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Dear Editor,

Enclosed is a new submission of a manuscript entitled: "Method for marker-less gene deletion by Floxed-Cassette Allelic Exchange Mutagenesis (FLAEM) in *Chlamydia trachomatis*" to be considered for publication in JoVE.

*Chlamydia* spp are important human pathogens whose obligate intracellular lifestyle has complicated direct genetic manipulation. Herein, we describe the protocol for Floxed-Cassette Allelic Exchange Mutagenesis that allows targeted marker-less gene deletion in *Chlamydia trachomatis*. This method utilizes the Cre-lox system to excise the selection cassette after allelic exchange mutagenesis. We have developed this method to alleviate cassette-induced polar effects, and to our knowledge, it is the first application of the Cre-lox system in a bacterium where cultivation in host cells is required. We feel the protocol will be useful to both chlamydiologists and a broad range of researchers. We look forward to hearing from you.

Thank you for your consideration.

Sincerely,

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