

**DISTRIBUTION AND DIVERSITY OF ZOOPLANKTON
AS BIOINDICATOR OF WATER QUALITY STATUS IN
AIR ITAM RESERVOIR, PENANG, MALAYSIA**

by

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for the degree of
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**TABURAN DAN KEPELBAGAIAN ZOOPLANKTON
SEBAGAI PENUNJUK BIOLOGI STATUS KUALITI AIR
DI EMPANGAN AIR ITAM, PULAU PINANG,
MALAYSIA**

oleh

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**Tesis yang diserahkan untuk
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UNIVERSITI SAINS MALAYSIA

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LIST OF ABBREVIATIONS

Abbreviation	Description
DO	Dissolved Oxygen
BOD	Biological Oxygen Demand
COD	Chemical Oxygen Demand
TSS	Total Suspended Solids
TDS	Total Dissolved Solids
pH	power of Hydrogen
NO ₂ -N	Nitrite-nitrogen
NO ₃ -N	Nitrate-nitrogen
NH ₄ -N	Ammonium-nitrogen
PO ₄ -P	Phosphate
SS	Suspended Solids
EC	Electrical conductivity
CCA	Canonical Correspondence Analysis
DOE	Department of Environment
WQI	Water Quality Index
INWQS	Interim National Water Quality Standards
s.d	Standard Deviation
Temp	Temperature
APHA	American Public Health Association
Sp.	Species
MPN	Most Probably Number

LIST OF SYMBOLS

Symbol	Description
°C	Degree Celsius
%	Percentage
ml/l	Milligram per liter
cm	Centimeter
µm	Micrometer
mm	Milimeter
m	Meter
µS/cm	Microsiemens per centimeter
ind/l	individu per liter
g/l	Gram per liter

**TABURAN DAN KEPELBAGAIAN ZOOPLANKTON SEBAGAI PENUNJUK
BIOLOGI STATUS KUALITI AIR DI EMPANGAN AIR ITAM, PULAU
PINANG, MALAYSIA**

ABSTRAK

Komposisi dan kelimpahan zooplankton, serta parameter kualiti air telah dikaji di Empangan Air Itam dari Mei 2006 sehingga Februari 2007. Kajian ini mengkaji struktur komuniti zooplankton sebagai penunjuk kualiti air di empangan tersebut, yang telah dibina untuk bekalan air domestik. Zooplankton telah disampel menggunakan jaring plankton Wisconsin dengan liang saiz 80 μm dari jarak vertikal lima meter. Parameter fizikokimia yang telah digunakan sebagai penunjuk kualiti air ialah oksigen terlarut (DO), suhu, pH, permintaan oksigen biologi (BOD), permintaan oksigen kimia (COD), konduktiviti, nitrat, nitrit, orthofosfat, jumlah pepejal terampai (TSS), jumlah pepejal terlarut (TDS), kejernihan air dan jumlah bakteria koliform. Lima stesen penyampelan telah ditempatkan termasuk tiga stesen terletak di empangan (Stesen A, B dan C) dan 2 stesen di bahagian hulu dan hilir (Stesen D dan E). Sebanyak 26 taxa, iaitu 18 Rotifera, enam Cladocera dan empat Copepoda, telah di catatkan. Spesies yang lazim dan dominan ialah *Polyarthra vulgaris*, *Ceriodaphnia cornuta*, *Moina micrura*, *Mesocyclops leuckarti* dan *Thermocyclops crassus*, manakala *Filinia opoliensis*, *Alona karua* dan *Tropocyclops prasinus* dicatatkan sebagai spesies yang kurang. Spesies tersebut merupakan penunjuk kepada pencemaran organik tingkat rendah. Bilangan dan kepelbagaian zooplankton yang rendah telah dicatatkan di Stesen D dan E. Analisis canonical correspondence (CCA) menunjukkan sumbangan kuat daripada $\text{NH}_4\text{-N}$ ($P=0.061$), suhu ($P=0.28$), $\text{PO}_4\text{-P}$ ($P=0.001$), $\text{NO}_2\text{-N}$ ($P=0.003$), $\text{NO}_3\text{-N}$ ($P=0.05$), TDS

($P=0.86$), COD ($P=1.68$), TSS ($P=0.018$) dan pH ($P=0.19$) terhadap assosiasi kelompok zooplankton dan parameter kualiti air. Indeks Kepelbagaian Shannon-Weiner paling tinggi di Stesen C (2.06 ± 0.08 bits/individu) dan terendah di Stesen D (0.90 ± 0.40 bits/individu). Indeks kualiti air (WQI) dihitung menggunakan WQI yang di cadangkan oleh Jabatan Alam Sekitar Malaysia (DOE). Julat WQI di Stesen A, B dan C di antara 92.02 – 93.64 diklasifikasikan sebagai keadaan air bersih sehingga sangat bersih. Stesen D dan E masing-masing menunjukkan nilai WQI 91.55 dan 88.46 di klasifikasikan sebagai air bersih. Purata nilai jumlah koliform di Stesen A, B dan C lebih rendah daripada Stesen D dan E. Nilai tertinggi adalah di Stesen E (96.10 ± 34.31 MPN/100 ml) dan terendah di Stesen B (2.29 ± 1.31 MPN/100 ml). Berdasarkan kepada analisis struktur komuniti zooplankton dan parameter kualiti air, dapat disimpulkan bahawa Empangan Air Itam berada dalam keadaan baik dan bersih sepanjang persampelan. Oleh itu, status kualiti air di empangan ini sesuai untuk bekalan air dan kegunaan manusia.

DISTRIBUTION AND DIVERSITY OF ZOOPLANKTON AS BIOINDICATOR OF WATER QUALITY STATUS IN AIR ITAM RESERVOIR, PENANG, MALAYSIA

ABSTRACT

Zooplankton composition and abundance, and water quality parameters were studied in Air Itam Reservoir from May 2006 to February 2007. This study investigates the community structure of zooplankton as indicators of water quality of the reservoir, which was constructed for domestic water supply. Zooplankton was sampled with a Wisconsin plankton net of 80 μm mesh size, from five meters vertical intervals. Physicochemical parameters used as the indicators of water quality were dissolved oxygen (DO), temperature, pH, biological oxygen demand (BOD), chemical oxygen demand (COD), conductivity, nitrates, nitrites, orthophosphates, total suspended solids (TSS), total dissolved solids (TDS), water transparency and total coliform bacteria. Five sampling stations were designated including three stations located in the reservoir (Stations A, B and C) and two stations in the upstream and downstream (Stations D and E) respectively. A total of 26 taxa, with 18 belong to Rotifera, six Cladocera and four Copepoda, were recorded. The most common and dominant species were *Polyarthra vulgaris*, *Ceriodaphnia cornuta*, *Moina micrura*, *Mesocyclops leuckarti* and *Thermocyclops crassus* while *Filinia opoliensis*, *Alona karua* and *Tropocyclops prasinus* were rare species recorded. These species are considered as the indicators of low level of organic pollution. Lower number and diversity of zooplankton were recorded at Stations D and E. A Canonical Correspondence Analysis (CCA) revealed a strong contribution of

NH₄-N ($P=0.061$), temperature ($P=0.28$), PO₄-P ($P=0.001$), NO₂-N ($P=0.003$), NO₃-N ($P=0.05$), TDS ($P=0.86$), COD ($P=1.68$), TSS ($P=0.018$) and pH ($P=0.19$) to the association of zooplankton assemblage and water quality parameters. Shannon-Weiner's Diversity Index was the highest at Station C (2.06 ± 0.08 bits/individual) and the lowest at Station D (0.90 ± 0.40 bits/individual). Water Quality Index (WQI) was calculated by using the WQI recommended by Malaysian Department of Environment (DOE). The WQI of Stations A, B and C which lies between 92.02 – 93.64 were classified as clean to very clean water conditions. Stations D and E recorded the WQI values of 91.55 and 88.46 respectively which were classified as clean. Mean total coliform at Stations A, B and C was lower than Stations D and E. The highest value was at Station E (96.10 ± 34.31 MPN/100 ml) and the lowest value was at Station B (2.29 ± 1.31 MPN/100 ml). Based on zooplankton community structural analysis and water quality parameters, it can be concluded that Air Itam Reservoir was in good conditions and clean throughout the sampling period. Therefore, the status of water quality in this reservoir is acceptable for water supply and human consumption.

CHAPTER 1

INTRODUCTION

Lakes and reservoirs are important water resources in Malaysia. Distribution of fresh water in the country consisted of 90 man-made lakes with multi purposes. Fifty five lakes are used for water supply and irrigation (61%), while another 35 lakes are used for hydropower, flood control, silt retention and recreational (39%). The total yearly supply of fresh water in the country has been estimated at 456 billion cubic meters. An average precipitation received monthly is between 20 mm to 700 mm, indicating the necessary of building the dams to supply fresh waters in this country (Omar et al., 2002).

An early report produced in 2005 on the eutrophication development claimed that more than 60 per cents out of 90 lakes in Malaysia were nutrient-rich or eutrophic, while the other remaining lakes were believed to be mesotrophic. It has been long well known that lakes and reservoirs play a very important role as the storage of a country's water resources. It would be a very serious concern for both the government and communities if the water quality deteriorates to a level that is dangerous for consumption. Accordingly, various preventive actions always need to be conducted, particularly with regard to the degradation issues so that measures to control and manage the water resources can be developed (Zati and Zulkifli, 2007).

The investigation of freshwater quality has become an important issue in many countries; especially due to concern that freshwater will be a scarce resource in the future so a water quality monitoring program is necessary for the protection of

freshwater resources (Pesce and Wunderlin, 2000). Lately, the reservoirs water quality has been deteriorating due to rapid industrialization in Malaysia. In Semenyih River Basin, Gasim et al. (2005) investigated the hydrological characteristics and water quality in the area in relation to land development in the surrounding areas. They summarized the variety of pollutants discharged into the Semenyih River is classified as sediment, organic chemicals, domestic waste and nutrients. This water quality degradation was influenced by the changes in the land-use and population growth. The increase of pollutants in the river not only became hazardous to biological life in the river, but also resulted in problems at the Semenyih Water Treatment Plant, so it is important that the land-use activities within the basin be carefully planned and controlled to preserve the water resource as well as the quality status.

In biomonitoring of aquatic ecosystem, the use of ecological methods is necessary due to the deterioration of water bodies through anthropogenic activities. The species composition, abundance, productivity and physiological conditions of the aquatic community may influenced by the quality of water. The structure and the composition of these aquatic communities is an indicator of water quality. Therefore, the use of physical, chemical and biological parameters to assess the quality of water can provide accurate information needed for appropriate water management (Iliopoulou-Georgudaki et al., 2003; Ocampo-Duque et al., 2006; Ramachandra et al., 2006).

Tropical aquatic ecosystems today are threatened by a number of serious environmental problems, which must be considered in the near future. Increasing water pollution causes not only the deterioration of water quality but also threatens human health and the balance of aquatic ecosystems, economic development and social

prosperity (Milovanovic, 2007). Without a detailed knowledge about seasonal changes in biota and water quality, it will very difficult to manage lakes and reservoirs (Nilssen, 1984). Limnological study that refers to the fresh and saline inland water bodies is important to provide information for conservation and management of aquatic ecosystems (Páez et al, 2001). The limnological studies had been done in many water bodies of the world to evaluate the water quality of aquatic systems. Many countries have introduced a plan to monitor and assess the pollution effects of aquatic system (Štambuk-Giljanović, 1999; Pesce and Wunderlin, 2002).

In Malaysia, limnological study had been carried out by many researchers and scientists from local universities, government authorities and non-government organizations to protect the freshwater ecosystem (Ho and Peng, 1997). Many programs of water quality monitoring had been conducted to improve and protect the freshwater resources, including the study of physical, chemical and biological impact on rivers and streams, lakes and ponds, reservoirs and wetlands. A study on aquatic organisms in biological monitoring to reflect upon water quality had been done by Wan Maznah and Mansor (2002). They studied the species composition of attached diatoms to test the utility and suitability of diatom community structure as an indicator of river pollution in Pinang River Basin. They found that the highest diversity value and species richness recorded at polluted station in the downstream with higher Saprobic Index values compared to upstream stations.

The monitoring of zooplankton, among various communities in aquatic ecosystem gives a fair idea of the status of all the communities because of the interrelationship they share in food webs. Therefore, biomonitoring results in directly

assessing the status of the entire water body (Wetzel, 2001; Ramachandra et al., 2006). Beside that, the evaluation of environmental impacts on organisms that are potentially bioindicators is an important tool of Limnology (Pinese et al., 2008).

Studies on zooplankton communities were also conducted by other researchers in Malaysia. Zooplankton is an important constituent of the food chain in freshwater environments and may be of interest in the prediction of long-term changes in lake ecosystems (Ferrara et al., 2002; Cadjo et al., 2007). So far, many studies on the zooplankton communities were only restricted to its distribution and taxonomy. The studies by Johan and Idris (2000), Johan et al. (2000), Rezai et al. (2000), among many others, can be cited in this respect. However, very few studies that used zooplankton community structure as an indicator to evaluate the changes of aquatic ecosystems in this country. According to Aubé et al. (2005), zooplankton community structure in the water body characterized a disturbed ecosystem. A study on zooplankton in freshwater conducted by Meor et al. (2002), reported the diversity of net-zooplankton population between the littoral and limnetic habitats of the embayment in Chenderoh Reservoir. They assumed that the total plankton density is higher in the littoral compared to the limnetic zone. Beside that, the high water current in the main channel of the embayment is important in determining the overall reservoir productivity.

The response of zooplankton to changes in the water quality (e.g. lake eutrophication, siltation, acidification, pollution by agriculture or logging) was investigated in many studies (Sousa et al., 2008). Moreover, according to Sousa et al. (2008) zooplankton is considered to be good indicators of water quality alterations and have a main role in energy flow and nutrient cycling in aquatic ecosystems.

The use of zooplankton for environmental characterization of lakes is potentially advantageous. Long-term monitoring of zooplankton community structure can be useful in detecting patterns and changes in species composition that may be related to changes in water quality. A routine monitoring program also helps to separate the ordinary effects of seasonal changes in the zooplankton community from changes caused by other factors (Escribano and Hidalgo, 2000; Beyst et al., 2001).

In the study of a natural ecosystem, many environmental factors can be evaluated to understand their interactions and assess the sustainability of the environment (Jonnalagadda and Mhere, 2001). In this study, microbiological parameter was also included to find the relationship with other parameters. The coliform group of bacteria can be defined as the principal indicators of purity of water for domestic, industrial and other uses (APHA, 1992).

Coliforms have been used as the standard indicator of recent fecal contamination under most conditions in temperate fresh water. In tropical regions, they have been used to monitor contamination levels despite reports that free-living coliforms may be indigenous to some tropical waters and cannot be distinguished from a fecal source. Microbiological health risks remain associated with many aspects of water use, including drinking water in developing countries, irrigation reuse of treated wastewater and recreational water contact (Craun, 1986; Grabow et al., 1991).

It has been reported that drinking water supplies have a long history of association with a wide spectrum of microbial infections (Grabow et al., 2000). Microbiological analysis for bacterial indicators namely, heterotrophic bacteria, total and

fecal coliforms, and physical parameters, pH, turbidity and temperature were assessed to check whether the distributed water as well as the water from dams and rivers was safe for drinking and other domestic uses. Freshwaters polluted by fecal discharges from human and animals may transport a variety of human pathogenic micro-organisms. Detection of all waterborne fecal pathogens is very difficult; therefore various indicators of fecal contamination are usually used to detect fecal pollution in natural waters (Servais et al., 2007, Plummer and Long, 2007).

Air Itam Reservoir is the smallest and oldest reservoir in Penang State, which was constructed for domestic water supply. So far, there was very little scientific study had been done there and there was no study on zooplankton ever conducted in the reservoir. Therefore, it is essential to study the zooplankton community structure to provide basic ecological information and to determine its water quality status in order to manage the reservoir and to assess the impact of environmental conditions surrounding the catchment area.

Considering the need for a specific study on zooplankton community in this reservoir, the objectives of the study are as follows:

1. To determine the community structure of the zooplankton at all sampling stations of Air Itam Reservoir.
2. To measure the quantitative and qualitative attributes of zooplankton to ascertain the water quality.
3. To determine the status of water quality of Air Itam Reservoir.

CHAPTER 2

LITERATURE REVIEW

2.1 Physicochemical parameters

According to Ivanova and Kazantseva (2006), species diversity in natural ecosystems is closely related to environmental conditions. The species diversity of zooplankton is depends on the chemical composition of water and is determined by a combined effect of several environmental factors rather than depends on a certain single factor. However, unless supported by experimental evidence, it is unlikely to demonstrate the existence of one-to-one causal relationships between zooplankton associations and environmental factors (Akin-Oriola, 2003).

There are various parameters included in this study to determine the factors that influence the species composition of zooplankton and water quality. Parameters measured were dissolved oxygen (DO), temperature ($^{\circ}\text{C}$), Secchi disk transparency, biochemical oxygen demand (BOD), chemical oxygen demand (COD), total suspended solids (TSS), total dissolved solids (TDS), pH, ammoniacal nitrogen (AN), phosphorus (PO_4^{3-}), nitrite ($\text{NO}_2\text{-N}$), nitrate ($\text{NO}_3\text{-N}$), and conductivity.

The concentration of dissolved oxygen is one of the most important parameters in water quality analysis. Dissolved oxygen (DO) is a crucial factor for aquatic life, and characterizes the health of an aquatic ecosystem (Adam et al., 2001; Radwan et al., 2003; USEPA, 2006; Lytras, 2007). DO is influenced by aquatic plants photosynthesis and respiratory activities of bacteria and animals, and the atmospheric oxygen-waters

interchange. Changes in DO concentrations can be an indicator of changing conditions in the water body (Bartram and Ballance, 1996; Jassby and Goldman, 2003).

The amount of oxygen required in the water depends on water temperature, the species of aquatic organisms and the stage of life with some organisms can be more tolerant of lower oxygen levels than others. However, for a short time, many species are able to survive in condition which is potentially harmful due to lack of oxygen, but they rarely survive if the condition lasts for many hours or days (Chapman, 1992; Radwan et al., 2003). At the concentration below 1 mg/l, anaerobic bacteria replace the aerobic bacteria (Štambuk-Giljanović, 1999) in which the latter causes anaerobic condition in water, and eventually adversely affects the aquatic organisms. The presence of large-bodied zooplankton, despite high planktivore abundance, demonstrates a successful creation of a low-oxygen refuge (Robert et al., 2004).

It is well known that temperature affects all chemical and biological processes in water. Temperature has wide-ranging effects on important factors such as solubility of oxygen and other certain chemicals, activity of bacteria and other aquatic organisms (Jassby and Goldman, 2003; Bellos and Sawidis, 2005). Sánchez et al. (2006) found that an increase in temperature contributes to an increase in the biological activity and decreasing the dissolved oxygen concentration. According to Pedrozo and Rocha (2005), temperature is considered the most significant factor that controls zooplankton abundance in lakes. In tropical lakes, water temperature is generally high but decreases with increasing altitudes (Bartram and Ballance, 1996). Density differences between depths are minimal because water temperature is almost constant.

Transparency is a characteristic of water quality of lakes and reservoirs, and it varies with the combined effects of color and turbidity (Bartram and Ballance, 1996). It is also affected by the concentrations of algae. The water transparency can be measured with a Secchi disk. The Secchi disk is obtained from the reflection of light through the disk surface, and it is affected by the absorption characteristics of the water and of dissolved and particulate matters in the water (Wetzel, 2001).

BOD represents the demand for oxygen capacity of organic materials in a water body. BOD values indicate the level of organic pollution in aquatic systems which adversely affect the water quality. Low values of BOD reflect a low level of organic pollution in the water body (Cude, 2001; Jonnalagadda and Mhere, 2001).

The chemical oxygen demand (COD) refers to the capacity of water to consume oxygen during the decomposition of organic matter and the oxidation of inorganic chemicals such as ammonia and nitrite. The COD is commonly used to measure the amount of organic pollutants in lakes and rivers, making COD become a very useful parameter of water quality (Chapman, 1992; Marofi, 2006).

The amount of particles that suspend in a water sample is called total suspended solids (TSS). The concentration of TSS is important to river and lake ecosystems for ecological and water quality reasons. According to Mitchell and Stapp (1992) the optical water quality will decrease if there are high concentrations of suspended solids as the result of reduced water clarity and decreasing light available to support photosynthesis. Predator-prey relationships will also be altered due to the suspended solids. Mitchell and Stapp (1992) further said that suspended solids in most fresh water systems come from

watershed, pollutant point and sediment resuspension. Therefore, high concentration of suspended solids can have impacts on water quality and deposition both in lakes and reservoirs.

Total dissolved solids (TDS) is defined as the total amount of all dissolved materials in water. The sources of dissolved solids can be composed of organic materials such as leaves, silt, plankton, industrial waste and sewage, while other sources are generated from urban wastes, fertilizer and pesticides (Health Canada, 2008). According to Jonnalagadda and Mhere (2001), high concentrations of TDS and TSS in the water increase the biological and chemical oxygen demands, which deplete the DO concentrations in the aquatic systems. High concentrations of TDS may also reduce water clarity, contribute to a decrease in photosynthesis and lead to an increase in water temperature (Chapman, 1996). Ivanova and Kazantseva (2006) claimed that TDS is a more constant parameter compared to pH and usually differs only between the bottom and surface layers of water. On the other hand, the pH of a water body may change during the growing season and even during the day.

Ammonium ($\text{NH}_4\text{-N}$) is an inorganic substance that is abundant in surface water, soil and easily released from decaying plant tissue, dead algae and other organic material. The main sources are from animal excretions. Ammonia will be oxidized to nitrite ($\text{NO}_2\text{-N}$) by bacteria. Bacterial oxidization of nitrite to form nitrate ($\text{NO}_3\text{-N}$), is called nitrification (Maketab, 1993; Cech, 2003; Landa et al., 2007). Under high pH conditions ammonia becomes toxic to many aquatic organisms (Wetzel, 2001).

pH is a useful indicator of the chemical balance in aquatic systems (Jonnalagadda and Mhere, 2001). pH and temperature are among the factors that influence zooplankton growth, reproduction and mortality (Gyllstrom and Hansson, 2004). The suitable range of pH for the existence of most biological life is typically pH 6 to 9 (Jarvie et al., 2006). Ivanova and Kazantseva (2006) found that water pH can affect the number of zooplankton species more than the other parameters. Zooplankton occurred in the simplest trophic structure in natural acid lake with pH < 5.5. pH in such lakes shifts to the alkaline range during repeated fertilizer used, resulting in the increase of species diversity in zooplankton and trophic structure becomes more complicated. Furthermore, a pH rise from 6.5 to 8.5 is favorable for an increase in zooplankton species diversity, provided all other conditions are also favorable.

Phosphorus is known as an important nutrient for the metabolic reactions of plants and animals. If it enters the water body, it will quickly bond to soil particles, making it temporarily unavailable to living organisms. Phosphorus naturally present in low levels, and usually the growth-limiting factor for algae. Phosphorus may enter a water body from organic matter, rock and sediments, and anthropogenic pollution such as sewage plant discharges and run-off from agricultural areas (Adam et al., 2001). In general, unpolluted lakes have phosphorus concentrations between 0.01 and 0.05 mg/l (Wetzel, 2001).

Nitrogen is a major protein component and may be sequestered in organic material. One major form of dissolved nitrogen is nitrate. Nitrate is the most highly oxidized form of nitrogen compounds commonly occur in natural waters, because it is a product of aerobic decomposition of organic nitrogenous matter. Nitrate can enter a

water body through fertilizers, precipitation, dry deposition, domestic and industrial effluents and elemental nitrogen fixation by bacteria and plants. Nitrate concentrations in unpolluted natural water usually range from undetectable levels to 10 mg/l (Wetzel, 2001; Ramachandra et al., 2006).

An ability of water to conduct an electric current is known as conductivity. The conductivity depends on the concentration of ions in solution (Bartram and Ballance, 1996), mobility, valence and relative concentrations and on the measure of temperature (Ramachandra et al., 2006).

2.2 Water Quality Assessment

Water quality is very important because constant access to good water quality is a condition necessary for life. Appropriate water quality is essential for sustainability of biological resources. The scientific evaluation of the water quality is a need for risk assessment and therefore can protect the water bodies from continuing deterioration (Bordalo et al., 2006; Absalon and Matysik, 2007).

The overall water quality is sometimes difficult to evaluate from a large number of water samples that contain concentrations of various parameters (Pesce and Wunderlin, 2000). A Water Quality Index (WQI) was formulated to provide a mechanism for presenting large quantities of water quality data and defines into a certain level of water quality. A WQI can reflect the collective influence of various parameters; therefore it can characterize the overall water quality of a water body (Štambuk-Giljanović, 1999; Swamee and Tyagi, 2000; Bordalo et al., 2006; Sanchez et al., 2006).

The use of WQI could be of particular interest for developing countries due to their cost-effective water quality assessment as well as the possibility of evaluating trends (Pesce and Wunderlin, 2000). The Department of Environment Malaysia (DOE) formulated six indicator parameters of water quality to determine the water quality index classification of this country, namely, dissolved oxygen (DO), biochemical oxygen demand (BOD), chemical oxygen demand (COD), ammoniacal nitrogen (AN), total suspended solids (TSS) and pH (DOE-UM, 1994).

Water quality in Malaysia is calculated and classified based on the guideline suggested by the Interim National Water Quality Standards (INWQS) (DOE, 2001). The WQI values above 92.7 represent very clean water quality. When the values of WQI are in the range of 76.5 – 92.7, the water is classified as clean, and if the values of WQI within the range of 51.9 – 76.5 the water is classified as moderate. WQI values in the range of 31.0 – 51.9 indicated slightly polluted water, while below 31.0 indicates severely polluted water (DOE, 2001). The suggested guidelines are summarized in Table 2.1, Table 2.2 and Table 2.3.

Table. 2.1 Interim National Water Quality Standards for Malaysia Department of Environment's Water Quality Index (DOE, 2001)

Parameters	Unit	Class I	Class II	Class III	Class IV	Class V
pH		6.5 - 8.5	6 - 9	6 - 9	5 - 9	5 - 9
DO	mg/l	> 7	5 - 7	5 - 7	3 - 5	< 3
BOD	mg/l	< 1	1-3	3-6	6-12	>12
COD	mg/l	< 10	10-25	25-30	50-100	> 100
Suspended solids	mg/l	< 25	25-50	50-150	150-300	> 300
Ammonia– N	mg/l	0.1	0.3	0.3	0.9	2.7
Water Quality Index		> 92.75	76.5-92.7	51.9-76.5	31.0-51.9	< 31.0

Table. 2.2 Water Quality Index (WQI) (DOE, 2001)

WQI Range	Pollution Degree
< 31.0	Severely Polluted
31.0 – 51.9	Slightly Polluted
51.9 – 76.5	Moderate
76.5 – 92.7	Clean
> 92.7	Very Clean

Table. 2.3 Interim National Water Quality Standard for Malaysia (INWQS) with relation to water quality parameters (DOE, 2001)

Parameters	Units	Class					
		I	IIA	IIB	III	IV	V
Ammoniacal- N	mg/l	0.1	0.3	0.3	0.9	2.7	> 2.7
BOD	mg/l	1	3	3	6	12	> 12
COD	mg/l	10	25	25	50	100	> 100
DO	mg/l	7	5-7	5-7	3-5	< 3	< 1
pH		6.5-8.5	6-9	6-9	5-9	5-9	-
Color	TCU	15	150	150	-	-	-
Conductivity	Mmhos/cm	1000	1000	-	-	6000	-
Floating		N	N	N	-	-	-
Odour		N	N	N	-	-	-
Salinity	ppt	0.5	1	-	-	2	-
Taste		N	N	50	-	-	-
TDS	mg/l	500	1000	-	-	4000	-
TSS	mg/l	25	50	50	150	300	> 300
Temperature	°C	-	Normal±2	-	Normal±2	-	-
Turbidity	NTU	5	50	50	-	-	-
E. Coli.	Coloni/100 ml	10	100	400	5000 (2000) ^E	5000 (2000) ^E	-
Total Coliform	Coloni/100 ml	100	5000	5000	50000	50000	>50000

2.3 Coliform Contamination

Coliform bacteria have been used as faecal indicator organisms to evaluate the microbial water quality in many countries, which might be indicators for human and animal waste pollution. Testing for coliform bacteria is cheap and simple compared to testing using specific organisms and pathogens. For more than a century, monitoring microbial water quality by measuring indicator bacteria to assess the health hazard in drinking and recreational waters has been carried out (Gauthier and Archibald, 2001; Hörman and Hänninen, 2006; Hoyer et al., 2006; Dulic et al., 2008).

Coliform bacteria are defined as Gram-negative, rod-shaped, nonsporulating, oxidase-negative and lactose-fermenting bacteria (Niemi and Niemi, 2000). That group of organisms are not pathogenic, but they are aerobic, facultatively anaerobic, that ferment lactose with gas formation and aldehyde/acid in 48 hours at 35°C incubation. The group includes faecal coliforms and bacteria of faecal origin, and also some bacteria that might be isolated from environmental sources. Coliform belongs to family Enterobacteriaceae that includes a large number of species, which includes genera *Escherichia*, *Klebsiella*, *Salmonella*, *Shigella*, *Aerobacter*, *Scrratia*, *Citrobacter* and *Proteus* (Bartram and Ballance, 1996; Hoyer et al., 2006). The purpose of coliform bacteria measurement on raw water is to identify any extreme values that can show dangerous sources of fecal contamination (Jassby and Goldman, 2003).

The major sources of contamination can enter a water body by direct discharges from humans and animals, or from urban and agricultural runoff, sewage treatment facilities, and farm animals (Agbogou et al., 2006). High levels of indicator bacteria

suggest the possible presence of pathogens that cause water-borne diseases such as gastroenteritis, bacillary dysentery, etc (Dulic et al., 2008).

2.4 Zooplankton in aquatic ecosystem

Aquatic ecosystem is one of the most productive and diverse assemblage of habitats in the world (Barnes, 1982). Zooplankton is planktonic animals, which are inseparable part of the aquatic ecosystem, and it fulfills a great variety of important functions. Zooplankton is unable to maintain their position by swimming against the physical movement of water (Parsons, 1982; Pliūraitė, 2003). Zooplankton play a central role in aquatic ecosystems (aquatic food webs) between primary producers (phytoplankton) and higher trophic levels (Banse, 1995). Many large animals feed on smaller zooplankton; thus form secondary consumers system, while these organisms are also consumed by other aquatic macrofauna (Ramachandra et al., 2006).

Furthermore, zooplankton is also the food of choice for larval insects and fishes, and they are extremely important in supporting higher level of food chain. Therefore, a healthy zooplankton community is a prerequisite for good fishery (Osborne, 2002). Zooplankton also functions as a bioindicator to detect the pollution of water body because of their wide distribution in nature and they inhabit all fresh water habitats in the world, including polluted and unpolluted water bodies (Mukhopadhyay et al., 2007).

2.5 Taxonomic classification of zooplankton

According to Osborne (2002), freshwater zooplankton is part of free-floating members of the animal kingdom that reside, feed, and reproduce in open water. Most of

animal phyla (Protista, Porifera, Coelenterata, Gastrotricha, Hydracarina, Ostracods, Rotifera, Copepoda, Cladocera, and larval members of insects and fishes) are represented in the zooplankton of freshwater lakes and streams with the main components are rotifers (Phylum Rotifera) and microcrustaceans (cladocerans, Suborder Cladocera and copepods, Order Copepoda).

2.5.1 Rotifers

Rotifers are a group of small or microscopic zooplankton which are characterized by corona at the anterior end, with a mouth near the ventral or posterior border. The corona is used for locomotion or to obtain food towards the mouth. The corona structure is usually the basic importance in the rotifers classification (Edmondson, 1992). Rotifers play a main role in energy transfer in various trophic status of lake (Whitman et al., (2002). According to Pedrozo and Rocha (2005) rotifers are considered opportunistic organisms, due to their ability to change densities in response to different temperatures. In a short time, several species of rotifers could survive high concentration of suspended materials due to their corona and mastax structures which are highly efficient at identifying and selecting materials to be ingested through sensorial bristles of the mouth in order to avoid organic particles. According to Conde-Porcuna et al. (2002), rotifer population dynamics are usually explained by exploitative competition, interference competition and predation.

2.5.2 Cladocerans

Among zooplankton, cladocerans are very important due to their beneficial and nutritive nature to higher members of fishes in the food chain (Haberman et al., 2007).

They are normally covered by carapace. Their common name and water fleas are usually identified by their two large second antennae. In addition, the antennae are also used for rowing through water. Cladocerans are filter feeders as they filter the water to trap organisms in it. However, they are very sensitive against low concentrations of pollutants. In heterotrophic systems, cladocerans are also recognized for making bacteria as their primary source of foods (Osborne, 2002; Ramachandra et al, 2006), and cladocera could obtain 16-21% of their carbon through ingestion of bacteria (Nogueira, 2001).

2.5.3 Copepods

Copepods are aquatic crustacean, the diminutive relatives of the crabs and shrimps (Johan, 2001). The main three suborders of copepods comprise of calanoids, cyclopoids and harpacticoids. Immature copepods are called nauplius and copepodid, which then develop to adults (Edmondson, 1992). Copepods are strongly motile than other zooplankton and generally they can survive harsher environmental conditions compared to cladocera. Copepods are known as good predator of other planktonic animals (Johan, 2001). Among the three orders of copepods, cyclopoid copepods are generally carnivorous on other zooplankton, fish larvae and also feed on algae, bacteria and detritus (Ramachandra et al, 2006).

2.6 Regional Distribution and Diversity of Zooplankton in Malaysian Freshwater Bodies

Rotifers have wide-spread distribution in all kinds of waters and have great abundance (Edmondson, 1992). Systematic chain of rotifers of Malaysia and Singapore,

and documentation of their occurrence are very meager in literature. Fernando and Zankai (1981) recorded 165 species with 16 species found in reservoir/dam from 500 zooplankton samples from East and West Malaysia and 38 samples from Singapore which were analyzed for rotifera.

Cladocera occur in all kinds of freshwaters. Cladocera are found in greater number in lakes and ponds than rivers. In the inland lakes at limnetic area, the cladocera has large population in term of number of individuals but less in species (Edmondson, 1992).

According to Idris and Fernando (1981), the Malaysian cladocera are poorly identified. There were a few works done on cladocerans in Malaysian waters. Cladocera are mostly planktonic, but some occur among the aquatic vegetation or algal mats and a few are benthic. The order Cladocera is represented by six family namely: *Sididae*, *Daphnidae*, *Moinidae*, *Bosminidae*, *Macrothricidae* and *Chydoridae*. In the study of zooplankton from all types of habitats in Malaysia and Singapore, using 500 samples, 63 Cladocera species were identified, with 29 species found in reservoir/dams and 10 species in streams habitats (Idris and Fernando, 1981).

Copepoda is the most dominant group in zooplankton community (Johan, 2001). The three suborders of copepods (Calanoida, Cyclopoida and Harpacticoida) are found in marine waters, fresh and other inland water bodies (Edmondson, 1992). According to Lai and Fernando (1978), the fresh water copepods are among the least recognized groups of zooplankton in Malaysia, although their existence has been beneficial as the source of fish food.

In the mean time, cyclopoid Copepoda are considered the most widely present microcrustaceans in freshwater. In the tropics, their occurrence is poorly documented. Several earlier studies on freshwater Cyclopoida of humid tropical Asia including Malaysia and Singapore (Dussart, 1974), as well as Burma (Lindberg, 1949), Cambodia (Lindberg, 1952), India (Kiefer, 1939; Lindberg, 1941) and Indonesia (Kiefer, 1933; Lidberg, 1954) have been carried out. More systematic studies are on Sri Lanka (Fernando, 1974) and on Philippine material (Mamaril and Fernando, 1978), and Thailand freshwater cyclopoida as well (Boonsom, 1970; Bricker et al., 1978). The occurrence of Cyclopoida in 528 samples from East and West Malaysia and 38 samples from Singapore were studied by Fernando and Ponyi (1981). A total of 15 species of cyclopoida were recorded in the samples of all habitats, with six species were found in reservoirs and seven species were recorded in rivers and streams water (Fernando and Ponyi, 1981).

2.7 Zooplankton as Bioindicator

Biological parameters can be used to assess the quality of water which provides information for water management (Iliopoulou-Georgudaki et al., 2003). Porto Neto (2003) defined bioindicators as the organisms that can characterize special conditions of an ecosystem as well as its natural modifications. Invertebrates are very significant as bioindicators of water conditions and they have been the basis of a number of hydrobiological studies during the last half century (Porto Neto, 2003). According to

Rusak et al. (2002), indicator species should be sensitive and shows specific responses to particular stressors as well as low baseline variability.

Aquatic organisms, e.g. zooplankton are among the most valuable indicators of pollution in aquatic systems. Zooplankton, as a monitoring tool for water bodies, has specifically been used in many ways to explore their usefulness in monitoring (Whitman et al., 2002). According to Bednarski and Morales-Ramírez (2004), zooplankton are considered to be the ecological indicators or bioindicators of water bodies because they can provide information on trends in environmental conditions and how these conditions affect the indicator itself.

Studies on zooplankton as bioindicator to assess the sustainability and health of aquatic ecosystems have been documented by a number of authors (e.g. Saksena, 1987; Attayde and Bozelli, 1998; Marneffe et al., 1998; Caramujo and Boavida', 2000; Stemberger et al., 2001; Lougheed and Chow-Fraser, 2002; Pinese et al., 2008; Hulyal and Kaliwal, 2008).

Zooplankton are suitable as bioindicator because they are highly sensitive to environmental conditions and have short life cycles so they can respond quickly to specific environmental changes (Porto Neto, 2003; Dulic et al., 2008). They are also inexpensive to collect (Kane and Culver, 2003), widely distributed geographically and are subject to intermediate trophic position in the food web in a lake ecosystem (Whitman et al., 2002).

Sládeček (1983) found that rotifers are very sensitive to water quality alterations and have been considered good indicators of water quality among the taxa of

zooplankton. According to Duggan and Barnes (2005), rotifers are considered an ideal bioindicators due to their discriminate responses to environmental conditions and have numerical dominance in the main zooplankton groups and are species rich, therefore they are suitable to be used in the ecological monitoring of water bodies. Duggan et al., (2001) also suggested that rotifers may provide useful bioindicators of lake trophic state based on their findings in 10 Rotorua lakes in North Island, New Zealand. In Brazil, rotifera is dominant in the most reservoirs with high density and species richness (Sampaio et al., 2002) because rotifera have competitive advantages of life cycle traits, feeding mechanisms and metabolisms than other main zooplankton groups such as Cladocera and Copepoda.

Ramachandra et al., (2006) summarized the advantages of zooplankton as bioindicators for the evaluation of water quality as follows:

- Zooplankton has a large number of species in any aquatic ecosystem; therefore they can follow the trends in water quality alterations.
- Samples of zooplankton can be collected easily and processed quickly.
- They have a relatively short cycle of reproduction, therefore could be analyzed for several generations within a relatively short period.
- Some of the common species can be easily cultured for experimental purposes.
- They respond to environmental changes more quickly than fishes that are usually used as indicator of water quality degradation.

The use of zooplankton as indicators of environmental conditions has been well studied. Whitman et al. (2002) assessed the Sleeping Bear Dunes National Lakeshore using limnetic zooplankton as a bioassessment tool. The study observed strong correlations between lake trophic status and zooplankton communities. The study also showed periodic peaks in zooplankton group throughout the season. In Europe, the size range of species has been used as an indicator of water quality. The existence of large species such as daphnids is a characteristic of better quality of waters (Sampaio et al., 2002).

Attayde and Bozelli (1998) used the indicator properties of zooplankton assemblages to evaluate aquatic ecosystem conditions. Target taxa were selected to be applied in different aquatic ecosystems by an analysis that can relate community composition change to environmental parameters. Therefore, the method can be widely used for environmental monitoring.

CHAPTER 3

MATERIALS AND METHODS

3.1 Study Area

The Air Itam Reservoir is the smallest reservoir in Penang State, Malaysia. It is one of the two reservoirs in Penang Island and can hold up to 550 million gallons of water. It is located in the middle of a tropical forest hill, constructed between 1958 and 1962. The reservoir was one of the first major engineering projects undertaken by the City Council of George Town after independence. Currently, it is used as a source of drinking water (water supply) for the people in Air Itam area (PBA, 2007).

The catchment area of the Air Itam Reservoir is surrounded by some recreational garden, farming and fruit orchards, as well as forest reserve. The reservoir is created by blocking the Air Itam River as the main river up and downstream of the reservoir. The average annual rainfall is 2300 mm/year, the maximum and minimum water level are 234.70 and 201.17 meters above sea level, respectively (PBA, 2007).

3.2 Sampling Stations

Five sampling stations were designated in this study area. Three stations were in the reservoir, namely A, B and C, one station was in the upstream (station D) and the last station was in the downstream (station E). Station A ($05^{\circ}. 23'.41.80''N$ $100^{\circ}. 15'. 42.23''E$) was located in limnetic zone near to the tower. The maximum depth of Station A was 30 m during the study period. Station B ($05^{\circ}. 23'. 37.69''N$, $100^{\circ}. 15'. 37.01''E$)