Comparison of the Use of Plasmid and PCR DNA on Microarray Chips

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Abstract

Using microarrays for high-throughput DNA detection is one of the fastest growing fields in biology today. However, as of yet there is no universal set of rules that researchers follow in designing experiments and determining results. Because of this there may be avenues of research that have not been utilized.

One of the methods that have not been reported is the use of probing for genes previously inserted within a plasmid. In previous experiments that utilized DNA detection with microarrays, the appropriate DNA has been extracted and amplified before being spotted on a microarray chip. This experiment is designed to test the effectiveness of spotting and probing plasmid DNA and comparing the result to spots of PCR DNA.

A favorable result would indicate a new path that researchers could take when preparing microarray chips. In certain experiments, the step of PCR could be removed, creating much faster, cheaper, and easier means to determine the result.

Preparation of Plasmid DNA

As is evident in this gel electrophoresis, the insert is very clean; when it is present, there is no smearing or question about whether the gene is or is not in the plasmid.

Preparation of PCR DNA

After the yeast genes were cloned into E. coli, the DNA was isolated and amplified using Polymerase Chain Reaction. Although PCR is a very acceptable method for obtaining large quantities of DNA, an increase in steps increases the chance that mistakes could be made. Note the smearing and uncertainty in interpretation of the electrophoresis gel at right.

Methods: Oligonucleotide Addition

The results were mixed, with some of the six microarray slides showing strong oligonucleotide binding to the PCR product . . .

Results

Plasmid . . . and other slides showing strong binding to the spotted plasmid.

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