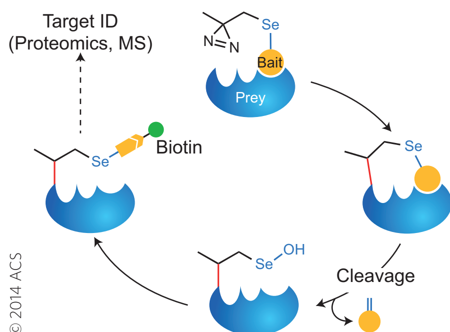
PROTEIN-PROTEIN INTERACTIONS

Bait and switch

J. Am. Chem. Soc. **136,** 11860–11863 (2014).



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Protein–protein interactions regulate many important cellular processes. However, their often transient nature means that identifying which proteins interact with which partners is far from trivial. One approach to identifying protein partners is to start with a known protein as a 'bait' protein, which is then used to covalently label its partner protein(s) — the sought-after, so-called 'prey' proteins. Expression of a bait protein within a cell enables it to form biologically relevant protein–protein interactions with its appropriate protein partners — which can then be labelled. Subsequent enrichment allows the prey proteins to be identified using techniques such as protein mass spectrometry. However, inefficient labelling can lead to important interactions being missed, and the labelling of protein pairs that interact non-specifically can lead to false partners being identified.

A team led by Peng Chen at Peking University, China, have now developed a tool for the crucial labelling step based on an unnatural amino acid. This amino acid, which can be genetically encoded into the bait protein, contains a selenium atom at the γ position and a diazirine group at the end of the side-chain. After the protein–protein interaction forms, the diazirine group can be irradiated with light, which induces crosslinking between the proteins, irrespective of their identity. Subsequently, the unnatural amino acid can be cleaved next to the selenium atom using H_2O_2, which separates the bait and prey proteins, but cleverly the prey protein is left with a selenic acid label. This can then be tagged with a fluorescent dye or with biotin, enabling enrichment and subsequent identification. Splitting the prey proteins from the bait simplifies the separation of the mixture of prey proteins, and also their subsequent identification.

To demonstrate the reliability and effectiveness of their approach the team profiled the binding partners of an *Escherichia coli* acid chaperone HdeA, under conditions of acid stress. Their analysis identified a number of previously known binding partners — showing that this method can reliably label prey proteins — and three new potential binding partners. *RJ*

PROTEIN FOLDING

Flip to unzip

Angew. Chem. Int. Ed. http://doi.org/f2tfb5 (2014)

Collagen, the most abundant mammalian protein, is characterized by a fibrillar

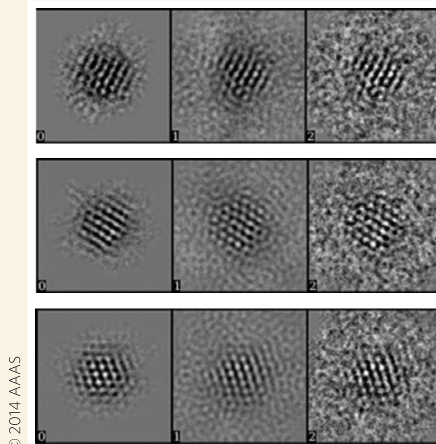
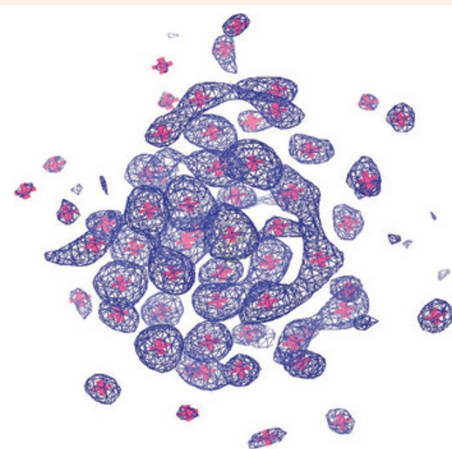
ELECTRON MICROSCOPY

Gold rush

Science **345,** 909–912 (2014)

For many molecules, organic and inorganic alike, X-ray crystallography is arguably the most reliable way to obtain detailed information on the positioning of atoms within the structure. However, crystallography, by definition, requires crystals, which are difficult or even impossible to obtain in many cases. Direct imaging techniques, like transmission electron microscopy (TEM), offer the ability to 'see' individual molecules and assemblies, but rarely do they give a clear atomic-level picture of a given structure

Now, a team made up of researchers from Finland, Japan and the USA, led by Roger Kornberg at Stanford University, have used sophisticated TEM techniques to determine the structure of a gold nanoparticle with atomic resolution. They achieved this without invoking any prior knowledge of the structure, or making any assumptions about the packing of the individual atoms within the cluster. A newly synthesized gold cluster, stabilized by thiolate ligands, was first analysed by mass spectrometry, photoelectron spectroscopy and thermogravimetric analysis to narrow down the number of possible structural formulae prior to imaging.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Subsequently, the team found that direct exposure to the electron beam disturbed the clusters, making imaging impossible, and so they turned to a technique commonly used for soft biological samples. This involved focusing the electron beam at an area adjacent to the cluster and imaging for a very short time in order to lower the dose of electrons. Images of nearly one thousand particles were taken and processed (three of the particle images are pictured, left) and the electron density averaged over all of them to give a map containing 68 peaks (pictured, right, with peaks highlighted in pink) — the number of gold atoms in the cluster. More strikingly, the packing of the atoms did not always fit with the face-centred cubic pattern that is normally associated with gold, demonstrating the power of this technique for elucidating surprising structural insights with atomic precision. *CH*

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