

Rapid and specific screening assay for KRAS oncogene mutation by novel gene amplification method

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OBJECTIVE: KRAS mutation attenuates treatment effect of cetuximab. The purpose of this study was to develop a rapid and specific method to detect KRAS mutation at codon 12 using novel gene amplification methods.

STUDY DESIGN: Previously, loop-mediated isothermal amplification (LAMP) assay was developed to amplify copies of particular DNA sequence. LAMP assay is rapid and specific method rather than polymerase chain reaction process. We modified LAMP assay by using peptide nucleic acid (PNA) to suppress an amplification of wild-type allele.

RESULTS: We successfully developed modified LAMP assay in several cultured cell lines. By using this assay, we can amplify DNA fragment of mutated KRAS gene within 20 minutes. Primers bracketing the nucleotides of KRAS codon 12 were designed so that all possible alleles would be amplified by this assay.

CONCLUSIONS: The modified LAMP assay is a rapid, and highly sensitive detection assay for cancer mutations.