengan partikel makanan yang terdapat di habitatnya serta parameter persekitaran yang boleh mempengaruhi pemilihan makanan kerang. Persampelan dijalankan sebulan sekali selama 10 bulan (Mei 2019 hingga Februari 2020) di Teluk Marudu. Sampel kerang, parameter persekitaran, dan sampel air (untuk analisis seston, klorofil-a, analisis kualiti dan nutrien air, fitoplankton dan zooplankton) diambil di kawasan persampelan sepanjang tempoh kajian. Sampel kerang diambil dari substrat mengunakan kerek iaitu sejenis alat mencari kerang semasa air surut. Sebanyak 750 sampel kerang digunakan untuk analisis indeks kondisi, histologi dan kandungan perut. Keputusan kajian menunjukkan kerang adalah dioecious dan populasi kerang adalah pencong sedikit ke arah kerang betina (1.058:1) dengan kehadiran hermafrodit pada frekuensi yang sangat rendah (0.4%). Kerang didapati menjalankan pemijahan sepanjang tahun dengan dua puncak utama (Jun hingga Julai dan November hingga Disember). Terdapat perbezaan signifikan ($p<0.05$) dalam indeks kondisi kerang di antara bulan. Walau bagaimanapun, tiada perbezaan signifikan ($p>0.05$) yang diperhatikan di antara indeks gonad setiap bulan dan di antara indeks gonad jantan dan betina. Indeks kondisi kerang di kawasan kajian berada dalam kategori sederhana gemuk dan menunjukan korelasi signifikan terhadap pelbagai faktor persekitaran manakala indeks gonad pula didapati hanya menunjukkan korelasi signifikan terhadap jumlah taburan hujan. Untuk analisis kandungan perut pula, sebanyak 1475 partikel makanan dijumpai di dalam perut kerang. Fitoplankton dan zooplankton masing-masing mewakili sebanyak 80.9% dan 19.1% dari jumlah keseluruhan partikel makanan yang dijumpai di dalam perut kerang. Sementara itu, diatom merupakan makanan dominan yang dimakan oleh kerang dengan 95.81% dan dinoflagelat pula hanya menyumbang 4.19% daripada jumlah keseluruhan fitoplankton yang dimakan oleh kerang. Lima jenis genera fitoplankton yang paling dominan dijumpai di dalam perut kerang sepanjang tempoh kajian adalah Nitzschia (24.71%), Coscinodiscus (23.38%), Cyclotella (13.70%), Pleurosigma (12.68%) dan Navicula (8.95%). Dendrogram persamaan menunjukkan bahawa komposisi fitoplankton dalam sampel kandungan air sekitar dan perut dikelompokkan secara berasingan. Keputusan analisis One-way ANOSIM yang membandingkan komposisi fitoplankton dalam sampel kandungan perut dan air sekitar menunjukkan perbezaan signifikan setiap bulan dengan purata keseluruhan $R: 0.717$, $p<0.001$. Secara amnya, persamaan antara sampel kandungan air sekitar dan perut adalah kurang daripada 50% dan dua kluster utama boleh dikenalpasti. Kluster 1 dengan persamaan kurang daripada 40% dan Kluster 2 dengan persamaan antara 40-50%. Kluster 1 (September 2019) menunjukkan perbezaan signifikan antara sampel kandungan air

sekitar dan perut dengan R: 0.924, p<0.008. Sementara itu, semua bulan kecuali September 2019 berada di dalam Kluster 2 dengan R: 0.788, p<0.008. Keputusan dari PERMANOVA dalam pemodelan linear berasaskan jarak (DistLM) menunjukkan bahawa kepadatan sel fitoplankton, indeks diversiti fitoplankton, klorofil-a dan saliniti secara signifikan (p<0.05) mempengaruhi tingkah laku pemakanan terpilih kerang. Populasi kerang di Teluk Marudu adalah seimbang dengan corak pembiakan yang aktif sepanjang tahun dan mempunyai peringkat 'resting' dan 'spent' yang singkat seperti kebanyakan bivalvia tropika yang lain disebabkan suhu tahunan yang tinggi dan bekalan makanan yang berterusan sepanjang tahun. Kerang menunjukkan tingkah laku pemakanan terpilih yang jelas dimana lebih banyak diatom bentik dipilih sebagai makanan disebabkan kandungan nutrisi, kuantiti, dan ketersediaan bentik diatom di habitat semula jadi kerang. Walaupun pelbagai parameter persekitaran mempengaruhi kerang dalam pelbagai aspek, saliniti merupakan parameter penting yang mempengaruhi bukan sahaja indeks kondisi dan indeks gonad tetapi juga tingkah laku pemakanan terpilih kerang. Perubahan drastik dalam saliniti mempengaruhi kondisi dan mencetuskan pemijahan kerang. Manakala pada saliniti tertentu, fisiologi pemakanan kerang, seperti kadar penapisan dan pengingesan, mencapai kadar optimum dan kerang secara aktif menapis makanan dari kolum air dan tingkahlaku pemakanan terpilih kerang menjadi lebih cekap.

LIST OF CONTENTS

CHAPTER 4 : RESULTS

CHAPTER 5 : DISCUSSION

LIST OF TABLES

LIST OF FIGURES

LIST OF SYMBOLS AND UNITS

<	-	smaller than
>	-	bigger than
^{o}C	-	Degree Celcius
%	-	Percentage
µL	-	Microliter
µm	-	Micrometer
m	-	Meter
cm	-	Centimeter
mm	-	Milimeter
L	-	Liter
ml	-	Milliliter
mg/l	-	Milligram per liter
µg/l	-	Microgram per liter
cells/ml	-	Cells per milliliter
m^3	-	Cubic meter
ppt	-	part per thousand

LIST OF ABBREVIATIONS

GF/C	-	Glass microfiber filters
HSD	-	Honest significant difference
TPM	-	Total Particulate matter
POM	-	Particulate organic matter
PIM	-	Particulate inorganic matter
OC	-	Organic content
WMW	-	Wet meat weight
DMW	-	Dried meat weight
ANOVA		Analysis of variance
HSD	-	Honestly significant difference
ANOSIM	-	Analysis of similarities
SIMPER	-	Similarity percentage analysis
PERMANOVA	-	Permutational multivariate analysis of variances
DistLM	**-**	Distance-based linear modeling

LIST OF APPENDICES

CHAPTER 1

INTRODUCTION

1.1 Asiatic Hard Clam, *Meretrix meretrix*

Asiatic hard clam, *Meretrix meretrix* is commonly found buried in muddy-sand bottoms of intertidal and sublittoral waters to a depth of about 20m. *M. meretrix* distribution is widespread in the Indo-West Pacific, from East Africa to the Philippines, north to Japan, and south to Indonesia (Poutiers, 1998). Besides *M. meretrix (*Linnaeus, 1758*)*, the genus *Meretrix* also consists of 8 other generally recognized species including *M. casta* (Chemnitz, 1782), *M. lusoria* (Roding, 1798), *M. lyrata* (Sowerby, 1851), *M. lamarckii* (Gray, 1853), *M. petechialis* (Lamarck, 1818)*, M. planisulcata* (Sowerby, 1851), *M. attenuate* (Dunker, 1862) *and M. ovum* (Hanley, 1845) (OBIS Indo-Pacific Molluscan Database, 2006). In general, *M. meretrix* taxonomy is classified as below.

Phylum: Mollusca
Class: Bivalvia
Sub-class: Autobranchia
Superorder: Imparidentia
Order: Venerida
Super Family: Veneroidea
Family: Veneridae (Gray)
Subfamily: Meretricinae
Genus: *Meretrix*
Species: *meretrix*
Scientific name: *Meretrix meretrix* (Linnaeus, 1758)

Meretrix meretrix is widely distributed along the coastal and estuarine areas in China, Korea, Vietnam, and India (Jayabal & Kalyani, 1986; Wang *et al*., 1993; Chowdhury *et al.,* 2019). The clam is widely cultured in countries like China (Tang *et al.,* 2006; Huang *et al*., 2016) to support the consumption demand. In Vietnam, the main contributor to the total production of this species is from the northern coastal provinces, and the clam is one of the indigenous mollusks in the region (Phuang *et al*., 2001). In Malaysia, *M. meretrix* can be found in Sarawak as mentioned in Hamli *et al*. (2012). Besides that, *M. meretrix* is reportedly found in Pahang (Abdul Halim *et al.,* 2018), Johor (Azmi *et al.,* 2014), and Penang (Abdul Halim *et al.,* 2019). The clam is also commonly distributed throughout Sabah coastal areas and estuaries. In the West Coast Sabah, the clam inhabited the rural estuary in Kota Belud and the urban estuary in Kota Kinabalu (Abdullah *et al.,* 2007). According to Tan *et al*. (2017), *M. meretrix* is the most dominant species in the sandy shoreline of Marudu Bay, Northeast Coast Sabah. While in the East Coast Sabah, the clam can be found in Tawau (Mohd Hamdan *et al.,* 2017) and is one of the species collected by the villagers from the fishing villages in Semporna as reported by Ridzwan and Kaswandi (1995).

Meretrix meretrix is a commercially important species in coastal areas of South and Southeast Asia (Liu *et al*., 2006). The community favours the clam for its good taste and high nutritional content including protein (9.39%), Eicosapentaenoic Acid, EPA (2.03%), and Docosahexaenoic Acid, DHA (6.06%) (Gifari, 2011). Besides the high protein content, *M. meretrix* is also reported to have valuable medical properties such as antioxidants, antitumors, to reduce swelling, detoxification, and other functions (Xie *et al.,* 2012). In recent years, many bioactive components such as peptides, enzymes, and enzyme inhibitors have been identified and purified from the clam which is responsible for its nutritional and medicinal functions (Xie *et al*., 2012).

Besides that, *M. meretrix* is also used as a bioindicator. They are filter feeders and thus able to accumulate toxic substances from water and sediment due to its high bioconcentration factors (BCFs) values especially for Cadmium and Zinc (Abdullah *et al*., 2007). Mollusks such as bivalves are good bioindicators of heavy metal pollution because they can collect the heavy metal elements in their tissue