

MOLECULAR BIOLOGY, NUCLEIC ACIDS & MOLECULAR MEDICINE

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Using single-stranded DNA for homology directed repair catalyzed by CRISPR/Cas9 activity

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This workshop session will feature many speakers who are actively working in the field of gene editing with a specific focus on the use of single-stranded DNA templates in combination with gene editing tools such as CRISPR/Cas9 to repair genetic mutations via homology directed repair. Speakers will describe the design and rationale for using varying types of single-stranded DNA molecules, ranging from short single-stranded oligonucleotides to long strands generated by multiple amplifications in vitro. The overarching objective of all these projects is to reverse a point mutation or to restore gene function by homologous fragment insertion. Each speaker will also detail experimental case studies in which their method of choice was either successful or unsuccessful in generating the predicted genetic outcome. What were the consequences of these failed attempts? The use of single-stranded DNA functioning as patching or bridging the resected section of DNA created by the double-stranded break directed by CRISPR/Cas9 to reduce allelic heterogeneity will also be discussed. The format of the workshop will be interactive and a healthy dose of participation by attendees will be highly encouraged. We hope to achieve an understanding of how to study homology directed repair in mammalian cells in the most effective way, both transformed and primary. We hope to define what is real, reproducible and robust? And what is also non-reproducible artefactual or fictional? so that gene editing can grow in a truly healthy fashion, driven by science and not by publicity.

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