Can HB-PNIPAM Polymer Functionalised with Autoinducer Molecules disturb QS in *Pseudomonas aeruginosa* PA01?

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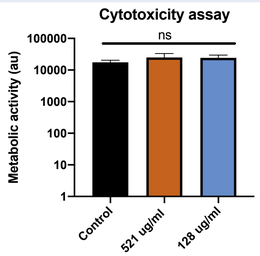
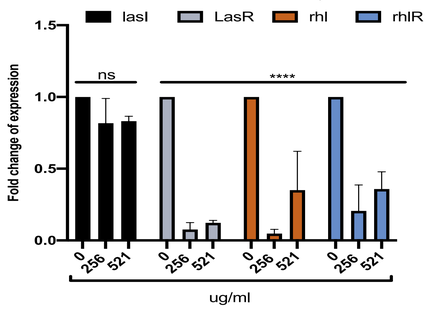
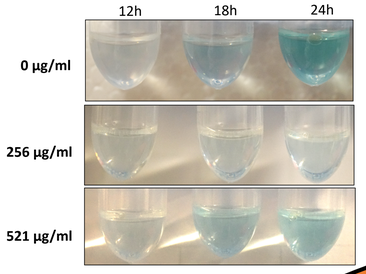
Background: Interfering with intercellular communication quorum sensing (QS) circuits in bacteria could present possible control over several QS-controlled virulence factors important in bacterial biofilm formation. This could limit infection without antibiotic use, circumventing antimicrobial resistance (AMR). We present an anti-quorum sensing polymer, HB-PNIPAM-HL, with uniquely functionalized chain ends targeting the HSL QS pathway in *P.aeruginosa*. **We aim to measure HB-PNIPAM-HL effects on several aspects of QS-controlled virulence in *P.aeruginosa*.**



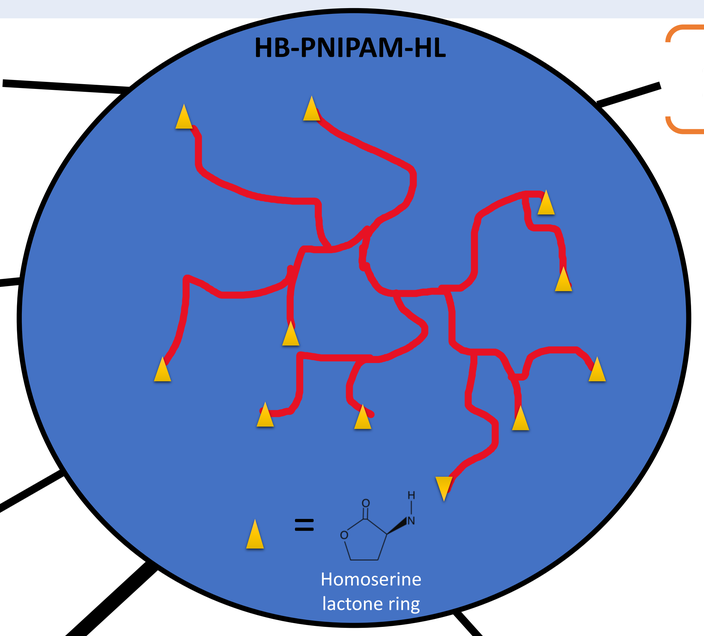
HB-PNIPAM-HL shows no cytotoxicity to human primary fibroblasts. Using the metabolic assay Presto Blue



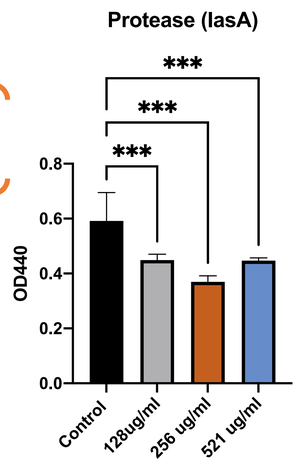
RT-PCR shows that HB-PNIPAM-HL decreases expression levels of QS genes (*lasR, rhlI, and rhlR*) in *P.aeruginosa* at 18h. Normalization with the reference gene *16sRNA*

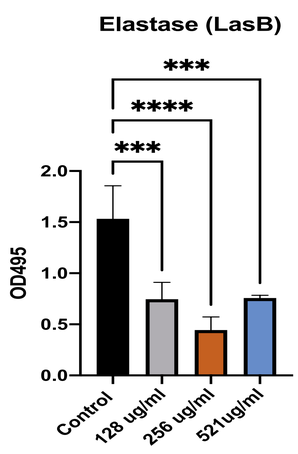


HB-PNIPAM-HL reduces the production of pyocyanin in *P.aeruginosa* after 24h. Pyocyanin has antibacterial properties and increases the fitness of *P.aeruginosa* in competition with other bacterial species.

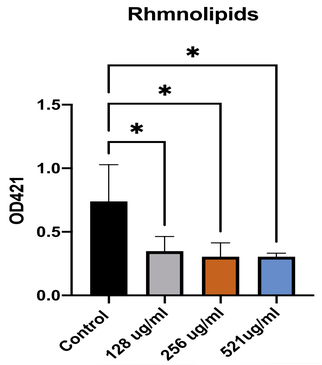


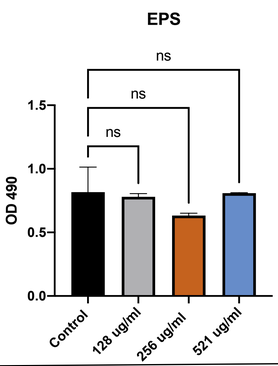
HB-PNIPAM-HL decreases protease and elastase production (LasA & LasB), enzymes that causes epithelial disruption, tissue penetration and endothelial damage, degrade elastin and collagen of host cells and induce tissue injury and haemorrhage. The proteolytic activity of the supernatant was evaluated using 2% azocasein after 18h incubation. Elastase activity of *P.aeruginosa* supernatant was evaluated by elastin Congo red after 18h incubation.





HB-PNIPAM-HL decreases Rhamnolipids production in *P.aeruginosa* after 18h. Rhamnolipids and EPS are important biofilm components that strengthen bacteria against unfavourable environments, i.e., antibiotics exposure.





Conclusions & Future Work

- **Results** suggest that HB-PNIPAM-HL can interfere with HSL-based QS signalling in *P.aeruginosa*.

- **Future work** will include:
  - Assessing **cytotoxicity in keratinocytes** (HaCaT).
  - Measuring the effects of the polymer on *P.aeruginosa* virulence in a **3D tissue-engineered model of skin infection**.
  - Analysing the **transcriptome** of gene expression patterns of *P.aeruginosa* after incubation with HB-PNIPAM-HL.

Acknowledgments

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