THE C677T MTHFR GENE POLYMORPHISM IN PATIENTS WITH MONOSOMY X AND ITS CYTOGENETIC VARIATION

POLIMORFISME C677T GEN MTHFR PADA PASIEN DENGAN MONOSOMI X DAN VARIASI SITOGENETIKNYA

Thesis
Submitted to fulfill the assignment and fit-out requisite
in passing Post-graduate program Majoring Genetics Counseling
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Declaration

I hereby declare that this submission is my own work and that, to the best of my knowledge and belief, it contains no material previously published or written by another person nor material which to a substantial extent has been accepted for the award of any other degree or diploma of the university or other institute of higher learning, except where due acknowledgement is made in the text.

Semarang, April 2012

Puspita Kusuma Dewi

Curriculum vitae

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Educations

- High School in SMU N I Salatiga (2000 - 2003)
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Scholarship Award

Beasiswa Unggulan BPKLN Ministry of Education and Culture, Republic of Indonesia, at KK Women's and Children's Hospital Singapore for clinical and laboratory attachment (2010-2012)

Work experiences

1. General Practitioner at Primary Health Centre, Welahan Jepara, Indonesia (2009)
2. General Practitioner at District Hospital, Jepara, Indonesia (2009)
3. Academic staff at Department of Biology, Faculty of Medicine Diponegoro University (2009 – present)

Seminars and Courses

1. Training Management of Emergency Patient, conducted by Faculty of Medicine Diponegoro University. in collaboration with Indonesian Medical Association, Semarang, Indonesia (June 2007)
2. Advanced Cardiac Life Support Training, conducted by Indonesian Heart Association, Semarang, Indonesia (November 2009)
3. Medical Genetic Course : From Basic to Clinic, conducted by Center for Biomedical Research (Cebior), Faculty of Medicine Diponegoro University in collaboration with Department of Human Genetics Radboud University Nijmegen Medical Centre, Semarang, Indonesia (January 2010)
4. Hematology-Medical Oncology Update : From Bench to Bed, conducted by Internal Medicine Department of Dr. Kariadi Hospital in collaboration with Radboud University Nijmegen Medical Centre, Semarang, Indonesia (January 2010)
5. 2nd International Seminar and Workshop on Fragile-X Autism and Related Disorders, conducted by Center for Biomedical Research (Cebior), Faculty of Medicine Diponegoro University in collaboration with Medical Investigation of Neurodevelopmental Disorders (MIND) Institute UC Davis, Semarang, Indonesia (August 2010)
6. 6th KKH Annual Scientific Meeting, Academic Excellence Towards Better Healthcare for Women and Children, conducted by KK Women's and Children's Hospital Singapore (July 2011)

Research Experiences

1. The difference of heart failure comorbidity on the elderly with on the adult in Kariadi hospital from January to December 2006. Paper for undergraduate research project (2006-2007)
2. Nuchal Translucency Thickness in the Screening of Congenital Heart Defects. Research collaboration with KK Women's and Children's Hospital Singapore (2011)

Publications

Dewi PK, Armalina D. MTHFR and MTR gene polymorphism as maternal risk factors for Down Syndrome. Poster presentation: Biomolecular and Cellular Anatomy. 6th APICA & 13th PIN-PAAI (Surabaya, July 2011)

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Table of Contents

List of Tables

List of Figures

Abbreviation List

5,10-methyleneTHF	: 5,10-methylenetetrahydrofolate
5-methylTHF	: 5-methyltetrahydrofolate
CEBIOR	: Center for Biomedical Research
DNA	: Deoxyribonucleic acid
DS	: Down Syndrome
DSD	: Disorders of Sex Development
FISH	: Fluorecense In Situ Hybridization
H-W	: Hardy-Weinberg
MGD	: Mixed Gonadal Dysgenesis
MTHFR	: Methylenetetrahydrofolate reductase
NTD	: Neural Tube Defect
PCR	: Polymerase Chain Reaction
RFLP	: Restriction Fragment Length Polymorphism
RNA	: Ribonucleic acid
SAM	: S-adenosylmethionine

List of Appendices

Abstract

Background: Abnormal chromosomal segregation in monosomy X and its variants was thought correlate with the C677T polymorphism in Methylenetetrahydrofolate reductase (MTHFR) gene by DNA hypomethylation of centromic and pericentromic regions. This study aims to describe the distribution of C677T MTHFR gene polymorphism of patients with monosomy X and its cytogenetic variants in Indonesian population

Methods: MTHFR gene polymophism was analyzed using PCR and RFLP in patients with monosomy X and its variants who underwent cytogenetic examination in CEBIOR, Semarang in 2000-2011. Genotype and allele frequency were calculated using Gene Counting Method. Statistical test for deviation from the Hardy-Weinberg Equilibrium was performed using Pearson's Chi-squared test.

Result: The frequencies of three genotypes were CC 0.76, CT 0.18, and T/T 0,06. The allele frequency for C is 0.84 and for T is 0.16. Genotype and allele frequency in this study is not in the Hardy-Weinberg equilibrium. There was no significant different in the TT genotype distributions considering mosaic and non-mosaic groups.

Conclusion: The C677T MTHFR gene polymorphism was found in patients with monosomy X and its variants, with distribution as follows: CC 0.76, CT 0.18, and T/T 0,06. The allele frequency for C is 0.84 and for T is 0.16 . The T allele frequency in patients with monosomy X and its variants was higher than in normal population, which will facilitate further investigations of the pathogenic effects of the gene.

Keywords: *MTHFR gene, polymorphism,chromosomal non disjuction, monosomy X*

Abstrak

Latar belakang: Abnormalitas pemisahan kromosom pada monosomi X dan variasinya dianggap berhubungan dengan adanya polimorfisme C677T pada gen MTHFR melalui hipometilasi DNA daerah sentromer dan parasentromer. Penelitian ini bertujuan untuk menggambarkan distribusi polimorfisme C677T gen MTHFR pada pasien dengan monosomi X dan variasi sitogenetiknya pada populasi Indonesia

Metode: Polimorfisme gen MTHFR dianalisis dengan menggunakan PCR dan RFLP pada pasien dengan monosomi X dan variasinya yang menjalani pemeriksaan sitogenetik di CEBIOR, Semarang pada tahun 2000-2011. Frekuensi genotipe dan alel dihitung dengan menggunakan *Gene Counting Method*. Uji statistik untuk deviasi terhadap *Hardy-Weinberg Equilibrium* dikerjakan dengan Uji Khi-kuadrat Pearson.

Hasil: Frekuensi ketiga genotipe adalah CC 0.76, CT 0.18, and T/T 0,06. Frekuensi alel untuk C adalah 0.84 dan untuk T adalah 0.16. frekuensi genotipe dan alel pada penelitian ini tidak berada pada *Hardy-Weinberg equilibrium.* Tidak terdapat perbedaan yang signifikan pada distribusi genotipe TT pada kelompok mosaik dan non-mosaik.

Simpulan: Polimorfisme C677T gen MTHFR ditemukan pada pasien dengan monosomi X dan variasinya, dengan distribusi berikut: CC 0.76, CT 0.18, and T/T 0,06. Frekuensi alel untuk C adalah 0.84 dan untuk T adalah 0.16. Frekuensi alel T pada pasien dengan monosomi X dan variasinya lebih tinggi dibandingkan dengan populasi normal, yang akan memfasilitasi penelitian lebih lanjut mengenai efek patogenik dari gen.

Kata kunci: gen MTHFR, polimorfisme, *"chromosomal non-dijunction",* monosomy X