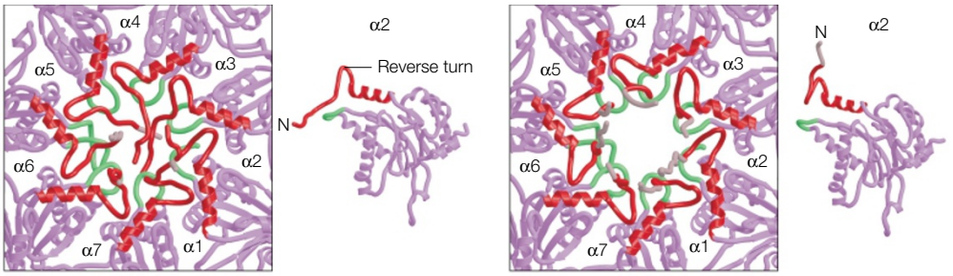
PROTEIN METABOLISM

Gates of destruction



Without PA26 **Bound to PA26**

An ancient Greek legend tells that the gate to Hades was guarded by Cerberus, a many-headed dog. But what guards the proteasome — a cellular underworld where proteins are sent to their deaths? Two structural studies, published in *Nature Structural Biology* and *Nature*, characterize the many-tailed beast at the proteasome's gate, and reveal the trick that the cell uses to tame it.

The working proteasome consists of a 20S core particle with a 19S regulatory particle at one or both ends. In the 20S core, four rings of seven subunits form a hollow chamber, with proteolytic β-subunit rings in the centre and α-subunit rings sealing each end. The seal itself, comprising a meshwork of highly divergent α-subunit amino-terminal tails, is broken when 19S particles are added, but we can only guess at how.

Michael Groll and colleagues suspected that the α3-subunit's tail holds the key to the gate, because it is the only tail that contacts all the others. They confirmed their suspicions by solving the structure of a 20S proteasome containing an α3-subunit lacking its amino terminus. This mutant 20S particle chomps its way through peptides without assistance from 19S particles, and its crystal structure reveals how: unlike in the wild-type 20S particle, where the seven amino-terminal tails have a fixed structure, the chamber's entrance is disordered in the mutant. The α3-tail therefore calls the others to order and, remarkably, it can do so even if it's added as a separate peptide.

One of the proteasome's tasks is to process antigens for presentation to T cells, but the optimal size of processed antigen is 8–10 amino acids — larger than the typical proteasome product. Interferon-γ induces the production of subunits that make proteasomes better suited to antigenic peptide production. One of these is trypanosome PA26 (PA28 in humans), which forms a heptameric 11S particle that can substitute for the 19S particle (although, unlike the 19S particle, it doesn't recognize the ubiquitin tags that target proteins for destruction). Frank Whitby and colleagues now show how the 11S particle opens the proteasome's gate. By binding a pocket in the interfaces between the α-subunits, the 11S particle puts pressure on a reverse turn at the end of each α-subunit's amino-terminal tail. This trips a conformational switch, causing the tails to point towards the 11S particle instead of obscuring the gate (see picture). By grabbing the dog by its tails, then, the 11S subunit opens the gates of destruction.

Both studies show that, irrespective of whether the α-subunits are in their open or closed conformations, the structures of the β-subunits remain unchanged, refuting the idea that gating might allosterically activate the the β-subunits. Does the 19S subunit use the same trick to open the gates? We'll need a hero bearing crystals before that legend can be told.

Cath Brooksbank

References and links

ORIGINAL RESEARCH PAPER Groll, M. *et al*. A gated channel into the proteasome core particle. *Nature Struct. Biol.* **7**, 1062–1067 (2000) | Whitby, F. G. *et al*. Structural basis for the activation of 20S proteasomes by 11S regulators. *Nature* **408**, 115–120 (2000)

FURTHER READING Pickart, C. M. & VanDemark, A. P. Opening doors into the proteasome. *Nature Struct. Biol.* **7**, 999–1001 (2000)

TRANSLATION

Unmasking the effects of CPEB

The best way to ensure the safe delivery of a protein is to produce it as close as possible to where it's needed. Reporting in *Cell*, Joel D. Richter and colleagues describe how this may be done for cyclin B1. By localizing the proteins responsible for translation of cyclin B1 messenger RNA to the mitotic spindle, production of this key player in cell division can be tightly controlled. And the results indicate that this regulation may be necessary for integrity of the mitotic apparatus.

Translation of cyclin B1 mRNA is regulated by cytoplasmic polyadenylation. This process is critical for the activation of different maternally inherited mRNAs during early development in many animals. It has been extensively studied in *Xenopus* oocytes where, in response to progesterone stimulation, the poly(A) tails of certain mRNAs (encoding, among them, several cyclins) are elongated.

A central player in polyadenylation is the cytoplasmic polyadenylation element-binding protein (CPEB), which recruits a factor that promotes the interaction between poly(A) polymerase and the end of the mRNA. Polyadenylation in turn triggers translation, and a key to this switch is maskin — a protein that was initially identified on the basis of its specific immunoprecipitation with CPEB.

Given that some mRNAs are concentrated in certain regions of *Xenopus* oocytes, Richter and colleagues wondered whether CPEB and maskin might be involved in mRNA localization as well as translation. To test this, they immunostained *Xenopus* oocytes at various stages of development with antibodies against the two proteins and found that both were especially concentrated at the cortex of the animal pole during late stages of oocyte development. Surprisingly, though, in the early embryo CPEB and maskin seemed to localize to structures resembling the mitotic spindle.

Closer analysis using antibodies against α-tubulin, CPEB and maskin confirmed this suspicion. At metaphase, the authors observed a gradient of CPEB and maskin along the length of the spindles, peaking at the area around the centrosomes.

So CPEB and maskin localize to spindles and centromeres, but what about the mRNAs that they regulate? Using *in situ* hybridization, the authors next showed that cyclin B1 mRNA is also localized to the animal pole of *Xenopus* oocytes and, more specifically, to spindles. This localization depends on CPEB, as cyclin synthesis was blocked in one-cell embryos injected with an antibody against CPEB. This treatment also caused embryos to divide three to five times more slowly than controls, and many of them showed spindle defects.

The conclusion, then, is that CPEB and maskin regulate the translation of cyclin B1, a process that is important not only for integrity of the mitotic apparatus, but for cell division as a whole. Local delivery, it seems, really is the safest option.

Alison Mitchell

References and links

ORIGINAL RESEARCH PAPER Groisman, I. *et al*. CPEB, maskin, and cyclin B1 mRNA at the mitotic apparatus: implications for local translational control of cell division. *Cell* **103**, 435–447 (2000)

FURTHER READING Richter, J. D. Cytoplasmic polyadenylation in development and beyond. *Microbiol. Mol. Biol. Rev.* **63**, 446–456 (1999)