TOOLbox

RefDIC

To integrate transcriptomic and proteomic information on the immune system, Osamu Ohara and colleagues at the RIKEN Research Center for Allergy and Immunology and the Kazusa DNA Research Institute (both in Japan) developed a database called reference genomics database of immune cells (RefDIC). The database features quantitative mRNA profiles of human and mouse immune cells and tissues as well as quantitative protein profiles of mouse immune cells. A query interface allows researchers to visualize and analyze the data. For example, various inputs such as a gene name or probe set IDs can retrieve mRNA annotations and heat maps. Protein profiling data for multiple genes also can be visualized. Because the protein profiles are derived from 2DE analyses, users can obtain the apparent p*I*s and molecular masses and other information. In addition, the entire 2DE gel image can be retrieved. Finally, both mRNA and protein profiling data can be viewed simultaneously for a single gene to see whether the amounts correlate. RefDIC is accessible at http://refdic.rcai.riken.jp. (*Bioinformatics* **2007**, *23*, 2934–2941)

PHOSIDA

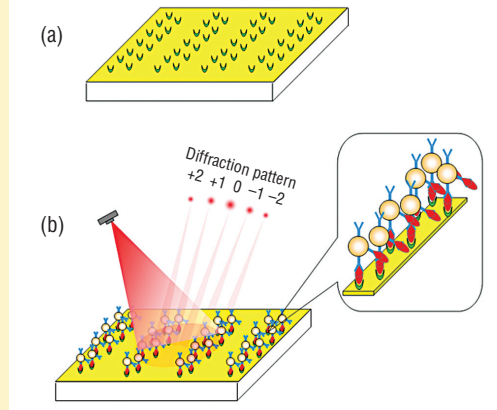
Matthias Mann and co-workers at the Max Planck Institute of Biochemistry have developed a phosphorylation site database called PHOSIDA. The database contains thousands of high-confidence phosphosites from large-scale proteomics studies. For many entries, quantitative and time-resolved data are available. The researchers studied the structures of phosphosites and found that, in agreement with previous reports, phosphosites are in highly accessible and flexible regions of proteins. With tools in PHOSIDA, phosphoproteins from many species can be aligned, and the color of each phosphosite indicates the degree of conservation. Although regions containing phosphosites generally were less conserved than other protein regions, Mann and co-workers determined that five amino acids on either side of a phosphosite were highly conserved. Finally, a support vector machine in PHOSIDA can predict phosphosites. (*Genome Biol.* **2007**, *8*, R250)

proach, alternate coatings of oppositely charged polyelectrolytes are placed onto memobeads. These coatings help position the beads for decoding without interfering with the bar-code reading process. In addition, the layers provide many sites for antibody attachments.

To characterize the LbL-coated memobeads, De Smedt and colleagues ran them through a battery of tests. Beads with antibodies against tumor necrosis factor (TNF-α) were incubated in various TNF-α solutions. The fluorescence of the beads was proportional to the TNF-α concentration and was stable for at least 20 days. When these beads were added to serum or a plasma–buffer mixture spiked with TNF-α, slightly lower sensitivities were observed. Next, two sets of beads with antibodies against TNF-α or P24 were added to a plasma–buffer mixture to test the multiplexing capabilities of the assay. TNF-α and P24 were specifically bound by the appropriate beads with no cross-reactivity. Finally, TNF-α and IL-2 were specifically detected in whole-blood samples spiked with TNF-α and IL-2. Unlike other immunoassays, the new test did not require any washes. The researchers say that the assay would be ideal as a microfluidic, point-of-care device because it is actually more sensitive when fewer beads are used. (*Anal. Chem.* **2008**, *80*, 80–89)

Immunomagnetic diffractometry for biomarker detection

In the quest for an improved biomarker detection method, Cagri Savran and colleagues at Purdue University and the Mayo Clinic have designed a new technique called immunomagnetic diffractometry. Their approach combines the immunomagnetic capture of an analyte on beads, in situ assembly of an optical diffraction grating, and measurement of the diffraction. In this method, magnetic beads capture the target from the serum sample and bind to a surface to form the diffraction gratings. By virtue of their size, the beads enhance the diffraction signal, so no further signal amplification or labeling is needed. The target chosen for the proof-of-principle study was the folate receptor (FR), a potential serum biomarker for cancer.



Immunomagnetic diffractometry FR assay. (a) Microcontact-printed F–BSA patterns; (b) FR–beads forming in situ diffraction grating. Laser illumination yields a characteristic diffraction pattern dependent on the density of the attached beads.

With microcontact printing, the researchers deposited alternating 15 μm lines of folate-coupled bovine serum albumin (F–BSA) onto a gold surface. Magnetic beads derivatized with the FR antibody (FR–beads) captured FR from serum and subsequently bound to the F–BSA lines. The bound molecules and beads effectively formed a grating that diffracted the incident laser radiation.

The researchers observed that the FR–beads attached specifically to the F–BSA on the surface. The packing density of the bound FR–beads intensified with increasing FR concentrations (700 fM to 11 nM). After creating a calibration curve, the researchers measured the FR concentration in the serum of cancer patients. The detection limit of immunomagnetic diffractometry was lower than that of several other biomarker assays such as ELISAs. Immunomagnetic diffractometry has the additional advantages of speed, robustness, low cost, and ease of miniaturization. The researchers say that the assay is applicable to other biomarkers. (*J. Am. Chem. Soc.* **2007**, *129*, 15,824–15,829)