

IAF MICROGRAVITY SCIENCES AND PROCESSES SYMPOSIUM (A2)  
Interactive Presentations - IAF MICROGRAVITY SCIENCES AND PROCESSES SYMPOSIUM (IP)

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PHENOTYPICAL CHANGES IN ESCHERICHIA COLI FROM CHRONIC EXPOSURE TO  
SIMULATED MICROGRAVITY**Abstract**

Humans have adapted alongside bacteria and will continue to do so while in space. However, the effects of chronic exposure to the space environment on bacteria are still being explored. This work uses a custom 2D clinostat that has been previously validated to simulate microgravity conditions, specifically for bacteria, that operates both the control and microgravity treatments simultaneously. The purpose of this research was to identify the effects of microgravity on *Escherichia coli* while specifically looking at changes in biofilm formation, antibiotic resistance, acidic pH, and osmotic stress. Preliminary data with simulated microgravity (SM) has demonstrated that *E. coli* exhibits permanent phenotypical changes after four days of exposure. To further analyze this trend, biofilm development was measured using two assays (i.e., Crystal Violet and XTT) on *E. coli* samples that were previously on the clinostat for 4 and 22 days. The cells were grown in 6-well plates for 24 hours before the assays were completed. Both assays showed a higher biofilm biomass and metabolic activity from cells grown in SM compared to gravity. Increased biofilm growth has been related to increases in antibiotic resistance. To explore this relationship, the Kirby-Bauer assay was performed with a set of eight antibiotics. Initial findings revealed a significant increase in the resistance to some antibiotics, in direct response to the SM environment. The third experiment, testing acidic pH and osmotic stress tolerance, consisted of growing *E. coli* in nutrient media acidified to pH 4 and with an NaCl concentration gradient ranging from zero to ten percent in one percent increments. Cells grown in SM demonstrated larger growth inhibition compared to control under acidic stress and became completely inhibited after 4