2.15 DNA microarray technology

Although all of the cells in the human body contain identical genetic material, but the same genes are not active in every cell. Studying which genes are active and which are inactive in different cell types helps scientists to understand both how these cells function normally and how they are affected when various genes do not perform properly. In the past, scientists have been able to conduct these genetic analyses on a few genes at once. With the development of DNA microarray technology, however, scientists can now examine how thousands of genes express and their product interact at any given time.

A **DNA microarray** (also commonly known as *gene chip*, *DNA chip*, or *gene array*) is a collection of microscopic DNA spots attached to a solid surface, such as glass, or silicon chip forming an array for the purpose of expression profiling, monitoring expression levels for thousands of genes simultaneously.

An *array* is an orderly arrangement of samples. In general, arrays are described as *macroarrays* or *microarrays*, the difference being the size of the sample spots. Macroarrays contain sample spot sizes of about 300 microns or larger and can be easily imaged by existing gel and blot scanners. The sample spot sizes in microarray are typically less than 200 microns in diameter and these arrays usually contain thousands of spots.

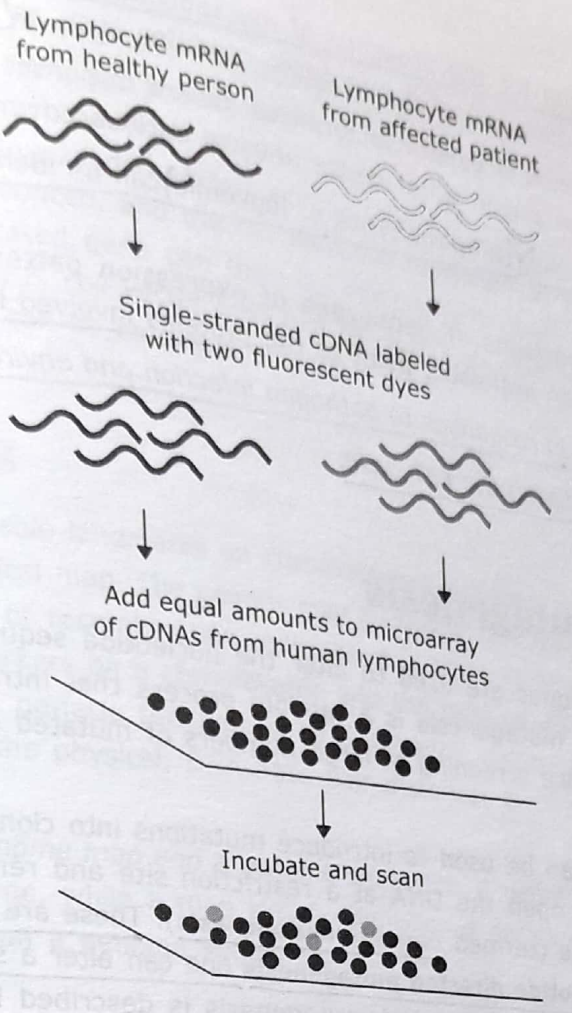

Figure 2.21 : DNA microarray.

A large number of DNA probe, each one with a different sequence, are immobilized at defined positions on a solid surface. The probe can be *synthetic oligonucleotides* (20–25 nt long), short DNA molecules, such as cDNA or PCR products. These can be spotted onto a glass microscope slide or a piece of nylon membrane (*low density arrays*) or on the surface of a wafer of silicon (*high density array*).

Base-pairing (i.e., A-T and G-C for DNA; A-U and G-C for RNA) or hybridization is the underlining principle of DNA microarray. Microarray technology evolved from Southern blotting, whereby fragmented DNA is attached to a substrate and then probed with a known gene or fragment. To determine which genes are turned on and which are turned off in a given cell, a researcher must first collect the messenger RNA molecules present in that cell. The researcher then labels each mRNA molecule by attaching a fluorescent dye. Next, the researcher places the labeled mRNA onto a DNA microarray slide. The messenger RNA that was present in the cell will then hybridize - or bind - to its complementary DNA on the microarray, leaving its fluorescent tag. A researcher must then use a special scanner to measure the fluorescent areas on the microarray.

If a particular gene is very active, it produces many molecules of messenger RNA, which hybridize to the DNA on the microarray and generate a very bright fluorescent area. Genes that are comparatively less active produce fewer mRNAs, which results in dimmer fluorescent spots. If there is no fluorescence, none of the messenger molecules have hybridized to the DNA, indicating that the gene is inactive. Researchers frequently use this technique to examine the activity of various genes at different times.

Principal applications of microarrays

- *Investigating cellular states and process.* Patterns of expression that change with cellular state or growth conditions can give clues to the mechanisms of processes such as sporulation, or the change from aerobic to anaerobic metabolism.

- *Diagnosis of disease.* Testing for the presence of mutation can confirm the diagnosis of a suspected genetic disease, including detection of a late-onset condition such as Huntington disease.
- *Drug selection.* Allows detection of genetic factors that govern responses to drugs, that in some patients render treatment ineffective and in other cause unusual serious adverse reactions.
- *Specialized diagnosis of disease.* Different types of leukemia can be identified from different patterns of gene expression.
- *Pathogen resistance.* Comparisons of genotypes of expression patterns, between bacterial strains susceptible and resistant to an antibiotic, point to the proteins involved in the mechanism of resistance.

- *Investigating cellular states in responses to pathogen infection and environmental change*
- *Investigating cellular states during the cell cycle.*