CHEMICAL BIOLOGY

An ‘on’ switch for proteins

Current methods for producing proteins that can be activated by light require knowledge of the protein’s active site, or can reduce the protein’s functionality. A technique that overcomes these issues has been devised. SEE ARTICLE P.509

KLAUS MICHAEL HAHN

Cells can activate the same proteins at different times or places to generate diverse effects — for example, the same enzymes can be involved in both cell growth and programmed cell death. Many cellular processes that depend on the timing and site of protein activity can be studied in living cells, by triggering localized protein activity and examining the effects. In the past few years, scientists have developed ‘photoactivation’ methods that allow protein functions to be switched on by light[1,2]. On page 509, Wang *et al.*[3] now describe a photoactivation strategy that is both broadly applicable and minimally perturbs normal protein functions.

Approaches for the photoactivation and photoinhibition of proteins are available, but it is often difficult to apply these without modifying some of the proteins’ activities. These methods work by manipulating amino-acid residues involved in the target protein’s mechanism of action, within the active site. However, the structure and active sites of many proteins are poorly understood, preventing such methods from being applied to many important systems.

Wang and colleagues report a method that they term computationally aided and genetically encoded proximal decaging (CAGE-prox). In CAGE-prox, a straightforward computational method is used to identify a position in a protein of interest at which the introduction of a bulky chemical group is likely to perturb the protein’s interaction with its substrate. The amino-acid residue at that position is then replaced with a tyrosine residue that has been modified[4] to carry a group that can be cleaved using light. Once installed, this bulky group blocks the protein’s activity until light irradiation ‘prunes’ it back to the normal tyrosine structure (Fig. 1), whereupon activity is restored.

The computational method requires no information about a protein’s mechanism of action. CAGE-prox can therefore be applied to an amazingly wide range of target proteins. The authors elected to use a modified tyrosine residue, rather than other amino acids that could be modified with a light-cleavable group, because they found that the normal tyrosine produced after cleavage proved least likely to perturb folding or normal protein binding.

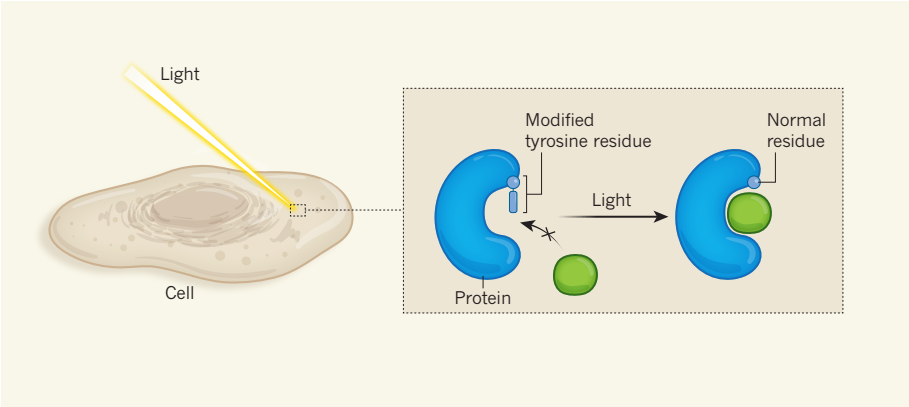

Figure 1 | A method for activating proteins using light. Wang *et al.*[3] report a technique that they call computationally aided and genetically encoded proximal decaging (CAGE-prox), which activates proteins in cells. In CAGE-prox, an amino-acid residue close to a protein’s active site is replaced by a modified tyrosine residue. The modified residue carries a bulky group on its side chain, which prevents the protein’s substrate (in this case, another protein; green) from binding in the active site. Light clips off the bulky side chain, leaving a normal tyrosine residue that allows substrate binding, thereby activating the protein.

One of the most striking advantages of CAGE-prox is its ability to produce an almost-native protein analogue — irradiation produces a protein that differs from the normal one by only a single amino-acid residue, which need not be in the active site. Other approaches have used light to link a fragment of an active protein to a second protein anchored at a specific site in the cell, thereby driving the fragment to specific regions[1]. Alternatively, engineered protein domains that change shape when irradiated have been inserted into a target protein to alter its conformation when illuminated, or have been positioned so that they block the target’s active site only in the dark[1,2]. These previously reported methods alter the overall structure of the target protein, thus potentially affecting its ability to interact normally with its biological partners.

By contrast, the surfaces used by wild-type proteins to mediate interactions with other cell components are retained almost intact in the CAGE-prox proteins after irradiation. This allows the modified proteins to target their normal binding partners in cells, and to be simultaneously activated at multiple locations in the same way as the wild-type protein.

It is perhaps surprising that the small changes associated with just one light-sensitive tyrosine residue can affect the interactions of a target protein so effectively. The success of Wang and colleagues’ method depends crucially on the ability to select the best site for modification. Encouragingly, the computational modelling used in CAGE-prox identified fewer than ten possible modification sites for each of the diverse proteins studied, limiting the number of amino-acid positions that had to be tested experimentally to find the most effective one.

The researchers used CAGE-prox not only to activate diverse protein structures, but also to control the sensitivity of kinase enzymes to inhibitors, thereby allowing modified and wild-type kinases to be inhibited independently. Just as impressively, Wang *et al.* used their method to activate proteins that have anticancer activity, and showed that light-triggered activation of these proteins inhibits tumour growth *in vivo* in mice.

In other photoactivation methods, a key amino-acid residue in the active site is identified and replaced by a modified version of that residue. By contrast, in CAGE-prox, the identified residue does not have to be in the active site and is always replaced by the same modified tyrosine residue. This tyrosine is introduced using a cell-based technique called unnatural-amino-acid mutagenesis[5,6]. The use of this technique could be seen as a weakness of Wang and co-workers’ approach, because unnatural-amino-acid

mutagenesis is not suitable for all cell types, and can require substantial optimization for each application. Furthermore, the covalent bond that is cleaved to remove the bulky side chain from the light-sensitive tyrosine residue can be broken only by using high-energy light (wavelengths of less than 400 nanometres), which is toxic to living cells. These are likely to be short-term obstacles, however, because many laboratories are actively pursuing and improving methods for altering proteins in cultured cells, and even *in vivo*.

With its remarkable simplicity and generality, CAGE-prox opens the door to studies of previously inaccessible cellular pathways, and of the spatio-temporal control of processes that determine cell behaviour. The range of applications that Wang *et al.* have already proved in principle for their technique is remarkable. No doubt, many more will soon follow. ■

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CONDENSED-MATTER PHYSICS

Superconductivity near room temperature

For a century, researchers have sought materials that superconduct — transport electricity without loss — at room temperature. Experimental data now confirm superconductivity at higher temperatures than ever before. SEE LETTER P.528

JAMES J. HAMLIN

Materials known as superconductors transmit electrical energy with 100% efficiency. They have a wide range of applications, such as magnetic resonance imaging in hospitals. However, these applications have been hampered, largely because the superconducting state exists only at temperatures well below room temperature (295 kelvin). On page 528, Drozdov *et al.*[1] report several key results that confirm that, when compressed to pressures of more than one million times Earth's atmospheric pressure, lanthanum hydride compounds become superconducting at 250 K — a higher temperature than for any other known material.

Superconductivity was first discovered[2] in 1911, in mercury cooled below 4 K. The temperature below which a material becomes superconducting is called the critical temperature. It was quickly appreciated that a state exhibiting zero electrical resistance could be tremendously useful, if materials that have critical temperatures much higher than 4 K could be found. Over the past century, as more superconductors have been discovered, the record for the highest critical temperature achieved has progressed, in fits and starts, towards the ultimate goal of room temperature.

In 2014, some of the authors of the current paper and their colleagues shattered the previous record[3] of 164 K. They found that hydrogen sulfide — a chemical responsible for the odour of rotten eggs — transforms into a superconductor at a temperature of about 200 K, when compressed to nearly 2 million times atmospheric pressure[4,5]. Then, in 2018, two independent research groups reported, almost simultaneously, that compressed lanthanum hydride compounds might exhibit superconductivity at even higher temperatures, ranging from 215 K to perhaps as high as 280 K (refs 6–8).

The common features of these hydrogen sulfide and lanthanum hydride superconductors are that they are rich in hydrogen and that superconductivity emerges only under pressures above about one million times atmospheric pressure. Under these extreme conditions, chemical bonds can be greatly altered, inducing the formation of compounds that are otherwise unstable. In the case of lanthanum hydride, high pressure seems to stabilize the formation of a compound, LaH_{10}, that has a much larger hydrogen content than is achievable at ambient pressure[9,10].

Drozdov *et al.* reached these extraordinarily high pressures (roughly half the pressure at Earth's core) using a device called a diamond anvil cell. This device could fit in the palm of your hand and generates pressure by squeezing a sample, contained by a thin metal foil, between two flattened diamonds (Fig. 1). This configuration severely limits the types of measurement that can be carried out, because the sample is tiny (of the order of 0.01 millimetres across) and is surrounded on all sides by the comparatively massive foil and diamonds. Moreover, to obtain electrical measurements, electrical leads need to make contact with the sample, but must remain electrically isolated from the foil.

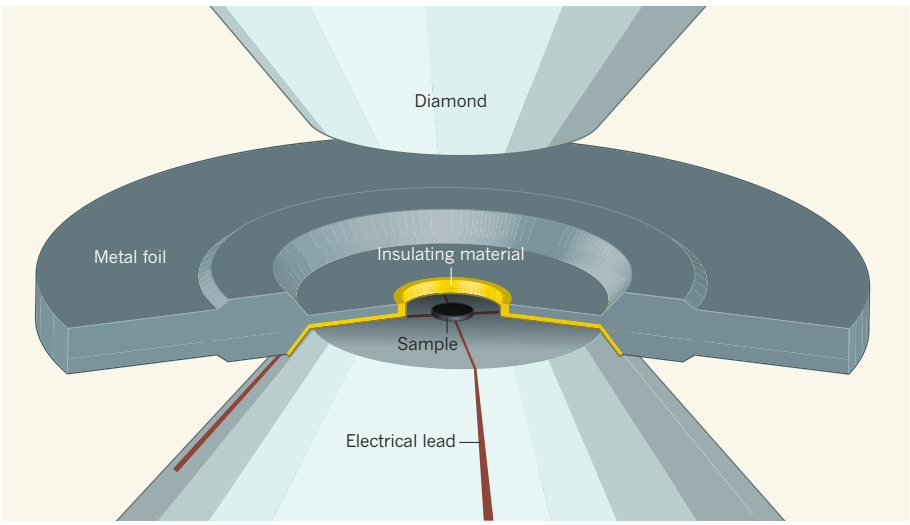

Figure 1 | Detection of high-temperature superconductivity. Drozdov *et al.*[1] report an experiment in which a tiny sample of lanthanum is enclosed inside a hole in a thin metal foil. The hole is filled with liquid hydrogen (not shown). Four electrical leads make contact with the sample but are electrically isolated from the foil by an insulating material. The sample is squeezed between two diamonds and transforms into lanthanum hydride at high pressure. The authors use this set-up to demonstrate that lanthanum hydride becomes superconducting at a temperature of 250 kelvin, under pressures greater than one million times Earth's atmospheric pressure.