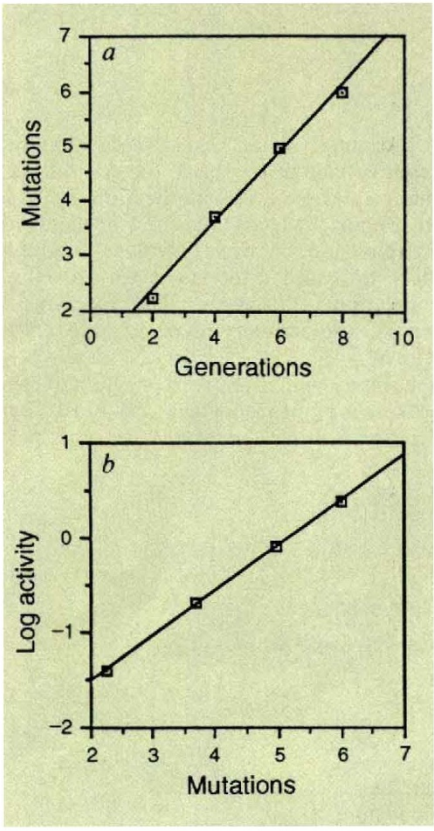

FIG. 2 *a*, The average number of mutations per sequence in the pool increases linearly with the number of generations. *b*, The log of the activity of the pool is a linear function of the average number of mutations per sequence in the pool.

selection, the average number of mutations in the surviving sequences is only 2.26; clearly, the vast majority of mutations are harmful and have been eliminated. Of course, clones with many more mutations are still present — they are just too rare to be seen. After this initial phase, the progress of the selection is remarkably uniform, with exponentially increasing activity and a steady linear increase in the average number of mutations (Fig. 2*a*). The gradual increase in the average number of mutations is due largely to the enrichment of progressively more active, but initially rarer, sequences that were present in the initial population. If a superior mutant sequence were present in the initial population, it would increase rapidly in numbers, and the subsequent creation of the same mutant sequence by new mutations would not contribute significantly to its abundance. Although the effect of new mutations is probably small after only eight generations, it seems quite likely that continued selection will lead to variants with so many beneficial mutations that they would not have been present in the original population.

The linear relationship between the log of the pool activity and the average number of mutations per sequence is surprising (Fig. 2*b*). The simplest interpretation of this correlation is that all the mutations contribute equally and independently to the increasing activity; why this should be so is less clear. This plot also tells us something about the shape of the activity 'hill' in sequence space: there seems to be at least one, and perhaps several, easily climbed ridges that lead gradually to a peak of activity. The fact that two of the mutations are found in mutually exclusive subsets of clones suggests that there are at least two different solutions to the problem of functioning with Ca^{2+}. The location and height of these peaks must await further generations of exploration.

How do the selected mutations change the ribozyme so that it can work with Ca^{2+} alone? It is by no means obvious how one could change a metal-ion binding site by simply changing the bases involved in direct interactions. Lehman and Joyce argue strongly from kinetic data and measured metal ion concentrations that the enzyme has not simply become so adept at binding Mg^{2+} that it can function in the low but unavoidable contaminating Mg^{2+}. Two more interesting possibilities are that the Mg^{2+}-specific site of the ribozyme has become modified so that it can bind either Mg^{2+} or the larger Ca^{2+} ion (radius 100 pm, compared with 72 pm for Mg^{2+}), or that it has become able to function without any divalent cation at all.

It is unlikely that any of the selected mutations are part of the critical metal-binding site of the ribozyme. Inspection of the three-dimensional model of Michel and Westhof[6], which is consistent with a remarkable number of other experimental tests, reveals that none of these mutations is close to the reactive centre. It seems more likely that they are at distant locations, and exert their effects by causing subtle changes in the geometry of the ribozyme, for example changing the twist of a helix, or the angle between two helices, the effects of which are then propagated to the binding site to displace the directly interacting bases to positions in space consistent with the binding of both Mg^{2+} and Ca^{2+}. Obviously, these experiments are crying out for a high resolution X-ray crystal structure for a proper interpretation. □

Jack W. Szostak is in the Department of Molecular Biology, Massachusetts General Hospital, and the Department of Genetics, Harvard Medical School, Boston, Massachusetts 02114, USA.

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DAEDALUS

Second thoughts

LAST week Daedalus proposed his encephalomagnet, a cunning quadrupolar affair with hollow pole pieces. Placed over the subject's head, it induces an intense local ring current in a small region of his brain. He then forcefully recalls whatever memory is stored at that location.

In effect, this is a sort of local electroconvulsive therapy (ECT). Instead of passing through much of the brain, with widespread disruptive effects, the current is neatly localized. Despite its fearful reputation, ECT relieves depression very effectively, though it does seem to cause some loss of memory. Encephalomagnetic therapy should be much more controllable.

Why does ECT cause memory loss? Memory is said to be stored in the synapses where one neuron connects with another. The triggering neuron releases a neurotransmitter into the synapse; this has some finite probability of launching a nerve impulse up the dendrite leading to the receiving neuron, which fires in its turn. The synapse 'learns' by altering this probability, so that all our knowledge is ultimately stored as a huge matrix of probabilities. Now ECT fires neurons by direct electrical stimulation. The resulting nerve impulse must travel out to all the ramifications of the neuron — even down the dendrite back to the receptor synapse, an event that never happens naturally. This unnatural backwards impulse, says Daedalus, must erase the firing probability stored at that synapse. Hence the loss of memory.

Memory loss is not altogether a bad thing. Daedalus recalls that dreadful verdict on the Bourbons: "They learnt nothing. They forgot nothing". Many creative people lose their spark in middle age: they know all the answers, but they don't notice that the questions have changed. So Daedalus's encephalomagnetic therapy may be a powerful rejuvenator. The local ring current could be used to explore round the brain until it stimulates some obsession, rigid misapprehension or area of total complacency. A cautious increase of current will then start to reset the local neurons, erasing their firing probabilities and freeing them to learn anew. The subject will find his certainties softening; new questions and ideas will begin to encroach on his previous unalterable convictions. He could even control the whole process himself. Artists, scientists, engineers and creative people generally should welcome this opportunity to renew their mental landscape. Sadly, the politicians, demagogues, academics and company chairmen most in need of the treatment will resist it fiercely.

David Jones