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Who's been nibbling on my PSD: Is it LTD?

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It is widely thought that long-term memory is stored by changes in synaptic strengths, but the mechanisms of the storage process remain unclear. One possibility is that the changes are purely modulatory, involving reactions such as phosphorylation of the receptors that mediate synaptic transmission. It had been generally assumed that such reactions are too transient to store long-term memory, however, both theory and experiment indicate that CaM-kinase, a kinase that can control glutamate receptors, may have special autophosphorylation properties that enable it to store long-term information. Evidence regarding the role of CaM-kinase in long-term potentiation (LTP) is reviewed elsewhere (Lisman, 1994). An alternative possibility is that changes in synaptic strength are determined by structural alterations, such as a change in the number of postsynaptic receptors. It has generally been assumed that activity-dependent structural changes are too slow to contribute to the early stages of plasticity. However, this assumption is unwarranted if one considers vesicle exocytosis as a means of adding new components to the presynaptic or postsynaptic membrane.

There has been considerable effort using anatomical methods to determine whether structural changes occur after LTP (Wallace *et al*, 1993). One type of study has compared hippocampal tissue in which LTP has been induced to control tissue. This necessarily involves statistical comparisons and there are many technical difficulties. Most studies have detected some structural changes, but there is no general agreement about the nature of these changes.

Another approach to studying anatomical plasticity is based on analysis of the diversity of synaptic structure in a given slice (reviewed in Lisman and Harris, 1993). Three-dimensional reconstructions show that even nearby synapses of Schaffer collaterals onto the same dendrite of a CA1 pyramidal cell can be highly heterogeneous in size. However, it remains unclear whether the growth process that underlies this diversity is an expression of LTP or independent of LTP.

One of the interesting conclusions that can be drawn from studying synaptic diversity is that the growth process is trans-synaptic (Lisman and Harris, 1993). Irrespective of the size of the synapse, the presynaptic grid and postsynaptic density have exactly the same lateral borders when viewed in cross-section. This and other data suggest that the growth process that accounts for synapse diversity is a trans-synaptic, structurally coordinated process. Viewed in cross-section, one would conclude that synapse construction is governed by a simple, well-ordered process.

And so it is surprising that when viewed *en-face*, the lateral borders of the synapse have a highly disordered and almost moth-eaten appearance. This is particularly true for large synapses, an example of which is shown in figure 1. Why should a structure that is so orderly arranged perpendicular to the membrane be so oddly and irregularly arranged in the plane of the membrane?

One possibility is that the moth-eaten shape of the synapse relates to the bidirectional control of synaptic strength. Physiological studies have found that some patterns of stimulation (1 Hz for

* Abbreviations: PSD, postsynaptic density; LTD, long-lasting depression; LTP, long-term potentiation.

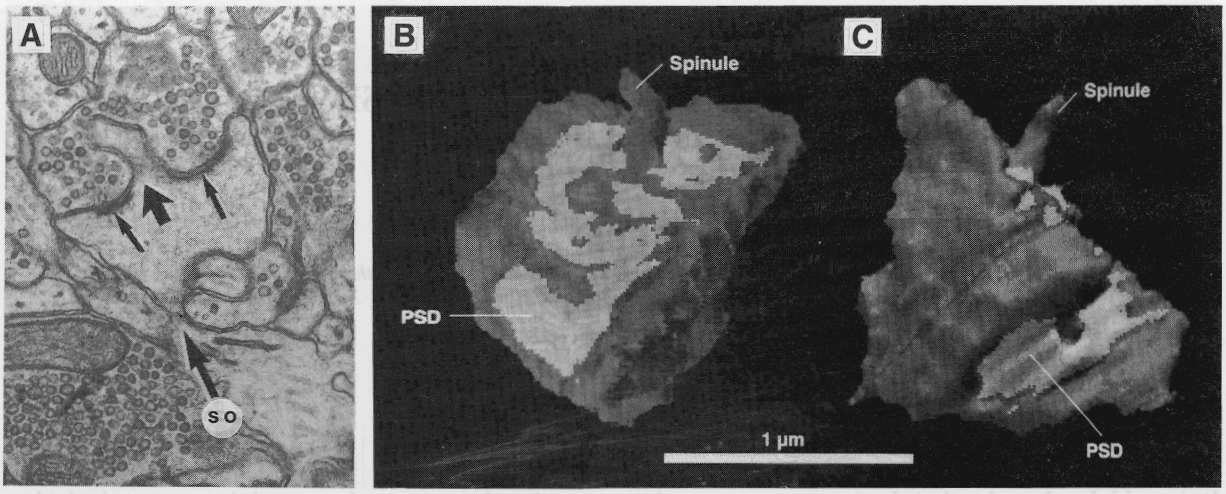

Fig 1. A. Cross-sectional view through a perforated postsynaptic density (small arrows) with a spinule (large arrow) in hippocampal area CA1 of the rat hippocampus. **B, C.** Three-dimensional reconstruction of the dendritic spine (gray) and the highly irregular PSD surface area. SO, spine origin with its parent dendrite, this connection is made in adjacent serial sections.

300 s) produce a long-lasting depression (LTD) of the same synapses that undergo LTP in response to other stimulation protocols (reviewed in Malenka and Nicoll, 1993). Let us assume that LTP and LTD are due to structural changes, but of opposite sign. One could then imagine that the complex shape of synapses resulted from the cumulative effects of periods of growth, during which the synapse expanded at its edge, and periods of constriction, during which the synapse was digested at its edges.

A somewhat different view is that the complex shape arises from a digestion process that is actually part of the LTP growth process itself. This possibility is suggested by recent evidence that LTP results in an increase in segmented synapses that have two separate active zones on single spine heads (Geinisman *et al*, 1993). If this increase arises from a splitting of a single synapse during LTP, then LTP must necessarily involve a digestive process. The highly complex shape of many PSDs may reflect this digestive process, even though complete splitting has not yet occurred.

What kind of data would be useful for clarification of these difficult issues? There remains some doubt as to which kind of structural change is specifically related to LTP. Furthermore, the question of whether LTD involves structural changes has not been addressed at all. Answers to these questions would be most convincing if they were based on observation of individual living synapses as they underwent LTP or LTD. A prerequisite of this approach would be the ability to identify which synapses are actually undergoing synaptic changes in living tissue. Recent experiments indicate that it may be possible to optically monitor individual synapses and extensions of this method may make it possible to determine whether LTP or LTD occurs at a given synapse (Malinow *et al*, 1994; Murphy *et al*, 1994). It could then be determined whether LTP or LTD was accompanied by changes in spine size. Since serial EM studies show a near perfect correlation between the size of the synapse and the size of the spine, one might infer that if the spine enlarges, the synapse enlarges also but such inferences will not really settle the issue. What is needed is a direct measurement of synapse size. It is hard to see how this could be measured optically, given that synapses are usually less than 0.2 micron in diameter (Harris and Stevens, 1989), below the resolution limit of the light microscope. Thus, for determination of actual synapse size, there is little alternative to using EM analysis a cross-slice comparisons. New stimulation protocols, statistical methods, and procedures for three-dimensional reconstruction will put the interpretation of structural results on a sounder footing. It is thus likely that the EM approach will be vital in determining who it is that's been nibbling on the PSD.

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