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DIAGNOSTICI E TECNOLOGIE BIOMEDICHE
BIOLOGIA MOLECOLARE

UNI EN ISO 9001
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DETECTION OF c-Kit EXON 11 MUTATIONS

AMPLI-c-KIT 11 Cat. n. 1425

Gastrointestinal stromal tumors (GISTs) are the most common mesenchymal tumors of the gastrointestinal tract. GISTs account for approximately 2% of all stomach tumors, 14% of all small intestine tumors, and 0.1% of colon tumors. Over 85% of GISTs express the KIT receptor (stem cell factor receptor, CD 117). KIT activation initiates a variety of intracellular signaling cascades that are involved in the regulation of proliferation, apoptosis and cellular adhesion. The majority GIST tumors contain a gain-of-function mutation in the *kit* protooncogene, leading to ligand-independent constitutive activation of the KIT receptor. Somatic mutations that result in constitutive activation of KIT receptor have been reported in a large number of GIST (50-95%). Mutations in the *kit* gene are most frequent in exon 11 and are less common in exons 9, 13 and 17. The presence of these mutations is related to a more aggressive tumoral phenotype.
Moreover, it has been recently discovered that a small subset of GISTs, wild-type for *kit* gene mutations, presents activating mutations in PDGF receptor gene (PDGFR).
The kit allows the detection of mutations in exon 11 of the *kit* gene. The detection of the mutations is carried out performing the amplification of the genomic region of interest with specific primers followed by the sequencing analysis. The characterization of the mutations of the kit gene represents an important prognostic and therapeutical factor as the activated KIT receptor is the target of the most effective therapy against GISTs.
Imatinib (Gleevec in USA; Glivec in Europa) is a strong competitive inhibitor of activated proteins as KIT, BCR-ABL and PDGFR α e β.

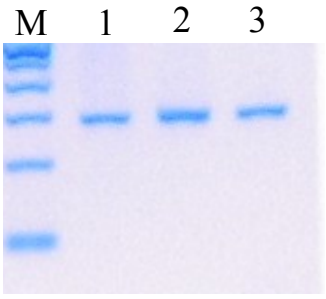
Principle of assay:
A) extraction of genomic DNA
B) amplification
C) detection on agarose gel
D) DNA sequencing.
Applicability: On extracted and purified genomic DNA from whole blood samples.
Tests: 50

REAGENTS AND STORAGE

AMPLIFICATION	
PCR mix exon 11	-20°C
H_2O sterile	-20°C
Taq Polymerase (5U/□l)	-20°C

Stability: over 18 months if correctly stored.

ANALYSIS OF RESULTS



1, 2, 3: exon 11 PCR fragment

The yield of amplification is a fragment of 303 bp. The sequence analysis reveals the presence or the absence of the mutations.

References:
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Cancer Research 64, 5913-5919 (2004).
Science 279 (1998) 577-580.
Cancer Control (2005) 12, 44-56.