



PCR: QUESTIONS & ANSWERS

Q. What is Polymerase Chain Reaction (PCR), Reverse Transcription-PCR (RT-PCR), Real-Time PCR?

A. PCR is a Nobel Prize Winning Technology. It was used to map the human genome and was the first FDA approved Molecular Technology. It utilized a 3-Step Process:

1. Denaturation – separating the DNA double strands
2. Annealing – binding or reconnecting 2-DNA segments
3. Extension – replicating or extending copies of the DNA sequence

RT-PCR is a variation of PCR designed for RNA applications. Real-Time PCR is a faster (PCR or RT-PCR) methodology

Q. What does PCR, Reverse Transcription-PCR (RT-PCR), Real-Time PCR do?

A. PCR is an extremely sensitive and specific method of identifying DNA from many different sample types. RT-PCR is a method of making complementary DNA from RNA. Real-Time PCR (can be PCR or RT-PCR) is a faster methodology that combines the amplification and detection steps used in PCR and RT-PCR, and provides very broad dynamic ranges for viral load assays.

Q. How is PCR used?

A. All PCR methods (PCR, Reverse Transcription-PCR & Real-Time PCR) are used to diagnose the presence of infectious agents, genetic mutations, or any specific sequence of base pairs.

Q. What is the difference between qualitative and quantitative test?

A. Qualitative tests provide “detected”/“not detected” (positive/negative) answers and are usually used to determine the presence or absence of a DNA Target. Quantitative tests provide the actual number of DNA targets present in a sample and are used for viral load tests.

Q. What is the difference in sensitivities between qualitative and quantitative assays?

A. In the past qualitative tests have been more sensitive than quantitative tests, but with the introduction of the TaqMan assays (Real-Time PCR) the quantitative tests have potential to be more sensitive than the qualitative tests.

Q. What is the difference between PCR testing and Antibody testing and how to interpret differences?

A. PCR tests detect and/or measure actual infectious agents (ie. bacteria or viruses). Antibody (Ab) tests simply monitor the body's response to an infectious agent. An Antibody test cannot tell you if a person is still infected, only that they have been exposed at some point in time.

