Fragile X: a class of its own

Just over a decade ago, **fragile X syndrome** — a common form of hereditary mental retardation — rose to notoriety by being the first disorder to be caused by the expansion of a trinucleotide repeat. Fragile X sufferers have more than 200 CGG repeats in the 5' untranslated region of the fragile X mental retardation 1 (*FMRI*) gene, compared with 60 or fewer in normal individuals. As in other similarly inherited conditions, the expanded disease alleles derive from phenotypically normal individuals that carry an intermediate number of unstable repeats, which in the case of fragile X ranges from 60 to 200.

Although this inheritance model has held true for years, some of these so-called 'premutation' carriers have now been found to develop a new form of progressive neurodegeneration — a phenotype that is unrelated to fragile X. This observation provided the starting point for the study by Peng Jin and colleagues. By modelling the condition in *Drosophila* they show that human premutation repeats alone can lead to

neurodegeneration, and that the effect depends on the abundance and length of the repeat. This is also the first time that neurodegeneration has been attributed to RNA alone.

To test whether the neurodegeneration phenotype could be pinned down to changes in CGG-repeat size, Jin and colleagues looked at the effect of expressing human repeats of different length in several *Drosophila* tissues. What they found was clear cut: moderate expression of 60 CGG repeats (rCGG₆₀) had no effect, but moderate expression of the longer premutation rCGG₉₀, or overexpression of either rCGG₆₀ or rCGG₉₀, caused progressive degeneration specifically in neuronal tissue. This tells us that longer repeats are more toxic than shorter (normal length) ones and that toxicity increases with transcript abundance. The result also tallies nicely with what is seen in premutation patients, who have high levels of *FMRI* mRNA.

Fragile X patients lack any *FMRI* message, so the mechanism that underlies the progressive neurodegeneration must be distinct from that which causes fragile X. Instead, it could resemble that of other neurodegenerative disorders, such as some forms of **spinocerebellar ataxia**, which are caused by repeat expansions in noncoding regions and have been blamed on RNA.

DEVELOPMENT

Compartments in the roof

In recent years, the roof plate of the neural tube has emerged as an important source of dorsal neural patterning signals. In *Nature Genetics*, Awatramani *et al.* now provide some unique insights into the development and patterning of the hindbrain roof plate and a related structure, the fourth ventricle choroid plexus. Their findings indicate additional roles for these structures in the anteroposterior patterning of the nervous system.

The authors designed a powerful 'dual recombinase' transgenic strategy to map the fate of cells at the intersection of two gene expression domains. Enhancer elements from genes that were known to be expressed in intersecting patterns in the developing hindbrain were used to control the expression of two different site-specific DNA recombinases. The system was designed such that in cells expressing both recombinases, a reporter gene was activated in a permanent and heritable manner.

First, Awatramani *et al.* clarified whether the roof plate is of neural or non-neural origin. They mapped the fate of a population

of cells in the dorsal hindbrain that transiently expressed the *Wnt1* gene. The roof plate does not normally express *Wnt1*, but in the transgenic embryos, the reporter (in this case, green fluorescent protein) was expressed in the roof plate. This indicated that the roof plate cells are indeed of neural origin, and that they are derived from the dorsal neural tube.

Next, Awatramani and colleagues used the dual recombinase strategy to label cells that expressed both *Wnt1* and a marker gene for a specific hindbrain segment (or rhombomere). Interestingly, they showed that the hindbrain roof plate is segmented along the anteroposterior axis, and that the cells derived from different rhombomeres do not mingle. This is in contrast to the floor plate — the ventral counterpart of the roof plate — where there is extensive mixing of cells derived from different rhombomeres.

The authors also used the dual recombinase technique to investigate the development of the choroid plexus, a convoluted ribbon-like structure that

But are we any closer to understanding what these repeat RNAs do? In both experimental flies and human premutation patients, nuclear aggregates are seen that also contain ubiquitin, and so it is possible that here the RNA repeats sequester vital proteins from their normal functions. The fact that the formation of these clumps can be reversed by overexpressing the chaperone heat shock protein 70 (Hsp70), which normally unravels or destroys badly-folded proteins, is intriguing: the involvement of the protein-degradation machinery could link these RNA-mediated defects to the larger class of protein-based neurodegenerative disorders, many of which are also reversed by Hsp70.

The creation of a *Drosophila* model for this disease has already paved the way for targeted genetic studies. Meanwhile, it looks like fragile X is back where it started — in a class of its own.

Tanita Casci, Senior Editor,
Nature Reviews Genetics

References and links

ORIGINAL RESEARCH PAPER Jin, P. *et al.* RNA-mediated neurodegeneration caused by the fragile X premutation rCGG repeats in *Drosophila*. *Neuron* **39**, 739–747 (2003)

WEB SITE

Stephen Warren's laboratory:
<http://www.emory.edu/WHSC/MED/GENETICS/visitors/warren.html>

invaginates from the roof plate. The choroid plexus is best known as a producer of cerebrospinal fluid, but it is also thought to have a dorsoventral patterning function in the hindbrain. The authors confirmed that the choroid plexus cells are descendants of the same *Wnt1*-expressing population as the roof plate, and that like the roof plate cells, they respect boundaries.

The restriction of roof plate and choroid plexus cells to lineage-restricted compartments with distinct gene expression profiles makes these structures strong candidates for providing signals for the generation of neuronal diversity along the anteroposterior axis. By analysing and manipulating the gene expression profiles of specific roof plate and choroid plexus compartments, it should be possible to define their anteroposterior patterning roles more precisely.

Heather Wood

References and links

ORIGINAL RESEARCH PAPER Awatramani, R. *et al.* Cryptic boundaries in roof plate and choroid plexus identified by intersectional gene activation. *Nature Genet.* **35**, 70–75 (2003)

FURTHER READING Caspari, T. & Anderson, K. V. Patterning cell types in the dorsal neural tube: what the mouse mutants say. *Nature Rev. Neurosci.* **4**, 289–297 (2003) | Zinyk, D. *et al.* Fate mapping of the mouse midbrain-hindbrain constriction using a site-specific recombination system. *Curr. Biol.* **8**, 665–668 (1998)

BEHAVIOURAL NEUROSCIENCE

Decisions, decisions

“You must learn to consider the consequences of your actions.” How many times did we hear that while growing up? Being able to predict the outcome of an action — and to know whether that outcome is desirable — is essential for us to function normally. New evidence supports the idea that a prelimbic corticothalamic circuit is crucial for the ability of rats to make use of specific associations between actions and outcomes in an instrumental conditioning task.

Corbit *et al.* set out to distinguish the roles of two thalamic nuclei in this task. The mediodorsal nucleus is part of a circuit that includes the prelimbic part of the prefrontal cortex, whereas the anterior thalamic nucleus is linked to the hippocampus. Previous studies had suggested that mediodorsal lesions affected instrumental behaviour, but in some cases the lesions also affected the anterior nucleus. This, coupled with evidence that the hippocampal circuit, which includes the anterior nucleus, is important for some forms of learning, led to suggestions that impairments in action–outcome learning might have arisen from anterior damage, rather than from the mediodorsal lesion itself. The new data confirm that it is the mediodorsal thalamus, not the anterior nucleus, that is responsible.

Rats with anterior thalamic lesions performed normally on all measures tested. By contrast, rats with mediodorsal lesions showed specific impairments. Although they could learn to press a lever for food pellets

and another for sucrose, they were less sensitive than control rats to manipulations of the outcome. For example, if trained rats are allowed to eat as much as they want before a test, they will normally press the lever that previously delivered food pellets less often than one that delivered the sucrose drink. Mediodorsal lesions reduced the sensitivity of the rats to this specific, satiety-induced devaluation of one reward.

In another test, the relationship between pressing one lever and delivery of its associated reward was altered by delivering one of the two rewards independently of lever pressing as well as when the lever was pressed. Unlesioned rats and rats with anterior thalamic lesions showed reduced lever-pressing when it was no longer necessary to receive the reward, but rats with mediodorsal lesions continued to press both levers equally.

Control experiments showed that the mediodorsal lesions did not prevent the rats from distinguishing between the two rewards or the two levers. Rather, it seems that they impair the ability of the rats to learn and use information about the link between an action and its outcome. Prelimbic cortical lesions have similar effects, supporting the idea that a prelimbic–mediodorsal circuit supports action–outcome learning in this kind of task.

Rachel Jones

References and links

ORIGINAL RESEARCH PAPER Corbit, L. H. *et al.* Lesions of mediodorsal thalamus and anterior thalamic nuclei produce dissociable effects on instrumental conditioning in rats. *Eur. J. Neurosci.* **18**, 1286–1294 (2003)