*Eur. J. Clin. Nutr.* **53,** 831–839 (1999).
3. Ashihara, H. & Crozier, A. *Adv. Bot. Res.* **30,** 117–205 (1999).
4. Mazzafera, P., Crozier, A. & Magalhães, A. C. *Phytochemistry* **30,** 3913–3916 (1991).
5. Kato, M. *et al. Plant Physiol.* **120,** 579–586 (1999).
6. Ross, J. R., Nam, K. H., D'Auria, J. C. & Pichersky, E. *Arch. Biochem. Biophys.* **367,** 9–16 (1999).
7. Stensvold, I., Tverdal, A., Solvoll, K. & Foss, O. P. *Prev. Med.* **21,** 546–553 (1992).
8. Hertog, M. G. L. *et al. Lancet* **342,** 1007–1010 (1993).
9. Vinson, J. A. & Dabbagh, Y. F. *FEBS Lett.* **433,** 44–46 (1998).
10. Leenen, R., Roodenburg, A. J. C., Tijburg, L. B. M. & Wiseman, S. A. *Eur. J. Clin. Nutr.* **54,** 87–92 (2000).
11. Fujiki, H. *J. Cancer Res. Clin. Oncol.* **125,** 589–597 (1999).
12. Lane, J. D. *et al. Psychosom. Med.* **60,** 327–330 (1998).
13. James, J. E. *Lancet* **349,** 279–281 (1997).

Neurobiology

# Kainate receptors and synaptic plasticity

Bortolotto *et al.*[1] report that the kainate subtype of glutamate receptor is essential for the plasticity of certain types of synaptic transmission in the brain, which is of interest as these receptors were previously not thought to initiate plastic processes. In particular, a new antagonist (LY382884) was shown to act selectively against the GluR5 type of kainate receptor: in the presence of LY382884, which reduces kainate-receptor-mediated postsynaptic responses by ~40%, long-term potentiation (LTP) at hippocampal mossy-fibre synapses could no longer be induced[1]. Here we argue that the available evidence does not support a major role for kainate receptors in the induction of mossy-fibre LTP.

It is well established that kainate receptors are also blocked by the ionotropic glutamate-receptor antagonists kynurenate and CNQX, and therefore, if Bortolotto *et al.* are correct, mossy-fibre LTP should also be blocked by these antagonists. Although they do provide evidence that this is the case, their results contradict earlier work showing that these antagonists, at the same or higher concentrations, are ineffective on mossy-fibre LTP[2–6].

In two of these studies[2,3], 10–20 mM kynurenate was found to have no effect, whereas Bortolotto *et al.*[1] report that 10 mM kynurenate blocks LTP completely (for reference, 1 mM kynurenate is sufficient to block the kainate-receptor-mediated excitatory postsynaptic current (e.p.s.c.)[7]). The others[4–6] showed that 10–20 μM CNQX, which also blocks the kainate-receptor-mediated e.p.s.c. by 40–80% (ref. 8), did not prevent induction of mossy-fibre LTP. In two cases[5,6], the entire experiment was done in the presence of 10 μM CNQX, the concentration used to block mossy-fibre LTP by Bortolotto *et al.*[1], and the increase in glutamate release that underlies mossy-fibre LTP was monitored using the NMDA-receptor-mediated e.p.s.c.

By studying LTP on the NMDA-receptor component of the e.p.s.c., which has properties identical to the LTP of the AMPA-receptor component[5,6], the potentiation can be assessed continuously after induction. We believe that this approach has better resolving power than the experiments of Bortolotto *et al.*[1], where LTP is presumed to be blocked on the basis that no potentiation is apparent three hours after induction and washout of the antagonist. Evidence using glutamate-receptor antagonists[2–6] would indicate that kainate receptors are not required for mossy-fibre LTP.

The results taken together suggest that the action of LY382884 on mossy-fibre LTP is either nonspecific or indirect. This is supported by expression data showing that the GluR5 subunit of kainate receptors is only weakly expressed in the dentate gyrus and in area CA3 of the hippocampus[9,10], and that most (about 90%) of the cells generating this weak signal are GABAergic interneurons[10], thought not to be directly involved in the induction of mossy-fibre LTP.

Given these combined results[2–6,9,10], we believe that GluR5 is unlikely to play a role in mossy-fibre synaptic transmission or plasticity; although LY382884 may antagonize GluR5-containing kainate receptors, its effect on mossy-fibre LTP is more likely to involve some other interaction of this drug with synaptic transmission.

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1. Bortolotto, Z. A. *et al. Nature* **402,** 297–301 (1999).
2. Castillo, P. E., Weisskopf, M. G. & Nicoll, R. A. *Neuron* **12,** 261–269 (1994).
3. Yeckel, M. F., Kapur, A. & Johnston, D. *Nature Neurosci.* **2,** 625–633 (1999).
4. Ito, I. & Sugiyama, H. *NeuroReport* **2,** 333–336 (1991).
5. Weisskopf, M. G. & Nicoll, R. A. *Nature* **376,** 256–259 (1995).
6. Tong, G., Malenka, R. C. & Nicoll, R. A. *Neuron* **16,** 1147–1157 (1996).
7. Castillo, P. E., Malenka, R. C. & Nicoll, R. A. *Nature* **388,** 182–186 (1997).
8. Vignes, M. & Collingridge, G. L. *Nature* **388,** 179–182 (1997).
9. Bahn, S., Volk, B. & Wisden, W. *J. Neurosci.* **14,** 5525–5547 (1994).
10. Paternain, A. V., Herrera, M. T., Nieto, M. & Lerma, J. *J. Neurosci.* **20,** 196–205 (2000).

*Bortolotto* et al. *reply* — Nicoll *et al.* challenge our finding that kainate receptors are involved in mossy-fibre LTP in the hippocampus[1]. Their argument is based on a discrepancy of our results with earlier work, as we show (our Fig. 5)[1] that two kainate-receptor antagonists, kynurenate and CNQX, can also block mossy-fibre LTP. We do not dispute that it may be possible to induce mossy-fibre LTP in the presence of kainate-receptor antagonists under certain conditions. For this reason, we neither stated nor implied that activation of kainate receptors is always necessary for mossy-fibre LTP[1].

Concerning the resolving power of our experiments, it is important to consider three factors. First, although the effect of CNQX on the induction of LTP had to be assessed three hours after the tetanus owing to the slow washout of CNQX, we often included a non-tetanized input as a control, which also invariably returned to the same, pre-CNQX, level[2]. Second, in our experiments with the selective AMPA-receptor antagonist GYKI53655, we also had to measure LTP three hours after washout of the antagonist, and LTP was always observed in the tetanized input[2]. Finally, the kynurenate experiments required only one hour for complete washout.

The relatively low expression of GluR5 messenger RNA in principal cells within the dentate gyrus and area CA3 was not a major concern for us because the relation between GluR5 gene expression and protein levels at this synapse is not known. In addition, as discussed previously[3], it is possible that the LY382884 class of compounds can also act on GluR5-lacking heteromeric assemblies of kainate receptors, such as GluR6-KA1, which have not been tested with these antagonists.

The fact that others have observed mossy-fibre LTP in the presence of kynurenate and CNQX has several possible explanations, the most likely being, we suspect, that involvement of GluR5-containing kainate receptors in mossy-fibre LTP can be bypassed in certain circumstances. To determine conditions under which LY382884 might fail to block the induction of mossy-fibre LTP, we have applied multiple trains at test intensity and find that LTP is always blocked by the antagonist; however, when we greatly increase the stimulus strength during the tetanus, some LTP remains[2].

Multiple routes for the induction of LTP at CA1 synapses, involving both NMDA[4] and mGlu[5] receptors, have already been reported. Crosstalk between receptors and their signalling cascades may make LTP more difficult to understand, but it provides synapses with a much richer array of mechanisms with which to build memories.

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1. Bortolotto, Z. A. *et al. Nature* **402,** 297–301 (1999).
2. www.bris.ac.uk/synaptic/info/kainate.html
3. Vignes, M. *et al. Neuropharmacology* **37,** 1269–1277 (1998).
4. Grover, L. M. & Teyler, T. J. *Nature* **347,** 477–479 (1990).
5. Bortolotto, Z. A., Bashir, Z. I., Davies, C. H. & Collingridge, G. L. *Nature* **368,** 740–743 (1994).