Heteroduplex Analysis Encyclopedia Article

Heteroduplex Analysis

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Heteroduplex Analysis

Heteroduplex analysis (HA) is a method of detecting sequence differences between a reference **DNA** and a test DNA (both usually produced by **polymerase chain reaction** amplification). It is commonly used to detect mutations in a **gene**, and is much simpler and cheaper than the related technique **denaturing high performance liquid chromatography** (DHPLC). It has been used to screen many disease genes in humans, including the breast **cancer** genes BRCA1 and BRCA2, the tuberous sclerosis genes TSC1 and TSC2, and FBN1, the gene that causes Marfan syndrome. It is also used to screen non-human DNA for mutations, such as the DNA from the human immunodeficiency **virus** (HIV).

The method requires test DNA (usually PCR-amplified DNA from a person who is thought to have a **mutation** in the gene of interest), and reference DNA (the same PCR-amplified fragment from a normal individual). The reference and test DNA are mixed, then heated to melt (denature) the duplex then cooled to reanneal the duplex. This allows heteroduplexes to form. These are DNA duplexes where one strand is from the reference DNA and the other strand is from the test DNA. If there is a difference between the two DNA sequences, then the two **bases** at that point cannot base pair--a DNA mismatch (DNA heteroduplex) is formed. The mixture is then subjected to electroporesis in a polyacrylamide gel. Any heteroduplex migrates more slowly through the gel as compared to its homoduplex counterparts, and so any sequence change can be detected by an extra band above the main DNA homoduplex band. Like most other DNA mutation detection techniques, the DNA must be sequenced to determine the exact position and type of **nucleotide** change.

The method has recently been adapted so that analysis is performed by chromatography or minuture electrophoresis on a `chip' instead of acrylamide gels. This will allow more samples to be screened in a given amount of time, and helps HA to be another method for high--throughput screening of patients by medical geneticists.