To: Course 7 students
Re: Full-time or Part-time UROP positions for In-Person and Remote Bioprocess Summer Projects
From: Jean-François Hamel, PhD
4/12/2021

If you are interested in exploring any of the projects, please Email me a copy of your resume and relevant information. Please note that you must secure summer housing outside the MIT dormitory system in order to pursue UROP work.

UROP PROJECT #1: Developing a Rational Methodology for the Golden Batch from Simulation and Experimentation

BACKGROUND

The proposed project aims to develop a methodology for creating the Golden Batch in the bioprocess field, and to evaluate Golden Batch candidates in real-time.

The "Golden Batch" is a terminology routinely used in the industry to refer to the "ideal batch," or a standard batch that can be used as a reference point for assessing the quality of future batches. In other words, the Golden Batch serves as a reference to determine whether future batches meet the standard or desired quality attributes (i.e. optimal cell density) within 3 standard deviations or 95% confidence interval limits (Figure 1). There have already been multiple studies on using data from Golden Batches to detect the good and bad batches during fermentations.[2-6] It is also possible to predict the average "golden batch" trajectory for desired properties as the fermentation progresses forward.[4] The "golden batch" approach has also been used to develop a predictive feeding rate control algorithm for an optimal biomass growth profile.[5]

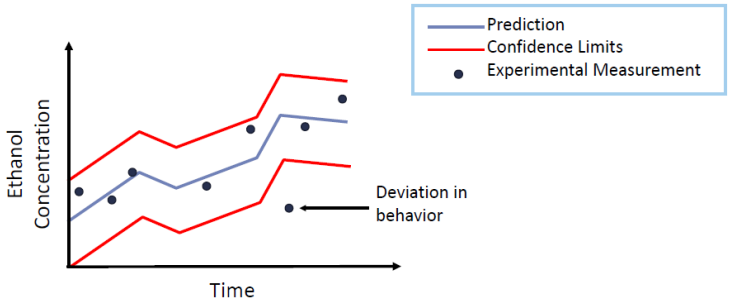

Figure 1. Models can help detect deviations in standard behavior (Janny Cai, Course X, Class'2019)

In these studies, the Golden Batches are usually already known, so the expected results can be predicted. However, in the biotechnology industry where people are constantly working with new strains of bacteria or organic material, the Golden Batch (or standard batch) is often unknown. However, determining the Golden Batch requires multiple experiments, which can be resource-intensive. Thus, one goal of the Golden Batch project is to determine an optimal number of runs and variables that would minimize resource usage.

Across these studies, there is significant variation in the number of bioreactor runs and the nature of measurements, off-line and online, used for generating a Golden Batch approach.

This project aims to develop a Golden Batch approach by implementing a multivariable linear regression model, which will use off-line and online capabilities (Figure 2).

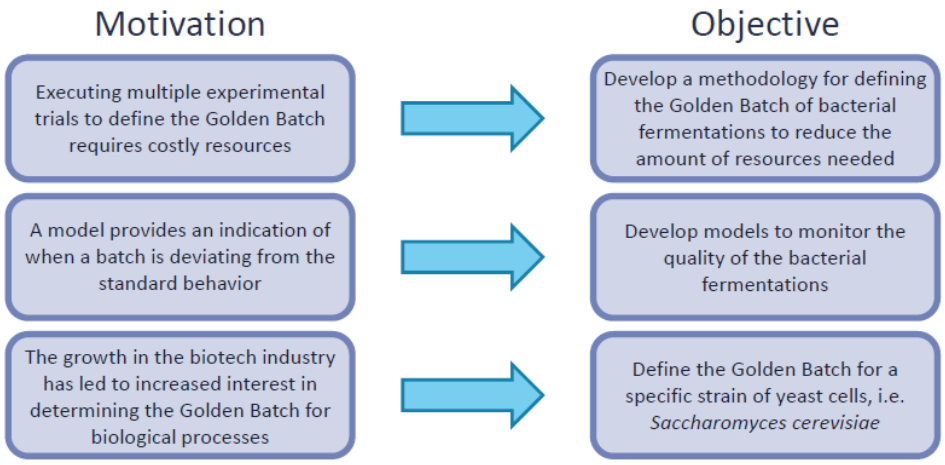

Figure 2. The Golden Batch is an ideal batch, which can be used as a reference to evaluate the quality of future batches (Janny Cai, Course X, Class'2019)

Team members located onsite and off-site will monitor and analyze a series of batches in real-time and perform post-run analysis and modeling. Overall, multiple data points will be collected from multiple fermentation runs, and the datasets will be used to develop a model for defining the Golden Batch. Once the Golden Batch model is defined, it can be used to assess the quality of future fermentation batches.

The Golden Batch project has many implications for the future of the biotechnology industry and research. It can help optimize and automate bioprocesses, and simultaneously reduce the consumption of materials during fermentations if the batch is deviating from standard behavior.

THE TEAM

The Summer Team will include Mathieu Medina (Course X, Class'2021) who will oversee the simulation and modeling work, UROP student(s) and Dr. Hamel. Mat is contributing to enhancing the bioprocess module for 10.28 (fall term). External support will be provided by OSI Soft (now part of AVEVA) for real-time data monitoring and archiving during Phase II (see below).

ON-SITE LAB WORK

The UROP student will study the effect of glucose concentration, pH and temperature on metabolism of *Saccharomyces cerevisiae*, under aerobic and anaerobic conditions. In Phase 1, shake-flask cultures will be designed and analyzed using the Design of Experiment (DoE) methodology (software: JMP). Off-line analysis will include measurements of glucose, ethanol cell concentration (by optical density and dry cell weight) and pH. Data will be used for simulation

studies and to design bioreactor experiments. In Phase II, shake-flask culture conditions will be selected for scale-up to the bioreactor, for batch and fed-batch studies. Similar parameters will be analyzed like done for the shake flask scale. However, pH will be monitored and controlled online, as well as dissolved oxygen and agitation rate. The data will be captured by OSI PI, a Data Archiving system, designed to be used remotely. Off-line data (e.g. glucose, ethanol, cell concentration and from simulation runs) can also be fed into the PI system for advanced analysis.

REMOTE SIMULATION WORK

The UROP student will investigate and implement computational simulation methods to describe mathematically data derived from on-site experiments, detailed above, and develop predictive transport and kinetics models[7] about cell growth and ethanol production (Figure 3).

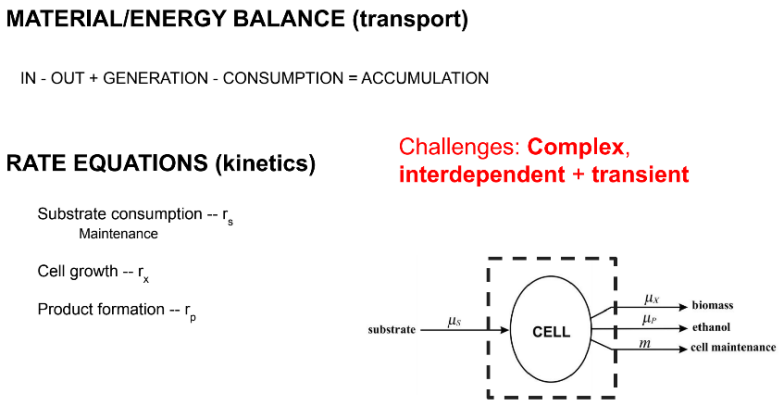

Figure 3. Yeast fermentation module (Mathieu Medina, Course X Class'2021)

A review and incorporation of relevant scientific literature will be performed to inform and refine simulation of in-lab experiments. Numerous software programs will be available to simulate the bioprocess, including but not limited to more familiar programs like Excel or MatLab, as well as less familiar programs specific to yeast fermentation simulation, such as the AnaBioPlus[8] software package, which will be instructively introduced to the student, using multimedia developed specifically for this project. In each stage of the project, different programs may be implemented as appropriate and as determined by the student. Simulation work will be integrated with on-site lab work to facilitate an improved understanding of the experimental data, inform ongoing work in the project, and develop a rational methodology for the Golden Batch.

UROP PROJECT #2: Process Intensification and Continuous Extraction of Biofuel

BACKGROUND

Recent efforts to improve robustness of small molecule chemical and pharmaceutical manufacturing have been revolving around continuous approaches. In this context chemicals are essentially reacted as they flow through channels instead of reacting in a large batch reactors. Adoption of continuous manufacturing has been fueled, among other things, by emerging technologies that offer opportunities to execute in a continuous mode all the traditional process steps can be performed in a classic batch reactor. One example is provided by liquid-liquid extraction (LLE), this separation technique involves first a mass transfer step of a solute between two immiscible phases and a subsequent phase separation. Technology developed at MIT and now commercialized by Zaiput Flow Technologies (an MIT ChemE spin off www.zaiput.com) enables continuous and effective phase separation. This coupled with an engineered mass transfer step (i.e. deploying static or active mixers) provides fully continuous and very effective LLE. The result is not only a continuous process capability but also an intensified one, in line with the most recent industrial trends (Figure 4).

On another front, bioprocessing is well known to be a powerful and widespread approach to chemical manufacturing; bioprocesses are being deployed in a growing number of applications to manufacture the most diverse molecules, not only in a pharmaceutical context.

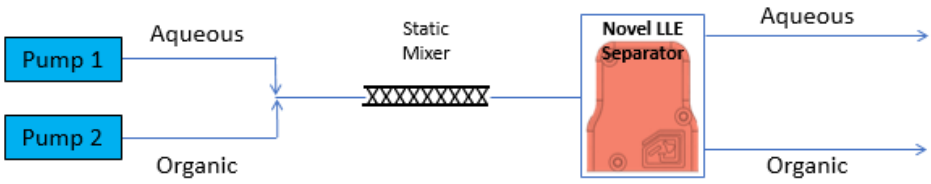

Figure 4. Coupling transport and separation steps in a novel liquid-liquid extractor (Andrea Adamo, Course X PhD, Zaiput LLC)

PROJECT DESCRIPTION

In this research project we propose to explore an intersection between bioprocessing and continuous liquid-liquid extraction. It can be speculated that continuous LLE can be used to alter the composition of the fermentation broth by continuously removing one or more chemical species. As an example, these can be products, thus skewing the bioprocess towards the product; as another example, removal of a toxic product, could, for instance, allow longer operation.

In this project the student will start first familiarizing oneself with continuous LLE using the Zaiput devices. Then this will be implemented in the context of a bioprocess. An engineer from Zaiput

will provide assistance and technical support. The work will entail contributing to the selection of adequate test cases, running the experiments, collecting and analyzing data. The project will be a great opportunity to use concepts from fluid mechanics, mass transfer in practical applications. Critical and original thinking to identify and address any performance issues will be an important part of the activities as well.

THE TEAM

The student will work with other UROP students and Dr. Hamel on site and receive training and external support from Zaiput engineers.

[1] P. Möree & J. Elfving Applications of Multivariate Data Analysis, OSIsoft Users Conference, 2013.
[2] L Chiang *et al*., Multivariate analysis for quality improvement of an industrial fermentation process, IFAC, 2004.
[3] A batch was declared "bad" when nonlinear relationships were observed between variables, while linear relationships were observed for most batches.
[4] J. Alves-Rausch *et al*., Real time in-line monitoring of large scale *Bacillus* fermentations with near-infrared spectroscopy Journal of Biotechnology 189 (2014) 120–128.
[5] O. Grigs *et al*., Model Predictive Feeding Rate Control in Conventional and…, *Chem. Biochem. Eng. Q.*, 30 (1) 47–60 (2016)
[6] R. Luttmann *et al*., Sequential/parallel production of potential Malaria vaccines – A direct way from single batch to quasi-continuous integrated production, Journal of Biotechnology 213 (2015) 83–96.
[7]Oliveira, Samuel C., *et al*. "Kinetic Modeling of 1-G Ethanol Fermentations." *Fermentation Processes*, 2017, doi:10.5772/65460.
[8]Oliveira, C. M., *et al*. "AnaBioPlus: a New Package for Parameter Estimation and Simulation of Bioprocesses." *Brazilian Journal of Chemical Engineering*, vol. 34, no. 4, 2017, pp. 1065–1082.