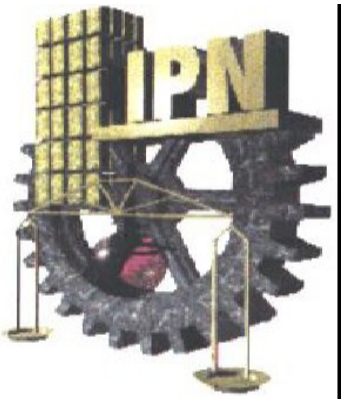
iGEM MEXICO 2007



"Biological implementation of algorithms"

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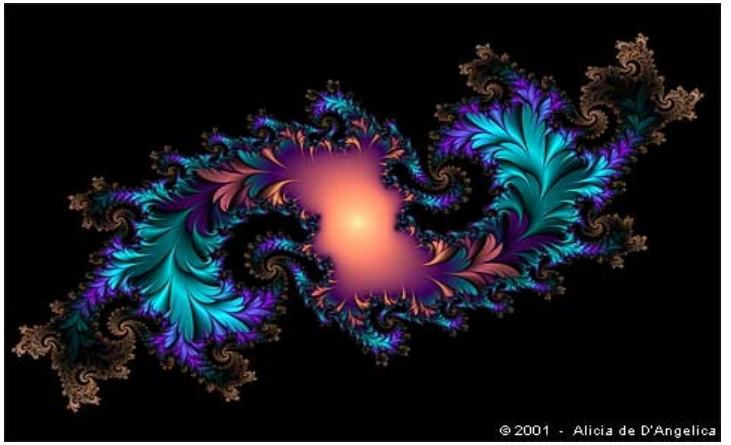
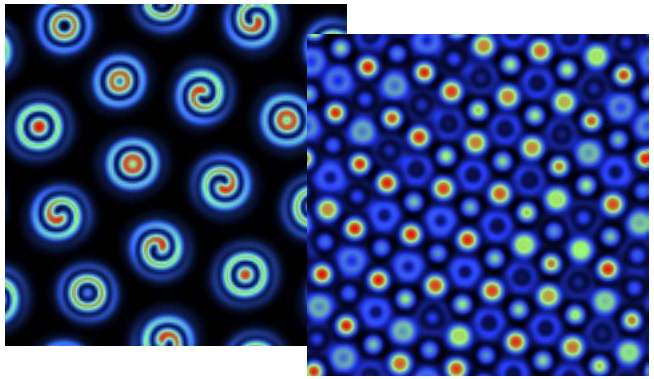
INTRODUCTION



- How structures emerge in living systems? Several mechanisms have been proposed, depending on the observed patterns: Turing patterns.
- It is still questioned if pattern formation and more generally, the appearance of functional structures can be understood by means of Turing patterns or more broadly, reaction-diffusion mechanims.
- One of the main goals of our project is to test different pattern formation mechanisms, not only Turing patterns, but also oscillatory and time varying structures.
- We propose that if the appropriate genetic construction is implanted in a colony of bacteria, the reaction-diffusion mechanism can be replaced by a genetic control system (Elowitz repressilator).
- In order to model these systems, we use Stochastic Pi-Calculus.

Our main interest is to reproduce complex patterns in biological systems

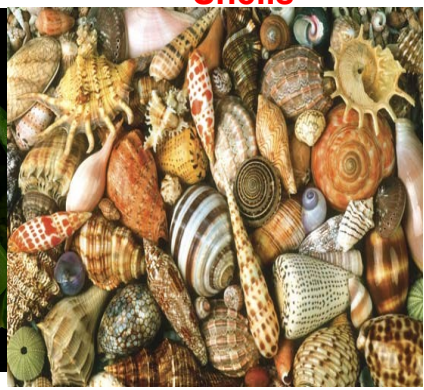


Oscillatory Turing pattern

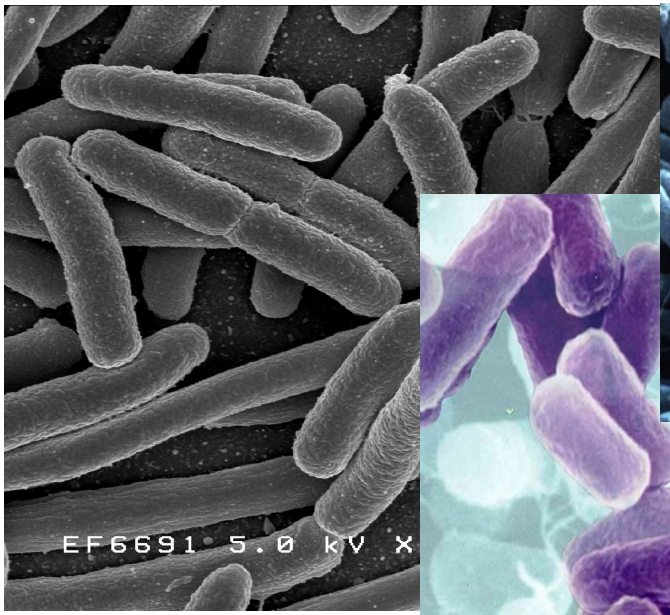
Parrot fish

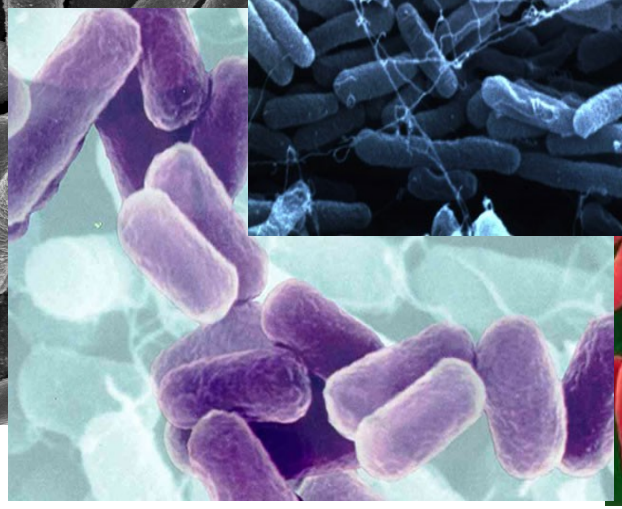
Plant fractal

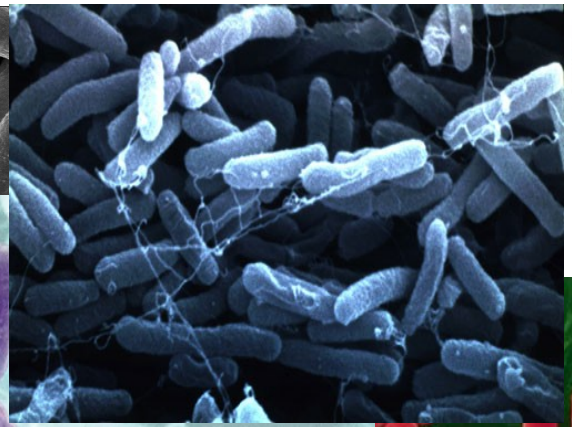
Shells

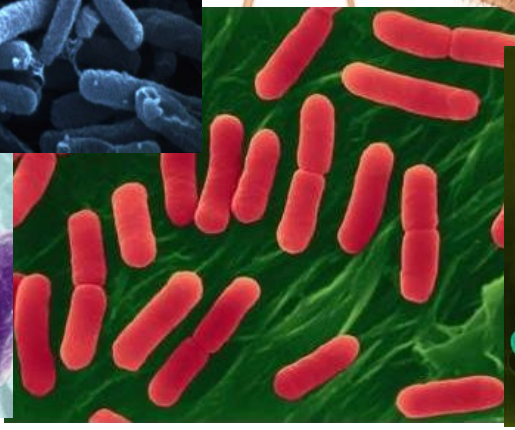


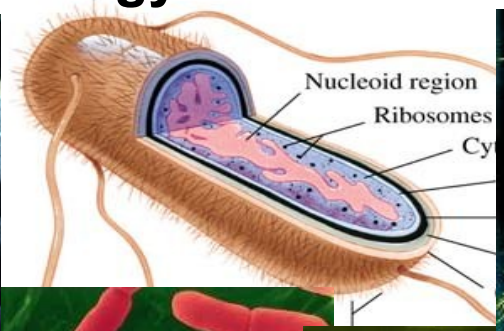
We chose *Escherichia coli* because is easily manipulated and there is a lot of information about its biology

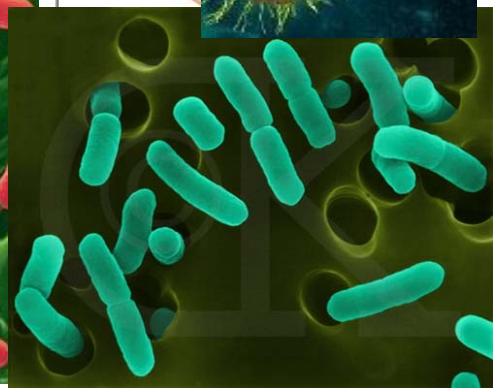




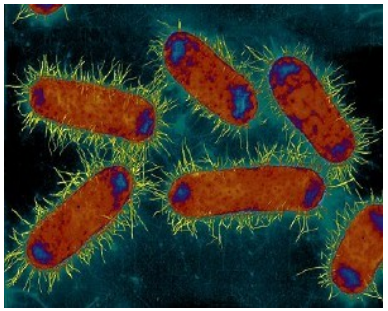








And is frequently used as a molecular biology model!!!!

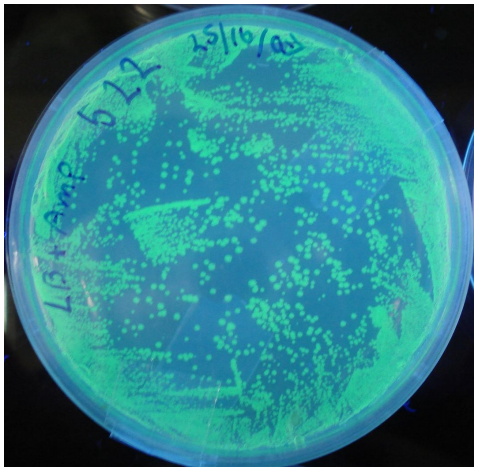


Do bacteria create complex patterns when cultivated on semisolid agar?

We transformed *E. coli* JM109 with biobricks Bba_I13521, Bba_I13522, Bba_J04430 and BBa_J04450.

Individual clones were visualized!!!

✓



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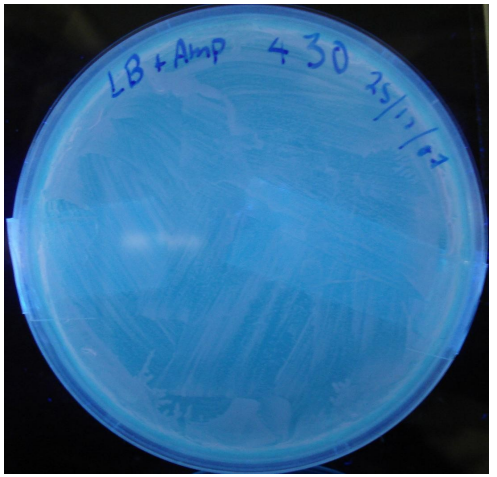

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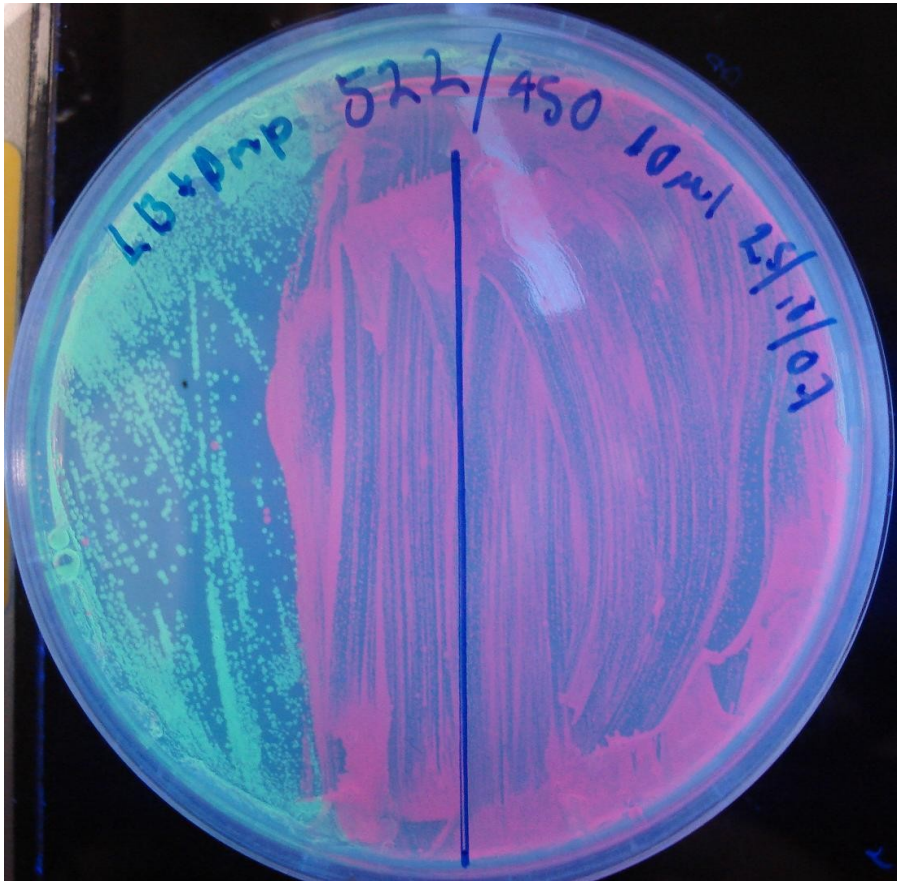
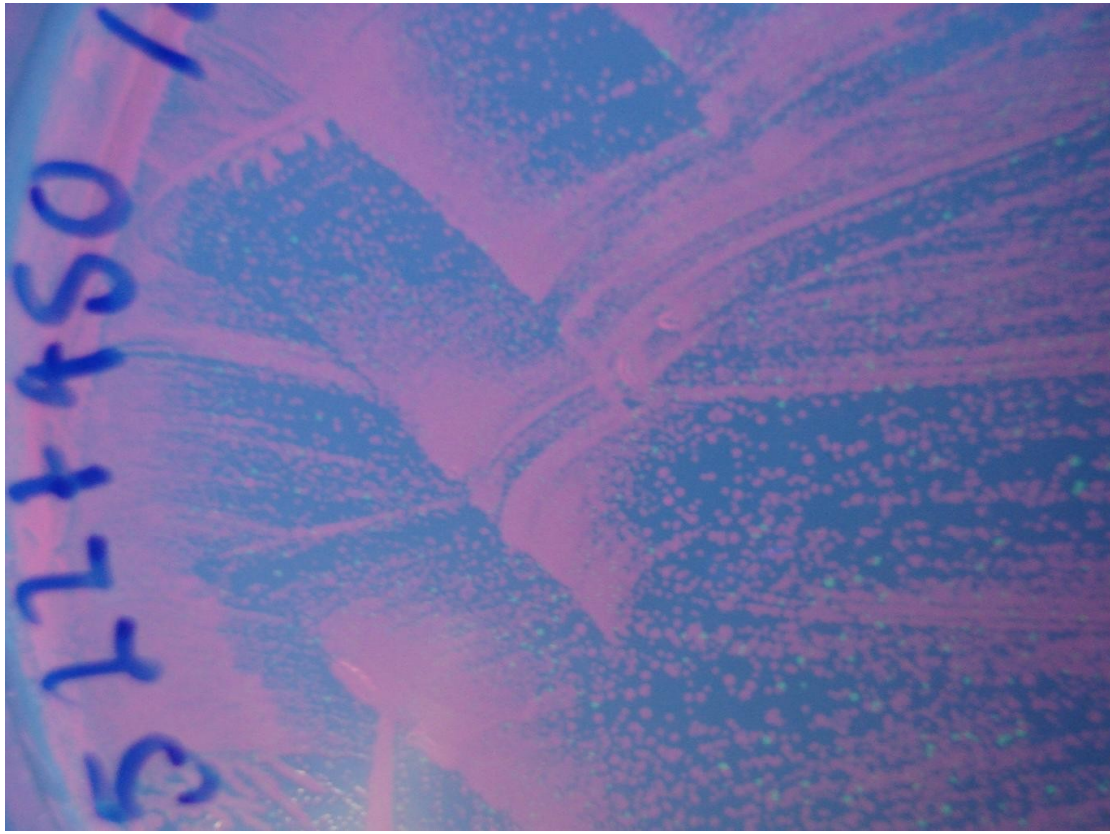
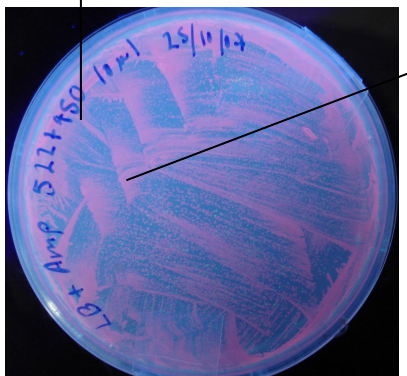
Too many bacteria!!!!!

Experiment

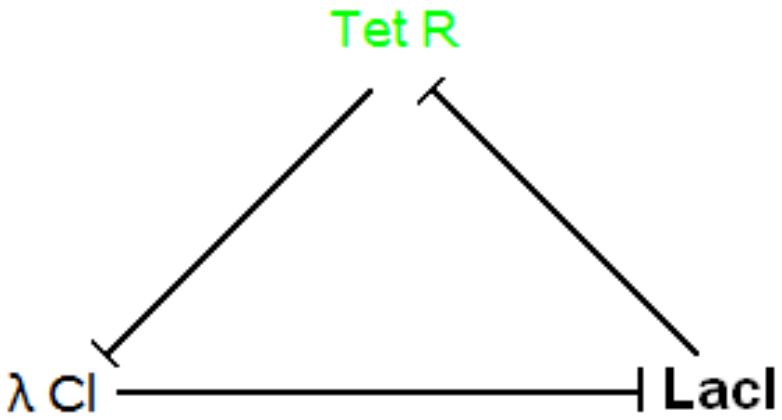
Plate *E. coli* that produce GFP and RFP separately and together...

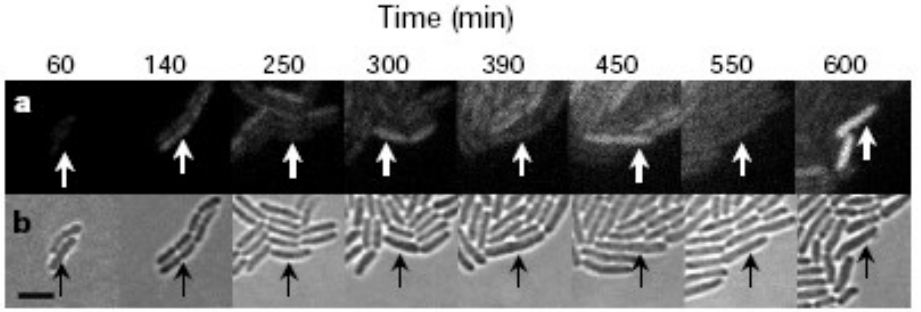




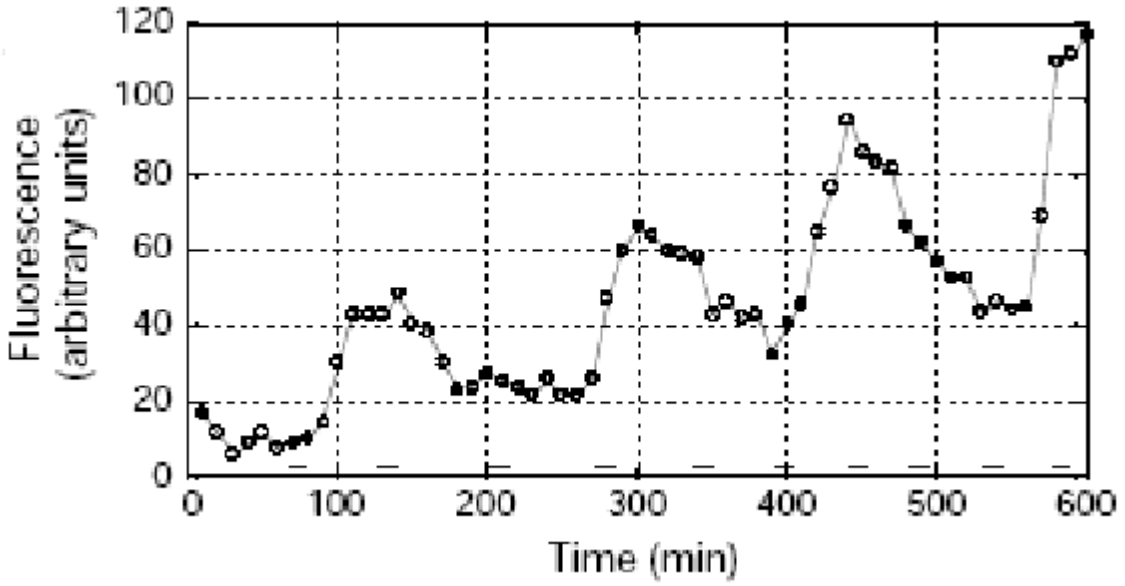


Elowitz

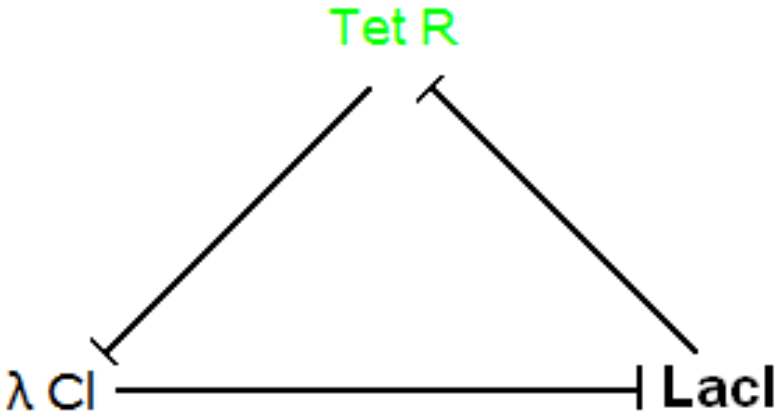


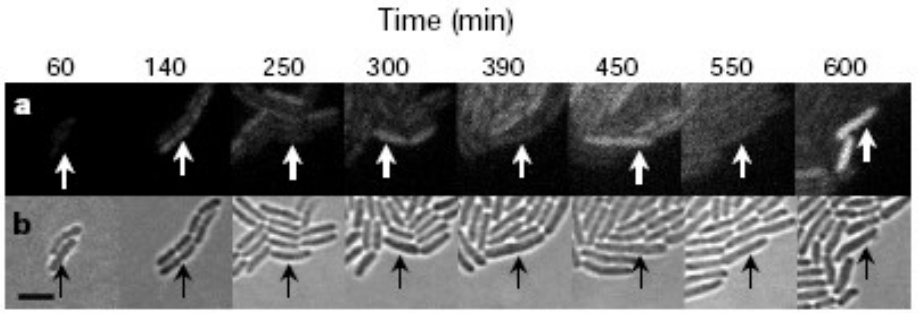


-Nature 403, 335 (2000)



Elowitz



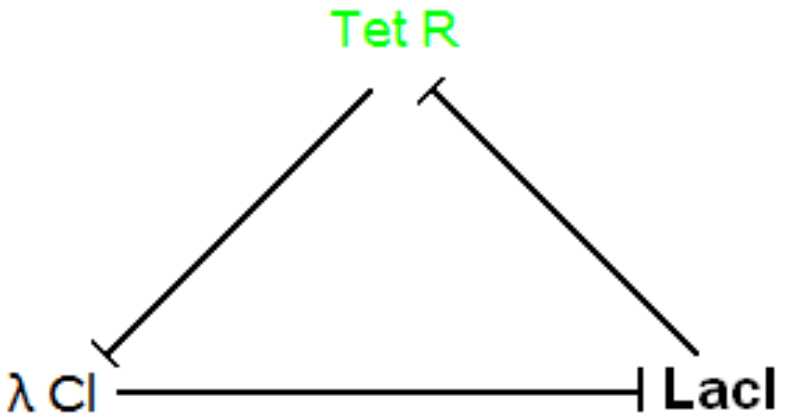


.Syncronyzation

-Positive control

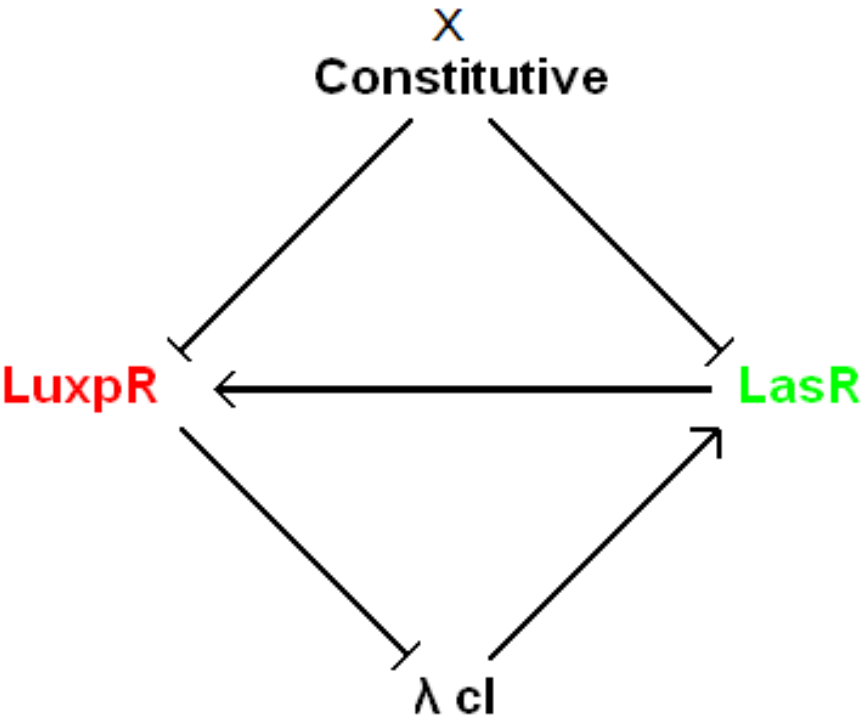
-Nature 403, 335 (2000)

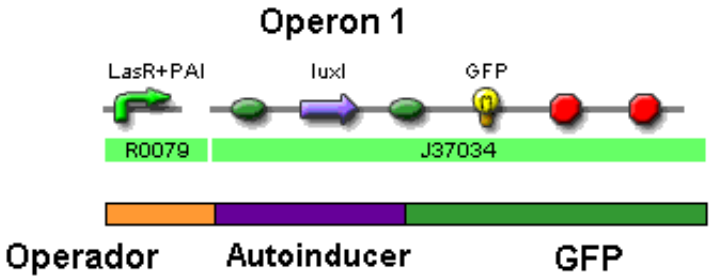
Elowitz



-Nature 403, 335 (2000)

UNAM

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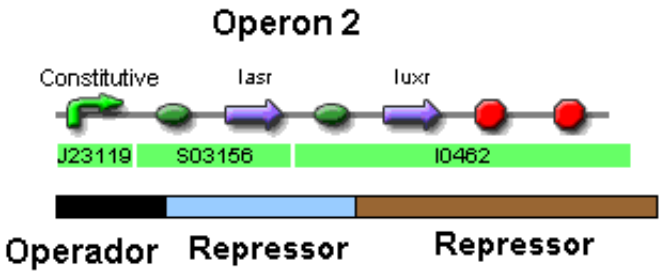
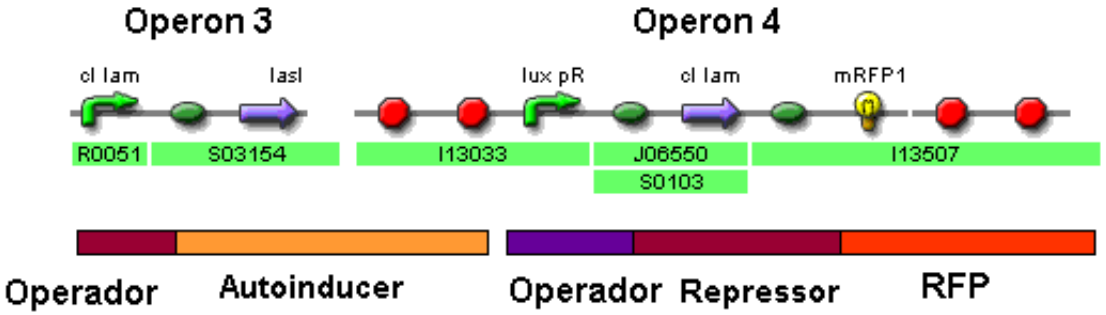
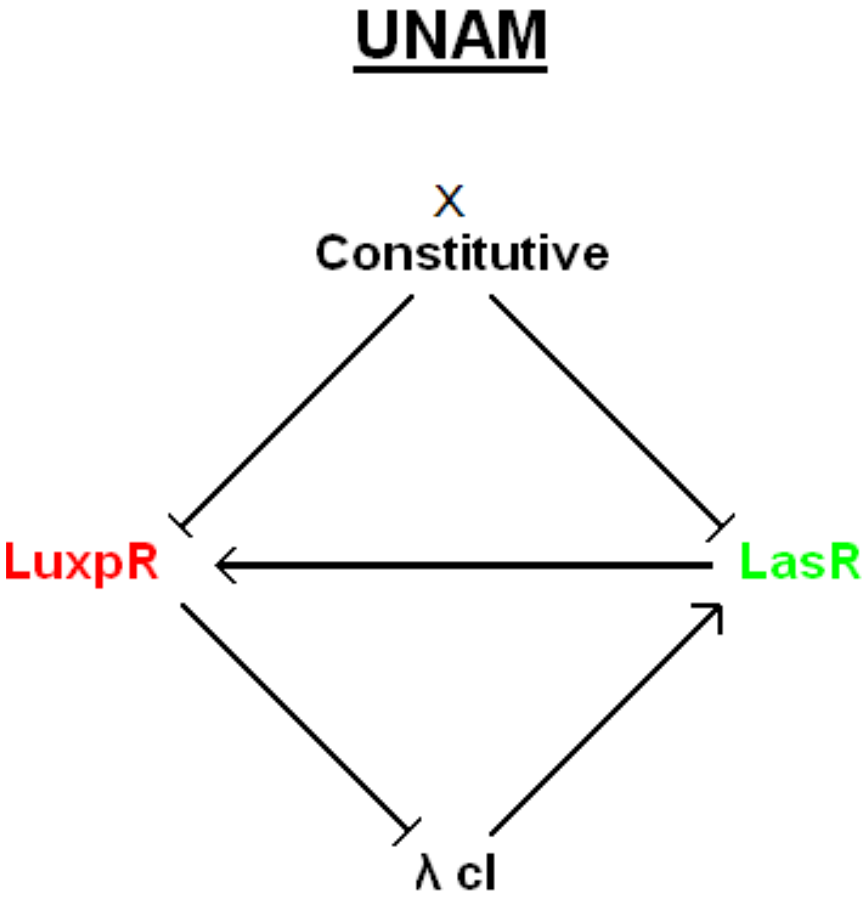
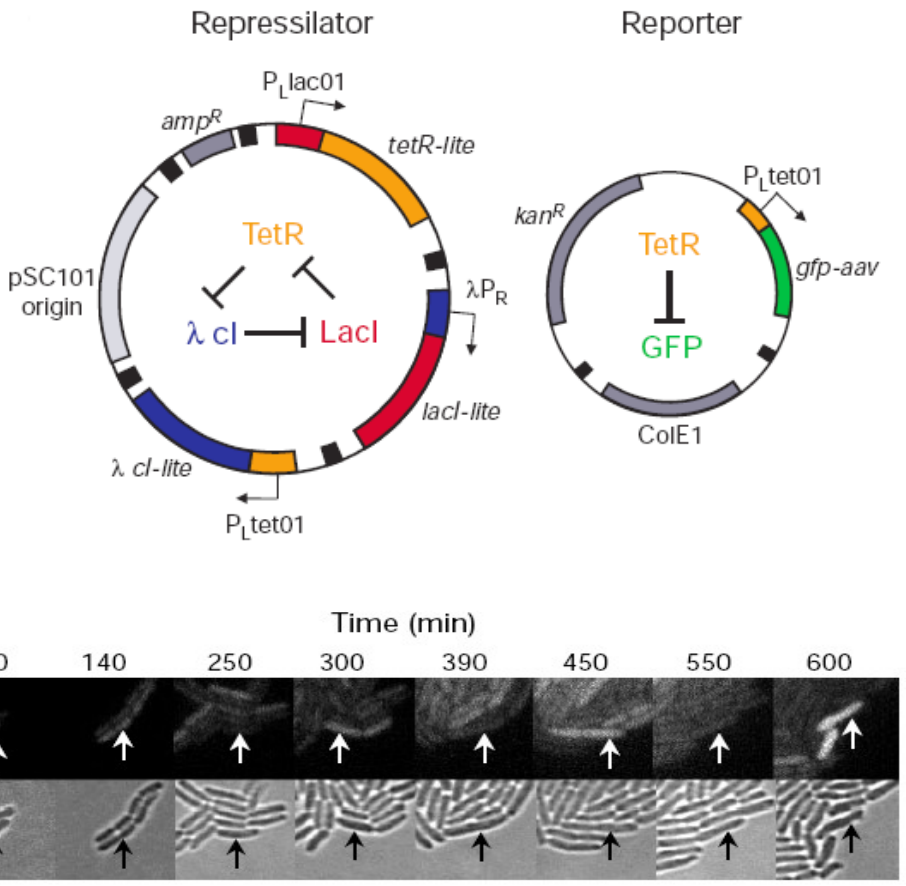
LasR+PAI
luxI
GFP
R0079
J37034
Operador
Autoinducer
GFP


Operon 2
Constitutive
lasr
luxr
J23119
S03156
I0462
Operador
Repressor
Repressor


Operon 3
Operon 4
cI lam
lasI
lux pR
cI lam
mRFP1
R0051
S03154
I13033
J06650
S0103
I13507
Operador
Autoinducer
Operador
Repressor
RFP

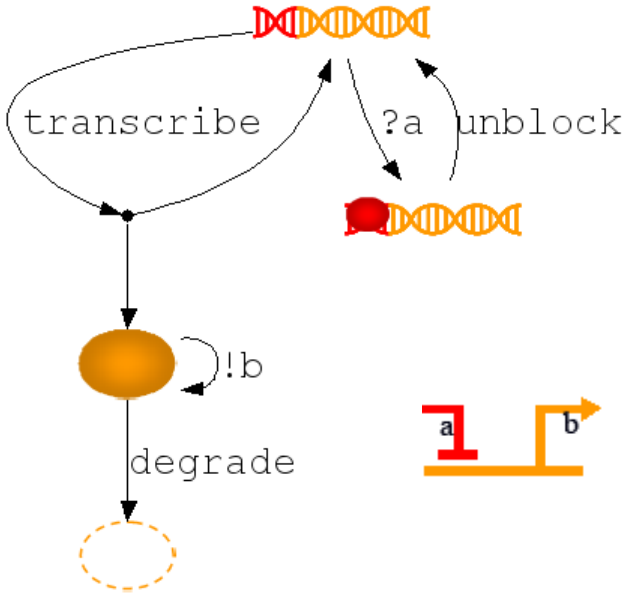

UNAM
X
Constitutive
LuxpR
LasR
λ cI

Stochastic Pi -Calculus



- Stochastic Pi-Calculus as modeling tool for biology.
- Stochastic Pi-Calculus is suitable to model kinetic processes in physical chemistry.
- The simulation algorithm was mapped to functional program code, which was used to implement the SPiM simulator.

Elowitz: Genes with Negative Control

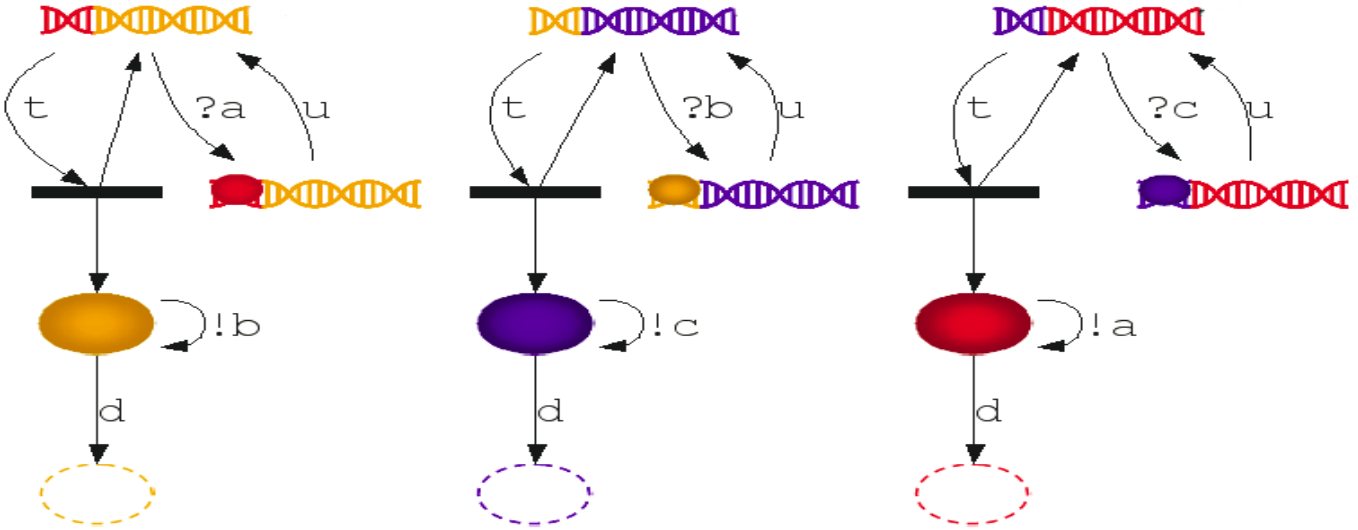


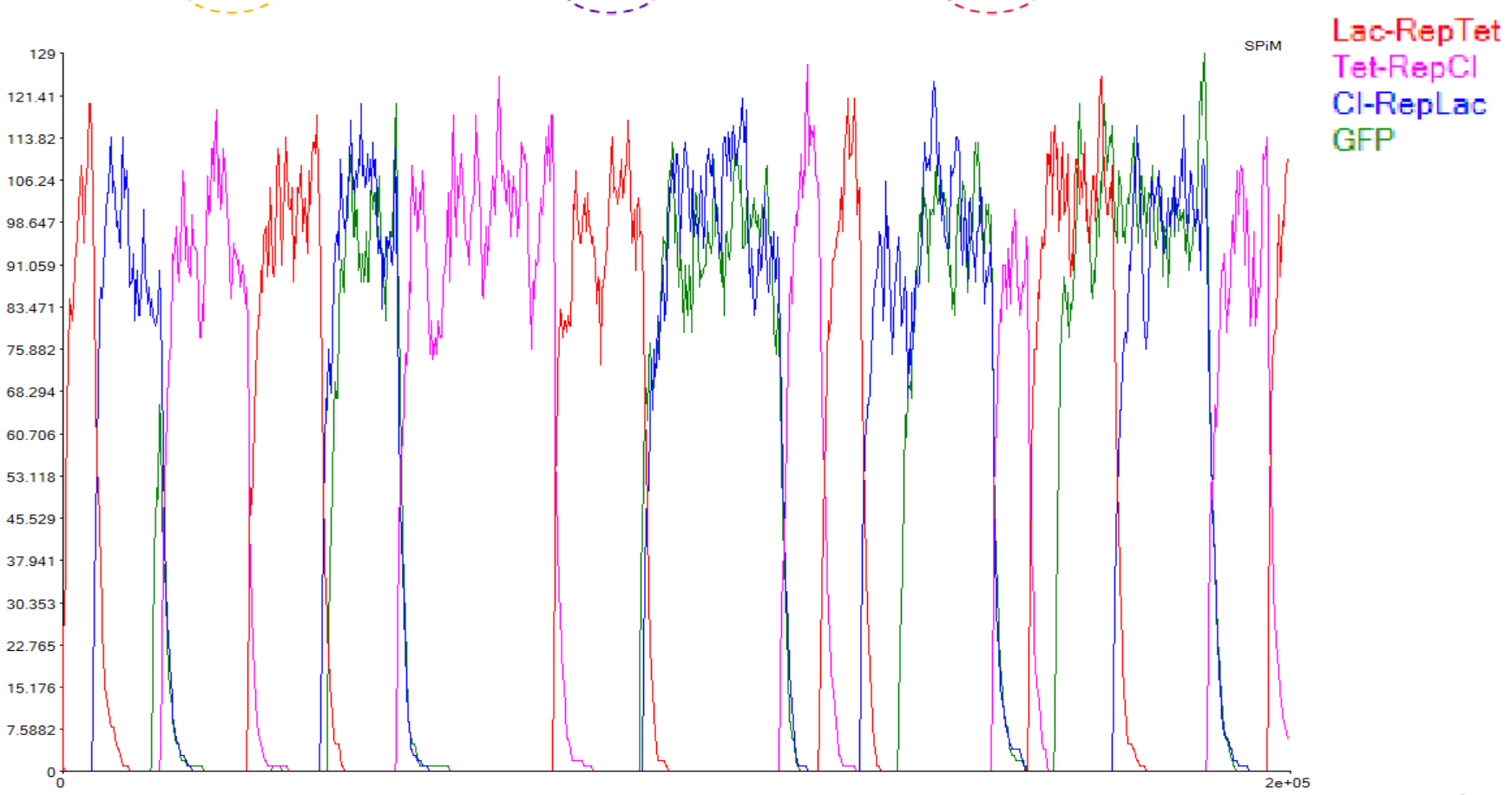
Stochastic Pi Calculus Construction:

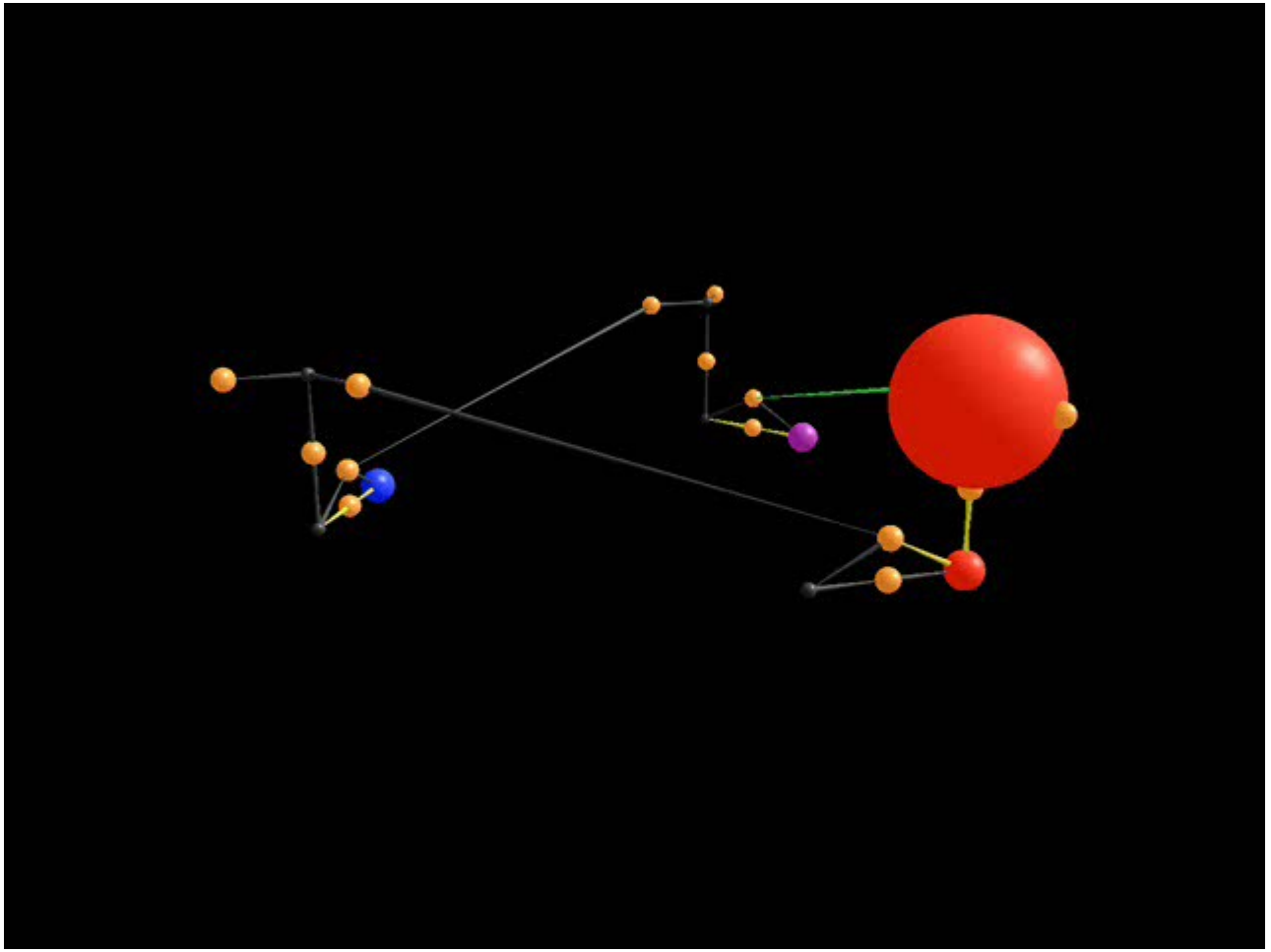
- $g(a,b) = ?a \,.\, g'.(a,b) + \tau_t \,.\, (P(b)|g(a,b)$
- $g'(a.b) = \tau_u \,.\, g(a,b)$
- $P(b) = !b \,.\, P(b) + \tau_d \,.\, 0$

Stochastic Pi-Machine code:

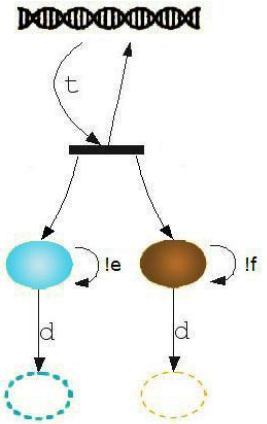
```
val transcribe = 0.1     val degrade = 0.001
val unblock = 0.0001     val bind=1,0

new a@bind:chan          new b@bind:chan
new c@bind:chan

let Neg(a:chan,b:chan) =
  do delay@transcribe;
     (Protein(b) | Neg(a,b))
  or ?a; Blocked(a,b)

and Blocked(a:chan,b:chan) =
  delay@unblock; Neg(a,b)

and Protein(b:chan) =
  do !b; Protein(b)
  or delay@degrade


run Neg(a,b) | Neg(b,c) | Neg(c,a))
```

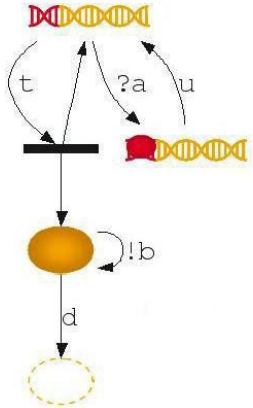


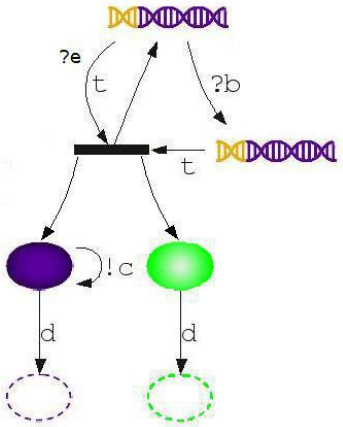


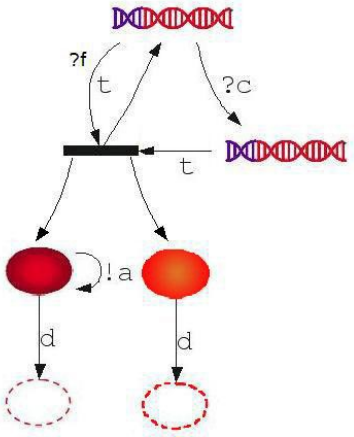


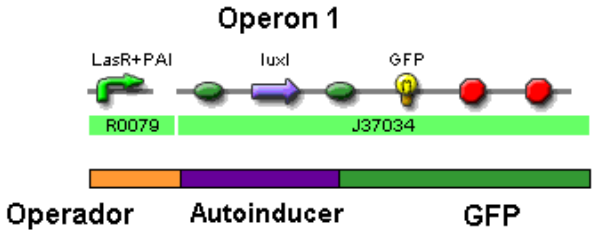
IGEM México Construction

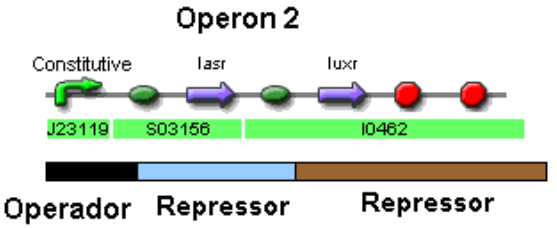


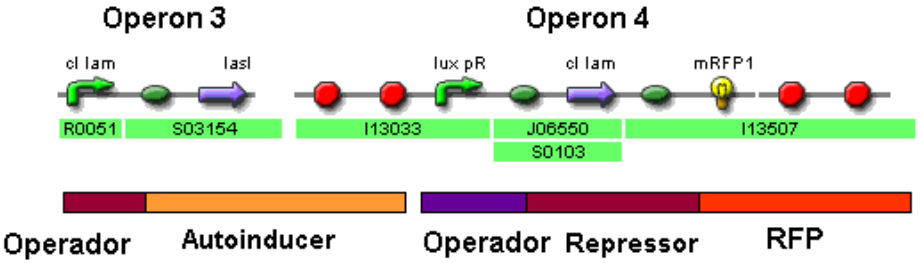


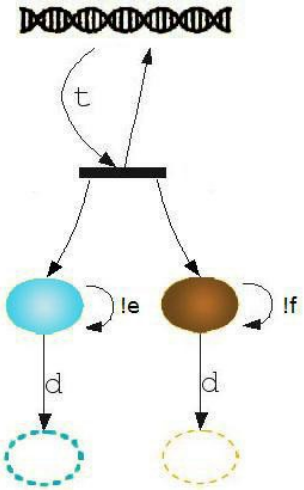


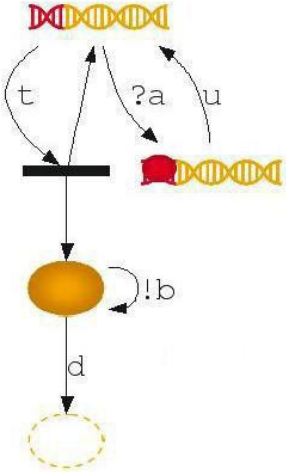


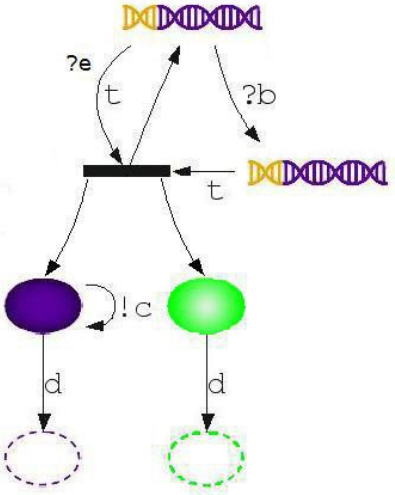


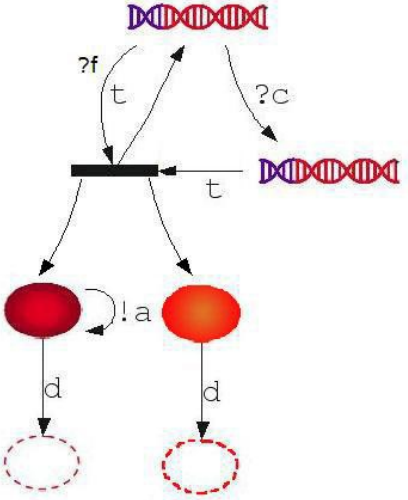






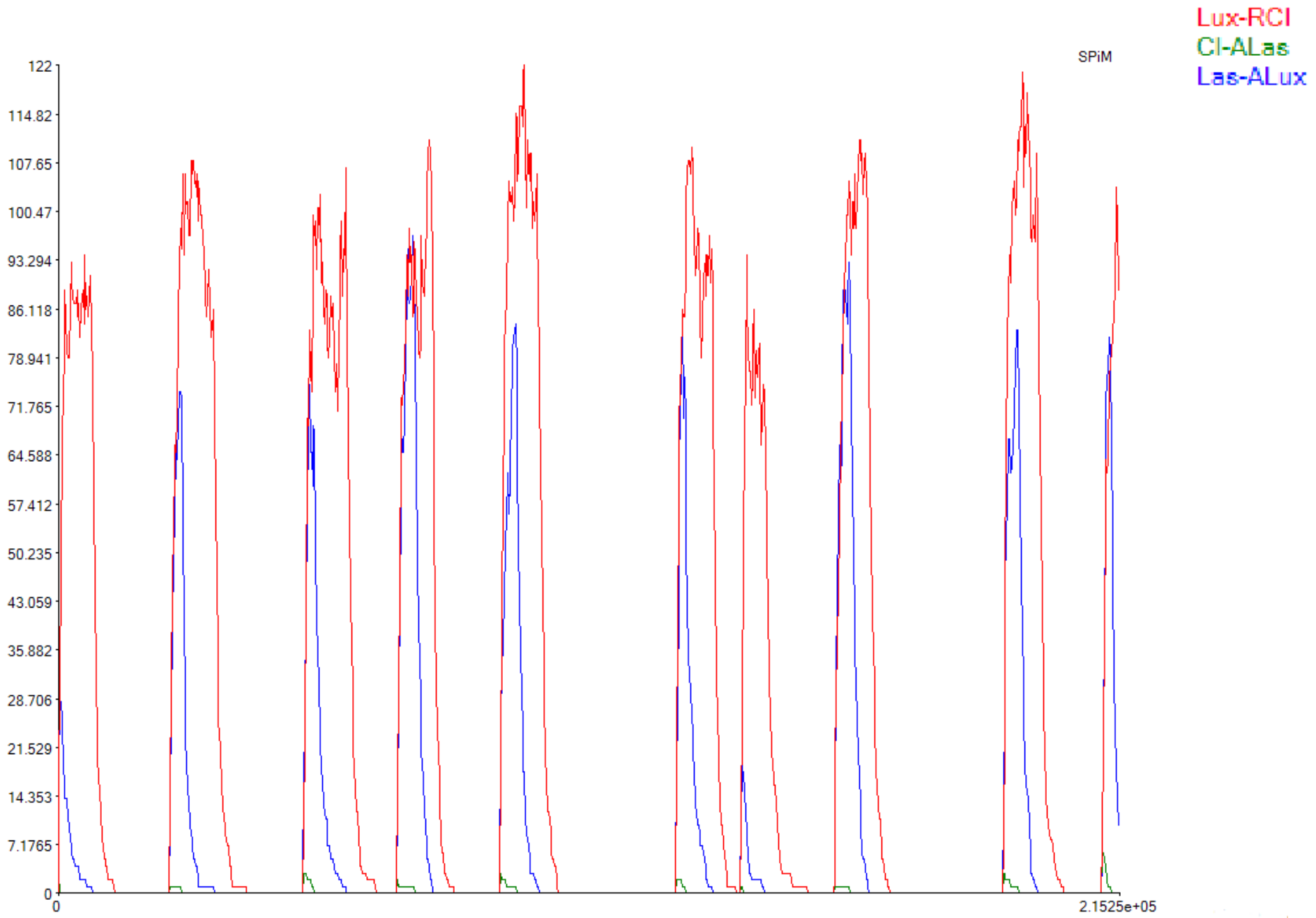


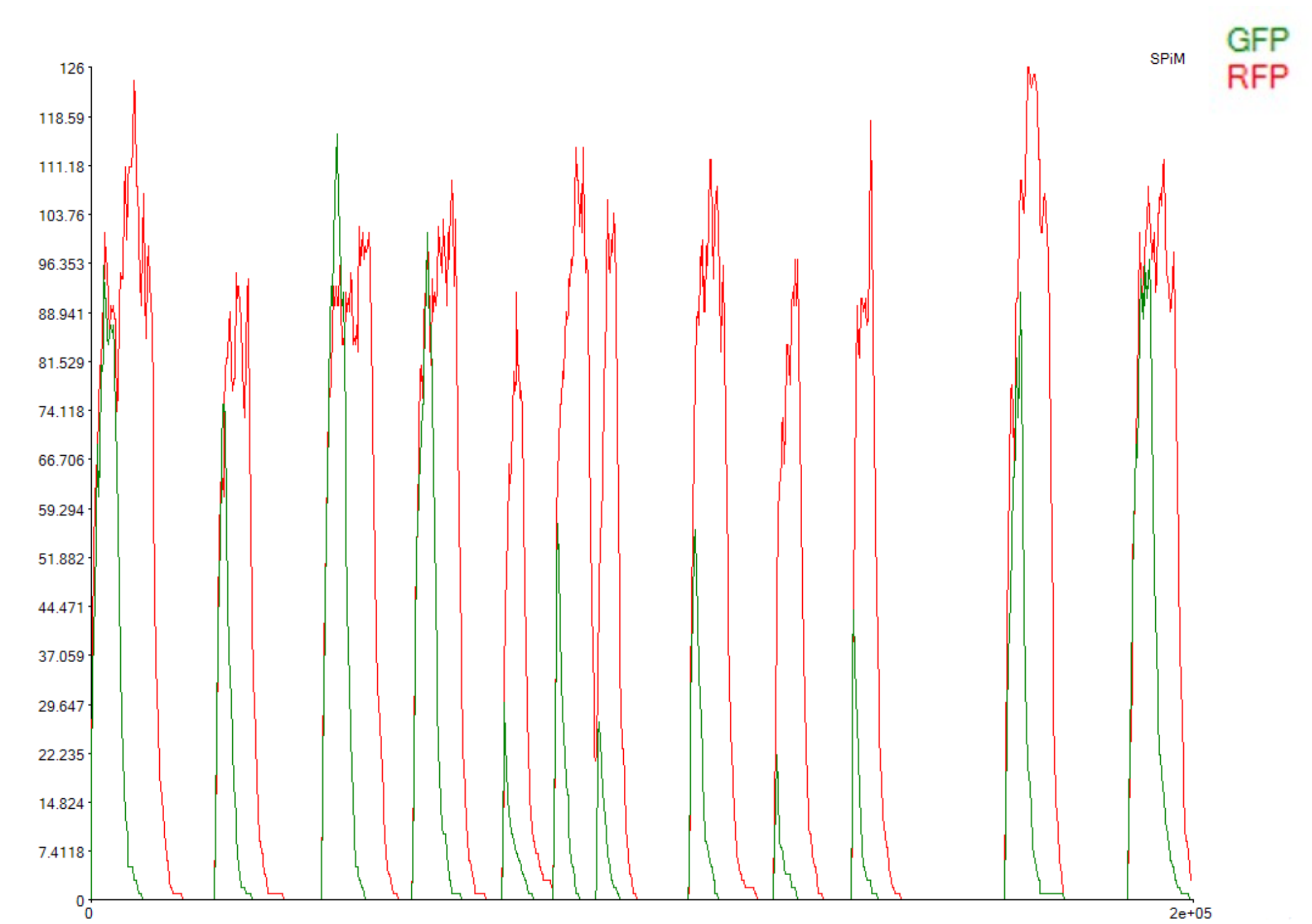




- g(a,b)= ?a . g'.(a,b)
  + τ_t . (P(b)|g(a,b)
  g'(a.b)= τ_u . g(a,b)
  P(b)= !b . P(b)
  + τ_d . 0
- g(b,c)= ?b . g'(b,c)
  + ?e.τ_t . (P(b)|GFP()|g(a,b))
  g'(b,c)= τ_u . (P(c)|GFP()|g(b,c))
  P(c)= !c . P(c)
  + τ_d . 0
  GFP()= τ_d . 0
- g(c,a)= ?c . g'(c,a)
  + ?f.τ_t . (P(a)|RFP()|g(c,a)
  g'(c,a)= τ_u . (P(a)|RFP()|g(c,a))

  P(a)= !a . P(a)
  + τ_d . 0
  RFP()= τ_d . 0
- X()= τ_t . (P(e)|P(f)|X())
  P(e)= !e . P(e)
  + τ_d . 0





Conclusion

- It is reasonable to suggest that the structures we observed in our experiments are in fact Turing patterns. More specifically of the activator-substrate type.

- We still have to measure how the constructions affect the division kinetics of the two strains.

- The simulations indicate that, under certain parameters, our construction might indeed be functional, since the model predicts particular oscillations of protein concentration levels.

- We intend to corroborate these results experimentally; and see if the colony of engineered cells with these construction gets synchronized to obtain Turing Patterns or at least another interesting structures.

References

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