

IDENTIFICATION OF POLYMORPHISM

G2677T/A del GENE MDR1

AMPLI-set-G2677T MDR1 RT

Cat. n.2.006RT

The ABCB1 gene, also known as multi-resistance gene (MDR1), is the best characterized molecular transport through its ability to confer a MDR phenotype to cancer cells that have developed resistance to drugs.

The MDR1 gene encodes P-glycoprotein (PgP), which plays an important role in the development of resistance to many anticancer chemotherapy in precluding the accumulation of neoplastic cells in cases of overexpression. More generally, the PGP is responsible for biliary excretion of many drugs and renal function and may modify the intestinal absorption or passage in the CNS. These protein structures seem to be some sort of intelligent gate through which the cells of the human body can regulate the entry or exit from the cytoplasm of specific substances. These factors may not only be indispensable for the life of the cell for several reasons not cross the cell membrane (hydrophobic substances and macromolecules), but the same products of cell metabolism which if not removed soon could undermine the vitality of corresponding cell and organ function. Two polymorphisms frequently found in the Caucasian population are located in exons 21 (2677G → T) and 26 (3435C → T). G2677T polymorphism in exon 21 is responsible for the amino acid substitution Ala893Ser, C3435T polymorphism in exon 26 causes a silent mutation, responsible for the alteration of protein expression levels of MDR1. These polymorphisms are associated with alterations in the pharmacokinetics of several drugs including digoxin, and many anticancer chemotherapy.

The kit allows the identification of the MDR1 gene G2677T polymorphism, using the technique of Real-time PCR. The research of this polymorphism is performed after amplification with specific primers and hybridization with a probe that recognizes an internal sequence. The kit used for detection of the G2677T polymorphism, the probe that recognizes the sequence wt (G allele) is conjugated to the FAM reporters, while recognizing the polymorphic sequence (allele T) is conjugated to a reporter Joe.

Principle of the method: A) extraction of genomic DNA
B) amplification and detection using real-time PCR equipment.

Applicability: On genomic DNA extracted and purified from whole blood samples.

Number of Tests: 24.

KIT CONTENTS AND STORAGE

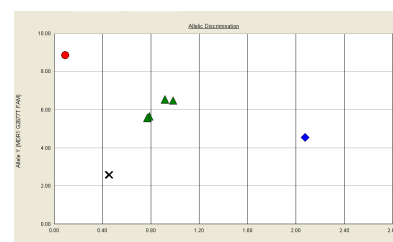
AMPLIFICATION	
PCR mix 2X	+4°C
H ₂ O sterile	-20°C
Primer-probe mix 20X	-20°C
Wild type control (GG)	-20°C
Mutated homozygous control (TT)	-20°C
Heterozygous control	-20°C

Stability: more than 18 months if properly stored.

ANALYSIS OF THE RESULTS

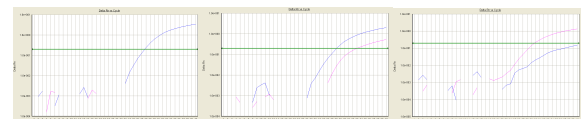
The analysis of the results will be made by the specific program (ALLELIC DISCRIMINATION) Real-time PCR instrument previously set. In any case, however, is also useful to analyze the graphs of Amplification PLOT, to ensure that the reaction has taken place correctly

Allelic discrimination MDR1 G2677T



Legend:
Red: Allele WT (G)
Green: Alleli G e T
Blu: Allele MUT (T)

Amplification plots MDR1 G2677T



Legend: Blu: Allele WT (G) Purple: Allele Mut (T)

References

- The Journal of Biological Chemistry (1990) 265 (1), 506-514.
- Genetics and Molecular Research (2010) 9 (1), 34-40.
- AAPS PharmSci (2002) 4 (4), article 29.
- The Journal of Pharmacology and Experimental Therapeutics (2001) 297:1137-1143.