

- 21 Ponting C.P. *et al.* (1997) The breast cancer gene product TSG101: a regulator of ubiquitination? *J. Mol. Med.* 75, 467–469
- 22 Koonin, E.V. and Abagyan R.A. (1997) TSG101 may be the prototype of a class of dominant negative ubiquitin regulators. *Nat. Genet.* 16, 330–331
- 23 Schultz, J. *et al.* (2000) SMART: A web-based tool for the study of genetically mobile domains. *Nucleic Acids Res.* 28, 231–234
- 24 Bateman, A. *et al.* (2000) The Pfam protein families database. *Nucleic Acids Res.* 28, 263–266

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## Condensing the RNA world

The track record of X-rays in revealing life's inner secrets is the envy of other photons. The most profound mysteries that have been investigated with them have been revealed first to the bravest and the most persistent people on earth. Few dared to purify a 1 500 000 Da complex containing 30 or more polypeptides and 990 000 Da of RNA. Fewer succeeded in crystallizing the large ribosomal subunit, and many turned to more sensible projects when they obtained poor crystals. By the time Ban *et al.*<sup>1</sup> obtained improved crystals of the large subunit from *Haloarcula marismortui* and found 132 heavy-atom positions for an osmium pentamine derivative and 84 heavy-atom positions for an iridium hexamine derivative, there was no turning back. The absence of three of the 31 polypeptides from the SwissProt database no longer elicited fear.

In the 20 years since Ada Yonath and co-workers reported the first crystals of the large subunit, new scientific and engineering disciplines, such as whole-genome sequencing, phylogenetic RNA alignment and secondary-structure prediction, and synchrotron X-ray crystallography came into practice; without these, a model of the large subunit could not have been built. Dominating the assembly is the 23S RNA macromolecule, whose six subdomains assemble into a large single domain. Of the 27 protein structures solved in the assembly, 21 were solved *de novo*; 13 have a nonglobular domain or are entirely extended in their native conformation. The nonglobular protein structures make no sense apart from the large subunit assembly and, in fact, resisted crystallization on their own.

Although Francis Crick proposed as early as 1968 that 23S RNA might catalyse peptide-bond formation, a huge body of genetic and biochemical work has been devoted to testing this hypothesis. Indeed, the notion that RNA once 'did it all' depends more significantly on

determining which enzymes in modern cells still 'do it' with RNA catalytic centers than on evolutionary speculation and reconstruction. Difference electron density maps of the large subunit bound to two substrate analogs now demonstrate that the N3 nitrogen of adenosine at position 2486 of 23S RNA is positioned to abstract a proton from the N terminus of the incoming aminoacyl tRNA (Ref. 2). In these X-ray structures, no amino acid side chain is within 15 Å of the condensation. The lack of direct involvement of protein in the reaction underscores the first impression of the large-subunit structure: that polypeptides are largely there to stabilize 23S RNA.

There is an amusing turnabout in the large-subunit story. Crystal structures of substrate analogs for peptide transfer are bound by RNA, proving that the ribosome is a ribozyme. The proposed mechanism (adenosine 2486 as a general base) is the

reverse of the mechanism for hydrolysis of an acylenzyme by serine proteases (histidine as a general base). Can the history of protein enzyme evolution be distilled to a counter attack of ribosomal protein synthesis? Revolutionary ribosomal RNAs make proteins, which then deploy an enhanced set of side chains as RNA mimics and function to break the bonds condensed in the RNA world.

- 1 Ban, N. *et al.* (2000) The complete atomic structure of the large ribosomal subunit at 2.4 Å resolution. *Science* 289, 905–920
- 2 Nissen, P. *et al.* (2000) The structural basis of ribosome activity in peptide bond synthesis. *Science* 289, 920–930

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## Fringe gives a saccharine to Notch

Post-translational modifications like phosphorylation, acetylation and fatty acid acylation of proteins are well-known strategies that cells use to regulate the activity of enzymes and maintain the binding properties of ligands and receptors. By contrast, glycosylation, where carbohydrates are attached to the side chains of amino acids, is important in the synthesis of many secreted and cell-surface proteins.

Moloney *et al.*<sup>1</sup> and Brückner *et al.*<sup>2</sup> now demonstrate that elongation of *O*-linked fucose on glycosylated proteins can act as an as-yet-unknown post-translational regulatory mechanism for modulating receptor–ligand interactions in signal transduction. Both groups studied the influence of glycosylation on the activity of Notch-induced signalling, which is important for the formation of tissue boundaries during development. Notch receptors are transmembrane proteins

with an extracellular domain of epidermal growth factor (EGF)-like repeats and are activated by two conserved families of ligand proteins, Jagged/Serrate and Delta.

Molony *et al.* show that Fringe, a modulator of Notch, possess a fucose-specific glycosyltransferase activity that catalyses the elongation of carbohydrates on the EGF repeats of the receptor protein. Using tritium-labelled saccharides they found that EGF-*O*-fucose is a highly specific target for Fringe. The elongation of the glycans on Notch led to an inhibitory effect on the activation of its ligand, Jagged1, as monitored by a luciferase-reporter assay. Brückner *et al.* demonstrate that Fringe displays its ability to modify Notch within the Golgi apparatus and that modulation of the receptor increases the binding activity to its ligand Delta. Both groups show that the Notch–ligand interaction is not affected when replacing the DxD motif (aa 236–238) in Fringe, required for the catalytic activity in many glycosyltransferases, by either NNN or DEE.