Modeling and Dynamic Optimization of protein and spore production by *Bacillus thuringiensis*

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This presentation has 5 sections

1- IMP-4 Citrus aims
2- Model Process
3- Control Strategy
4- Results
5- Conclusions

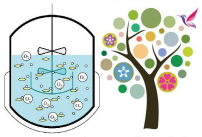
IPM-4-CITRUS Project

HORIZON 2020 FUNDED

Marie Slodowska Curie Action

Research & Innovation Staff Exchange

IPM-4-CITRUS



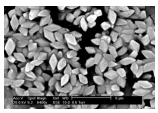
11 PARTNERS

6 COUNTRIES

4 YEARS DURATION

INTEGRATED PEST MANAGEMENT

- ✓ **Understanding & sensitising stakeholders** about the health risks related to citrus pests
- ✓ **Developing an alternative IPM approach**
- ✓ based on biological control



STRAIN USED:

Bacillus thuringiensis kurstaki BLB1 and LIP

Citrus

TARGETED PEST:

insect larvae

Phyllocnistis citrella & Prays citri

This project has received funding from the European Union's Horizon 2020 Research and Innovation programme under Grant Agreement N° 734921

BIOTECH

BIOACTIVITY

TRANSFER

Interdisciplinary

Intersectoral

International

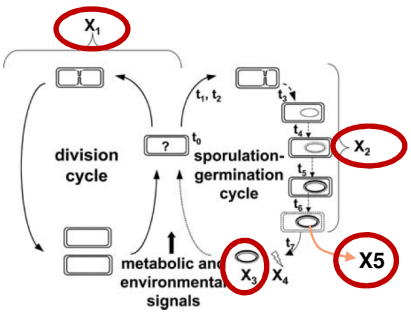
GOALS :

- ➢ Strengthening Academia & Industrial collaborations
- ➢ Optimising bioproduction processes
- ➢ Developing new biopesticides in the Mediterranean region

HOW :

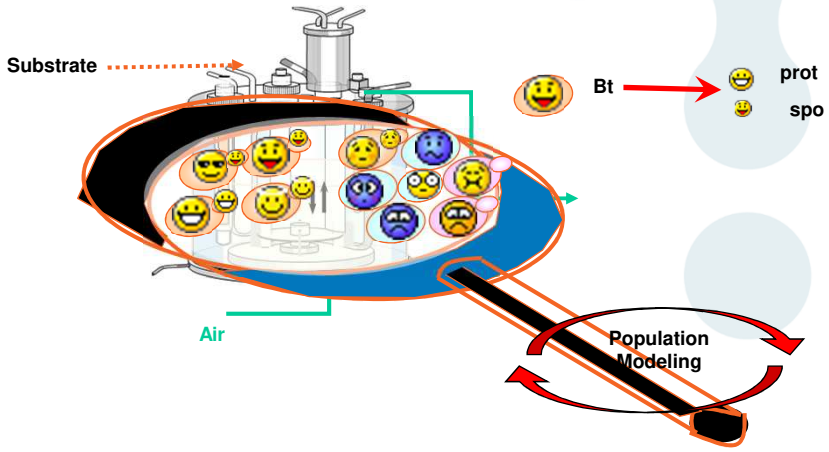
- ➢ Feasibility study for future spin-off activities and new production lines,
- ➢ Benchmarking the opportunities & obstacles related to bringing innovative ideas to the market.

Physiological states in a spore-forming bacterium (application to Bt kurstaki)



X1, X2, X3 and X4: cell quantification [cell],[DM] morphology
X5 : δ-endotoxin quantification [Crystal], [Protein] crystal morphology

Bioprocess Model Challenges



METHODES: Model for *B. THURINGIENSIS (FedBatch and Sequential Process)*

$$\frac{dX}{dt} = \mu * X - k_d * X - X\frac{Qin}{V} \quad (1)$$

$$\frac{dS}{dt} = -\frac{\mu * X}{Y1} - S\frac{Qin}{V} + Sin * Qin \quad (2)$$

$$\mu = \mu_{max}\frac{S}{(Kc * X) + S} \quad (3)$$

Model 1

$$\frac{dPro}{dt} = \frac{X * Y2}{Y1} - Pro\frac{Qin}{V} \quad (4)$$

$$\frac{dSpo}{dt} = \frac{X * Y3}{Y1} - Spo\frac{Qin}{V} \quad (5)$$

Model 2

$$\frac{dPro}{dt} = \frac{X * Y2}{Y1} + \alpha - Pro\frac{Qin}{V} \quad (6)$$

$$\frac{dSpo}{dt} = \frac{X * Y3}{Y1} + \beta - Spo\frac{Qin}{V} \quad (7)$$

METHODES: Control Strategies

Case 1 (Feed batch Optimisation)

$$max_{Qin}\left(\frac{(Pro * V + Spo * V)}{t_{end}}\right)$$

Subject to: $\begin{cases} Eq.(1-7) \\ 1 < V < 10L \\ 0.01 < Qin < 0.4L/h \end{cases}$

Were t_{end} is fermentation final time.

t_c=2 h

Case 2 (Sequential Batch Optimisation)

$$max_{Gluin,V,t_{end1},t_{end2}}\left(\frac{(Pro * V + Spo * V)}{t_{end}}\right)$$

Subject to : $\begin{cases} Eq.(1-7) \\ 1 < V < 10L \\ 15 < S_{in} < 25 \\ 1 < t_{end1} < t_{end2} < 45h \end{cases}$

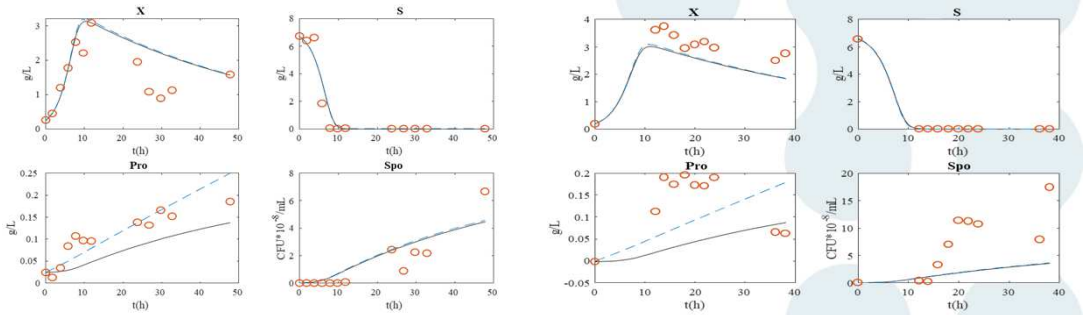
Were t_{end1} and t_{end2} are the fermentation final time for first and second sequential batches.

Results

Optimized parameter values from Lip strain

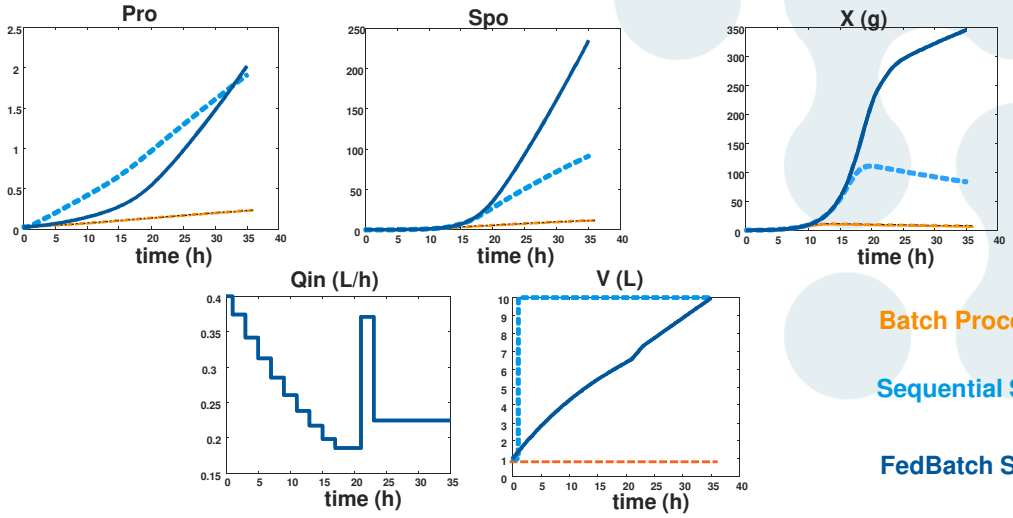
Parameter	Lip	
	Model 1	Model 2
μ max (h^{-1})	0,3966	0,3916
Kc	0,6899	0,5794
Kd (h^{-1})	0,0189	0,0193
Y1 ($g_{Biomass}/g_{Glucose}$)	0,4866	0,4956
Y2 ($g_{Pro}/g_{Glucosa}/h$)	0,0005	0,0001
Y3 (CFU*10^{-5}/$g_{Glucose}$/h)	0,0213	0,0218
Alpha (g/L/h)	-	0,0042
Beta (CFU*10^{-5}/L/h)	-	0,0002

RESULTS: MODEL SIMULATIONS



(-) Model 1, (- -) Model 2 and (o) **experimental data.**
X (biomass g/L), S (glucose g/L), Prot (Proteins g/L) and Spo (Spores CFU*10^{-8}/mL)

RESULTS: Control Strategy



Batch Process

Sequential Strategy

FedBatch Strategy

CONCLUSIONS

Both models followed the biomass and substrate dynamics.

Model 2 fitted better all data specially the proteins production

Fed-batch strategy had the best proteins and spores productivity with high biomass productivity

Perspectives:

- **To adapt control strategies with industrial substrates**
- **To propose some Soft Sensors to monitoring bioprocess**