Gene editing technology continues to have a major impact on drug discovery and development research. Scientists utilize these tools to not only edit DNA for purposes of cell and gene therapies, but also to study disease states and their molecular targets and pathways, as well as to produce stable cell lines and reagent tools to further accelerate these activities. By utilizing gene editing technologies, companies can accelerate discovery and development activities in pharmacology, drug metabolism, and toxicology to support important advances in pharmaceutical research.

Gene editing tools such as Cas-CLOVER act as “molecular scissors” to introduce targeted double-strand breaks in genomic DNA for knockouts, knock-ins and base-pair edits. Such tools have a wide range of applications including cell-line engineering, animal model creation, genomic screening, and cell/gene therapies. Knock-out screening methods to identify novel molecules is fast becoming one of the most widely used gene editing applications in drug discovery (1).

Cas-CLOVER is unique compared to other gene editing tools as it combines the efficiency of CRISPR/Cas9 with the specificity of TALENs:

- **High efficiency**: Cas-CLOVER is recruited to the target-site by guide RNAs (gRNAs) in the same way Cas9 is recruited. Therefore, it has high on-target activity comparable to the CRISPR/Cas9 technology.

- **High specificity**: Cas-CLOVER utilizes two gRNAs which recruit two dCas9-Clo51 nucleases to initiate targeted disruption. The dCas9-Clo51 nuclease is fully dimeric, meaning activity or cutting only occurs where a pair of nucleases bind correctly to genomic DNA. No off-target cutting was detectable by deep sequencing.

- **Easy to Use**: The Cas-CLOVER technology can be integrated into any gene editing application or workflow with only a simple design change. With clear and independent issued intellectual property, Cas-CLOVER has simple and convenient licensing terms for R&D and commercial applications.

The specificity of Cas-CLOVER enables multiple rounds of targeting at one locus to increase indel frequency without introducing the risk of unwanted off-target mutations. Shown here are indel frequencies of 26% and 43% at the CHO GS locus for one and two rounds of targeting, respectively. These on-target frequencies in CHO cells are higher than reported for ZFN and comparable to those of CRISPR/Cas9.

Using Cas-CLOVER to discover novel therapeutics to treat disease is an exciting next step into the future of gene editing. Whether it’s utilizing the ability to create cellular and animal models to mimic and study diseases, or to screen for the next therapeutic breakthrough, gene editing in drug discovery and development is the future.

## Technology Applications

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Hera Biolabs is offering clear commercial freedom to operate and simple accessible licenses to commercial users. We are interested in special collaborations with academia and other research groups that can result in methods to make gene editing in drug discovery and development more precise and accessible to the scientific community.