POPULATION DENSITY, HUMAN-CROCODILE CONFLICT AND GENETIC VARIATION AMONG SALTWATER CROCODILE, CROCODYLUS POROSUS IN SARAWAK

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POPULATION DENSITY, HUMAN-CROCODILE CONFLICT AND GENETIC VARIATION AMONG SALTWATER CROCODILE, *CROCODYLUS POROSUS* IN SARAWAK

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DECLARATION

The work presented in this thesis, to the best of my knowledge and belief, original and my own work, except as acknowledge in the text. I hereby declare that no portion of the work referred to this thesis has been submitted in support of an application for another degree or qualification to this or any other university or institution of higher learning.

_________________________________________

(MOHD IZWAN ZULAINI BIN ABDUL GANI)

Date: 27 March 2014
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LIST OF ABBREVIATIONS

° Degree
π Nucleotide diversity
µg Microgram
µL Microliter
µM Micromolar
AGE Agarose Gel Electrophoresis
AIC Aikake Informations Criterion
AMOVA analysis of Molecular Variance
AZA The American Association of Zoos and Aquariums
BI Bayesian Inference
BLAST Basic Local Alignment Search Tool
BPPs Bayesian posterior probabilities
bp Base pair
C Celsius
CIA Chloroform Isoamyl Alcohol
CITES Convention on International Trade in Endangered Species of Wild Fauna and Flora
cm Centimeter
CTAB Cetyltrimethyl ammonium bromide
Da Nucleotide divergence
DNA Deoxyribonucleic Acid
dNTP Deoxyribonucleotide
EDTA Ethylenediaminetetraacetic acid
Fst Population Subdivision
GPS Global Positioning System
g Gram
IBD Isolation by distance
IUCN International Union for Conservation of Nature and Natural Resources
KCl Potassium Chloride
km Kilometer
kg Kilogram
MgCl₂ Magnesium Chloride
min Minute (time)
mm Millimeter
mM Milimolar
mL Mililiter
ML Maximum-Likelihood
MP Maximum-Parsimony
mtDNA Mitochondrial Deoxyribonucleic Acid
MWC Matang Wildlife Centre
Nm Gene Flow
Nst Nucleotide Subdivision
<table>
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<td>NCBI</td>
<td>National Center for Biotechnology Information</td>
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<tr>
<td>NGO</td>
<td>non-governmental organization</td>
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<tr>
<td>NJ</td>
<td>Neighbour-Joining</td>
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<tr>
<td>PBS</td>
<td>Phosphate buffered saline</td>
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<td>Polyacrylamide Gel Electrophoresis</td>
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<td>rpm</td>
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<tr>
<td>rRNA</td>
<td>Ribosomal Ribonucleic Acid</td>
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<tr>
<td>sec</td>
<td>Second (time)</td>
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<tr>
<td>SSRs</td>
<td>Short Sequence Repeats</td>
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<tr>
<td>SFC</td>
<td>Sarawak Forestry Corporation</td>
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<tr>
<td>TAE</td>
<td>Tris-Acetate-ethylenediaminetetraacetic</td>
</tr>
<tr>
<td>UPGMA</td>
<td>Unweighted Pair Group with Arithmetic Averages</td>
</tr>
<tr>
<td>UV</td>
<td>Ultraviolet</td>
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Study of Population Density, Human-Crocodile Conflict and Genetic Variation among Saltwater Crocodile, *Crocodylus porosus* in Sarawak

Mohd Izwan Zulaini Bin Abdul Gani

**ABSTRACT**

The objective of this study aims to (i) determine the distribution and density of *Crocodylus porosus* in rivers of western Sarawak, (ii) assess socio-economic profile and matters related to human-crocodile conflicts within population of local peoples and (iii) analyze genetic variations of *C. porosus* in Sarawak based on microsatellite data. Surveys of *C. porosus* population had been carried out using night spotting techniques in Batang Samarahan, Sibu Laut River and Bako River. The highest mean relative density was recorded in Bako River (3.19±8.24 non-hatchling/km), followed by Sibu Laut River (0.77±0.51 non-hatchling/km) and Batang Samarahan (0.60±0.33 non-hatchling/km). Pilot study showed that dependency of local communities toward rivers and their awareness toward crocodile issues are relatively high. Based on microsatellite data using *Cj127* primer, result showed limited success on determining the relationship among *C. porosus* from different areas in Sarawak. However using *Cj16* primer, phylogenetic and network analysis showed distinct clades based on geographical areas. Population genetic analyses show gene flow occurs and high number of migrants per generations in population of *C. porosus* in Sarawak suggesting they are panmictic population.

**Keywords:** Crocodiles, *C. porosus*, density, Panmictic population, Microsatellite.
Kajian tentang Ketumpatan Populasi, Konflik Manusi-Buaya dan Variasi Genetik dikanalangan Populasi Buaya Air Masin *Crocodylus porosus* di Sarawak

Mohd Izwan Zulaini Bin Abdul Gani

ABSTRAK

Objektif kajian ini bermatlamat untuk (i) menentukan taburan dan kepadatan *Crocodylus porosus* di beberapa sungai di barat Sarawak, (ii) menilai profil sosio-ekonomi dan perkara-perkara yang berkaitan dengan konflik manusia-buaya di kalangan populasi penduduk tempatan dan (iii) menganalisis variasi genetik *C. porosus* di Sarawak berdasarkan data mikrosatelit. Tinjauan *C. porosus* telah dijalankan menggunakan teknik pengesanan malam di Batang Samarahan, Sungai Sibu Laut dan Sungai Bako. Min ketumpatan relatif tertinggi dicatatkan di Sungai Bako (3.19±8.24 non-hatchling/km) diikuti oleh Sungai Sibu Laut (0.77±0.51 non-hatchling/km) dan Batang Samarahan (0.60±0.33 non-hatchling/km). Kajian perintis menunjukkan kebergantungan yang penduduk tempatan kepada sungai dan juga kepekaan mereka terhadap isu-isu buaya yang agak tinggi. Berdasarkan data mikrosatelit menggunakan primer Cj127, keputusan analisis menunjukkan kejayaan yang terbatas dalam menentukan hubungan di kalangan *C. porosus* daripada kawasan-kawasan yang berbeza di Sarawak. Walaubagaimanapun, dengan menggunakan primer Cj16, keputusan dari analisis filogenetik dan Network menunjukkan klad tersendiri berdasarkan kawasan-kawasan geografi. Analisis populasi genetik menunjukkan berlakunya aliran gen dan jumlah migrasi per generasi yang tinggi dalam populasi *C. porosus* di Sarawak menunjukkan bahawa mereka adalah populasi yang panmictic.

CHAPTER I
GENERAL INTRODUCTION

1.1 General Introduction

Within Class Reptilia, Order Crocodilia can be divided into three extant families: (i) Alligatoridae which includes alligators and the caimans, (ii) Crocodylidae, which includes the true crocodiles and (iii) Gavialidae that most likely contains two gharial species (Rodriguez, 2007; de Silva, 2013). All crocodilians species share almost similar morphological, anatomical, and physiological features. They are nocturnal carnivorous opportunistic predators, whose diet depends on their developmental stage, species and potential prey diversity (Martin, 2008). All crocodilian species are also amphibious and may be considered as totally water dependent since they can only mate in water. They can live in various aquatic habitats such as forest streams, rivers, marshes, swamps, elbow lakes and each species usually can be found in a specific zoogeographical region.

There are two species of crocodiles can be found in Sarawak namely saltwater crocodiles (Crocodylus porosus) and Malayan gharials (Tomistoma schlegelli) (Cox and Gombek, 1985). Interestingly, Cox and Gombek (1985) also included third species of crocodile, the Siamese crocodiles (Crocodylus siamensis). However, there is no concrete evidence that can support the presence of this species in the state although this species is found in East Kalimantan, Borneo (Cox, 2004).

Among the local people in Sarawak, C. porosus usually refer as “buaya katak” while other species, T. schlegelli, locally known by the name “buaya jejulong” (Ritchie and Jong, 2002). Presently, C. porosus is more abundant compared to T. schlegelli. They can be found in almost all major river basins including large river system,
mangroves estuaries and inland freshwater swamps in the state (Tisen and Ahmad, 2010). In contrast, *T. schlegelli* are difficult to sight in the wild and reports have indicated that this species can be found in upper tributaries of Batang Sadong, Baram River, Kimena River and a few other peat swamp areas (Stuebing *et al.*, 2004).

Nearly extinct due to overhunting since 1950’s until late 1980’s, *C. porosus* was primarily hunt for its lucrative hides and meats. *Crocodylus porosus* always become the centre of attention in the state as this species had caused many fatalities towards local people. *C. porosus* has been terrorized people who live along the river in Sarawak and conflicts between human and crocodile always occur since Rajah Brooke era (Ritchie and Jong, 2002). However, the conflicts was at its peak when a large white-backed crocodiles known by the name “Bujang Senang” attacked a man in one of the Batang Lupar tributary in early 80’. Since then, the story about “Bujang Senang” had become household word among the local people in the state (Ritchie and Jong, 2002).

Crocodiles play important roles both for human and environment. For human, crocodile bring great fortune and increase economy as the high prize and demand for the crocodile leather. Lucrative crocodile based leather product such as cloths, handbags, shoes and others cause many people hunting this animals and even illegally poaching them. According to Martin (2008), crocodile population have been depleting until the mid 60’s because of uncontrolled hunting of these animals. As for environment, crocodiles plays important role in food web as they act as main predator which helps in keeping wetland ecosystem healthy. When a wetland habitat is healthy, the fishery is considered to be healthy too. They are also very important for freshwater ecosystems during the dry season as they maintain waterholes that are used as reservoir for many arthropods, crustacean, fishes and amphibians (Martin, 2008).
1.2 Problem statements

*Crocodylus porosus* has been listed in Appendix I in the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) and categorized as Lower Risk / least concerned by the International Union for Conservation of Nature and Natural Resources (IUCN) Red List of Threatened Species 2012. In Malaysia, this species is listed as protected animal by the Wildlife Conservation Act 2010 and Sarawak Wild Life Protection Ordinance 1998. Appendix I CITES stated that killing and exporting of *C. porosus* either its eggs, meat, leather or by product is prohibited (Landong and Zaini, 2010). Since there are some reports saying that the population of *C. porosus* in Sarawak is overpopulated, there is an urgent need to carry out proper research to determine status of *C. porosus* population in Sarawak so that Sarawak could propose to down list *C. porosus* from Appendix I to Appendix II CITES.

Patchy information on crocodile’s population in Sarawak had caused difficulty in management of the human-crocodile conflicts as well as carrying out sustainable management of the resource (crocodiles and crocodile-based products). Data on density and distribution of crocodiles for many rivers in Sarawak are still inadequate to make any conclusion on crocodile population status in the state. Lately, many reports on crocodile attack in Sarawak had captured public attention. The incident had triggered fear among the local people in the state. The crocodile attacks in many rivers in Sarawak had provoked the villagers which led them to hold a grudge on crocodiles and sometimes foolishly taking matters into their own hands. Thus, a systematic management of crocodile-human conflicts is needed to ensure safety both humans and crocodiles. Studies about crocodile population alone are considered as too biocentric and the information will not be enough to solve human-crocodile conflicts in Sarawak. Information on local people socio-economic profile who live along the rivers and their dependency towards rivers are
essential to develop management plans to minimize crocodile attacks toward humans and livestock.

Chaeychomsri et al. (2008) had done study on C. siamensis population using microsatellite data and they had been successful in monitoring work of captive individual which has been released to the wild population. Genetic studies on C. porosus in Sarawak had been documented. For example, Abdullah et al. (2010) studied population structure of C. porosus in Sarawak using Cytochrome b and 12S rRNA. However, Abdullah et al. (2010) reported of unresolved molecular phylogenetic of C. porosus from Sarawak due to very slow evolution of both genes. Therefore, this study is designed to obtain microsatellite data in order to resolve population structure of C. porosus in Sarawak. Furthermore, genetic information data pertinent can be used for the development of management plan by identifying conservation units for many threatened and endangered species (Moritz, 1999).
1.3 Objectives

The objectives of this study are to:

1. determine the distribution and density of *C. porosus* in three selected rivers in western Sarawak.
2. analyse genetic variations and population structure of *C. porosus* in Sarawak based on microsatellite data.
3. assess socio-economic profile and matters related to human-crocodile conflicts within local people population along the three selected rivers.

Upon completion of this study, data obtained could help relevant agencies in Sarawak to further carried out sustainable management of *C. porosus*, besides helping in addressing the human-crocodile conflicts in Sarawak.