NATIONAL PH.D. PROGRAM IN AUTONOMOUS SYSTEMS

Production of microorganisms in bioreactors:

from modelling to control systems.

Ph.D. candidate

Antonio Spallone

Cycle

XL

Tutors

Fabrizio Carteni (Tutor)

Mario Di Bernardo (Tutor)

Stefano Mazzoleni (Company tutor)

1. Description of the research program

Bioreactors are sophisticated devices designed to facilitate biological and biochemical processes under meticulously monitored and tightly regulated environmental and operational conditions. These conditions include parameters such as pH, temperature, pressure, nutrient supply, and waste removal, all of which are critical for the optimal performance of the bioreactor.

One critical phenomenon affecting bioreactor efficiency is self-inhibition (SI), where microorganisms create unfavorable conditions that limit their own growth and reduce yields. Mechanisms of SI include nutrient depletion, accumulation of pathogens, and changes in microbial community composition. A recently discovered SI mechanism is inhibition by self-DNA (DNA originating from conspecifics), which affects bacteria, fungi, and plants by inhibiting growth and activity. The self-DNA is produced through various processes involving the natural lifecycle and environmental interactions of organisms including cell death, decomposition, and environmental stresses. Mazzoleni et al. conducted a study on the growth of the yeast Saccharomyces cerevisiae on a glucose substrate, comparing the various growth phases of the yeast under three different conditions: virgin substrate, substrate inoculated with self-DNA, and substrate inoculated with extra-DNA. The results showed that self-DNA fragments with lengths ranging from 120 to 3000 nucleotides led to a reduction in the yeast growth rate, a significant extension of the diauxic lag phase, and a delay in reaching the stationary phase.

These findings suggest that self-DNA inhibition is an important factor to consider in bioreactor control systems to optimize performance. Further research in this area is needed to better understand and mitigate the effects of self-inhibition, ultimately improving the efficiency and yield of bioreactor-based processes.

Objectives:

- Construct a model of microorganism growth in bioreactors that accounts for the inhibitory effect of self-DNA.
- Develop a control strategy to maximize production while minimizing production costs.
- Design the bioreactor system.
- Validate the model and control through simulation and physical testing.

2. Schedule of the research activities

First academic year

	Description	**Period**	**Activity abroad**
Bibliographic review	Analysis of existing research and publications on the self-inhibition, model of microorganisms and plant growing in bioreactors and control strategies for process optimization and efficiency.	1/11-28/2	NO
Mathematical model prototype	Definition of a mathematical model of the system to be developed during the project for controlling the inhibitory effect of self-DNA.	1/1-31/5	NO
Prototype	Design and implementation of the system, scaled based on the data obtained from the model. In the initial phase, the system will be kept at a smaller scale to allow for a simpler and more efficient development process.	1/12-31/5	NO
Prototype testing	Prototype testing on a scaled production system of microorganisms or plants in bioreactors.	1/5-31/10 **company**	NO

Second academic year

	Description	Period	Activity abroad
Prototype testing	Prototype testing on a scaled production system of algae or plants in bioreactors.	1/11-31/3 **company**	NO
Mathematical model real system	Modeling of the entire system, including the bioreactor and the filtration of the inhibitory effect.	1/11-28/2	NO
Control strategy	Implementation of the control strategy aimed at maximizing efficiency and minimizing energy consumption.	1/12-31/10	NO
Testing on real system	Testing of the filtration system and control strategies on a real production system of microorganisms and plants in bioreactors.	1/05-31/10 **abroad**	YES

Third academic year

	Description	**Period**	Activity abroad
Scale-up prototype	Scale-up of the prototype as possible with the goal of approaching industrial-scale usage.	1/11-31/03 **abroad**	YES
Model validation	Validation of the model on a larger scale.	1/11-31/03 **abroad**	YES
Thesis writing	Writing of the final report.	1/03-31/10	NO

3. Training and research activities plan

First academic year

	Description	Period	Final Exam	ECTS
A. Ph.D. courses	Deep Reinforcement Learning for Control of Autonomous Systems	Jan	No	0.5
	Modeling and simulation of biosystems	Jan	Yes	2
	How to prepare a research paper and present experimental data	Sep-Nov	Yes	6
	Stochastic Differential Equations	May-Jun	Yes	4
B. Master's degree courses	Biological systems modeling	Nov-Jan	Yes	6
C. Soft skill courses				
D. Participation to seminars	It will be chosen later based on the topics covered during the doctoral work.	/		1
E. Participation to international congresses or workshops	Conference on Decision and Control - Milan	Dic 16-19		4
	It will be chosen later based on the topics covered during the doctoral work.	/		4
F. Presentation of research products at international congresses or workshops				
	TOTAL OF ECTS FOR TRAINING ACTIVITIES			27.5

G. Individual research activity	The individual work will be carried out according to the plan outlined in the paragraph above.	Nov-Oct		27.5
H. Supervision of students				
I. Integrative teaching activities				
J. Preparation of manuscripts for conferences or journals	The final months of the first year will also be dedicated to writing an article on the preliminary results that I will obtain.	Mar-Oct		5
	TOTAL OF ECTS FOR RESEARCH ACTIVITIES			32.5
	TOTAL OF ECTS			**60**

Second academic year

	Description	**Period**	**Final Exam**	**ECTS**
A. Ph.D. courses				
B. Master's degree courses				
C. Soft skill courses				
D. Participation to seminars	It will be chosen later	/		1
E. Participation to international congresses or workshops	It will be chosen later	/		6
F. Presentation of research products at international congresses or workshops	It will be chosen later	/		2
	TOTAL OF ECTS FOR TRAINING ACTIVITIES			9
G. Individual research activity	The individual work will be carried out according to the plan outlined in the paragraph above.	Nov-Oct		35
H. Supervision of students	Support for students participating in the courses "Modeling of Biological Systems" and "Nonlinear Dynamics and Control."	/		2
I. Integrative teaching activities	I will hold a seminar on self-DNA inhibition and the modeling of the dynamics involved.	/		1
J. Preparation of manuscripts for conferences or journals	Time allocated to writing an article on the results I will obtain.	Nov-Oct		13
	TOTAL OF ECTS FOR RESEARCH ACTIVITIES			51
	TOTAL OF ECTS			**60**

Third academic year

	Description	**Period**	**Final Exam**	**ECTS**
A. Ph.D. courses				
B. Master's degree courses				

C. Soft skill courses				
D. Participation to seminars				
E. Participation to international congresses or workshops	It will be chosen later	/		6
F. Presentation of research products at international congresses or workshops		/		2
	TOTAL OF ECTS FOR TRAINING ACTIVITIES			8
G. Individual research activity	The individual work will be carried out according to the plan outlined in the paragraph above.	Nov-Oct		30
H. Supervision of students	Support for students participating in the courses "Modeling of Biological Systems" and "Nonlinear Dynamics and Control."	/		2
I. Integrative teaching activities	I will conduct a seminar on self-DNA inhibition, focusing on the modeling of the underlying dynamics, the control techniques that will be adopted, and the corresponding results.	/		2
J. Preparation of manuscripts for conferences or journals	Time allocated to writing an article on the results I will obtain.	Nov-Apr		18
	TOTAL OF ECTS FOR RESEARCH ACTIVITIES			52
	TOTAL OF ECTS			**60**

4. List of the publications written by the candidate in the triennium

Antonio Spallone

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Dr. Fabrizio Cartenì

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Dr. Mario Di Bernardo

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