

*TaKaRa Ex Taq*<sup>®</sup> DNA Polymerase (Cat.# RR001A)

*TaKaRa Ex Taq*<sup>®</sup> DNA Polymerase Hot-Start Version (Cat.# RR006A)

*TaKaRa LA Taq*<sup>®</sup> DNA Polymerase (Mg<sup>2+</sup> free buffer) (Cat.# RR002A)

*TaKaRa LA Taq*<sup>®</sup> DNA Polymerase with GC Buffer (Cat.# RR02AG)

## Frequently Asked Questions: *TaKaRa Ex Taq*<sup>®</sup> and *TaKaRa LA Taq*<sup>®</sup> DNA Polymerase Enzymes

Thousands of scientists worldwide have relied on *TaKaRa Ex Taq*<sup>®</sup> and *TaKaRa LA Taq*<sup>®</sup> DNA polymerases for robust amplification of their targets of interest. *TaKaRa Ex Taq* polymerase combines the performance of *TaKaRa Taq* with 3' to 5' proofreading ability, resulting in an enzyme system optimized for high yield and sensitivity. *TaKaRa LA Taq* polymerase also possesses 3' to 5' proofreading capability and is designed to allow amplification of very long targets.

Answers to frequently asked questions about *TaKaRa Ex Taq* and *TaKaRa LA Taq* DNA Polymerases are presented here. For additional information, refer to the technical literature and web pages for the products listed at the top of this FAQ.

### Comparing *TaKaRa Ex Taq* and *TaKaRa LA Taq* DNA Polymerases

#### Q1: What is the difference between *TaKaRa Ex Taq* and *TaKaRa LA Taq* polymerases?

A1: *TaKaRa Ex Taq* DNA Polymerase is optimized for generating high yield of product with high sensitivity and efficiency. Products up to 20 kb in length can be amplified from genomic DNA with this enzyme.

*TaKaRa LA Taq* DNA Polymerase is optimized for amplification of long targets (long range PCR). Products of up to 40 kb in length can be amplified from  $\lambda$  phage DNA and products up to 30 kb in length can be amplified from genomic DNA templates with this enzyme.

#### Q2: What is the fidelity of *TaKaRa Ex Taq* DNA Polymerase and *TaKaRa LA Taq* DNA Polymerase?

A2: Both enzymes have proofreading capability (3' to 5' exonuclease activity), and therefore have higher accuracy than standard *Taq* DNA polymerase. Using the Kunkel method, *TaKaRa Ex Taq* polymerase showed approximately 3-fold higher fidelity and *TaKaRa LA Taq* showed approximately 5-fold higher fidelity than standard *Taq*. However, fidelity varies significantly depending on the reaction conditions and target DNA amplified by PCR. If optimization of amplification efficiency fidelity is required, we recommend increasing the amount of template used (the upper limit of template concentration will result in smeared bands) and reducing the number of cycles (to 22-25).

#### Q3: Can I use a primer that contains inosine (dITP) with these enzymes?

A3: For enzymes with 3' to 5' exonuclease activity (e.g., *TaKaRa Ex Taq*, *TaKaRa LA Taq*, and *TaKaRa Z-Taq*<sup>™</sup> DNA Polymerase), using a primer that contains inosine significantly decreases the reaction efficiency. If using an inosine-containing primer, we recommend using *TaKaRa Taq*<sup>™</sup> DNA Polymerase (Cat. # R001A) for PCR amplification.

### Using *TaKaRa LA Taq* DNA Polymerase with GC Buffer

#### Q4: Two different buffers are provided with *TaKaRa LA Taq* DNA Polymerase with GC Buffer (Cat. # RR02AG). How do you select the proper buffer?

A4: GC Buffer II has a stronger DNA denaturation effect than GC Buffer I. Therefore, we recommend trying GC Buffer I first. If amplification products are not obtained with GC Buffer I, try GC Buffer II.

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**Q5: Is it possible to obtain long PCR products using *TaKaRa LA Taq* DNA Polymerase with GC Buffer?**

A5: Long PCR is possible in some cases. For targets with 50% GC composition, using GC Buffer I allowed amplification of products up to 35 kb long from  $\lambda$  DNA, and products up to 17.5 kb long from human genomic DNA template. For templates with high GC content (~70% GC), amplification of at least 2 kb has been confirmed.

**Q6: Are there any general recommendations for primer design when amplifying regions with high GC content using *TaKaRa LA Taq* DNA Polymerase with GC Buffer?**

A6: Primers with high T<sub>m</sub> values are recommended. Also, consider using primers that are 25 - 30 nucleotides in length. The 3' terminal sequence of the primer is important; therefore, avoid high GC content at the 3' ends of both upstream and downstream primers. Finally, avoid complementarity within and between primers to reduce the formation of primer-dimers.

**Q7: Can the GC Buffers included with *TaKaRa LA Taq* DNA Polymerase with GC Buffer be used for amplification of targets that are not GC rich?**

A7: GC Buffer I has been used for successful amplification of templates with approximately 50% GC content (i.e., products up to 17.5 kb long from human genomic DNA and up to 35 kb long from  $\lambda$  DNA). However, for templates with low GC content, using GC Buffer (particularly GC Buffer II) may decrease the amplification efficiency. For amplification of templates with normal levels of GC content, we recommend using LA PCR Buffer II, which is included with *TaKaRa LA Taq* DNA Polymerase (Cat. # RR002A).