

Title: Determination Of Bile Acid Tolerance Of *Clostridium sporogenes* and *Clostridioides difficile*

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*Clostridioides difficile* infection (CDI) is the leading cause of nosocomial infection in the United States, impacting nearly half a million people each year. Antibiotics are a major risk factor for CDI as they alter the gut microbiota and decrease colonization resistance to this pathogen. It is hypothesized that commensal *Clostridia* are able to prevent *C. difficile* growth due to their ability to make inhibitory secondary bile acids from primary bile acids. The bile acid-inducible (*bai*) operon is necessary to carry out 7 $\alpha$ -dehydroxylation of deconjugated primary bile acids – cholic acid (CA) and chenodeoxycholic acid (CDCA) thereby converting them into the secondary bile acids deoxycholic acid (DCA) and lithocholic acid (LCA), respectively. A recent study was able to knock in the *bai* operon into *C. sporogenes* or MF001. To determine the relationship between *C. sporogenes* wildtype (WT), MF001, and *C. difficile*, we will measure the minimum inhibitory concentrations (MICs) and growth kinetics of these strains in the presence of bile acids CA, CDCA, DCA, and LCA at different concentrations. *C. sporogenes* WT and MF001 have similar MICs in CA (>10 mM), CDCA (0.96 mM), and DCA (1.25 mM). Whereas, *C. difficile* has different MICs in CA (~5 mM), and CDCA (~0.625 mM). These MICs will inform future experiments that will determine how *C. difficile* growth is affected by secondary bile acids made by MF001. These findings will help further our understanding of how commensal bacteria are able to inhibit *C. difficile* in the gut.