

**Figure 1 | Models for partial agonism.** Agonists bind to receptors to produce an effect, such as the opening of an ion channel. **a**, The occupancy model proposes that agonist binding opens the channel immediately. But this model does not explain how a so-called partial agonist can bind to produce a complex in which the probability of channel opening is low. **b**, A two-step mechanism has therefore been proposed<sup>2</sup> in which an inert receptor–agonist complex forms first, after which the channel opens. Partial agonists were thought to be less effective than full agonists in the channel-opening step. **c**, Lape *et al.*<sup>1</sup> show that another step is required, in which the inert complex enters a state of readiness — the flip state — before channel opening. Partial agonists are less effective than full agonists at inducing the flip state, but bring about channel opening just as quickly.

functional states. Unfortunately, no complete high-resolution structures have been determined for these receptors in known functional states. The crystal structures of putative flip states will be even more difficult to obtain, as

such states are necessarily short-lived so that the protein can respond rapidly to transient changes in agonist concentration. Optical or resonance techniques (such as nuclear magnetic resonance) might fill this gap in the future.

The authors' insight<sup>1</sup> into the mechanistic basis of partial agonism will quell some angst among pharmacologists, but it also generates ideas about other consequences of the flip state. For example, it has been reported that mutations in the glycine receptors of mice reduce the ability of glycine to open the associated ion channels, because the receptor is less able to 'flip' after binding the agonist<sup>7</sup>. It is also known that some compounds enhance the effects of partial agonists, and can even sometimes convert them to full agonists. Perhaps these compounds affect the entry of the channel to the flip state. Similarly, certain inhibitors of agonists might work by reducing the stability of the flip state.

Lape *et al.*<sup>1</sup> have answered a long-standing question by showing that the flip state can explain partial agonism. Future research will reveal what other aspects of receptor function are shaped by the properties of this state. ■

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## MATERIALS SCIENCE

# Protein gels on the move

April M. Kloxin and Kristi S. Anseth

**Light-induced reactions enable three-dimensional objects to be built from simple compounds. Proteins have been added to the list of building blocks, and the resulting gels move in response to environmental cues.**

The ancient Egyptians were arguably among the first photochemists, because they used light-induced reactions as part of their mummification process. They wrapped bodies in linens that had been dipped in a substance called bitumen of Judea. On exposure to sunlight, the molecules in the bitumen reacted to form crosslinks with each other, yielding a hard, protective coating that preserved the mummy for centuries<sup>1</sup>. Variations of this crude polymerization process are still in use today. Latter-day scientists can choose from thousands of different light-reactive monomers, and use sophisticated laser techniques to turn them into polymers that have diverse applications — for forming printing plates for newspapers, for example, or for dental restoration.

Reporting in *Proceedings of the National Academy of Sciences*, Kaehr and Shear<sup>2</sup> describe

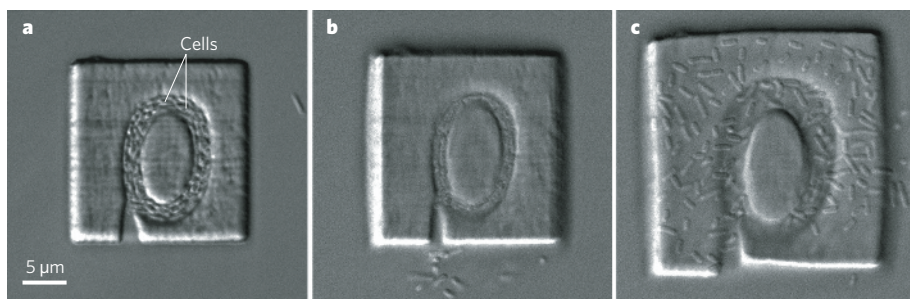
a fresh twist to this technique. They have generated complex materials by using lasers to induce crosslinking reactions in proteins. Remarkably, the resulting three-dimensional objects change shape in response to their environment. Such systems might find use as moving parts in miniaturized devices, with applications in medical diagnostics or remote environmental sensing.

The authors' approach relies on advances in a technique known as three-dimensional light patterning. Traditionally, this involves 'drawing' a pattern with a narrow laser beam across liquid monomers on a surface. The monomers react to form solid polymers at the places where the laser strikes, so that three-dimensional objects can be built up one layer at a time. A faster process is also now used, in which an entire layer of polymer is made at once by passing

light through a stencil — known as a mask to those in the trade — of the desired pattern.

Kaehr and Shear combine these two approaches by raster-scanning a narrow laser beam across a mask on a protein solution, to create solid, three-dimensional structures. This allows complex macroscopic structures to be made without the elaborate and expensive apparatus required to raster-scan precise shapes with a laser. Using their technique, the authors can expeditiously generate objects with dimensions as small as 100 nanometres, and of any desired shape.

To date, proteins have not found much use as building blocks for three-dimensional photo-fabrication processes, because commonly used photochemical reactions typically denature or destroy the molecules. An unexpected breakthrough came from studies of photodynamic therapy, a medical procedure that uses molecules called photosensitizers to absorb light and so destroy tumours. The mechanism of action was thought to involve photochemical reactions that created covalent links between proteins (or between proteins and other molecules) in tumour cells, leading to cell damage. The crosslinking was generally considered to be uncontrolled, occurring randomly between the various kinds of amino acid. But studies on proteins<sup>3</sup>, and on synthetic macromolecules



**Figure 1 | Dynamic microchambers for cell culture.** Kaehr and Shear<sup>2</sup> have prepared gels that change shape in response to environmental cues, for example pH. The materials can be made into components for various devices, such as chambers for culturing cells. **a**, This doughnut-shaped chamber, in a bath at pH 7, contains cultured *Escherichia coli* cells. **b**, A change of acidity (to pH 12.2) causes compression of the chamber and the release of a few cells. **c**, Eventually, the chamber is completely disrupted, and all the cells escape into the surrounding environment.

containing selected amino-acid side chains<sup>4</sup>, conclusively demonstrated that crosslinking of only specific amino acids occurs in the presence of a photosensitizer on exposure to light.

Kaehr and Shear<sup>2</sup> exploited this controlled crosslinking process to make protein-based materials that have highly defined shapes and microstructures. Using their patterning technique in conjunction with a photosensitizer, they 'wrote' three-dimensional structures by crosslinking proteins in solution using spatially directed light. The entire process takes only a few minutes, and the resulting materials expand and contract, open and close, or flex in response to changes in their environment (such as changes in pH) — just as naturally occurring proteins do.

The rich chemistry of proteins, and the ease with which their properties can be modified by changing their amino-acid sequences, make this a versatile approach for synthesizing designer components for devices. Proteins can respond to several triggers, such as external changes in temperature or ionic concentration. It is also possible to control the number of molecular interactions between different proteins, to integrate several proteins into a single material, and to make three-dimensional protein structures at different scales. And, unlike synthetic macromolecules (polymers), proteins can be used that have uniform size, amino-acid sequence and three-dimensional structure. The combination of functional flexibility and the ability to make identical copies of each structure opens up many potential applications for crosslinked proteins as device components.

The authors demonstrated an application of their materials by using them to make dynamic enclosures for culturing cells (Fig. 1). The enclosures trap cells, expand as the cells multiply, and subsequently release the cultured cells in response to a change in pH. In the future, the properties of the protein gels used to make the microchambers could be modified by forming hybrid materials with synthetic polymers, and by using proteins that respond to a specific chemical signal. This approach has recently been demonstrated<sup>5</sup> in a similar material, in which a modified version of the protein

calmodulin was trapped within a polymer network. The volume of the resulting gel changed when calcium ions or ligand molecules bound to the protein.

So what does the future hold for Kaehr and Shear's sophisticated protein gels? As potentially useful biomaterials, opportunities abound. The sequence, concentration and spatial presentation of biological signals within the material — such as binding pockets or the active sites of enzymes — can easily be varied, and the crosslinked proteins can be designed so that an external stimulus exposes such embedded features. And by using enzymes as a basis

for the gels, materials could be designed that amplify chemical signals or generate multiple responses from a single input.

Nevertheless, it won't all be plain sailing. Many proteins denature in response to changes in temperature or other environmental cues, and it isn't known whether all crosslinked proteins will undergo reversible changes in response to such signals. But as the repertoire of proteins that can be used in these materials expands, a tantalizing array of prospects can be envisaged: customized gels with passive and active molecular barriers to sift or release soluble chemical signals; systems that combine several design elements to mimic the complexity of naturally occurring tissues; or even actuators that have dynamic responses reminiscent of muscle contraction. ■

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## ALZHEIMER'S DISEASE

# The latest suspect

Rudolph E. Tanzi and Lars Bertram

**Many genetic mutations and several environmental factors contribute to Alzheimer's disease. Yet another disease risk gene, one that is involved in calcium regulation, has been added to the mix.**

Alzheimer's disease is characterized by neurodegeneration in several brain areas, with the hippocampus being one of the first regions affected. Accumulation of aggregates of the amyloid- $\beta$  peptide, for example in the form of senile plaques, is another feature of this disorder. Dreses-Werringloer *et al.*<sup>1</sup> set out to search for genes that are preferentially expressed in the hippocampus and lie in chromosomal regions that have been linked<sup>2</sup> to the risk of developing Alzheimer's disease. Writing in *Cell*, they report the identification of a new Alzheimer's disease risk gene that not only meets both of these criteria, but also seems to modulate extracellular levels of amyloid- $\beta$ .

The authors call this gene *CALHM1* (for calcium homeostasis modulator 1), because its product seems to be involved in calcium homeostasis. They show that increased expression of the *CALHM1* protein, which they find

to be localized in the cell membrane and in the membrane of an intracellular organelle known as the endoplasmic reticulum, enhances the entry of calcium into the cell and thus its concentration in the cytoplasm. They find that this results in changes in the processing of the amyloid precursor protein that lead to reduced levels of amyloid- $\beta$  peptide.

Intrigued by these observations, the authors resequenced *CALHM1* from a small number of patients with Alzheimer's disease and healthy controls. They identify a single nucleotide polymorphism (variation) in this gene, which, at the protein level, results in the substitution of the proline amino acid at residue 86 with a leucine residue (P86L). In normal subjects, the gene version carrying this polymorphism (the leucine allele) seems to occur with a frequency of about 20%. In the patients with Alzheimer's, however, it occurs nearly twice as frequently. The authors then tested this polymorphism