3.4 How can we study the effect of viruses on cell pathways?

Welcome to a new video on viral diagnosis. Surely you know that when a virus infects a cell, it produces a series of molecular imbalances, which make that the cell synthesizes molecules in response to infection, either as defence or to inform other cells about the infection. In this video we'll talk about microarrays, a technique used to know which cell routes are affected by the viral infection.

Microarrays or microchips or DNA chips, that they are all synonymous, have many applications in research, for example, to develop drugs, to study the expression of genes related with the viral pathogenicity, etc. But there are other microchips that can be used in diagnosis, to identify new viruses, to monitor patients treated with antiviral agents, or to control the quality of vaccines and confirm that they are exempt of contaminating micro-organisms. So, as you can see, the uses are innumerable. By the way, microarrays designed to diagnose many viruses are called virochips.

They consist of a membrane that can be nylon, silicone or a simple glass slide. The DNA of many genes is sorted according to a specific pattern and immobilized in this membrane. These fragments of DNA, known as probes or oligomers, are arranged in dots or microscopic spots. When a virus infects a cell, a series of signals are triggered. They reach the nucleus and activate specific genes. The result is the synthesis of mRNA that goes into the cytoplasm. Well, this mRNA (or after reverse transcription into complementary DNA) hybridizes with the DNA attached to the solid matrix.

Two-channel microarrays

The typical technique involves the extraction of the mRNA from infected and non-infected cells. With the enzyme RT that we saw in previous videos, the mRNA is reverse transcribed into cDNA, using different colour-labelled dNTPs for the infected cells and for the non-infected cells, so that they are marked with different fluorescence colour. When these cDNAs are added to the matrix with thousands of DNA sequences, in the right conditions, if there is this complementarity between the probe of the microarray and the cDNA, they both hybridise. As they have different colour, we can be determine with which probes cDNA from infected cells and from non-infected hybridizes, and thus determine the expression of genes which triggered the viral infection. Don't forget that in the microchip there are thousands of probes, so in the same experiment we can see the expression of many different genes, which are identified by their position on the chip. Although it may seem complicated, microarrays can be customized depending on the purpose for which they will be used. And don't worry, that they are not manually read: there are microarray readers and specialized programs to interpret the fluorescent signals.

One-channel microarrays

A single fluorochrome colour can also be used especially if we want to quantify the amount of mRNA because thus we will see absolute values, which we will compare with the intensity of fluorescence that provides DNA diluted at known concentrations. In this way, the results can be compared between different laboratories or obtained at different times.

In this video we have seen an introduction to microarrays a very powerful technology, with many applications. I am sure that you have many questions. There is a video in English in the additional material that will solve many of them.

In addition, as this ends this section of diagnosis through nucleic acids, don't forget to do the corresponding exercises.

Thank you very much for your attention.