

ความสัมพันธ์ของ single nucleotide polymorphism ในยีน vascular endothelial growth factor กับการเกิดโรคและความรุนแรงของโรคสะกดเงินในประเทศไทย

นายสุรศักดิ์ อ้อยงสะถิต

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ลิขสิทธิ์ของจุฬาลงกรณ์มหาวิทยาลัย

THE ASSOCIATION BETWEEN SINGLE NUCLEOTIDE POLYMORPHISM WITHIN
VASCULAR ENDOTHELIAL GROWTH FACTOR GENE WITH DISEASE
SUSCEPTIBILITY AND SEVERITY OF PSORIASIS IN THAI POPULATION

Mr. Surasak Yooyongsatit

A Thesis Submitted in Partial Fulfillment of the Requirements
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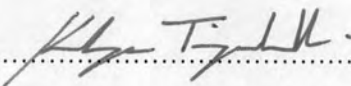
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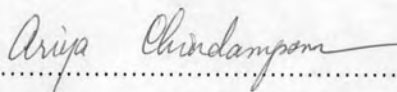
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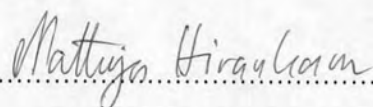
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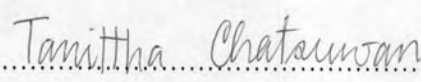

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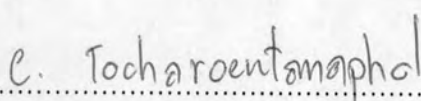
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(Associate Professor Nattiya Hirankarn, M.D., Ph.D.)


.....Member
(Tanittha Chatsuwon, Ph.D.)


.....Member
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สุรศักดิ์ อยู่ยงละถิต : ความสัมพันธ์ของ Single nucleotide polymorphism ในยีน Vascular endothelial growth factor กับการเกิดโรคและความรุนแรงของโรคสะเก็ดเงิน ในประชากรไทย (THE ASSOCIATION BETWEEN SINGLE NUCLEOTIDE POLYMORPHISM WITHIN VASCULAR ENDOTHELIAL GROWTH FACTOR GENE WITH DISEASE SUSCEPTIBILITY AND SEVERITY OF PSORIASIS IN THAI POPULATION) อ. ที่ปรึกษา: ผศ.พญ.ดร. จงกลณี วงศ์ปิยะบวร, อ. ที่ปรึกษาร่วม: รศ. พญ.ดร. ญัญฉิยา หิรัญกาญจน์, 123 หน้า

โรคสะเก็ดเงินเป็นโรคผิวหนังเรื้อรัง ลักษณะของผื่นคือ เป็นผื่นนูนหนาสีแดงขอบเขตชัดเจนปกคลุมด้วยสะเก็ดเงิน จัดเป็นโรคภูมิคุ้มกันตนเองชนิด T-cell โดยไซโตไคน์มีบทบาทสำคัญในการดำเนินโรค พบว่าปัจจัยทางพันธุกรรมและปัจจัยทางสิ่งแวดล้อมมีส่วนเกี่ยวข้องกับพยาธิสภาพของโรค มีรายงานการศึกษาทางพันธุกรรมจำนวนมาก ซึ่งส่วนใหญ่ทำโดยวิธี linkage analysis และ association study รายงานอื่นที่เกี่ยวข้องกับโรคสะเก็ดเงินมากมาย หลายรายงานสรุปว่ายีน VEGF เป็นยีนที่มีตำแหน่งอยู่บนโครโมโซม 6p21 มีบทบาทสำคัญในโรคที่มีพื้นฐานเกี่ยวกับการสร้างหลอดเลือดใหม่ เช่น ในมะเร็งชนิดต่างๆ รวมทั้งโรคสะเก็ดเงินด้วย งานวิจัยนี้ศึกษาความสัมพันธ์ระหว่างความหลากหลายของยีน VEGF และความเสี่ยงต่อการเกิดโรคสะเก็ดเงิน ใช้วิธี PCR-SSP และ PCR-RFLP โดยศึกษาแบบ population-based case-control จากผู้ป่วยโรคสะเก็ดเงินจำนวน 154 คน แบ่งเป็นผู้ป่วยที่เกิดโรคก่อนอายุ 40 ปี (early-onset) จำนวน 102 คน และผู้ป่วยที่เกิดโรคเมื่ออายุ 40 ปี หรือมากกว่า (late-onset) จำนวน 52 คน เปรียบเทียบกับกลุ่มคนปกติจำนวน 234 คน ผลการศึกษาพบว่ารูปแบบของ -460TT หรือ -460TC มีความสัมพันธ์กับความเสี่ยงต่อการเกิดโรคสะเก็ดเงินในช่วงอายุน้อยกว่า 40 ปี อย่างมีนัยสำคัญทางสถิติ ($p=0.0450$, $OR=4.67$, $95\%CI=1.03-29.52$) นอกจากนี้เป็นที่น่าสนใจว่า เมื่อวิเคราะห์รูปแบบ haplotype ของยีน VEGF พบว่า CTG haplotype มีความสัมพันธ์กับความเสี่ยงต่อการเกิดโรคสะเก็ดเงิน การเกิดโรคในช่วงอายุน้อยกว่า 40 ปี และความรุนแรงของโรค อย่างมีนัยสำคัญทางสถิติ ($p=0.0307$, $OR=1.40$, $95\%CI=1.03-1.91$, $p=0.0098$, $OR=1.58$, $95\%CI=1.11-2.24$, $p=0.0475$, $OR=1.70$, $95\%CI=1.01-2.87$) ตามลำดับ และนอกจากนั้น CTG/CTG หรือ CTG/other haplotype ยังพบว่ามีความสัมพันธ์กับความเสี่ยงต่อการเกิดโรคสะเก็ดเงิน และการเกิดโรคในช่วงอายุน้อยกว่า 40 ปี อย่างมีนัยสำคัญทางสถิติเช่นเดียวกัน ($p=0.0078$, $OR=1.81$, $95\%CI=1.16-2.84$, $p=0.0028$, $OR=2.20$, $95\%CI=1.29-3.74$) ตามลำดับ นอกจากนี้ไม่พบความแตกต่างของระดับ VEGF ใน plasma ของผู้ป่วยระหว่างกลุ่ม haplotypes ที่แตกต่างกัน จากผลการศึกษาครั้งนี้ สรุปได้ว่า CTG haplotype และ -460C/T polymorphism สามารถใช้เป็นเครื่องหมายของยีนในการกำหนดความเสี่ยงต่อการเกิดโรคสะเก็ดเงินในช่วงอายุน้อยกว่า 40 ปีในประชากรไทย

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ลายมือชื่ออาจารย์ที่ปรึกษา..... Jaykan Uayriyaku

ลายมือชื่ออาจารย์ที่ปรึกษาร่วม.....

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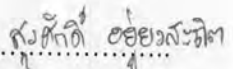
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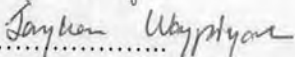
SURASAK YOOYONGSATIT: THE ASSOCIATION BETWEEN SINGLE
NUCLEOTIDE POLYMORPHISM WITHIN VASCULAR ENDOTHELIAL GROWTH
FACTOR GENE WITH DISEASE SUSCEPTIBILITY AND SEVERITY OF
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JONGKONNEE WONGPIYABOVORN, M.D., Ph.D. THESIS CO- ADVISOR:
ASSOC. PROF. NATTIYA HIRANKARN, M.D., Ph.D., 123 pp.

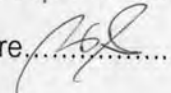
Psoriasis is a common chronic skin disease that is characterized by sharply demarcated erythematous plaque covered with silvery scales. Psoriasis is a T-cell-mediated skin autoimmunity, in which cytokines, play important role to be a key element in the disease progression. Psoriasis is a multifactorial disease requiring environmental trigger and genetic susceptibility factors to become manifested. Many genetic studies by linkage analysis and association study implicated that various genes are related to psoriasis. However, many studies of vascular endothelial growth factor (VEGF) gene, located on chromosome 6p21, is proposed to play important parts in pathogenesis of various diseases with angiogenesis basis such as breast cancer, cutaneous malignant melanoma and autoimmune disease including psoriasis. In this study, the association between psoriasis susceptibility and three functional single nucleotide polymorphisms (SNPs) in VEGF promoter and exon1 (at position -1557 C/A, -460 C/T and +405 C/G) were investigated. Genotyping was performed by polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) and PCR-sequence specific primer (PCR-SSP) methods. In population-based case-control study, allele and genotype frequencies of each marker were compared between 234 unrelated healthy volunteers and 154 chronic plaque psoriasis patients (102 early-onset and 52 late-onset psoriasis). The result of this study demonstrated that the -460 TT or CT compared to CC genotype were found to be significantly risk associated with patients with early-onset psoriasis compared with normal controls ($p=0.0450$, $OR=4.67$, $95\%CI=1.03-29.52$). Interestingly, haplotype analysis revealed that the CTG haplotype was found to be significantly associated with psoriasis patients, early -onset and severe psoriasis compared with normal controls ($p=0.0307$, $OR=1.40$, $95\%CI=1.03-1.91$, $p=0.0098$, $OR=1.58$, $95\%CI=1.11-2.24$, $p=0.0475$, $OR=1.70$, $95\%CI=1.01-2.87$, respectively). Moreover, The CTG/CTG or CTG/other genotype compared to those other genotypes were significantly associated with psoriasis patients and early-onset psoriasis compared with normal controls ($p=0.0078$, $OR=1.81$, $95\%CI=1.16-2.84$, $p=0.0028$, $OR=2.20$, $95\%CI=1.29-3.74$, respectively) in the dominance mode of inheritance. Moreover, VEGF plasma concentration was not significantly different between groups of haplotype in psoriasis patients. The CTG haplotype and -460 C/T polymorphism can be used as a genetic marker for early-onset psoriasis in Thais.

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Student' signature..... 

Advisor' signature..... 

Co-advisor' signature..... 

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ABBREVIATIONS

APCs	Antigen-Presenting Cells
bp	base pair
CD	Cluster of Differentiation
95% CI	95% Confidence Interval
°C	degree Celsius
CTL	Cytotoxic T Lymphocyte
et al	et alii
IL	Interleukin
IFN	Interferon
kDa	Kilodalton
HLA	Human Leukocyte Antigen
μl	microliter
μg	microgram
ml	milliliter
mM	millimolar
MW	molecular weight
ng	nanogram
NK	Natural Killer
OR	Odd Ratio
PCR	Polymerase Chain Reaction
RFLP	restriction fragment-length polymorphism
RAPD	Random Amplified Fragment-length polymorphism
SSP	sequence specific primer
SDS	Sodium Dodecyl sulfate
SNP	Single Nucleotide Polymorphism

TNF	Tumor necrosis factor
U	Unit
SCID	Severe Combine Immunodeficiency Mice
PSORS	Psoriasis Susceptibility loci
VEGF	Vascular Endothelial Growth Factor
Kb	Kilo Base Pair
PLGF	Placental Growth Factor
PASI	Psoriasis Area and Severity Index
NKT	Natural Killer T Cell
KC	Keratinocyte
TARC	Thymus and Activation-regulated Chemokine
MIG	Monokine Induced by Interferon-Gamma
IP	Interferon Inducible Protein
MDC	Macrophage-Derived Chemokine
RANTES	Regulated on Activation, T-cell Expressed and Secreted
CXCR	Chemokine-related Receptor
CCR	Chemokine Receptor
MIP	Macrophage Inflammatory Protein
TGF	Transforming Growth Factor
IGF	Insulin-like Growth Factor
KGF	Keratinocyte Growth Factor
NGF	Nerve Growth Factor
MHC	Major Histocompatibility Complex
CDSN	Corneodesmosin
HCR	Coiled-coil α -Helical Rod protein
cM	Centi-Morgan
PIGF	Placental Growth Factor
flt-1	fms-like tyrosine kinase

Flk-1/KDR	Fetal liver kinase 1-murine homologue/Kinase insert Domain containing Receptor-human homologue
EDTA	Ethylene-Diamine-Tetra-acetic Acid
TF	Transcription Factor