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## BIOCHEMISTRY

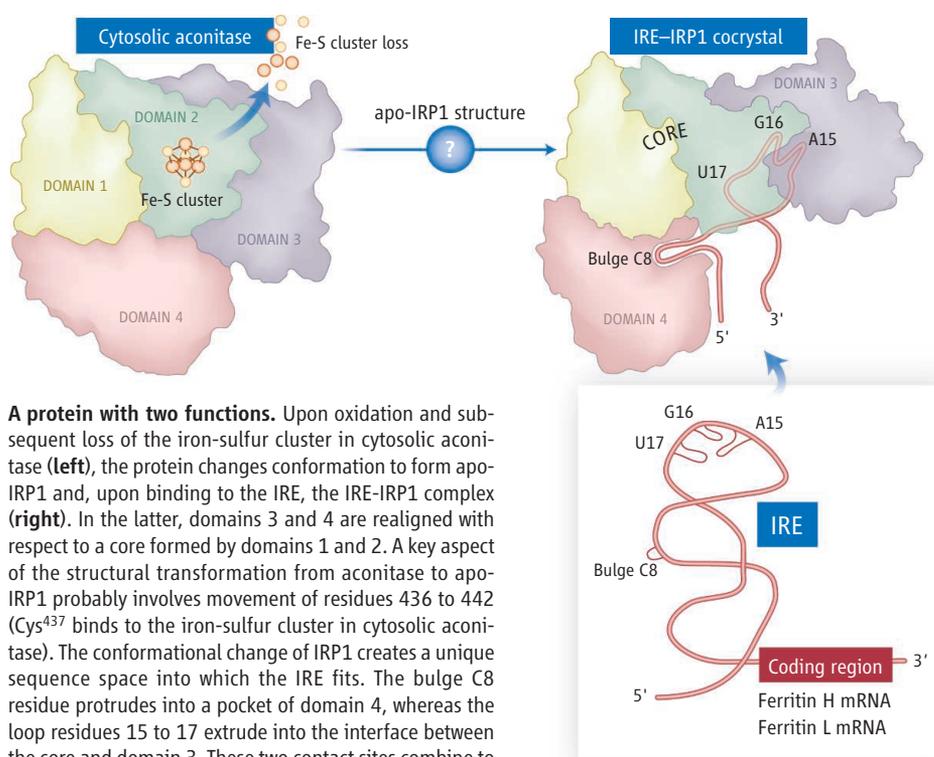
## If the RNA Fits, Use It

Tracey A. Rouault

Cells and organisms use a wide variety of regulatory mechanisms to sense and respond to changes in the extracellular environment. Cells can regulate gene expression at several steps after a gene has been transcribed. On page 1903 of this issue, Walden *et al.* (1) shed light on one such posttranscriptional regulatory mechanism. They show how a single protein—iron regulatory protein 1 (IRP1)—responds to changing conditions by performing two entirely different functions.

In iron-replete cells, IRP1 functions as an aconitase enzyme, which interconverts citrate and isocitrate in the cytosol (2, 3). However, in cells that are iron-depleted and oxidatively stressed, the fragile and exposed iron-sulfur cluster in the aconitase active site disassembles and is lost from the protein. The protein transforms into apo-IRP1, which can bind RNA stem-loops within transcripts of iron metabolism genes known as iron-responsive elements (IREs) (see the figure) (4). Walden *et al.* now report the crystal structure of IRP1 bound to a ferritin IRE. Together with the previous structure of cytosolic aconitase (3), this cocrystal structure reveals how apo-IRP1 binds with high affinity to IREs, whereas cytosolic aconitase does not.

Cytosolic aconitase has four domains. Residues from each domain contribute to the enzymatic active site, including three cysteines that bind to the iron-sulfur cluster. In the IRE-IRP1 cocrystal, domain 4 has moved and rotated relative to its position in cytosolic aconitase (see the figure). Domain 3 has also substantially shifted its position relative



**A protein with two functions.** Upon oxidation and subsequent loss of the iron-sulfur cluster in cytosolic aconitase (left), the protein changes conformation to form apo-IRP1 and, upon binding to the IRE, the IRE-IRP1 complex (right). In the latter, domains 3 and 4 are realigned with respect to a core formed by domains 1 and 2. A key aspect of the structural transformation from aconitase to apo-IRP1 probably involves movement of residues 436 to 442 (Cys<sup>437</sup> binds to the iron-sulfur cluster in cytosolic aconitase). The conformational change of IRP1 creates a unique sequence space into which the IRE fits. The bulge C8 residue protrudes into a pocket of domain 4, whereas the loop residues 15 to 17 extrude into the interface between the core and domain 3. These two contact sites combine to establish specific high-affinity binding of the IRE to IRP1.

to the central core formed by domains 1 and 2, opening up a hydrophilic cavity between the core and domain 3.

Previous structural and mutagenesis studies of IREs, which have conserved structural and sequence elements but are not identical in different transcripts, indicated that the most important residues for high-affinity binding to IRP1 would be the unpaired residues of the terminal loop and an unpaired cytosine that interrupts the double-helical structure of the upper and lower stems. In the cocrystal, these unpaired residues contribute to two discrete binding sites between the IRE and IRP1. In one site, the terminal-loop residues A15, G16,

A crystal structure of RNA bound to the IRP1 protein explains how this protein can perform two entirely different functions.

and U17 interact with residues in the cavity between the core and domain 3. In the second binding site, separated by 1.0 nm from the first, the C8 bulge residue fits into a pocket of domain 4 (see the figure).

The structure of the IRE in the complex is similar to its structure in solution, except that the purine bases of the terminal loop (residues A15 and G16) reorient from a tucked position to extrude into the cavity between the core and domain 3. IRE is thus an ideal binding partner for apo-IRP1, because it can bind to apo-IRP1 while minimally reorganizing its terminal loop.

Apo-IRP1 accumulates in cells that cannot

The author is with the Cell Biology and Metabolism Branch, National Institute of Child Health and Human Development, Bethesda, MD 20892, USA. E-mail: trou@helix.nih.gov

retain an intact iron-sulfur cluster because of iron depletion and/or because of oxidative degradation of the cluster. The conformation of IRP2, a duplicated gene important in regulation, probably mimics that of apo-IRP1, although there are differences in its IRE binding site (5). Similar but not identical IREs found in many different transcripts can be regulated by either IRP1 or IRP2. These transcripts encode proteins involved in iron sequestration, red-cell heme biosynthesis, and iron export. An iron-starved cell would be expected to benefit from repressing translation of these transcripts. IREs are also found in an isoform of the iron importer, divalent metal transporter (DMT1), and transferrin receptor (TfR1), where stabilization of the TfR transcript by IRP binding increases TfR synthesis and iron uptake.

The appearance of nonidentical IREs in transcripts of multiple genes suggests that numerous independent evolutionary selection events created the IRP-IRE posttranscriptional regulatory network in mammalian cells. Because IREs are located in the untranslated

ends of transcripts (see the figure), they can vary without affecting the protein sequence. Therefore, untranslated regions can develop new stem-loops or other features that favorably alter cellular iron conditions. Short RNA sequences such as stem-loops can provide good ligands for a wide variety of targets, in part because their shape and contact sites can vary appreciably, while stable underlying conformations are readily maintained (6). Aconitase arose early in evolution (7), and some researchers postulate that life itself arose in an iron-sulfur world (8), but the IRE-IRP system likely developed later in evolution and matured into an important regulatory system mainly in mammals.

The conformational shift of IRP1 from its cytosolic aconitase form to the apo-conformation thus appears to have created a new sequence space that could be sampled over time by mRNA sequences. mRNAs that contained IREs were repeatedly selected and retained, because by binding to apo-IRP1, they improved the ability of organisms to respond to iron deprivation. A new regulatory

system likely took shape not through creation of new regulatory genes, but by enabling one of the oldest work-horses in the cell—aconitase—to acquire a new regulatory function. In the words of François Jacob, “Evolution does not produce novelties from scratch. It works on what already exists, either transforming a system to give it new functions or combining several systems to produce a more elaborate one” (9).

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## EARTH SCIENCE

# A Submarine Volcano Is Caught in the Act

William W. Chadwick Jr.

In April 2006, Maya Tolstoy, a geophysicist at Columbia University, received some good news and some bad news during a research expedition at sea. The submarine volcano that she and her colleague Felix Waldhauser had been monitoring for years had recently erupted. This was exciting, because only a handful of other deep-sea eruptions have been detected (1), and it was the first time ocean-bottom seismometers were in place during such an event. However, two-thirds of the instruments were stuck in the new lava on the sea floor (see the figure). Would the remaining third yield the data needed to gain new insights into this fundamental but poorly understood geological process?

In the end, the good news outweighed the bad. The instruments that were recovered provided some remarkable results, as Tolstoy *et al.* report on page 1920 of this issue (2). Also, this may only be the first installment in

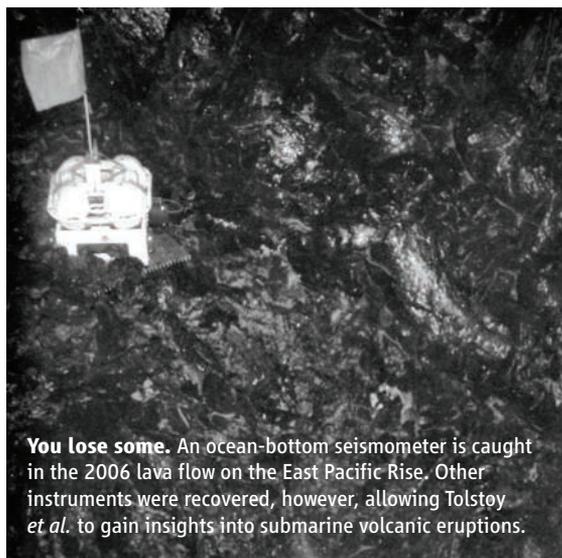
this story, because there is hope that more instruments can be rescued from the sea floor.

The eruption took place on the East Pacific Rise, a huge ridge on the ocean floor west of Mexico where two of Earth's giant tectonic

Ocean-bottom seismometers have recorded the seismic activity associated with a submarine volcanic eruption, revealing important differences from eruptions on land.

plates gradually spread apart. As the plates spread, molten rock rises in the gap to feed periodic eruptions on the sea floor, creating new ocean crust. Three quarters of Earth's volcanism takes place along such seafloor spreading centers, but we know very little about these events. Seismometers on land generally cannot sense them, because they are too far away and the associated earthquakes are too small. Before 1990, not a single eruption was documented on the mid-ocean ridge system, even though many probably occur each year. Since then, swarms of small earthquakes associated with seafloor spreading events have been detected by remote hydrophone arrays (1, 3) and by local networks of ocean-bottom seismometers such as those used by Tolstoy *et al.*

We learn the most about Earth's active processes such as eruptions when we can observe them as they are happening. This is the philoso-



**You lose some.** An ocean-bottom seismometer is caught in the 2006 lava flow on the East Pacific Rise. Other instruments were recovered, however, allowing Tolstoy *et al.* to gain insights into submarine volcanic eruptions.

The author is at Oregon State University/NOAA, Hatfield Marine Science Center, Newport, OR 97365, USA. E-mail: william.w.chadwick@noaa.gov