

**ASSOCIATION BETWEEN INTERLEUKIN-10 (IL-10)
PROMOTER POLYMORPHISMS AND NON HODGKIN
LYMPHOMA IIN MALAYSIAN POPULATION**

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ABSTRACT

Non-Hodgkin Lymphoma is one of the malignant diseases that has the most rapidly increasing incidence in many countries. IL10 is a potent cytokine, which has been considered to be involved in pathogenesis of non-Hodgkin lymphoma. Furthermore allelic variations in the genes that has been implicated in inflammation, such as interleukin 10 are candidate risk factors for NHL. This study was been conducted as a case-control experiment on 320 patients with NHL, 285 clinically normal subjects for *IL10*-1082 G/A SNP and 312 patients with NHL, 284 clinically normal subjects for *IL10*-819 C/T SNP. After DNA extraction, Polymerase Chain Reaction (PCR) was carried out to amplify two promoter Single Nucleotide Polymorphisms (SNPs), *IL10*-1082 G/A (rs1800870) and *IL10*-819 C/T (rs1800871) of the *IL10* gene. PCR products were subjected to BseRI and MslI for genotyping of the *IL10*-1082 G/A and *IL10*-819 C/T respectively by analyzing the Restriction fragment length polymorphism (RFLP) patterns. Hardy-Weinberg equilibrium (HWE) of the genotype distribution in control population was examined with chi-square test. Association of the two SNPs with NHL in each of the three main ethnic groups (Malays, Chinese, and Indian) in Malaysia was examined using Fisher's exact test under allelic, dominant inheritance and recessive models. Genotype and allele frequencies of *IL10*-1082 G/A and *IL10*-819 C/T in all controls were in HWE. The results have indicated that *IL10*-1082 G/A polymorphism was not associated with NHL incidence. Conversely *IL10*-819 C/T was associated with risk of NHL only in the Malay population under the allelic and

dominant inheritance model ($P=0.0235$, $P=0.0297$ respectively). However, further studies with extended sample size are required in order to confirm the association between *IL10*-819 C/T and NHL in Malaysian Malays.

ABSTRAK

Non-Hodgkin Lymphoma adalah salah satu penyakit malignan yang mempunyai insiden yang paling pesat meningkat di banyak negara. IL10 adalah cytokine mujarab, yang telah dianggap sebagai terlibat dalam patogenesis limfoma non-Hodgkin. Tambahan pula allelic variasi dalam gen yang telah terbabit dalam keradangan, seperti interleukin 10 adalah faktor risiko calon untuk NHL. Kajian ini telah dijalankan sebagai uji kaji kes-kawalan ke atas 320 pesakit dengan NHL, 285 klinikal biasa subjek untuk IL10-1082 G / A SNP dan 312 pesakit dengan NHL, 284 mata pelajaran untuk IL10-819 C / T Taggart klinikal biasa. Selepas pengekstrakan DNA Polymerase Chain Reaction (PCR) telah dijalankan untuk menguatkan dua polimorfisme penganjur Nukleotida Tunggal (SNPs), IL10-1082 G / A (rs1800870) dan IL10-819 C / T (rs1800871) gen IL10. Produk PCR tertakluk kepada BseRI dan MslI untuk genotyping IL10-1082 G / A dan IL10-819 C / T masing-masing dengan menganalisis serpihan Sekatan panjang polymorphism (RFLP) corak. Keseimbangan Hardy-Weinberg (HWE) pengagihan genotip dalam kawalan penduduk telah diperiksa dengan ujian khi-kuasa. Persatuan SNPs dua dengan NHL dalam setiap satu daripada tiga kumpulan etnik utama (Melayu, Cina, dan India) di Malaysia telah diperiksa menggunakan ujian yang tepat Fisher di bawah pusaka allelic, dominan dan resesif model. Genotip dan frekuensi allele IL10-1082 G / A dan IL10-819 C / T dalam semua kawalan di HWE. Keputusan telah menyatakan bahawa IL10-1082 G / A polymorphism tidak dikaitkan dengan insiden NHL. Sebaliknya IL10-819 C / T telah dikaitkan dengan risiko NHL hanya di kalangan

penduduk Melayu di bawah model pusaka allelic dan dominan ($P = 0,0235$, $P = 0,0297$ masing-masing). Walau bagaimanapun, kajian lanjut dengan saiz sampel lanjutan diperlukan untuk mengesahkan persatuan antara IL10-819 C / T dan NHL Melayu Malaysia

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TABLE OF CONTENTS

ABSTRACT	I
ACKNOWLEDGEMENT	V
TABLE OF CONTENTS	VI
LIST OF FIGURES	VIII
LIST OF ABBREVIATIONS	X
1 INTRODUCTION	1
2 Objective	5
3 LITERATURE REVIEW	6
3.1 Cancer.....	6
3.2 Lymphoma	7
3.3 Non Hodgkin Lymphoma (NHL).....	8
3.3.1 NHL Causes	10
3.3.2 Symptoms of NHL.....	10
3.3.3 Treatments of NHL.....	10
3.3.4 Incidence of NHL in the world	11
3.3.5 Incidence of NHL in Malaysia.....	12
3.4 Single Nucleotide polymorphisms (SNPs).....	13
3.5 IL10 protein characteristic	15
3.6 IL-10 gene	16
3.7 Heritable production of IL10 protein and polymorphisms.....	17
3.8 IL10 (gene/protein) is associated with disease.....	19
3.9 IL10 protein and cancer.....	20
3.10 SNP genotyping methods	24
3.10.1 Allele discrimination methods	24
3.10.2 Signal Detection Method	26
4 Materials and Methods	27
4.1 Study Design	27
4.2 DNA Extraction.....	28
4.2.1 Blood DNA Extraction	28
4.2.2 Swab DNA Extraction	30
Material	31
4.3 DNA quantitation and qualification	31
4.3.1 Estimation of optical density (OD).....	32

4.3.2 Agarose gel electrophoresis	32
4.4 Selection of SNPs.....	33
4.5 Polymerase Chain Reaction (PCR)	34
PCR condition.....	36
4.5.1 Gel Electrophoresis for PCR Product	36
4.6 Restriction Fragment Length Polymorphism (RFLP) Analysis	37
4.6.1 Gel Electrophoresis for RFLP Digestion Products	38
Material	39
4.7 DNA Sequencing.....	40
4.8 Statistical analysis	41
5 RESULTS.....	43
5.1 Study population	43
5.2 DNA extraction and quantitation	44
5.3 PCR amplification of the promoter region IL10 gene.....	45
5.4 PCR-RFLP	46
<i>IL10</i> -1082 G/A.....	46
4.3.2 <i>IL10</i> -819 T/C	48
5.5 DNA sequencing	49
5.6 Statistical analyses.....	53
5.6.1 Chi-square (χ^2) test of Hardy-Weinberg equilibrium (HWE).....	53
5.6.2 Association of <i>IL10</i> -1082G/A with NHL	53
5.6.3 Association of <i>IL10</i> -819 C/T with NHL.....	54
6 DISCUSSION.....	59
6.1 Genomic DNA extraction and quantification.....	60
6.2 Genotyping method.....	60
6.3 Statistical analysis	61
7 CONCLUSION.....	64
REFERENCES:	65
APPENDIX B	78
APPENDIX C	79

LIST OF FIGURES

FIGURE 5-1: ETHIDIUM-BROMIDE STAINED 0.8% (w/v) AGAROSE GEL OF EXTRACTED GENOMIC DNA	45
FIGURE 5-2: PCR AMPLIFICATION OF THE PROMOTER REGION OF <i>IL10</i> GENE	46
FIGURE 5-3: ETHIDIUM-BROMIDE STAINED 3% (w/v) AGAROSE GEL OF PCR-RFLP PATTERN OF <i>IL10</i> 1082 G/A	47
FIGURE 5-4: ETHIDIUM-BROMIDE STAINED 3% (w/v) AGAROSE GEL OF PCR-RFLP PATTERN OF <i>IL10</i> 819 T/C	48
FIGURE 5-5: THE SEQUENCING RESULT OF HOMOZYGOUS WILD TYPE GENOTYPE OF <i>IL10</i> -819 C/T SNP (TT) ON THE REVERSE STRAND ON DNA.	50
FIGURE 5-6: THE SEQUENCING RESULT OF HOMOZYGOUS VARIANT GENOTYPE OF <i>IL10</i> -819 C/T SNP (CC) ON THE REVERSE STRAND ON DNA.	50
FIGURE 5-7: THE SEQUENCING RESULT OF HETEROZYGOUS GENOTYPE OF <i>IL10</i> -819 C/T SNP (CT) ON THE REVERSE STRAND OF DNA.	51
FIGURE 5-8: THE SEQUENCING RESULT OF HOMOZYGOUS WILD TYPE GENOTYPE OF <i>IL10</i> -1082 G/A SNP (AA)	51
FIGURE 5-9: THE SEQUENCING RESULT OF HETEROZYGOUS GENOTYPE OF <i>IL10</i> 1082 SNP G/A ON THE REVERSE STRAND OF DNA.	52
FIGURE 5-10: THE SEQUENCING RESULT OF HOMOZYGOUS VARIANT GENOTYPE OF <i>IL10</i> -1082 G/A SNP (GG) ON THE REVERSE STRAND OF DNA.	52

LIST OF TABLES

TABLE 4-4-1: DETAILS OF IL-10 SNPs SELECTED.....	34
TABLE 4-4-2: DETAILS OF PRIMER SEQUENCES, PCR AMPLICON SIZES	35
TABLE 4-4-3: REACTION MIXTURE FOR PCR AMPLIFICATION OF IL10 -1082 AND IL10 -819 SNPs.	35
TABLE 4-4-4: PCR CONDITION	36
TABLE 4-4-5: MASTER MIX FOR BSERI RESTRICTION ENZYME	38
TABLE 4-4-6: MASTER MIX FOR Ms1I RESTRICTION ENZYME	38
TABLE 4-4-7: DIGESTED FRAGMENTS YIELD SIZE.....	39
TABLE 5-1: ETHNICITY DISTRIBUTION OF 320 CASES AND 285 CONTROL SUBJECTS FOR IL10- 1082 G/A SNP	44
TABLE 5-2: ETHNICITY DISTRIBUTION OF 312 CASES AND 284 CONTROL SUBJECTS FOR IL10- 819 C/T SNP.....	44
TABLE 5-5-3: ASSOCIATION OF IL10 1082 G/A AND -819 C/T SNPs WITH NHL IN CHINESE POPULATION	56
TABLE 5-5-4: ASSOCIATION OF IL10 1082 AND 819 SNPs WITH NHL IN MALAYS POPULATION	57
TABLE 5-5-5: ASSOCIATION OF IL10 1082 AND 819 SNPs WITH NHL IN INDIAN POPULATION	58

LIST OF ABBREVIATIONS

%	Percentage
C ⁰	DegreeCelsius
µg	Microgram
µL	Microliter
µM	Micromolar
A	Adenine
AD	Alzheimer's disease
AIDS	Acquiredimmunodeficiency syndrome
ASO	allele-specificoligonucleotide
Bp	basepair
bcl2	B-cell lymphoma
C	Cytosine
CI	Confidence Interval
CHO	Chinese hamster ovary
CLL	Chronic lymphocyticleukemia
Df	Degree of freedom
dH ₂ o	Distilled water
DLBCL	Diffuse large B-cell Lymphoma
DNA	Deoxyribonucleic acid
dNTP	Deoxynucleoside triphosphate
EBV	Epstein-Barr Virus
EDTA	Ethylenedinitrilotetraacetic acid
Ets	E-twenty-six specific
FFP	freedom from progression

LIST OF ABBREVIATIONS

FL	Follicular lymphoma
g	gram
G	Guanine
HCV	Hepatitis C virus
HD	Hodgkin's disease
HET	Hetrozygous
HIV	Human immunodeficiency
HL	Hodgkin's lymphoma
H-pylori	Helicobacter pylori
HTLV-1	human T-lymphotropic virus type 1
HWE	Hardy-Weinberg equilibrium
IARC	International Agency for Research on Cancer
IL10	Interleukin 10
Ig	Immunoglobulin
L	Liter
LPS	Lipopolysaccharide
M	Molar
MAF	Minor Allele Frequency
MALDI-TOF	Matrix-assistedlaser esorption/ionization time of Flight
mg	Milligram
Mgcl ₂	Magnesium chloride
Min	Minutes
mL	Milliliter
MLP	Multi-locus probe
mM	millimolar
mRNA	Messenger RNA

LIST OF ABBREVIATIONS

MHC	MajorHistocompatibility Complex
MMPs	Matrix metalloproteases
MS	multiple sclerosis
NaCl	Sodium chloride
NCI	National Cancer Institute
NCBI	National Center for Biotechnology
ng	Nanogram
NHL	Non-Hodgkin's Lymphoma
NK	Natural Killer
NO	Nitric oxide
OD	Optical density
OS	overall survival
OR	Odds Ratio
PCR	Polymerase Chain Reaction
Pf	Primer forward
Pr	Primer reverse
RA	Rheumaoid Arthritis
RCLB	Red cell lysis buffer
RE	Restriction enzyme
RFLP	Restriction Fragment Length Polymorphism
RNA	Ribonucleic acid
Rpm	Rounds per minute
SCID	Severe Combined Immunodeficiency
Sec	Second
SDS	Sodium dodecyl sulfata
SNP	Single nucleotide Polymorphism
SEER	Surveillance, Epidemiology, and End Results

LIST OF ABBREVIATIONS

SLE	Systemic lupus erythematosus
SLP	paradigms and single-locusprobe
Sp1	Specificity Protein 1
Sp3	Specificity Protein 3
T	Thymine
TB	Tuberculosis
TBE	Tris- base EDTA
Th1	T-helper subtypes 1
Th2	T-helper subtypes 2
TIMP	tissue inhibitors of metalloproteases
Tm	Melting temperture
TNF-a	tumour necrosis factor a
TNP	Tumor necrosis factor
Tr 1	Treg Cell 1
USA	United State America
UTR	Untranslated region
UV	Ultraviolet
V	Volt
V/V	Volume per volume
VAR	Variant
WHO	World Health Organization
W/V	Weight per Volume
WT	Wild type
χ^2	Chi-square