DNA Microarrays

Benefits and Drawbacks

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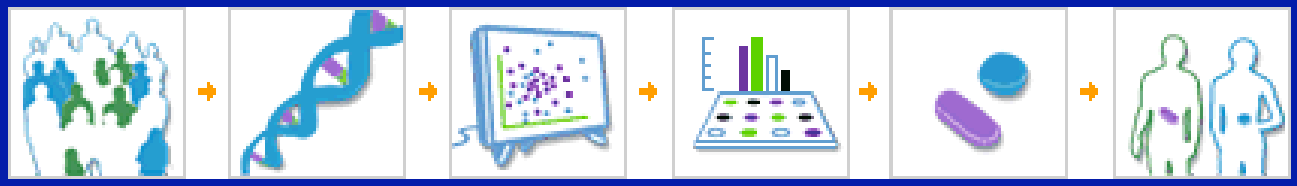
What is a DNA Microarray?

- A basic premise of drug research is that differences in gene expressions (which ones are active) will provide clues about where to look for drugs. DNA microarrays are a fundamental tool for doing such differential gene expression analysis

- Microarray is a sliver of glass or silicon robotically studded with two dimensional orderly arrangement of thousands of DNA spots. It provides a medium for matching known and unknown DNA samples based on base-pairing rules (hybridization) and automating the process of identifying the unknowns

Need for Microarrays

- Despite the recent flood of new biological data from the human genome sequencing, scientists are struggling to answer many basic questions
- There is a need to accelerate figuring out which genes are active, not by tackling one gene one experiment at a time, but implementing a massively parallel process
- Microarrays allow researchers information on thousands of genes simultaneously - a dramatic increase in thorough output: months to days

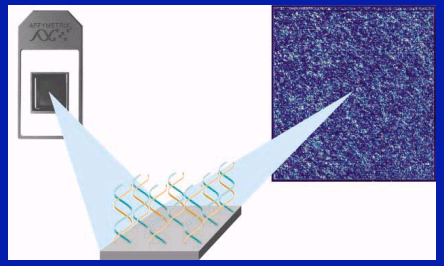


How does a DNA Microarray work?

Microarray works by exploiting the property of a given mRNA molecule to bind specifically to, or **hybridize** to, the DNA template from which it originated

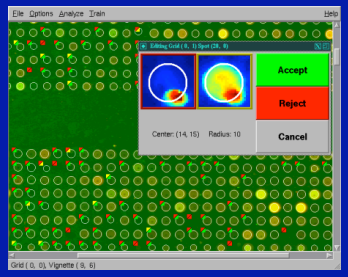
In a single experiment, using an array with many DNA samples from known genes, we can determine the expression levels of hundreds or thousands of genes within a cell by measuring the amount of mRNA bound to each site on the array

The amount of mRNA, fluorescently labeled, bound to the spots on the microarray is precisely measured using a computer, generating a profile of gene expression in the cell



DNA MicroArray

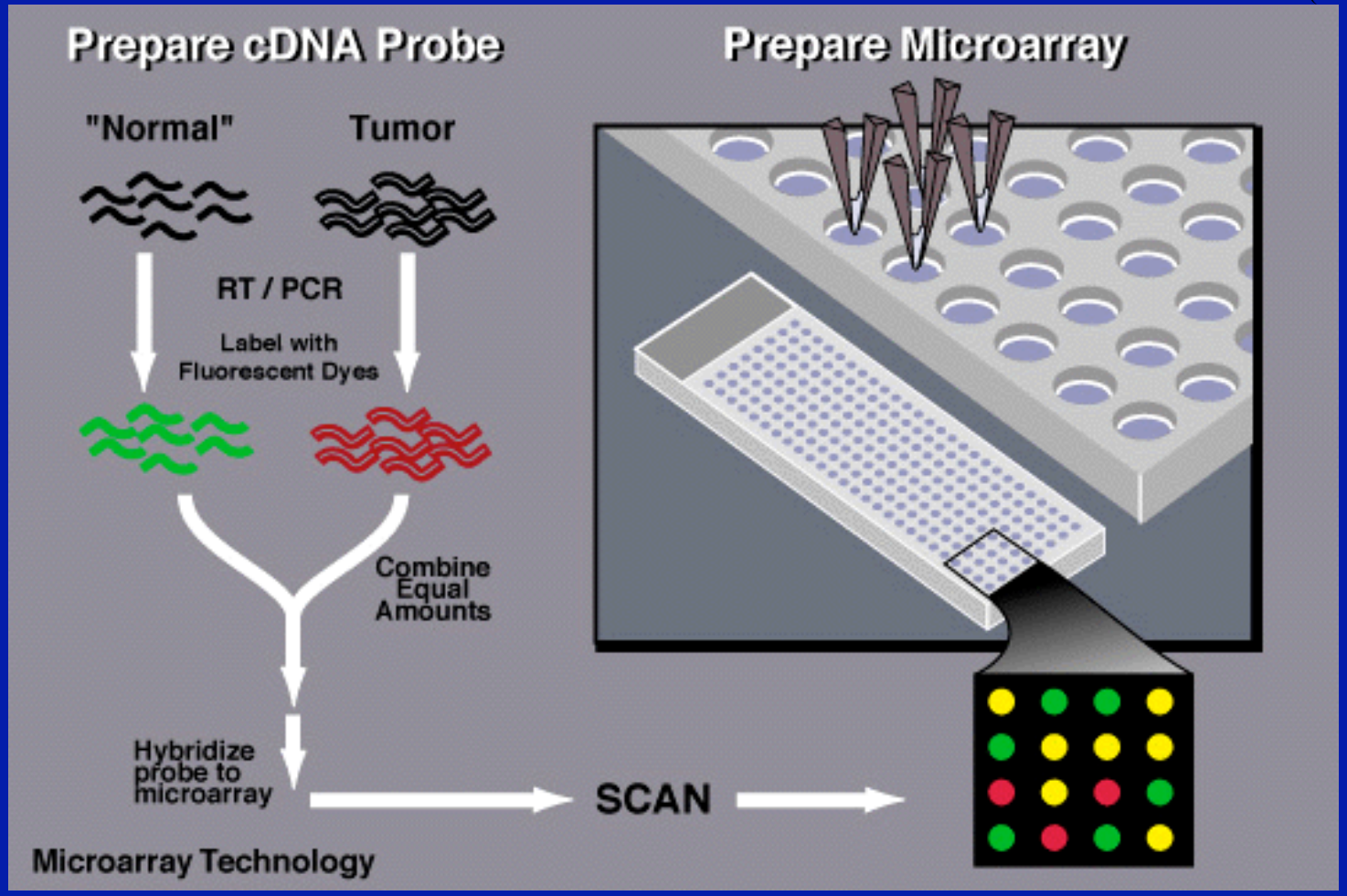
MicroArray Analyzer



Profile

Hybridization is the Key

Source: National Human Genome Research Institute (NHGRI)



Analyzing the location and intensity of fluorescent signals, one can determine the level of activity of each gene

Types of DNA Microarrays

- cDNA Microarrays
  - each spot contains a cDNA clone from a known gene (PCR product)
  - better matching results because of longer length cDNA
  - initially developed at Stanford University

- Oligo(nucleotide) Microarrays
  - made by prefabricated or synthesizing single stranded oligo from known database
  - Since oligos are usually short, the density of these arrays is much higher
  - Traditionally called GeneChips by commercial suppliers, such as Affymetrix, Agilent

Benefits of DNA Microarrays

- Gene Discovery
- Gene Expression
- Disease Diagnosis
- Drug Discovery and toxicology

Drawbacks of MicroArrays

- Lack of standardization
- Inadequate computer based tools
- Statistical problems
- Insufficient quality
- Insufficient sensitivity
- Lack of reproducibility
- Variability of outcome
- Fidelity of gene expression data
- Effects of probe length
- Cost

Lack of Standardization

- Data collected from different microarray platforms can't be accurately compared
- Absence of a unified "language" for exchange of microarray data between different groups
- Microarray Gene Expression Data Society, MGED, has developed guidelines for the publication of DNA microarray data called MIAME, minimal information
- Microarray Markup Language, developed by MGED, is an attempt to provide a standard platform for submitting and analyzing the microarray expression data generated by different laboratories around the world

Inadequate Computer Tools

- Interpreting microarray experiments is very taxing – too much, and too little information
- Lack of turnkey bioinformatics models and tools inhibit quick and efficient analysis of the large data sets created within each experiments
- Statistical problems ranging from image analysis to pattern discovery and classification exist.
- Efforts are being made to improve standards of experimental design, data presentation and analysis – advance bioinformatics

Insufficient Quality, Sensitivity and Reproducibility

- Ways to characterize the quality of microarrays to ensure full functionality are lacking
- Poor sensitivity and background noise forces amplification which may not give the researcher a true picture
- Array to array coefficients of variation is high lowering the reproducibility of microarray results
- Hybridization process still involves a great deal of manual handling. Microfluidics combined with microarrays has the promise of enhancing ease of use and reproducibility of results

Fidelity of Gene Expression Data

- Some researchers have shown that for complex diseases a considerable discrepancy exists between the differentially expressed genes identified on oligo arrays arrays and those identified on cDNA microarrays
- Separation of genes causally involved in a disease from innocent bystander genes whose expression levels have been secondarily altered by primary changes elsewhere is a challenge
- In addition, different bioinformatic protocols applied to the same microarray data yield quite different gene sets making clinical decisions difficult

Cost

- Initial cost of designing and fabricating custom microarrays and analyzer is high but falling; commercial DNA arrays retail for a few hundreds dollars
- Replication of experiments to minimize statistical variability of results can be cost prohibitive
- Use of fabrication processes and procedures from the semiconductor industry are helping bring the cost down

Summary

- DNA microarrays give researchers the opportunity to analyze vast amounts of genetic information in a single experiment.
- The use of DNA microarrays holds great promise in our understanding of genes and their impact on disease, drug discovery and development
- DNA microarrays like any other emerging technology has shortcoming and drawbacks that have been identified and addressed by the biotechnology community

References

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