

Intra-Microcolony Spatial Positioning Affects Antibiotic Susceptibility in *Neisseria gonorrhoeae*

Abouelkheir, Mahmoud

Bacteria are rarely found alone in natural environments. Instead, they are usually located within complex structures composed of extracellular matrices called biofilms. Many species of bacteria have thin hair like organelles called type IV pilus (tfp), which are integral to bacterial motility and biofilm formation. These tfp are used by bacteria to interact with their environments and other surrounding cells. To understand the role of spatial positioning within microcolony formations, this experiment indirectly observes the diffusion of small molecules (antibiotics) throughout biofilm structures. This is significant in understanding bacterial persistence in nature, and gaining a closer insight into the role of forces within biofilms. This experiment focuses on the the mixing of both wild type *Neisseria gonorrhoeae* (WT GC) and its derivative Δ pilT mutants, and measuring their survival rates post antibiotic selection exposure. WT GC forms tight microcolonies within the center of the mixed formation, whereas Δ pilT GC cells are relegated to the periphery of the formation (Poenisch, 2016). Once the mixed microcolony formation is formed, one hour antibiotic selections are administered at their minimum inhibitory concentrations for GC. Through serial dilution, the mixed microcolonies are plated and cell count is noted. Results heavily suggest spatial positioning is a main factor in antibiotic susceptibility for GC. WT survival rates outpace Δ pilT survival for each antibiotic selection.

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