

## EcAMSst: Effect of Space-Flight on Antibiotic Resistance of a Pathogenic Bacterium and its Genetic Basis

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Human immune response is compromised in space and incidence of urinary tract infections (UTI) in astronauts has been reported. We have found that the causative agent of UTI, the uropathogenic *Escherichia coli*, becomes more resistant to gentamicin (Gm), which is commonly used to treat this disease, under modeled microgravity conditions (MMG), the increase being controlled by the stress response master regulator,  $\sigma^s$ . While the wild type bacterium becomes virtually invincible under MMG, the strain missing this sigma factor barely survives. We report here preparatory ground work for testing this finding in space flight on a nanosatellite.

We have shown that the effect of Gm treatment on culture viability is directly correlated to increased Alamar Blue (AB) reduction; we have identified conditions to keep the experimental elements – the bacterial cultures, Gm, and AB – in a state of viability and potency to permit successful spaceflight experimentation given the necessary constraints. Spaceflight kinetics of AB reduction will be transmitted from the satellite via telemetry.

The PharmaSat hardware previously used for space experimentation with yeast was modified to permit studies with bacteria by reducing the filter pore size and increasing fluidics volume to enable more fluid exchanges. Several verification tests have been run using the nanosatellite's flight software and prototype hardware. Cells were grown to stationary phase to induce the  $\sigma^s$ -controlled stress resistance and treated with Gm. Without Gm, the mutant took longer than the wild type to reduce the AB; this time difference increased almost 8 fold at 55  $\mu\text{g/mL}$  Gm concentration. Thus, using flight hardware the mutant shows similarly increased sensitivity to Gm compared to the wild type to that found in our pilot microtiter plate experiments. Previous inflight experiments have given contradictory results concerning bacterial antibiotic resistance; none has yet explored the involvement of specific genes in this phenomenon. With our system ready to fly in late 2015/early 2016, these questions can be approached.

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