



Heritable gene editing by targeted delivery of Cas9 nuclease to the germline in bumble bees (*Bombus impatiens*)

Author(s): Rong Ma, Rong Ma , Vanessa M. Macias , Allyson M. Ray , Christina M. Grozinger , Jason L. Rasgon

Institution(s): Department of Entomology, Pennsylvania State University, University Park, PA, USA ; Department of Entomology, Pennsylvania State University, University Park, PA, USA ; Department of Entomology, Pennsylvania State University, University Park, PA, USA ; Department of Entomology, Center for Pollinator Research, Pennsylvania State University, University Park, PA, USA ; Department of Entomology, Pennsylvania State University, University Park, PA, USA ; Department of Entomology, Pennsylvania State University, University Park, PA, USA

CRISPR/Cas is a powerful genome engineering tool currently available in a limited range of model plants and animals. Current approaches rely on the delivery of Cas9 ribonucleoprotein complex to arthropod eggs by embryonic microinjection. However, embryonic microinjection is challenging, limited to a small number of species amenable to laboratory rearing, and inefficient even in optimized taxa. Receptor-Mediated Ovary Transduction of Cargo (ReMOT Control) is a newly-developed technique that allows straight-forward and flexible use of CRISPR/Cas9-mediated genome modification. Using ReMOT Control, the Cas9 ribonucleoprotein complex has been successfully delivered to the insect germline by injection into adult female mosquitos, resulting in “in utero” editing of the developing germline. We are currently expanding the technology into a broad spectrum of arthropod systems, including bumble bees (*Bombus impatiens*) and honey bees (*Apis mellifera*). Optimization of ReMOT Control technologies will allow researchers to genetically manipulate diverse insect species—including those where egg-injection and rearing are particularly challenging—with greater efficiency and lower costs, thereby greatly expanding the potential for functional genomics studies.