Development of a CRISPR-based antimicrobial for the targeted destruction of plant pathogens.

W. Evan Braswell[1], Jennie Fagen[2], and Chase Beisel[2]

[1]U.S. Department of Agriculture APHIS PPQ CPHST Mission Laboratory, Edinburg, Texas, USA

[2]Department of Chemical and Biomolecular Engineering, North Carolina State University, Raleigh, North Carolina, USA

Abstract: Over 80 years ago, penicillin changed the world; no longer were minor infections a likely cause of death. Today, the United Nations has declared antimicrobial resistance to be a world health crisis with the Secretary General stating that antimicrobial resistance represents a "fundamental, long term threat to human health, sustainable food production and development". Overuse in medicine and agriculture has led to the evolution of resistance genes that confer resistance to all known antimicrobials. Worse, the broad-spectrum action of current antimicrobials induce substantial selection pressure on diverse bacterial community members along with the intended pathogenic target thereby accelerating the rate of resistance evolution. In an effort to minimize use of traditional antimicrobials, we present an alternative model for antimicrobial action based the bacterial immune system CRISPR-Cas. We review the mechanisms by which CRISPR-Cas systems act and present a framework for converting them into antimicrobial systems for targeted destruction of plant pathogens. This approach is aimed at circumventing current modes of resistance and restricting action to individual species. We review data demonstrating the specificity and effectiveness of these systems in *E. coli* and discuss progress made towards this goal by focusing on efforts to target *Candidatus* Liberibacter asiaticus, the causative agent of Huanglongbing, and *Xylella fastidiosa*, the causative agent of Pierce's disease. Finally, we discuss the challenges of, and propose solutions to, delivery of such a novel antimicrobial to the site of infection.