Nuthin' but a 'G' thang

G protein–coupled receptors (GPCRs) are membrane proteins involved in a broad range of biological processes, and a considerable proportion of clinically used drugs elicit their biological effect(s) via a GPCR. Structural information about GPCRs is quite limited, as the X-ray crystal structures of only two human protein receptors have been solved (the β_2 adrenergic and A_{2A} adenosine receptors). Wu *et al.* now report several X-ray crystal structures of CXCR4, a chemokine receptor that is involved in cancer metastasis and is a key mediator of HIV-1 entry into human cells. The structures are of the GPCR in the presence of IT1t, a drug-like small molecule, or CVX15, a short cyclic peptide that can function as an anti-metastatic therapeutic agent and an inhibitor of HIV-1 entry. The authors identify a number of differences between the structure of CXCR4 and the previously reported structures of GPCRs, including the observation that the ligand-binding pocket of CXCR4 is much larger and closer to the extracellular surface of the protein. The binding sites of IT1t and CVX15 overlap, though CVX15—which is much larger than IT1t—occupies more of the ligand-binding pocket and induces conformational changes in several adjacent helices. CXCR4 is known to form homo- and heterodimers *in vivo*, and the authors propose that ligand binding to one CXCR4 monomer could induce conformational changes in other CXCR4-associated proteins, modulating receptor function. Additional work is needed to characterize the stoichiometries, structures and *in vivo* functions of these multimeric complexes, especially because this may reveal new ways to combat HIV infection. (*Science*, published online 7 October 2010; doi:10.1126/science.1194396) *JMF*

Telomerase RNA round the bend

Telomeres shorten as a consequence of DNA replication, which can cause genome instability. The addition of new repeats to the telomere ends is the job of the telomerase enzyme. Telomerase contains an RNA component, the telomerase RNA (TR), and much remains unknown about its structure and how it catalyzes RNA template-driven repeat synthesis. Various regions of the core domain RNA, the minimal region of TR that is required for telomere repeat synthesis, have been determined. A key region important for catalytic function, conserved in location but not in sequence across vertebrate TRs, is the region around J2a/b, a 5-nucleotide (5nt) bulge connecting two conserved helices for which there is little structural information. Using NMR-based techniques, Feigon and colleagues now find that the human TR 5nt bulge has a crucial role in forming the tertiary structure of the core TR. Mutagenesis data indicate that the formation of the 5nt bulge is required for telomere repeat synthesis. In addition, further structural analyses indicate that the helices flanking the bulge flex relative to one another. Using this piece of the structure, the authors build a full model of the structure of the core region of the TR and find that the 5nt bulge is likely key to forming the overall architecture. The size of the modeled core domain is suggestively similar to the size of the putative beetle telomerase structure. Finally, the structure in and around the 5nt bulge is strikingly similar to that surrounding a 5nt bulge in the Hepatitis C Virus internal ribosomal entry site, indicating that this may be a new class of RNA structural element. (*Proc. Natl. Acad. Sci. USA*, published online 21 October 2010; doi/10.1073/pnas.1013269107) *SL*

Written by Inês Chen, Joshua M. Finkelstein, Maria Hodges & Sabbi Lall

Multiple toxicity

Bacillus anthracis, the bacterium that causes the disease anthrax, produces large quantities of the toxins lethal factor (LF) and edema factor (EF) that are able to enter host cells. Anthrax infection can be fatal owing to the vascular leakage and shock these factors promote. How the toxins act biochemically is reasonably clear, but how they disrupt cellular functions during the final stages of infection is not. Using *Drosophila melanogaster* as a model system, Guichard *et al.* demonstrate that the Rab11/Sec15 exocyst, which mediates the last step of the endocytic recycling pathway, is a novel target for EF and LF. These two toxins work in concert to block Rab11/Sec15-dependent endocytic recycling: EF reduces the levels of Rab11, indirectly blocking vesicle formation by its binding partner Sec15, and LF prevents the formation of Sec15 vesicles. As a result, signaling by the Notch ligands Delta and Serrate is reduced, as is the level of cadherin expression at adherens junctions. Similar effects were noted in human endothelial cells. Loss of adhesion probably partly explains the loss of endothelial barrier integrity in toxin-treated cells. The effect of Notch inhibition is less well understood and could be direct (for example through Notch-Delta–dependent adhesion) or indirect (perhaps through cytokine production). (*Nature* **467**, 854–858, 2010) *MH*

Unfolding costs

Molecular chaperones facilitate the correct folding of proteins. Some act on newly synthesized polypeptides; others can promote the refolding of misfolded species to their native state. Refolding a misfolded protein would seem less expensive than degrading it and synthesizing a new one. However, determining the energetic costs of refolding a misfolded polypeptide has been difficult, as classical enzyme kinetics studies are hampered by substrate heterogeneity—most misfolded proteins form polydisperse oligomers and precipitates—and by the fact that refolding assays have so far required a molar excess of chaperones over substrates. Now Goloubinoff and colleagues use a homogeneous misfolded substrate to directly measure kinetic parameters and ATP consumption for unfolding or refolding. The authors find that a luciferase variant can be converted into an inactive, monomeric form by freeze-thaw cycles. The stably misfolded luciferase could be unfolded by urea and spontaneously refolded to its native state. A chaperone system (the bacterial Hsp70, DnaK, together with its cochaperone DnaJ and nucleotide-exchange factor GrpE) could also promote the native refolding in an ATP-fueled process, under saturating substrate conditions. This allowed the measurement of kinetic parameters and the dissection of the process into distinct mechanistic steps. The authors propose that one DnaK molecule consumes five ATPs to promote the refolding of one misfolded luciferase to its native state. DnaK acts as a bona fide ATP-fueled unfoldase, and the released unfolded intermediate can spontaneously refold into its native or misfolded states. The data suggest that the former occurs with a 20% frequency. Even so, compared to the cost of degrading and resynthesizing luciferase (estimated to be around 3,000 ATPs), using a molecular chaperone to rescue a misfolded protein does seem like a huge bargain for the cell. (*Nat. Chem. Biol.*, published online 17 October 2010, doi:10.1038/nchembio.455) *IC*