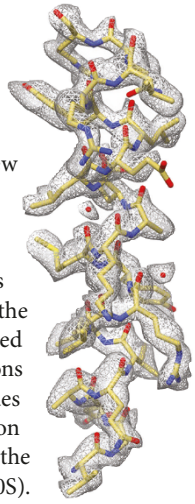
A barrier-breaking resolution



Substantial technical advancements have recently allowed single-particle cryo-EM to achieve resolutions comparable to those of X-ray crystallography in protein-structure determination. New direct electron detectors with faster acquisition rates and higher signal-to-noise ratios are supplanting charge-coupled-device cameras, and new algorithms enable correction of sample movements induced by the electron beam, to further boost the signal-to-noise ratio. These developments have led to a surge of cryo-EM–based protein reconstructions below 4-Å resolution. Carragher and colleagues have now reported the cryo-EM reconstruction of a protein complex below 3-Å resolution, for the *Thermoplasma acidophilum* 20S proteasome (T20S). By multiple rounds of particle selection to account for motions due to beam exposure and to eliminate particle images with high angular uncertainty, they sorted ~50,000 particles for T20S reconstruction at 2.81-Å resolution. Carragher and colleagues built the atomic model by rigid-body fitting of the crystal structure of T20S into the cryo-EM map, performing iterative refinement with Rosetta and improving the model by manual adjustments with Coot. The quality of the T20S cryo-EM map is akin to that achieved by X-ray crystallography at comparably high resolution, and it allows unambiguous assignment of side chains, establishment of different rotameric conformations and identification of ordered water molecules. This level of detail opens the possibility for cryo-EM to compete with X-ray crystallography for *de novo* determination of protein structures. (*eLife* doi:10.7554/eLife.06380, 11 March 2015) *CD*

The yin and yang of eIF3

During initiation of eukaryotic translation, the eIF3 complex acts as a scaffold that mediates recruitment of the 40S ribosomal subunit to the 5′ UTRs of cellular mRNAs. However, eIF3's functions seem to go beyond this general scaffolding role, and eIF3 promotes translation of hepatitis C viral RNA by directly binding a structured RNA element. Furthermore, eIF3 affects cellular differentiation and growth and is linked to various cancers, thus suggesting that it may direct the translation of specific cellular mRNAs. Cate and colleagues now provide PAR-CLIP analyses demonstrating that 4 of the 13 eIF3 subunits indeed interact directly with RNA. Genome-wide sequencing revealed that eIF3 was predominantly associated with the 5′ UTRs of mRNAs involved in cell growth processes. *JUN* and *BTG1* 5′ UTRs were among the top eIF3-binding targets, and reporter assays showed that, as expected from eIF3's canonical function, the eIF3-binding site of *JUN* was required for translation. Surprisingly, *BTG1*'s eIF3-binding site conferred translational repression. To understand how eIF3 could act as a positive or negative regulator of translation, the group used SHAPE analyses and showed that the eIF3-binding sites occurred in conserved stem-loop structures. Although eIF3 could bind the *JUN* 5′-UTR stem-loop directly, it appeared to require additional factors to bind the *BTG1* 5′ UTR, thus suggesting that eIF3's modes of binding to the RNA stem-loops may underlie its ability to activate or repress translation. Thus, this study uncovers a new form of translational control for cellular mRNAs and describes an unexpected dual role for eIF3. (*Nature* doi:10.1038/nature14267, 6 April 2015). *SG*

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Mammalian mitoribosomes revealed

Mitochondrial ribosomes, or mitoribosomes, are responsible for synthesis of mitochondrial membrane proteins. The structures of the large subunit (39S) from porcine and human mitoribosomes were described in 2014. Now the same investigators complete the picture with the structures of the full 55S mitoribosome. Amunts, Brown *et al.* determined the structure of the human mitoribosome at 3.5-Å resolution by cryo-EM and observed nascent polypeptide being folded in the exit tunnel. Greber, Bieri, Leibundgut *et al.* presented the cryo-EM structure of the small subunit (28S) of the porcine mitoribosome at 3.6-Å resolution and an atomic model of the entire mitoribosome with mRNA and tRNAs bound, based on a 3.8-Å cryo-EM map and data from cross-linking and MS analysis. The findings from both groups confirmed that the mammalian 55S mitoribosome is very different from its bacterial 70S counterpart, with a reduced rRNA core and numerous ribosomal proteins covering the surface almost completely. The small and large subunits interact less extensively than in bacterial 70S, to result in increased conformational flexibility. In fact, the small subunit could be observed in different orientations by both groups. The mRNA channel, responsible for binding leaderless mitochondrial transcripts, features remodeled entry and exit sites. Finally, the structures explain why certain polymorphisms in the mammalian mitoribosome rRNA lead to hypersensitivity to aminoglycoside antibiotics: those mutations allow base-pairing that would yield a mitoribosome more similar to the bacterial one and restore the binding pockets for aminoglycosides. (*Science* **348**, 95–98, 2015; *Science* doi:10.1126/science.aaa3872, 2 April 2015) *IC*

SETX attenuates antiviral transcription

Innate immune responses are crucial for defense against infectious agents, but the inherent danger of pathological inflammation and tissue damage highlights the need for rigorous control mechanisms. van Bakel, Marazzi and colleagues now uncover an unanticipated role for the helicase senataxin (SETX) in inhibiting the cellular transcriptional response to viral infection. The authors show that SETX specifically promotes premature promoter-proximal termination at genes whose expression is dependent on the interferon regulatory transcription factor IRF3. Interestingly, Sen1, the yeast ortholog of SETX, was known to track along nascent RNA and induce early termination at noncoding-RNA loci. Accordingly, SETX is able to bind RNA derived from the 5′ ends of antiviral genes, and expression of wild-type SETX, but not mutants lacking either ATPase or RNA-binding activity, increases the amount of short RNAs associated with transcription start sites. SETX-mediated termination might thus depend on sequence-specific recognition of target RNAs or be driven by interaction with structural elements formed at nascent transcripts. In addition, SETX interacts with the transcription-initiation factor TAF4, previously proposed to coordinate transcriptional responses by acting as a binding platform for both positive and negative regulators. Importantly, congenital SETX mutations have been linked to neurodegenerative diseases, and the authors now show that patient-derived cell lines display an augmented antiviral response, an observation that can be recapitulated in a *Setx*-knockout mouse model. These results lend support to the idea that excessive inflammation might contribute to neurological disorders. (*Nat. Immunol.* doi:10.1038/ni.3132, published online 30 March 2015). *AS*