SPATIO-SEASONAL VARIATIONS OF WATER QUALITY AND ANURANS AT MALIAU BASIN, SABAH

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ABSTRACT

Forested watersheds play an important role in supporting aquatic organisms by providing food and habitats. Freshwater organisms inhabit rivers that flow through forested regions and respond to all environmental stresses. Thus, aquatic organisms are useful bioindicators to evaluate and monitor river health and river water quality. Maliau Basin is relatively untouched and lacks scientific documentation. The objectives of this study were to determine spatial variations of physico-chemical water quality parameters, optical parameters, and anurans at Maliau Basin; to evaluate the seasonal variations of physico-chemical water quality parameters, optical parameters and anurans; and to investigate the relationship between physico-chemical water quality parameters, optical parameters and anurans as biological indicators. A total of 12 stations were chosen consisting of four streams representing for different land uses that are altered habitat (AH), main river (MR), secondary forest (SF) and pristine forest (PF) during four sampling occasions (December 2014, March, May and September 2015). Physico-chemical water quality parameters pH, dissolved oxygen, temperature, conductivity, total suspended solids (TSS) and total dissolved solids (TDS) were recorded *in-situ*. UV-visible absorption coefficients at wavelengths 254nm, 340 nm, and 410nm (a_{254}, a_{340}, and a_{410}) and spectral slope were also determined. Anurans were collected at the same stations for water samples via time-constrained opportunistic examination. Discriminant analysis showed that temperature, TSS, and a_{340} were dominant at AH and MR. For seasonal variations, DO, pH and a_{340} exhibited differences amongst four occasions. A total of 266 individuals representing 20 anuran species were recorded from the four sampling sites. Low diversity score was observed at AH (H'= 0.861) and MR (H'= 0.818), as compared to forest type habitat (SF and PF) (H'= 1.303 to 1.571). The anuran community showed low seasonal variations (H' = 1.10 – 1.18). Assessment of the relationships between anurans and water quality parameters was performed using the canonical correspondence analysis. *Limnonectes leporinus* and *Phrynoidis juxtaspera* were dominant in high temperature and conductivity, and negatively associated with aromatic/fresher dissolved organic matter content, suggested that these species have the potential to act as biological indicators for disturbed habitats.

ABSTRAK

VARIASI SPATIAL DAN MUSIM KUALITI AIR, KATAK DAN KODOK DI MALIAU BASIN, SABAH

Kawasan tadahan air di hutan memainkan peranan penting bagi menyokong organisma akuatik dengan menyediakan makanan dan habitat. Organisma air tawar di sungai-sungai yang mengalir melalui kawasan hutan menerima pelbagai tekanan alam sekitar. Oleh itu, organisma akuatik boleh digunakan sebagai penunjuk untuk menilai dan memantau kesihatan sungai dan kualiti air sungai. Kawasan Pemuliharaan Lembangan Maliau adalah salah satu kawasan pemuliharaan yang kekurangan dokumentasi penyelidikan. Objektif kajian ini adalah untuk mengenalpasti variasi kualiti air, katak dan kodok di Maliau Basin dari aspek ruang dan musim. Di samping itu, kajian ini juga mengkaji hubungan di antara kualiti air dan katak dan kodok. Empat sungai telah dipilih bagi mewakili empat jenis habitat iaitu kawasan terbuka (AH), sungai utama (MR), hutan sekunder (SF), dan hutan asli (PF). Kajian ini telah dijalankan dari Disember 2014 sehingga September 2015. Parameter fiziko-kimia, iaitu pH, oksigen terlarut (DO), suhu, kekonduksian, jumlah pepejal terampai (TSS) dan jumlah pepejal terlarut (TDS) telah direkodkan secara in-situ. Koefisien penyerapan gelombang UV-vis (a_{254}, a_{340} and a_{410}) dan kecerunan spektra telah dinilaikan dengan spectrophotometer. Persampelan anuran dijalankan di stesen yang sama dengan kaedah pemeriksaan oportunistik. Analisis diskriminan menunjukkan parameter suhu, TSS dan a_{340} dominan di AH and MR, manakala DO, pH dan a_{340} menunjukkan variasi musim. Sebanyak 266 individu yang mewakili 20 species anuran telah direkodkan. Diversiti katak dan kodok yang rendah didapati di AH (H′ = 0.861) dan MR (H′ = 0.818) berbanding dengan SF dan PF (H′ = 1.303 – 1.571). Diversiti anuran tidak menunjukkan perbezaan yang ketara antara empat kali persampelan (H′ = 1.10 – 1.18). Penilaian mengenai hubungan katak dan kodok dengan parameter kualiti air dengan analisis Canonical Correspondence Analisis (CCA) menunjukkan taburan spesies Limnonectes leporinus, dan Phrynoidis juxtaspera berkorelasi positif dengan sungai mempunyai nilai suhu dan kekonduksian, dan berkadar songsang dengan kandungan bahan organik aromatik. Ini menunjukkan kedua-dua spesies ini berpotensi sebagai penunjuk kepada habitat terganggu.

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LIST OF ABBREVIATIONS

a_{254}	-	UV-Vis absorption coefficient at 254 nm
a_{340}	-	UV-Vis absorption coefficient at 340 nm
a_{410}	-	UV-Vis absorption coefficient at 410 nm
AH	-	altered habitat
CCA	-	Canonical Correspondence Analysis
CDOM	-	chromophoric/coloured dissolved organic matter
DA	-	discriminant analysis
DO	-	dissolved oxygen
DOC	-	dissolved organic carbon
DOM	-	dissolved organic matter
EEM	-	fluorescence excitation-emission matrix
GF/F	-	glass microfibre filter with 0.7-micron nominal pore size
HCl	-	hydrochloric acid
HDPE	-	high-density polyethlene
ITBC	-	Institute for Tropical Biology and Conservation
IUCN	-	The International Union for Conservation of Nature
MBCA	-	Maliau Basin Conservation Area
MR	-	main river
NOM	-	natural organic matter
OM	-	organic matter
POM	-	particulate organic matter
PF	-	primary forest
$S_{275-295}$	-	spectral slope for the interval of 275 to 295 nm
SF	-	secondary forest
TDS	-	total dissolved solids
TOM	-	total organic matter
TSS	-	total suspended solids
UV-Vis	-	ultraviolet-visible absorbance spectroscopy
UV-A	-	ultraviolet of relatively long wavelengths (320-400 nm)
UV-B	-	ultraviolet of relatively long wavelengths (280-320 nm)

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CHAPTER 1

INTRODUCTION

1.1 Background

Rivers are important across nations as water resources for both human usage, and to sustain the well-being of wildlife (Vörösmarty *et al.*, 2010). Anthropogenic activities lead to destructive consequences on water quality within this vital ecosystem. There is currently an exponential increase in water quality related research due to its vital importance to both humans and wildlife (Varol *et al.*, 2012; Wang *et al.*, 2012). Assessing spatial-temporal variations of river water quality has become an essential aspect of determining the characteristics and status of the aquatic environment (Gu *et al.*, 2015; Vizcaíno *et al.*, 2016). Organic pollution and eutrophication of surface water are currently of great environmental concern worldwide (Yang *et al.*, 2008). The presence of chemical pollutants and excessive nutrients in the rivers can adversely impact the functionality of the ecosystems (Rhind, 2009), such as fish mortality, causing critically low dissolved oxygen content, loss of biodiversity, and loss of aquatic plant beds.

In the highly complex aquatic ecosystem, physical and chemical properties of water are known to interconnect with each other (Makinde *et al.,* 2015). For example, the increase of surface water temperature can result in the decrease of dissolved oxygen in the water (Mokhtar *et al.*, 2009). On the other hand, the increase of surface water temperature is related to microbial metabolism (Scofield *et al.*, 2015), and their exponential growth can eventually lead to algal blooms that further deplete the dissolved oxygen in the water body (Piontkovski *et al.*, 2012; Wells *et al.*, 2015). Detailed water quality assessment is crucial to determine the current status of the rivers for better management of natural ecosystems in order to sustain a high diversity of organisms (Dudgeon, 2010; He *et al.*, 2014).

Dissolved organic matter (DOM) is a dynamic and heterogeneous mixture of chemical compounds that widely exist in terrestrial and aquatic ecosystems (Maie *et al.*, 2012). It represents an important source of carbon and other imperative nutrient resources that determine the availability and diversity of living organisms with its significant role in various biogeochemical and ecological processes (Richey, 2005). DOM has become the most studied fraction of natural organic matter as its sources and characteristics have been extensively investigated along with its interactions and degradation that occur on both spatial and temporal scales (Hudson *et al.*, 2007).The origin of DOM can be categorized as either allochthonous or autochthonous (Findlay and Sinsabaugh, 1999; Pagano *et al.*, 2014). The allochthonous DOM generally referring to the organic matters with the terrestrial origin and enters the fluvial ecosystem through soil leaching, leachates from decomposing flora and fauna, and geographical activities (Hudson *et al.*, 2007). Furthermore, DOM with autochthonous origin is synthesized *in-situ* by aquatic microorganisms, algal by-products, and break down of fine particles by aquatic invertebrates (Hudson *et al.*, 2007; Pagano *et al.*, 2014). On the other hand, the anthropogenic source of organic pollutants can increase the DOM input into the aquatic system, these organic matters from human activities included the wastewater, agricultural runoff, and industrial discharges (Reyes and Crisosto, 2016). DOM with varying sources in the aquatic system contains a wide range of organic compounds in different chemical structure and reactivity (Goldman, 2011). DOM is having numerous ecological impact to the aquatic life, for example, the high concentration of DOM can deplete the dissolved oxygen contents in the water bodies due to microbial oxidation of these DOM.

Amphibians spend at least part of their life cycle in or connected to water, thus water quality is critically important to the well-being of amphibians, especially anurans. Sparling (2009) illustrated that the physico-chemical properties of water, namely dissolved oxygen, temperature, pH, salinity, organic matters, conductivity, and the existence of pollutants can influence anurans' survival, growth, maturation, and physical development. Although DOM can cause disastrous consequences if present in high concentration, its ability to absorb solar radiation is considered beneficial to anurans. Ultraviolet B radiation was known to adversely affect the physiology of anurans (Pahkala *et al.,* 2002), DOM acts to attenuate the impact of light that penetrating the aquatic ecosystem (Diamond *et al.*, 2005; Forsström *et al.*, 2015), and thus providing protection for both the adults and tadpoles. However, the correlation between DOM concentration and anuran's species composition and richness are still inadequately documented.

1.2 Context and Relevance of Study

Maliau Basin Conservation Area (MBCA) is widely known as "Sabah's Lost World", one of the most important wilderness areas left in Malaysia. MBCA is protected under Sabah State laws as Class I (Protection) Forest Reserve under Sabah Enactment 1968 (Caldecott, 2002). The relatively untouched wilderness of MBCA with insufficient scientific documentations attract the interest of naturalist, biologist, and conservationist (Dubgaard, 2002). The rivers of MBCA were known for its tea-coloured water. This colouration resulted from humic substances that leach from the heath forest vegetation (Harun et al., 2010). This blackwater is commonly associated with low productivity, high acidic, low ion concentration, low transparency, and low dissolved and suspended solids (Winemiller et al., 2008). This had attributed to low and limited fish species and abundance recorded in MBCA (Hazabroek et al., 2004). However, anurans – semi-aquatic organisms were recorded in abundance and variety (Yayasan Sabah, 2014), implying that anurans would be better biology indicators for freshwater ecosystem at MBCA.

Documentation regarding the water quality of the streams in MBCA is relatively scarce and not updated. This issue needs to be resolved in order to better understand the status quo. The latest water quality works published were carried out by Mokhtar *et al.* (2009) at Eucalyptus River, and Harun *et al.* (2010) at Giluk River, Takod-Akob River, Ginseng River, and Maliau Falls. There are several rivers with different habitat characteristics within Maliau Basin that were previously not subjected to any detailed spatial and temporal water quality assessment. Any changes in the water quality occur in these rivers will be untraceable if the current status is not documented in detail. Thus, this study aimed to provide more information by updating the current status of river water quality and to document the diversity of stream anurans.

The essential objective of ecologists is to understand what biotic environmental factors that shape the spatial patterns of a species. The geographical distribution of anuran species is determining by local environmental suitability and their tolerance towards the fluctuation of these environmental factors. MBCA documented with 53 anuran species from the expeditions and zoological surveys from 1998 to 2005 which carried out at different locations (Ahmad and Wong, 1998; Traeholt, 2001; Jomitin, 2002; Lakim *et al.*, 2002; Kueh and Maryati, 2005a; Matsui *et al.*, 2014). Most of these recorded species are having a lifestyle closely related to water, either as a stream bank sitter or as a stream breeder. Therefore, this study seeks to determine the correlations between water quality (for example DOM, water temperature, conductivity, and dissolved oxygen concentration) and anuran species composition at several localities with different habitat types. This information might be useful to establish stream anurans as biological indicators for the habitat types where they are found.

1.3 Objectives

Water quality parameters, as well as anuran diversity and occurrence, are known to fluctuate with time and space. To get more precise and accurate information about the river water quality and anurans at Maliau Basin, a relatively intense sampling programme would be required, and have to take place at the different streams with various characteristics. The objectives of this study were:

i. To determine spatial variations of physico-chemical water quality parameters (pH, temperature, conductivity, dissolved oxygen, total suspended solids, total dissolved solids, and salinity), optical parameters (a_{254}, a_{340}, a_{410}, and $S_{275-295}$) and anurans at Maliau Basin;
ii. To evaluate the seasonal variations of physico-chemical water quality parameters (pH, temperature, conductivity, dissolved oxygen, total suspended solids, total dissolved solids, and salinity), optical parameters (a_{254}, a_{340}, a_{410}, and $S_{275-295}$) and anurans; and
iii. To investigate the relationship between physico-chemical water quality parameters, optical parameters, and anurans as biological indicators.

CHAPTER 2

LITERATURE REVIEW

2.1 Tropical Flowing Water

Heavy seasonal rainfall favors by Hadley circulation created numerous well-watered landscaped within the tropical regions (Lewis Jr., 2008). While these streams or rivers occur in complex variety and the fluvial ecosystem differ in various additional features. Depth and velocity of flow, water chemistry as well as metabolic rates within the tropical stream or rivers are well-defined seasonally. However, the water chemistry can be altered by several environmental factors, for instance, the suspended and dissolved solids of the tropical fluvial ecosystem can be influenced by local topography (Armijos *et al.*, 2013; Lewis, 2008). In addition, seasonality fluctuations on the concentration of dissolved and suspended solids, organic matter, and nutrients are determined by the seasonal variation of water discharge (Armijos *et al.*, 2013; Lambert *et al.*, 2016). In brief, the tropical rivers and streams are stable thermally comparing to those located in temperate regions but show seasonality fluctuations in water chemistry driven mainly by local topography (Boulton *et al.*, 2008).

Rivers are vital to the survivorship of both mankind and wildlife by providing essential ecological services (Vörösmarty *et al.*, 2010). Several ecological processes provided by rivers or streams are indispensable for the ecosystem, for instances, the streams direct the path of food webs. These essentially important ecosystem services provided by streams or rivers include water supply for domestic, industrial, agricultural and aquaculture uses, hydropower, waste disposal, navigation, recreational enjoyment, and spiritual fulfillment (Dudgeon, 2012). At the same time, rivers influencing the carbon and nutrients cycle, as well as processing and transporting materials with terrestrial origin towards the aquatic ecosystems (Lambert *et al.*, 2016). Despite the contribution and importance of streams and rivers to the overall ecological functionality, anthropogenic disturbances often cause devastating consequences on water quality that are not only impacting the well-being of wildlife but also directly affecting the health of mankind (Dudgeon, 2010). Freshwater ecosystems are exclusively subjecting to various pressures, for example, the water abstraction, industrial and domestic effluents, the spread of invasive species, altered hydrology, habitat degradation, and overharvest of the resources within the ecosystem (Dudgeon *et al.*, 2006; Dudgeon, 2010; 2012; Rundle, 2002).

All the threats mentioned above are having catastrophic impacts on the ecological balance of the fluvial ecosystem. The biological diversity that inhibiting the freshwater ecosystems is encountering much greater declines than is seen in much of terrestrial ecosystems due to the anthropogenic disturbances (Dudgeon *et al.*, 2006; Mohmad *et al.*, 2015). If human pressures continue to accelerate and biodiversity continues its descending trend, the prospects for freshwater ecosystems are alarming and perhaps catastrophic. Revenga and Kura (2003) emphasized that multiple interacting threats are leading to biodiversity crisis that is currently experiencing by freshwater biota. In-stream alterations of habitats included the construction of dams, channelization, and activities that are harmful to the aquatic environment operate along the water's edge are currently destabilizing the river banks. In addition, the changes of land use affect the natural hydrology that eventually leads to secondary consequences for physical processes and the biota inhabiting the areas (Dudgeon *et al.*, 2006; Mohmad *et al.*, 2015). Moreover, climatic changes that directly alter the precipitation rate, ambient temperature, and run-off patterns have indirect effects on various aspects of the lotic ecosystem function (Vörösmarty *et al.*, 2000).

Habitat degradation caused by the changes of land use and logging activities is the most significant threat to biodiversity and ecosystem function in most human-impacted river systems (Iwata *et al.*, 2003). Land-use change is an integrator of many anthropogenic activities that have a disastrous impact on the functionality of the stream ecosystems (Dudgeon, 2012). For example, the logging practices and road building associated with agricultural activities are most likely to cause alteration on flow variability and sediment delivery toward the streams (Lorion and Kennedy, 2009). The vegetation located in the riparian regions are having remarkable influence on the fluvial ecosystem, the presence of vegetation can stabilize the river banks that prevent erosion, moderates water temperature through shading, filter nutrients, and sediments, and influence the energy pathways by regulating the inputs of particulate organic matter and govern the light availability (Iwata *et al.*, 2003; Singh and Mishra, 2014). Therefore, the loss of riparian vegetation is commonly accompanied by bank erosion, silt deposition, higher water temperature, and altered food webs. Allan (2004) stated that sedimentation caused by land-use changes can impair the substrate suitability for periphyton and biofilms production that can be further translated into depleting the food resources for the aquatic ecosystem. The degradation of primary production and food quality eventually causes bottom-up effects through food webs, moreover, decreases in periphyton and biofilms populations are believed to result in starvation of tadpoles (section 2.7.2). Furthermore, riparian clearing reduces shading that consequently leads to increases in stream temperature (Iwata *et al.*, 2003) and at the same time logging can reduce sediment trapping that most likely to result in the bank and channel erosion that further impact the aquatic environment (Singh and Mishra, 2014).

Due to the importance of water quality towards the well-being of both human and wildlife, there is an exponential increase in water quality related studies which assess the status of the rivers in different localities. In addition, research being carried out to determine the impacts of human disturbance on the water quality that are having critical influences on the health of both human and wildlife (Varol *et al.*, 2012; Wang *et al.*, 2012). By utilizing the information collected regarding the spatial and temporal variations of the water quality, stream ecologist can have a better understanding on the status quo of the water quality. The information collected by stream ecologist can, therefore, inform the land manager to synthesize a better planning in order to manage the natural resources.

2.2 Spatial and Seasonal Variations of Water Quality

The physical and chemical elements of the water are interdependent within the complex fluvial ecosystems, and these complex arrays of chemical and physical factors are highly heterogeneous at both spatial and seasonal scales (Rier and Stevenson, 2002; Fatema *et al.*, 2014). Makinde *et al.* (2015) stated that the interactions of these physical and chemical properties are the primary determinant factors for the observed fluctuations in the water quality. Surface water quality parameters varied seasonally mainly due to the differences in land use types, degradation of riverine vegetation, and rain events (Poudel *et al.*, 2013). While the spatial heterogeneity of water quality mainly determined by local environmental conditions, namely the light intensity, water velocity, temperature, and discharge rate (Fatema *et al.*, 2014).

Water temperature is increasingly being recognized as the highly sensitive variable of water quality, it can influence other physical-chemical water quality parameters, and influence the survival, growth rates, timing of life history and metabolism of aquatic organisms (Hannah and Garner, 2015). The water temperature of the inland aquatic ecosystems is expected to be remarkably influenced by precipitation rate, land use and rate of evaporation (Malmqvist *et al.*, 2008). In addition, the spatial and temporally variability in heat flux and hydrological processes within the river system created heterogeneity in river temperature (Hannah and Garner, 2015). Water quality parameters are interdependent as mentioned previously, for example, the raise in water temperature will deplete the dissolved oxygen concentration in the aquatic ecosystem by reducing the solubility of oxygen in the water (Mokhtar *et al.*, 2009). Moreover, the increase of surface water temperature is positively correlated to microbial metabolism, and their exponential growth can eventually lead to algal blooms that further exhaust the dissolved oxygen capacity of the water body (Wells *et al.*, 2015). Precipitation rate that influences the flow rate and evaporation rate can subsequently influence the salt concentration and conductivity of the stream water (Gelca *et al.*, 2014). On the other hand, the conductivity of the water was influenced by the local topography, for example, the concentration of inorganic salts and organic material within the water bodies determined the conductivity value (Basu and Lokesh, 2013). In addition, highly mineralized groundwater inflows will raise the conductivity and salinity of the stream water, while this depends on local topography and the solubility of riverbed rock's inorganic minerals (Zaidi and Pal, 2015).

The leachates derived from the riparian vegetation are among the most readily available organic carbon for the stream microbiota. While the concentration