LAPORAN AKHIR GERAN PENYELIDIKAN
USM JANGKA PENDEK

TAJUK:

Genetic Relationship among the Negrito (Semang) Tribes of Peninsular Malaysia: the Microsatellite Approach

1 Mei 2007 – 30 April 2009

Dr. Hoh Boon Peng

SENARAI SEMAKAN UNTUK BUKU LAPORAN AKHIR GERAN USM JANGKA PENDEK

NAMA PENYELIDIK UTAMA	:	**Dr. Hoh Boon Peng**
NAMA CO-RESEARCHER	:	**Prof. Madya Dr. Zilfalil Bin Alwi** **Dr Endom Ismail (UKM)**
TAJUK GERAN	:	**Genetic Relationship among the Negrito (Semang) Tribes of Peninsular Malaysia: the Microsatellite Approach**
NO.AKAUN	:	

SENARAI SEMAKAN SEMASA PENYERAHAN BUKU LAPORAN AKHIR
(Sila Tandakan (4) Pada Kotak Yang Berkenaan)

NO.	PERKARA	ADA	TIADA
1.	Borang Laporan Akhir Projek Penyelidikan USM Jangka Pendek	✓	
2.	Borang Laporan Hasil Penyelidikan, PPSP	✓	
3.	i) Salinan Menuskrip	✓	
	ii) Salinan surat/email bukti penghantaran kepada mana-mana journal	✓	
4.	Penyata Perbelanjaan (Financial Statement) (Sila dapatkan daripada Jabatan Bendahari)	✓	
5.	Laporan Komprehensif (termasuk kertas persidangan atau seminar dan penerbitan saintifik hasil daripada projek ini)	✓	
6.	Surat pemakluman penghantaran Laporan Akhir ke Bhg. Penyelidikan	✓	

Nota:

* Sila buat 3 salinan buku laporan Akhir
* No. 1-5 - Perlu dimasukkan dalam Buku Laporan Akhir
* No.6 - Hantar terus Kepada Pn. Che Merah Ismail (RCMO) hanya salinan kepada Bhg. R&D, PPSP

My doc/checklist borang2/sue

UNIVERSITI SAINS MALAYSIA

LAPORAN AKHIR PROJEK PENYELIDIKAN JANGKA *PENDEK*

FINAL REPORT OF SHORT TERM RESEARCH PROJECT

Sila kemukakan laporan akhir ini melalui Jawatankuasa Penyelidikan di Pusat Pengajian dan Dekan/Pengarah/Ketua Jabatan kepada Pejabat Pelantar Penyelidikan

1. Nama Ketua Penyelidik: *Name of Research Leader* Hoh Boon Peng

☐ Profesor Madya/ *Assoc. Prof.* ☑ Dr./ *Dr.* ☐ Encik/ *Mr/Ms*

2. Pusat Tanggungjawab (PTJ):
School/Department

Pusat Pengajian Sains Perubatan

3. Nama Penyelidik Bersama: *Name of Co-Researcher* *Prof. Madya Dr. Zilfalil Alwi*
Dr Endom Ismail (UKM)

4. Tajuk Projek:
Title of Project

Genetic Relationships among the Negrito (Semang) Tribes of Peninsular Malaysia: the Microsatellite Approach

5. Ringkasan Penilaian/*Summary of Assessment*	**Tidak Memuaskan** *Inadequate* 1	2	**Boleh Diterima** *Acceptable* 3	**Sangat Baik** *Very Good* 4	5
i) Pencapaian objektif projek: *Achievement of project objectives*	☐	☐	☐	☑	☐
ii) Kualiti output: *Quality of outputs*	☐	☐	☐	☑	☐
iii) Kualiti impak: *Quality of impacts*	☐	☐	☑	☐	☐
iv) Pemindahan teknologi/potensi pengkomersialan: *Technology transfer/commercialization potential*	☐	☑	☐	☐	☐
v) Kualiti dan usahasama : *Quality and intensity of collaboration*	☐	☐	☑	☐	☐
vi) Penilaian kepentingan secara keseluruhan: *Overall assessment of benefits*	☐	☐	☐	☑	☐

6. **Abstrak Penyelidikan**
(Perlu disediakan di antara 100 - 200 perkataan di dalam **Bahasa Malaysia dan juga Bahasa Inggeris**. Abstrak ini akan diguatkan dalam Laporan Tahunan Bahagian Penyelidikan & Inovasi sebagai satu cara untuk menyampaikan dapatan projek tuan/puan kepada pihak Universiti & masyarakat luar).

Abstract of Research
(An abstract of between 100 and 200 words must be prepared in Bahasa Malaysia and in English). This abstract will be included in the Annual Report of the Research and Innovation Section at a later date as a means of presenting the project findings of the researcher/s to the University and the community at large)

REFER ATTACHMENT (Research Abstract and Comprehensive Technical Report)

7. **Sila sediakan laporan teknikal lengkap yang menerangkan keseluruhan projek ini.**
[Sila gunakan kertas berasingan]
Applicant are required to prepare a Comprehensive Technical Report explaning the project.
(This report must be appended separately)

REFER ATTACHMENT (Research Abstract and Comprehensive Technical Report)

Senaraikan kata kunci yang mencerminkan penyelidikan anda:
List the key words that reflects your research:

Bahasa Malaysia	Bahasa Inggeris
Penanda Genetik Mikrosatelit	Microsatellite Markers
Orang Asli Negrito	Orang Asli Negrito
Heterozigotisiti	Heterozigotisity

8. **Output dan Faedah Projek**
Output and Benefits of Project

(a) * **Penerbitan Jurnal**
Publication of Journals
(Sila nyatakan jenis, tajuk, pengarang/editor, tahun terbitan dan di mana telah diterbit/diserahkan)
(State type, title, author/editor, publication year and where it has been published/submitted)

1. Hoh B.P , Mohd Redhuan M.N , Nur Shafawati A.R , Zilfalil B.A. Genetic Variation among the Negrito (Semang) Tribes of Northern Peninsular Malaysia: A Microsatellite Approach. XX International Congress of Genetics, Berlin, Germany, 2008
2. Hoh BP, Nur Shafawati AR, Yam YY. Characterization of Seven (CA)n Microsatellite Markers among theThree Negrito Aborigines of Peninsular Malaysia. Russian Journal of Genetics. (Manuscript under review)

(b) **Faedah-faedah lain seperti perkembangan produk, pengkomersialan produk/pendaftaran paten atau impak kepada dasar dan masyarakat.**
State other benefits such as product development, product commercialisation/patent registration or impact on source and society.

This study helps in our understanding of pre-historic human migration in the Malay Archipelago which until today remains largely a mystery besides providing information for forensic profiling and the identification of complex diseases which may be prevalent in certain populations.

* Sila berikan salinan/*Kindly provide copies*

(c) **Latihan Sumber Manusia**
Training in Human Resources

i) Pelajar Sarjana:
Graduates Students
(Perincikan nama, ijazah dan status)
(Provide names, degrees and status)

ii) Lain-lain: 1 Bachelor final year project
Others

9. **Peralatan yang Telah Dibeli:**
Equipment that has been purchased

21/12/08

Tandatangan Penyelidik
Signature of Researcher

Tarikh
Date

Komen Jawatankuasa Penyelidikan Pusat Pengajian/Pusat
Comments by the Research Committees of Schools/Centres

Projek disiapkan dan mencapai objektif kajian.

22/3/09

16150 Kubang Kerian, Kelantan.

TANDATANGAN PENGERUSI
JAWATANKUASA PENYELIDIKAN
PUSAT PENGAJIAN/PUSAT
Signature of Chairman
[Research Committee of School/Centre]

Tarikh
Date

BORANG LAPORAN HASIL PENYELIDIKAN
PPSP

Tajuk geran: Genetic Relationship among the Negrito (Semang) Tribes of Peninsular Malaysia: the Microsatellite Approach
Penyelidik: Dr. Hoh Boon Peng
Jenis geran: USM-Short Term
Tempoh geran: 2 Tahun

Jenis laporan: Laporan Kemajuan [ ] Alatan di beli [ ] Ya : nyatakan.....
Laporan Akhir*: [✓] [✓] Tidak

OBJEKTIF SPESIFIK KAJIAN (sama spt dalam proposal asal)	**SECARA RINGKAS TERANGKAN PENCAPAIAN/HASIL**	**OBJEKTIF TERCAPAI ATAU TIDAK**
1. Generate the genotype data of the 10 selected autosomal microsatellite markers among the three Negrito tribes of Kelantan state	62 samples were collected and PCR optimizations were carried out on the 10 selected autosomal microsatellite markers. 7 were successfully amplified and genotyped. Due to time constrain, genotpying on the remaining 3 markers were not carried out.	Tercapai
2. To compare the allele frequencies and heterozygosities of these 3 tribes with other ethnic groups from the publicly available data	Allele frequencies and heterozygosities of the 7 markers were calculated among the 3 tribes and compared with the publicly available data (CEPH data). It was shown that the heterozygosities of the populations studied were lower than expected, suggesting the effect of inbreeding	Tercapai
3. To examine the Hardy-Weinberg Equilibrium of the population studied	Hardy-Weinberg Equilibrium was tested on the populations, found that majority of the markers were not in Hardy-Weinberg Equilibrium, further supporting the effects of inbreeding	Tercapai
4.		

** Laporan Akhir perlu disertakan salinan manuskrip dan surat yang dihantar kepada mana-mana jurnal untuk penerbitan.*

Nama Penyelidik Utama (PI): Dr Hoh Boon Peng
Tarikh: 22/12/08

RESEARCH ABSTRACT AND COMPREHENSIVE TECHNICAL REPORT

Abstrak Penyelidikan

Salah satu pendekatan dalam pengenalpastian gen penyakit kompleks ialah kajian terhadap population pedalaman seperti Orang Asli kerana pengaruh persekitaran yang kurang dan kandungan genetik yang kurang kompleks. Penanda genetik telah berkembang pesat terutama penanda genetik mikrosatelit kerana ia mempunyai nilai heterozigot yang tinggi, panjang jujukan yang berulang dan tahap polimorfisma yang tinggi. Suku kaum Negrito dipercayai orang asli pertama menduduki Semenanjung Malaysia dalam 10,000 tahun yang lalu. Kaum ini terbahagi kepada enam kumpulan iaitu Kensiu, Jahai, Bateq, Lanoh, Mendriq dan Kintak. Oleh itu, kajian ke atas kumpulan etnik ini memberi manfaat kepada pemetaan genetik serta pemahaman lebih mendalam dalam evolusi dan proses biologi sesuatu penyakit.

Objektif kajian ini adalah untuk menghasilkan data genotip untuk 10 penanda mikrosatelit antara 3 kumpulan sub-etnik Negrito di negeri Kelantan, iaitu, Bateq, Jahai dan Mendriq, dan seterusnya membandingkan frekuensi alel tersebut dengan kumpulan etnik yang lain dari pangkalan data umum. 10 penanda mikrosatelit yang mempunyai nilai heterozigot yang tinggi dipilih (Appendix I) dan tujuh telah digunakan. Nilai heterozigot yang terdapat pada sampel dikaji berbanding dengan pangkalan data umum, kecuali lokus D1S243, mencadangkan kecenderungan "kacukan dalaman" dan hanyutan genetik telah berlaku pada populasi Negrito. Kajian yang melibatkan penanda genetik dan sampel yang lebih banyak harus dijalankan untuk mengesahkan keputusan tersebut. Kajian ini juga akan bermanfaat dalam usaha memahami pra-sejarah migrasi manusia di arkipelago Melayu yang tidak diketahui sehingga kini, di samping memberi informasi kepada profil forensic dan penentuan penyakit kompleks yang mungkin lebih cenderung terhadap sesetengah populasi.

Research Abstract

One of the approaches to identify susceptibility genes for complex disorders is to focus on genetically isolated populations such as the Orang Asli populations rather than the

general population, because the environmental variation may be lower and the genetic make-up of these populations is expected to be less complex. Various markers have been used for genetic linkage mapping. Microsatellite markers are of interest to many researchers as they are more polymorphic and hence more informative (typically having more than ten alleles). Hence, they offer greater statistical power for the data analysis, whether by parametric analysis of large families, or nonparametric analysis of sib-pairs etc. The Negritos (Semang), believed as the first Orang Asli group to arrive in Peninsular Malaysia more than 10,000 years ago. They are divided into six subgroups, namely, Kensiu, Bateq, Mendriq, Jahai, Lanoh and Kintak. Therefore, studying this ethnic group could be useful for fine scale mapping as well as better understanding for the evolutionary and biological process leading to certain disease.

The objective of the study is to generate the genotype data of 10 selected microsatellite markers among the 3 Negrito tribes in Kelantan state, and later to compare the allele frequencies with other ethnic groups from publicly available data. 10 markers with the highest heterozygosities were chosen (Appendix I) and seven were applied. The observed heterozygosities of the seven loci were found to be significantly lower than those found in the published database, except for D1S243, suggesting the possibility of inbreeding and genetic drift in the Negrito population. A larger scale investigation using more samples and markers should be done to confirm the results. Such study would also help in our understanding of pre-historic human migration in the Malay Archipelago which until today remains largely a mystery besides providing information for forensic profiling and the identification of complex diseases which may be prevalent in certain populations.

Comprehensive Technical Report

Official consent was obtained from the JHEOA and the discrict representative respectively. Samples were collected from Pos Lebir, Kuala Lar, Sg Rual and Aring. A total of 105 samples were collected. However, only 62 were included into the study after a stringent sample inclusion criteria. This includes 18 Bateqs, 14 Mendriqs and 30 Jahais. The numbers of individuals in the tribes are decreasing and majority of them practice

consanguineous marriage, or inter-tribe marriage. Currently, there are about 1000 individuals in the Jahai tribe; 700 in Bateq tribe; and less than 80 individuals in Mendriq tribe. Thus, it is of reasonable that the sample collection is rather lesser than that of expected.

10 markers with the highest heterozygosities were chosen (Appendix I). Optimizations were done and 7 of the primers were successful. These markers were amplified on the samples and the fluorescent-labeled fragments were electrophoresed through ABI3100 and analyzed using GeneScan analysis software. The allele frequencies of the loci were calculated. Heterozygosities of the markers and possible error in genotyping or allele calling were checked and tested with Hardy-Weinberg's Equilibrium. Due to time constraint, the remaining STR primers were not further optimized.

The observed heterozygosities of the seven loci were found to be significantly lower than those found in the published database, except for D1S243. The lower observed heterozygosity among the Negritos compared to other published data suggested the possibility of inbreeding and genetic drift in the Negrito population in general. Analysis with a low value of Fst (0.0694) indicating the high genetic similarity among the three tribes, supported the suggestion; while the number of individuals in the tribes further proven the findings in this study.

If carried out using sufficient sample sizes and loci, a study such as this would be helpful in understanding pre-historic human migrations in the Malay Archipelago which currently remains largely unknown. On top of that, it may contribute additional information to forensic profiling and the identification of complex diseases which may be prevalent in certain populations.

Remark: This study was expected to be completed by March 2009. However, it is ended earlier than expected due to my resignation as an academic staff in School of Medical Sciences, USM, effective from January 2009.