应用新型荧光定量PCR技术检测FGFR2基因SNP

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摘　要：目的：探讨一种目前较为新颖的通过Rotor Gene-3000荧光定量PCR仪进行Tm-shifting荧光定量PCR扩增测定单核苷酸多态性（SNP）的可行性。方法：以913份乳腺肿物患者成纤维细胞生长因子受体2(FGFR2)基因rs2981582位点（T→C）作为研究对象，用荧光染料SYBR Green I 标记DNA，并在设计引物时在所研究基因的特异性引物5＇末端连接上不同长度的尾，使不同基因型标本的PCR融解曲线的高峰出现在不同位置，最终可通过对PCR融解曲线的分析进行SNP分型测定。结果：纯合子C/C基因型融解曲线峰出现在≤84.60C处，特殊情况下稍向右偏移，但不超过85.10C，峰型较尖。纯合子T/T基因型融解曲线峰出现在≥87.50C处，特殊情况下稍向左偏移，但不小于870C，峰型较尖。杂合子T/C基因型融解曲线峰出现在前两者之间，峰型较平。 结论：实验结果表明，用本方法检测大批量人群标本的SNP结果符合遗传平衡定律，且操作简便，检测耗时短，结果特异且费用较为低廉，适合于进行大规模样品的SNP快速测定。

关键词：荧光定量PCR,单核苷酸多态性,FGFP2基因,乳腺癌

The determine method of FGFR2 gene SNP based on fluorescent quantitative PCR

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Abstract：Objective To study the feasibility of a novel single nucleotide polymorphism(SNP) typing method based on Tm-shifting fluorescence quantitative PCR . Method The site of rs2981582(T→C) in FGFR2 gene in patients with breast cancer was selected and its DNA were marked by using fluorescent dye SYBR Green I, moreover, it was connected with different length tails at 5＇-end of the specific genotypes primers. The peaks of different genotypes in melt curve would appear at its respective sites, so we could distinguish the SNP by the melt curve . Result In melt curve, the peak of C/C genotype appeared at the point less than or equal to 84.6℃, under special circumstances, it would slightly move to right, but not exceeding 85.1℃, it's a sharp peak. The peak of T/T genotype appeared at the point more than 87.5℃, under special circumstances, it would slightly move to left, but not less than 87℃, it's also a sharp peak. The peak of T/C genotype appeared between the two peaks above-mentioned, it's a smooth peak. Conclusion The results showed that the method was accord with heredity regular and the method was specific, easy to operate, low-expensive and suitable for SNP typing detection in large scale samples.

Key words：real-time fluorescent quantitative polymerase chain reaction,SNP,FGFP2,breast cancer