

The cap-snatching endonuclease of influenza virus polymerase resides in the PA subunit

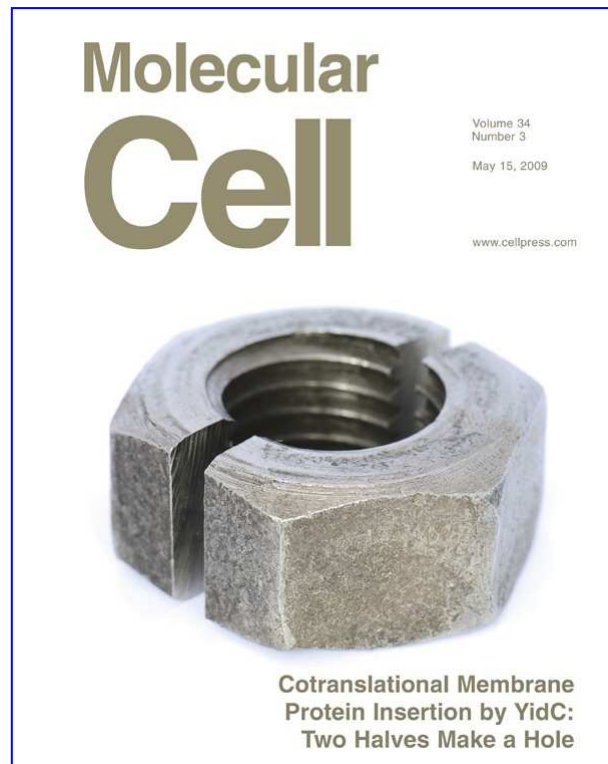
by Dias A, Bouvier D, Crepin T, McCarthy AA, Hart DJ, Baudin F, Cusack S, Ruigrok RW.

[Nature, April 2009 - 458:914-8](#)

The influenza virus polymerase, a heterotrimer composed of three subunits, PA, PB1 and PB2, is responsible for replication and transcription of the eight separate segments of the viral RNA genome in the nuclei of infected cells. The polymerase synthesizes viral messenger RNAs using short capped primers derived from cellular transcripts by a unique 'cap-snatching' mechanism. The PB2 subunit binds the 5' cap of host pre-mRNAs which are subsequently cleaved after 10–13 nucleotides by the viral endonuclease, hitherto thought to reside in the PB2 or PB1 subunits.

Here we describe biochemical and structural studies showing that the amino-terminal 209 residues of the PA subunit contain the endonuclease active site. We show that this domain has intrinsic RNA and DNA endonuclease activity that is strongly activated by manganese ions, matching observations reported for the endonuclease activity of the intact trimeric polymerase. Furthermore, this activity is inhibited by 2,4-dioxo-4-phenylbutanoic acid, a known inhibitor of the influenza endonuclease. The crystal structure of the domain reveals a structural core closely resembling resolvases and type II restriction endonucleases. The active site comprises a histidine and a cluster of three acidic residues, conserved in all influenza viruses, which bind two manganese ions in a configuration similar to other two-metal-dependent endonucleases. Two active site residues have previously been shown to specifically eliminate the polymerase endonuclease activity when mutated. These results will facilitate the optimisation of endonuclease inhibitors as potential new anti-influenza drugs.

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YidC and Oxa1 Form Dimeric Insertion Pores on the Translating Ribosome

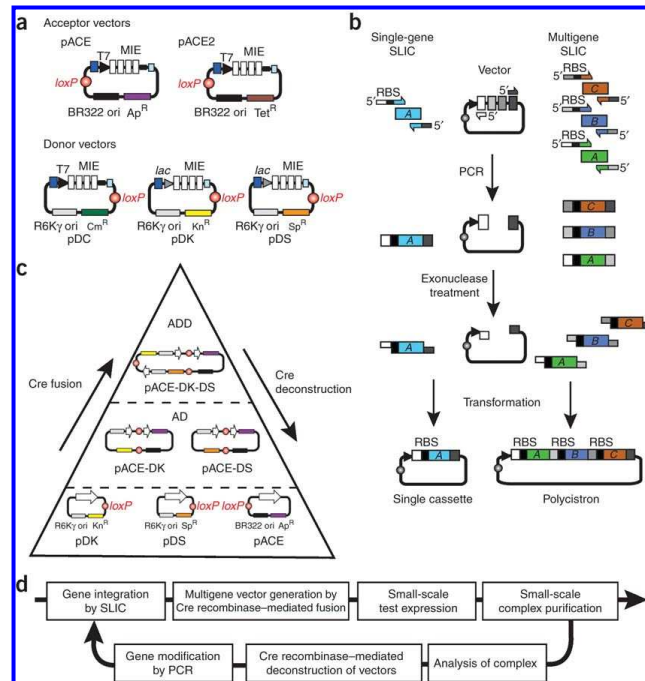
By Rebecca Kohler, Daniel Boehringer, Basil Greber, Rouven Bingel-Erlenmeyer, Ian Collinson, Christiane Schaffitzel, Nenad Ban

► [Molecular Cell, May 2009 – 34 : 344-353.](#)

Members of the YidC/Oxa1/Alb3 protein family insert membrane proteins in bacteria, mitochondria, and chloroplasts. In this issue of Molecular Cell, Kohler et al. (p. 344) report the three-dimensional structures of YidC and Oxa1 bound to translating ribosomes as dimers of two-fold symmetry. The dimeric channel forms a central pore, and it can open laterally to release transmembrane helices into the lipid bilayer, analogous to the functioning of the two halves of the SecY monomer. The image showing a nut cut into halves symbolizes this bipartite assembly. Image: Basil Greber, ETH Zurich.

corresponding authors:

[Christiane Schaffitzel](#) (EMBL/UVHCI - Grenoble) & [Nenad Ban](#) (ETH - Zurich).



Getting a grip on complexes: EMBL scientists develop first fully automated pipeline for multiprotein complex production

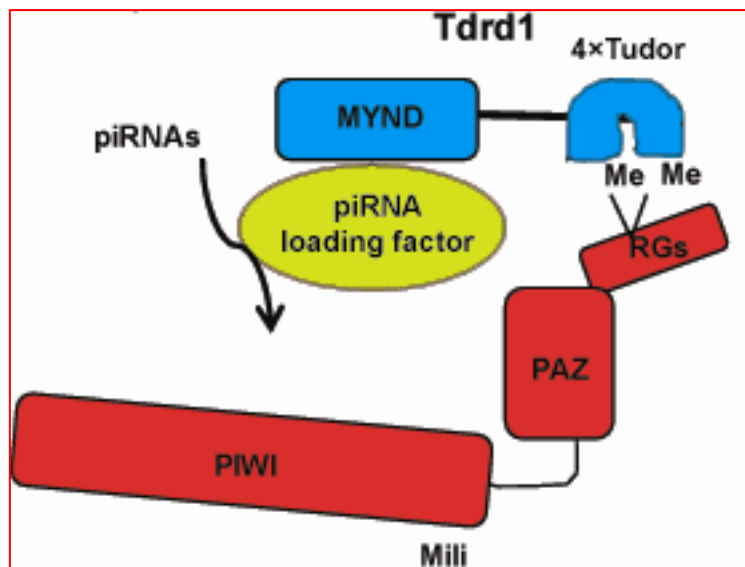
By C. Bieniossek, Y. Nie, D. Frey, N. Olieric, C. Schaffitzel, I. Collinson, C. Romier, P. Berger, T. J Richmond, M. O Steinmetz & I. Berger

Nature Methods May 2009

Most cellular processes are carried out by molecular machines that consist of many interacting proteins. These protein complexes lie at the heart of life science research, but they are notoriously hard to study. Their abundance is often too low to extract them directly from cells and generating them with recombinant methods has been a daunting task. A new technology to produce multiprotein complexes, developed by researchers at the EMBL in Grenoble, France, and the Paul Scherrer Institute [PSI] in Villigen, Switzerland, now makes the biologist's life easier.

Researchers of the groups of Imre Berger (EMBL) and Michel Steinmetz (PSI) describe ACEMBL, the first fully automated pipeline for the production of multiprotein complexes. Requiring much less effort and materials, the new pipeline will speed up structural studies of protein complexes and will allow deciphering as yet elusive molecular mechanisms of health and disease. ACEMBL can produce complexes consisting of different types of components, including protein, RNA and other biomolecules. Currently designed to express proteins in the standard system *Escherichia coli*, the automated pipeline will in future be adapted for complex production in eukaryotic cells. This will allow the study of even larger, more complicated complexes of human origin, including many promising drug targets. The system has already attracted commercial interest.

Corresponding author: Imre Berger (EMBL) & Michel Steinmetz (PSI)



Loss of the Mili-interacting Tudor domain-containing protein-1 activates transposons and alters the Mili-associated small RNA profile

By Michael Reuter, Shinichiro Chuma, Takashi Tanaka, Thomas Franz, Alexander Stark & Ramesh S Pillai.

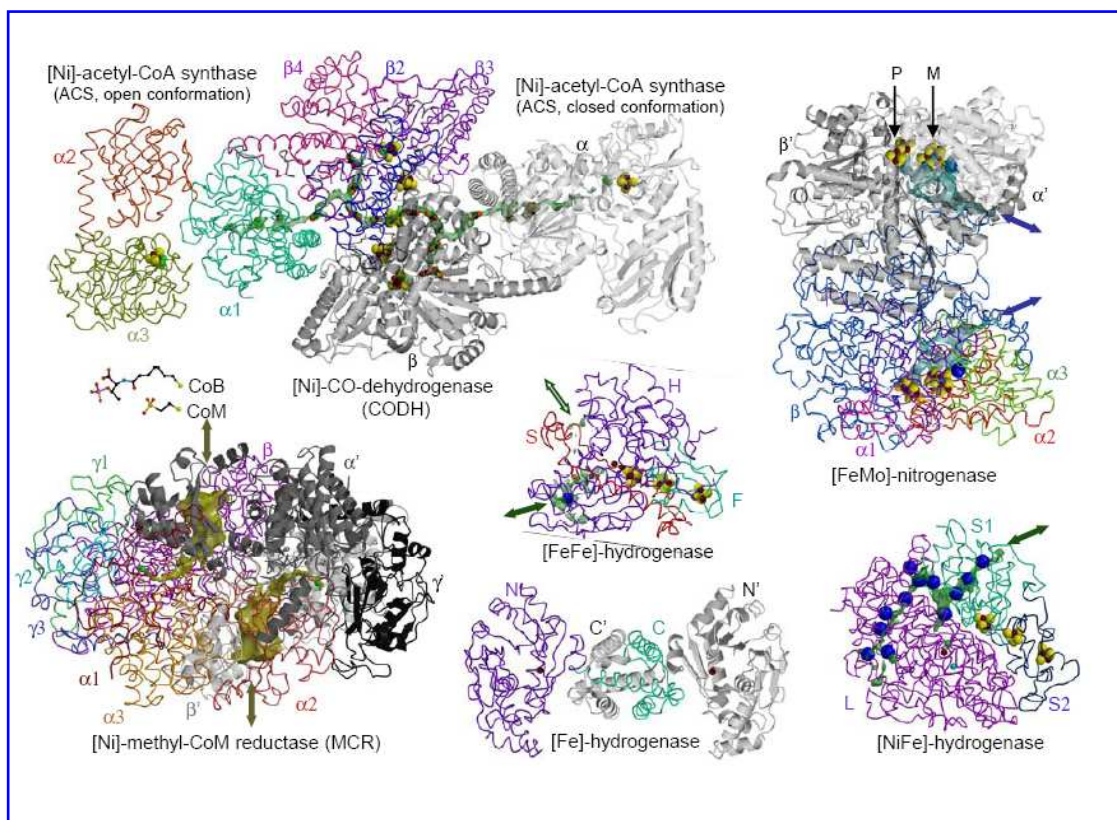
[NATURE STRUCT. & MOL. BIOL. JUNE 2009, 16 page 639](#)

Piwi proteins and their associated Piwi-interacting RNAs (piRNAs) are implicated in transposon silencing in the mouse germ line. There is currently little information on additional proteins in the murine Piwi complex and how they might regulate the entry of transcripts that accumulate as piRNAs in the Piwi ribonucleoprotein (piRNP).

Pillai's group and collaborators isolated Mili-containing complexes from adult mouse testes and identified Tudor domain-containing protein-1 (Tdrd1) as a factor specifically associated with the Mili piRNP throughout spermatogenesis. Complex formation is promoted by the recognition of symmetrically dimethylated arginines at the N terminus of Mili by the tudor domains of Tdrd1. Similar to a Mili mutant, mice lacking Tdrd1 show derepression of L1 transposons accompanied by a loss of DNA methylation at their regulatory elements and delocalization of Miwi2 from the nucleus to the cytoplasm. Finally, they show that Mili piRNPs devoid of Tdrd1 accept the entry of abundant cellular transcripts into the piRNA pathway and accumulate piRNAs with a profile that is drastically different from that of the wild type.

These data suggest that Tdrd1 ensures the entry of correct transcripts into the normal piRNA pool.

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Structure-function relationships of anaerobic gas-processing metalloenzymes.

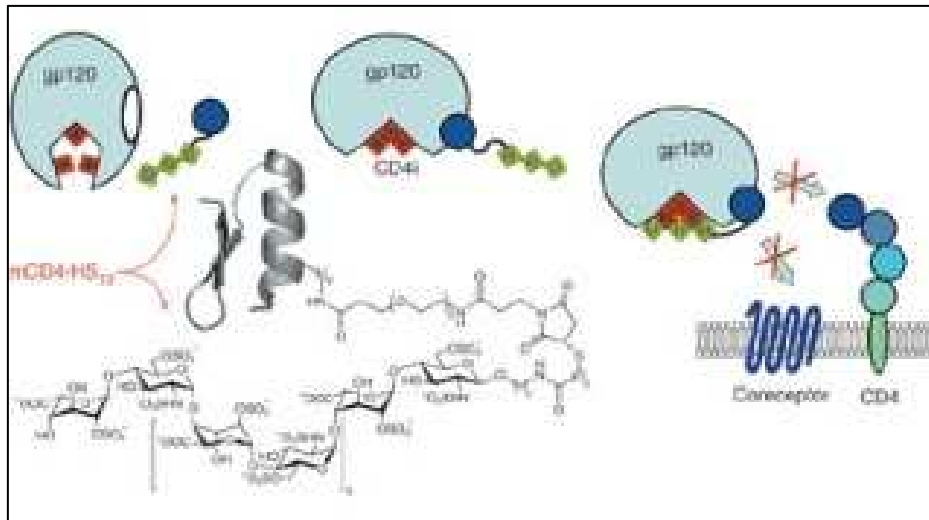
by

Fontecilla-Camps JC, Amara P, Cavazza C, Nicolet Y, Volbeda A.

[Nature 2009 Aug 2009 - 460:814-22.](#)

Reactions involving H₂, N₂, CO, CO₂ and CH₄ are likely to have been central to the origin of life. This is indicated by the active-site structures of the enzymes involved, which are often reminiscent of minerals. Through the combined efforts of protein crystallography, various types of spectroscopy, theoretical calculations and model chemistry, it has been possible to put forward plausible mechanisms for gas-based metabolism by extant microorganisms. Although the reactions are based on metal centres, the protein matrix regulates reactivity and substrate and product trafficking through internal pathways, specific ligation and dielectricity.

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Sugar and peptide combines to fight HIV

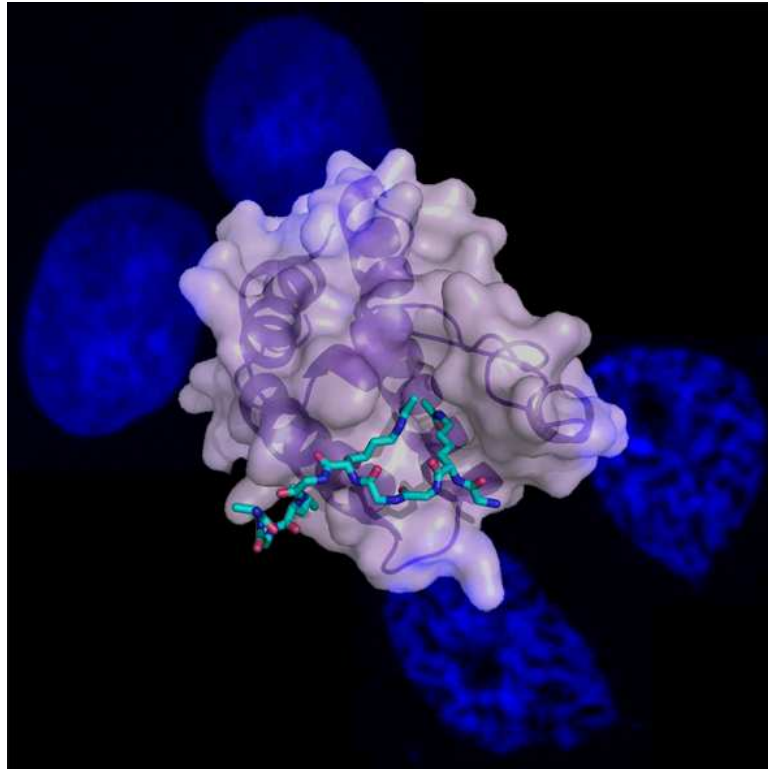
By Baleux F., Loureiro-Morais L., Hersant Y., Clayette P., Arenzana-Seisdedos F., Bonnaffé B. and Lortat-Jacob H.

A synthetic CD4-heparan sulfate glycoconjugate inhibits CCR5 and CXCR4 HIV-1 attachment and entry.

[Nature Chem Biol. SEP 2009](#)

HIV normally enters cells when gp120, a protein on the surface of HIV, binds to receptors on the host cell. In this process, gp120 first binds to one receptor - CD4 - which causes the structure of the protein to change so that it can bind to a second receptor, such as CCR5 or CXCR4. Following several studies showing that this second receptor binding site also interact with heparan sulphate, Hugues Lortat-Jacob and colleagues have now made a molecule that contains a sequence of amino acids to mimic CD4 and a sequence of carbohydrates to mimic heparan sulfate to bind to both sites on gp120. This new strategy results in very effective inhibition of HIV infection in cellular assays, and so may have implications for further antiviral efforts.

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The crystal structure of the bromodomain from Brdt in complex with a diacetylated histone tail. The background shows Brdt causes chromatin, which normally appears diffuse (two nuclei in top left), to clump together and become compact (bottom right).

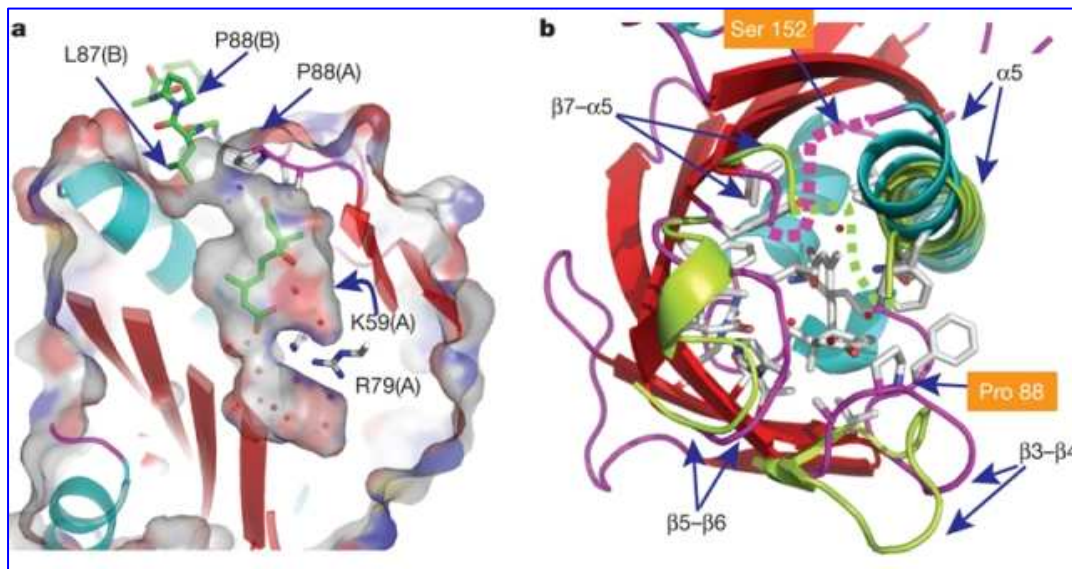
Cooperative binding of two acetylation marks on a histone tail by a single bromodomain.

By Morinière J, Rousseaux S, Steuerwald U, Soler-López M, Curtet S, Vitte AL, Govin J, Gaucher J, Sadoul K, Hart DJ, Krijgsveld J, Khochbin S, Müller CW, Petosa C.

NATURE October 2009

The recognition of histone post-translational modifications by effector modules such as bromodomains is a key step in many chromatin-related processes. Although effector-mediated recognition of single post-translational modifications is well characterized, combinatorial readout of histones bearing multiple modifications is poorly understood. Here, a distinct mechanism of combinatorial readout for the mouse TAF1 homologue Brdt, a testis-specific member of the BET protein family, is reported.

Corresponding author. Christoph Mueller (EMBL)



The abscisic acid receptor PYR1 in complex with abscisic acid

By Julia Santiago, Florine Dupeux, Adam Round, Regina Antoni, Sang-Youl Park, Marc Jamin, Sean R. Cutler, Pedro Luis Rodriguez & José Antonio Márquez.

[Nature](#), 8th November 2009

The plant hormone abscisic acid (ABA) has a central role in coordinating the adaptive response in situations of decreased water availability as well as the regulation of plant growth and development. Recently, a 14-member family of intracellular ABA receptors, named PYR/PYL/RCAR, has been identified. These proteins inhibit in an ABA-dependent manner the activity of a family of key negative regulators of the ABA signalling pathway: the group-A protein phosphatases type 2C (PP2Cs). Here we present the crystal structure of *Arabidopsis thaliana* PYR1, which consists of a dimer in which one of the subunits is bound to ABA. In the ligand-bound subunit, the loops surrounding the entry to the binding cavity fold over the ABA molecule, enclosing it inside, whereas in the empty subunit they form a channel leaving an open access to the cavity, indicating that conformational changes in these loops have a critical role in the stabilization of the hormone–receptor complex. By providing structural details on the ABA-binding pocket, this work paves the way for the development of new small molecules able to activate the plant stress response.

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