Improvement of bioengineering courses through systems biology and bioprocess modeling

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Abstract

This joint project applied MATLAB and Simulink to improve courses for Metabolic Engineering, Parameter Estimation for Engineering, Process Dynamics and Control, and a Process Control Laboratory in the Department of Energy, Environmental, and Chemical Engineering (EECE) at Washington University in St. Louis (WUSTL) in 2011-2013. The project also improved the existing bioengineering courses in the Department of Biosystems & Agricultural Engineering (BAE) at Michigan State University (MSU),specifically Microbial Systems Engineering and Analysis of Biological Systems in 2012~2014. Both MATLAB and Simulink were used in these courses. More than 100 undergraduate and graduate students per year from both universities have been enrolled in the classes in the past two years.

The teaching approach was to introduce MATLAB and Simulink to bioengineering courses. Using computational modeling tools, students developed the equations for various applications in systems biology and bioengineering, solved the forward and the inverse problems, used Simulink to perform process control/design of engineering projects, and finally optimized bioprocesses (both static and dynamic modes) using MATLAB tool boxes. Moreover, students were exposed to real experiments in the bio-reaction lab where data were collected. For all courses, each student had a MathWorks-supplied license to use all necessary toolboxes.

The class material is designed to teach bioengineering students multiple-scale modeling skills in both bioprocess engineering and systems biology so that they can have a holistic understanding of both scale-up fermentation engineering design and the microbial metabolism in response to bioreactor heterogeneity. Such skills are especially valuable for students looking to work in bioprocessing companies. Assessment was made through homeworks, projects, exams on MATLAB/Simulink, comments from students and other instructors, and feedback from alumni. Several of the students have now implemented MATLAB/Simulink in their research, introducing new methods to their advisors. Outcomes included a webpage with slides and notes posted for public access; two journal articles published, one book chapter published and two submitted, and one graduate course on MATLAB/Simulink that became a required course for graduate students in the Department of BAE at MSU.

Introduction

Modeling is used to summarize data, optimize, allow rapid predictions for what-if" scenarios, and to design processes. Over the past decade, computer modeling and simulation have become much more widely used in the industry. The explosion of interest in modeling is due in part to access to better and lower-cost software, and to the potential savings in effort, time, and cost in experimental work. Both undergraduate and graduate engineering students need to have some proficiency in using modeling and technical computing software before they enter the job market. Therefore, the goal of this project was to improve the existing engineering courses in Chemical and Biosystems Engineering by using MATLAB and Simulink. The students' experience of writing code in MATLAB and arranging a system in Simulink was excellent preparation for understanding the computational algorithms. Teaching bioengineering students computational and simulation skills related to kinetics and systems biology can improve their knowledge for future careers in industry. Ultimately, students will develop systems engineering skills to solve problems in the nascent biotechnology industry.

Background and Course Description

Industrial biotechnology often uses microorganisms and enzyme catalysts to synthesize useful products. The benefits of biological reactions in large quantities cannot be realized without using systems biology (knowledge at the molecular level), dynamic control and modeling theory (knowledge at the process level). The critical frontiers for bioprocess engineering students are direct experience with systems biological analysis, bioreactor operations and the modeling of dynamic behavior of metabolic reactions under controlled variables. The MathWorks grant-funded project of systems biology and kinetic process modeling relied on fundamental knowledge in biology, chemistry, mathematics, statistics, kinetics, and chemical process engineering, which was integrated into the curriculum for four major courses at WUSTL and MSU.

1. ***Metabolic engineering*** (ChE596) at WUSTL focuses on analysis of complex interactions in biological systems and introduction of metabolic changes to achieve desired cellular properties [1]. Currently, numerous chemical compounds, ranging from pharmaceuticals to biofuel, have been produced with the aid of biological tools. The ability to efficiently synthesize natural or synthetic products requires a systems-level understanding of metabolism as well as metabolic responses in different fermentation conditions. This class teaches molecular tools for pathway modifications, systems biology, and metabolic modeling. The class also teaches the methods to solve the underdetermined flux balance models to describe microbial ethanol fermentation and predict product yield using different carbon substrates. There are total of 40 graduate/undergraduate students who participated in the 2011~2014 classes.

2. ***Process Control (ChE 462) and Process Control Laboratories (ChE 463)*** at WUSTL, teaches chemical engineers process control theory and educates the students on control techniques employed in industry. Process control (ChE462) focuses on the control dynamics and model simulation of chemical processes [2]. The Control laboratories

(ChE463) consist of 5 control experiments using a state-of-the-art EMERSON electronic controller and workstations to control processes such as flow, level, pressure and temperature. Real-time process data are available in EXCEL and MATLAB (including Simulink). A bioprocess laboratory (a sixth control experiment) is set up to focus on the bioreactor operation and modeling (~30 graduate/undergraduate students/year). Students learn about Programmable logic controllers from an adjunct professor from a local chemical company, and use control theory and modeling skills to resolve the actual problems related to industry.

3. ***Bioprocess Engineering (ChE 453)*** at WUSTL, teaches chemical engineers fermentation engineering theory and bio-separations. The class focuses on the enzyme kinetics, fundamental of microbiology, bio-reaction modeling, mass transfer limitation, metabolic models (stoichiometry models and flux balance models), process scale-up and product separations [3]. In the class, there are three-week computer labs that cover the training of students in using three MATLAB tool boxes (1.curve fitting, 2. partial differential equation, and 3. optimization). Students will also learn the use of MATLAB (fmincon function) to predict optimal control parameters for bio-reactions.

4. ***Microbial Systems Engineering*** (BE 360) at MSU trains biosystems engineers how to design, model, and simulate bioengineering processes. Topics include application of engineering fundamentals, biological principles, and computational tools to the analysis of microbial processes; kinetic analysis of biological processes, modeling of microbial processes, unit operations and scale-up. Applications to biofuel and food production are given. MATLAB and Simulink were used in this course. Development of a fermentation laboratory exercise, and more extensive modeling experience with MATLAB and Simulink enhanced the student experience. There were 30, 42, 35, and 35 undergraduate students in 2011, 2012, 2013, and 2014, respectively.

5. ***Analysis of Biological Systems*** (BE 835) at MSU. This graduate-level course is in two parts: 1) Numerical techniques and the forward problem; and 2) Parameter estimation and inverse problems. Other topics within these two include optimal experimental design, sequential parameter estimation, model discrimination, and Monte Carlo simulation. Students are tested on being able to use MATLAB to solve systems of ODEs and single PDEs when all parameters are given (the forward problem), and to estimate these parameters when experimental data are given (the inverse problem). There were 15, 17, and 14 students in 2011, 2012, and 2013, respectively. Each year, 3-5 of the students are from other departments, such as School of Packaging and Chemical Engineering.

Teaching Approach

Both MATLAB and Simulink were used in these courses to estimate parameters for fermentation kinetics (a system of ordinary differential equations), to numerically solve kinetic models (ode functions), to simulate the bioethanol/biomass production (Systems Biology Toolbox), and to model a bioreactor combining mass and heat transfer (Basic Simulink).

In each course, students were taught the principles, equations, and numerical solution methods before using MATLAB and Simulink, to avoid a "black-box" approach. MATLAB and Simulink were presented as tools that make the solving of the equations faster and more convenient. Parameters were given to solve forward problems first. For the inverse problem, simulated or real data were given to estimate the parameters. Simulink was used to solve systems of ODEs, and then to optimize the process based on selected parameters or variables. The details for each course are given below.

1. ***Metabolic Modeling Development and Solutions***

Students at WUSTL have both undergraduate and graduate courses to teach MATLAB/Simulink skills to develop and solve biological problems. For example, flux analysis is an important systems biology tool for physiological prediction of enzymatic rates in metabolic networks, and allows knowledge-based design of cellular functions. The cell-wide quantification of intracellular fluxes can be performed via Flux Balance Analysis (FBA), which uses the stoichiometry of the metabolic reactions and a series of biological constraints to obtain the feasible fluxes. In both metabolic engineering and bioprocess classes, students have learned how to develop the FBA models using MATLAB to solve the underdetermined flux model using the function (fmincon).

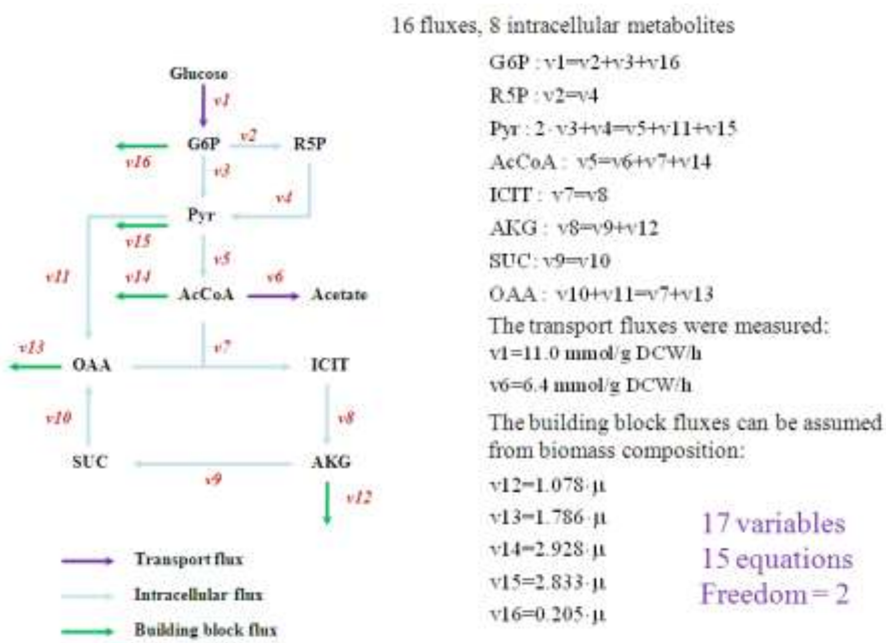


Figure 1: A simplified FBA model to demonstrate *E.coli* aerobic growth. The model requires an objective function (maximize μ) to predict the cell metabolism towards optimal biomass growth (MATLAB “fmincon” can be used to resolve this problem).

2. ***Process Dynamics and Control and Process Control Laboratories***

In both classes, steady and unsteady-state behavior of chemical processes, fundamental feedback and feedfoward control strategies, and modern control theory and applications were taught. After taking this course, students not only understood process control theory and laboratory operations, but also learned the skills for developing models to analyze and predict the process dynamics. During the semester, students learned both Simulink and MATLAB in the computer lab for about one month. Students had team projects to use the bioreactor for ethanol and butanol fermentations, and developed models to describe the biomass and alcohol production data (Figure 2a). They completed a computer project on alcohol fermentation using the actual experimental data provided by the instructor. Students developed the kinetic models (using ordinary differential equations) and perform the parameter fitting and statistical analysis using MATLAB (ode45 coupled with nlinfit functions) (Figure 2). There are four to five variables in the model including glucose, biomass, alcohol, acetate (as the inhibitory byproducts) and nitrogen sources (yeast extract). Students have to compare the parameters obtained from different fermentation conditions to estimate the alcohol production under influences of oxygen level, substrate concentration, and chemical inhibitions. The project will also ask students to incorporate the control loop (PID control) to simulate the operation of bioreactor fermentation under different oxygen conditions. According to students’ feedback, the computer lab is the most valuable part of this class since it develops their computational skills for their future academic and industrial jobs.

Biomass $$\frac{d(XV)}{dt} = V\mu_{max}X\frac{S}{S+K_S}\frac{[O]}{K_O+[O]}\frac{N}{K_N+N} - k_d X$$

Ethanol $$\frac{d(PV)}{dt} = VX\frac{S}{\frac{[O]}{K_I}+S+K_S}$$

Sugar $$\frac{d(SV)}{dt} = FS_f - Y_{S/X}\frac{d(XV)}{dt} - Y_{S/P}\frac{d(PV)}{dt}$$

Nitrogen source $$\frac{d(NV)}{dt} = -\frac{\mu X}{Y_{X/N}}$$

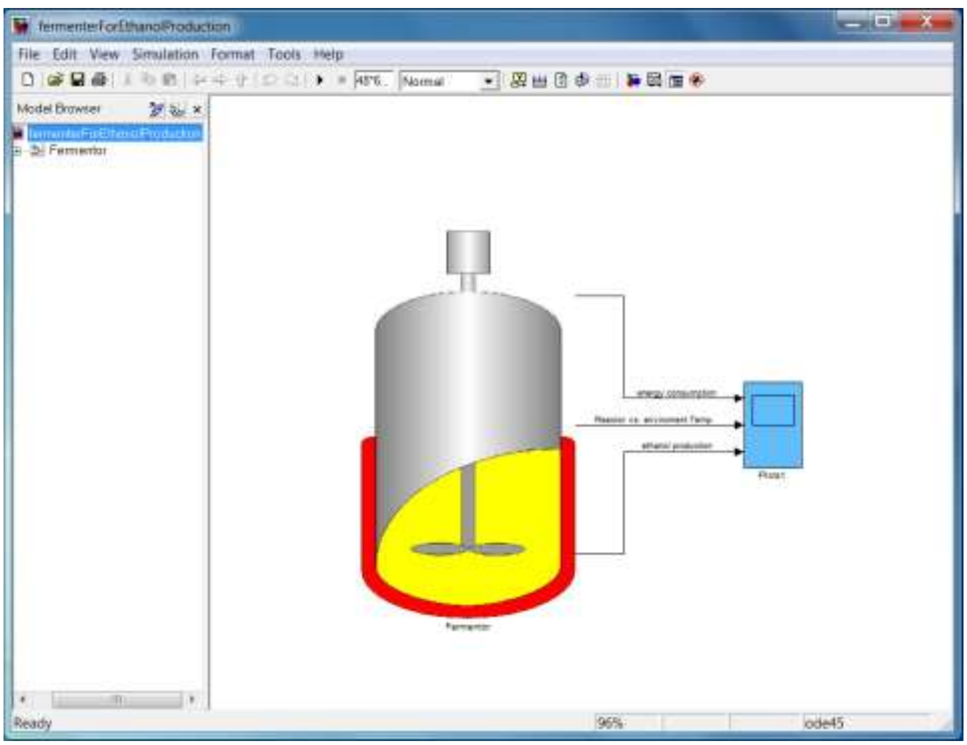
Total mass $$\frac{dV}{dt} = F$$

a) Ethanol fermentations using yeast. b) Kinetic model for ethanol fed-batch fermentation

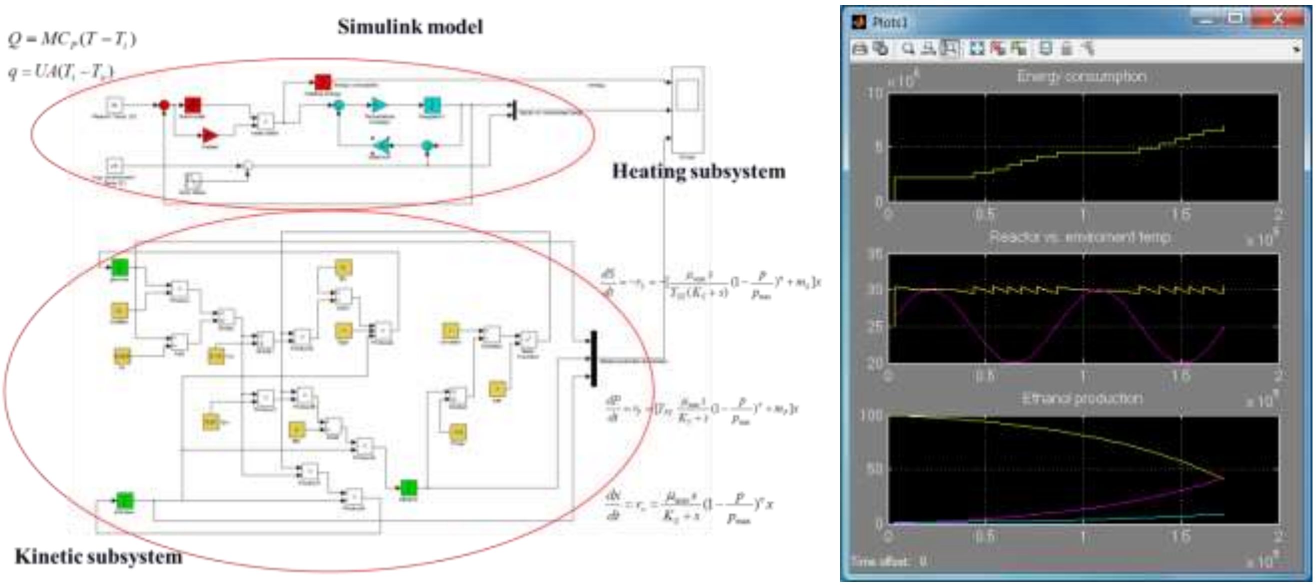
Figure 2: Fermentation lab and modeling at Washington University in St. Louis.

3. ***Microbial Systems Engineering*** (BE 360)

The approach this course took was integrating the introduction of microbial processes with mathematical modeling of microbial kinetics. The course started with lecturing applications of microbial systems in environmental, food, and energy industries. After discussion of fundamental microbial physiology and mass/energy balance of microbial processes, microbial kinetics was introduced in the class. Due to the complicated nature of microbial kinetics, a system of differential equations was required to describe the kinetics. Often, there are no algebraic solutions for such kinetics. Students used mathematical modeling to connect the engineering and microbiology components. The MATLAB function ode45 and a solution approach for a system of differential equations were introduced to students. A demonstration fermentation lab was given to apply the modeling. All parameters such as specific growth rate constant, maintenance coefficients, product/biomass yield, and inhibition coefficient that were needed for the modeling were obtained from the demonstration lab. Students were required to construct a Simulink® model to describe a complicated microbial process using the parameters from the demonstration lab (Fig. 3). According to the students' responses, this approach enhanced the students' understanding of how to use mathematical tools to find solutions for real-world applications.



(a)



(b) (c)

Figure 3. Simulink® model of a fermentor of yeast ethanol fermentation
(a) Model interface; (b) Simulink® model; (c) Simulation results

4. ***Analysis of Biological Systems*** (BE 835).

All lectures were taught in a computer lab where each student had two monitors. Class was held Tuesday and Thursday, 4:10-5:30pm. The first half of the course, the forward problem, was taught using this textbook [4] . The main topics taught were numerical techniques to integrate the area under functions; to do root-finding, and to solve systems of ODEs for initial-value and boundary-value problems for linear and nonlinear ODEs, using ode45 and the finite-difference method. The finite-difference method for a heat-conduction PDE was taught for Cartesian coordinates. Students were then assigned homework to write two MATLAB codes for the PDE in radial coordinates with two different boundary conditions.

The instructor's screen was shared live on the student's screen, so that students could see the instructor's code and simultaneously test it on their other screens using MATLAB. The lectures were based on PowerPoint slides [5] for each chapter, supplied by Mathworks. The first 4 lectures were an introduction to coding in MATLAB, Chapters 2-4. After the students had a working knowledge of MATLAB coding, a typical lecture would consists of the instructor giving a powerpoint lecture explaining the concept for that day, such as how initial-value ODEs are set up and solved in MATLAB. The instructor would stop at appropriate intervals to demonstrate how to run the MATLAB code so the students could try it and ask questions. The instructor had uploaded all slides and code examples so that students would have all materials when each class started.

After the students had already mastered how to use ode45 to solve an initial-value ODE, Simulink was introduced. Students were expected to know how to use Simulink to solve systems of initial-value ODEs.

The second half of the course, Parameter Estimation, was based on [6] and an update of Chapter 6 that has MATLAB code in it [7]. The instructor developed his own notes and powerpoint slides to give lectures and show how to run the MATLAB code. The main topics were parameter estimation by ordinary least squares (OLS) with ode45 and nlinfit, sequential estimation; matrix formulation and statistics for the parameter errors; model discrimination, and optimal experimental design. Because the MATLAB code for these topics was fairly long and complicated, the instructor supplied most of these as generic codes, and went through each cell in the code to make sure the students understood it. Students were expected to know how to modify the code for any homework or exam problem.

Assessment

Assessment was made through homeworks, projects, exams on MATLAB/Simulink, required numerical student evaluations, and comments from students and other instructors. More than 90% of the students entering all of the courses described have very little to no MATLAB coding or Simulink experience. Based on student class evaluations, all our courses received very positive feedback. For example, 2013 Bioprocess Engineering Class (ChE453) at WUSTL has a class evaluation of 6.2/7 (far above the school average 4.8/7). The student evaluation for the instructor of BE 835 in 2013 was 3.91 out of 4.0, a high score. Several of the students have now implemented MATLAB/Simulink in their research, introducing new methods to their advisors. Many students at WUSTL (>50) from the bioprocess engineering and process control classes) went to graduate school. Several graduate students from Metabolic Engineering Class found academic jobs at universities and National Laboratories. Their current job requires use of MATLAB extensively.

Outcomes included a webpage with slides and notes posted for public access; a new website on parameter estimation using MATLAB; one journal article submitted by a student based directly on his data and what he learned in the course with instructor Dolan; and one graduate course on MATLAB/Simulink that became a required course for graduate students at MSU. The instructors Tang and Liao are preparing two book chapters (One chapter is on Enzyme kinetics, and the other chapter is on Metabolic and Bioreactor Models) for a new text book "Bioenergy: Principles and Applications". The aim of the book will bridge the gap between bioenergy education and industrial applications. A third book chapter based on the parameter estimation section of BE 835 was recently published [8].

Some details are given below.

1. ***Metabolic engineering*** (ChE596) and ***Bioprocess Engineering*** (ChE453)

One homework was dedicated to the flux modeling component of the course ChE596. Students were asked to develop a simple FBA model with 20 reactions in the central metabolisms to describe the alcohol fermentation pathways. One homework was dedicated in ChE453, which requires the use of MATLAB optimization (e.g. "fmincon") to predict the optimal bioreactor conditions during fermentation process.

2. ***Process Control and Process Control Laboratories*** (ChE 462/463)

Two homework and two computer projects were assigned for students to practice modeling skills using MATLAB and Simulink. They learned parameter estimation using Excel and the MATLAB curve fitting Toolbox; kinetic modeling (using ode45, ode23 and ode15s funcitons); Simulink (building and running simulations); Parameter fitting and Process Optimization (using fmin function and nlinfit). One class project example was posted on **Youtube** by students: http://www.youtube.com/watch?v=kL-qoKvNesU

3. ***Microbial Systems Engineering*** (BE 360)

Two homeworks were dedicated to the modeling component of the course. Students were asked to independently derive a system of differential equations to describe the kinetics of two microbial processes, a bacterial denitrification process and a yeast ethanol fermentation process. The students were also required to use MATLAB to find numeric solutions for them. The instructor found that the students were intrigued by using the mathematical tools they just learned to model real-world applications.

4. ***Analysis of Biological Systems*** (BE 835).

Weekly homeworks counted for 50% of the class, because students can learn MATLAB best by doing many example problems. Some difficulties doing in-class exams on the computer included unplanned technical difficulties with certain computers, and the time required to debug codes. After teaching the course twice, the instructor found that if an in-class midterm exam (25%) is used, it should be either short-answer principle-based, or have a limited number of straightforward coding questions, or both. A take-home midterm is being considered as a better option. For the final exam (25%), both a take-home exam and a project with 15-minute presentation were tried in two separate classes. The instructor preferred the project, because students will use the inverse problems methods to estimate parameters for the students' data or data selected from the literature, giving the students real-world experience.

Finally, students from these bioengineering classes at WUSTL and MSU helped submit several research articles and book chapters:

- Kinetic modeling and isotopic investigation of isobutanol fermentation by two engineered *Escherichia coli* strains. ***Industrial & Engineering Chemistry Research.*** 2012. 51 (49): 15855–15863.
- Construction of a parsimonious kinetic model to capture microbial dynamics via parameter estimation. Inverse Problems in Science and Engineering. 2014. 22(2): 309~324.

- Book Chapter 14. Microbial metabolisms and metabolic modeling for biofuel production in "Bioenergy: Principles and Applications". Under review.

- Book Chapter 15. Enzymatic hydrolysis in "Bioenergy: Principles and Applications". Under review.

- "Chapter 7: Parameter Identification Under Dynamic Temperature Conditions in Inactivation Kinetics", *Progress on Quantitative Approaches of Thermal Food Processing*, New York, New York: Nova Science Publishers, 2012. Published.

Outcomes

1. Both undergraduate and graduate engineering students are proficient in MATLAB and Simulink, making them more competitive for jobs.
2. Slides and course syllabus are posted for free use at the website: http://tang.eece.wustl.edu/MATLAB_WUSTL.htm
3. Journal articles published (on ***Industrial & Engineering Chemistry Research, Inverse Problems in Science & Engineering***) or) by student using the methods learned in the course.
4. BE 835 selected in 2012 as a required course for graduate students in the department.
5. Increased use of MATLAB and Simulink in undergraduate projects and graduate research, and improvement of the quality of the academic research.

Conclusions

All four classes were significantly improved by teaching and hands-on problem solving with MATLAB and Simulink. The individual student licenses provided by Mathworks ensured that toolboxes could be used at all times. This class, including multiple-scale modeling education, bridges the gap between the biological science and bioprocess engineering, as shown in the diagram (Figure 4). Such education will train the students with diverse skills and expertise on biotechnology applications.

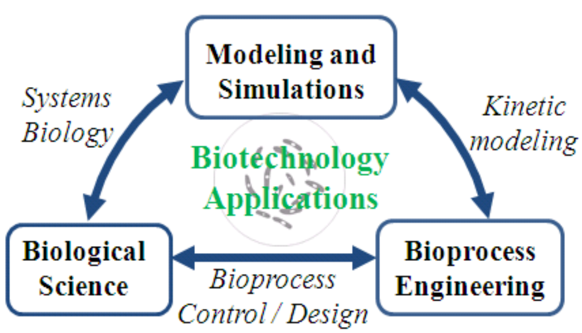


Figure 4: The overall goal of course development.

Acknowledgments

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