

# Optimizing CRISPR-Cas9 genetic modification in *Phyllobates* poison-dart frogs

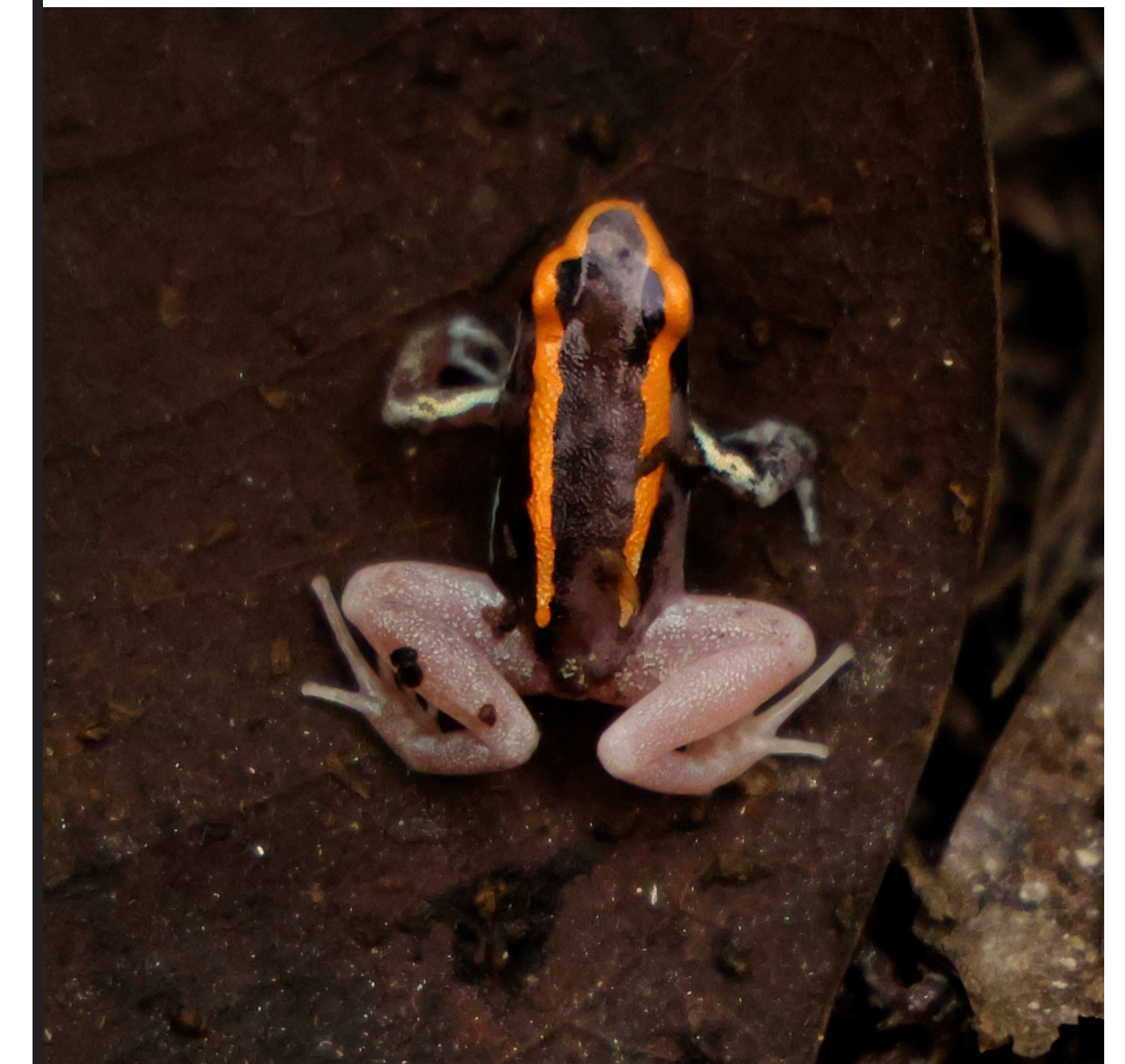
Chris Talbot, Roberto Márquez

Department of Ecology and Evolutionary Biology, University of Michigan

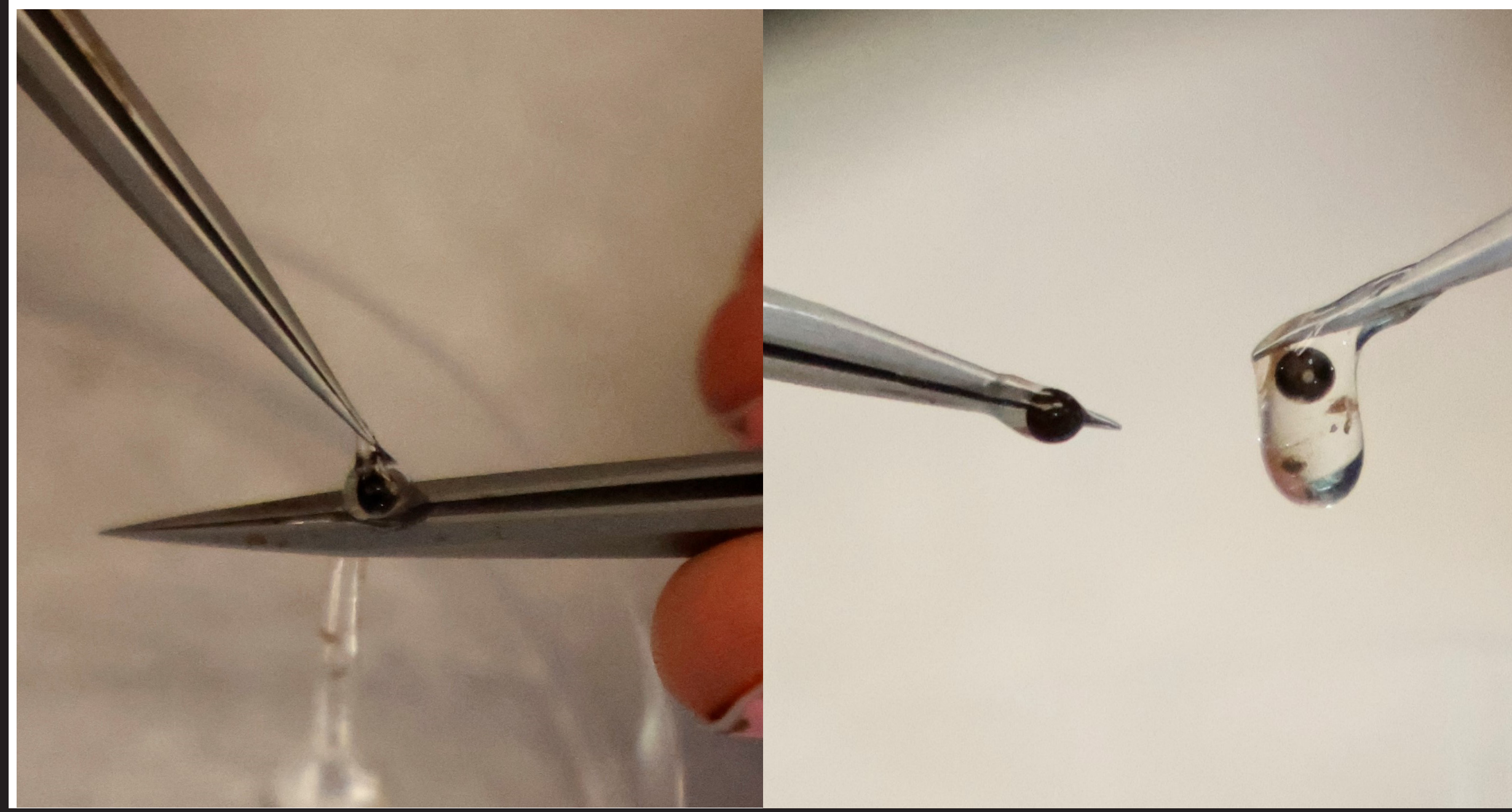


## Introduction

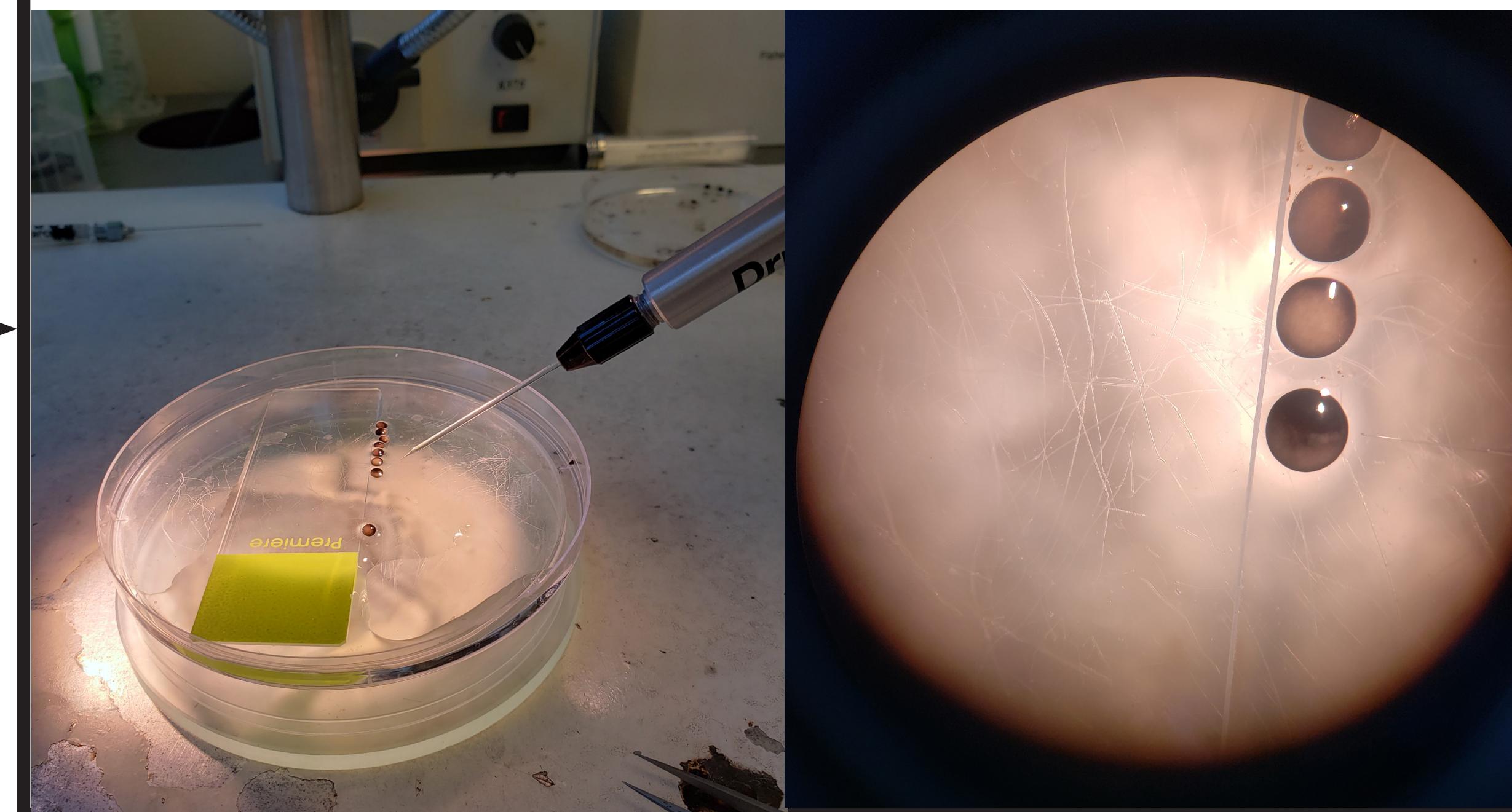
- Genetic modification allows us to explore the connection between genotype and phenotype.
- CRISPR-Cas9, a bacterial system for defense against bacteriophages, can be used to perform gene knockouts in embryos.
- CRISPR is relatively quick and cheap, allowing for genomic experimentation in non-model organisms.
- Knocking out *slc45a2* gene is known to create amelanistic frogs, allowing us to easily identify successfully modified frogs.
- Creating a procedure for CRISPR-Cas9 knockouts in *Phyllobates* may be generalizable to other non-model amphibians.



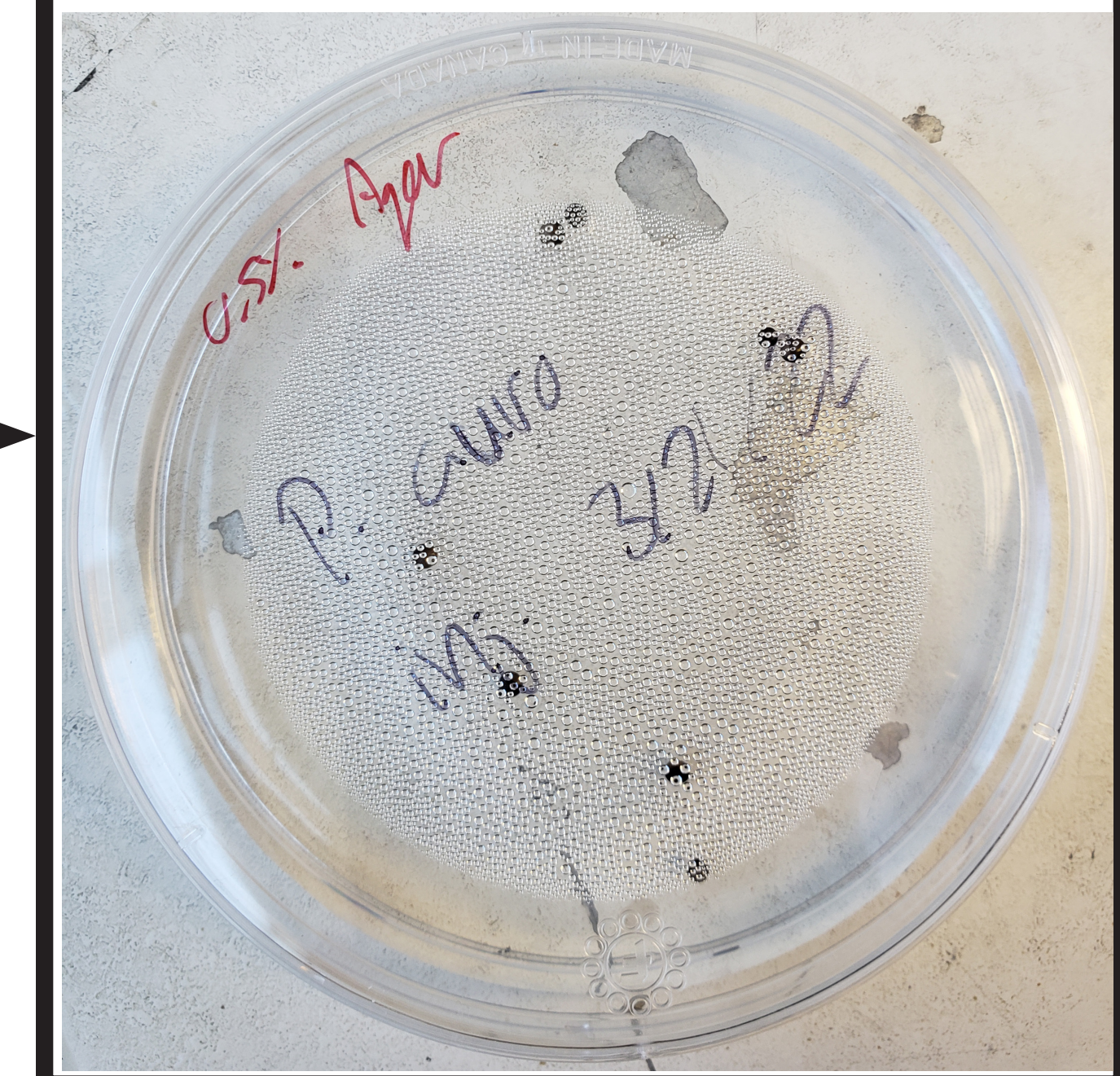
## Dejellinging



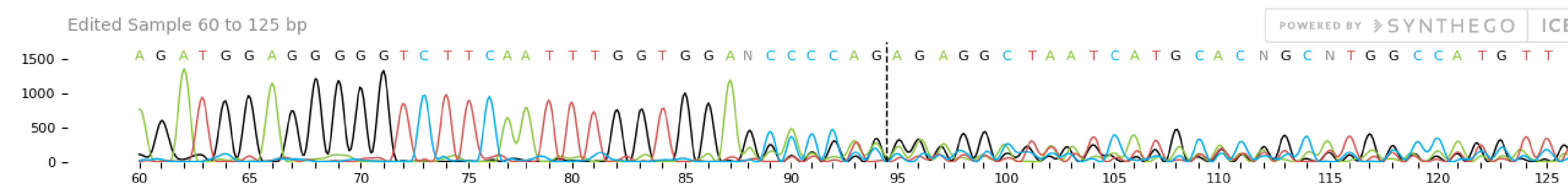
## Microinjection



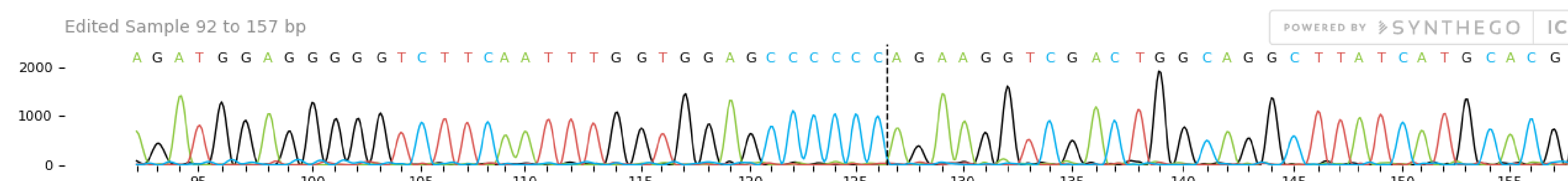
## Rearing



## Results



Sometimes our methods work, cutting the genome in the expected location...



But we are still seeing inconsistencies and need further experimentation to determine the cause.

## Future directions

Evaluate trade-offs between genome cutting efficiency and long-term survivorship for varying concentrations of CRISPR-Cas9 solution.

## Acknowledgements

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