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Original Article

POLYMORPHISM *OF ORGANIC CATION TRANSPORTER 1 (OCT1*) IN INDONESIAN CANCER PATIENTS

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ABSTRACT

Objective*: Organic Cation Transporter1 (OCT1)*, was known as the highly polymorphism of polyspesific transporter. The genetic variability of *OCT1* might alter the drug response, such as metformin, imatinib, levodopa and 5-Hydroxytriptamine Receptor Antagonist (5-HTRA) drugs. This study was aimed to know the variability of *OCT1* gene in Indonesian Cancer patients.

Methods: The SNPs of *OCT 1*; rs12208357, rs55918055, rs34130495, rs34059508 and amino acid substitution Meth420del were selected. Genotypes were established using Taqman assays and analysed on ABI 7500 realtime PCR System from Applied Biosystems according to manufacture's protocol of allelic discrimination.

Results: There were no genetic variations found in the 202 Indonesian cancer patients according to the 4 SNPs included in this study and only 1.5% heterozygous were found in the Met420del. This finding is consistent with the previous studies in Japannese and Chinnese population.

Conclusion: The further study is warranted to do the sequencing analysis which could find the most possible variants in Indonesian cancer patients.

Keywords: *OCT1*, Indonesian, Cancer.

INTRODUCTION

Organic Cation Transporter1 (OCT1), encoded by *SLC22A1*, was located in chromosome 6q26 and was known as largest superfamily of transporters [1]. *OCT1* was majorly expressed in the liver, it means that the variations of this gene would affect the transport of the drug to the liver [2]. Some of the mechanism was proposed as the mechanism which could explain the role of *OCT1* polymorphisms to the drug response. The most possible mechanism was probably related to the drug disposition in the liver which relevant to the pharmacodynamics matter [3]. For the drugs which are being the substrate of *OCT1* and are metabolized majorly by CYP2D6 which was known highly polymorphism, probably need individualization of dose administration in different ethnicities.

In the previous studies of Caucasian subjects, the polymorphisms of *OCT1* could alter the metformin, imatinib and levodopa response [4-8]. Moreover, the newest study in Caucasian cancer patients found that the polymorphisms of *OCT1* could affect the patients response to ondansetron and tropisetron as antiemetic drugs [9]. Ondansetron and tropisetron, the 5-Hydroxytriptamine Receptor Antagonists (5-HTRA), are known as the antiemetics used in chemotherapy-induced nausea vomiting (CINV). In the previous study of Indonesian cancer subjects, the polymorphisms of *5-HT3B* gene receptor, *CYP2D6* and *Multi Drug Resistance1* (*MDR1*) gene were not associated to the ondansetron response. However, there were still 30% patients experienced CINV after treated by highly emetogenic cytostatic agent [10]. Thus, we should consider the *OCT1* polymorphisms as the predictor of drug response, besides considering the subject characteristic.

The previous study in Japannese and Chinese subjects with diabetes melitus disease suggested that new nonsynonymous variants found could affect the metformin response. However, the nonsynonymous variants found in the Caucasian subjects were not found in their study [11]. Moreover, the previous study in Korean population also showed the nonsynonymous variants similar to the Vietnamese and Chinese and different variants from the Caucasians [12].

This study was aimed to find the variations of *OCT1* gene in Indonesian cancer patients which could be related to the 5-HTRA drug response, since the association between the drug response and the other genes in the previous study were not found [10].

MATERIALS AND METHODS

SNPs selection

Five SNPs of *OCT 1* were selected from the study of Tzvetkov et al [9]; rs12208357 (amino acid substitution arginine61-to-cycteine), rs55918055 (amino acid substitution cycteine88-to-arginine), rs34130495 (amino acid substitution glycine401-to-serine), rs34059508 (amino acid substitution glycine465-to-arginine) and unknown rs number (amino acid substitution methionine420-to-deletion methionine). This study has been approved by the ethic committee of Medicine Faculty, University of Gadjah Mada, Yogyakarta. This study has been permitted by Faculty of Medicine Gadjah Mada University, Yogyakarta.

DNA collection

The deoxyribonucleic acid (DNA) was collected from 202 saliva samples of Indonesian cancer patients. Saliva was collected at least one hour after eating meal. The patients were asked to rinse their mouth before the saliva collection with plain water. After the saliva was collected in the saliva container, the container was closed and mix gently for 10 seconds. The containers were put in the room with ambient temperature.

DNA extraction

40µl DNA purifier was added to 1000 µl saliva solution in the microcentrifuge tube and vortex the tube for 5 seconds. The tube was incubated in the ice for 10 minutes then it was centrifuged in the room temperature for 5 minutes at 13.000 rpm. 1000 µl supernatant was transferred into clean tube and was added by 1000µl-100% ethanol. The solution was mixed gently for 10 times and allowed in the room temperature for 10 minutes or until the DNA was fully precipitated. The supernatant was removed carefully and 100 µl TE buffer was added to DNA pellet. The solution was vortexed for 5 seconds then incubated in the room temperature for 1-2 days. DNA was quantified using Nanodrop (Isogen, Maarssen, The Netherlands).

Allelic Discrimination

Genotypes were established using Taqman assays and analysed on ABI 7500 realtime PCR System from Applied Biosystems (Nieuwerkerk aan den IJssel, The Netherlands) according to the protocol of allelic discrimination. The solution for allelic discrimination were contained of Taqman Universal PCR Master Mix, 2X, (12.50 µl) and SNP genotyping assay (1.25 µl) for 96 wells plate in each reaction. 13.75 µl solution was pipette in to each well of the plate. The plate was performed for pre – read and amplification running of allelic discrimination. The amplification running used the condition of : 2 minutes of 50ºC and 10 minutes of 95 ºC for initial steps and 40 cycles of 15 seconds 95 ºC, 40 cycles of 1 minutes 60 ºC for PCR. This system is known as defaults times and temperature in ABI 7500 realtime PCR System from Applied Biosystems. After the amplification procedures, the post-read assay was performed in the machine to get the genotype data.

Statistical analysis

The genotype frequencies were assessed for deviations from Hardy Weinberg equilibrium and the genotype distributions did not deviate from Hardy Weinberg equilibrium.

RESULTS

We did not find the heterozygous and homozygous mutant over the rs12208357 (amino acid substitution arginine61-to-cycteine), rs55918055 (amino acid substitution cycteine88-to-arginine), rs34130495 (amino acid substitution glycine401-to-serine), rs34059508 (amino acid substitution glycine465-to-arginine).

However, there were 1.5% heterozygous found in unknown rs number (amino acid substitution methionine420-to-deletion methionine). The Table 1 showed the finding of this study.

Table 1: It shows the genotyping assay result over the five SNPs of *OCT1*

SNP	Variants	% from 202 subjects
rs12208357	Wild type; CC	100
rs55918055	Wild type; TT	100
rs34130495	Wild type; GG	100
rs34059508	Wild type; GG	100
Met420del	Wild type; ATG	98.5
	Heterozygous; del/ATG	1.5

C: cytosine; T: Thymine; A: Adenine; G: Guanine; del: deletion; Met: methionine

DISCUSSION

This study was aimed to undrstand the variability of *OCT1* gene in Indonesian Cancer patients. Our finding was consistent with the previous study in Japannese and Chinnese. This previous study found the other new nonsynonymous variants which associated to the metformin response. The Q97K, P117L and R206C could alter the pharmacokinetic parameters of metformin [11]. In addition, the previous study of Korean population, there were four SNPs involved in their study, such as; F160L, M408V, P283L and P341L. The first two SNPs did not cause functional changes, however the remaining SNPs were found as the decreased functional activity. The allele frequency of P283L among Korean population, Vietnamese, Chinnese and Caucasians were similar. However, the allele frequency of P341L between Korean population and Caucasian was significant different [12].

This recent study was inconsistent with the previous study in Caucasian cancer patients, which found the haplotype of similar SNPs involved and phenotyped the haplotypes as active function allele and decrease functional alleles. The authors also found the association among the phenotypes 5-HTRA drug response [9]. The small number of heterozygous in Met420del which is found in this current study should be replicated in the next further study with the larger sample size. Therefore, the next further study is warranted to find the new nonsynonymous variants in Indonesian subjects. The decrease functional variants found in the previous studies in Korean population, Japannese and Chinese were proposed to explore in Indonesian cancer patients.

CONCLUSION

The further study is warranted to do the sequencing analysis which could find the most possible variants in Indonesian cancer patients. To consider the similar SNPs involved in previous studies of Japannese, Chinnese and Korean population would be a challenge for the pharmacogenetic studies in Indonesian population.

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Conflict of interest

The authors did not have conflict of interest.

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