



**UNIVERSITI PUTRA MALAYSIA**

**PRODUCTIVITY AND COMPOSITION OF MICROALGAE IN FISH TANK  
IN DIFFERENT MALAYSIAN WEATHER CONDITIONS**

**HAYFAA MOHAMMED SAHAB**

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By

**HAYFAA MOHAMMED SAHAB**

**Thesis submitted to the School of Graduate Studies, Universiti Putra  
Malaysia, in Fulfilment of the Requirements for the Degree of  
Doctor of Philosophy**

**January 2018**

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## **DEDICATION**

**To my late father and dear mother who illuminated my life with care and love.**

**To my supervisor Dr. Hisham, I was not one of your sons to feel happy for me, nor your relative to feel sorrow for me but my rejoice is yours, my sadness is yours. The real persuasions for all this are your love and care for religion.**

**To all members of my family and my friends.**



Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirements of the degree of Doctor of Philosophy

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By

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**January 2018**

**Chairman : Hishamuddin Omar. PhD**  
**Faculty : Science**

Climate change is affecting every country all over the world such as Malaysia. Climate change is the result of global warming which causes changes in daily weather conditions and water bodies. Many scientists study the water quality and microalgae separately from influence of weather conditions. However, there are few researches about effect of different stocking density of fish under different weather conditions on water quality parameters, mixed microalgae species composition, diversity, productivity, biochemical composition, and toxicity of mixed microalgae. Therefore, the objectives of this study was to see effects of changing weather conditions on water quality, diversity and succession of microalgae, primary productivity, biochemical composition, and toxicity of mixed microalgae in different stocking density of fish (Control (no fish), 10, 20, 40, and 80 fish). The weather conditions were monitored three times daily in the morning (8.00-9.00), noon (12.00-13.00), and afternoon (16.00-17.00). The weather is categorized into mix, wet, and dry conditions based on weather scoring. The sampling of five tanks were done every two days and performed outdoor; it's carried out for water quality, microalgae species and diversity, microalgae biomass, productivity, biochemical composition, and toxicity of mixed microalgae. Statistical analysis were done using one way ANOVA, Factorial ANOVA for microalgae parameters, and XLSTAT for toxicity test of microalgae. The mean of water quality parameters of the five tanks during all weather conditions were: water temperature (27.71, 27.83, 27.77, 27.79, 27.96 °C), pH (7.25, 7.03, 7.05, 6.96, 7.04), electrical conductivity (0.15, 0.15, 0.15, 0.16, 0.16 mS/cm), total dissolved solids (0.09, 0.09, 0.09, 0.09, 0.10 mg/L), dissolved oxygen (5.44, 5.31, 5.1, 4.62, 4.13 mg/L), salinity (0.06, 0.06, 0.06, 0.07, 0.07 ppt), secchi disk depth (0.40, 0.17, 0.19, 0.20, 0.22 m), nitrate-nitrogen (0.93, 0.21, 0.21, 0.23, 0.21 mg/L), orthophosphates (0.03, 0.14, 0.14, 0.16, 0.15 mg/L), ammonium-nitrogen (0.012, 0.045, 0.046, 0.048, 0.053 mg/L), alkalinity (41.89, 47.70, 46.76, 44.17, 48.17 mg CaCO<sub>3</sub>/L), total nitrogen (0.23, 0.16, 0.16, 0.17, 0.17 mg/L), total phosphorus (0.008, 0.049, 0.046, 0.049, 0.048 mg/L), TN: TP (32.72,

3.46, 3.45, 3.39, 3.52) respectively. A total of 55 species belonging to 6 divisions (Chlorophyta, Cyanophyta, Chrysophyta, Dinophyta, Bacillariophyta, and Euglenophyta) have been collected from 5 tanks in different weather conditions. The division Chlorophyta was the most dominant in five tanks within different weather conditions comprising of 62.2, 80.3, 81.3, 78.3, and 84.3% in control, tanks 1(10 fish), 2 (20 fish), 3 (40 fish), and 4 (80 fish) respectively. The density of microalgae was high in dry weather conditions but was low in wet weather. The total number of species of microalgae in wet and mix weather conditions were higher than number of species in dry weather conditions. Tank 4 (80 fish) showed the highest chlorophyll concentration of 11.51  $\mu\text{g/L}$  followed by tanks 3, 2, 1, and control [(40, 2, 10 fish) and control (no fish)] 10.75, 9.38, 8.71, and 2, 56  $\mu\text{g/L}$  respectively. The primary productivity was high in tank 4 with high stocking density of fish in dry weather conditions. For biochemical components, the protein contents were high in tanks 4, 3, 2, 1 (49.69, 46.35, 42.82, and 37.55 %) respectively, while the lipid and carbohydrate showed fluctuation rates with (6.83, 8.93, 3.04, 1.78 %), (8.50, 4.55, 1.91, and 2.77%) in tanks (1, 2, 3, and 4) respectively. The percentage of brine shrimp mortality was 70% in concentration 1000  $\mu\text{g/mL}$  in dry weather conditions, while the lowest percentage was 10% in 1  $\mu\text{g/mL}$  in wet weather conditions. The average response of *Artemia* sp. mortality rate against variable dose of extracted mixed microalgae was 538.90, 1256.21, 1444.66, 1222.00 and 1254.13  $\text{mg/L}$  in tanks (control, 1, 2, 3, and 4). The environmental factors such as temperature and light intensity were the main factors that increased the primary productivity of microalgae, whereas the reduction of light intensity in mix and wet weather due to cloud covers led to decrease in microalgae primary productivity. The conclusion of this study demonstrates that temperature and light intensity have impacted the water quality parameters, microalgal diversity and productivity. Typically, the mixed microalgae grown in the fish tank were not toxic for brine shrimp. The relationship between different stocking density of fish and different weather conditions have influenced on the water quality and microalgae growth.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan ijazah Doktor Falsafah

## **PRODUKTIVITI DAN KOMPOSISI MIKROALGA DALAM TANGKI IKAN DALAM KEADAAN CUACA DI MALAYSIA**

Oleh

**HAYFAA MOHAMMED SAHAB**

**Januari 2018**

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Perubahan iklim kini memberi kesan kepada setiap negara di seluruh dunia termasuk Malaysia. Perubahan iklim disebabkan oleh pemanasan global telah menyebabkan perubahan dalam keadaan cuaca harian dan badan air. Ramai saintis telah mengkaji kualiti air dan mikroalga secara berasingan mengenai pengaruh keadaan cuaca. Walau bagaimanapun hanya terdapat sedikit kajian mengenai kesan ketumpatan stok ikan yang berbeza di bawah keadaan cuaca yang berlainan pada parameter kualiti air, komposisi spesies mikroalga campuran, kepelbagaian, produktiviti, komposisi biokimia, dan ketoksikan mikroalga campuran. Sehubungan dengan itu, objektif kajian ini adalah untuk mengetahui kesan keadaan cuaca yang berubah-ubah terhadap kualiti air, kepelbagaian dan penggantian mikroalga, produktiviti primer, komposisi biokimia, dan ketoksikan mikroalga campuran dalam ketumpatan stok ikan yang berbeza (Kawalan (tiada ikan), 10, 20, 40 dan 80 ikan). Keadaan cuaca dipantau tiga kali setiap hari iaitu pada waktu pagi (8.00-9.00), tengah hari (12.00-13.00), dan petang (16.00-17.00). Cuaca dikategorikan kepada keadaan campuran, basah, dan kering berdasarkan skor cuaca. Persampelan untuk lima tangki dijalankan setiap dua hari dan dilakukan di luar; ia dijalankan untuk kualiti air, spesies dan kepelbagaian mikroalga, biojisim mikroalga, produktiviti, komposisi biokimia, dan ketoksikan campuran mikroalga. Analisis statistik dilakukan menggunakan ANOVA sehala, ANOVA faktorial untuk parameter mikroalga dan XLSTAT untuk ujian toksisiti mikroalga. Purata parameter kualiti air lima tangki semasa semua keadaan cuaca adalah: suhu air (27.71, 27.83, 27.77, 27.79, 27.96 ° C), pH (7.25, 7.03, 7.05, 6.96, 7.04), kekonduksian elektrik (0.15, (0.09, 0.09, 0.09, 0.09, 0.10 mg / L), oksigen terlarut (5.44, 5.31, 5.1, 4.62, 4.13 mg / L), kemasinan (0.06 (0.40, 0.17, 0.19, 0.20, 0.22 m), nitrit-nitrogen (0.93, 0.21, 0.21, 0.23, 0.21 mg / L), orthofosfat (0.03, 0.14, 0.14, 0.16, 0.15 mg / L), ammonium-nitrogen (0.012, 0.045, 0.046, 0.048, 0.053 mg / L), alkali (41.89, 47.70, 46.76, 44.17, 48.17 mg CaCO<sub>3</sub> / L), 0.16, 0.16, 0.17, 0.17 mg / L), jumlah fosforus (0.008, 0.049, 0.046, 0.049, 0.048 mg / L), TN: TP (32.72, 3.46, 3.45, 3.39, 3.52). Sebanyak 55 spesies mewakili 6 bahagian

(Chlorophyta, Cyanophyta, Chrysophyta, Dinophyta, Bacillariophyta, dan Euglenophyta) berjaya dicam dari 5 tangki dalam keadaan cuaca yang berbeza. Bahagian Chlorophyta adalah yang paling dominan dalam lima tangki dalam keadaan cuaca yang berbeza yang terdiri daripada 62.2, 80.3, 81.3, 78.3, dan 84.3% masing-masing untuk tangki kawalan, tangki 1 (10 ikan), 2 (20 ikan), 3 (40 ikan), dan 4 (80 ikan). Ketumpatan mikroalga adalah tinggi dalam keadaan cuaca kering dan rendah dalam cuaca basah. Jumlah spesis mikroalga dalam keadaan cuaca basah dan cuaca campuran adalah lebih tinggi berbanding dalam keadaan cuaca kering. Tangki 4 (80 ikan) menunjukkan kepekatan klorofil tertinggi 11.51  $\mu\text{g} / \text{L}$  diikuti oleh tangki 3, 2, 1 dan kawalan [(40, 2, 10 ikan) dan kawalan (tiada ikan)] 10.75, 9.38, 8.71, 2, 56  $\mu\text{g} / \text{L}$  masing-masing. Produktiviti primer adalah tinggi di tangki 4 dengan ketumpatan stok ikan yang tinggi dalam keadaan cuaca kering. Bagi komponen biokimia, kandungan protein adalah tinggi dalam tangki 4, 3, 2, 1 (49.69, 46.35, 42.82, dan 37.55%), manakala lipid dan karbohidrat menunjukkan kadar fluktuasi dengan (6.83, 8.93, 3.04, 1.78% , (8.50, 4.55, 1.91, dan 2. 77%) masing-masing dalam tangki (1, 2, 3, dan 4). Peratusan mortal udang air garam adalah 70% dalam kepekatan 1000  $\mu\text{g} / \text{mL}$  dalam keadaan cuaca kering, manakala peratusan terendah adalah 10% dalam 1  $\mu\text{g} / \text{ml}$  dalam keadaan cuaca basah. Tanggapan purata kadar mortar udang air garam terhadap dos pembolehubah mikroalga bercampur ialah 538.90, 1256.21, 1444.66, 1222.00, dan 1254.13  $\text{mg} / \text{L}$  dalam tangki (kawalan, 1, 2, 3, dan 4). Faktor persekitaran seperti suhu dan keamatan cahaya adalah faktor utama yang meningkatkan produktiviti primer mikroalga, sedangkan pengurangan keamatan cahaya dalam cuaca campuran dan cuaca basah akibat awan menyebabkan penurunan produktiviti primer mikroalga. Kesimpulan kajian ini menunjukkan bahawa suhu dan intensiti cahaya memberi kesan terhadap ciri-ciri kualiti air, kepelbagaian mikroalga dan produktiviti. Biasanya mikroalga bercampur yang terdapat di dalam tangki ikan adalah tidak toksik kepada *Artemia* sp.. Hubungan antara ketumpatan stok ikan yang berbeza dan keadaan cuaca yang berbeza mempengaruhi kualiti air dan pertumbuhan mikroalga.



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This thesis submitted to the Senate of Universiti Putra Malaysia has been submitted as fulfilment of the requirement for the degree of the Doctor of Philosophy. The members of the Supervisory Committee were as follows:

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# CHAPTER 1

## INTRODUCTION

### 1.1 Background

The environmental degradation has been serious issue since late eighteenth and nineteenth centuries. The life style of human and industrial revolution had a great impact on the degradation of the global environment. Global warming is considered as one of the biggest and dangerous contemporary environmental problems that pose a threat to life on the planet due to the devastation it caused on the ecosystem (Riebeek, 2010). Many scientists have pointed out that the production of carbon dioxide by human activities and greenhouse gases emissions trap the solar heats radiations and preventing it to escape from earth surface.

Increasing in the global warming led to climatic change due to rise in atmospheric temperature through burning of fossil fuels (Yao et al., 2013). Climatic changes have several damaging effects like seasonal changes, floods and changes in global weather situation. Several climatic change impacts have been observed; for example glacier retreat, changes in the timing of seasonal events, changes in agricultural productivity (Hansen and Cramer, 2015) and biodiversity throughout increasing the extension of some plants and animal species, loss of habitat, and disruption of aquatic life (Brennan & Owende, 2010). In some places of the world, the rain distribution is the probable outcome of climatic changes. All this changes is caused by emissions of greenhouse gases.

Higher temperatures in strategic producing zones classically result in reduced yields of required crops whereas high weather events for example droughts, floods, and storms can aggravate food security problems. In changing climate, production gains are based on projected rainfall and temperature changes (Yadav et al., 2011).

Freshwater naturally contain different inorganic elements such as carbon, nitrogen and organic compounds which derived from decaying biological materials. Any freshwater body has a separately pattern of physical and chemical features which that effect on water quality.

As all freshwater bodies are interconnected, from the atmosphere to the sea, by means of the hydrological cycle, so these conditions are heavily influenced by climate, geographic location, and type of water body which is determine largely by the climatic changes. The carbon dioxide and oxygen are considered important components in aquatic ecosystem, so both respiration and photosynthesis play a big role to control concentration of these components. In Malaysia, the weather conditions vary due to different temperature and light intensity within the day.

The government, economist, social, cultural rights and scientific researchers are forced to deal with the challenge when designing policies to adapt to climatic changes (Levy and Patz, 2015) to face up a future food crisis and mass emigration as outcome of widespread water shortages due to global warming.

There are various approaches were proposed to reduce carbon dioxide emission such as bioenergy crops by removing carbon dioxide from the air during growth and store it in crop biomass and soil, biofuel production (FAO, 2008). To achieve those aims, the countries must understand the interactions between water, energy, and food under changing climate.

Hence it must look into appropriate and economical way and at the same time environmentally friendly to reduce carbon dioxide emission, so the scientist and researchers preferred using microalgae as a key solution to deal with this problem (Singh et al., 2012). Choosing microalgae cultivation is as a result of their ability to capture carbon and storage processes and utilizing sunlight to convert energy (Chisti, 2007).

Furthermore, microalgae can be grown in different habitats and are capable to tolerate diverse environmental conditions like high salinity, temperature and pH. The processes of microalgae growth can take place in open raceway ponds and closed bioreactor systems (Chen et al., 2011). On the other hand, microalgae are considered as probable candidates for wastewater treatment (Selvaratnam et al., 2015) because of the ability of the microalgae to sequester carbon dioxide from the atmosphere through photosynthesis (Oswald and Golueke, 1966) and oxygenate the treated effluent (Silva et al., 2015). A microalga is considered a primary producer in food chain, fixer carbon, and the main source for oxygen production.

## **1.2 Justification and objectives of the study**

Many reports have revealed that the abundance of microalgae in ponds differ with environmental factors changes like light, temperature, pH and salinity (Teoh et al., 2010). In the past, most studies carried out in the small tanks and samplings were obtained monthly. Numerous studies have shown that monocultures are difficult to maintain, but mixed cultures of two or more strains species or higher taxa may increase and stabilize productivity (Novovesxa et al., 2016). Most microalgae studies are conducted under laboratory conditions (Exton et al., 2013). Although there are some studies monitoring microalgae combination with fish but the studies are limited to different stocking density of fish in changing weather conditions. Furthermore, the expansion of aquaculture is also limited due to land costs. Some studies focus on improve water quality in aquaculture under laboratory-scale conditions based on the principle of waste nutrients recycling (Kuhan et al., 2010). There are limited studies on the effect of fluctuation of weather conditions on microalgae proximate composition.



Variations in proximate composition in algae were studied seasonally in some present studies (Khairy and El-Shafay, 2013) but without mentioning the use of fish. The linkages between toxic algae and climate change in fish ponds are also limited.

Therefore the aim of the present study is to demonstrate daily weather and limnological changes in fish tanks and to show impacts of these changes on species composition, diversity, and succession of mixed microalgae, also proximate composition and toxicity of microalgae. On the other hand there is no need to change water because using biofloc technique reduces water change rates in different aquaculture systems. Therefore, the objectives of this study are:

1. To records growth and diversity of mixed microalgae and identify dominant species in outdoor fish tanks under different weather conditions.
2. To measure physico-chemical water parameters in outdoor fish tanks under variable weather conditions.
3. To compare the productivity of mixed microalgae in fish tanks under different weather conditions.
4. To monitor the biochemical composition and toxicity of mixed microalgae in fish tanks under changing weather conditions.



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## LIST OF PUBLICATONS

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