ADDICTION

Of mice and morphine

The development of tolerance and addiction to morphine is thought to involve 'anti-opioid' systems that include some types of glutamate receptor. In the *Journal of Neuroscience*, Inoue *et al.* show that both tolerance and dependence require a specific receptor subtype, but that they are mediated by different brain areas.

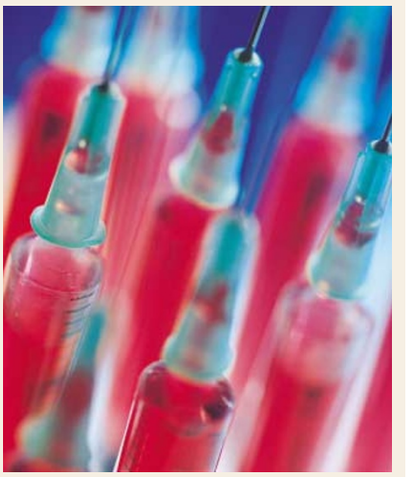
The authors investigated the effects of morphine in mice lacking the gene for the NR2A subunit (also called GluRε1) of the *N*-methyl-D-aspartate (NMDA) receptor. The NR2A-knockout mice showed an increase in the acute analgesic effect of morphine, but the ability of morphine to induce tolerance or dependence was reduced in these mice.

In control mice, chronic treatment with morphine that was sufficient to induce tolerance also caused an increase in levels of NR2A in certain areas of the brain: the periaqueductal grey, the ventral tegmental area and the nucleus accumbens.

To investigate the roles of these areas more fully, Inoue *et al.* induced expression of NR2A by electroporating the gene into specific nuclei in the brains of adult mice. Expression of NR2A in either the periaqueductal grey or the ventral tegmental area — but not the nucleus accumbens — rescued the ability of morphine to induce tolerance in the mutant mice.

Induction of dependence by a different treatment protocol also increased NR2A expression in normal mice, but in this case the increase was only in the nucleus accumbens. Consistent with this, knockout mice in which NR2A was expressed in the nucleus accumbens became susceptible to morphine dependence.

The new results support the idea that an increase in NR2A in specific brain areas might contribute to the development of tolerance and dependence by acting as part of an 'anti-opioid' system. Local electroporation of receptor genes into the brains of knockout



mice could represent a useful approach for investigating these and other effects.

Rachel Jones

References and links

ORIGINAL RESEARCH PAPER Inoue, M. *et al.* Locus-specific rescue of GluRε1 NMDA receptors in mutant mice identifies the brain regions important for morphine tolerance and dependence. *J. Neurosci.* **23**, 6529–6536 (2003)

FURTHER READING Nestler, E. J. Molecular basis of long-term plasticity underlying addiction. *Nature Rev. Neurosci.* **2**, 119–128 (2001)

NEUROTECHNIQUES

Waste disposal under the spotlight

Destruction of key regulatory proteins by the ubiquitin/proteasome system is crucial for many regulatory processes in the cell, and this system has been proposed to have a role in a number of disorders, including cancer and neurodegenerative diseases. Despite the existence of animal models for a number of these diseases, such as Alzheimer's and Parkinson's diseases, *in vivo* data concerning the ubiquitin/proteasome system are lacking. To address this need, Dantuma and colleagues have developed a model for *in vivo* quantitative analysis of the degradative machinery by generating transgenic mice that carry a green fluorescent protein (GFP) reporter with a constitutively active degradation signal, according to work published in *Nature Biotechnology*.

The process of removing intracellular proteins involves a complex network of enzymes that link multiple N-terminally linked ubiquitin molecules to the protein substrate, which targets the substrate for unfolding and subsequent degradation by the proteasome. To dissect this process *in vivo*, Lindsten *et al.* selected a GFP-reporter fused to an N-terminally linked ubiquitin that serves as an acceptor for further ubiquitin molecules, thereby automatically delivering the GFP to the proteasome for degradation. The GFP fusion protein was tolerated at high concentration in mammalian cells. Proteasome inhibitors given to the transgenic animals expressing this reporter substrate resulted in substantial accumulation of GFP in multiple tissues, confirming the *in vivo* functionality of the reporter. Furthermore, analysis of the transgenic animals revealed that accumulation of the reporter was induced in primary neurons by an aberrant ubiquitin that is found in Alzheimer's disease.

The role of the ubiquitin/proteasome system in diverse disorders, as well as its identification as a possible therapeutic target, makes these GFP-transgenic animals an important tool for monitoring the status of the ubiquitin/proteasome system in physiological and pathological conditions.

Melanie Brazil, Senior Editor,
Nature Reviews Drug Discovery

References and links

ORIGINAL RESEARCH PAPER Lindsten, K. *et al.* A transgenic mouse model of the ubiquitin/ proteasome system. *Nature Biotechnol.* 20 July 2003 (doi:10.1038/nbt851)