conversion takes place in fractions of a second, resulting in H_2 productivities of, typically, 200 g of H_2 per kilogram of catalyst per hour[2]. Productivity at this level means that the production costs of H_2 (expressed as the cost in US dollars per joule of calorific value contained in the molecule) are two to three times higher than for natural gas[4,5]. However, taking into account the transportation and distribution costs of natural gas, and how pure you want the H_2 to be, the comparative cost of H_2 production can vary.

Biomass, the product of photosynthesis, could become a feedstock of choice for H_2 production, even in the absence of environmental premiums. At present, two strategies are being intensively pursued. The first is fractionation, which leads to the isolation and depolymerization of constitutive biomass fractions. An advantage of this approach is that the production of marketable goods (such as chemicals and fibres) can be integrated with the conversion of particular residual fractions to H_2, as well as to other biofuels or to bioenergy[6]. In fact, in the absence of subsidies or carbon credits (through which countries can trade off carbon-producing industry against carbon-absorbing activities, such as the planting of crops), co-products from biomass are necessary to make bioenergy or biofuels economically viable[7].

The second approach is through biosyntheses that produce H_2 directly. This requires photosynthesis to be conducted, as in algae, either with the removal of oxygen (to avoid poisoning the natural hydrogenases), or with oxygen-tolerant hydrogenases, which would need genetic engineering[8]. Alternatively, certain wet biomass fractions can be fermented by hydrogen-producing microorganisms in anaerobic environments where methanogenic pathways are suppressed[9]. Efforts in all of these directions are being pursued actively in academic and industrial laboratories.

Realistically, the introduction of a clean fuel such as H_2 is most likely to be achieved if the production technology can be integrated within the existing infrastructure used for natural-gas reforming. To use biomass as the source of H_2, several objectives must be achieved. To begin with, reaction and reactor productivities must match those achieved in hydrocarbon reforming. According to the results of Cortright *et al.*[1], this is almost achieved with glycerol as substrate, but in the case of sugars as substrate much lower water–sugar ratios for aqueous-phase reforming will need to be used. Heat inputs to balance the endothermic reforming reaction must also be equivalent to those used in hydrocarbon reforming. In steam reforming, a steam–carbon molar ratio of between 3 and 5 is normally used, so for aqueous-phase reforming, an energy-equivalent liquid-water–carbon molar ratio of between 7.5 and 12.5 is needed, at an operating temperature of 260 °C.

Practical feedstocks should be chosen, such as sugar-containing hydrolysates (and not just high-purity, model sugars) or glycerol-containing liquors derived from residual fats with minimum purification. These feedstocks should preferably be obtained from high-productivity biomass crops, with little or no use of synthetic fertilizers. The biomass should also be able to yield co-products that will improve the economics of the process. Crops such as sugar cane (in tropical climates), as well as switchgrass and hybrid poplar (in temperate climates), might well be suitable.

Biofuels are becoming a viable component of tomorrow's energy mix. If the appropriate feedstocks are considered, biomass refineries seem uniquely suited to providing a variety of products — food, fibres and chemicals as well as biofuels — to satisfy society's needs in a sustainable manner. ■

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Cognitive neuroscience

The molecules of forgetfulness

Alcino J. Silva and Sheena A. Josselyn

Not everything that we learn is useful, so the brain needs a mechanism to prevent itself being burdened by unhelpful details. The molecular details of this mechanism are now being uncovered.

Studies of the molecular and cellular foundations of cognitive processes have come of age with the development of techniques that allow genes to be over-expressed, deleted or modified in mice. These altered animals have been studied from a variety of aspects simultaneously by molecular biologists, neurophysiologists and psychologists. The result is the birth of a field that is unravelling the basis of learning, remembering[1], and now — as a paper in this issue shows — forgetting. On page 970, Genoux and colleagues[2] report that an enzyme known as protein phosphatase 1 (PP1) actively suppresses memories in mice, both during and after a learning exercise.

Like other biological processes, memory is regulated by yin-and-yang-like interactions between molecules with opposing functions — in this case, protein phosphatases and kinases, which respectively remove and add phosphate groups on target proteins, thereby altering their properties.

To examine the role of these interactions in forming and maintaining memories, Genoux *et al.*[2] generated mice that express a natural inhibitor of PP1, called inhibitor 1. When phosphorylated, this protein binds to PP1 and prevents it from working. The authors regulated the expression of inhibitor 1 in mice by using the reverse tetracycline transactivator system[3]: simply feeding the mice a tetracycline-like compound switches on the gene encoding the inhibitor, enabling PP1 activity to be blocked at will.

To test the animals' memory, the authors trained them in an object-recognition task that takes advantage of the natural propensity of mice and other rodents to investigate new objects more avidly than familiar ones. Their memory for objects depends on the hippocampus — a brain structure that shows robust expression of the inhibitor.

The authors' initial results help to explain one of the earliest discoveries of modern experimental psychology. In landmark experiments published in 1885, Hermann Ebbinghaus[4] showed that distributing training into several sessions results in stronger memories than equivalent amounts of training crammed into a single session (students take note!). Genoux *et al.* now find that massed training triggers more PP1 activity than distributed training. Remarkably, when the authors switched on inhibitor 1 specifically during massed training, blocking the PP1 activity, this type of training became as effective as distributed training. So PP1 apparently suppresses memory formation during massed training — but how?

One of PP1's targets is a gene-transcription factor called CREB, which becomes inactive when dephosphorylated by PP1. When phosphorylated, CREB directs the transcription of genes with specific CREB-binding DNA sequences in their control regions[5], and so is needed for proteins to be

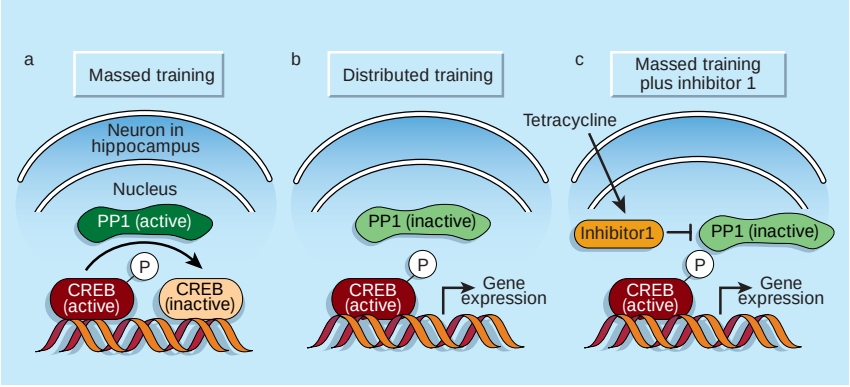

Figure 1 **Learning and forgetting — a tale of molecular antagonism. Genoux *et al.*[2] studied the molecular and cellular basis of memory by inserting into mice a gene that is activated, particularly in the brain's hippocampus, by feeding the mice the drug tetracycline. The protein produced from this gene, inhibitor 1, represses an enzyme, protein phosphatase 1 (PP1).** a, **When the mice were trained in a certain learning task in the absence of tetracycline, with the training crammed into one session, PP1 was active, and dephosphorylated (thereby inactivating) the gene-transcription factor CREB (a circled 'P' represents phosphorylation). This blocked the expression of genes needed for long-term memory formation.** b, **If training was distributed among more sessions, PP1 activity was lower, CREB was active, and the appropriate genes could be expressed.** c, **If inhibitor 1 was induced during massed training, repressing PP1, the levels of active CREB rose to those seen during distributed training in** b.

produced from these genes. Since the late 1960s, it has been known that blocking protein synthesis results in memory deficits that emerge within hours of training[6]. Later studies of species ranging from the marine mollusc *Aplysia* to flies, mice and rats have put this finding on a more detailed molecular footing, showing that CREB is required for the long-term processes involved in memory formation[7].

It now appears that one of the ways in which inhibitor 1 enhances memory during massed training is by blocking the PP1-dependent dephosphorylation of CREB that would normally occur (Fig. 1). That alters CREB's activity, which Genoux *et al.* studied by looking at the CREB-regulated expression of an inserted gene encoding β-galactosidase — an enzyme that produces a characteristic blue colour from its substrate. CREB activity (as judged by β-galactosidase expression) was higher after distributed training than massed training when inhibitor 1 was not switched on. But when activated, the inhibitor abolished this difference, increasing CREB function during massed training to levels comparable to those during distributed training.

Earlier work[8–10] showed that genetic manipulation of CREB also affects the learning regimen required to form long-term memories. In general, decreasing CREB levels increases the numbers of trials and the length of inter-trial intervals required for such memories to be formed, whereas increasing CREB levels has the opposite effect. Genoux *et al.*'s results suggest that the balance between the levels of active CREB and PP1 during training determines whether long-term memories are laid down.

The authors also found that inhibiting PP1 had almost immediate effects on memory formation. But these effects are unlikely to involve CREB-dependent transcription, which is an inherently slow process. Instead, these results point to molecules implicated in the early stages of memory formation, such as calcium/calmodulin-dependent protein kinase II (CaMKII)[11]. During learning, this enzyme becomes activated by increases in calcium levels inside neurons, with the result that several critical signalling molecules at neuronal junctions (synapses) are phosphorylated, and CaMKII itself becomes constitutively active. Such events are thought to be central to the changes in neuronal communication required for learning and memory. Indeed, genetic and pharmacological manipulations that interfere with CaMKII impair learning[11]. Genoux *et al.* now show that inhibiting PP1 results in higher levels of activated CaMKII. So, during massed training PP1 suppresses both short-term and long-term processes involved in learning and memory formation.

Can this phosphatase also suppress memories after they have formed? Learning is thought to trigger molecular changes (short- and long-term) that facilitate neuronal communication and are important for memory. Just as digital information is encoded by altering the grooves in a compact disc during burning, so memories are encoded by altering synapses in the brain during training. By means not yet understood, neurons can decode these changes and use them to reconstruct the learned information. Unlike a compact disc, however, the brain seems to store information in a highly dynamic manner: psychological studies suggest that the brain cares more about the usefulness of stored memories than their fidelity. Most of what we perceive and process is quickly discarded, and what we do remember tends to fade and change with time. There is a good reason for this: flawless recall would burden the brain with useless details, perhaps at the expense of storing and recalling information that is useful.

Genoux and colleagues have found that PP1 is involved in forgetting. The authors trained mice to find a submerged platform in a round pool of opaque water — another task that requires the hippocampus. To test the animals' memory, Genoux *et al.* then removed the platform. The mice initially searched for the platform in the correct quadrant; but with time, memory faded and performance declined. The authors found that blocking PP1 activity during training accelerated learning, but did not affect memory decline. But impairing PP1 activity after learning all but halted the memory decline for four to six weeks. Interestingly, studies[12] of an inserted CaMKII gene showed that interfering with its function after training destabilizes memory. So the antagonistic interactions between PP1 and CaMKII that are seen during learning may also be important later on, in maintaining memories.

Previous neurophysiological studies have shown that PP1, CaMKII and CREB modulate the changes in synaptic strength that are thought to underlie learning and memory[13]. These results mirror Genoux *et al.*'s molecular and behavioural findings. Similar examples of convergence between different types of study are now common in work on cognition at the molecular and cellular levels, showing that we are well on our way to unravelling the biology of learning and forgetting. ■

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