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GENOME EDITING

4 BIG QUESTIONS

Despite the popularity of genome-editing techniques, researchers are still grappling with the known unknowns of the technologies. Here are four of their most pressing questions.

BY WILL TAUXE

	QUESTION	WHY IT MATTERS	WHAT WE KNOW	NEXT STEPS
1	***How much can we reduce the off-target effects of genome editing?***	Unintentional edits can occur where a similar or identical target DNA sequence appears elsewhere in the genome. These off-target edits can frustrate the use of genome editing as a lab tool, and may cause side effects if the technique is used as a therapy.	The frequency of off-target effects varies among the three genome-editing technologies. TALENs produce the fewest off-target edits because they use a longer stretch of target DNA than ZFNs or CRISPR–Cas9 (see page S4).	The specificity of CRISPR–Cas9 can be increased by adjusting the guide RNA, which leads Cas9 to its target, and Cas9's structure. Bioinformatics can predict where off-target effects are most likely to occur and evaluate their consequences.
2	***Which diseases are suitable targets for genome editing?***	The more diseases that can be addressed through genome editing, the greater the technology's potential to relieve the disease burden.	Genome editing has had some success in combating HIV in people with the infection (see page S8), providing hope for those with other non-inherited diseases. Encouraging results have also been seen in models of certain monogenic diseases (see page S10).	To expand the range of diseases amenable to genome editing, researchers need better ways to deliver the technology to the right cells. CRISPR–Cas9 is too large to fit inside the vector adeno-associated virus. CRISPR systems in different bacteria may offer smaller alternatives.
3	***Can the phenotypic effects of genome editing be accurately predicted?***	For gene editing to be successful, researchers need to be able to determine the effect that making small changes to DNA, or to its packaging, has on the chemical components and physical properties of cells.	Several approved drugs (that do not edit the genome) treat conditions such as epilepsy and cancer by causing chemical modifications to DNA that do not change the order of its bases, or by altering DNA's packaging. But no one knows which of the alterations lead to these outcomes.	Researchers have modified genome-editing tools to make epigenetic changes. By investigating the changes caused by precise edits, they hope to gain a better understanding of the role of epigenetics in gene expression, and hence phenotype.
4	***Should we edit the human germ line?***	Making heritable edits has the potential to prevent diseases from being passed down the generations. But 'permanent' changes are risky if we do not have a full understanding of human gene expression. There is also the potential for misuse.	Edits to the genomes of non-viable human embryos have established proof-of-principle, although there is a high failure rate. If viable embryos are edited, implanting them and bringing them to term is just a short step away.	Scientists need to engage with governments and invite informed public discussion to draw up rigorous guidelines that govern research and clinical procedure. Systems must then be put in place to ensure that these guidelines are followed.

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