

Pseudomonas Aeruginosa: Virulence vs Biofilm Formation

Laura Brustenga i Moncusí

joint work in progress with Elisenda Feliu

August 18, 2021

KØBENHAVNS
UNIVERSITET



Pseudomonas Aeruginosa

- Ubiquitous
- Multidrug, antibiotic resistant (**biofilm**)
- A leading cause of healthcare-acquired infections
- Considered the paradigm for negative regulation in multikinase networks



Figure: Illustration of *P. aeruginosa* by Kateryna Kon.

Antibiotic resistance: Biofilm formation

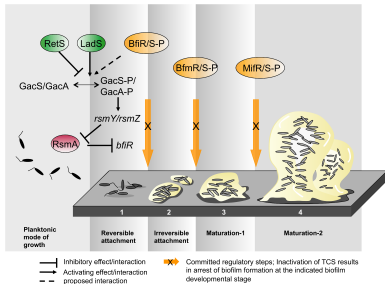


Figure: Schematic illustration of biofilm development of *P. aeruginosa* (CC BY 4.0 (PS09)).

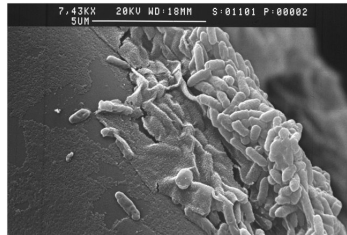


Figure: Formation of *P. aeruginosa* biofilms observed by scanning electron microscopy (CC BY 4.0 (BRA⁺14)).

Pseudomonas Aeruginosa: Biofilm formation

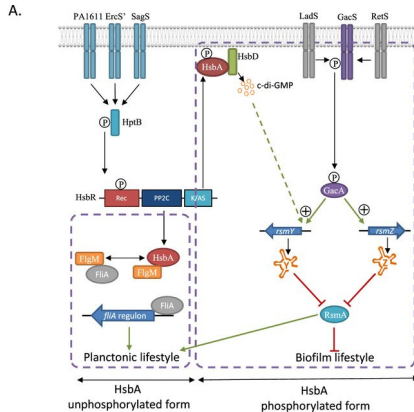


Figure: Schematic illustration of part of the regulatory network of life style control of *Pseudomonas aeruginosa* (CC BY (BBH⁺19)).

RetS inhibits GacS

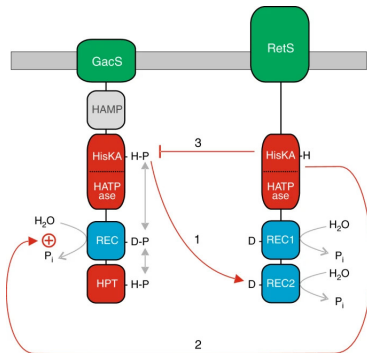
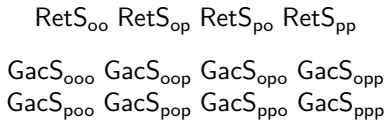


Figure: The three mechanisms used by RetS to inhibit GacS signalling.

(CC BY (FWFJ⁺18)).

Mechanism 1 (complete but too big)

Spices:



Solo intersections RetS:

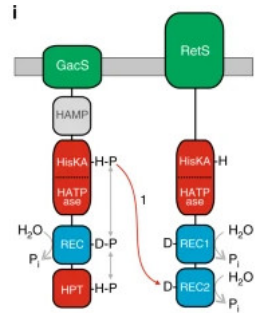
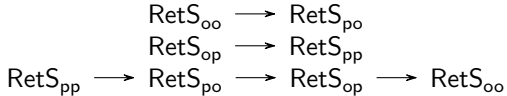


Figure: RetS inhibiting GacS:
Mechanism 1 (CC BY (FWFJ⁺18)).

Mechanism 1 (complete but too big)

Solo intersections GacS:

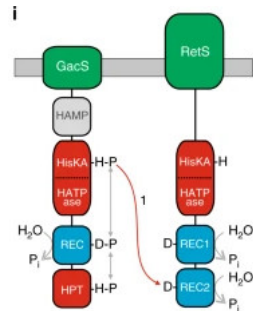
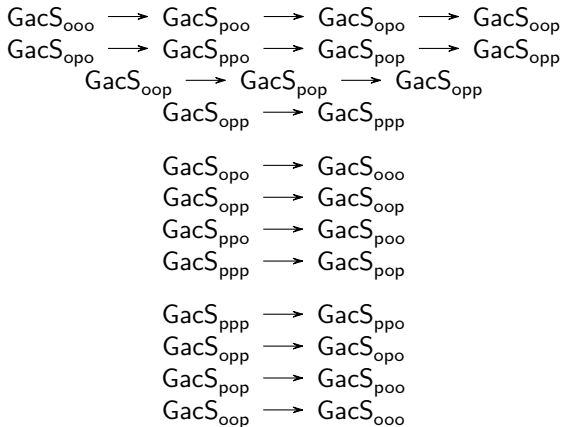
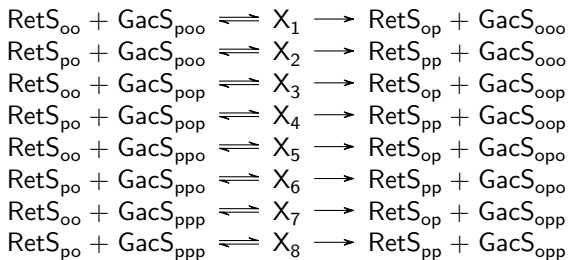


Figure: RetS inhibiting GacS:
Mechanism 1 (CC BY (FWFJ⁺18)).

Mechanism 1 (complete but too big)

Mechanism 1:



We have:

Spices: $4+8+8=20$

Reactions: $5+17+24=65$

Too big for *Real Semi-Algebraic* Geometry!

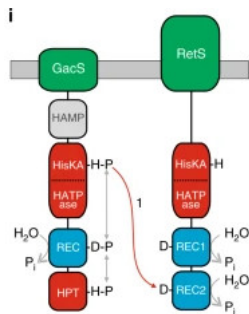
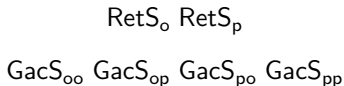


Figure: RetS inhibiting GacS:

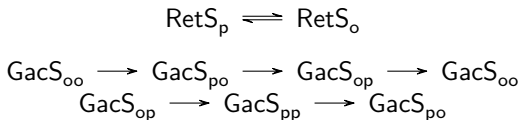
Mechanism 1 (CC BY (FWFJ⁺18)).

Mechanism 1 (simplified)

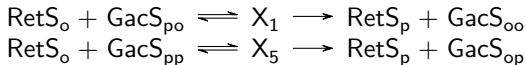
Spices:



Solo reactions:



Mechanism 1:



Spices: 2+4+2=8

Reactions: 2+7+6=15

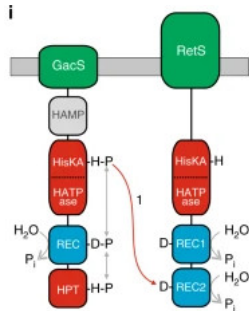


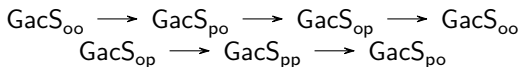
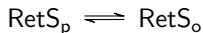
Figure: RetS inhibiting GacS:
Mechanism 1 (CC BY (FWFJ⁺18)).

Mechanism 2 (simplified)

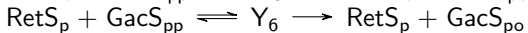
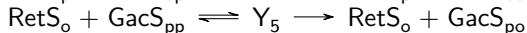
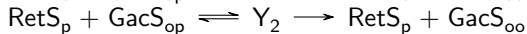
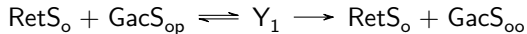
Spices:



Solo reactions:



Mechanism 2:



Spices: 2+4+4=10

Reactions: 2+7+12=21

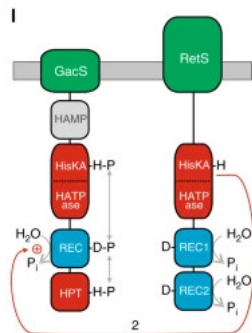
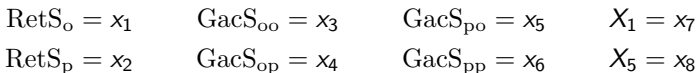


Figure: RetS inhibiting GacS:
Mechanism 2 (CC BY (FWFJ⁺18)).

Equations Mechanism 1



Assuming mass-action kinetics:

$$\dot{x}_1 = -k_{10}x_1x_5 - k_{11}x_1x_6 + k_1x_2 - k_2x_1 + k_{12}x_7 + k_{13}x_8$$

$$\dot{x}_2 = -k_1x_2 + k_2x_1 + k_8x_7 + k_9x_8$$

$$\dot{x}_3 = -k_3x_3 + k_6x_4 + k_8x_7$$

$$\dot{x}_4 = k_4x_5 - k_5x_4 - k_6x_4 + k_9x_8$$

$$\dot{x}_5 = -k_{10}x_1x_5 + k_3x_3 - k_4x_5 + k_7x_6 + k_{12}x_7$$

$$\dot{x}_6 = -k_{11}x_1x_6 + k_5x_4 - k_7x_6 + k_{13}x_8$$

$$\dot{x}_7 = k_{10}x_1x_5 - k_8x_7 - k_{12}x_7$$

$$\dot{x}_8 = k_{11}x_1x_6 - k_9x_8 - k_{13}x_8$$

Equations Mechanism 2

$$Y_1 = x_7$$

$$Y_2 = x_8$$

$$Y_5 = x_9$$

$$Y_6 = x_{10}$$

Assuming mass-action kinetics:

$$\dot{x}_1 = -k_{12}x_1x_4 - k_{14}x_1x_6 + k_1x_2 - k_2x_1 + k_8x_7 + k_{10}x_9 + k_{16}x_7 + k_{18}x_9$$

$$\dot{x}_2 = -k_{13}x_2x_4 - k_{15}x_2x_6 - k_1x_2 + k_2x_1 + k_9x_8 + k_{11}x_{10} + k_{17}x_8 + k_{19}x_{10}$$

$$\dot{x}_3 = -k_3x_3 + k_6x_4 + k_8x_7 + k_9x_8$$

$$\dot{x}_4 = -k_{12}x_1x_4 - k_{13}x_2x_4 + k_4x_5 - k_5x_4 - k_6x_4 + k_{16}x_7 + k_{17}x_8$$

$$\dot{x}_5 = k_3x_3 - k_4x_5 + k_7x_6 + k_{10}x_9 + k_{11}x_{10}$$

$$\dot{x}_6 = -k_{14}x_1x_6 - k_{15}x_2x_6 + k_5x_4 - k_7x_6 + k_{18}x_9 + k_{19}x_{10}$$

$$\dot{x}_7 = k_{12}x_1x_4 - k_8x_7 - k_{16}x_7$$

$$\dot{x}_8 = k_{13}x_2x_4 - k_9x_8 - k_{17}x_8$$

$$\dot{x}_9 = k_{14}x_1x_6 - k_{10}x_9 - k_{18}x_9$$

$$\dot{x}_{10} = k_{15}x_2x_6 - k_{11}x_{10} - k_{19}x_{10}$$

Switch virulence vs biofilm

For mechanism 1, we have that:

$$\begin{aligned}\dot{x}_1 + \dot{x}_2 + \dot{x}_7 + \dot{x}_8 &= 0 \\ \dot{x}_3 + \dot{x}_4 + \dot{x}_5 + \dot{x}_6 + \dot{x}_7 + \dot{x}_8 &= 0\end{aligned}$$

Given the total amounts RetS_{TOT} and GacS_{TOT} , the solutions/dynamics are contained in the affine space:

$$\begin{aligned}x_1 + x_2 + x_7 + x_8 &= \text{RetS}_{\text{TOT}} \\ x_3 + x_4 + x_5 + x_6 + x_7 + x_8 &= \text{GacS}_{\text{TOT}}\end{aligned}$$

“Switch” between biofilm and virulence/planktonic mode:

Question1(multistationarity): Are there more than one steady state on such affine space?

Question2(bistability): Are at least two of them asymptotically stable?

Multistationarity

Proposition

Mechanism 1 is multistationary if and only if

$$k_1 k_3 k_4 k_{13} + k_1 k_3 k_6 k_{13} + k_1 k_4 k_6 k_{13} + k_3 k_4 k_5 k_{10} + \\ k_3 k_4 k_{10} k_{13} + k_3 k_6 k_{10} k_{13} + k_4 k_6 k_{10} k_{13} < k_4 k_5 (k_1 k_{10} + k_3 k_{13} + k_{10} k_{13})$$

Mechanism 2 is multistationary if and only if \mathbf{k} satisfies even a bigger condition.

Proof.

Apply procedure elaborated by Conradi, Feliu, Mincheva and Wiuf in “Identifying parameter regions for multistationarity” (CFMW17).

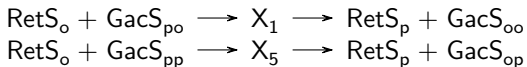
Caution: A parametrization of the steady states variety is required. □

Problem1: Not so insightful conditions.

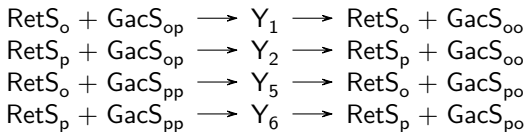
Problem2: Deciding stability is computationally unfeasible.

Even simpler models

Mechanism 1:



Mechanism 2:



Consider all the possible networks without some of the intermediate species X_1 , X_5 , Y_1 , Y_2 , Y_5 or Y_6 .

Multistationarity

Mechanism 1:

Intermediates	Multist.
00	yes
01	yes
10	yes
11	yes

Mechanism 2:

Intermediates	Multist.
00.00	no
00.01	yes
00.10	yes
00.11	yes
01.00	no
10.00	no
11.00	no
01.01	yes
01.10	yes
01.11	yes
10.01	yes
10.10	yes
10.11	yes
11.01	yes
11.10	yes
11.11	yes

Observation: For mechanism 2, it is only RecS inhibiting GacS_{pp} which enables multistationarity!

Now, stability becomes computationally feasible (but for the next talk...).

References I

- [BBH⁺19] Sophie Bouillet, Moly Ba, Laetitia Houot, Chantal lobbi-Nivol, and Christophe Bordi, *Connected partner-switches control the life style of pseudomonas aeruginosa through rpos regulation*, Scientific Reports **9** (2019), no. 1.
- [BRA⁺14] Hayette Benamara, Christophe Rihouey, Imen Abbas, Mohamed Amine Ben Mlouka, Julie Hardouin, Thierry Jouenne, and Stéphane Alexandre, *Characterization of membrane lipidome changes in pseudomonas aeruginosa during biofilm growth on glass wool*, PLoS ONE **9** (2014), no. 9, e108478.
- [CFMW17] Carsten Conradi, Elisenda Feliu, Maya Mincheva, and Carsten Wiuf, *Identifying parameter regions for multistationarity*, PLOS Computational Biology **13** (2017), no. 10, e1005751.

References II

- [FWFJ⁺18] Vanessa I. Francis, Elaine M. Waters, Sutharsan E. Finton-James, Andrea Gori, Aras Kadioglu, Alan R. Brown, and Steven L. Porter, *Multiple communication mechanisms between sensor kinases are crucial for virulence in pseudomonas aeruginosa*, Nature Communications **9** (2018), no. 1.
- [PS09] Olga E. Petrova and Karin Sauer, *A novel signaling network essential for regulating pseudomonas aeruginosa biofilm development*, PLoS Pathogens **5** (2009), no. 11, e1000668.