Electroporation delivery of CRISPR/Cas9 system improves efficiency and throughput

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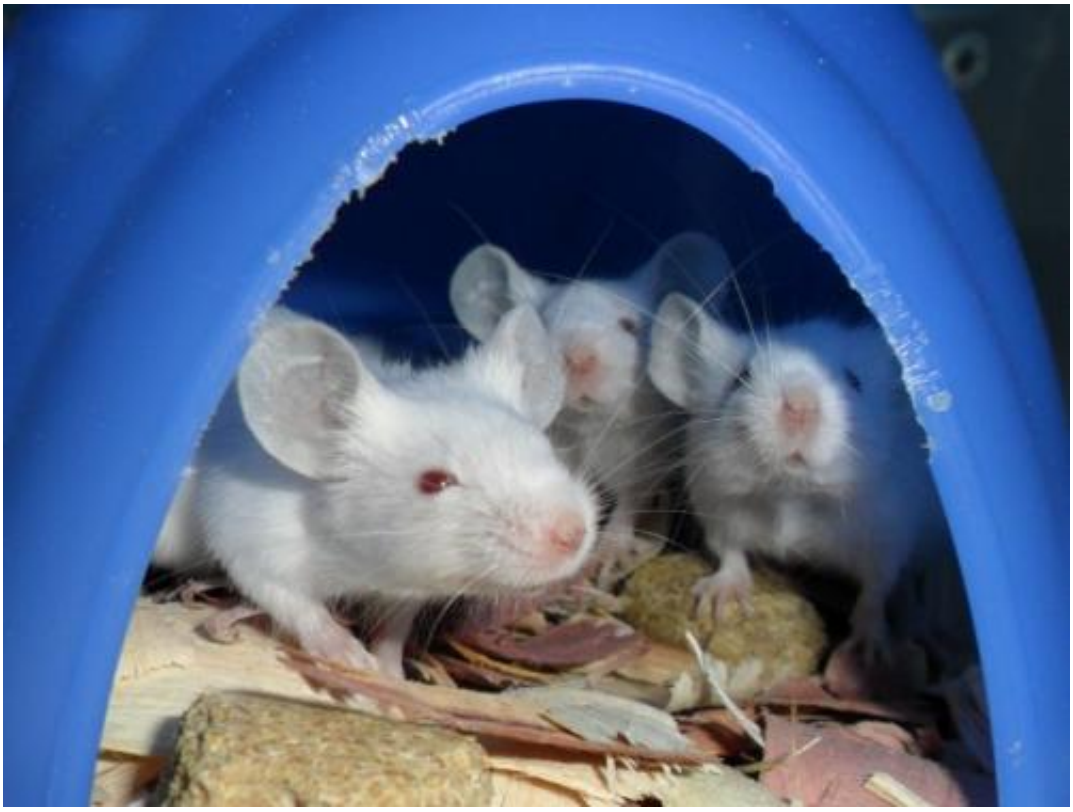

Credit: Martha Sexton/public domain

Jackson Laboratory researchers have shown that using an electric current to deliver the CRISPR/Cas9 system, in order to engineer genetic changes in laboratory mice, is highly efficient and significantly improves the system's throughput.

CRISPR/Cas9 has significantly enhanced the precision, speed and ease with which experimental organisms can be genetically modified in order to create models of human diseases. Mice carrying mutations in single or multiple genes or other modifications can be created in one step by injecting the CRISPR/Cas9 system into zygotes (the cells formed by the union of egg and sperm that proceed to divide and create a living organism). The technique is effective, but it requires microinjection, a technically demanding and time-consuming process that can only be done one zygote at a time.

In a paper highlighted in the June issue of the journal *Genetics*, Distinguished Visiting Professor Haoyi Wang, Ph.D., JAX associate director of genomic engineering technologies Wenning Qin, Ph.D., and colleagues demonstrated that electroporation—using an electric current to increase the permeability of the cell membrane—can be a faster, higher throughput alternative to microinjection.

"Development of the electroporation protocol brings significantly higher efficiency and higher throughput genome engineering in animal models," says Wang, who as a postdoctoral associate in the laboratory of Rudolf Jaenisch at MIT was involved in the early development of CRISPR technology. "Through this technique we were able to dramatically increase processing from one zygote at a time through microinjection to 20 to 50 zygotes simultaneously."

Thanks to this increase in efficiency, Wang adds, "we may now be in a position to generate mouse models of human disease alleles with unprecedented efficiency and throughput."

The researchers first used a mild acid solution to weaken the zona pellucida, a protective layer surrounding the zygote's membrane, and then tested several voltages to find one that delivers the CRISPR/Cas9 components inside the cell without causing it harm.

In addition to faster throughput, the researchers' electroporation technique may also be less invasive and damaging to the zygotes, as they achieved a birthrate comparable to that of untreated zygotes and nearly twice as high as the injected zygotes. The researchers are currently working to further refine the technique and optimize it for a variety of inbred mouse strains.

More information: Qin et al.: Efficient CRISPR/Cas9-Mediated Genome Editing in Mice by Zygote Electroporation of Nuclease. *Genetics*, June 2015, dx.doi.org/10.1534/genetics.115.176594

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