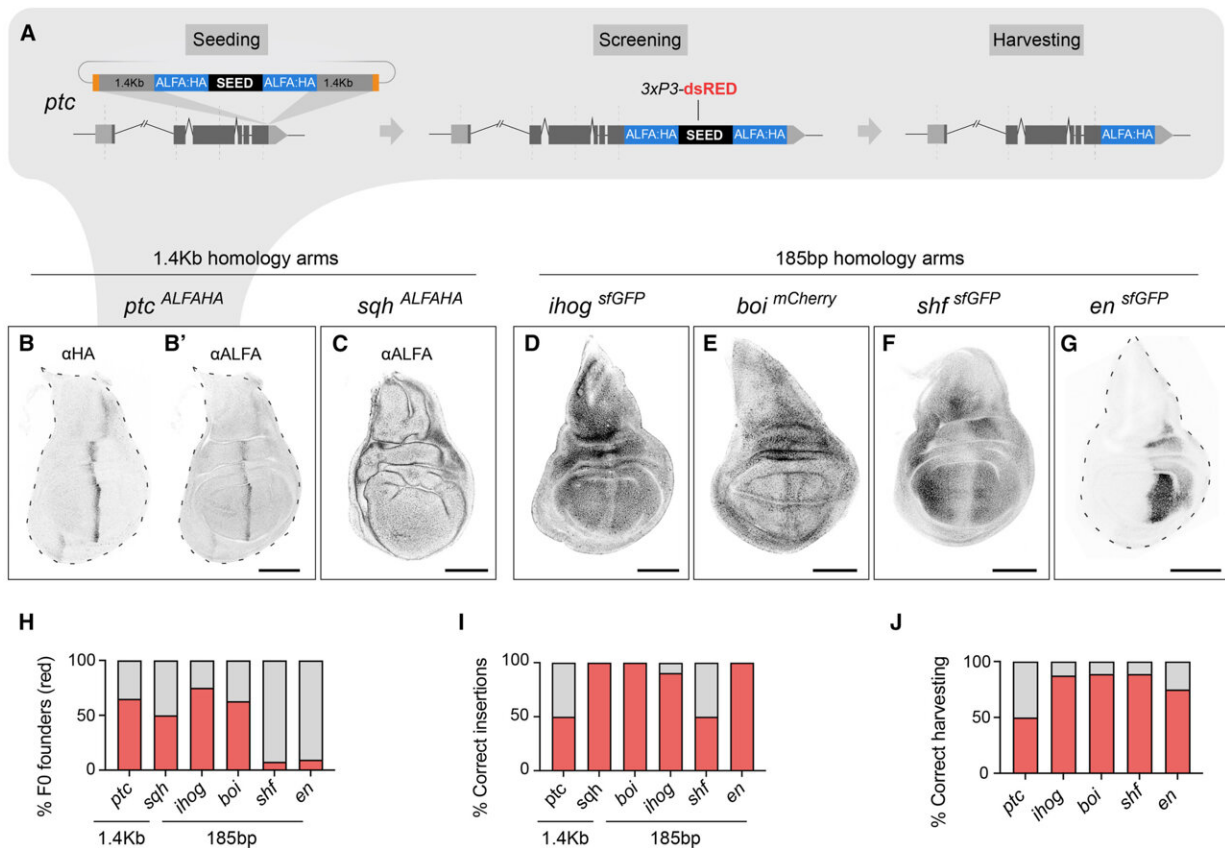


Precise genetics: New CRISPR method enables efficient DNA modification

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Precise knockin generation using SEED/Harvest. Credit: *Developmental Cell* (2024). DOI: 10.1016/j.devcel.2024.06.004

With the revolutionary CRISPR/Cas technology, the DNA of living organisms can be precisely altered. Using a guide RNA that recognizes a

specific DNA sequence, Cas9 protein is recruited to that sequence and cuts the DNA. This targeted cut allows the DNA to be repaired or altered at this specific location.

Prof. Markus Affolter's team at the Biozentrum, University of Basel, has now developed a new method called SEED/Harvest in the fruit fly (*Drosophila melanogaster*). This method combines the CRISPR-Cas9 technique with the Single-Strand Annealing (SSA) [repair pathway](#), enabling genome-wide changes to be carried out more efficiently and without leaving unwanted scars. The study has been [published](#) in *Developmental Cell*.

The SEED/Harvest method proceeds in two steps. In a first step, the researchers introduced a [marker gene](#) into the desired DNA site within a protein-coding region. This marker is placed at the targeted location and is used to isolate successful modifications.

In a second step, the marker is excised and the DNA breakpoints are repaired by the Single-Strand Annealing (SSA) repair pathway. "This enables us to cut the DNA seamless while maintaining its full function," explains first author Gustavo Aguilar. "The combination of both methods makes it possible to mark any desired [protein](#) in the genome without [collateral damage](#), allowing us to study the functions of proteins in living organisms."

"Since we would like to introduce and analyze changes in the DNA throughout the genome for our research, the method must be both precise and efficient," explains Affolter. "And the SEED/Harvest method is both. It combines the most robust screening of successful insertions and all the advantages of seamless tagging."

One of the advantages of the SEED/Harvest method is that proteins can be labeled in specific tissues and cell types. "We can now control and

determine in various tissues and developmental stages when and where genes are activated or inactivated," adds Aguilar. This opens up new possibilities for research to investigate the dynamics of proteins systematically in living cells in real-time.

This method is not only significant for genetics and biotechnology. "The SEED/Harvest method could also be of interest for [medical research](#), for example, to identify defects caused by disease genes," says Affolter.

More information: Gustavo Aguilar et al, Seamless knockins in *Drosophila* via CRISPR-triggered single-strand annealing, *Developmental Cell* (2024). [DOI: 10.1016/j.devcel.2024.06.004](https://doi.org/10.1016/j.devcel.2024.06.004)

Provided by University of Basel

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