W1-1

Pyrosequencing: Principle and application

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There is a growing demand for characterization of naturally occurring variations, particularly Single Nucleotide Polymorphism (SNP) in the human genome. Pyrosequencing, a real-time pyrophosphate detection technology, is a novel sequencing approach for high-throughput analysis of SNP. This method is based on indirect lumonometric quantification of the pyrophosphate that is released as a result of nucleotide incorporation onto an amplified template. The SNP analysis can efficiently be performed by Pyrosequecing using a highly automated sequencing instrument for 96 samples within 10 to 20 minutes. The resulting pyrogram data are evaluated by comparison of peak heights, which are proportional to the number of incorporated nucleotides. In addition to the SNP genotyping, this system can be used for the mutation detection, deletion/insertion analysis, sequencing of the short DNA fragments, bacterial typing, viral typing, forensic analysis, etc.

W1-2

Methods for Generating siRNA

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Many researchers are using small interfering RNAs (siRNAs) to reduce the expression of specific mammalian genes. siRNAs are short, double stranded RNA molecules that can target mRNA of a specific sequence for degradation via the RNA interference pathway. Applications of siRNAs include functional genomics, drug target validation, and cell signaling pathway analysis. Currently there are five methods for generating siRNAs for gene silencing studies. Each of these methods has its its advantages and drawbacks. The best method for generating siRNAs depend on the goals of the experiment.