Different aspects of mathematical modeling applied to systems biology

9.45 h: Welcome address (Alessandro Verri and Silvio Parodi)

10-11 h: **Boris Kholodenko** (Systems Biology Ireland, University College Dublin)
Signaling ballet in four dimensions

11-11.20 h: Coffee break

11.20-12 h: **Alberto D'Onofrio** (European Institute of Oncology, IFOM, Milan)
The strange case of Dr Bounded and Mr Gaussian: a bistable tale

12-12.40 h: **Annalisa Barla** (Department DISI, Genoa University)
Parameter space exploration within dynamic simulations of signaling networks

12.40-14.00 h: Lunch

14.00-14.40 h: **Marco Antoniotti** (Department of Bioinformatics, University Milan Bicocca)
Colonic Crypts simulations: survey and developments.

14.40-15.20 h: **Barbara Di Camillo** (Information Engineering Department, University of Padova)
Understanding genotype-phenotype interaction through pathway analysis

15.20-16.00 h: **Giuseppe Jurman** (Fondazione Bruno Kessler, Trento)
Spectral distances for (biological) network comparison

16.00 Open discussion: perspectives and challenges

Abstracts

Signalling ballet in four dimensions

Boris N. Kholodenko, Systems Biology Ireland, University College Dublin, Belfield, Dublin, Ireland

Extracellular information received by plasma membrane receptors, such as G-protein coupled receptors (GPCRs) and receptor tyrosine kinases (RTKs), is encoded into complex temporal and spatial patterns of phosphorylation and topological relocation of signaling proteins. Processing and integration of this information through protein kinase cascades leads to important decisions that determine cell's fate. Aberrant information processing by RTKs is a leading cause of many human diseases that range from developmental defects to cancer, chronic inflammatory syndromes and diabetes. We employ computational and experimental approaches to reveal kinetic and molecular factors that control the spatio-temporal dynamics of RTK signaling networks, including transient versus sustained phosphorylation patterns, bistable dynamics and oscillations of the protein phosphorylation state.

Cells have developed mechanisms for precise sensing of the positional information. We show that the spatial separation of opposing reactions in covalent-modification cycles results in the intracellular gradients of protein activities. These gradients provide positional cues for pivotal cellular processes, such as mitosis, motility and migration. The membrane confinement of initiating kinase (e.g., Ras/Raf in the MAPK cascade) and cytosolic localization of phosphatases can result in precipitous spatial gradients of phosphorylated kinases down the cascade, with high concentration near the membrane and low in the perinuclear area. This suggests a need for additional (besides diffusion) mechanisms that facilitate signaling to distant targets. These mechanisms include vesicular and non-vesicular trafficking of phosphorylated kinases driven by molecular motors. Rapid survival signals in neurons might be transmitted by waves of protein phosphorylation emerging in kinase/phosphatase cascades, such as MAPK, PI3K/Akt and GTPase cascades.

Cells respond to countless external cues using a limited repertoire of interconnected signalling pathways. Using modeling and experiments, we unravel how epidermal growth factor (EGF) and heregulin (HRG), induce distinct none-or-all responses of the transcription factor c-Fos by activating the extracellular regulated kinase (ERK) pathway. Although EGF and HRG induce transient versus sustained ERK activation in the cytoplasm, the nuclear ERK activity and the resulting c-fos mRNA expression are transient for both ligands due to induced nuclear dual-specificity phosphatases. Our data demonstrate that the distinct c-Fos responses arise from ligand-dependent, spatiotemporal control of ERK activity emerging from transcriptional negative feedback and cytoplasmic-signalling-to-protein-expression feedforward loops.

In addition to mechanistic modeling, a top-down approach to inferring the structure of cellular signaling and gene networks will be presented. We demonstrate how dynamic connections leading to a particular network node can be retrieved from experimentally measured network responses to perturbations influencing other nodes.

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Parameter space exploration within dynamic simulations of signaling networks

Annalisa Barla, Department of Computer and Information Science, University of Genova

At the scale of biochemical interactions, when dealing with signaling-networks of the size of the MIM (Molecular Interaction Map) in this work, we are clearly over the breaking point of mental intuition of a field expert. The pathways reconstructed in our present MIM are downstream of TGF-β, Wnt and EGF-family growth factors, they include about 60 basic molecular species (proteins and other small molecules).
Our signaling-network comprises about 390 modified species and complexes; it involves more than 800 reactions (reversible and catalytic reactions). To our knowledge, this is probably one of the largest signaling-networks ever simulated at the biochemical-interaction scale level. Our signaling-network reconstructs important molecular controls related to the G0 – G1 transition of cell cycle. Because of the crucial decisional role of this network region, mutations of dominant and recessive onco-proteins (genes) are often found in many cancers, including colorectal cancer. Effects of space compartmentalization (for instance cytoplasmic versus nuclear space) have been mostly ignored at this stage of MIM development. Dynamic network simulations can be considered a crucial support for an *a posteriori* qualitative comprehension of the behavior of a network of this degree of complexity. In order to explore robustness / sensitivity to perturbations of the network, we perturbed one or a small number of species at a time, observing the effects induced on the remaining species. We introduced combinations of 10x and 10/

perturbations in multiple species (up to five), for about 60 total molecular concentrations. Perturbation of rates is not considered in this presentation. Perturbed species were obviously never coincident with perturbing species. When single species or pairs act as perturbator molecules, the expected wall-clock time required to evaluate the ODEs varies from few min to several hours (using a standard desktop PC - 3.00 GHz, CPU, 4.00GB RAM). For three perturbing species several days are required. Indeed, when considering more than three perturbing species, the problem becomes quickly intractable on a single standard desktop computer.
To improve our strategy we devised a targeted quasi-random sampling procedure capable of a more efficient exploration of the parameter space. The underlying idea is to make explicit use of the available knowledge regarding the network structure. We implemented perturbations of the concentrations concerning about 60 consumable basic molecular species following a random exploration of the parameter space.
We obtained significant indications about sensitivity/robustness of the network described in our MIM. We were able to note an interesting feature characterizing the network: the effect of a perturbing species becomes weaker and weaker for perturbed molecules that are positioned at increasing distances from the perturbing molecules.

Understanding genotype-phenotype interaction through pathway analysis
Barbara Di Camillo, Information Engineering Department, University of Padova

Genome Wide Association Studies (GWAS) analyze all or most of the genes of different individuals to see how much the genes vary from individual to individual. Different variations are then associated with different quantitative traits, such as diseases. Although these studies have successfully identified a number of significant SNP-disease associations, they were able to explain only a small fraction of disease hereditability. One of the reasons is that complex pathologies, such as diabetes are indeed heterogeneous and multi-causal, as a result of the alteration of multiple regulatory pathways and of the interplay between different genes and the environment, rather than referable to a single dysfunctional gene like in monogenic diseases. In this context, identification of functional pathways underlying the observed phenotype is a major challenge in Systems Biology.
We have developed a method to identify pathways associated to the disease in GWAS, based on the Entropy concept. In particular, Joint Entropy between pairs of SNPs belonging to the same functional pathway is calculated and the difference in cases vs control is calculated and averaged across the pairs of genes in the pathway. This allows ranking pathways based on how widely the mapped SNPs are associated to the disease. Our approach is sensitive enough to identify rare variants affecting different SNPs associated to the same pathway.
As a proof of concept we have applied our method to the WTCCC type 1 and 2 diabetes GWAS dataset.