CERTIFIED COMPANY
UNI EN ISO 9001, UNI EN ISO 13485

DETECTION OF -31delC POLYMORPHISM IN CDA GENE (CITIDINE DEAMINASI)

AMPLI-CDA -31delC Cat. n.2.019RT

Capecitabine is a 5-fluorouracil (5-FU) prodrug widely used in gastro-intestinal, head-neck and breast cancer treatment. The activation from prodrug to active molecule occurs through a 3 steps enzymatic process). a) Capecitabine is metabolized by carbossilesterase-2 (CES2), b) Capecitabine is metabolized by CDD (citidine deaminase), c) timidine phosphorilase (TP) converts the molecule in 5-FU. 5-FU inhibits the timilate sintetase (TS) and finally it is catabolized by diidrossipirimidine deidrogenase.
The Hand-Foot syndrome (HFS), which shows the desquamation of keratinized areas of hands and feet, it is one of the most important adverse event restricting the use of Capecitabine to the interruption of treatment and it is present in 30% of treated patients.
A study valued the correlation between HFS induced by Capecitabine and the presence of polymorphisms in gene involved in its metabolism. The 41% of patients developed a grade 3 HFS and they showed a significative association with 451C>T polymorphism in CDA gene. Particulary, T allele was associated to an increased development of HFS. The association between 451C>T polymorphism and mRNA expression of CDA in an experiment led on EBV lymphoblastoid cell lines was not demonstrated. Other genetic variants of CDA promoter region have been investigated. A polymorphism, -31delC (rs3215400), associated to HFS and expression of CDA, has been discovered. Carriers of C allele have a lower risk of development grade 3 HFS, compared to homozygous patients. The deleted allele leads to the deletion of a transcriptional site E2F. CDA genotype showing -31delC polymorphism shows a significative association with increased risk to develop HFS due to Capecitabine treatment.
The kit allows the detection of -31delC (rs 3215400) of CDA gene using Real Time PCR technique. An amplification with specific primers and hybridization with a probe recognizing an internal sequence allows to detect the -31delC polymorphism. The probe is linked to two different fluorophores (reporter dye and quencer dye). The relapse of the quencher causes an increase of reporter fluorescence directly proportional to the yield of PCR products (Real Time quantitation PCR).
The probe recognizing Allele C insertion is conjugated to FAM reporter, whereas the probe recognizing Allele C deletion is conjugated to VIC/Joe reporter.

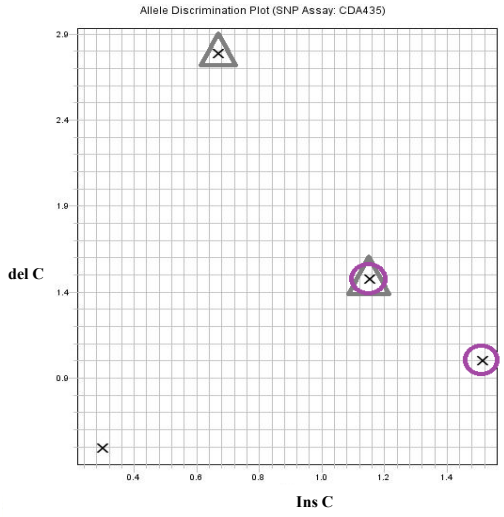
Principle of the method: a) extraction of genomic DNA;
b) amplification and detection using real-time PCR equipment;
Applicability: of genomic DNA extracted and purified from whole blood samples.
Number of Tests: 24.

KIT CONTAINS AND STORAGE

AMPLIFICATION	
Mix PCR 2X	-20°C
Mix Primers-Probes 20X CDA -31delC	-20°C
CDA ins C WT Control	+ 4°C
CDA ins C/ -31del C Heterozigous control	+ 4°C
CDA -31del C Homozigous control	+ 4°C
H_2O sterile RNase/DNase FREE	-20°C

Stability: more than 18 months if properly stored.

ANALYSIS OF RESULTS



Purple: ins C sample
Grey-Purple: insC/delC heterozigous sample
Grey: del C homozigous sample

References:
Clin. Cancer Res. 17, 2006-13, 2011
Curr Drug Metab 9,336-343, 2008
Hum Genet 119, 276-283, 2006