Introduction

The Agilent QuikChange Lightning Multi Site-Directed Mutagenesis Kits lets you rapidly and efficiently create point mutations in plasmid DNA at up to five sites simultaneously. QuikChange Lightning Multi Kits are optimized for plasmids up to 8 Kb. The QuikChange Lightning technology employs a simple, 3-step protocol (Figure 1) that makes it easier and faster than other available methods or kits. Other methods are more cumbersome and prone to human error as they often require subcloning, single-stranded DNA rescue, or in vitro methylation.

The QuikChange Method

QuikChange kits have provided researchers with a fast, easy, and efficient non-PCR method to reliably perform site-directed mutagenesis since 1996. Other commercially-available kits utilize PCR-based techniques, which can propagate errors with each successive round of thermal cycling. The QuikChange method uses a linear amplification strategy with only the parental strand serving as the DNA template. Combining this with our highest fidelity polymerases leads to a significant reduction in unwanted second-site errors. The existence of such errors is likely to complicate and delay downstream screening and analysis.

Figure 1. QuikChange Lightning Multi Site-Directed Mutagenesis Kit method.
The Lightning Advantage

The ability to create mutations at multiple sites simultaneously provides users of QuikChange Lightning Multi Kits a significant savings in both time and effort compared to traditional methods. Conventional techniques to perform site-directed mutagenesis at five sites (along with accompanying miniprep and sequencing reactions) take 15 days to complete, but with QuikChange Lightning Multi the methods can be completed in only 3 days.

The QuikChange Lightning Multi Kits contain specially designed enzymes to shorten the time necessary to complete our signature 3-step protocol. Extension times for the thermal cycling process have been reduced by 75%, and digestion of the non-mutated parental template has been decreased to only five minutes. From the adjacent graph, when introducing multiple point mutations into three different template sizes, there is a significant reduction in thermal cycling and digestion times when comparing QuikChange Lightning Multi to QuikChange Multi (Figure 2). The QuikChange Lightning Multi Kits provide the same performance and ease-of-use as previous versions but with the added benefit of saving valuable research time.

Featured Application: Engineering Mutant Clone Collections

The QuikChange Lightning Multi Kits are well-suited for constructing a diverse collection of engineered mutant clones. Such a collection can be created and then screened for mutants with improved activity using the appropriate functional assay. For example, the system may be used to create mutant collections containing all possible amino acids at one site as well as combinations of different amino acids at multiple sites.

Ordering information

Our QuikChange kits provide all of the necessary components, including enzymes and competent cells, to successfully and rapidly perform site-directed mutagenesis.*

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*The Commercial QuikChange Lightning Multi Site-Directed Mutagenesis Kits include a limited use license which is required for purchases by commercial entities.

Related Products

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