EFFECT OF STOCKING DENSITY ON THE GROWTH PERFORMANCE OF RED TILAPIA AND WATER QUALITY IN ZEOLITE SUPPLEMENTED CLOSED SYSTEM

By

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THESIS DECLARATION

I declare that this thesis entitled "Effect of Stocking Density on the Growth Performance of Red Tilapia and Water Quality in Zeolite Supplemented Closed System" is the result of my own research except as cited in the references. The thesis has not been accepted for any degree and is not concurrently submitted in candidature of any other degree.

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LIST OF ABBREVIATIONS

ADWG	Average Daily Weight Gain
BOD	Biochemical Oxygen Demand
COD	Chemical Oxygen Demand
DO	Dissolved Oxygen
DWG	Daily Weight Gain
FAO	Food and Agriculture Organization
FCR	Feed Conversion Rate
GFSI	Global Food Security Index
LG	Length Gain
MANOVA	Multivariate Analysis of Variance
PH	Potential Hydrogen
SR	Survival Rate
SGR	Specific Growth Rate
TDS	Total Dissolved Solid
TAN	Total Ammonia Nitrogen
WG	Weight Gain

LIST OF SYMBOLS

%	Percentage
<	Less Than
℃	Celsius
℃F	Celsius Fahrenheit
Al	Aluminium
AlO_4^-	Alumina Ion
Al_2O_3	Aluminium Oxide
B	Boron
CO_2	Carbon Dioxide
Ca^{2+}	Calcium Ion
CaO	Calcium Oxide
Fe_2O_3	Iron (III) Oxide
g	Gram
g/L	Gram Per Liter
Ge	Germanium
K_2O	Potassium Oxide
K^+	Potassium Ion
L	Liter
Li^+	Lithium Ion
m^3	Cubic Meter
mg/ L	Milligram Per Liter
mL	Milliliter
Mg^{2+}	Magnesium Ion
MgO	Magnesium Oxide
N	Nitrogen
Na^+	Sodium Ion
Na_2O	Sodium Oxide
NH_3	Ammonia
$NH_3 - N$	Ammonia Nitrogen

$NO_2 - N$	Nitrite Nitrogen
$NO_3 - N$	Nitrate Nitrogen
NO_2^-	Nitrite Ion
NO_3^-	Nitrate Ion
NH_4^+	Ammonium Ion
Na^+	Sodium Ion
Si	Silicon
SiO_2	Silicon Dioxide
SiO_4	Silica
TiO_2	Titanium (IV) Oxide
TAN	Total Ammonia Nitrogen

Effect of Stocking Density on the Growth Performance of Red Tilapia and Water Quality in Zeolite Supplemented Closed System

ABSTRACT

Increasing in fish density in the culture systems increases the fish production; however the intensification in cage culture system resulted in massive mortality in fish. The main objective of this study is to determine the effect of stocking density, (5, 10, 15, 20, and 25 fry/aquarium) on water quality and growth parameters of fresh water aquarium fish, red tilapia, *Oreochromis niloticus*, at different stocking density supplemented with zeolite. This study also aimed to compare the water quality and growth performance of fish in treatments with and without zeolite. Red tilapia fry (12.90±0.01 g, 81.95±0.02 cm) were stocked into aquarium (40×20×20 cm). Five treatments (containing zeolite) with two replicates and one replicate (without zeolite) were used: T_1- 5, T_2- 10, T_3- 15, T_4- 20, and T_5- 25 fry/aquarium. Fish were fed twice a day with 2% of total biomass. The water quality parameters of each aquarium were monitored by using YSI model 556 multi-parameter and UV-VIS spectrophotometer. Weight and length of red tilapia was measured every two weeks by using electronic balance and vernier calipers. After 9 weeks, the zeolite treatments which recorded the highest final mean weight was T_1 (40.27±15.87) while in T_5 (17.26±0.15) lowest mean weight was recorded. Specific growth rate and length gain in T_1 (1.74%, 24.38 mm) was significantly ($p<0.05$) higher than other treatments. Based on the water quality recorded, significant differences ($p<0.05$) were found in all water quality parameters among treatments except salinity. On the other hand, there was no significant difference ($p>0.05$) in growth parameters between the treatments with and without zeolite. The water quality recorded in zeolite supplemented treatments was better than in control. The findings of this paper will be useful for the practitioners to understand the best practice for stocking density in zeolite supplemented closed system.

Kesan Ketumpatan Stok kepada Prestasi Pertumbahan Tilapia Merah dan Kualiti Air dalam Sistem Tertutup yang Mengandungi Zeolit

ABSTRAK

Pertambahan ketumpatan stok akan menambah pengeluaran ikan tetapi ketumpatan yang tinggi dalam sistem kultur akan menyebabkan kebanyakkan ikan mati. Objektif utama kajian ini adalah mengkaji kesan ketumpatan ikan (5, 10, 15, 20, dan 25ekor/akuarium) kepada kualiti air dan prestasi pertumbuhan tilapia merah dalam sistem tertutup yang mengandungi zeolit. Kajian ini juga dijalankan untuk membandingkan kualiti air dan prestasi pertumbuhan tilapia ikan antara rawatan air yang mengandungi dan tidak mengandungi zeolite. Tilapia merah (12.90±0.01 g, 81.95±0.02 cm) telah dimasukkan dalam dalam akuarium (40×20×20 cm). Lima rawatan yang mengandungi zeolite dengan dua ulangan dan lima rawatan yang tanpa mengandungi zeolite dengan satu ulangan telah dijalankan: T_1- 5, T_2- 10, T_3- 15, T_4- 20, and T_5- 25 ekor/akuarium. Tilapia diberi makan sebanyak dua kali sehari dengan 2% daripada jumlah biomas. Parameter kualiti air dalam setiap akuarium dipantau dengan menggunakan YSI 556 pelbagai parameter dan spektrofotometer UV-VIS. Berat dan panjang tilapia merah diukur setiap dua minggu dengan menggunakan keseimbangan elektronik dan caliper vernier. Selepas 9 minggu, antara rawatan yang mengandungi zeolite, T_1 (40.27±15.87) mencatatkan berat yang paling tinggi dan T_5 (17.26±0.15). Dari segi kadar pertumbuhan dan penambahan panjang, T_1 (1.74%, 24.38 mm) mempunyai perbezaan yang ketara dengan rawatan yang lain ($p<0.05$). Berdasarkan kualiti air yang direkodkan, semua parameter kualiti air mempunyai perbezaan yang ketara antara semua rawatan kecuali kemasinan air. Selain daripada itu, parameters pertumbuhan antara rawatan didapati tidak ada perbezaan yang ketara ($p>0.05$). Kualiti air dalam rawatan yang mengandungi zeolite lebih baik daripada yang tidak mengandungi zeolite. Penemuan kertas ini akan berguna bagi para pengamal untuk memahami amalan terbaik untuk menumpuk ketumpatan ikan dalam sistem tertutup yang mengandungi zeolite.

CHAPTER 1

INTRODUCTION

1.1 Background of Study

According to United Nations Department of Economic and Social Affairs, the food demand is expected to increase by 70% by 2050 due to population growth. It is estimated that the world population will reach 9.7 billion by 2050 (FAO & Aquaculture, 2007). Most of the population will exist in developing countries where living standards are rapidly rising, and food needs such as meat and dairy products will increase to meet basic dietary needs (Mitch Hunter, 2017). Two-thirds of the world's 1 billion hungry people live in Asia and the Pacific (Timmer, 2014). GFSI is used to measure food security in terms of availability, affordability, food quality and security. In Malaysia, Global Food Security Index (GFSI) declined from 69.4 in 2016 to 66.2 in 2017. It is predicted that Malaysia will become a victim of the food crisis in the near future if it does not pay serious attention to the issue of food productivity (Razak, Sahilla, Amir, Abas, & Idris, 2013).

Developing countries are the nations with the highest fish consumption. Fish is a source of protein to humans and animals (Safaa M., 2012). Food and Agriculture Organization (FAO) showed that Malaysia is one of the top fish consumption countries in Asia which is double the average in China and Thailand. Fish accounting for 85% of Malaysia's total seafood production. Ministry of Agriculture stated that the rate of fish consumption in Malaysia is higher than the rate of meat consumption (Ibrahim, Mohd Khan, Norrakiah, & Intan Fazleen, 2014). The FAO estimates that by 2030, an additional 37 million tons of fish will be needed each year to meet global demand (Barraza, 2010). The main source of Malaysian fish is capture fisheries. In Malaysia, the decline of capture fisheries stock is attributed to overfishing and environmental degradation caused by many anthropogenic activities (Chowdhury & Khairun, 2015). The fish supply from capture fisheries, therefore cannot meet the growing demand for fish food in Malaysia. Therefore, aquaculture is the method used by Malaysia to increase fish production (Barraza, 2010).

Aquaculture is the world fastest-growing food-producing sector over the last two decades. It is an efficient resource for providing animal protein and improving nutrition (Tacon & Metian, 2013). It has contributed 45 % of all the fish consumed by humans by today. In Malaysia, aquaculture contributed 302,886 tonnes of fish in 2012 (A. Yusoff, 2015). According to FAO's forecast, global aquaculture production should reach 102 million tons by 2050 in order to keep current levels of per capita fish consumption at a minimum and reduce the exploitation pressure on stocks of capture fisheries (FAO & Aquaculture, 2007).

Tilapia is one of the most important farmed fish in all aquaculture in the 21st century. It is showed that over 90 percent of tilapia is produced in developing countries, especially in Asia (Ferdous et al., 2017). According to statistics, more than 80% of the world's tilapia is produced in Asia (Eknath & Hulata, 2009). In the past decades, it has become one of the major species of fisheries sector in Asian countries including China, Thailand, Vietnam, Indonesia, Malaysia, Philippine, Bangladesh and Sri Lanka due to its rapid growth rate, high market demand and increasing consumer acceptance (Ferdous et al., 2017). In 2013, the production of red tilapia in Malaysia was 90 % of the total tilapia production (Rahman, Zambry, Basha, Kamarzaman, & Chowdhury, 2013).

As the population grows, food demand will change and emerging economies will need more meat. At the same time, limited resources such as water will have to be managed sustainably. Water is the most important agent in aquaculture. Consideration of the water quantity and water quality used in aquaculture is needed to monitor. In aquaculture, the water quality parameters that are commonly monitored are potential hydrogen (pH), temperature, dissolved oxygen (DO), alkalinity, ammonia (NH_3), nitrites (NO_2^-), nitrate (NO_3^-) and turbidity (Moogouei, Karbassi, Monavari, Rabani & Taheri Mirghaed, 2010).

1.2 Problem Statement

In Malaysia, capture fisheries supply more than 70% of fish for human consumption. However, overexploitation has led to a decline in fish production in the past few decades. The reduction of capture fishery has contributed to the increase in need for aquaculture in tilapia to compensate for the gap between supply and demand (Iliyasu & Mohamed, 2016). According to Department of Fisheries (2010), the main tilapia species of the freshwater cage culture system is red hybrid tilapia, which produced 5,664.42 tonnes in 2010 (Najiah et al., 2012). To increase the fish production, fish density in the culture system increases. However, Department of Fisheries (2012) reported that the intensification of the red tilapia in cage culture system resulted in a cumulative mortality rate of approximately 20,556 red tilapias in February 2012 (Najiah et al., 2012). The massive mortality of red tilapia is due to high-density farming. High density of fish lower the water quality and make the cultured fish more susceptible to outbreaks of disease, *Streptococcus* (Najiah et al., 2012). A large number of red tilapia deaths resulted in fish supply unable to meet local demand. This shows that the stocking density of fish farms is very important and must be regularly monitored and modified according to the size of the fish. Therefore, this study was conducted to investigate the effect of stocking density on the growth performance of red tilapia and water quality in the closed system. In advance to improve the quality of water and feed, zeolites were added to the closed system.

1.3 Objectives

The objectives of this study are:

1. To determine the physicochemical properties such as pH, temperature, DO, salinity, Total Dissolved Solids (TDS), turbidity, Biochemical Oxygen Demand (BOD), Chemical Oxygen Demand (COD), nitrites, nitrate and ammonia nitrogen ($NH_3 - N$) at different stocking densities in zeolite supplemented closed system.

2. To compare the growth performance of red tilapia at different stocking densities in zeolite supplemented closed system.

3. To compare the water quality and growth performance of red tilapia between control and zeolite treatments.

1.4 Scope of Study

The scopes of study are:

1. To determine the parameters of pH, temperature, DO, salinity and TDS of zeolite supplemented culture water by using an YSI Model 556 Multiparameter Meter and turbidity by using a 200 P Turbidimeter.

2. To determine the parameters COD, nitrites, nitrates and ammonia nitrogen by using UV-VIS Spectrophotometer DR 6000 and for BOD will be analyzed by using test kits HQ40D Biochemical Oxygen Demand Meter.

3. To investigate the effect of stocking densities on the growth performance [(weight gain (WG), length gain (LG), average daily weight gain (ADWG), specific growth rate (SGR), survival rate (SR)] of red tilapia (*Oreochromis niloticus*) in zeolite supplemented culture water.

1.5 Significance of Study

In this study, natural zeolite was used in aquariums with different stocking densities to remove the unwanted particles and ammonia. It helps to adjust the water quality, stimulate the growth of plankton and function as natural food for fish. Low-cost, high-tolerance zeolites that are tolerance to temperature and chemical changes are suitable sources of material to make water suitable for fish survival. This study determined the best practice for stocking density in zeolite supplemented closed system and help aquaculture industry to improve the productivity of red tilapia.

CHAPTER 2

LITERATURE REVIEW

2.1 Tilapia and Red Tilapia (*Oreochromis niloticus*)

The native countries of tilapia are Africa and the Middle East. Therefore, Tilapia's name comes from thiape, which is African language for fish. Tilapia is one of a genus of fishes located under the Cichlidae family (Safaa M., 2012). Cichlidae is a diverse group of fishes. Nearly hundreds of species of cichlid fishes in the genera *Tilapia, Oreochromis* and *Sarotherodon* are commonly known as Tilapia. Prior to this, *Tilapia* is considered to be a large genus because there are about 40 species of this genus. Now, many species of Tilapia have been moved to the genera *Oreochromis* and *Sarotherodon*. All these three genera are categorized according to their reproductive behavior. All *Tilapia* species are nest builders; all species in *Oreochromis* and *Sarotherodon* are mouth brooders (Yadav, 2006). The three most important *Tilapia* species in aquaculture industry are *Oreochromis niloticus*, *Oreochromis mossambicus*, and *Oreochromis aureus* (Verster, 2017).

Tilapia is a very important fish genus in production, capture and aquaculture sector. Tilapia is a species which is suitable for estuaries and freshwater aquaculture (Rahman et al., 2013). However, they are primarily farmed in freshwater. Their desirable characteristics make them suitable for aquaculture including high growth rate, highly resistant to diseases, tolerance to a wide range of environmental conditions such as poor water quality (high pH, ammonia and nitrite concentration, and low dissolved oxygen), extreme water temperatures, and high salinities (Ferdous et al., 2017).

Red Tilapia is one of the species in genus *Oreochromis*. In the late 1960s, the first red tilapia hybrid was produced in Taiwan between the *O.nilocitus* and *O.massambicus*. In the 1970s, second red tilapia strain was developed in Florida between *O. hornorum* and *O. massambicus* (Hamzah, Nguyen, Ponzoni, Kamaruzzaman, & Subha, 2008). The Asian developed red tilapia strain consists of the *O. nilocitus* and *O. massambicus* gene pools. In Malaysia, red tilapia accounts for about 90% of total tilapia production. Red tilapia is faster-growing tilapia in the world due to its special characteristics. It achieves top production among the tilapia due to its shorter cultivation period and wide range of tolerant to high temperature than other tilapia.

Red tilapia has been cultured by freshwater aquaculture in Malaysia (Ng, 2009). It is widely cultured in ponds, cages, and pen as well as tanks culture systems. The red tilapia is cultured either in monoculture or polyculture. In Malaysia, cages culture of red tilapia in freshwater dams, former mining pools, rivers, irrigation canals and reservoirs using the semi-intensive and intensive method are practiced (Iliyasu, Mohamed, & Terano, 2016). Intensive culture of red tilapia in tanks also practiced in Malaysia.

Advantages of cages culture system are it requires low capital investment and has the high flexibility of management compared to ponds and tanks. On the contrary, tank culture requires high capital investment because of high construction and production costs (complete commercial diet, aeration, recycling system). Tank culture of tilapia also poses a higher risk of major fish mortality due to disease outbreaks and electrical failures (Gupta & Acosta, 2004). For tank culture systems, stocking density is an important factor in ensuring optimal fish productivity because it is directly related to physiological, physical and chemical parameters such as growth rate, water quality, physiological ability, nutrient and culture system type, and biochemical stage.

2.2 Effect of Stocking Density on Growth

Stocking density refers to the number of specific types of animals per unit area (Abudabos, Samara, Elsayeid, Al-Ghadi, &Al-Atiyat, 2016). Stocking density is an important aspect that affects the survival, behavior, growth performance, food quality, production of fish and water quality. It has been reported that the stocking density has a negative effect on growth rates of fish that depend on density. Three important factors affecting fish growth are fish size, water quality and feeding rate (Chowdhury & Khairun, 2015). Growth performance of fish was evaluated based on the Specific Growth Rate [SGR], Feed Conversion Rate [FCR], Survival Rate [SR], and Daily Weight Gain [DWG] (Ronald, Gladys, & Gasper, 2014).

The increase of stocking density reduces the weight gain, ADWG, and specific growth rate of fish. Daudpota et al. (2014) established that the red tilapia (hybrid) cultured in lowest stocking density *hapa* achieved highest weight gain, daily weight gain, and specific growth performance. Ronald et al. (2014) noted that a reduction of stocking density in a pond with 1000 fry/m^3 Nile Tilapia has greatest weight gain, daily weight gain and specific growth rate than 5330 fry/m^3. The low growth performance of fish with higher stocking densities is due to voluntary appetite suppression, increased competition for food and limited space (Chakraborty & Banerjee, 2010). Fish stocks that exceed the carrying capacity will result in slower fish growth due to lack of food, and will also stress fish because of low dissolved oxygen and high ammonia content (Daudpota et al., 2014). Hashim, Chong, Fatan, Layman, and Ali (2002) reported that the optimal stocking density in a culture system is depend on the size of fish, type and level of nutrient inputs, culture period, rate of water exchange, and possibly of aeration.

2.3 Effect of Stocking Density on Water Quality

Water quality is needed as an indicator of the chemical, physical and biological characteristics of water (Myers, 2014). Water quality is an important factor affecting aquaculture production. Toxic un-ionized ammonia is the main pollutant causing deteriorating of water quality in aquaculture systems, especially for intensive or closed aquaculture system (Yusoff, Banerjee, Khatoon, & Shariff, 2011). Al-Harbi and Siddiqui (2000) reported that increase of fish density in a fish tank may result in high concentration of ammonia nitrogen $(NH_3 - N)$, nitrite nitrogen $(NO_2 - N)$, and low concentration of oxygen. In general, water quality is greatly affected by fish density and rate of feed input (Al-Harbi & Siddiqui, 2000).

In fact, stocking density directly related to total ammonia nitrogen concentration (TAN: ammonia, ammonium ion). In culture system with high fish density, high levels of TAN from unconsumed feed and fish excrement is break down into nitrite (NO_2^-) and nitrate (NO_3^-) by nitrifying bacteria. The conversion of ammonia nitrogen into ionized (NH_4^+) and un-ionized (NH_3) form depends on pH and temperature of the water. Result found that the increase in pH and temperature has increased the concentration of un-ionized ammonia in the water. High levels of ammonia are not only toxic to fish but also reduce the DO in the water. This is because large amount of oxygen is required to convert ammonia into nitrite and nitrate forms. Oxygen deficiency in culture system will cause fish to become tense. However, it was found that the stocking density had no effect on the dissolved oxygen concentration due to the fish reared in the continuous flow cell (Al-Harbi & Siddiqui, 2000). Besides, the high concentration of nitrite also causes fish suffered from brown blood disease as nitrite competes with oxygen binds to hemoglobin to form methaemoglobin. This condition can cause fish to suffocate (Tilak, Veeraiah, & Milton Prema Raju, 2007).

In addition, increasing the density of aquaculture systems will also increase the concentrations of carbon dioxide (CO_2) released through the gills and decomposition of excreta. The high concentration of CO_2 will reduce the pH of the culture water. As conclusion, the pH of the water was influenced by the concentration of TAN and CO_2 (Eshchar, Lahav, Mozes, Peduel, & Ron, 2006). The high stocking density of fish in ponds often exacerbates the problem of water quality and sediment degradation (Daudpota et al., 2014).

2.4 Zeolite

Zeolites (Clinoptilolite) are hydrated aluminosilicates of the alkaline and alkaline-earth metals (Virta, 2001). Zeolite has a highly microporous unique crystalline framework and a high surface area of several hundred square meters per gram of zeolite.

Zeolite is made up of TO_4 tetrahedral units (T may be Si, Al, B, Ge, etc.). Each T atom is connected to four oxygen atoms and each oxygen atom is connected to two T atoms to form chains. The chains connect to each other to form rings. The three-dimensional structure of the zeolite extends to form a framework structure (Hoseinzadeh, 2011).

The zeolite framework is usually composed of tetrahedral units of Silicon oxide, SiO_4 (neutral) and Aluminium oxide, AlO_4^- (negatively charged). The negatively charged is balanced by an external cation such as sodium (Na^+), lithium (Li^+) and calcium (Ca^{2+}) ions. These cations are located in the channels of aluminosilicate framework and can be easily substituted. This unique zeolite structure makes it a good ion exchanger (Hoseinzadeh, 2011). The adsorption-desorption capabilities of zeolite allow charged particles to be quickly absorbed and released.

Zeolite has two specifications, granule, and powder type. The chemical compositions of zeolite are silicon oxide, aluminium (III) oxide, iron (III) oxide, calcium oxide, magnesium oxide, potassium oxide, sodium oxide and phosphorus (IV) pentoxide (Sheppard & Gude, 1969). Zeolite can be divided into two types, which is natural and synthetic zeolite. There are about 50 types of natural zeolites. The most common natural zeolites used are analcime, clinoptilolite, chabazite, erionite, phillipsite, mordenite, ferrierite. More than 150 types of synthetic zeolites; the most common are Beta, Silicalite-1, ZSM-5, Linde Type F, and Linde Type L. They are classified according to their crystal structure and chemical composition (Virta, 2001). The most common natural zeolite used in aquaculture is clinoptilolite and chabazite (Ghasemi, Sourinejad, Kazemian, & Rohani, 2016).

Due to its high adsorption efficiency, a zeolite is widely used as inorganic adsorbents in many fields such as agriculture, animal husbandry, environmental management, chemical industry and aquaculture (Ghasemi et al., 2016). It was found that zeolite capacity loss after ten to eleven regenerations with clean salt water when saturated with cations (Abdel-Rahim, 2017). Factors that affect the ability of zeolites to remove ammonium ions in water are the presence of organics in the wastewater, the ionic strength of the wastewater, the hardness and salinity of the water, and the flow of water (Ghasemi et al., 2016).

In agriculture, zeolite acts as an adsorbent, keeping the fertilizer in the soil by preventing it from leaching or evaporation. This increases the likelihood of plants absorbing nutrients. In animal husbandry, zeolites, as animal feed additives, can reduce the amount of food needed and increase its value quality. For environmental management, zeolites act as ion exchangers to improve the quality of water by purifying sewage from sewage systems. It also helps remove heavy metals and radioactive ions from industrial wastewater. In the chemical industry, zeolites absorb harmful gases released during the petrochemical process (Mumpton & Fishman, 1977).

In aquaculture industry, zeolite functions to improve the water quality of fish farm and fish transportation tanks by removing unwanted particles and ammonia produced by decaying excrement and remaining food (Mumpton & Fishman, 1977). Besides, zeolite also acts as a feed additive to provide natural food for fishes and shrimps as it contains many kinds of mineral materials. This enhances fish growth by increasing nutritional parameters (Mumpton & Fishman, 1977). Şahin, Aral, and Öz

(2016) carried out an experiment to investigate the effect of clinoptilolite on aquarium water. The results showed that the turbidity, ammonia and TAN levels in the clinoptilolite aquarium were lower than in the control group at the end of 12 days. Ghiasi and Jasour (2012) also conducted a study to determine the effects of natural zeolite on water quality, growth performance and nutritional parameters of Angel (*Pterophyllum scalare*). Results of the study showed that the ammonia and hardness of water in the aquarium decreased as the zeolite levels increased. The result also revealed that the final weight of fish in the aquarium with 10 and 15g/ L zeolite were significantly higher than the fish in the 0 and 4g/ L zeolite aquarium (Ghiasi & Jasour, 2012).

CHAPTER 3

MATERIALS AND METHODS

3.1 Materials

This study was conducted at the Aquaculture Laboratory of University Malaysia Kelantan campus Jeli. 225 fry of red tilapia (*Oreochromis niloticus*) with average initial weight and length of 12.90 ±0.01g/fish and 81.95mm/fish were purchased from the fish nursery in Kelantan, Malaysia. Commercial feed (35% crude protein) and anti-chlorine crystals were purchased from One Diq enterprise, Kampung Jeli, Kelantan. On the other hand, zeolites granule (Clinoptilolite) having a size between 0.2 and 0.4 cm was purchased from the Tunas Abadi online store. The chemical composition of the zeolite is shown in Table 3.1. In this study, the tap water was used for fish farming.

Table 3.1: Chemical Composition of zeolite

Elements	%
SiO_2	71.10
Al_2O_3	13.12
Fe_2O_3	0.97
TiO_2	0.19
CaO	1.54
MgO	0.95
Na_2O	0.90
K_2O	2.40

3.2 Methodology

3.2.1 Experimental Design

225 fry of red tilapia were distributed to 15 aquariums containing 30 liters of tap water and acclimatized for two weeks. After the acclimation period, 5 fry of red tilapia were placed in an aquarium containing 30 liters of tap water. Tap water is a municipal water supply containing chlorinated water. In order to make the water harmless to the fish, 0.5 grams of anti-chlorine crystals are added to each aquarium to remove chlorine or chloramine from the tap water. 450 grams of zeolite granule were then placed in a non-woven fabric drain filter and suspended in the water column. Clinoptilolite is used as adsorbent for pollutants in the aquariums. Thereafter, the aquarium is supported by an aerator diffuser to circulate water. The circulation helps to keep the water in the aquarium well oxygenated by moving water from the bottom to the surface to pick up oxygen and release carbon dioxide. Another four treatments with fish densities of T_2= 10, T_3= 15, T_4= 20 and T_5= 25 were prepared using the same method. All treatments were duplicated to reduce errors during the measurement. Next, a control treatment for each stocking density without zeolite was prepared. Fishes was fed a commercial feed containing 35% crude protein twice daily for 9 weeks. The aquarium settings are shown in Figure 3.1.

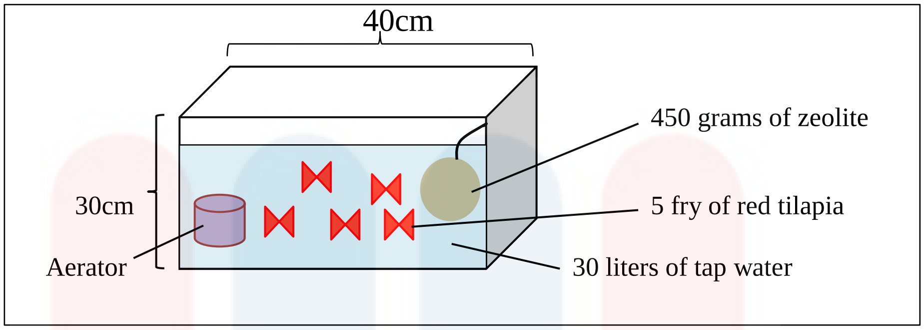

Figure 3.1: Aquarium setting

Water samples from each aquarium were tested weekly to monitor the effect of zeolite on water quality parameters. All of the water samples were collected from each aquarium by using 500 mL polyethylene bottles. On the other hand, the weight and length of the fish were measured every two weeks. This is because there was no significant difference in initial body weight, body length and final body weight and length if data were collected weekly. The experimental schedule is shown in the Figure 3.2. In the duration of the experiment period, zeolite was replaced with new zeolite every month because zeolite may be saturated in a month at very high ammonia levels especially in a closed system.

Week 1 Week 2 Week 3 Week 4 Week 5 Week 6 Week 7 Week 8 Week 9

Week 1, 2, 3, 4, 5, 6, 7, 8, 9: Water quality of treatment water in each aquarium was monitored.

Week 2, 4, 6, 8 **:** The weight and length of the fish was measured.

Figure 3.2: Experimental schedule

3.3 Fish Feeding and Culture

The fishes were fed with 2% of their body weight twice (at 9am and 5pm) a day at 8 hour interval for 63 days. The amount of feed is adjusted according to average weight of the fish in each aquarium. In order to investigate the average weight of fish, five fish in each aquarium were randomly selected every two weeks and their weight and length were measured. The remaining feed and feces in each aquarium are cleaned once a week. The water in each aquarium is replaced with pre-treated pipe water every week. On other hand, daily inspections are also carried out to remove dead fish.

3.4 Fish Sampling

The fish in each aquarium are randomly selected, weighed and released back to the aquarium every two weeks. During the sampling process, 15% of the stocked fish from each aquarium was scooped out with a scoop net. After drying with a towel, their weight and length is measured. In the eighth week, the weight and length of all fish in the aquarium were measured. The number of fish was also calculated at the end of the experiment.

3.5 Data Collection and Analysis

The weight and length of the fish in each aquarium were measured every two weeks using electronic scale and vernier calipers. The measurement of weight was used to determine to evaluate the growth performance of red tilapia by calculating the SGR (%), WG (%) and ADWG of fish in each aquarium. Survival rate (%) was also calculated by determining the number of fish in each aquarium at the end of the experiment. Equations 3.1, 3.2, 3.3, 3.4, and 3.5 were used to calculate the growth parameters.

Specific Growth Rate, SGR (%) = 100 [($In\ W_t - In\ W_0$)/ t] (3.1)

Weight Gain, WG (%) = 100 ($W_t - W_0$)/ W_0 (3.2)

Length Gain, LG (mm) = $L_{t} - L_0$ (3.3)

Average daily weight gain, ADWG = ($W_t - W_0$)/ t (3.4)

Survival Rate, SR (%) = (Final number of fish/ Initial number of fish) $\times 100$ (3.5)

Where W_t and W_0 are final and initial weight (g), L_t and L_0 are final and initial length, and t is time in days from stocking to harvesting.

3.6 Water Quality Analysis

Physical parameters [temperature (℃), salinity (ppt), pH, DO (mg/L), TDS (mg/L) and turbidity (NTU)] were measured by using an YSI Model 556 Multi-parameter Meter and a 2100 P Turbidimeter. While chemical parameters [COD (mg/L), Ammonia (mg/L), nitrite (mg/L), nitrate (mg/L), and BOD (mg/L)] were analyzed by using UV-VIS Spectrophotometer DR 6000 and HACH HQ40d. The instruments used to monitor the water quality parameters are shown in Table 3.2.

Table 3.2: Instrument to monitor the water quality parameters

Instruments / Parameters	YSI Model 556 Multiparameter Meter	UV-VIS Spectrophotometer DR 6000	2100 P Turbidi-Meter	HACH HQ40d
Physical Parameters	- Temperature - Salinity - pH - Dissolved Oxygen (DO) - Total Dissolved Solid (TDS)	-	- Turbidity	-
Chemical Parameters	-	- Chemical Oxygen Demand (COD) - Nitrite nitrogen ($NO_2 - N$) - Nitrate nitrogen ($NO_3 - N$) - Ammonia nitrogen (NH_3-N)	-	- Biochemical Oxygen Demand (BOD)

3.7 Nitrate Analysis

First, the program 353 N, Nitrate MR PP in UV-VIS Spectrophotometer was started. For the preparation of the sample, a sample cell was filled with 10 mL of sample. Then, the content of one Nitrate 5 Reagent Powder Pillow was added to the sample cell. A 1 minute reaction time was started by using instrument timer. The sample cell was closed and was shaken vigorously until the timer expires. After that, a 5 minutes reaction time was started. For the preparation of blank, a second sample cell was filled with 10 mL of sample. After the timer expired, the blank was cleaned and inserted into the cell holder. ZERO was pushed and the display was showed in 0.00 mg/L. Then, the prepared sample was cleaned and inserted into the cell holder within 2 minutes after the timer expired. READ was push and the result was shown in mg/L.

3.8 Nitrite Analysis

First, the program 371 N, Nitrite LR PP in UV-VIS Spectrophotometer was started. For the preparation of sample, a sample cell was filled with 10 mL of sample. Then, the content of NitriVer 3 Reagent Powder Pillow was added into the sample cell. The sample cell was swirl to mix. Next, a 20 minutes reaction time was started. After the timer expired, a blank was prepared. For the preparation of blank, a second sample cell was filled with 10 mL of sample. After that, the blank was cleaned and inserted into the cell holder. ZERO was pushed and the display was showed in 0.00 mg/L. Next, prepared sample was cleaned and inserted into the cell holder. READ was push and the result was shown in mg/L.

3.9 Nitrogen, Ammonia Analysis

First, the program 385 N, Ammonia, Salic in UV-VIS Spectrophotometer was started. For the preparation of blank, a sample cell was filled with 10 mL of deionized water. For the preparation of sample, a second sample cell was filled with 10 mL of sample. Then, the content of one Ammonia Salicylate powder was added to each sample cell. Sample cells were closed and shaken to dissolve the reagent. Next, a 3 minutes reaction time was started by using instrument timer. After the timer expired, the content of one Ammonia Cyanurate powder pillow was added to each sample cell. Sample cells were closed and shaken thoroughly in order to dissolve the reagent. Then, the solution was waited for 15 minutes to complete the reaction. Lastly, the reading of the sample was taken after the blank solution.

3.10 Chemical Oxygen Demand (COD) Analysis

100 ml of water sample was homogenized for 30 seconds in a blender. Then, the homogenized sample was poured into a 250 mL beaker and stirred gently with a magnetic stir plate. The DRB 200 was turned on and preheats to 150 ℃. After that, 2 mL of sample was pipetted into the vial for the selected range at an angle of 45 degrees. For the blank vial of selected range, 2 mL of deionized water was added into the vial by using a clean pipet. Then, the vials were closed tightly. Next, vials were rinse with water and wipe with a tissue. The vial was inverted gently several times to mix and then inserted into the preheated DRB200 Reactor. The lid was closed and the sample was heated for two hours. After two hours, the DRB 200 was turned off the vial was left

about 20 minutes to cool to 120 ℃ or less. Each vial was inverted several times while it was still warm and then was put in a tube rack to cool to room temperature. Then, program 431 COD was started in a UV-VIS Spectrophotometer. The blank vial was cleaned and inserted into the cell holder and ZERO was pressed. After that, the prepared sample was cleaned and inserted into the cell holder. READ was pressed and result was shown.

3.11 Biochemical Oxygen Demand (BOD) Analysis

To prepare the dilution water, 3 L of distilled water was added to 3 L BOD bottle. BOD bottle filled with distilled water were then put into the chiller for overnight. BOD bottles were taken out from chiller. 3 L of BOD Nutrient Buffer Pillow was added to the BOD bottle. BOD bottle was inverted several times to mix. For the preparation of the sample, 100 mL of water was added into a BOD bottle. After that, the dilution water was added to the water sample up to 300 mL. For the preparation of blank, another BOD bottle was filled with 300 mL of prepared dilution water. To prevent air bubbles, the water was pour down the inner surface of the bottle. The dissolved oxygen in the blank and water sample were measured by using HQ40D Portable Meter Kit. Next, the prepared sample bottles was kept in an incubator at 20 ℃ (68 ℉) for 5 days. After 5 days, the remaining dissolved oxygen in the prepared sample was measured. Ensure that the prepared sample contained a minimum DO concentration of 1.0 mg/L after incubation to obtain accurate results.

3.11.1 Calculation of the Concentration of BOD

$$\mathbf{BOD_5\ mg/L} = (D_1 - D_2) / \mathrm{P} \qquad (3.5)$$

Where BOD_5= BOD value from the 5-day test (mg/L)

D_1= DO of the prepared sample immediately after preparation in mg/L

D_2= DO of the prepared sample after incubation in mg/L

P= Decimal volumetric fraction of the sample used

3.12 Statistical Analysis

All data collected were subjected to statistical analysis and analyze by using SPSS version 20 program. Two way analysis of variance (MANOVA) was used to evaluate the effects of stocking densities on water quality and the growth performance of red tilapia in zeolite supplemented closed system at the five stocking densities. In addition, MANOVA was used to analyze the effect of zeolite on water quality and growth performance of fish between treatments with and without zeolite. Then, a post hoc test using Tukey's multiple range tests, with $p < 5\%$ significance levels were used to evaluate the differences among treatment means.

CHAPTER 4

RESULTS AND DISCUSSIONS

4.1 Effect of Stocking Density on Growth Performance of Red Tilapia in Zeolite-containing Treatments

The growth performance of red tilapia in different treatments in terms of initial number and final mean number (n), mean weight, mean length, weight gain (WG), length gain (LG), average daily weight gain (ADWG), specific growth rate (SGR), and survival rate (SR) were calculated and are presented in Table 4.1. The effect of stocking density on growth performance of red tilapia was investigated in the experiment.

Table 4.1: Growth parameters (mean ± SD) of red tilapia in the zeolite treatments at different stocking densities.

Growth Parameters	Treatment 1	Treatment 2	Treatment 3	Treatment 4	Treatment 5
Initial number (n)	5	10	15	20	25
Final mean number (n)	3.5 ±2.12	8.5 ±2.12	14.5 ±0.71	19 ±1.41	23 ±1.41
Mean initial weight (g)	12.90 ±0.01	12.90 ±0.01	12.90 ±0.01	12.90 ±0.01	12.90 ±0.01
Mean final weight (g)	40.27 ± 15.84^{b}	23.17 ± 0.7^{ab}	21.47 ± 0.00^{ab}	19.63 ± 2.18^{ab}	17.26 ± 0.15^{a}
Mean initial length (mm)	81.95 ±0.02	81.95 ±0.02	81.95 ±0.02	81.95 ±0.02	81.95 ±0.02
Mean final length (mm)	106.33 ± 2.46^{c}	97.33 ± 5.44^{b}	93.13 ± 0.33^{ab}	90.66 ± 4.63^{ab}	87.83 ± 2.01^{a}
Weight gain, WG (%)	212.17 ± 122.78^{a}	79.65 ± 5.43^{a}	66.43 ± 0.00^{a}	52.17 ± 16.88^{a}	33.76 ± 1.15^{a}
Length gain (mm)	24.38 ± 2.46^{b}	15.38 ± 5.44^{ab}	11.18 ± 0.33^{ab}	8.71 ± 4.63^{a}	5.88 ± 2.01^{a}
Average daily weight gain, ADWG (g)	0.4345 ± 0.2541^{a}	0.1631 ± 0.0112^{a}	0.1360 ± 0.0000^{a}	0.1069 ± 0.0346^{a}	0.0692 ± 0.0023^{a}
Specific growth rate, SGR (% per day)	1.74 ± 0.64^{b}	0.93 ± 0.05^{ab}	0.81 ± 0.00^{ab}	0.66 ± 0.17^{ab}	0.46 ± 0.01^{a}
Survival rate, SR(%)	70 ± 42.43^{a}	85 ± 21.21^{a}	96.67 ± 4.72^{a}	95 ± 7.07^{a}	96 ± 5.66^{a}

Values with different superscript within a row are significant difference (p<0.05).

4.1.1 Length and Weight

At the beginning of the experiment, there was no significant difference ($p > 0.05$) in initial weight and length of red tilapia under different treatments. Significant difference ($p<0.05$) were observed among five treatments in final mean length and weight when compared using MANOVA. The results showed that with the increase of stocking density, the final mean length and weight showed a downward trend. It was found that fry stocked in T_1 exhibited the highest mean final length and weight (106.33 and 40.27) while fry stocked in T_5 recorded the lowest (87.83 and 17.26). The final mean weight and length observed at high stocking density were low; this may be due to insufficient acquisition of feed, low availability of oxygen and increased competition for food and the space for fish movement. These results are in agreement with the findings obtained by Chakarborty and Banerjee (2010) who revealed that the increased fish biomass of Nile tilapia in cages had a significant negative effect on the final mean weight. Ferdous, Hossain, and Jaman (2017) reported that Monosex tilapia in hapa at a low density had a better growth than at a higher density. The lower growth performance of tilapia at higher stocking density may be caused by voluntary appetite suppression, more energy is expended on intense antagonistic behavioral interaction between fish, increased competition for food and living space, and increased stress due to reduction in space availability (Ferdous et al.).

At sampling 3 (week 6) and 4 (week 8), there was a significance difference in mean length between four samplings ($p<0.05$). Tukey's test indicated a highly significant different ($p<0.05$) between T_1 and the rest of the treatment. The relationship between treatments and mean length of the four samplings are shown in Figure 4.1.

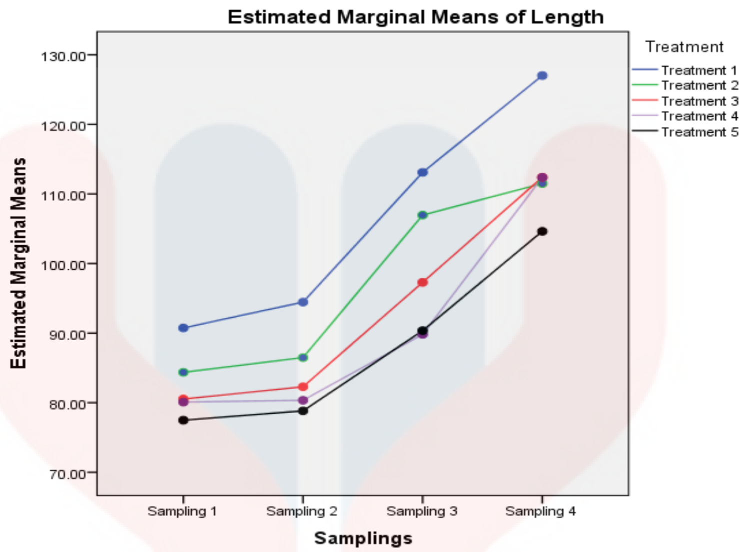

Figure 4.1: Relationship between mean lengths among treatments for the four samplings.

On the other hand, at sampling 4 (week 8), there was a significance difference in mean weight of all the four samplings ($p<0.05$). Tukey's test showed that there was no significant ($p>0.05$) different in mean weight between T_1 and the rest of the treatment except T_5. Figure 4.2 shows the relationship between treatments and mean weight of the four samplings.

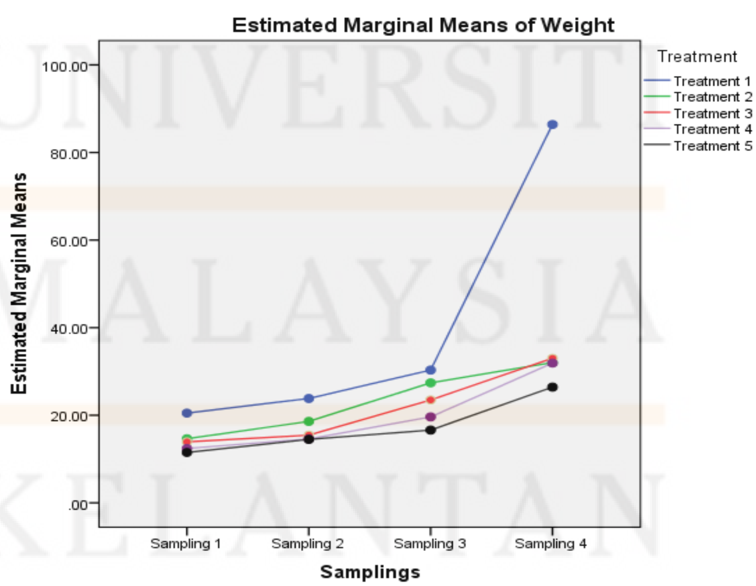

Figure 4.2: Relationship between mean weights among treatments for the four samplings.

4.1.2 Specific Growth Rate

In the current study, there was a significant different ($p<0.05$) in specific growth rate (SGR) between the five treatments (refer to Table C.2 in appendix). As can be seen from Table 4.1, the mean specific growth rate of red tilapia in different treatments was between 0.46 and 1.74. The significantly ($p<0.05$) highest SGR values (1.74) was recorded in T_1 while the lowest (0.46) was recorded in T_5. The low growth rate in T_5 may be due to increased crowding effect of fish, making it difficult for the fish to move to reach the food, thus reducing the feeding rate. It can be seen that it is more difficult to ensure uniform distribution of food at high stocking densities. These results are consistent with the results obtained by Dambo and Rana (1993), who reported that SGR was significantly affected by stocking density. Figure 4.3 showed that SGR decreased with increasing stocking densities.

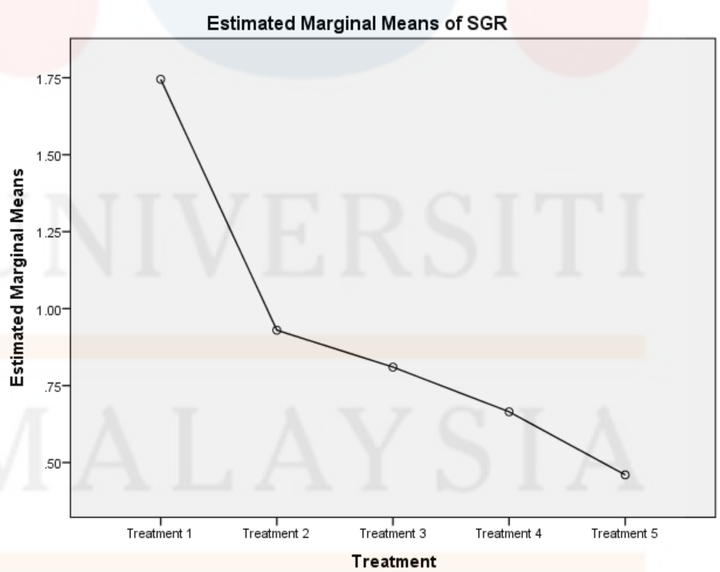

Figure 4.3: Comparison of specific growth rate between treatments.

4.1.3 Weight Gain and Length Gain

The final mean weight gain of red tilapia in different treatments ranged between 33.76 and 212.17. The weight gain of fish was statistically similar ($p>0.05$) at different stocking densities (refer to Table C.2 in appendix). The highest and lowest final weight gain of red tilapia was recorded in T_1 (212.17) and T_5 (33.76) respectively. Figure 4.3 showed an inverse relationship between stocking density and weight gain in five treatments.

The final length gain of individual fish in different treatments ranged between 5.88 and 24.38. There was a significant difference ($p<0.05$) at different stocking density (refer to Table C.2 in appendix). The length gain of red tilapia in T_1 (24.38 ±2.46) was significantly highest and in T_5 (5.88 ±2.01). Figure 4.4 showed an inverse relationship between stocking density and length gain in five treatments.

The low growth at high stocking densities may be due to social interaction through competition for food and living space; this may lead to increased stress, resulting in increased energy demand and decreased in weight gain. Similar observations were made by Ofor and Afia (2015), who found that the weight gain of hybrid catfish was not affected by stocking density. The weight gain and length gain in T_1 higher than all others, it can be assumed that there is metabolic savings and low energy consumption at this density. These findings were similar to those reported by Rahman (2016), who revealed that the Monosex male tilapia stocked at the lowest densities achieved optimal weight gain.

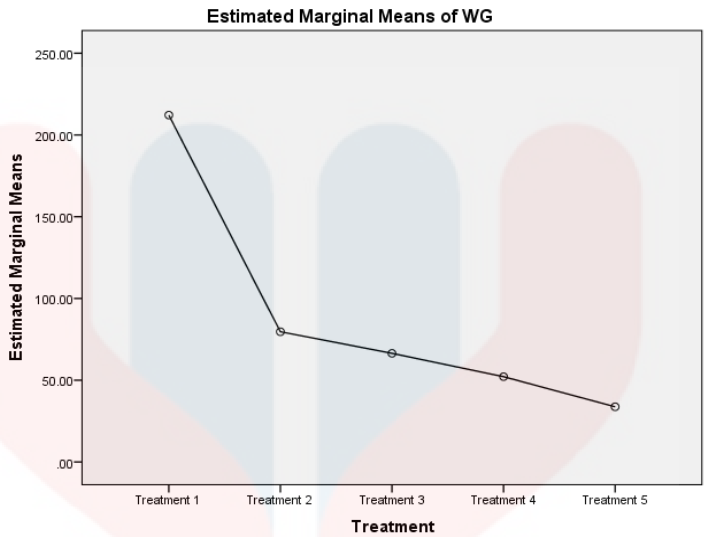

Figure 4.4: Comparison of weight gain between treatments.

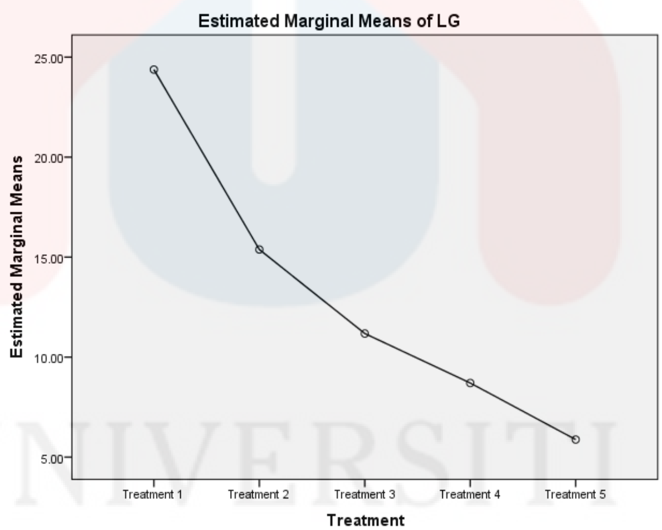

Figure 4.5: Comparison of length gain between treatments.

4.1.4 Survival Rate

In this study, there was no significance ($p>0.05$) different in SR between treatments (refer to Table C.2 in appendix). Tukey's test showed that there was no significant different in SR between T_1 and the rest of the treatment. Figure 4.4 showed the relationship between stocking density and survival rate of red tilapia. The results showed that survival rate for all the treatments were above 70%. Fish reared in T_3 recorded the highest percent survival of 96.67% while the fish population reared in T_1 showed the lowest percent survival of 70%. It can be seen that survival rate did not show a significant decline as stocking density increased. This may be because the stocking density is not as high as that commonly used in aquaculture. The stocking densities have not reached the threshold at which food availability and competition among individuals impacted growth rate. These results are in agreement with Rahman (2016) who reported that the mortality of Nile tilapia in cages was not dependent on stocking density. The results of current study showed that, survival rates increase with high stocking density. Similar results were obtained by Quattara, Teugels, Douba, and Philippart (2003), who showed 98%, 96% and 100% survival rates (50 fish/m^3, 100 fish/m^3 and 150 fish/m^3). The lower survival rate in T_1 could be attributed to the inhibition of proper feeding of smaller fish due to the presence of larger fish. Consequently, the high survival rate of red tilapia at high stocking density in this study showed the ability of to survive in poor conditions (including high density) and the amenability of this fish to the intensive culture system.

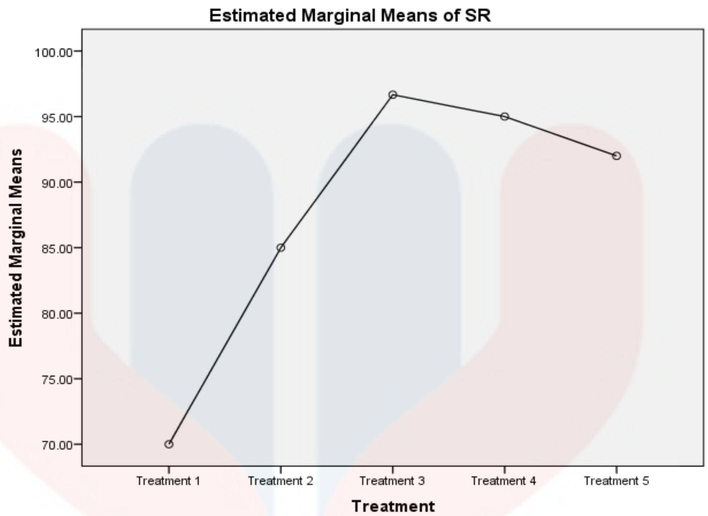

Figure 4.6: Comparison of survival rate between treatments.

In general, the growth performance of red tilapia in zeolite systems decreased as the stocking density of fish increased. Based on the growth performance parameters recorded in this study, it was found that red tilapia stocked in the T_1 of the lowest stocking density (5 fish) had the highest growth performance than the fish in other treatments. Compare to other treatments, T_1 recorded the highest final length and weight, weight and length gain, SGR among the treatments. As conclusion, T_1 with the lowest fish density is the most suitable stocking density for the red tilapia.

4.2 Effect of Stocking Density on Water Quality Parameters in Zeolite-containing Treatments

The overall mean values for water quality parameters are given in Table 4.2. Significant differences ($p<0.05$) were found in all water quality parameters among treatments except salinity (refer to Table C.3 in appendix). Figure 4.6 and 4.7 showed the physical and chemical parameters of treatment water at different stocking density.

Table 4.2: Water quality parameters (mean value±S.E.) in different treatments with zeolite.

Parameters	Treatment				
	1	2	3	4	5
Temperature (℃)	25.73 ± 0.84^{ab}	25.65 ± 0.91^{a}	25.63 ± 0.89^{a}	26.04 ± 0.85^{c}	25.96 ± 0.80^{bc}
Dissolved oxygen (mg/L)	3.74 ± 0.84^{c}	2.92 ± 0.85^{abc}	3.71 ± 0.96^{bc}	2.67 ± 0.86^{ab}	2.54 ± 0.89^{a}
pH	6.24 ± 0.25^{a}	6.48 ± 0.31^{abc}	6.39 ± 0.46^{ab}	6.73 ± 0.55^{bc}	6.80 ± 0.47^{c}
Salinity (ppt)	0.08 ± 0.02^{a}	0.11 ± 0.02^{a}	0.12 ± 0.02^{a}	0.13 ± 0.03^{a}	0.14 ± 0.01^{a}
Total dissolved solids (mg/L)	118.20 ± 25.5^{a}	148.10 ± 22.1^{b}	163.20 ± 28.7^{bc}	180.30 ± 32.20^{cd}	189.40 ± 14.40^{d}
Turbidity (NTU)	17.88 ± 4.72^{a}	33.21 ± 14.53^{ab}	43.69 ± 17.27^{bc}	70.17 ± 50.59^{d}	56.59 ± 9.07^{cd}
Chemical oxygen demand (mg/L)	55.20 ± 21.7^{a}	88.80 ± 26.1^{b}	94.80 ± 44.4^{b}	106.10 ± 31.3^{bc}	124.10 ± 9.3^{c}
Biochemical oxygen demand (mg/L)	13.06 ± 3.16^{a}	15.34 ± 3.97^{b}	15.52 ± 2.55^{b}	17.45 ± 1.65^{c}	18.89 ± 1.72^{c}
Nitrite (mg/L)	2.744 ± 2.053^{a}	3.459 ± 2.049^{ab}	3.155 ± 1.817^{ab}	2.812 ± 2.074^{bc}	3.441 ± 1.811^{c}
Nitrate (mg/L)	8.78 ± 6.9^{a}	11.72 ± 7.82^{ab}	10.12 ± 5.43^{ab}	12.68 ± 10.15^{ab}	15.37 ± 21.26^{b}
Ammonia (mg/L)	2.34 ± 1.43^{a}	5.19 ± 2.4^{ab}	4.84 ± 3.84^{ab}	7.94 ± 2.46^{bc}	11.60 ± 9.07^{c}

Values with different superscript within a row are significant difference ($p<0.05$).

The maintenance of a good water quality is important to ensure optimum growth of culture organisms. Temperature outside the optimal range can act as stressors and fish cannot feed actively as non-stressed situation (Stickney, 2005). This would affect the growth of aquatic organisms. Temperature would influence all the chemical and biological processes in an aquaculture operation. In the current study, the mean temperature was stable around 25.63 to 26.04 °C. Makori, Abuom, Kapiyo, Anyona, and Dida (2017) stated that the preferred temperature range for optimum tilapia growth in ponds was between 25 and 27 °C. A research by Devi, Padmavathy, Aanand, and Aruljothi (2017) showed that a temperature range of 25 to 32 °C is ideal for tropical fish farming. This indicated the water temperature in this study is suitable and ideal for red tilapia culture.

pH is another important physical parameter that controls the amount of soluble ions in the water body. Boyd and Lichtkoppler (1979) reported that an acidic pH of treatment water would reduce the growth rate, metabolic rate and other physiological activities of fishes. In this study, the mean values of pH showed a narrow range of variation between treatments, which was ranged from 6.30 to 6.80. Figure 4.6 showed the pH value increased with the increased of stocking density. This may be due to the accumulation of ammonia increased at the highest stocking density. Makori, Abuom, Kapiyo, Anyona, and Dida (2017) stated that the optimal pH for tilapia is between 6.5 and 9. All pH values obtained in this study were in slightly acidic and neutral in all treatments which indicate good productivity.

In this study, the mean salinity value ranged from 0.08 to 0.14 ppt. Figure 4.6 showed a slightly increased in salinity when the stocking density increased. The mean value of salinity was found no significance difference ($p>0.05$) between different treatments although T_5 recorded the highest salinity level (0.14). Based on National Water Quality Standard for Malaysia, the suitable range for tolerance species is lower than 2 ppt. All the mean salinity values obtained in this study was within the range which indicated the water conditions suitable for tilapia fish culture.

On the other hand, the mean value of total dissolved solids was ranged between 118.2 and 189.4 mg/L. Figure 4.6 showed that the total dissolved solids was increased with the increasing stocking density and the highest value (189.4) was found at highest density. While the mean values of turbidity was ranged between 17.88 and 70.17. Zweigh (1989) reported that the suitable range of turbidity for fish culture was between 20-30 NTU. But the turbidity levels in current study showed little higher comparatively with Zweigh (1989) findings.

Dissolved oxygen (DO) is the most critical water quality parameter for fish and nitrifying bacteria that convert fish waste into non-toxic state. In the current study, the highest mean dissolved oxygen concentrations were found in T_1 (3.74) and the lowest value was recorded in T_5 (2.54). Figure 4.6 showed the DO values gradually decreased with the increasing of stocking density. Similar findings were observed by Murugesan, Soundarapandian, and Manivannan (2011) who reported that a decreasing in DO concentrations at high stocking density of fish was attributed to the gradual increase of biomass. From this study, it was found that the DO levels in T_2, T_4, and T_5 were recorded below 3 mg/L although all the treatments were continuously aerated throughout the study period. However, it did not show mass mortalities in any treatments but may affect the growth rate of fish. This is because tilapia would be able to tolerate dissolved oxygen levels of less than 0.3 mg/L. The low dissolved oxygen in the treatments may be due to insufficient aeration as aerator in the aquarium do not create current across entire aquariums, thus basically the same water being cycled through the aerator repeatedly. Boyd and Lichtkoppler (1979) stated that the concentration of DO below 3.5 mg/L is undesirable for fish farming. Stickney (2005) reported that the acceptable range of dissolved oxygen was 5 mg/L, the fish may be stressed when DO lower than 3 mg/L and exists hypoxia when less than 2 mg/L.

Biochemical oxygen demand (BOD) is the measurement of the amount of oxygen needed to decompose organic waste in water. On the other hand, chemical oxygen demand (COD) refers to the amount of oxygen needed to oxidize all organic and inorganic matters in the water by strong oxidant. The higher the BOD and COD value, the lower the water quality. According to INTERIM Water Quality Standards for Malaysia, the optimum levels for BOD and COD for aquaculture should be less than 6 and 50 mg/L. According to Figure 4.7, the value of BOD ranged between 13.06 and 18.89 while the COD values ranged between 55.2 and 124.1, far beyond the range given by the standard. This may be due to insufficient oxygen supply or accumulation of metabolic waste products and feed residuals that caused by low water exchange frequency. However, the standard is based on the ideal range of all species in aquaculture but does not reflect the acceptable tolerance limits for tilapia. High BOD and COD value would cause fish to be stressed, suffocated and possibly die.

Ammonia is the major product of metabolic waste and nitrogenous waste in intensive aquaculture production. It exists in equilibrium between two forms in water: ionized (NH_4^+) and un-ionized (NH_3) forms. Conversion of ammonia into toxic unionized form (NH_3) depends on the temperature and pH of water. The higher the temperature and pH in the water, the more the toxic unionized (NH_3) ammonia is formed. According to Figure 4.7, the mean values of ammonia significantly increased as the fish density increased and mean ammonia concentrations ranged from 2.34 to 11.60 mg/L. According to Makori, Abuom, Kapiyo, Anyona, and Dida (2017) the optimal range of ammonia concentrations for tilapia growth was 0.02-0.05 mg/L. The ammonia concentration in the current study is higher than the optimal range reported by Makori, Abuom, Kapiyo, Anyona, and Dida (2017), this may be due to the low frequency of water exchange, only once a week and caused the ammonia accumulate in the treatments. As a result, the high concentration of ammonia in the treatments was greater than the amount that the zeolite can handle.

Nitrite is highly toxic to fish even in small quantity. High concentration of nitrite reduces the ability of haemoglobin to transport oxygen because nitrites combine with haemoglobin to produce methaemoglobin (Timmons, Ebeling, Wheaton, Summerfelt, & Vinci, 2001). Fish will be asphyxiated when loss haemoglobin (Stickney, 2005). Timmons et al. (2001) reported that the nitrite levels for tilapia should be maintained below 1 mg/L. Jiménez-Ojeda, Collazos-Lasso, and Arias-Castellanos (2018) reported that tilapia begin to die when nitrite levels reach 5 mg/L. In the present study, the mean nitrite concentrations ranged from 2.744 to 3.459 which were lower than the lethal level but higher than the permission limit. The high concentrations of nitrite in the treatments may be due to insufficient aeration. The low oxygen levels may slow down or even stop the nitrification process to covert the nitrite into nitrate and this may lead to the accumulation of toxic nitrite. Figure 4.7 showed that the nitrite levels increased as the stocking density increased.

Nitrate is non-toxic to fish even in large concentration but the accumulation of nitrate can cause stressed to fish and affect the growth rate. The mean value of nitrate concentrations for the current study ranged from 8.78 to 15.37. The highest nitrate content was found in T_5 (15.37), which has the highest stocking density. Boyd (2004) stated that the desired nitrate concentration for aquaculture is between 0.2 and 10 mg/L. The nitrate concentrations obtained in the current study were beyond the optimal range for fish culture. Figure 4.7 revealed that the concentrations of nitrate was increased with the increased of stocking density. Similar findings were observed by Khatune-Jannat et al. (2012), who found that nitrates concentrations increased as the stocking density of ornamental fish increased.

In general, the water quality of treatment water in zeolite supplemented closed system deteriorated with the increasing of stocking density. Based on the recorded water quality, the T_1 of the lowest stocking density (5 fish) is the most suitable stocking density for the red tilapia, because the water quality is better than other treatments. Compared to other treatments, T_1 recorded the higher concentrations of dissolved oxygen and lower levels of toxic ammonia, nitrite, and nitrate, which is more ideal for tilapia culture.

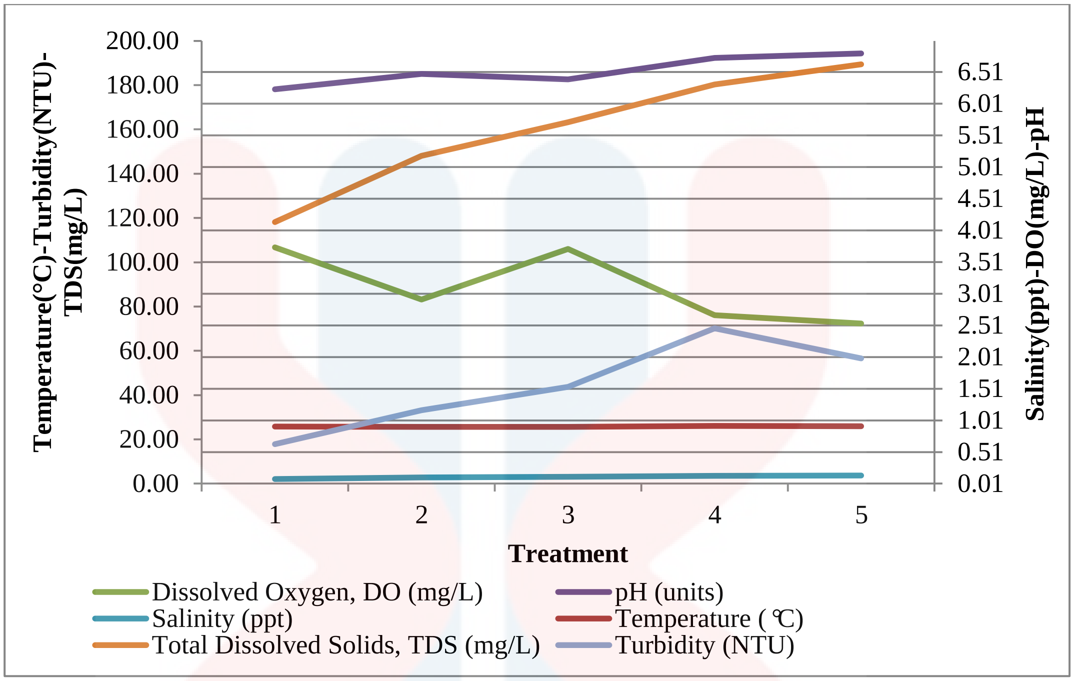

Figure 4.7: Comparison of physical water quality parameters between different treatments with zeolite.

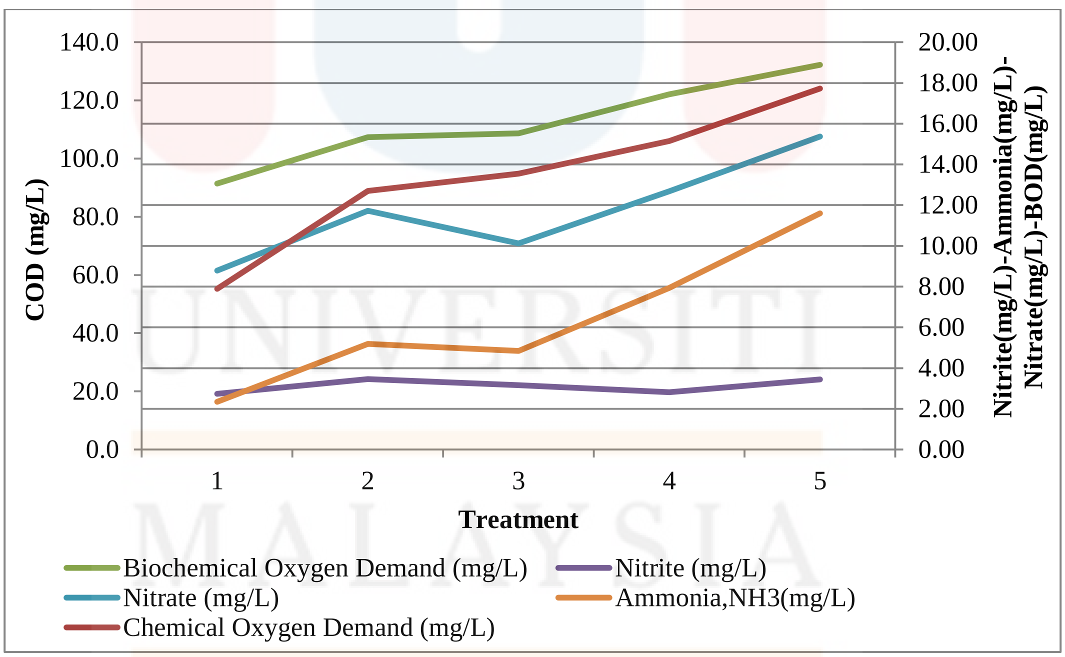

Figure 4.8: Comparison of chemical water quality parameters between different treatments with zeolite.

4.3 Comparison between Control and Zeolite Treatments

4.3.1 Effect of Zeolite on Growth Performance of Red Tilapia

The mean values of growth parameters of red tilapia in the control treatments was calculated and recorded in Table 4.3.

Table 4.3: Growth parameters of red tilapia in the control treatments under different stocking densities.

Growth Parameters	Treatment 1	Treatment 2	Treatment 3	Treatment 4	Treatment 5
Initial number (n)	5	10	15	20	25
Final mean number (n)	5	5	12	7	17
Mean initial weight (g)	12.90	12.90	12.90	12.90	12.90
Mean final weight (g)	27.64	25.99	19.16	22.48	18.85
Mean initial length (mm)	81.95	81.95	81.95	81.95	81.95
Mean final length (mm)	101.08	99.05	90.63	92.07	89.65
Weight gain, WG (%)	114.26	101.47	48.53	74.26	46.12
Length gain (mm)	23.34	20.87	10.59	12.35	9.4
Average daily weight gain, ADWG (g)	0.2340	0.2078	0.0994	0.1520	0.0944
Specific growth rate, SGR (% per day)	1.12	1.11	0.63	0.88	0.6
Survival rate, SR (%)	100	50	80	35	68

The growth performance of red tilapia in control treatments was compared with that of the zeolite. The initial mean length and mean weight of fish stocked in control and zeolite groups was same. The difference between the fish in control and zeolite was not considered as significance in terms of growth performance ($p>0.05$) when tested with MANOVA. This indicated that usage of zeolite did not influence the growth performance of red tilapia. Similar findings were found by Onder Yildirim, Turker, and Bilgin Senel (2009) who found no significant difference ($p>0.05$) in growth performance between Tilapia zillii fed diets containing and without zeolite. From the results, it was found that T_1 with zeolite recorded the highest values in mean final weight and length, weight gain, length gain, and growth rate among the treatments. However, the mean values of these growth parameters recorded by T_4 and T_5 with zeolite were lower than the control. This may be due to the fact that the survival rate of the T_4 and T_5 with zeolite is much higher than the control. Therefore, the number of fish per unit area in the zeolite treatment was greater than that of the control. Aksungur, and Kutlu (2007) reported that high stocking densities leads to increased stress, resulting increased in energy demand and causing a reduction in growth rates and feed utilization. According to Figure 4.12, all treatments with zeolite except T_1 had higher survival rate than control. The low survival rate in T_1 with zeolite may be due to the inhibition of proper feeding of smaller fish due to the presence of larger fish. During the experiment, it was found that one fish was much larger than the rest of the fish in the aquarium.

In general, usage of zeolite did not influence the growth of red tilapia. Based on the growth performance parameters recorded in this study, it was found that red tilapia stocked in treatments with zeolite had the highest growth performance than the stocked in control treatments.

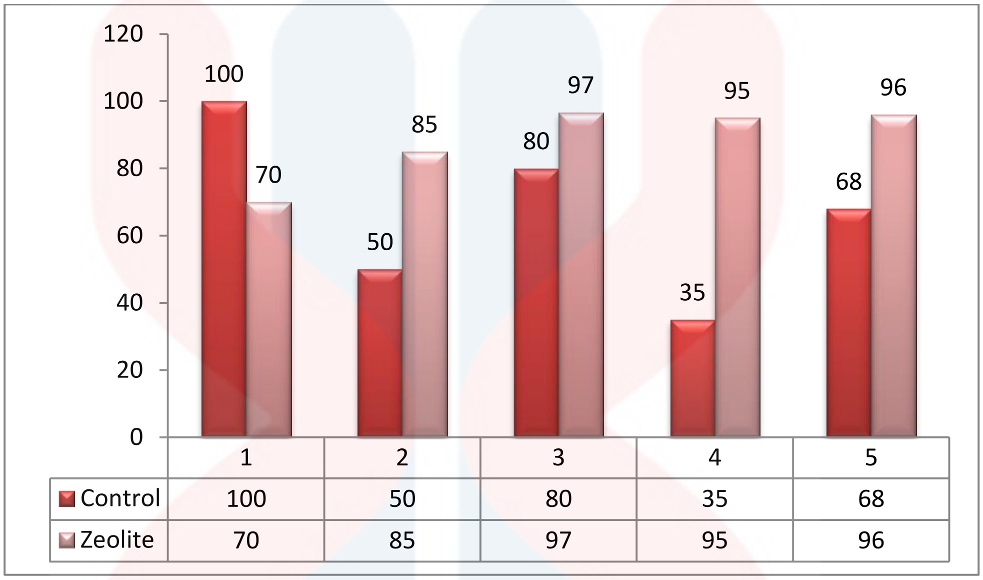

Figure 4.9: Comparison of survival rate (%) between control and zeolite treatments.

4.3.2 Effect of Zeolite on Water Quality

The mean values of water quality parameters for control treatments are listed in Table 4.3. The results showed there was significant differences ($p<0.05$) in temperature, DO, pH, COD, BOD, nitrite, nitrate, and ammonia between control and zeolite groups (refer to Table D.3 in appendix). However, there was no significant difference ($p>0.05$) in salinity, total dissolved solids, and turbidity between zeolite and control groups (refer to D.3 in appendix).

Table 4.4: Water quality parameters (mean value±S.E.) between control groups at different stocking densities.

Parameters	Treatment				
	1	**2**	**3**	**4**	**5**
Temperature (℃)	25.77 ±0.93^{a}	26.16 ±0.75^{a}	26.29 ±0.86^{a}	26.24 ±0.80^{a}	26.65 ±0.80^{a}
Dissolved oxygen (mg/L)	3.61 ±1.13^{a}	2.95 ±1.20^{ab}	2.70 ±1.08^{ab}	2.36 ±1.30^{ab}	1.52 ±0.54^{b}
pH	5.99 ±0.26^{a}	6.25 ±0.36^{ab}	6.31 ±0.81^{ab}	6.42 ±0.44^{ab}	6.70 ±0.42^{b}
Salinity (ppt)	0.09 ±0.01^{a}	0.11 ±0.02^{b}	0.13 ±0.02^{bc}	0.12 ±0.02^{b}	0.15 ±0.01^{c}
Total dissolved solids (mg/L)	125.94 ±16.99^{a}	156.50 ±20.10^{b}	181.61 ±22.29^{bc}	164.22 ±23.01^{b}	206.28 ±19.33^{c}
Turbidity (NTU)	15.71 ±5.53^{a}	30.04 ±10.7^{ab}	44.13 ±16.68^{bc}	42.67 ±17.05^{bc}	57.11 ±10.71^{c}
Chemical oxygen demand (mg/L)	91.30 ±45.5^{a}	133.90 ±44.9^{ab}	167.00 ±28.8^{b}	174.50 ±31.5^{b}	181.70 ±22.7^{b}
Biochemical oxygen demand (mg/L)	14.80 ±3.07^{a}	16.33 ±2.98^{ab}	18.69 ±1.86^{b}	17.93 ±3.56^{ab}	19.46 ±2.19^{b}
Nitrite (mg/L)	3.343 ±2.301^{a}	4.278 ±1.187^{ab}	3.628 ±1.871^{ab}	4.284 ±0.938^{b}	3.778 ±1.674^{b}
Nitrate (mg/L)	13.29 ±8.9^{a}	25.78 ±18.07^{a}	17.73 ±21.56^{ab}	20.59 ±19.61^{ab}	20.67 ±20.61^{b}
Ammonia (mg/L)	7.78 ±1.9^{a}	10.14 ±2.78^{ab}	12.76 ±3.87^{bc}	9.73 ±2.9^{ab}	15.88 ±4.63^{c}

Values with different superscript within a row are significant difference (p<0.05).

Results showed that the concentration of ammonia nitrogen, nitrite, and nitrate in zeolite treatments was significantly ($p<0.05$) lower in compare with control treatments. These results indicated that it is possible for the zeolite to effectively remove ionized ammonium, nitrite, and nitrate ions from the water. These results are in agreement with Danabas and Altun (2011) who revealed that zeolite is able to reduce the ammonia, nitrite, and nitrate concentration in the concentrate ponds. Yousefian et al. (2010) also reported that the application of zeolite reduce the concentration of ammonia.

Zeolites remove ammonium ions in the water by ion-exchange process. In water, the negatively charge zeolites were neutralized by exchangeable cations (normally K^+, Na^+, Ca^{2+}, and Mg^{2+}). Zeolite exchanges the weakly bound sodium ions for ammonium ions present in the water and shifts the ammonia equilibrium away from toxic unionized ammonia, thus reducing the levels of toxic unionized ammonia (Ghiasi & Jasour, 2012). Ammonium ions replaced the sodium ions in the zeolite channel because zeolite has higher selectivity toward ammonium ion (Rahmani, Mahvi, Mesdaghinia, & Nasseri, 2004). It was reported by Jorgensen and Weatherley (2008) that zeolite can be used for removing ammonium from wastewater.

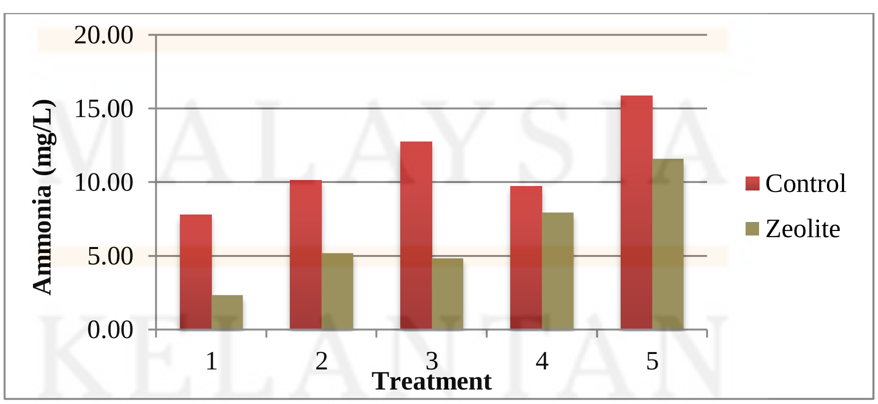

Figure 4.10: Comparison of ammonia concentration between control and zeolite treatments.

In this study, the mean concentrations of BOD and COD in zeolite treatments were significantly ($p<0.05$) lower than control treatments. These findings are in agreement with Ghiasi and Jasour (2012) who reported that zeolite reduce the concentrations of BOD and COD in the treatment water. BOD and COD is the measurement of oxygen required to decompose feed residuals and metabolic waste products produced by fish. During decomposition, nitrifying bacteria require oxygen to convert the ammonia produced by the fish to nitrate by nitrification. Ammonia and ammonium are first converted to nitrite and then nitrite is converted to nitrate. In the treatments containing zeolite, some ammonium ions in treatment water were removed by zeolite. Therefore, the decreased of total ammonium nitrogen in treatments with zeolite had indirectly reduce the oxygen consumed by nitrifying bacteria to transform the ammonium ions into nitrite and nitrate ions.

Based on the recorded water quality, treatments with zeolite is more suitable for the red tilapia culture, because the water quality is better than other treatments. Compare with the control, zeolite supplemented closed system had higher concentrations of dissolved oxygen and lower levels of toxic ammonia, nitrite, nitrate, BOD, and COD , which is more ideal for tilapia culture.

CHAPTER 5

CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

In conclusion, the objectives of this experiment are achieved. This research is vital to investigate the effect of stocking density on the water quality and growth performance of fish in zeolite supplemented closed system. The physico-chemical properties of treatment water in zeolite supplemented closed system including pH, temperature, DO, salinity, TDS, turbidity, BOD, COD, NO_2N, NO_3N, and NH_3 were determined. Results showed that T_1 of the lowest stocking density (5 fish/aquarium) were the most suitable stocking density for the red tilapia, because T_1 recorded the highest concentrations of dissolved oxygen and lowest levels of toxic ammonia, nitrite, and nitrate.

Besides, the growth performance of red tilapia at different stocking densities in zeolite supplemented closed system was also determined. It was found that there was a significant differences ($p<0.05$) in specific growth rate and length gain among treatments. According to the results recorded, red tilapia stocked in the T_1 of the lowest stocking density (5 fish) had the highest growth performance than the fish in other treatments. Compared to other treatments, T_1 recorded the highest final length and weight, weight and length gain, SGR. As conclusion, T_1 with the lowest fish density is the most suitable stocking density for the red tilapia.

Next, the water quality and growth performance of red tilapia between control and zeolite treatments were also compared. Based water quality parameters recorded, there was no significant difference ($p>0.05$) among zeolite and without zeolite in salinity, total dissolved solids, and turbidity. On the other hand, it was found that there was no significant differences ($p>0.05$) in growth parameters among zeolite and control.

5.2 Recommendation

This study was carried out to determine the effect of stocking density on growth performance of red tilapia and water quality in the closed system. Water quality parameters in each aquarium were monitored every week. However, there are some elements that would also affect the water quality and growth performance did not evaluated in this study. The research can be extended by evaluating other important elements in water such as phosphorus and total suspended solids.

In the current study, zeolite was added into different stocking density treatments to improve water quality. The research can be extended using other types of ion exchangers including activated carbons, and probiotics products such as effective microorganisms. Therefore, the efficiency of different ion exchangers in improving water quality can be assessed and compared.

Lastly, the duration of the experiment should be extended to a longer period. This is because the size of fish stocked in the aquarium is still small within two months. Therefore, the results obtained in this research only reflect the ideal stocking density for small fish in the aquarium but not for large fish. In fact, the ideal stocking density is affected by the carrying capacity (the stocking density of fish that an aquarium can sustain) of an aquarium which depends on the fish size. The carrying capacity for the small fish is higher than the large fish. This showed that the ideal stocking density for large fish is not same as small fish. Therefore, the experiment period need to be extended to determine the ideal stocking density for large fish.

REFERENCES

Alaeldein M. Abudabos, Emad M. Samara, Elsayeid O.S. Hussein, Mu'ath Q. Al-Ghadi & Raed M. Al-Atiyat. (2016). Impacts of Stocking Density on the Performance and Welfare of Broiler Chickens. Italian Journal of Animal Science, 12 (1), 66-71.

Al-Harbi, A. H., & Siddiqui, A. Q. (2000). Effects of Tilapia Stocking Densities on Fish Growth and Water Quality in Tanks. *Asian Fisheries Science*, *13* (2000), 391–396.

Abdel-Rahim, M. M. (2017). Sustainable Use of Natural Zeolites in Aquaculture: A Short Review. Oceanography & Fisheries Open Access Journal. Sustainable Use of Natural Zeolites in Aquaculture, *2* (4), 1-5.

Aksungur N., Akbulut B. and Kutlu I. (2007). Effects of stocking density on growth performance, survival and food conversion ratio of Turbot (Psetta maxima) in the net cages on the southeastern coast of the Black Sea. Turkish J. Fish. Aquat. Sci., 7: 147-152.

Barraza, C. A. H. (2010). *Analyses Of Productivity Of Nile Tilapia (Oreochromis Niloticus), Red Tilapia (O . Niloticus X O . Mossambicus) And Pacific White Shrimp (Litopenaeus Vannamei) Polyculture In A Recirculating System (Doctoral dissertation, University of Arizona, Tucson, United States.*

Boyd, C. E., & Lichtkoppler, F. (1979). Water Quality Management in pond fish culture Research and Development Series, *22*(22).

Boyd CE. Farm-level issues in aquaculture certification, 2004. Tilapia.http://www.aces.edu/dept/fisheries/aquaculture/documents/WWFboyd.pdf

Chakraborty, S. B., & Banerjee, S. (2010). Effect of Stocking Density on Monosex Nile Tilapia Growth during Pond Culture in India. *International Journal of Biological, Biomolecular, Agricultural, Food and Biotechnological Engineering*, *4* (8), 646–650.

Chowdhury, M. A. K., Bureau, D. P., Bose, M. L., & Dey, M. M. (2007). Relevance of a rapid appraisal approach to identify locally available feed ingredients to small-scale nile tilapia (Oreochromis niloticus). Aquaculture Economics and Management, 11 (2), 151-169.

Daudpota, A. M., Kalhoro, I. B., Shah, S. A., Kalhoro, H., & Abbas, G. (2014). Effect of stocking densities on growth , production and survival rate of red tilapia in hapa at fish hatchery Chilya Thatta , Sindh , Pakistan. *Journal of Fisheries*, *2* (3), 180–186.

Department of Fisheries, 2010. Annual fisheries statistics 2010. Department of Fisheries, Ministry of Agriculture and Agro-Based Industry, Malaysia.

Department of Fisheries, 2012. News letter 2012. Department Fisheries, Ministry of Agriculture and Agro-Based Industry, Malaysia.

DAMBO, W. B., & RANA, K. J. (1993). Effect of stocking density on growth and survival of Oreochromis niloticus (L.) fry in the hatchery. *Aquaculture Research, 24*(1), 71–80. https://doi.org/10.1111/j.1365-2109.1993.tb00829.x

Danabas, D., & Altun, T. (2011). Effects of zeolite (Clinoptilolite) on some water and growth parameters of rainbow trout (Oncorhynchus mykiss Walbaum, 1792). *Digest Journal of Nanomaterials and Biostructures*, *6*(3), 1111–1116.

Devi, P. A., Padmavathy, P., Aanand, S., & Aruljothi, K. (2017). Review on water quality parameters in freshwater cage fish culture. *International Journal of Applied Research*, *3*(5), 114–120. Retrieved from www.allresearchjournal.com

Eshchar, M., Lahav, O., Mozes, N., Peduel, A., & Ron, B. (2006). Intensive fish culture at high ammonium and low pH. *Aquaculture*, *255* (1–4), 301–313.

Eknath, A. E., & Hulata, G. (2009). Use and exchange of genetic resources of Nile tilapia (Oreochromis niloticus). *Reviews in Aquaculture*, *1* (3–4), 197–213.

FAO, & Aquaculture, F. and. (2007). *The State of World Fisheries and Aquaculture 2006*. https://doi.org/10.5860/CHOICE.50-5350

Ferdous, J., Hossain, M. M., Jaman, H. U., Rupom, A. H., Tonny, N. I., & Jaman, A. (2017). Psidium Guajava Leaf Extracts Fed to Mono-sex Nile Tilapia Oreochromis Niloticus Enhance Immune Response Against Pseudomonas fluorescens. European Journal of Clinical and Biomedical Sciences, *3* (1), 34–42.

Ghiasi, F., & Jasour, M. S. (2012). Performance and Nutritional Parameters of fresh water aquarium fish , Angel, *2*(1982), 22–25.

Gupta, M. V, & Acosta, B. O. (2004). A review of global tilapia farming practices. Aquaculture, *Vol. IX* (January 2004), 7–12.

Ghasemi, Z., Sourinejad, I., Kazemian, H., & Rohani, S. (2016). Application of zeolites in aquaculture industry: A review. *Reviews in Aquaculture*, 1, 1–21.

Ghiasi, F., & Jasour, M. S. (2012). The Effects of Natural Zeolite (Clinoptilolite) on Water Quality, Growth Performance and Nutritional Parameters of fresh water aquarium fish , Angel (Pteropgyllum scalare). International Journal of Research in Fisheries and Aquaculture, *2* (1982), 22–25.

Hamzah, A., Nguyen Hong Nguyen, Ponzoni, R. W., Kamaruzzaman, B. N., & Subha, B. (2008). Performance and survival of three red tilapia strains (Oreochromis spp.) in pond environment in Kedah state, Malaysia. *8th Symposium on Tilapia in Aquaculture*, 199–211.

Hashim, R., Chong, A. S. ., Fatan, N. A., Layman, N., & Ali, A. (2002). Evaluation of Commercial Shrimp Grow-Out Pellets as Diets for Juvenile Southern Rock Lobster , Jasus edwardsii. *Journal of Applied Aquaculture*, *12* (3), 43–57.

Hoseinzadeh Hejazi, S. A. (2011). *Characterization of natural zeolite membranes for molecular hydrogen/carbon dioxide separations by single gas permeation. (master's thesis, University of Alberta, Ottawa, Canada).* Retrieved from https://search.proquest.com/docview/1220485524?accountid=51152

Ibrahim, A. B., Mohd Khan, A., Norrakiah, A. S., & Intan Fazleen, Z. (2014). Fresh water aquaculture fish consumption in Malaysia and heavy metals risk exposure to consumers. *International Food Research Journal*, *21*(6), 2109–2113.

Iliyasu, A., & Mohamed, Z. A. (2016). *Evaluating contextual factors affecting the technical efficiency of freshwater pond culture systems in Peninsular Malaysia: A two-stage DEA approach*. *Aquaculture Reports*, 3, 12-17.

Iliyasu, A., Mohamed, A. Z., & Terano, R. (2016). *Comparative analysis of technical efficiency for different production culture systems and species of freshwater aquaculture in Peninsular Malaysia*. *Aquaculture Reports*, 3, 51-57.

Jiménez-Ojeda, Y. K., Collazos-Lasso, L. F., & Arias-Castellanos, J. A. (2018). Dynamics and use of nitrogen in biofloc technology - BFT. *AACL Bioflux*, *11*(4), 1107–1129.

Jorgensen, T.C. and Weatherley, L.R. (2008) Continuous ion-exchange removal of ammonium ion onto clinoptilolite in the presence of contaminants. Asia-Pac. J. Chem. Eng. 3, 57–62.

Khatune-Jannat, M., Rahman, M. M., Bashar, M. A., Hasan, M. N., Ahamed, F., & Hossain, M. Y. (2012). Effects of stocking density on survival, growth and production of Thai climbing perch (Anabas testudineus) under fed ponds. *Sains Malaysiana*, *41*(10), 1205–1210.

Murugesan, R., Soundarapandian, P., & Manivannan, K. (2011). Fisheries and Aquatic Science. *Fisheries and Aquatic Science*, *3*(1), 82–86. https://doi.org/10.1146/annurev.ecolsys.110308.120220

Mitch Hunter. (2017). We don’t need to double world food production by 2050-here’s why. Retrieved April, 20, 2018, from http://theconversation.com/we-dont-need-to-double-world-food-production-by-2050-heres-why-74211

Moogouei.R, Karbassi A.R, Monavari S.M, Rabani M., & Taheri Mirghaed A. (2010). Effect of the selected physico-chemical parameters on growth of rainbow trout (Oncorhynchus mykiss) in raceway system in Iran. Iranian Journal of Fisheries Sciences, *9* (2), 245–254.

Md. Arif Chowdhury & Khairun Yahya. (Eds.). (2012). Sustainable seafood production status in Malaysia and comparison with the world. Pulau Pinang, Malaysia: Center for Marine and Coastal Studies.

Myers, D. N. (2014). Why monitor water quality?. Retrieved March, 15, 2018, from http://water.usgs.gov/owq/WhyMonitorWaterQuality.pdf

Mumpton, F. A., & Fishman, P. H. (1977). The application of natural zeolites in animal science and aquaculture. *Journal of Animal Science*, *45*(5), 1188–1203.

Makori, A. J., Abuom, P. O., Kapiyo, R., Anyona, D. N., & Dida, G. O. (2017). Effects of water physico-chemical parameters on tilapia (Oreochromis niloticus) growth in earthen ponds in Teso North Sub-County, Busia County. *Fisheries and Aquatic Sciences*, *20*(1), 1–10. https://doi.org/10.1186/s41240-017-0075-7

Najiah, M., Aqilah, N. I., Lee, K. L., Khairulbanyyah, Z., Mithun, S., Jalal, K. C. A., … Nadirah, M. (2012). Massive mortality associated with Streptococcus agalactiae infection in cage-cultured red hybrid tilapia Oreochromis niloticus in Como River, Kenyir Lake, Malaysia. *Journal of Biological Sciences*, *12*(8), 438–442.

Onder Yildirim, Turker, A., & Bilgin Senel. (2009). in Fish Diet on Water Quality , Growth Performance, *18*(9), 1567–1571.

Ofor, C. O., & Afia, O. E. (2015). Effect of Stocking Densities on Growth and Feed Utilization of Hybrid Catfish Effect of Stocking Densities on Growth and Feed Utilization of Hybrid Catfish (Clarias gariepinus X Heterobranchus longifilis) Fed at 1 % Body Weight, (October).

Ouattara, N., Teugels, G., Douba, V., & Philippart, J. (2003). Aquaculture potential of the black-chinned tilapia, Sarotherodon melanotheron (Chiclidae): comparative study of the effect of stocking density on growth performance of landlocked and natural populations under cage culture conditions in Lake Ayame (Cote d'Ivoire). Aquaculture Research, 32, 1223-1229.

Rahman, S. A., Zambry, H., Basha, S., Kamarzaman, S., & Chowdhury, A. J. K. (2013). The potential role of red tilapia (Oreochromis Niloticus) scales: Allergic reaction test in mice. *Journal of Applied Pharmaceutical Science*, *3*(10), 45–50.

Roberts H.., 2010. Fundamentals of ornamental fish health. Ames, Iowa: Wiley-Blackwell.

Rahman, M. (2016). Effects of stocking density on growth and production performance of Monosex male tilapia (Oreochromis niloticus) in earthen ponds. *International Journal of Fisheries and Acquatic Studies*, *4*(3), 267–271.

Rahmani, A. R., Mahvi, A. H., Mesdaghinia, A. R., & Nasseri, S. (2004). Investigation of ammonia removal from polluted waters by Clinoptilolite zeolite. *International Journal of Environmental Science & Technology*, *1*(2), 125–133. https://doi.org/10.1007/BF03325825

Ronald, N., Gladys, B., & Gasper., E. (2014). The Effects of Stocking Density on the Growth and Survival of Nile Tilapia (Oreochromis niloticus) Fry at Son Fish Farm, Uganda. *Journal of Aquaculture Research & Development*, *5*(2), 1-7.

Stickney, Robert.R., (2005). AQUACULTURE: AN INTRODUCTORY TEXT. UK: CABI Publishing.

Safaa M. Ezzat, R. M. El. (2012). The Economical Value of Nile Tilapia Fish "Oreochromis niloticus" in Relation to Water Quality of Lake Nasser, Egypt. *Journal of American Science*, *8*(9), 234–247.

Sheppard, R. a, & Gude, a J. (1969). Chemical Composition and Physical Properties of the Related Zeolites Offretite and Erionite. *American Mineralogist*, *54*(1967), 875.

Şahin, D., Aral, O., & Öz, M. (2016). The Effect of Natural Zeolite Clinoptilolite on Aquarium Water Conditions. *Hacettepe Journal of Biology and Chemistry*, *2*(44), 203–203.

Timmer, C. P. (2014). Food Security in Asia and the Pacific: The Rapidly Changing Role of Rice. *Asia and the Pacific Policy Studies*, *1*(1), 73–90.

Tacon, A. G. J., & Metian, M. (2013). Fish Matters: Importance of Aquatic Foods in Human Nutrition and Global Food Supply. *Reviews in Fisheries Science*, *21*(1), 22–38.

Tilak, K. S., Veeraiah, K., & Milton Prema Raju, J. (2007). Effects of ammonia, nitrite and nitrate on hemoglobin content and oxygen consumption of freshwater fish, Cyprinus carpio (Linnaeus). *Journal of Environmental Biology*, *28*(1), 45–47.

Timmons, M.B., Ebeling, J.M., Wheaton, F.W., Summerfelt, S.T., Vinci, B.J., 2001. Recirculating Aquaculture Systems. Cayuga Aqua Ventures, 126 Sunset Drive, Ithaca, NY 14850. 650 pp.

Verster, N. (2017). *Comparison of Growth Rates of Tilapia Species (Oreochromis Mossambicus and Oreochromis Niloticus) Raised in a Biofloc and a Standard Recirculating Aquaculture (Ras) System* (master's thesis). Ghent University,St. Pietersnieuwstraat, Belgium.

Virta, R. L. (2001). *Zeolites*. *U.S. Geological Surveys Yearbook*.United States: U. S. Environmental Protection Agency.

Yusoff, A. (2015). Status of resource management and aquaculture in Malaysia. In M. R. R. Romana-Eguia, F. D. Parado-Estepa, N. D. Salayo, & M. J. H. Lebata-Ramos (Eds.), In *International Workshop on Resource Enhancement and Sustainable Aquaculture Practices in Southeast Asia 2014* (pp. 52–65). Malaysia: Institute of Bioscience.

Yadav, C. N. R. (2006). Tilapia -An Introduction and Prospects of its Culture in Nepal. *Our Nature*, *4*, 107–110.

Yusoff, F. M., Banerjee, S., Khatoon, H., & Shariff, M. (2011). *Biological Approaches in Management of Nitrogenous Compounds in Aquaculture Systems. In Fatimah (Ed), Dynamic Biochemistry, Process Biotechnology and Molecular Biology* (pp. 21-33). Serdang: Global Science Books.

Yousefian, M.., Hedayatifard, M., Farabi, S.V., Bigom Norouzian, M., Nikkho, M., Makhtomi, Ch. and Noori, A. (2010). The effects of zeolite on growth parameters of common carp of Caspian Sea. Journal of Fisheries, 4(3), 101-109

Zweigh RD (1989) Evolving water quality in a common carp and blue tilapia high production pond. Hydrobiologia 171: 11-21.

APPENDIX A

Weekly values for each water quality parameter, standard deviation (SD), minimum (Min), maximum (Max), and average values of different treatment water in control treatment. T1= 5 fish, T2= 10 fish, T3= 15 fish, T4= 20 fish, T5= 25 fish.

A.1 Physical Parameters

Temperature (°C)													
	1	2	3	4	5	6	7	8	9	SD	Average	Min	Max
T1	25.97	25.91	26.06	27.50	26.29	25.13	25.60	24.04	25.42	0.93	25.77	24.04	27.50
T2	26.30	26.40	26.55	27.20	26.86	25.65	26.12	24.65	25.73	0.75	26.16	24.65	27.20
T3	26.41	26.54	26.65	27.76	26.86	25.75	26.15	24.65	25.89	0.86	26.29	24.65	27.76
T4	26.63	26.67	26.74	27.34	26.67	25.68	25.99	24.64	25.83	0.80	26.24	24.64	27.34
T5	26.85	26.87	27.00	27.95	27.21	26.08	26.48	25.10	26.35	0.80	26.65	25.10	27.95
Dissolved oxygen (mg/L)													
T1	4.77	5.24	4.64	3.92	2.88	3.53	2.44	3.20	1.90	1.13	3.61	1.90	5.24
T2	4.25	3.64	4.53	1.53	0.91	3.52	2.76	3.07	2.34	1.20	2.95	0.91	4.53
T3	3.40	2.99	3.87	3.77	2.75	3.38	2.04	1.00	1.15	1.08	2.70	1.00	3.87
T4	3.84	3.94	3.33	1.08	0.36	3.37	2.08	1.81	1.44	1.30	2.36	0.36	3.94
T5	1.54	2.56	1.91	1.41	1.60	1.82	1.12	0.86	0.91	0.54	1.52	0.86	2.56

Potential hydrogen (pH)													
T1	6.22	6.24	6.18	5.85	5.66	6.10	6.25	5.78	5.63	0.26	5.99	5.63	6.25
T2	6.89	5.99	6.03	6.48	6.71	5.87	6.07	6.03	6.17	0.36	6.25	5.87	6.89
T3	6.34	5.94	5.87	5.43	5.38	5.87	7.19	7.43	7.36	0.81	6.31	5.38	7.43
T4	6.49	5.83	5.97	6.60	6.66	5.98	6.59	6.44	7.25	0.44	6.42	5.83	7.25
T5	7.00	5.93	6.30	6.49	6.57	6.90	7.07	6.81	7.26	0.42	6.70	5.93	7.26
Salinity (ppt)													
	1	2	3	4	5	6	7	8	9	SD	Average	Min	Max
T1	0.08	0.09	0.08	0.08	0.08	0.09	0.12	0.09	0.11	0.01	0.09	0.08	0.12
T2	0.10	0.10	0.10	0.11	0.14	0.10	0.14	0.12	0.11	0.02	0.11	0.10	0.14
T3	0.14	0.12	0.11	0.12	0.12	0.13	0.14	0.16	0.15	0.02	0.13	0.11	0.16
T4	0.13	0.12	0.14	0.12	0.12	0.09	0.10	0.12	0.13	0.02	0.12	0.09	0.14
T5	0.14	0.14	0.14	0.14	0.14	0.15	0.16	0.16	0.18	0.01	0.15	0.14	0.18
Total Dissolved Solids (mg/L)													
T1	117.00	118.00	113.00	110.00	116.00	122.50	159.00	129.00	149.00	16.99	125.94	110.00	159.00
T2	143.00	138.00	143.00	148.00	192.00	145.00	186.00	166.50	147.00	20.10	156.50	138.00	192.00
T3	190.00	164.00	152.00	163.00	172.00	176.00	188.00	217.00	212.50	22.29	181.61	152.00	217.00
T4	186.00	171.00	189.00	164.00	161.00	124.00	131.50	167.50	184.00	23.01	164.22	124.00	189.00
T5	195.00	193.00	192.00	191.00	186.00	210.00	226.00	223.50	240.00	19.33	206.28	186.00	240.00
Turbidity (NTU)													
T1	13.90	12.10	10.60	9.55	13.50	13.95	20.40	25.60	21.75	5.53	15.71	9.55	25.60
T2	21.70	21.10	16.00	32.50	52.30	27.45	30.80	37.80	30.75	10.70	30.04	16.00	52.30

T3	46.80	35.40	19.30	42.60	50.20	30.45	61.75	36.15	74.55	16.68	44.13	19.30	74.55
T4	33.30	62.70	39.00	58.80	35.50	22.00	32.15	29.55	71.00	17.05	42.67	22.00	71.00
T5	66.80	52.60	53.30	34.80	53.50	56.45	59.25	70.55	66.75	10.71	57.11	34.80	70.55

A.2 Chemical Parameters

	Chemical oxygen demand (mg/L)												
	1	2	3	4	5	6	7	8	9	SD	Average	Min	Max
T1	36.0	90.0	71.0	73.0	103.0	74.0	98.0	75.5	201.0	45.5	91.3	36.0	201.0
T2	61.0	114.0	79.0	180.0	172.0	181.0	154.5	108.0	156.0	44.9	133.9	61.0	181.0
T3	171.0	201.0	146.0	187.0	132.0	149.0	185.0	129.0	203.0	28.8	167.0	129.0	203.0
T4	178.0	210.0	206.0	195.0	157.0	141.0	166.0	118.5	199.0	31.5	174.5	118.5	210.0
T5	172.0	191.0	205.0	161.0	134.0	200.0	199.0	186.0	187.0	22.7	181.7	134.0	205.0
	Biochemical oxygen demand (mg/L)												
T1	8.19	14.01	15.63	16.86	12.69	15.42	15.57	15.48	19.32	3.07	14.80	8.19	19.32
T2	10.62	15.03	16.77	19.56	19.53	14.91	15.66	15.15	19.77	2.98	16.33	10.62	19.77
T3	16.41	18.42	20.70	20.40	20.22	16.84	16.81	17.55	20.88	1.86	18.69	16.41	20.88
T4	17.52	21.24	23.07	19.71	19.32	12.64	13.18	15.45	19.29	3.56	17.93	12.64	23.07
T5	16.68	23.16	21.54	19.41	20.37	17.47	17.79	17.79	20.97	2.19	19.46	16.68	23.16
	Nitrite (mg/L)												
T1	5.070	4.580	5.090	5.000	4.440	5.025	0.450	0.420	0.011	2.301	3.343	0.011	5.090
T2	5.190	4.600	5.200	4.790	4.270	5.190	4.730	2.060	2.470	1.187	4.278	2.060	5.200
T3	4.860	4.540	5.170	4.560	4.630	5.210	0.225	1.965	1.495	1.871	3.628	0.225	5.210
T4	4.970	4.420	5.070	3.700	4.270	5.025	5.140	3.655	2.306	0.938	4.284	2.306	5.140
T5	4.520	4.650	5.120	4.440	4.510	5.345	0.150	2.830	2.436	1.674	3.778	0.150	5.345

Nitrate (mg/L)													
	1	2	3	4	5	6	7	8	9	SD	Average	Min	Max
T1	31.90	18.00	15.50	15.00	12.00	14.75	5.50	3.00	4.00	8.90	13.29	3.00	31.90
T2	70.00	35.50	19.00	11.50	14.50	20.75	14.25	26.00	20.50	18.07	25.78	11.50	70.00
T3	70.00	29.50	22.00	6.50	4.50	10.00	10.50	3.50	3.05	21.56	17.73	3.05	70.00
T4	68.00	17.00	31.00	5.00	10.50	13.75	21.00	15.50	3.55	19.61	20.59	3.55	68.00
T5	71.00	26.00	28.00	9.50	8.50	6.00	8.00	20.00	9.05	20.61	20.67	6.00	71.00
Ammonia (mg/L)													
T1	6.40	5.65	9.60	6.20	5.90	7.10	10.30	8.90	10.00	1.90	7.78	5.65	10.30
T2	8.70	10.00	13.40	9.50	13.10	4.00	11.40	10.50	10.70	2.78	10.14	4.00	13.40
T3	10.20	11.10	10.40	13.40	11.60	12.80	22.60	10.20	12.50	3.87	12.76	10.20	22.60
T4	10.60	7.60	11.80	12.60	12.20	3.20	9.40	10.40	9.80	2.90	9.73	3.20	12.60
T5	13.40	12.40	17.60	15.60	15.20	16.20	27.20	12.10	13.20	4.63	15.88	12.10	27.20

APPENDIX B

Weekly values for each water quality parameter, standard deviation (SD), minimum (Min), maximum (Max), and average values of different treatment water with zeolite.

T1= 5 fish, T2= 10 fish, T3= 15 fish, T4= 20 fish, T5= 25 fish.

B.1 Physical Parameters

	Temperature (°C)												
	1	2	3	4	5	6	7	8	9	SD	Average	Min	Max
T1	25.70	25.70	25.98	27.29	26.48	25.22	25.74	24.34	25.11	0.84	25.73	24.34	27.29
T2	25.71	25.71	26.19	27.32	26.26	25.16	25.58	24.22	24.69	0.91	25.65	24.22	27.32
T3	25.55	25.40	26.21	27.39	26.18	25.35	25.45	24.28	24.86	0.89	25.63	24.28	27.39
T4	25.66	25.65	26.64	27.70	26.71	25.77	25.92	24.76	25.59	0.85	26.04	24.76	27.70
T5	26.06	26.06	26.42	27.43	26.42	25.56	25.84	24.54	25.33	0.80	25.96	24.54	27.43
	Dissolved oxygen (mg/L)												
T1	5.30	4.28	3.78	4.32	3.27	3.96	2.67	3.40	2.69	0.84	3.74	2.67	5.30
T2	3.84	3.90	4.14	3.09	2.58	2.53	2.05	2.17	1.98	0.85	2.92	1.98	4.14
T3	5.16	4.11	5.01	3.34	3.30	3.79	3.32	3.40	2.02	0.96	3.71	2.02	5.16
T4	3.63	3.71	3.51	2.90	2.05	3.00	2.01	1.67	1.57	0.86	2.67	1.57	3.71
T5	3.57	3.44	3.69	3.02	1.76	2.37	1.71	1.79	1.52	0.89	2.54	1.52	3.69

Potential hydrogen (pH)													
T1	6.39	6.18	6.24	5.93	5.98	5.98	6.65	6.23	6.55	0.25	6.24	5.93	6.65
T2	6.73	6.08	6.20	6.41	6.23	6.34	6.81	6.55	6.98	0.31	6.48	6.08	6.98
T3	6.61	6.10	5.91	6.35	5.84	6.07	6.55	7.07	7.04	0.46	6.39	5.84	7.07
T4	7.19	6.41	6.34	6.50	6.20	6.17	6.71	7.54	7.52	0.55	6.73	6.17	7.54
T5	7.04	6.70	6.30	6.38	6.17	6.67	7.21	7.36	7.43	0.47	6.80	6.17	7.43
Salinity (ppt)													
	1	2	3	4	5	6	7	8	9	SD	Average	Min	Max
T1	0.07	0.07	0.07	0.08	0.09	0.10	0.10	0.10	0.12	0.02	0.08	0.07	0.12
T2	0.11	0.09	0.09	0.10	0.09	0.11	0.13	0.14	0.12	0.02	0.11	0.09	0.14
T3	0.11	0.09	0.11	0.11	0.12	0.12	0.13	0.12	0.17	0.02	0.12	0.09	0.17
T4	0.13	0.10	0.11	0.12	0.13	0.12	0.15	0.17	0.18	0.03	0.13	0.10	0.18
T5	0.15	0.14	0.12	0.14	0.13	0.13	0.15	0.15	0.16	0.01	0.14	0.12	0.16
Total Dissolved Solids (mg/L)													
T1	91.0	87.0	92.5	110.5	118.5	133.5	133.0	134.5	163.5	25.5	118.2	87.0	163.5
T2	158.5	127.0	122.0	131.0	128.0	145.0	175.5	175.5	170.0	22.1	148.1	122.0	175.5
T3	151.0	122.0	143.0	149.0	162.0	158.5	180.5	180.5	222.5	28.7	163.2	122.0	222.5
T4	177.5	138.0	150.0	162.0	176.5	165.5	206.5	206.5	240.5	32.2	180.3	138.0	240.5
T5	200.0	188.5	165.0	182.5	181.5	178.0	198.0	198.0	213.0	14.4	189.4	165.0	213.0
Turbidity (NTU)													
T1	18.95	16.25	17.03	10.70	14.95	13.02	23.45	23.29	23.29	4.72	17.88	10.70	23.45
T2	25.20	16.50	33.45	18.80	26.10	24.35	49.80	52.35	52.35	14.53	33.21	16.50	52.35

T3	27.85	19.70	29.45	36.30	38.10	52.65	56.35	66.40	66.40	17.27	43.69	19.70	66.40
T4	29.15	23.35	32.75	35.60	33.05	83.20	103.40	145.50	145.50	50.59	70.17	23.35	145.50
T5	54.25	42.90	48.55	55.20	48.10	70.25	64.50	62.80	62.80	9.07	56.59	42.90	70.25

B.2 Chemical Parameters

	Chemical oxygen demand (mg/L)												
	1	2	3	4	5	6	7	8	9	SD	Average	Min	Max
T1	20.5	60.5	57.0	54.5	74.5	40.0	83.0	29.5	77.5	21.7	55.2	20.5	83.0
T2	35.5	77.5	88.0	101.5	85.0	75.5	129.0	98.0	109.5	26.1	88.8	35.5	129.0
T3	45.5	81.0	75.5	77.5	71.5	106.0	99.5	93.0	203.5	44.4	94.8	45.5	203.5
T4	42.0	95.0	101.5	129.0	98.0	111.0	140.0	146.5	91.5	31.3	106.1	42.0	146.5
T5	135.0	108.5	128.5	124.0	116.0	126.0	137.0	116.0	125.5	9.3	124.1	108.5	137.0
	Biochemical oxygen demand (mg/L)												
T1	7.02	10.47	12.41	14.36	13.38	14.51	13.98	12.74	18.66	3.16	13.06	7.02	18.66
T2	7.50	11.70	15.02	18.36	18.53	16.37	15.98	13.92	20.66	3.97	15.34	7.50	20.66
T3	12.77	13.88	14.78	16.80	19.56	13.13	15.96	13.64	19.20	2.55	15.52	12.77	19.56
T4	15.39	16.59	19.08	17.46	20.19	16.17	17.79	15.63	18.71	1.65	17.45	15.39	20.19
T5	15.69	18.92	21.39	19.77	20.15	17.72	18.44	17.73	20.24	1.72	18.89	15.69	21.39
	Nitrite (mg/L)												
T1	4.410	3.860	4.115	4.760	3.655	3.765	0.049	0.040	0.040	2.053	2.744	0.040	4.760
T2	5.105	4.595	5.140	5.010	4.515	4.410	0.019	1.083	1.258	2.049	3.459	0.019	5.140
T3	4.760	4.565	4.995	3.010	4.105	4.250	0.065	1.127	1.515	1.817	3.155	0.065	4.995
T4	4.185	4.490	4.290	4.320	3.495	4.310	0.059	0.080	0.075	2.074	2.812	0.059	4.490
T5	4.470	4.185	4.610	5.070	3.860	5.065	0.995	0.110	2.600	1.811	3.441	0.110	5.070

Nitrate (mg/L)													
	1	2	3	4	5	6	7	8	9	SD	Average	Min	Max
T1	25.50	10.00	6.00	10.25	9.00	5.75	2.25	7.50	2.75	6.90	8.78	2.25	25.50
T2	23.50	18.50	14.50	17.75	15.50	3.50	2.75	4.70	4.75	7.82	11.72	2.75	23.50
T3	21.50	16.50	8.50	9.50	7.50	6.75	5.25	5.60	10.00	5.43	10.12	5.25	21.50
T4	37.00	15.75	12.25	15.75	9.00	6.75	5.15	9.85	2.65	10.15	12.68	2.65	37.00
T5	70.00	14.50	14.25	15.75	5.75	11.50	1.63	1.77	3.20	21.26	15.37	1.63	70.00
Ammonia (mg/L)													
T1	3.05	0.40	0.58	1.78	2.35	2.35	5.20	2.33	3.05	1.431182	2.34	0.40	5.20
T2	3.85	3.05	1.75	3.50	5.40	5.80	6.15	8.30	8.95	2.403441	5.19	1.75	8.95
T3	2.70	2.25	2.15	1.95	1.55	4.90	5.80	11.60	10.65	3.841856	4.84	1.55	11.60
T4	9.10	5.50	5.55	6.80	9.85	5.30	11.05	6.80	11.50	2.464379	7.94	5.30	11.50
T5	10.80	8.15	6.90	7.05	8.45	8.55	12.30	6.92	35.30	9.073927	11.60	6.90	35.30

APPENDIX C

Table C.1: Statistically difference in final mean length and weight between the mean of treatments with zeolite. Mean difference is significant at the 0.05 level. Mean were tested by MANOVA and ranked by Tukey's multiple range test.

Source	Dependent Variable	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Samplings	Length	6289.868	3	2096.623	65.479	.000	.908
	Weight	4543.234	3	1514.411	7.472	.002	.528
Treatment	Length	1660.841	4	415.210	12.967	.000	.722
	Weight	2684.101	4	671.025	3.311	.031	.398
Samplings * Treatment	Length	247.552	12	20.629	.644	.781	.279
	Weight	2782.016	12	231.835	1.144	.382	.407

Table C.2: Overall comparison of specific growth rate (SGR), weight gain (WG), survival rate (SR), and length gain (LG) between different treatments with zeolite. Mean difference is significant at the 0.05 level. Mean were tested by ANOVA and ranked by Tukey's multiple range tests.

Source	Dependent Variable	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Treatment	SGR	1.939	4	.485	5.418	.046	.813
	WG	40350.897	4	10087.724	3.277	.113	.724
	SR	945.452	4	236.363	.502	.738	.287
	LG	415.135	4	103.784	8.467	.019	.871

Table C.3: Overall comparison of water quality parameters between different treatments with zeolite. Mean difference is significant at the 0.05 level. Mean were tested by MANOVA and ranked by Tukey's multiple range tests.

Source	Dependent Variable	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Week	Temperature	57.719	8	7.215	76.274	.000	.888
	DO	54.731	8	6.841	5.424	.000	.360
	pH	11.297	8	1.412	7.190	.000	.428
	Salinity	47934.964	8	5991.871	8.157	.000	.459
	TDS	42640.500	8	5330.062	8.189	.000	.460
	Turbidity	26394.553	8	3299.319	5.902	.000	.380
	COD	29378.289	8	3672.286	4.955	.000	.340
	BOD	433.930	8	54.241	12.815	.000	.571
	Nitrite	291.580	8	36.448	57.948	.000	.858
	Nitrate	7596.405	8	949.551	18.582	.000	.659
	Ammonia	816.516	8	102.064	5.530	.000	.365
Treatment	Temperature	2.591	4	.648	6.849	.000	.262
	DO	23.645	4	5.911	4.687	.002	.196
	pH	4.016	4	1.004	5.112	.001	.210
	Salinity	157.870	4	39.468	.054	.995	.003
	TDS	58170.572	4	14542.643	22.342	.000	.537
	Turbidity	29548.634	4	7387.159	13.215	.000	.407
	COD	46430.378	4	11607.594	15.663	.000	.449
	BOD	355.887	4	88.972	21.021	.000	.522
	Nitrite	8.204	4	2.051	3.261	.016	.145
	Nitrate	524.432	4	131.108	2.566	.045	.118
	Ammonia	828.542	4	207.136	11.223	.000	.368

APPENDIX D

D.1: Overall comparison of final mean length and weight between zeolite and control treatments.

Source	Dependent Variable	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Treatment	Length	1183.630	4	295.907	1.278	.298	.131
	Weight	1249.383	4	312.346	1.879	.137	.181
Control_Zeolite	Length	3.125	1	3.125	.013	.908	.000
	Weight	23.639	1	23.639	.142	.708	.004

D.2: Overall comparison of specific growth rate (SGR), weight gain (WG), survival rate (SR), and length gain (LG) between zeolite and control in different treatments.

Source	Dependent Variable	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Treatment	SGR	1.062	4	.266	5.209	.069	.839
	WG	18776.821	4	4694.205	3.641	.119	.785
	SR	871.186	4	217.796	.401	.801	.286
	LG	199.385	4	49.846	8.301	.032	.892
Control_Zeolite	SGR	.003	1	.003	.057	.824	.014
	WG	354.501	1	354.501	.275	.628	.064
	SR	1116.615	1	1116.615	2.056	.225	.339
	LG	44.184	1	44.184	7.358	.053	.648

D.3: Overall comparison of water quality parameters between zeolite and control.

Source	Dependent Variable	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Treatment	Temperature	3.387	4	.847	16.088	.000	.459
	DO	28.745	4	7.186	20.398	.000	.518
	pH	4.279	4	1.070	9.094	.000	.324
	Salinity	.031	4	.008	40.912	.000	.683
	TDS	57177.683	4	14294.421	48.108	.000	.717
	Turbidity	21079.804	4	5269.951	16.777	.000	.469
	COD	69377.406	4	17344.351	21.343	.000	.529
	BOD	282.895	4	70.724	18.746	.000	.497
	Nitrite	6.573	4	1.643	2.412	.056	.113
	Nitrate	728.163	4	182.041	2.608	.042	.121
	Ammonia	713.583	4	178.396	12.715	.000	.401
Control_Zeolite	Temperature	3.986	1	3.986	75.730	.000	.499
	DO	5.339	1	5.339	15.154	.000	.166
	pH	.849	1	.849	7.215	.009	.087
	Salinity	.000	1	.000	.713	.401	.009
	TDS	1123.600	1	1123.600	3.781	.056	.047
	Turbidity	915.084	1	915.084	2.913	.092	.037
	COD	70280.278	1	70280.278	86.481	.000	.532
	BOD	43.611	1	43.611	11.559	.001	.132
	Nitrite	12.400	1	12.400	18.204	.000	.193
	Nitrate	1396.336	1	1396.336	20.003	.000	.208
	Ammonia	534.800	1	534.800	38.117	.000	.334

APPENDIX E

E.1: UV-VIS Spectrophotometer

E.2: HACH HQ40d used to test BOD

E.3: Heating water samples by DRB200 Reactor for COD reading

E.4: YSI 556 Multiparameter