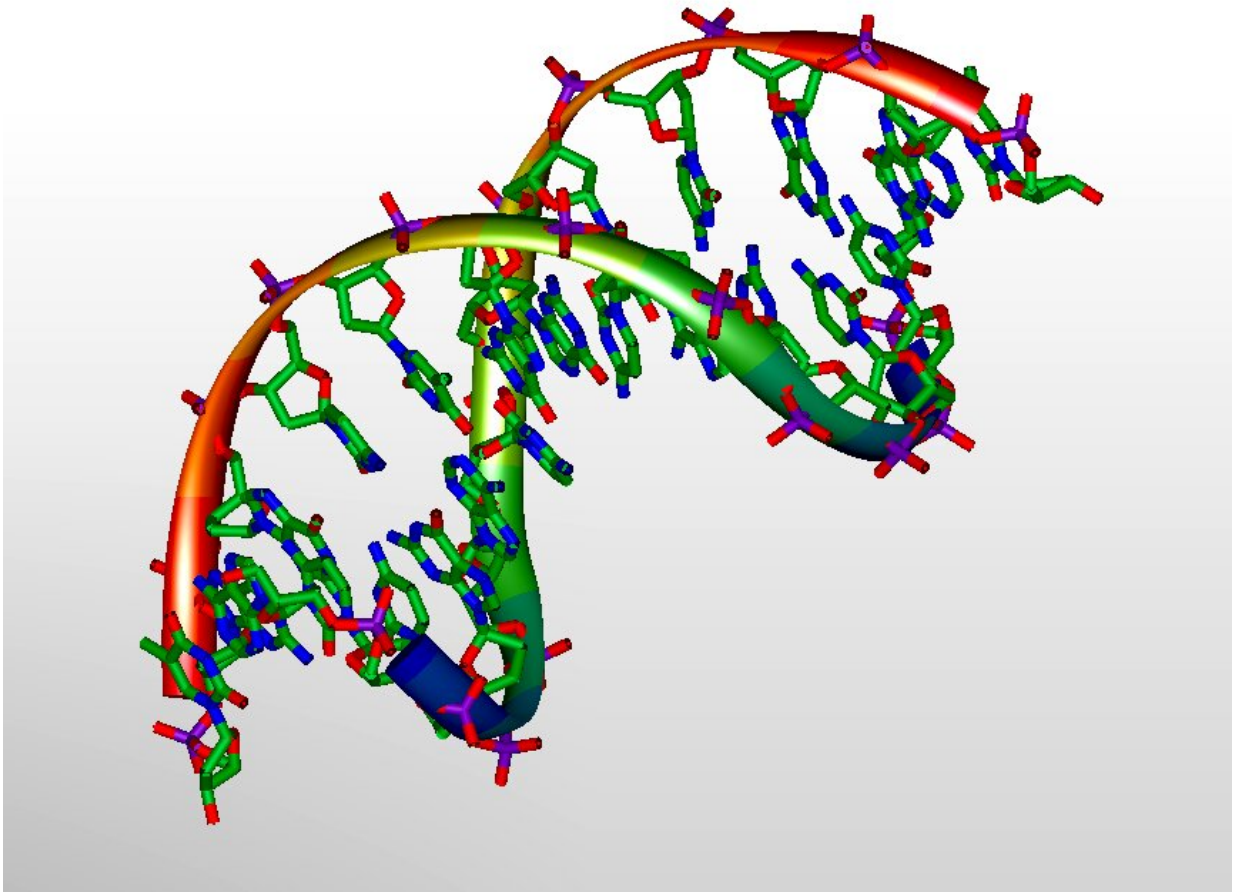
New gene-editing technique offers scientists ability to 'turn on' enzymes that cause DNA base mutations

October 28 2021



3D-model of DNA. Credit: Michael Ströck/Wikimedia/ GNU Free Documentation License

Targeted mutations to the genome can now be introduced by splitting specific mutator enzymes and then triggering them to reconstitute, according to research from the Perelman School of Medicine at the University of Pennsylvania. Led by graduate student Kiara Berríos under the supervision of Rahul Kohli, MD, Ph.D., an associate professor of Infectious Diseases at Penn, and Junwei Shi, Ph.D., an assistant professor of Cancer Biology, the investigations uncovered a novel gene editing technique that offers superior control compared to other existing techniques and has the potential to be used in-vivo. The technique has been patented, and the research is published in the latest issue of *Nature Chemical Biology*.

Base editors are one of the latest and most effective ways to achieve precise gene editing. In DNA targeted by base editors, C:G base pairs in DNA can be mutated to T:A or A:T base pairs can be turned to G:C. The base editors use CRISPR-Cas proteins to locate a specific DNA target and DNA deaminase enzymes to modify and mutate the target. Nevertheless, there was no way to trigger mutations at specific times or keep the editor in check to prevent undesired mutations.

The Penn researchers found that DNA deaminases can be divided into two inactive pieces, which can then be put back together using a small cell-permeable molecule called rapamycin. The new split-engineered base editors (seBEs) system can be introduced and lay dormant within a cell until the small molecule is added, at which point the base editing complex can be rapidly "turned on" to alter the genome.

"Our newly created split-engineered base editors really offer new potential for both research and therapeutics," Kohli said. "Since we can control the time mutations are made, there is a possibility to use these seBEs in vivo to model diseases by altering a gene, similar to how scientists control the timing of gene knockouts, and even potentially someday offer clinicians the ability to control editing of a patient's genes

for treatment purposes."

"Splitting DNA deaminase can also work outside of base editors," said Shi. "As a cancer researcher, I see this technique as having potential in controlling genetic changes that cause cancer development and growth. It could also be used to identify vulnerabilities in cancer cells."

Kohli's and Shi's labs plan to build on this research by applying controllable genome editing to cell-based screen research and by adding a layer of spatial control to accompany temporal control. A strength of the researchers' approach is that the controllable split enzyme system can also be partnered with other new developments in the rapidly expanding CRISPR/Cas field to newly gain regulatory control over these various base editing strategies.

More information: Kiara N. Berríos et al, Controllable genome editing with split-engineered base editors, *Nature Chemical Biology* (2021). DOI: 10.1038/s41589-021-00880-w

Provided by University of Pennsylvania