DISSECTING THE ROLE OF THE SMALL RNA ERS A IN PSEUDOMONAS AERUGINOSA MOTILITY AND BIOFILM REGULATION

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Background
ErsA is a novel Pseudomonas aeruginosa small RNA responsive to infection-relevant host stimuli. Its transcription is under the control of the alternative sigma factor σ²² (AlgT/U), implicated in bacterial virulence. In strain PAO1, ErsA exerts a direct negative post-transcriptional regulation in an incoherent feed-forward loop with σ²² on the bi-functional enzyme AlgC, which is involved in the biosynthesis of sugar precursors for alginate and polysaccharide exoproducts.

Objectives
We aim at the characterization of novel target genes regulated by ErsA to expand the knowledge about the ErsA-based regulatory network.

Methods
We employed different approaches: i) bioinformatics analysis, ii) in vitro electrophoretic mobility shift assays, iii) in vivo translational fusions based on the gfp reporter gene, iv) RNA-seq, and v) biofilm and motility assays on mutant strains.

Conclusions
Our results comprise:

i) In silico analyses identified new putative target genes mainly involved in biofilm formation and exopolysaccharides production.

ii) Among these targets, we specifically validated the interaction between ErsA and amrZ mRNA, both in vitro and in vivo.

iii) The RNA-seq analysis showed that some of the genes whose expression is positively controlled by ErsA are important for biofilm development (i.e. genes belonging to the pel operon, algD).

iv) In line with the other results, the ErsA deletion mutant shows a hyper-motile phenotype compared to the wildtype and develops a thin and flat biofilm.

Overall, our results suggest that the small RNA ErsA might represent a relevant post-transcriptional regulator in biofilm development at different levels, likely interacting with different mRNA targets.