Bioengineering and Systems Biology

Article *in* Annals of Biomedical Engineering · August 2006

DOI: 10.1007/s10439-006-9119-3 · Source: PubMed

3 authors, including:

Raimond L Winslow
Johns Hopkins University
243 PUBLICATIONS **9,853** CITATIONS

Annals of Biomedical Engineering, Vol. 34, No. 2, February 2006 *(© 2006)* pp. 257–264
DOI: 10.1007/s10439-005-9047-7

Bioengineering and Systems Biology

TREY IDEKER,[1] L. RAIMOND WINSLOW,[2] and A. DOUGLAS LAUFFENBURGER[3]

[1]Department of Bioengineering, University of California at San Diego; [2]Department of Biomedical Engineering, The Johns Hopkins University, and [3]Biological Engineering Division, Massachusetts Institute of Technology

(Received 11 April 2005; accepted 23 November 2005; published online: 11 February 2006)

A field known as Systems Biology is emerging, from roots in the molecular biology and genomic biology revolutions—the succession of which has led biomedical scientists to recognize that living systems can be studied not only in terms of their mechanistic, molecular-level components but also in terms of many of them simultaneously. This prospect of understanding how biological entities function through the framework of integrated operation of component parts holds extraordinary promise for medical applications, as well for broader societal applications such as the environment, agriculture, materials/manufacturing, and national defense.

Some definitions of Systems Biology are available. Ideker *et al.*[22] suggest the following: "Systems Biology does not investigate individual genes or proteins one at a time, as has been the highly successful mode of biology for the past 30 years. Rather, it investigates the behavior and relationships of all the elements in a particular biological system while it is functioning." A description by Kitano[27] is that "To understand biology at the system level, we must examine the structure and dynamics of cellular and organismal function, rather than the characteristics of isolated parts of a cell or organism." The National Institute of General Medical Sciences at NIH[1] provides a slightly different perspective: "Systems Biology seeks to predict the quantitative behavior of an *in vivo* biological process under realistic perturbation, where the quantitative treatment derives its power from explicit inclusion of the process components, their interactions, and realistic values for their concentrations, locations, and local states."

Systems Biology can also be defined operationally, as by the MIT Computational & Systems Biology Initiative, in terms of the "4 M's"—Measurement, Mining, Modeling, and Manipulation—illustrated schematically in Fig. 1 (see http://csbi.mit.edu/). In this post-genomic era, Measurement can be undertaken in a high-throughput, multivariate manner using various kinds of array technologies. Because this multivariate data then is relatively recalcitrant to hypothesis generation by means of unaided human intuition, computational algorithms for Mining the data to generate hypotheses concerning the potential interpretation of these data sets is necessary. In order to consequently develop new predictions for experimental test (or design), computational Modeling is required for similar reason: unaided human intuition likely cannot produce effective predictions concerning complex, interconnected, nonlinear molecular systems. Finally, in order to test those model predictions or create a new technology or product, molecular-level manipulation is needed, employing genetic, biochemical, or materials interventions. Thus, Systems Biology involves a multivariate approach comprising topological and dynamical properties and aimed ultimately at quantitative prediction, for basic scientific understanding or technological design. It must be noted that the complexity of living systems does not reside solely in the number of components and interactions treated, nor in their associated structural and physicochemical properties, but also in the hierarchical connection across space and time scales from gene-level to cell-level to tissue-level to organism-level to population-level (see Fig. 2).

It must be emphasized that Systems Biology is not merely a contemporary manifestation of traditional bioengineering, despite the similarity of the "4 M's" approach to engineering perspective. The crucial difference is that the kinds of measurement and manipulation in modern Systems Biology is at the molecular level, and the data sets being generated and considered are highly-multivariate because of the existence now of high-throughput experimental assays at the genomic and proteomic levels. Systems Biology is aimed at true molecular and cellular mechanism underlying operation of biological systems, rather than phenomenological description to which higher levels of organization (e.g., tissue, organ, and organism) are restricted. Thus, we specifically and categorically restrict our definition of Systems Biology to require molecular-level information. Moreover, we emphasize that Bioengineering does not uniquely encompass this new field, but rather is one of the key disciplines required along with various others (e.g., molecular/cell biology, genetics, biochemistry, mathematics, and computer science) to move it forward.

Address correspondence to A. Douglas Lauffenburger, Biological Engineering Division, Massachusetts Institute of Technology

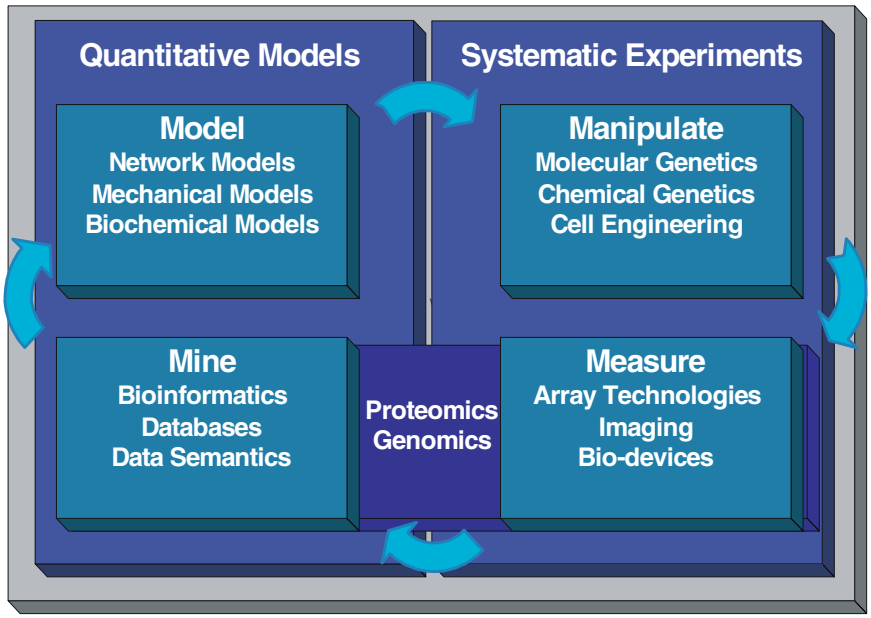

FIGURE 1. Operational definition of Systems Biology in terms of the 4 M's: measurement, mining, modeling, manipulation (see http://csbi.mit.edu/).

As evidence of the enormous impact systems thinking has had on biology, consider that in the last 3 years it has led to an explosion of new research institutes, companies, conferences, and academic departments, all having the words "systems biology" in the title or mission statement. Several journals are now either entirely devoted to reporting systems biology research or are sponsoring regular sections devoted to current issues in systems or computational biology. And under the leadership of Elias Zerhouni, the National Institutes of Health released a "roadmap" for 21st century medicine that includes interdisciplinary science and integrative systems biology as core focus areas.[47]

STATE OF THE ART IN SYSTEMS BIOLOGY RESEARCH

Gene-Protein-Metabolite Networks

One of the most exciting trends in modern biology involves the use of high-throughput genomic, proteomic, and metabolomic technologies to construct models of complex biological systems and diseases. While the notion of systems science has existed for some time,[2,5] these approaches have recently become far more powerful due to a host of new "omic" technologies that are high-throughput, quantitative, and large scale.[50] These technologies typically depend on knowing the complete DNA sequences in the organism's

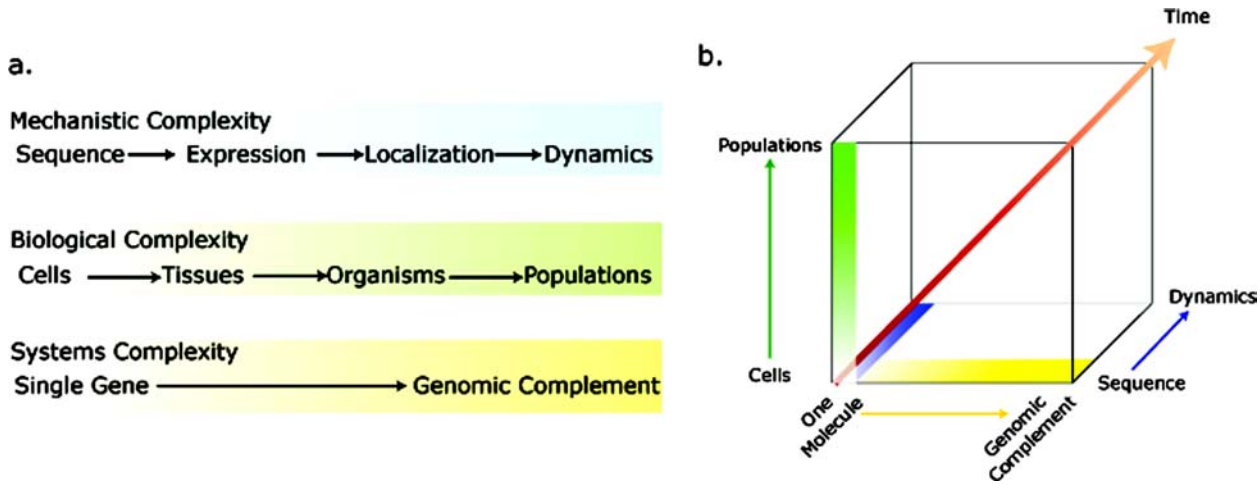

FIGURE 2. Illustration of the multiple dimensions biological systems complexity.

genome. For instance, DNA microarrays involve spotting thousands of these gene sequences on a solid substrate to bind and detect the complementary RNAs. Global changes in RNA expression can be measured with DNA microarrays,[10] and networks can be inferred in terms of gene expression effects (e.g., Lee *et al.*[30]; Gardner *et al.*[14]). Another group of technologies gives us insight into how these molecules interact with one another to form a large and complex intracellular network. For instance, procedures such as yeast two-hybrid or chromatin immunoprecipitation are being applied systematically to screen for "all" the protein–protein or protein–DNA interactions that occur in a cell at a particular condition or point in time. The resulting network of interactions yields information on how the cell transmits information in response to stimuli and dynamically forms the molecular machines required for life (e.g., Bar-Joseph *et al.*[4]; Haugen *et al.*[21]; Said *et al.*[39]). The more technically-challenging quantitative measurement of changes in protein abundance, protein phosphorylation state, and metabolite concentrations is also advancing with protein arrays, mass spectrometry, and NMR among other sophisticated techniques (e.g., Gygi *et al.*[20]; Zhou *et al.*[49], Griffin *et al.*[17]; Nielsen *et al.*[33]; Zhang *et al.*[48]). A crucial conceptual point that is becoming evident is that the most effective Systems Biology studies will incorporate data from heterogeneous assays, enabling greater depth of penetration into network operation (e.g., Griffin *et al.*[18]; Gaudet *et al.*[15]).

Cell Engineering

The point of these biomolecular machines and networks, of course, is to carry out and regulate cell behavioral functions such as metabolism, proliferation, death, differentiation, and migration. Because of the complexity of these processes, a Systems Biology perspective may be anticipated to be productively applicable to understanding of how they are governed by the constituent molecular properties and interactions. This area of endeavor, termed *cell engineering*, has a long-standing history in Bioengineering and its importance should only grow vigorously as the experimental measurement capacity and throughput accelerates in the omics era. There is a good present foundation in development of useful approaches to quantitative understanding of the operation of metabolic pathways and signaling networks at the molecular level (e.g., Price *et al.*[36]; Levchenko[31]; Gilman and Arkin[16]; Asthagiri and Lauffenburger[3]; Lauffenburger and Linderman[29]). It will be necessary to connect gene-level transcriptional networks with protein-level posttranscriptional networks (Harbison *et al.*, 2004), and the interplay of these two is certainly dynamic and two-way in nature (Alm and Arkin, 2003). A less well-resolved issue, however, is to develop models for understanding how the operation of these molecular pathways and networks relate to the cell-level behavioral functions they underlie and regulate. This task raises exceptional difficulties because the connections between regulatory signaling pathways and downstream functional mechanisms are poorly identified at this point in time. Accordingly, in the near-term relational models are likely to be especially productive in relating molecular-level signals to cell-level behavioral responses (e.g., Janes *et al.*[25]; Sachs *et al.*[38]).

Integrative Systems Physiology

A further imperative challenge is to understand how behavior at the level of genome, proteome and metabolome determines physiological function at the level of not only cells but tissues and organs. Because of the inherent complexity of real biological systems, the development and analysis of highly integrative computational models based directly on multiscale experimental data is necessary to achieve this understanding. We refer to this model-based approach as *integrative systems physiology*.

In addition to contributing to our basic understanding of subcellular function, application of high-throughput experimental technologies to identification of the cause, diagnosis and possible treatment of human illness will have a profound impact on the conduct of basic medical research. While currently in a nascent stage, it will soon be common for clinical research studies to collect genetic, transcriptional, proteomic, multimodal imaging and clinical data from every patient in large, carefully selected cohorts sharing a specific disease diagnosis. The first such studies directed at cardiovascular disease and cancer are in fact already underway. The goal will be to use these multiscale biomedical data sets to uncover novel insights regarding disease mechanisms across hierarchical levels of biological organization, to identify biological markers which correlate with different disease states and interindividual differences in disease risk and to suggest more effective therapeutics targeted to meet the needs of the individual. As our knowledge advances, there is no question but that integrative computational models of biological systems will become an intrinsic part of the decision making process in clinical research, diagnosis and treatment, ushering in an era of *computational medicine*.

A notable example of integrative modeling spanning from the level of molecular function to that of tissue and organ, with applications to physiological function in both health and disease, is modeling of the heart. The first models of the cardiac action potential (AP) were developed shortly after the Hodgkin-Huxley model of the squid AP and were formulated in order to explain the experimental observation, that unlike neuronal APs, cardiac APs exhibit a long duration plateau phase.[34] Over subsequent years, these models have been extended and now describe properties of voltage-gated membrane currents and transport and exchange processes regulating intracellular ion concentrations,[32] mechanisms of calcium-induced calcium-release,[24]

cross-bridge cycling and force generation,[37] mitochondrial ATP production and its regulation,[9] and β-adrenergic signaling pathways and their actions on target proteins.[40] Models have now been developed for canine, guinea pig, human and rabbit ventricular myoyctes, sino-atrial node cells, and atrial myocytes. These cells models have been integrated into large-scale, biophysically and anatomically detailed models of electrical conduction in the cardiac ventricles which have been used to investigate the molecular basis of life-threatening arrhythmias.[41,46] Such model development is not limited to the heart. As an additional example, anatomically-based models and fluid dynamics simulation of airflow in the lungs and bronchial tree are under development and approaches to physiologically-based modeling of respiratory function have been proposed to investigate transport phenomena and particle distribution within the airways. It is clear that integrative modeling of physiological systems will continue to develop over time, encompassing an expanding range of cell types as well as tissues and organs.

PROSPECTS FOR BIOENGINEERING CONTRIBUTIONS

Since systems biology depends so strongly on the interplay between new technology and basic biological science, Bioengineering cannot help but play a central role. New technology development will be crucial across several areas. First, "better, faster, cheaper" methods will be needed for characterizing molecules and molecular interactions. For instance, although current technology led to sequencing of a single "Human Genome", we are still quite far from the day in which every patient's genome can be sequenced and analyzed. Second, new computational methods are needed to integrate and analyze all of the genomic and post-genomic data, and here the technological gap is even bigger. New data sets are being generated at a rate that far outpaces our ability to interpret the results. To address this challenge, mathematical, computer-aided models will be needed to organize all of the global measurements at different levels into models of cells and tissues. Bioengineering will undoubtedly play a strong role in both areas, just as it has in the past.

Similarly, there are multiple ways in which research in biomedical engineering will drive the disciplines of integrative systems physiology and computational medicine. The first is in development of novel technologies and the improvement of existing technologies for collection of data on physiological function. The challenge is that while rapid advances are being made in development of new technologies for analysis of the genome, proteome and metabolome, methods for investigating the physiological function of cells, tissue and organ remain, for the most part, notoriously low-throughput. The emerging disciplines of micro and nanofabrication as applied to "laboratory on a chip" technologies will have significant impact in key areas of physiological data collection (e.g., Burns *et al.*[7]; Fritz *et al.*[13]; Savran *et al.*[42]; Wang *et al.*[44]). The first generation of high-throughput whole-cell assays for studying gene- and protein-level properties on cell physiological function are becoming available.[35,45] A next crucial challenge is to extend these kinds of high-throughput assay techniques to tissue physiology *in vitro*, likely by engineering tissue surrogates not for medical implants but for basic biology as well as pharmacology/toxicology purposes.[19,43] Powerful computational analysis methods aimed at predicting effects of molecular therapeutics are becoming available (e.g., Kunkel *et al.*[28]; diBernardo *et al.*[11]), so their application to the most effective experimental systems will provide a crucial synergy. Continued development of novel molecular imaging technologies for high-spatial resolution mapping of peptide and protein distributions in tissue samples, magnetic resonance imaging methods for mapping ion concentrations and metabolite levels in living tissue and organs and fluorescence resonance energy transfer (FRET) and related methods for measuring protein interactions in living systems—both *in vitro* and *in vivo*—will also be important.[6] Magnetic resonance and tomographic imaging systems are now being used for reconstruction of tissue and organ geometry and micro-anatomic structure, but continued development of these technologies to increase speed of data acquisition and spatial resolution, and especially to enhance capabilities for specific molecular-level imaging[26] will be vital to the development and application of quantitative models of physiological function.

Finally, in order to confront problems in integrative systems physiology and computational medicine, it will be necessary to develop novel methods for representing, storing, and querying heterogeneous multiscale experimental and simulation data sets and model descriptions. Those who take on this task will need a broad understanding of biology, principles of informatics and modern approaches to computational modeling of biological systems. This will indeed be among the most challenging and exciting tasks confronting biomedical engineers as we move forward.

EDUCATIONAL PROGRAMS

Not surprisingly for such a high-visibility field, Systems Biology has spurred interest from myriad researchers, some just starting their careers, others well established but looking for a "piece of the action". So, what is the best plan for students interested in a career in Systems Biology? Because of the need to couple computational analysis techniques with systematic biological experimentation, more and more universities are offering Ph.D. programs that integrate both computational and biological subject matter.

Because Bioengineering lies at the interface of these two disciplines, it is poised to play a strong, and very possibly dominant, role in systems biology education. A number

of graduate-level programs in systems biology are already affiliated with Bioengineering (Table 1). Several of these programs, such as the Computational & Systems Biology Initiative at MIT, include "systems biology" directly in the name but include in its core curriculum several Bioengineering subjects. Others, such as the Systems Biology syllabus within Bioengineering at UCSD, are significant courses of study offered from within a Department of Bioengineering. A number of institutions outside of Bioengineering also offer significant programs of study, such as Harvard Medical School, the Institute for Systems Biology, Oxford University, and Biocentrum Amsterdam.

Given the pace of the field, it is certainly too early to endorse a particular syllabus as the correct or best option. However, the study of Systems Biology must lead to a rigorous understanding of both experimental biology and quantitative modeling. Programs might require that all students, regardless of background, perform hands-on research in both computer programming and in the wet laboratory. Required coursework in biology typically includes genetics, biochemistry, molecular and cell biology, with lab work associated with each of these. Coursework in quantitative modeling might include probability, statistics, information theory, numerical optimization, artificial intelligence and machine learning, graph and network theory, and nonlinear dynamics. Of the biological coursework, genetics is particularly important, because the logic of genetics is, to a large degree, the logic of systems biology. Of the coursework in quantitative modeling, graph theory and machine-learning techniques are of particular interest, because systems approaches often reduce cellular function to a search on a network of biological components and interactions.[12,23] A course of study integrating life and quantitative sciences helps students to appreciate the practical constraints imposed by experimental biology and to effectively tailor research to the needs of the laboratory biologist. At the same time, knowledge of the major algorithmic techniques for analysis of biological systems will be crucial for making sense of the data.

An alternative to pursuing a cross-disciplinary program is to tackle one field initially and then learn another in graduate school. Examples would include choosing an undergraduate major in bioengineering and then obtaining a Ph.D. in molecular biology, or starting within biochemistry then pursuing graduate coursework in bioengineering and systems biology. This leads to a common question: when contemplating a transition, is it better to switch from quantitative sciences to biology or vice versa? Although some feel that it is easier to move from engineering into biology, the honest answer is that either trajectory can work. Some practical advice is that if coming from biology, it is best to start by becoming familiar with Unix, Perl, and Java before diving into more complex computational methodologies. If coming from the quantitative sciences, an effective strategy is to jump into a wet laboratory as soon as possible.

TABLE 1. Selected programs in systems biology programs highlighted in red involve strong participation of Bioengineering faculty.

(a) Graduate Programs w/ Sys. Bio. Courses
- Europe
  - Flanders and Ghent University
    - Department of Plant Systems Biology
    - http://www.psb.ugent.be/
  - Max Planck Institutes
    - Inst. of Molecular Genetics
    - Inst. of Dynamics of Complex Systems
    - http://lectures.molgen.mpg.de/
    - http://www.mpi-magdeburg.mpg.de/
  - University of Rostock
    - Systems Biology & Bioinf. Program
    - http://www.sbi.uni-rostock.de
  - University of Stuttgart
    - Systems Biology Group
    - http://www.sysbio.de/
- Asia
  - A*Star Bioinformatics Institute, Singapore
    - http://www.bii.a-star.edu.sg/
  - University of Tokyo
    - Graduate School of Information Science and Technology
    - http://www.i.u-tokyo.ac.jp/index-e.htm
- North America
  - Cornell, Sloan-Kettering, and Rockefeller Universities
    - Physiology, Biophysics & Systems Biology
    - Program in Comp. Biology and Medicine
    - http://www.cs.cornell.edu/grad/cbm/
    - http://biomedsci.cornell.edu
  - Massachusetts Inst. of Technology
    - Div. of Biological Engineering, Computational and Systems Biology Initiative (CSBi)
    - http://csbi.mit.edu/
  - Princeton University
    - Lewis-Sigler Inst. for Integrative Genomics
    - http://www.genomics.princeton.edu
  - Stanford University
    - Medical Informatics (SMI) and BioX
    - http://smi-web.stanford.edu/
  - U. C. Berkeley
    - Graduate Group in Comp. & Genomic Biology
    - http://cb.berkeley.edu/
  - U. C. San Diego
    - Dept. of Bioengineering
    - http://www-bioeng.ucsd.edu/
  - University of Toronto
    - Program in Proteomics and Bioinformatics
    - http://www.utoronto.ca/medicalgenetics/
  - University of Washington
    - Dept. of Bioengineering, Dept. of Genome Sciences
    - http://www.gs.washington.edu/
  - Virginia Tech
    - Program in Genetics, Bioinf. & Comp. Biology
    - http://www.grads.vt.edu/gbcb/phd_gbcb.htm
  - Washington University
    - Computational Biology Program
    - http://www.ccb.wustl.edu/

(b) Short courses
- Berlin Graduate Program
  - Dynamics & Evolution of Cellular and Macromolecular Processes
  - http://www.biologie.hu-berlin.de/

TABLE 1. Continued.

Biocentrum Amsterdam
Molecular Systems Biology Course
http://www.science.uva.nl/biocentrum/
Cold Spring Harbor Laboratory
Course in Computational Genomics
http://meetings.cshl.org/
Institute of Systems Biology
Introduction to Systems Biology and Proteomics
Informatics Courses
http://www.systemsbiology.org
University of Oxford
Genomics, Proteomics & Beyond
http://www.conted.ox.ac.uk/cpd/biosciences/courses/short_courses/Genome_Analysis.asp
(c) Emerging initiatives
German Systems Biology Research Program
http://www.systembiologie.de/
Harvard University
Department of Systems Biology
http://sysbio.med.harvard.edu/
Manchester Interdisciplinary Biocentre (MIB)
http://www.mib.umist.ac.uk/
U. Texas Southwestern
Program in Molecular, Comp. & Systems Biology
Integrative Biology Graduate Program
http://www.utsouthwestern.edu/utsw/home/education/integrativebiology/

And what for all of this training? What jobs are new systems biologists likely to find? With the formation of myriad new academic departments and centers, the academic job market is booming. On the other hand, biotechnology firms and "big pharma" have been more cautious about getting involved.[8] However, most agree that in the long term systems approaches promise to influence drug development in several areas: (a) target identification, in which drugs are developed to target a specific molecule or molecular interaction within a pathway; (b) prediction of drug mechanism of action (MOA), in which a compound has known therapeutic effects but the molecular mechanisms by which it achieves these effects are unclear; and (c) prediction of drug toxicity and properties related to absorption, distribution, metabolism, and excretion (ADME/Tox). In all of these cases, the key contribution of systems biology would be a comprehensive blueprint of cellular pathways used for identifying proteins at key pathway control points, or proteins for which the predicted perturbation phenotypes most closely resemble those observed experimentally with a pharmacologic or toxic agent.

Looking toward higher levels of living systems behavioral hierarchy, students preparing for research careers in integrative systems physiology should build a strong foundation in core life sciences, mathematics and engineering. It is particularly useful to be immersed in life sciences courses which present biological principles in the context of mathematical models and engineering methodologies. An example of such a course is the year-long course entitled "Physiological Foundations of Biomedical Engineering" offered at the Johns Hopkins University. Foundation courses in mathematics could include ordinary and partial differential equation theory as well as probability theory and stochastic processes. While not commonly available, introductory course work in nonlinear dynamical systems theory would be valuable. Students may also opt to build a strong foundation in a core engineering discipline such as mechanical, chemical or electrical engineering.

Students pursuing any aspect of computational or systems biology at the graduate level face the hard fact that they *must* be as deeply educated in relevant areas of the life sciences as their biological colleagues, and they *must* be as strong in appropriate areas of engineering and mathematics as their colleagues in traditional areas of engineering and mathematics. Students will only be successful in this endeavor if they have a true love for both their chosen areas of biology and math/engineering concentration. The broad discipline of quantitative modeling of biological systems is one that is developing rapidly and is seeing increased representation in bio- and biomedical engineering departments, life sciences departments and traditional engineering departments. Students may therefore undertake combined experimental and modeling research or modeling research conducted in collaboration with experimental investigators with reasonable confidence that they will be able to find an academic department which appreciates and supports the particular balance they have chosen between modeling and experimentation

The discipline of computational medicine poses exciting new educational prospects that have yet to be tapped. Bio- and biomedical engineering is seeing increased popularity as the chosen research discipline of students in Medical Scientist Training Programs. At the Johns Hopkins University School of Medicine, several graduating medical students each year choose to delay entry into residency programs in order to pursue a year of research. This presents an ideal opportunity for these students to receive focused, in-depth training in quantitative aspects of integrative systems physiology, so that they may then bring these methods to their area of clinical interest.

CONCLUDING NOTE

In our view, Bioengineering is an ideal discipline for address of questions posed in the realm Systems Biology, well-suited to contribute experimental measurement and manipulation techniques along with computational mining and modeling methods, which taken all together can generate and test hypotheses in multivariate, dynamic, and quantitative manner. We anticipate that in this way, Bioengineering can have a major impact on basic understanding of living systems in terms of underlying, complex

molecular mechanisms, and on generating significant advances in diagnosis, treatment, and prevention of human disease.

REFERENCES

[1]Anderson, J. http://www.nigms.nih.gov/funding/systems.html, 2003.

[2]Ashby, R. General systems theory as a new discipline. *Gen. Syst. Yearbook* 3:1958.

[3]Asthagiri, A. R., and D.A. Lauffenburger. Bioengineering models of cell signaling. *Annu. Rev. Biomed. Eng.* 2:31, 2000.

[4]Bar-Joseph, Z., G. Gerber, T. I. Lee, N. Rinaldi, J. Yoo, F. Robert, D. Gordon, E. Fraenkel, T. Jaakkola, R. A. Young, and D. K. Gifford. Computational discovery of gene modules and regulatory networks. *Nat. Biotech.* 21:1337, 2003.

[5]Bertalanffy, L.V. General Systems Theory: Foundations, Development, Applications. Penguin, 1973.

[6]Buehler, C., J. Dreessen, K. Mueller, P. T. So, A. Schilb, U. Hassiepen, K. Stoeckli, and M. Auer. Multiphoton excitation of intrinsic protein fluorescence and its application to pharmaceutical drug screening. *Assay Drug Dev. Technol.* 3:155, 2005.

[7]Burns, M. A., B. Johnson, S. Brahmasandra, K. Handiqute, J. Webster, M. Krishnan, T. Sammarco, P. Man, D. Jones, D. Heldsinger, C. H. Mastrangelo, and D. T. Burke. An integrated nanoliter DNA analysis device. *Science* 282:484, 1998.

[8]Cohen, H. Systems biology: A pale beacon for biotechs. *Scientist*. 17. 2003.

[9]Cortassa, S., M. Aon, E. Marban, R. L. Winslow, and O'Rourke. An integrated model of cardiac mitochondrial energy metabolism and calcium dynamics. *Biophys. J.* 84:2734, 2003.

[10]DeRisi, J. L., V. R. Iyer, and P. O. Brown. Exploring the metabolic and genetic control of gene expression on a genomic scale. *Science*, 278:680, 1997.

[11]DiBernardo, D., M. Thompson, T. Gardner, S. Chobot, E. Eastwood, A. Wojtovich, S. Elliott, S. Schaus, and J. J. Collins. Chemogenomic profiling on a genome-wide scale using reverse-engineered gene networks. *Nat. Biotech.* 23:377, 2005.

[12]Friedman, N. Inferring cellular networks using probabilistic graphical models. *Science* 303:799, 2004.

[13]Fritz, J., E. B. Cooper, S. Gaudet, P. K. Sorger, and S. R. Manalis. Electronic detection of DNA by its intrinsic molecular charge. *Proc. Natl. Acad. Sci. USA* 99:14142, 2002.

[14]Gardner, T., D. diBernardo, D. Lorenz, and J. J. Collins. Inferring genetic networks and identifying compound mode of action via expression profiling. *Science* 301:102, 2003.

[15]Gaudet, S., K. A. Janes, J. G. Albeck, E. A. Pace, D. A. Lauffenburger, and P. K. Sorger. A compendium of signals and responses triggered by prodeath and prosurvival cytokines. *Molec. Cell. Proteomics*, in press, 2005.

[16]Gilman, A., and A. P. Arkin. Genetic "code": Representations and dynamical models of genetic components and networks. *Annu. Rev. Genomics Hum. Genet.* 3:341 2002.

[17]Griffin, J. L., C. J. Mann, J. Scott, C. C. Shoulders, and J. K. Nicholson. Choline containing metabolites during cell transfection: An insight into magnetic resonance spectroscopy detectable changes. *FEBS Lett.* 509:263, 2001.

[18]Griffin, T. J., S. P. Gygi, T. Ideker, B. Rist, J. Eng, L. Hood, and R. Aebersold. Complementary profiling of gene expression at the transcriptome and proteome levels in Saccharomyces cerevisiae. *Mol. Cell. Proteomics* 1:323, 2002.

[19]Griffith, L. G,, and G. Naughton. Tissue engineering—current challenges and expanding opportunities. *Science* 295:1009, 2002.

[20]Gygi, S. P., B. Rist, S. A. Gerber, F. Turecek, M. H. Gelb, and R. Aebersold. Quantitative analysis of complex protein mixtures using isotope-coded affinity tags. *Nature Biotech.* 17:994, 1999.

[21]Haugen, A. C., R. Kelley, J. B. Collins, C. J. Tucker, C. Deng, C. A. Afshari, J. M. Brown, T. Ideker, and B. Van Houten. Integrating phenotypic and expression profiles to map arsenic-response networks. *Genome Biol.* 5:R95, 2004.

[22]Ideker, T., T. Galitski, and L. Hood. A new approach to decoding life: Systems Biology. *Annu. Rev. Genomics Hum. Genet.* 2:343, 2001.

[23]Ideker, T., and D. A. Lauffenburger. Building with a scaffold: Emerging strategies for high- to low-level cellular modeling. *Trends Biotech.* 21:255, 2003.

[24]Jafri, S., J. J. Rice,, and R. L. Winslow. Cardiac Ca^{2+} dynamics: The roles of ryanodine receptor adaptation and sarcoplasmic reticulum load. *Biophys. J.* 74:1149, 1998.

[25]Janes, K. A., J. R. Kelly, S. Gaudet, J. G. Albeck, P. K. Sorger, and D. A. Lauffenburger. Cue-signal-response analysis of TNF-induced apoptosis by partial least squares regression of dynamic multivariate data. *J. Comp. Biol.* 11:544, 2004.

[26]Jasanoff, A. Functional MRI using molecular imaging agents. *Trends Neurosci.* 28:120, 2005.

[27]Kitano, H. Computational systems biology. *Nature* 420:206, 2002.

[28]Kunkel, E. M, Dea, A. Ebens, E. Hytopoulos, J. Melrose, D. Nguyen, K. Ota, I. Plavec, Y. Wang, S. Watson, E. C. Butcher, and E. L. Berg. An integrative biology approach for analysis of drug action in models of human vascular inflammation. *FASEB J.* 18:1279, 2004.

[29]Lauffenburger, D. A., and J. J. Linderman. *Receptors: Models for Binding, Trafficking, and Signaling*. Oxford: Oxford University Press, 1993.

[30]Lee, T. N. Rinaldi, F. Robert, D. Odom, Z. Bar-Joseph, G. Gerber, N. Hannett, C. Harbison, C. Thompson, I. Simon, J. Zeitlinger, E. Jennings, H. Murray, D. Gordon, B. Ren, J. Wyrick, J. Tagne, T. Volkert, E. Fraenkel, D. K. Gifford, and R. A. Young. Transcriptional regulatory networks in Saccharomyces cerevisiae. *Science* 298:763, 2002.

[31]Levchenko, A. Dynamical and integrative cell signaling: Challenges for the new biology. *Biotech. Bioeng.* 84:773, 2003.

[32]Luo, C. H., and Y. Rudy. A dynamic model of the cardiac ventricular action potential: I. Simulations of ionic currents and concentration changes. *Circ. Res.* 74:1071. 1994.

[33]Nielsen, U. B. M. H. Cardone, A. J. Sinskey, G. MacBeath, and P. K. Sorger. Profiling receptor tyrosine kinase activation by using Ab microarrays. *Proc. Natl. Acad. Sc. USA* 100:9330, 2003.

[34]Noble, D. Cardiac action and pace maker potentials based on the Hodgkin-Huxley equations. *Nature* 188:495, 1960.

[35]Perlman, Z., M. Slack, Y. Feng, T. J. Mitchison, L. F. Wu, and S. J. Altschuler. Multidimensional drug profiling by automated microscopy. *Science* 306:1194, 2004.

[36]Price, N. D. J. A. Papin, C. H. Schilling, and B. O. Palsson. Genome-scale microbial *in silico* models: The constraints-based approach. *Trends Biotech.* 21:162, 2003.

[37]Rice, J. J. M. S. Jafri, and R. L. Winslow. Modeling short-term interval-force relations in cardiac muscle. *Am. J. Physiol.* 278:H913, 2000.

[38]Sachs, K. O. Perez, D. Pe'er, D. A. Lauffenburger, and G. P. Nolan. Causal protein signaling networks derived from multiparameter single-cell data. *Science* 308:523, 2005.

[39]Said, M. R. T. J. Begley, A. V. Oppenheim, D. A. Lauffenburger, and L. D. Samson. Global network analysis of phenotypic effects: protein networks and toxicity modula-

tion in *Saccharomyces cerevisiae*. *Proc. Natl. Acad. Sci. USA* 101:18006, 2004.

[40]Saucerman, J. J. L. L. Brunton, A. P. Michailova, and A. D. McCulloch. Modeling beta-adrenergic control of cardiac myocyte contractility in silico. *J. Biol. Chem.* 278:47997, 2003.

[41]Saucerman, J. J. S. N. Healy, M. E. Belik, J. L. Puglisi, and A. D. McCulloch. Proarrhythmic consequences of a KCNQ1 AKAP-binding domain mutation: Computational models of whole cells and heterogeneous tissue. *Circ. Res.* 95:1216, 2004.

[42]Savran, C. A. S. M. Knudsen, A. D. Ellington, and S. R. Manalis. Micromechanical detection of proteins using aptamer-based receptor molecules. *Anal. Chem.* 76:3194, 2004.

[43]Sivaraman, A. J. K. Leach, S. Townsend, T. Iida, B. J. Hogan, D. B. Stolz, R. Fry, L. D. Samson, S. R. Tannenbaum, and L.G. Griffith. A microscale in vitro physiological model of the liver: Predictive screens for drug metabolism and enzyme induction. *Curr. Drug Metab.*, in press, 2005.

[44]Wang, Y.-C. A. L. Stevens, and J. Han. Million-fold preconcentration of proteins and peptides by nanofluidic filter. *Anal. Chem.* 77:4293, 2005.

[45]Wheeler, D. B. Carpenter, A. E,., and D. M. Sabatini. Cell microarrays and RNA interference chip away at gene function. *Nat. Genetics.* 37:S25. 2005.

[46]Winslow, R. L., D. F. Scollan, A. Holmes, D. K. Yung, J. Zhang, and M. S. Jafri. Electrophysiological modeling of cardiac ventricular function: from cell to organ. *Annu. Rev. Biomed. Eng.* 2:119, 2000.

[47]Zerhouni, E. Medicine. The NIH Roadmap. *Science* 302:63, 2003.

[48]Zhang, Y., A. Wolf-Yadlin, P. Ross, D. Pappin, J. Rush, D. A. Lauffenburger,, and F. M. White. Time-resolved mass spectrometry of tyrosine phosphorylation sites in the EGF receptor signaling network reveals dynamic modules. *Mol. Cell. Proteomics*, in press, 2005.

[49]Zhou, H., J. D. Watts, and R. Aebersold. A systematic approach to the analysis of protein phosphorylation. *Nature Biotech.* 19:375, 2001.

[50]Zhu, H., and M. Snyder. "Omic" approaches for unraveling signaling networks. *Curr. Opin. Cell Biol.* 14:173, 2002.