IN BRIEF

TECHNIQUES & APPLICATIONS

RNA visualization in live bacterial cells using fluorescent protein complementation

Valencia-Burton, M. *et al. Nature Meth.* **4**, 421–427 (2007)

Localization and visualization of RNA in live bacterial cells is now possible thanks to a new method that uses a protein complementation assay. An RNA-binding protein (eIF4A) was split into two domains, each of which was fused to inactive fragments of a marker green fluorescent protein (GFP). Simultaneous binding of the eIF4A fragments to an RNA aptamer on a target RNA brings the two inactive fragments of GFP together, resulting in reconstitution of fluorescence and detection of the RNA of interest. Experiments with three different RNAs in *Escherichia coli* validated the technique: fluorescence depended on target RNA expression and could be quantified according to the level of gene expression; background was much lower than with similar techniques using a direct GFP reporter; and the complex assembled on the target RNA did not seem to interfere with its function or localization. The technique revealed spatial organization of transcription in *E. coli*. Untranslated message accumulated at the cell poles, which could be a storage site for unused RNAs.

BACTERIAL PATHOGENESIS

Borrelia burgdorferi intercepts host hormonal signals to regulate expression of outer surface protein A

Scheckelhoff, M. R. *et al. Proc. Natl Acad. Sci. USA* **104**, 7247–7252 (2007)

The agent of Lyme disease, *Borrelia burgdorferi*, is spread by biting *Ixodes* ticks and infects mammalian hosts to complete its replication cycle. The borrelial outer-surface protein A (OspA) is required for tick colonization. OspA is highly expressed by tick-borne bacteria but is downregulated in mammalian hosts. When ticks feed on infected mammals the OspA protein is upregulated and *B. burgdorferi* re-colonizes ticks. Scheckelhoff and colleagues showed that the mammalian stress hormones epinephrine and norepinephrine induce OspA expression in *B. burgdorferi*. Blocking the action of these hormones downregulated OspA *in vitro* and reduced the uptake of bacteria from infected mice, but only at the later stages of infection, not at the peak of bacterial dissemination. Further work will be needed to pinpoint how *B. burgdorferi* senses host hormones to aid completion of its infectious cycle.

VIROLOGY

Bluetongue virus VP4 is an RNA-capping assembly line

Sutton, G. *et al. Nature Struct. Mol. Biol.* 08 April 2007 (doi:10.1038/nsmb1225)

Capping of eukaryotic messenger RNAs protects the 5′-end of the RNA from degradation and promotes translation. Eukaryotic RNA viruses cap their genomic RNAs, often with their own enzymes. Bluetongue virus, a reovirus with a double-stranded genomic RNA, uses a single viral protein (VP4) to catalyse the four reactions that are needed to form an RNA cap. Solving the structure of VP4 to 2.5 Å has revealed how this enzyme works. Four active sites, defined by structural features and binding of ligands and products using soaks and co-crystallization, had a linear arrangement within the VP4 dimer. Bluetongue virus retains the capping enzyme in the core, unlike the turreted reoviruses which have a capping cage of five enzymes around the transcript. This structure provides further insights into the different capping solutions that viruses have evolved.