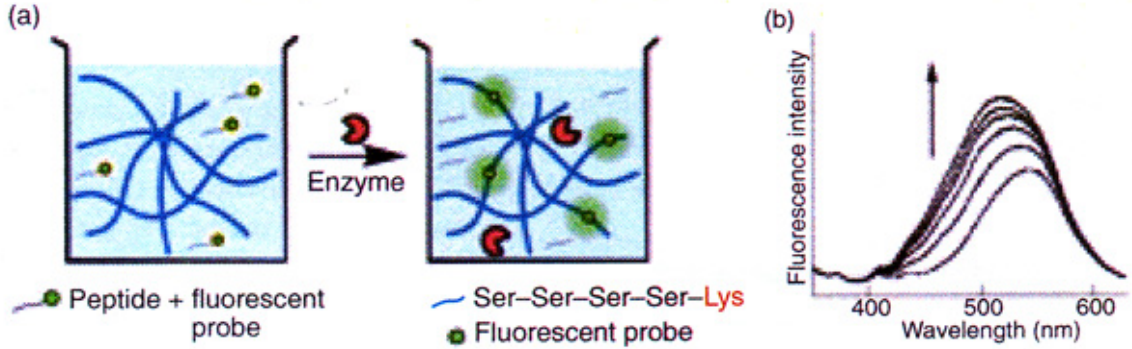
Protein microarrays made from hydrogel

Conventional microarrays are dry devices, but proteins function best in a wet environment. So Itaru Hamachi and colleagues at Kyushu University (Japan) chose the middle road and made semiwet protein microarrays using a supramolecular hydrogel.

The hydrogel uses a low-molecular-weight hydrogelator that works at the very low concentration of 0.1 wt%. The researchers estimate that 1 molecule of gelator immobilizes about 38,000 molecules of water. The proteins or peptides are trapped in semiwet aqueous cavities created in the gel matrix.

A handy feature of the hydrogel is that a strong blue shift occurs when hydrophobic probes are added. The researchers created a substrate from a hydrophilic peptide joined to a hydrophobic fluorescent probe. When lysyl endopeptidase (LEP) cleaved the substrate, the probe migrated from the aqueous cavities to the hydrophobic domains, where its fluorescence increased twofold and its emission shifted. The formerly pinkish-yellow hydrogel turned green.



Color-changing hydrogel. (a) An enzyme cleaves a substrate made from a hydrophilic peptide and a hydrophobic fluorescent probe, which induces (b) a color shift. (Adapted with permission. Copyright 2003 Macmillan Publishers Ltd.)

To make microarrays, the researchers spotted the hydrogel onto slides. Only spots injected with LEP turned green. Similar assays were conducted with a substrate cleaved by V8 protease and with another cleaved by either LEP or chymotrypsin. In addition, the researchers conducted an inhibitor assay, in which LEP was injected into the hydrogel first, followed by the candidate inhibitors, and finally the substrate. In this case, spots with poor inhibitors turned green.

The researchers note that unlike other protein- or peptide-based hydrogels, this one is made from a purely synthetic small molecule that exists as a single component, and gel formation is spontaneous. (*Nat. Mater.* **2003**, doi:10.1038/nmat1034)