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A fast SSP-PCR method for genotyping the ATP-binding cassette subfamily B member 1 gene C3435T and G2677T polymorphisms in Chinese transplant recipients

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ABSTRACT

Aim. P-glycoprotein, the product of the ATP-binding cassette subfamily B member 1 (ABCB1) gene (or the so-called multidrug resistance 1 gene), is an ATP-driven efflux pump contributing to the pharmacokinetics as well as the pharmacokinetics of drugs that are P-glycoprotein substrates, such as tacrolimus. This paper describes the development of a new method for detection of the 3435C/T and 2677G/T/A single nucleotide polymorphisms of the ABCB1 gene. The method is a simple sequence-specific primer polymerase chain reaction (SSP-PCR).

Methods. 158 Chinese health checkup examinees and 214 transplant recipients were included in the study. Genomic DNA was extracted from peripheral blood and amplified with SSP-PCR to detect the 3435C/T and 2677G/T/A mutations in ABCB1. The SSP-PCR condition was optimized, and the PCR results were compared with those of DNA sequencing.

Results. In the optimized condition, the two polymorphisms could be clearly distinguished after one-step PCR and electrophoresis. The ABCB1 3435C/T and 2677G/T/A genotypes of the subjects were scanned, and allele-specific bands were successfully amplified by SSP-PCR, which were in full accordance with the results of sequencing.

Conclusion. As a fast, simple and inexpensive genotyping tool, the method would be practicable in large clinical studies on interindividual pharmacokinetics.

Key words: ATP-binding cassette subfamily B member 1 (ABCB1), single nucleotide polymorphisms (SNPs), sequence-specific primer polymerase chain reaction (SSP-PCR).

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