

## 1. Keynote I

Monday, November 5, 2001

09:00 - 10:00

TBA

Al Gilman, University of Texas Southwestern Medical Center and Alliance for Cellular Signaling

## 2. Modeling Signal Transduction

Monday, November 5, 2001

10:20 - 12:15

### (1) Reverse-engineering the gene regulation network of *E. coli*

Uri Alon, Weizmann Institute

### (2) Modeling molecular events in a small volume of living cytoplasm

Dennis Bray, University of Cambridge

In a recent study, we proposed an atomic level structure for a lattice of chemotaxis receptors in coliform bacteria. A unique feature of this model was that it created a small compartment between the plasma membrane and an extended hexagonal lattice of the signaling proteins CheA and CheW. The proposed compartment is 20–30 nm deep, perhaps 300–500 nm wide, and contains a thicket of extended coiled-coils forming the cytoplasmic domains of the chemotactic receptors. The compartment is not closed, and should be freely accessible to cytoplasmic proteins diffusing in from the lateral borders or through 10 nm diameter pores in the hexagonal lattice. Despite the absence of sealed boundaries, however, there is reason to think that this minute volume of bacterial cytoplasm will be highly enriched in two diffusible proteins, CheR and CheB, which are responsible for adaptation in the bacterial system. We are currently using computational methods to explore the possible movement of these enzymes through the “adaptation compartment”. In particular we examined the possibility that these molecules might progress from receptor to receptor by swinging from one flexible region to another, like a monkey swinging through trees (“molecular brachiation”). We also attempted to predict temporal changes in protein conformation within such a molecular lattice. Could conformational changes in one receptor spread to neighboring receptors? If so, by what route and what will be the likely consequences for cellular behavior? Conclusions reached in this analysis are likely to lead to a clearer picture of the physiology of the chemotactic response in bacteria. They will also provide clues to the operation of other “privileged compartments” in both bacteria and eucaryotic cells.

For publications and additional information see <http://www.zoo.cam.ac.uk/comp-cell>.

### (3) Mechanisms of directional sensing in eucaryotic cells

Peter N. Devreotes, Johns Hopkins University

Chemotaxis plays a key role in immune response, wound healing, angiogenesis, and embryogenesis. Research in the last fifteen years in *D. discoideum* has shown that chemoattractants, like many hormones, neurotransmitters, and odorants are sensed by receptors that activate heterotrimeric G-proteins. Our strategy is to use the genetics of *D. discoideum* to discover mechanisms by which cells sense chemical gradients and to apply this information to other eukaryotic cells.

With GFP-tagging, we are studying the dynamic distribution of the receptors, G-protein subunits, and effectors in single living cells during chemotaxis and persistent stimulation. While receptors and G-proteins are uniformly distributed, PH-domains are recruited to the leading edge of cells in shallow gradients of chemoattractant. Thus, the decision for directional sensing is made downstream of the G-protein cycle but upstream of the motile machinery of the cell. The confinement of signaling to the leading edge can be explained in terms of a balance between local excitation and global inhibition processes.

## 3. Software and Theory in Systems Biology

Monday, November 5, 2001

13:45 - 17:00

### (1) Monte Carlo simulation of realistic cellular microphysiology

Joel R. Stiles, Pittsburgh Supercomputing Center, Carnegie Mellon University, and University of Pittsburgh

Biological systems function at widely different scales, and computational studies range from space-filled atomic resolution (e.g., molecular dynamics) to models that are independent of spatial organization (e.g., biochemical network dynamics). At the subcellular-to-cellular level, microphysiological processes involve simultaneous diffusion and reaction of participating molecules, and realistic ultrastructure can sometimes be a critical component of the model. It is increasingly important that simulations delineate *not just a cell's average behavior, but also its variability and propensity to switch between different operating modes and/or to fail*.<sup>1</sup>

Simulations run at atomic resolution are not feasible on microphysiological scales of space (nm to  $\mu\text{m}$ ) and time ( $\mu\text{s}$  to ms), so other methods are required to reduce computation and yet retain essential elements of subcellular topology and molecular interactions. I will discuss the basic theory and design of MCell (Monte Carlo cell) and related software presently under development at the Pittsburgh Supercomputing Center ([www.mcell.psc.edu](http://www.mcell.psc.edu)) and the Salk Institute ([www.mcell.cnl.salk.edu](http://www.mcell.cnl.salk.edu)). With MCell simulations, large-scale 3-D reconstructions (polygon meshes) from serial electron micrographs or electron tomography can be used to represent cell and organelle membranes, and the defined spaces and structures

then can be populated with diffusing and static molecules that interact probabilistically. For *space-independent* unimolecular transitions (e.g., conformational changes), events are decided using methods somewhat similar to those introduced by Gillespie.<sup>2</sup> For *space-dependent* bimolecular associations (e.g., ligand binding), however, MCell's algorithms are a unique and highly optimized integration of Brownian Dynamics diffusion methods and probabilistic testing for second order transitions.<sup>3</sup> Details about individual molecular structure are ignored, but the functional impact of individual molecular positions and densities within realistic subcellular topologies are included explicitly.

While MCell *can* be used with spatially complex models, it does not *have* to be; rather, it can easily be used with molecules distributed uniformly or in simple inhomogeneous arrangements. Moreover, stochastic MCell simulations can be run on biochemical reaction networks both with and without explicit simulation of 3-D diffusion; so spatial *vs.* chemical kinetic contributions to stochastic variability and simulation predictions can be addressed directly. I will illustrate our approach to microphysiological modeling using several examples that increase in scale, realism, and complexity (up to hundreds of serial sections, requiring surface meshes that include millions of polygons and molecules). Examples will show how subcellular topology and molecular organization can have a significant, counter-intuitive impact on signal variability within the clinically important context of synaptic jitter.

Supported by NIH RR-06009, NSF ITR-0086092, and NSF IBN-9603611.

- [1] McAdams, H. H., and Arkin, A. Simulation of prokaryotic genetic circuits. *Annu. Rev. Biophys. Biomol. Struct.*, 27:199–224, 1998.
- [2] Gillespie, D. T. Exact stochastic simulation of coupled chemical reactions. *J. Phys. Chem.*, 81:2340–2361, 1977.
- [3] Stiles, J. R., and Bartol, T. M. Monte Carlo methods for simulating realistic synaptic microphysiology using MCell. In: *Computational Neuroscience: Realistic Modeling for Experimentalists*, ed. De Schutter, E. CRC Press, Boca Raton, pp. 87–127, 2001.

## (2) SBW/SBML: a software platform and standard for systems biology

Hiroaki Kitano<sup>1</sup>, Hamid Bolouri<sup>1,2,3</sup>, Andrew Finney<sup>1,3</sup>, Herbert Sauro<sup>1,3</sup>, Michael Hucka<sup>1,3</sup>, and John Doyle<sup>1,3</sup>

<sup>1</sup>ERATO Kitano Symbiotic Systems Project

<sup>2</sup>University of Hertfordshire

<sup>3</sup>Control and Dynamical Systems, California Institute of Technology

Research on systems biology requires a range of software resources. While some systems biology software, mostly simulators, have been developed, they suffer from the problem of not being compatible with one another. This situation has impeded the development of large-scale models by limiting the availability of tools for a given software environment. The goals of the Systems Biology Markup Language (SBML) and the Systems Biology Workbench (SBW) are to rectify this situation by creating (1) a standard model representation language based on

XML (SBML) that will facilitate model sharing and exchange; and (2) a simple, open-source, application integration framework (SBW) that will allow communication among different simulation/analysis tools. These resources are expected to promote future collaborations in systems biology among both software developers and researchers. In this talk, the philosophy of SBML/SBW will be described along with concrete examples from models of G-protein signaling systems.

## (3) Representing and analyzing biological function with aMAZE, a database of molecular interactions and processes

Shoshana J. Wodak<sup>1,2</sup>, Christian Lemer<sup>2</sup>, Avi Naim<sup>1</sup>, Yong Zhang<sup>1</sup>, Gaurab Mukherjee<sup>1</sup>, Didier Croes<sup>2</sup>, Lorenz Wernisch<sup>3</sup>, David Gilbert<sup>4</sup>, and Jacques van Helden<sup>2</sup>

<sup>1</sup>European Bioinformatics Institute (EBI), Hinxton, Cambridge

<sup>2</sup>Service de Conformation des Macromolécules Biologiques, Université Libre de Bruxelles

<sup>3</sup>Birkbeck College, University of London

<sup>4</sup>Department of Computing, City University

Determining the biological function of a myriad of genes, and understanding how they interact to yield a living cell, is the major challenge of the post genome-sequencing era. The complexity of biological systems is such, that this cannot be envisaged without the help of powerful computer systems capable of representing and analysing the intricate networks of physical and functional interactions between the different cellular components.

Here we present the aMAZE database (<http://www.amaze.ulb.ac.be/>), representing an effort in this direction. Its data model embodies general rules for associating molecules and interactions into large complex networks that can be analysed using graph theory methods. It deals with a large variety of processes, including metabolic pathways, protein-protein interactions, gene regulation, transport and signal transduction. These processes are mapped into their spatial localisation. The ability of representing simultaneously several functional classifications is also provided. A distinct feature of aMAZE is its Object-Oriented, modular and open user interface. Queries are invoked through dedicated modules, data can be linked to external sources, interactively browsed and transferred between modules, and new modules can be readily added. In addition to typical queries, available modules currently include, a custom-built Diagram Editor for the automatic layout, display, and interactive modification of pathway diagrams, and procedures for analysing network graphs. Specialised tools facilitating pathway annotation will be available shortly.

The aMAZE system or others like it, should help the biologists in understanding, analysing, and ultimately modelling, complex cellular networks.

- [1] van Helden, J., Naim, A., Mancuso, R., Eldridge, M., Wernisch, L., Gilbert, D. & Wodak, S. J. Representing and analysing molecular and cellular function using the computer. *Biol. Chem.*, 381(9–10):921–35, 2000.
- [2] van Helden, J., Naim, A., Lemer, C., Mancuso, R., Eldridge, M. & Wodak, S. From molecular activities and processes to biological function. *Briefings in Bioinformatics*, 2(1):98–93, 2001.

#### **(4) Metabolic control analysis: a tool for understanding cellular behaviour, control and regulation**

Jan-Hendrik S. Hofmeyr, University of Stellenbosch

Metabolic control analysis is a powerful quantitative framework for understanding the relationship between the steady-state properties of a (bio)chemical reaction network as a whole and the properties of its component reactions. Although in essence it is a typical sensitivity analysis of a dynamical system, the stoichiometric structure of reaction networks gives it a character of its own. The responses of steady-state fluxes and concentrations to perturbations of the individual steps of the network are quantified as control coefficients, which exhibit useful summation and connectivity properties. Combined, these properties allow control coefficients, which are systemic functions, to be expressed in terms of elasticity coefficients, which are the variable kinetic orders of the individual steps in the system. It will be shown how the analysis of such expressions yields deep insight into how metabolic design, function, control and regulation are interrelated. As a point of departure, the behavior of a typical supply-demand system (i.e., metabolic blocks that produce and consume a product such as ATP or, say, an amino acid) is analyzed by means of rate characteristics, which show how high-level functions such as flux control and homeostatic regulation are distributed over supply and demand. Furthermore, control analysis has been extended to deal with multi-level hierarchical systems in a way that clearly distinguishes between control within the individual levels and the regulatory interactions between levels. This extension will be applied to the metabolic systems used as examples in this lecture. The accompanying paper attempts to capture in a nutshell the detailed derivation from first principles of all of the important theorems of control analysis starting with the general kinetic model of a reaction network.

#### **(5) Alternative designs for a genetic switch: analysis of switching times using the piecewise power-law representation within Biochemical Systems Theory**

Michael A. Savageau, University of Michigan

Genes within developmental circuits are thought to be switched discontinuously ON or OFF in response to developmental stimuli, whereas some other genes are thought to be switched in a continuously variable fashion. We have previously identified criteria that distinguish between discontinuous and continuous genetic switches for an inducible catabolic pathway. According to these criteria, induction via the product of the induced system results in a discontinuous ON-OFF switch with hysteresis, whereas induction via a substrate or intermediate of the induced system results in a continuously variable switch. Thus, genetic systems can exhibit either discontinuous or continuous switch behavior, depending on the circuitry with which the molecular elements of the system are interconnected. These two types of switches exhibit several additional characteristics, beyond their qualitatively distinct behaviors, that influence their natural selection. These characteristics include threshold value, magnitude of the input signal required for switching, magnitude of the corresponding output signal, duty cycle, switching time, and robustness. In order to characterize the biological design principles governing such switches we have developed

mathematical models of generic gene circuits and analyzed their behavior. Here we report the results of a comparative study designed to identify essential differences in switching time and robustness. This study has been greatly facilitated by use of the piecewise power-law representation, which was first developed by systems engineers in the 1940s and adapted for biochemical systems in the early 1970s. With this approach we have been able to derive analytical expressions for switching time and robustness that otherwise, with other forms of nonlinear representation, would require an empirical approach involving computer solution of the differential equations. When the alternative designs are made as nearly equivalent as possible, by the method of Mathematically Controlled Comparison, we find that the switching times for the continuous case are less than that for the corresponding discontinuous case. These results will be discussed in the specific context of the inducible lactose operon of *Escherichia coli*.

## **4. Keynote II**

Tuesday, November 6, 2001

09:00 - 10:00

### **Extracting biological information from genome-wide gene expression studies**

David Botstein, Stanford University

## **5. Experimental Technologies for Systems Biology**

Tuesday, November 6, 2001

10:20 - 12:15

### **(1) Quantitative proteome analysis: new technology and applications**

Ruedi Aebersold, Hookeun Lee, David Han\*, Michael Wright, Huilin Zhou, Tim Griffin, Sam Purvine, David Goodlett

Institute for Systems Biology

\*University of Connecticut

A number of powerful technologies now permit the determination of complete genome sequences as well as the systematic and quantitative measurement of gene expression at the mRNA and protein levels. It is the premise of "functional genomics" technologies that they will significantly contribute to the mechanistic understanding of biological processes, either by themselves, if applied in a discovery mode, or in combination with traditional hypothesis-driven research approaches. Proteomics is a preferred functional genomics technology because its focus is proteins, the most significant class of molecules affecting biological structure, function, and control.

In this presentation we will discuss a new approach to quantitative proteome analysis and show results from selected applications of

the technology to microbial and mammalian cell systems. The technology is based on a new class of chemical reagents termed isotope coded affinity tags (ICAT™) (Gygi S. P., Rist B., Gerber S. A., Turecek F, Gelb M. H., Aebersold R., *Nature Biotechnol* 1999, 17:994–9). The reagents and the tandem mass spectrometry-based analytical process allow the precise quantitation and identification of large numbers of proteins in complex mixtures rapidly and sensitively. The need to separate and analyze extremely complex peptide mixtures challenges the separation sciences. Optimized peptide separation protocols connected on-line with mass spectrometers will be discussed. The applications will document the performance of the method to examine changes in protein profile in yeast cells induced by metabolic shifts, to measure quantitative differences in the cell surface protein profile in mammalian cells, and to detect and quantify changes in protein phosphorylation profiles in cell lysates.

## (2) Quantitative gene expression profiling in the brain

David J. Lockhart, Aventa Biosciences

## (3) Dynamic properties of cellular signaling networks

Tobias Meyer, Stanford University

# 6. Modeling Cellular Physiology

Tuesday, November 6, 2001

13:50 – 16:45

## (1) Modeling cell cycle controls: an example of the “last step” of computational molecular biology

John J. Tyson<sup>1</sup> and Bela Novak<sup>2</sup>

<sup>1</sup> Virginia Polytechnic Institute and State University

<sup>2</sup> Budapest University of Technology and Economics

The fundamental goal of molecular cell biology is to understand cell physiology in terms of the information encoded in the cell's genome. In principle, we know how this information is translated into functional proteins that carry out most of the interesting chores in a living cell. But to make a firm connection between molecular events and cell behavior involves many challenging computational problems at every step along the way. The “last step” of computational molecular biology, from networks of interacting proteins to the physiological responses of a cell to its environment, is an especially challenging problem that has received little attention so far.

A nice example of this challenge is the cell cycle: the sequence of events by which a growing cell duplicates all its components and partitions them more-or-less evenly between two daughter cells. In the last 12 years, molecular biologists have made great progress in identifying the genes, proteins and molecular interactions that control the basic events of the cell cycle (DNA synthesis and mitosis). The control system is so complex that its behavior

cannot be understood by casual, hand-waving arguments. Using biochemical kinetics and dynamical systems theory, my research group converts hypothetical molecular mechanisms of cell cycle control into quantitative computational models that can be analyzed and simulated to predict their behavior. By comparing the simulated behavior of models to the observed behavior of cells, we gain new insights into how the control system works. The approach is generally applicable to any complex gene-protein network that regulates some physiological characteristics of a living cell.

[1] Novak & Tyson, *J. Cell Sci.*, 106:1153–1168, 1993.

[2] Tyson et al., *Trends Biochem. Sci.*, 21:89–96, 1996.

[3] Borisuk & Tyson, *J. Theor. Biol.*, 195:69–85, 1998.

[4] Chen et al., *Molec. Biol. Cell*, 11:369–391, 2000.

## (2) A systems engineering approach to the bacterial cell cycle: defining the genetic circuitry

Lucy Shapiro, Stanford University

## (3) Receptor, ligand, and signaling pathway dynamics in the Epidermal Growth Factor system

Doug A. Lauffenburger, MIT

The Epidermal Growth Factor receptor signaling system is involved in various aspects of tissue development, physiology, and pathology, helping to regulate a spectrum of cell functions including survival, proliferation, and migration. It also exhibits a set of interesting facets that help it serve as an apparent example paradigm for aiding conceptual understanding of a number of other receptor signaling systems, including a multi-receptor family, a multi-ligand family, autocrine/paracrine generation, and spatial compartmentation effects. Thus, it represents an excellent candidate for a quantitative systems-oriented analysis, combining experimental and modeling approaches. In this talk I will present an overview of an expanding range of work we have aimed in this manner at learning about how the EGF system operates, primarily in collaboration with Dr. H. S. Wiley (formerly University of Utah, now Pacific Northwest National Laboratory).

## (4) Understanding what we already know about the living cell: cellular bioinformatics, computational biochemistry, and the Silicon Cell

Hans V. Westerhoff, Boris N. Kholodenko<sup>1</sup>, Christof Francke, Frank Bruggeman, Jan Lankelma, Hans (JJHM) van Beek, Barbara M. Bakker and Jacky L. Snoep<sup>2</sup>

BioCentrum Amsterdam, <sup>1</sup>Thomas Jefferson University, and

<sup>2</sup>University of Stellenbosch

Bioinformatics is directed at obtaining biological understanding from large amounts of molecular biological data. At the structural level, homology between primary structures of proteins may be examined, or by comparison to homologous domains of known structure, structures may be predicted. At the level of cell function, it is the processes more than structures that matter. There Computational Biochemistry calculates process rates, or

control properties from known kinetic data. Calculating non-equilibrium structures in the living cell then extends the topic to Cellular or Integrative Bioinformatics.

It is impossible to predict kinetic properties of an enzyme from its amino-acid sequence, or even from its 3-D structure. The only way to calculate function is to start from empirical enzyme kinetic and thermodynamic data and from physical chemical principles. It is often suggested that it may be impossible to calculate the behavior of the living cell from all its molecular properties. Sure enough errors might accumulate and many kinetic properties are simply unknown. On the other hand we shall here show that to calculate highly important properties of living cells, one only needs a limited amount of information and only some of the enzymes. In addition the living cell is organized in functional modules which should alleviate its analysis.

It is in this manner that we are now beginning to construct *in silico* replicas of living cells, i.e., Silicon Cells. We shall point out why in such an enterprise of Integrative Bioinformatics, quantitative experimentation is a crucial factor. The examples we shall discuss are computational as well as experimental and include sugar and energy metabolism in *S. cerevisiae*, *E. coli* and *T. brucei*.

## 7. Keynote III

Wednesday, November 7, 2001 09:00 - 10:00

### Network bioinformatics: from effect detection to comparative regulation

Adam P. Arkin, Lawrence Berkeley Laboratories and University of California, Berkeley

## 8. Genomics and Systems Biology

Wednesday, November 7, 2001 10:00 - 12:45

### (1) Protein interactions

David Eisenberg, Ioannis Xenarios, Joyce Duan, Lukasz Salwinski, Edward Marcotte, Matteo Pellegrini\*, Michael J. Thompson\*, and Todd Yeates

UCLA-DOE Laboratory of Structural Biology and Molecular Medicine

\*Present address: Protein Pathways

Networks of protein interactions control the lives of cells, yet we are only beginning to appreciate the nature and complexity of these networks. We have taken two approaches to the study of protein networks. The first is to infer functional interactions between pairs of proteins by combining four methods: Rosetta Stone (fused domains), Phylogenetic Profiles (correlated occurrence of pairs of proteins in genomes), Gene Neighbor

(separation of pairs of protein-encoding genes on chromosomes), and analysis of DNA microarray signals. This combination produces networks of protein functional interactions.

The second approach is to reconstruct networks from published studies in the scientific literature. This Database of Interacting Proteins (<http://dip.doe-mbi.ucla.edu>), DIP for short, has now grown to thousands of interactions, and provides a second type of network of cellular protein interactions. The network from DIP is of physically interacting proteins, whereas the network of functional interactions is broader, including information on metabolic interactions.

A new level of information added to the DIP is that pertaining to Protein States given in LiveDIP. Gene-encoded proteins can exist in various states of covalent modification, oligomerization, alternative splicings, and cellular localization. Protein interactions often depend on these states. To describe the interactions realistically, it is necessary to include the states and the transitions between these states.

### (2) Experimental evolution in yeast: a progress report

Jun-Yi Leu, Russell Dorer, Dawn Thompson, Sandra Arciniegas, and Andrew Murray

Harvard University

We have begun to study experimental evolution for two reasons: 1) recent advances in genomics should allow us to perform experiments that in combination with theory can give us fundamental insights into how evolution works, and 2) studying how functional modules change under selective pressure may provide new insights into general principles that govern their structure and function. Specific experiments are being performed to speciate yeast in the laboratory, identify mutations that increase evolvability, and determine the costs and benefits of sex. All of these have given promising preliminary results and we hope that at least one of them will have advanced to presentability!

### (3) Towards a DNA sequence-based *cis*-regulatory network model for a major developmental specification process in the sea urchin embryo

Eric H. Davidson, R. Andrew Cameron, Jonathan P. Rast, Andrew Ransick, Paola Oliveri, Chiou-Hwa Yuh, Cristina Calestani, Veronica Hinman, Takuya Minokawa, Ochan Otim, C. Titus Brown, Carolina Livi, Pei Yun Lee, Peter Clarke, Elbert Branscomb<sup>1</sup>, Leroy Hood<sup>2</sup>, Lee Rowen<sup>2</sup>, Hamid Bolouri<sup>3</sup>, Alistair Rust<sup>3</sup>

Caltech, <sup>1</sup>Joint Genome Institute, <sup>2</sup>Institute for Systems Biology, and <sup>3</sup>University of Hertfordshire

Network models that convey the regulatory logic encoded in the genome are essential to understanding the functional relationships among genes in large developmental systems. Here we describe progress on an interdisciplinary approach to solving the genomic regulatory network underlying specification of the gut and of various mesodermal components in the sea urchin embryo. About 50 genes are at present functionally linked in this network. An initial step was discovery of many previously unknown genes

involved in endomesoderm specification and differentiation by application of novel microarray technology. The architecture of the regulatory network is being established on the basis of computational and experimental results obtained by: (a) functional knockouts and negatively acting forms of regulatory genes followed by determination of effects on many other genes; (b) isolation of BACs from two different species containing the relevant genes, sequence acquisition, and annotation of sequence features; (c) identification of putative *cis*-regulatory elements by computational interspecific sequence comparison; (d) experimental *cis*-regulatory analysis; and (e) determination of spatial changes in gene expression in normal development, and after perturbation by functional gene knockouts. The resulting network model is a work in progress, but it already provides powerful causal insight into this genomically controlled developmental process, which is a major component of the overall embryonic process.

#### (4) Genomics, proteomics, and systems biology

Leroy Hood, Institute for Systems Biology

The Human Genome Project has changed the landscape of biology. A variety of new mantras emerging from this effort are changing how we practice biology and medicine: 1) biology is an informational science; 2) high-throughput biology is creating new databases of information; 3) computer science, mathematics, and statistics are essential for capturing, storing, analyzing, displaying, modeling, and dispersing biological information; and 4) comparative genomics permits one to deduce the logic of life for different organisms and compare them. These all lead to the view that systems approaches will dominate biology and medicine of the 21<sup>st</sup> century. The systems approach will be illustrated with a simple model system, galactose utilization in yeast. The Institute for Systems Biology has been created to exploit and extend the power of systems biology.

## 9. Fresh Perspectives in Systems Biology

Wednesday, November 7, 2001 14:25 - 17:15

#### (1) A mathematical vision of TNF receptor interaction

B. Schoeberl<sup>1</sup>, E. D. Gilles<sup>1</sup>, P. Scheurich<sup>2</sup>, M. Fotin<sup>2</sup>, G. Mueller<sup>2</sup>, and H. Wajant<sup>2</sup>

<sup>1</sup>Max-Planck-Institute for Dynamics of Complex Technical Systems

<sup>2</sup>University of Stuttgart

Tumor necrosis factor (TNF) induces a broad spectrum of cellular responses like differentiation, immune response, or programmed cell death (apoptosis) via a highly complex signaling network. For example, TNF induced signal transduction is controlled by two distinct membrane receptors, TNFR1 and TNFR2, and a variety of intracellular signaling molecules. TNF induced apoptosis is largely attributed to TNFR1. The role of TNFR2 in TNF mediated apoptosis remains less well understood, although a

positive cooperation has been demonstrated. Using in this work an integrated approach of mathematical modeling in combination with experimental data, we have investigated the mechanisms by which TNFR2 might cooperate with TNFR1 induced apoptosis in HeLa cells.

In the case of the antiapoptotic pathway the intracellular adaptor molecule TRAF2 is believed to play a key role in receptor crosstalk. In order to gain a better understanding of the dynamics of TNF signaling and the TNFR1/TNFR2 crosstalk we have developed several mathematical models of the signaling pathways which enabled us to test hypotheses. We find that the apoptotic crosstalk of TNFR1 and TNFR2 does in fact depend on TRAF2 depletion, but must also rely on additional TNFR2 dependent, TRAF2 independent, mechanisms. With the help of mathematical modeling we propose a new regulatory principle of TNF receptor crosstalk based on the adaptor molecule RIP. RIP might play a key role in the TNFR1/TNFR2 crosstalk by regulating the balance between the apoptotic and gene inductive pathway. RIP concentration therefore influences NF- $\kappa$ B activation and thus NF- $\kappa$ B induced antiapoptotic gene products. Among these, c-FLIP and cIAP are of special interest, as these are major regulators of caspase activation.

As it has been shown that mitochondria are involved in the apoptotic pathway in HeLa cells, we have included this pathway into our model. With the help of computational simulation we investigate the possible role of the mitochondrial pathway for caspase activation. Our computational simulations show that the mitochondrial pathway delays caspase activation although the maximum level of activated caspase molecules is increased.

#### (2) Spatial sensing of chemotactic gradients: a reaction-diffusion model

J. Krishnan, P. A. Iglesias, and L. Ma

Johns Hopkins University

This talk analyzes a spatial gradient-sensing model for eukaryotic chemotaxis. The model is in the form of a reaction-diffusion system and comprises two enzymes  $A$  and  $I$ , whose production is mediated by the external signal  $S$ , as well as a response element,  $R$ . This model possesses the property of adaptation to spatially homogeneous signals: the response to spatially homogeneous signals is independent of the concentration of the signal. The response to spatially inhomogeneous signals results in a spatially inhomogeneous response element. We consider the response of the cell in two cases: (a) a disk-shaped cell in 2-D in a constant gradient of chemoattractant, and (b) a spherical cell in 3-D in a constant gradient of chemoattractant. The response of the cell is studied both analytically and numerically.

#### (3) Robustness vs. identifiability of regulatory modules?

J. Stelling and E. D. Gilles, Max-Planck-Institute for Dynamics of Complex Technical Systems

Invariant input-output behavior and pathway redundancy provide a competitive advantage for cellular survival. Therefore robustness of cellular control circuits seems to be a general, essential feature. The same property however provides a

challenge to the investigator because it hinders the estimation of kinetic parameters and thus the quantitative understanding of cellular regulation. The current study investigated the value of mathematical modeling in the elucidation of cellular control circuits under these constraints. A subsystem responsible for mitotic control in budding yeast cell cycle regulation was chosen as an example. On the basis of scarce quantitative experimental data we were able to develop a detailed mechanistic model. The model showed desirable descriptive and predictive character. For instance, model predictions agreed well with experimental observations without additional parameter adaptation. Furthermore, the model allowed for specifying barely characterized regulatory interactions. Determination of parameter estimation accuracy subsequently showed that the information content of the data available was much higher than generally expected. Combined data for wild-type and four variants with only five measured variables lead to acceptable estimates for about half of the 114 kinetic parameters in this complex model. Analysis also proved that the combination of data from the unperturbed and the disturbed control circuit was essential for this outcome. Parameter estimation accuracy additionally gives a quantitative measure for robustness, which in our case strongly supports the concept of highly optimized tolerance, i.e., of the co-existence of robustness and fragility in cellular control. A differentiated view on robustness and identifiability is thus required, but based on our experience, realistic models of cellular control are possible despite limited quantitative data.

#### **(4) Modeling biological responses using gene expression profiling and linear dynamical systems**

Claudia Rangel<sup>1</sup>, [David L. Wild](#)<sup>2</sup>, Francesco Falciani<sup>3</sup>, Zoubin Ghahramani<sup>4</sup>, and Alessia Gaiba<sup>3</sup>

<sup>1</sup>Claremont Graduate University, <sup>2</sup>Keck Graduate Institute, <sup>3</sup>Lorantis Limited, <sup>4</sup>University College, London

Linear dynamical systems are a subclass of dynamic Bayesian networks used for modeling time series data which assume the existence of a hidden state variable, from which we can make noisy measurements, which evolves with Markovian dynamics. We have applied these methods to the analysis of highly replicated gene expression microarray time series data with the intention of building testable hypotheses about the causal influences between gene expression events involved in the activation of human T cells.

#### **(5) Comparison of the small molecule metabolic pathways in *Escherichia coli* and *Saccharomyces cerevisiae*: nonorthologous displacements, gene fusions and protein interactions**

Oliver Jardine and [Sarah A. Teichmann](#), University College, London

We determined the domain structure and protein families of the enzymes in small molecule metabolism in *Escherichia coli* and *Saccharomyces cerevisiae* (yeast) using a combination of structural assignments and sequence comparisons. This allowed us to compare the evolutionary relationships between the proteins in the two organisms to determine the extent of conservation in pathways that are present in both yeast and *E. coli*. Among the 48 pathways and 232 enzymes shared between the two organisms, we identified twelve cases of non-orthologous displacement, where the enzymes carrying out identical functions belonged to entirely different protein families. Among the majority of enzymes which are conserved, we studied whether the subunit composition and gene structure were the same, looking for cases of gene fusions or fissions. We found fourteen cases where there is a multifunctional enzyme in one organism carrying out functions that are catalysed by several proteins in the other organism. Ten of the multifunctional enzymes were in the eukaryote yeast as expected, but another four of these multifunctional enzymes were in *E. coli*. In multifunctional enzymes, one of the advantages is the co-localisation of multiple active sites. In order to gain an insight into the role of protein interactions in metabolic pathways, we analysed the experimental data on protein-protein interactions available for the enzymes in yeast. In agreement with the small number of gene fusions identified in yeast, the extent of physical protein interactions between the enzymes is also limited, and most of the cases observed are between enzymes that are within a few reaction steps of each other with a pathway.

#### **(6) Developmental simulations with Cellerator**

Bruce E. Shapiro and [Eric D. Mjolsness](#), Jet Propulsion Laboratory

We describe how to perform developmental simulations with Cellerator. Biochemical reactions, specified in Cellerator with a compact, arrow-based notation, are automatically translated into the appropriate ordinary differential equations. These reactions can be combined into modules, leading to a natural graph-based hierarchical implementation. We demonstrate how the paradigm of organisms-as-graphs can represent the basic features of developing tissue, and propose a variable-structure graph-based algorithm to describe simple developmental processes. In particular, we show how such a variable-structure system (VSS) can be implemented using a pre-packaged fixed-structure differential equation solver.