the presence of ORF1p. We examine several Ty3 NC mutants to identify the roles of functional regions of the protein in its chaperone activity. This research was supported in part by funding from INSERM and ANRS (France).

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The Codes of "Non-Coding" ncRNA in Epigenetics: Episcription and Hermeneutics of the Genome in the Entangled Cancer-Angiogenesis-Tolerance Epigenome

Josef H. Wissler.
ARCONS Institute for Applied Research & Didactics, Postfach 1327, D-61231 Bad Nauheim, jhw@arcons-research.de, Germany.

OBJECTIVE: The genome as 3D-nucleic acid-[glyco-]protein nanomachine orchestrates translation of genetic [Mendelian] and epigenetic [non-Mendelian] information into phenotypical structural patterns. It is not restricted to transcription *and* translation of DNA scripts into proteins which matter only 2% of transcriptional output. By episcription, same genotypes [e.g. twins] deliver plethoras of not identical epigenetic phenotype variations. Functions of *endogenous RNA* ***isolated*** from total transcriptional output upon cell activation were investigated in vascular remodeling and tolerance phenomena. Structural codes *[3D-episcripts]* for epigenetic phenotyping were found escaping deciphering, so far. ***METHODS:*** Wissler et al, Protides Biol.Fluids **34**:517-536,1986; Materialwiss.Werkstofftech.**32**:984-1008,2001; Ann.N.Y.Acad.Sci.**961**:292-297,2002; **1022**:163-184,2004; **1137**:316-342,2008. ***RESULTS: Functional small ncRNA*** [<200n] were found upon cell activation by extrinsic environmental factors, including mitogenic, cell-mediated immune memory, metabolic [hypoxia] and [physical] shear stress reactions. They comprise conventional models for epigenetic remodeling variations directed rather to proteinaceous gene expression and regulation than genomic DNA sequence changes. Some edited, modified, *redox- and metalloregulated small hairpin* nc-shRNA bioaptamers are ***not complimentary*** to protein-coding transcripts, but feature 3D-episcripts fitting or misfitting to distinct protein conformers. Some address homologous helix-nucleating structural [proteomic] domains, termed **K**/**R**xxx**H** [**K**/**R**3**H**], i.e.-t/s/x**K**/**R**/q/n/hxxx**H**/y/n/q/e/d/r/kx$_{7\text{-}9}$h/xx$_{7\text{-}9}$h/xx$_{5\text{-}20}$**K**/**R**/q/n/e/h- with accessory canonical basic [R/K]$_n$, R/K-zipper, SR/K/RS, EF-hand and/or HxxxH/y/n/q segments, shared in several epigenetic regulator proteins entangled in growth, metabolic syndrome, vascularization, cancer epi- and genetic information indexing of the epigenome. ***CONCLUSIONS:*** Results suggest epigenetic [non-Mendelian] codes consisting of two different associated imprints: [1] Nucleic acid 3D-episcripts which some are not directly retranslatable to protein-coding transcripts. [2] Defined domains in epigenetic regulator protein and nucleic acid matrices as interaction address with [1] comprising all mutational, variant, polymorphism, infectious [viral] and Mendelian disease implications. This suggests tolerated mess-chaotic tumor vascularization associated with *bioaptamer disorders* in ncRNA-switched proteinaceous genetic and epigenetic processes.

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Linking Yeast Transcription Factor Structural Class and Detailed Binding Preferences with in vivo Regulatory Functions

Rachel Patton McCord[1], Cong Zhu[2], Trevor W. Siggers[2], Martha L. Bulyk[2].
[1]Harvard University, Cambridge, MA, USA, [2]Brigham and Women's Hospital and Harvard Medical School, Boston, MA, USA.

Cellular responses to internal and external stimuli through changes in gene expression are in part controlled by the binding of regulatory transcription factors (TFs) to specific sequences of DNA. These TFs belong to a wide variety of DNA binding domain (DBD) structural classes. Sequence specificity is diverse between structural classes, but TFs within each class often have apparently redundant sequence preferences. To understand how cells use these regulators to coordinate networks of responses, it is essential to determine how regulatory function is shared and partitioned between factors of similar and different structures and DNA binding specificities. Are certain structural classes better suited for certain functions? Are detailed differences in binding preferences among apparently similar TFs relevant to *in vivo* function? Using genome-wide datasets, we have described trends in biological function and regulatory mechanisms within TF structural classes in the yeast *Saccharomyces cerevisiae*. These trends suggest general ways in which TF function may be distributed across structural classes according to the biophysical constraints dictated by each DBD structure. Such analyses do not show, however, how specific details of an individual TF's binding specificity might affect its biological function. New data from protein binding microarrays (PBMs) provide such detailed TF binding preference information at all possible 8 base-pair DNA sequences. By combining these PBM data with *in vivo* binding locations measured by chromatin immunoprecipitation (ChIP-chip) experiments, we can infer the functional importance of specific types of TF binding sites (ie low and high affinity sites). This and other analyses made possible by the high resolution PBM data, when combined with observed functional trends in structural classes, will demonstrate how the cell utilizes both general and specific biophysical TF properties to accomplish cellular functions.

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Dissecting the High Rate Constant for the Binding of a Ribotoxin to the Ribosome

Sanbo Qin, Huan-Xiang Zhou.
Florida State University, Tallahassee, FL, USA.

Restrictocin belongs to a family of site-specific ribonucleases that kill cells by inactivating the ribosome. The restrictocin-ribosome binding rate constant was observed to exceed 10^{10} $M^{-1}s^{-1}$ [1]. We have developed a transient-complex theory to model the binding rates of protein-protein and protein-RNA complexes [2, 3]. The theory predicts the rate constant as $k_a = k_{a0} \exp(-U_{el}^*/k_BT)$, where k_{a0} is the basal rate constant for reaching the transient complex, located at the outer boundary of the bound state, by random diffusion, and U_{el}^* is the average electrostatic interaction energy within the transient complex. Here we applied the transient-complex theory to dissect the high restrictocin-ribosome binding rate constant. We found that the binding rate of restrictocin to the isolated sarcin/ricin loop is electrostatically enhanced by ~300-fold, similar to results found in protein-protein and protein-RNA complexes [2, 3]. Ribosome provides an additional 5000-fold rate enhancement. Two mechanisms work together to provide the dramatic additional enhancement. First, with the ribosome the transient complex is formed with relative separations and orientations where local electrostatic interactions with sarcin/ricin loop are particularly favorable. Second, distant parts of the ribosome provides additional electrostatic attraction with the ribotoxin. Our results quantitatively rationalize the experimental results for salt dependences and mutational effects of the binding rates of restrictocin with the isolated sarcin/ricin loop and the ribosome.

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Insight into the Roles of the 140-149 Catalytic Loop and the Zinc-Binding Domain for HIV-1 Integrase Activity

Kevin Carayon, Li Na, Olivier Delelis, Francoise Simon, Jean-Francois Mouscadet, Jean-Francois Mouscadet, Jean-Claude Brochon, **Eric Deprez**.
CNRS, Cachan, France.

Compounds targeting HIV-1 integrase multimerization process represent an interesting approach with likely no cross-resistance with other compounds such as strand-transfer inhibitors (INSTI). To get deeper insight into the role of the zinc-binding domain into the multimerization process, we have studied the effect of a zinc ejector, DIBA-1(2,2′-dithiobisbenzamide-1), on integrase activity. In presence of Mg^{2+}, DIBA significantly decreases the Hill number characterizing integrase-DNA interaction, as measured by fluorescence anisotropy, with only modest effects on the overall affinity, suggesting that zinc ejection primarily perturbs protein-protein interactions and then the nature of the active oligomer. In presence of Mn^{2+}, we found that the DNA-binding of integrase was less cooperative and therefore the Mn^{2+}-dependent 3′-processing is not expected to be influenced by zinc ejection. Nevertheless, the Mn^{2+}-dependent disintegration activity performed by truncated integrase lacking the zinc-binding domain remains sensitive to DIBA-1. One residue, Cys56, was identified as playing an important role for DIBA efficiency in the Mn^{2+} context. In contrast, no effect of the C56 mutation was observed in presence of Mg^{2+}, suggesting that, in this context, DIBA primarily inhibits integrase by a zinc ejection mechanism. A catalytic mutant Q148H was also studied due to its key role in the resistance to Raltegravir, an INSTI currently used in clinic against viruses resistant to other antiretroviral compounds. An additional mutation, G140S, was found to be associated with Q148H in Raltegravir-resistant patients. We found that resistance was mainly due to Q148H as compared to G140S. However, Q148H alone ''freezes'' integrase into a catalytically inactive state. In contrast, the conformational transition from the inactive to the active form is rescued with the double mutation G140S/Q148H. Consequently, when combined together, G140S/Q148H lead to a highly resistant integrase with improved catalytic efficiency.

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Single Molecule Measurements Of The Role Of Tetramer Opening In LacI-mediated DNA Looping

Danielis Rutkauskas[1], **Francesco Vanzi**[1], Hongli Zhan[2], Kathleen S. Matthews[2], Francesco S. Pavone[1].
[1]European Laboratory for Nonlinear Spectroscopy (LENS), Sesto Fiorentino (FI), Italy, [2]Rice University, Houston, TX, USA.