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CCR5Δ32 POLYMORPHISM NOT DETECTED IN HIV PATIENTS IN VOLUNTARY COUNSELING AND TESTING MOEWARDI GENERAL HOSPITAL SURAKARTA, INDONESIA

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ABSTRACT

Background: The CCR5Δ32 polymorphism (a naturally occurring 32-bp deletion in CCR5) influences the ability of HIV-1 to infect the target cells. Homozygosity for CCR5Δ32 prevents infection of HIV-1 R5 strain, while the heterozygous is associated with lower plasma viral load and delayed progression to acquired immune deficiency syndrome (AIDS). However, there is no report about the presentation of CCR5Δ32 polymorphisms in Indonesian HIV patients. The aim of this study is to detect CCR5Δ32 polymorphisms in Indonesian HIV patients, especially in Voluntary Counseling and Testing Moewardi General Hospital Surakarta, Indonesia. In an ongoing molecular epidemiology study of blood borne virus, 154 HIV patients in Moewardi General Hospital Surakarta were used for the study. The blood samples were collected during November – December 2011. The blood samples were aliquoted and fractionated. The DNA was extracted from all blood samples, and subjected for the PCR assay to detect the presentation of CCR5Δ32 polymorphisms. Internal amplification control was included in all assays. PCR products were analyzed in 3% agarose. The results showed that CCR5Δ32 polymorphism was not detected in all blood samples. So it can be concluded that all patients in this study had the CCR5 wild type.

Key words: CCR5Δ32, HIV, Indonesia.

INTRODUCTION

Human immunodeficiency virus (HIV) entry into target cells is a multi-step process involving binding of the viral glycoprotein, *Env*, to its receptor CD4 and a co-receptor of CCR5 or CXCR4. Understanding the means by which HIV enters cells has led to the identification of the CCR5Δ32 polymorphism that confers resistance to infection in homozygous individuals, and has also resulted in the development of entry inhibitors-small molecule antagonists that block infection at the entry step (Didigu & Doms, 2012; Grivel *et al.*, 2011). A natural deletion of 32 bases in CCR5 (CCR5Δ32) gene resulting in truncated protein product (Liu *et al.*, 2012). People homozygous for CCR5Ä32 are naturally resistant to R5 HIV infection and the heterozygous state is associated with up to 2–4 years delay in disease progression (Gupta & Padh, 2012). This polymorphism was found in Caucasians and in Chinese state high (Liu *et al.*, 2012) and exists at allele frequencies of typically 10% in European populations (Hyde *et al.*, 2010). However, to our knowledge, the presence of CCR5Δ32 polymorphisms

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in Indonesian HIV patients has not been reported. The research aims to detect the CCR5Δ32 polymorphism in HIV patients in Indonesia, especially in Surakarta, Indonesia.

MATERIALS AND METHODS

In November – December 2011, HIV patients from Surakarta and its surrounding area of Voluntary Counseling and Testing Moewardi General Hospital Surakarta (n= 154) were invited for the ongoing blood borne virus molecular epidemiology study. The ethical issues of this blood sample collection had been approved by the institutional ethical committee review boards of the Faculty of Medicine of Sebelas Maret University and Dr. Moewardi General Hospital, Surakarta, Indonesia. Blood samples collected from the patients were fractionated, aliquoted and kept frozen until further analysis. All the procedures were conducted according to the principles of the Declaration of Helsinki.

The nucleic acid was extracted from all blood samples using High Pure PCR Template Preparation Kits (Roche Diagnostic, Mannheim, Germany) and subjected for the PCR assay to detect the presence of CCR5Δ32 polymorphisms using primer set as described previously (Huang *et al.*, 1996), by Amplitaq Gold® 360 DNA Polymerase Kit (Invitrogen, Carlsbad, CA). Human β-globin was included as internal amplification control in all assays. PCR products were analyzed in 3% Agarose.

RESULTS AND DISCUSSION

Using the primer set used the length for CCR5 wild type is 189 bp, but for CCR5Δ32 is 157 bp (Huang *et al.*, 1996). The human β-globin PCR product is 100 bp and used as internal amplification control to exclude the false negative results. All genomic DNA were successfully isolated and amplified, showed by the presentation of two expected bands in 3% agarose after electrophoresis, 100 bp and 189 bp bands, and no sample showed 100 bp and 157 bp bands. Therefore, in this present study, all patients in the study had the CCR5 wild type, and no CCR5Δ32 polymorphism was detected. Our results indicated that CCR5Δ32 polymorphism maybe not exist in Indonesia population, at least in HIV patients involved in the study. However, due to small number of patients enrolled in the study and only HIV patients from Surakarta and its surrounding area were assayed in the present study, a bigger and more adequate sampling method is needed to clarify the presentation of CCR5Δ32 polymorphism in Indonesian.

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