SYNAPTIC PHYSIOLOGY

Counting glutamate receptors

A team from The Cold Spring Harbor Laboratory led by Karel Svoboda has quantified the number of *N*-methyl-D-aspartate receptors (NMDARs) that open during low frequency synaptic transmission in the hippocampus of rats. The number is surprisingly low.

The new study, published in *The Journal of Neuroscience*, used novel imaging assays of calcium transients in individual dendritic spines to estimate the number of NMDARs activated at single synapses. For the experiments, CA1 pyramidal neurons were loaded with a calcium indicator. Changes in fluorescence evoked by synaptic stimulation were measured using two-photon laser scanning microscopy. Under the experimental conditions, transient changes in calcium concentration in individual spines were due to the opening of NMDARs at single synapses.

Synaptic stimulation randomly produced both large amplitude fluorescence changes ('successes') or no change ('failures'). In general, failures might be the result of two experimentally indistinguishable events — failure of glutamate release, and failure of channels to open following glutamate release. Consistently, partial block of NMDARs using the high-affinity antagonist D-CPP increased the number of failures.

On the basis of this observation, the authors derived an equation to calculate the number of NMDARs that opened following transmitter release, based on the probability of failures in the presence and absence of D-CPP. The probability of failure was tracked by fluorescence imaging of calcium transients, as described above. Concomitant measurement of whole-cell current provided an estimate of the fraction of NMDARs that were blocked by D-CPP. Plugging these data into their equation led the authors to conclude that, at most synapses, fewer than five NMDARs open at some point after transmitter release. At the peak of the synaptic response, the average number of open NMDARS is less than one.

Using these estimates, in combination with measurements of excitatory postsynaptic potentials in the presence and absence of D-CPP, the team determined the number of AMPARs (α-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid receptors) that probably open in response to low frequency stimulation. As for NMDARs, the average number of open AMPARs was small — about ten for most synapses.

Previous studies have indicated that the potency of synapses is correlated with their distance from the soma. To test whether these differences in potency are due to variations in the number of receptors per synapse, Svoboda and colleagues plotted their estimates of the numbers of open NMDARs against the position of synapses on the dendritic tree. No significant correlation was detected. Nor was the number of open NMDARs strongly correlated with spine volume, indicating that changes in NMDAR-mediated calcium concentrations are greater in smaller spines. As such, spine size might influence susceptibility to different forms of synaptic plasticity.

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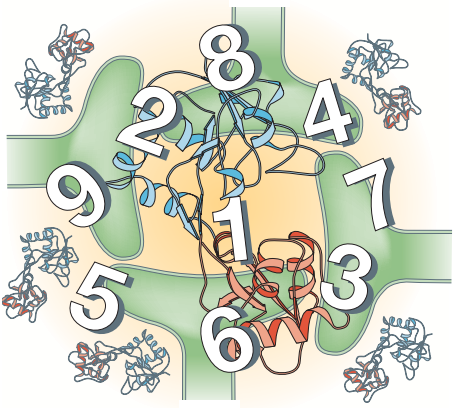

References and links

ORIGINAL RESEARCH PAPER Nimchinsky, E. A. *et al.* The number of glutamate receptors opened by synaptic stimulation in single hippocampal spines. *J. Neurosci.* **24**, 2054–2064 (2004)

FURTHER READING Madden, D. R. The structure and function of glutamate receptor ion channels. *Nature Rev. Neurosci.* **3**, 91–101 (2002)

REPAIR

Bridging the gap

Attempts to repair spinal cord injuries have focused largely on encouraging axons to regenerate through the lesion. However, spinal cord lesions create a hostile terrain for axon growth, owing to the presence of myelin-associated growth inhibitors and glial scar tissue. To overcome these obstacles, Campos and colleagues have developed a new approach in which a healthy nerve is re-routed to bypass the injury.

The T13 motor nerve emerges from the spinal cord at the level of the thirteenth thoracic vertebra, and it normally innervates the lower abdominal muscles. In adult rats, Campos *et al.* detached the distal end of the nerve from its muscle target and inserted the cut end into the spinal cord at the lumbosacral level. Over time, they found that the nerve extended axonal processes into the spinal cord tissue and established new synaptic connections. Moreover, electrical stimulation of the inserted nerve evoked contractions in the muscles of the lower back or hindlimb, depending on the site of insertion.

The authors investigated whether a re-routed T13 nerve could elicit functional recovery after a spinal injury. They made a hemisection in the spinal cord at the level of lumbar vertebrae 2/3 (L2/3). On its own, this injury produced spasticity and muscle wasting in the hindlimb that was ipsilateral to the hemisection. However, if the T13 nerve was grafted into the spinal cord at the L3/4 level, there was a marked recovery of hindlimb mobility, and muscle wasting was reduced.

So, Campos *et al.* have shown that the T13 nerve can establish a new circuit with neurons at the lumbosacral level of the spinal cord, thereby restoring the lines of communication between the brain and the hindlimb after spinal injury. In these preliminary experiments, the spinal hemisection and T13 graft were carried out simultaneously, but it will be interesting to find out whether a nerve bypass graft can also restore function to paralysed limbs in cases of chronic spinal cord injury.

Heather Wood

References and links

ORIGINAL RESEARCH PAPER Campos, L. *et al.* Engineering novel spinal circuits to promote recovery after spinal injury. *J. Neurosci.* **24**, 2090–2101 (2004)

FURTHER READING Filbin, M. T. Myelin-associated inhibitors of axonal regeneration in the adult mammalian CNS. *Nature Rev. Neurosci.* **4**, 703–713 (2003) | Silver, J. & Miller, J. H. Regeneration beyond the glial scar. *Nature Rev. Neurosci.* **5**, 146–156 (2004)