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Recommended Citation

Roser, Andrew; Tracy, Declan; Johnson, Henry; and Giorno, Rebecca, "Role of Spore-Associated Inosine-Uridine Nucleoside Hydrolase IunA in Bacillus anthracis Spores" (2019). ANS Research Symposium. 42.

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Role of spore-associated inosine-uridine nucleoside hydrolase IunA in *Bacillus anthracis* spores

Andrew Roser¹, Declan Tracy², Henry Johnson³, and Rebecca Giorno⁴

Bacillus anthracis spores are the infective particle in anthrax disease. The three outer layers of the spore that we study (exosporium, interspace, and coat) are involved in germination which influence our ability to decontaminate environments safely. A combination of two germinants, a nucleoside and an amino acid, trigger germination in B. anthracis. The metabolically inactive spores contain enzymes, one of which, called inosine-uridine-preferring nucleoside hydrolase (IunH), breaks down the cogerminant inosine into D-ribose and hypoxanthine. A putative inosine-uridine nucleoside hydrolase, IunA, has been identified and its impact, along with IunH, on germination and spore associated inosine hydrolase activity was studied. Spore-associated inosine hydrolase activity and germination kinetics were measured in wild-type, iunA, and iunH mutant spores and compared. iunH mutant spores lack hydrolase activity while the iunA mutant spores have reduced activity. Preliminary data suggests both iunH and iunA mutant spores have an enhanced germination rate compared to wild-type. The results show that IunA does have an impact on spore-associated inosine hydrolase activity and spore germination, but to a lesser extent than IunH. iunA mutant spores have an exosporium assembly defect. Given that IunH is also an exosporium protein, it is unclear if the resulting iunA phenotypes are due to the absence of IunA or improper assembly of other exosporium proteins such as IunH. Clarifying the role of IunA as either an enzyme or structural protein will affect future studies related to identifying inosine-uridine hydrolase inhibitors that may help to design better therapeutics and decontamination strategies.

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