

## BOX 2 GOING WITH THE FLOW

DNA arrays are traditionally printed on glass or glass-like slides, hybridized to a fluorescently labeled target, and the slide is read by a scanner that can detect multiple fluorescent signals. An alternative method for making arrays is the Continuous Flow Microspotter developed by scientists at the University of Utah and sold by Wasatch Microfluidics. It uses a parallel network of microchannels to cycle small volumes of fluid samples over a surface, binding the samples to the surface in an array of spots. The spots are sealed at the surface so they are physically protected from one another. "You can start with a very dilute solution of the material you want to spot on the array and run it back and forth on the surface until you have the concentration you want," says Bruce K. Gale of the University of Utah. "Variation between spots is less than 5%," he adds. One of the company's arrays, which became available last month, can hold 48 spots. "The technology works especially well for proteins that don't bind well to surfaces and denature when they dry out. The device keeps them wet until they are ready to use," says Gale. The platform has been used for spotting proteins, DNA, cells and lipids by changing the coating on the slides.

MetriGenix has also developed a microarray device based on a three-dimensional matrix of microchannels. Similarly to Biacore's platform, MetriGenix Flow-Through Chip allows molecular interactions between probes and targets to occur

in flow. Microchannels connect the upper and lower faces of the chip. As fluids containing different samples pass through these channels, binding reagents attach to the walls of channels, and target molecules are then captured in a fully automated process. Multiple spots are deposited in a regular grid to permit parallel analysis of up to 400 spots per sample. Because the binding reagents cover a wider surface area than they would on more traditional two-dimensional microarrays, binding occurs faster, resulting in shorter assay times. The technology has been used for protein arrays and pathogen detection. "We sell array systems, which include the flow-through chip and all the necessary reagents for hybridization," says Michael L. Cohen, CEO for MetriGenix. "The researchers just have to add their sample."

At the detection end, CombiMatrix has developed an electrochemical detection method as an alternative to traditional fluorescence-based platforms. "We have developed a smaller less expensive detection system that can measure 12,000 features on a microarray using electrochemical signals," says Tognotti. "The whole machine is the size of a toaster and can be taken out into the field." According to Tognotti the assay is slightly more sensitive than fluorescence detection and does not produce any images. "The data comes straight out of the instrument as electronic signals. You can get very quick answers in 40 seconds," he explains.

adenoviruses, several subtypes of influenza A virus, 12 other common respiratory pathogens, and 6 US Centers for Disease Control and Prevention category A bioterrorism pathogens known to cause flu-like symptoms. Two years ago, Affymetrix started working on developing a microarray capable of detecting hundreds of different bacteria and viruses from the US National Institute of Allergy and Infectious Diseases high-priority pathogen list.

### Where transcription factors go

Gene expression analysis provides only a partial glimpse into how transcription is regulated. Additional information can be gained by determining the precise locations the binding sites for regulatory proteins on genomic DNA. Chromatin immunoprecipitation (ChIP), an assay used to detect interactions between proteins and DNA, has been combined with DNA microarray technology in a method dubbed ChIP-on-chip or location analysis. In this technique, a protein is cross-linked to DNA upon binding. The protein is then used as a tag to pull out the bound DNA by means of an antibody. The DNA is then eluted, labeled and hybridized to a genomic tiling array, which consists of

DNA fragments spaced at regular intervals across the entire genome. The DNA fragment that 'lights up' identifies the genomic position of the protein binding site. This type of array provides key information not only about transcriptional start sites, but also DNA replication, modification and repair.

NimbleGen was the first company to offer a microarray service for custom ChIP-on-chip assays. Additionally, NimbleGen offers "both *de novo* tiling and targeted arrays for specific regions of the genome," says Emile F. Nuwaysir, vice president for business

development. "Our platform is the only high-density long-oligonucleotide arrays for covering up to 40 megabases of genomic DNA per array. It gives a maximum number of data points."

One of the most recent validations of ChIP-on-chip was performed by Richard Young's group at the Whitehead Institute for Biotechnology. The study identified DNA segments bound by a master regulator of human embryonic stem cells differentiation<sup>3</sup>. In January 2005 Agilent Technologies purchased Computational Biology Corp.,



The Biacore T100 system. (Courtesy of Biacore.)