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TUFF TAQ™ HOT START DNA POLYMERASE
PROTOCOL AND TECHNICAL DATA SHEET

TUFF TAQ™ HOT START DNA POLYMERASE

High Specificity, Yield & Sensitivity Even After 3 Months At Room Temperature

KEY FEATURES:

99% active after 1 month at room temperature

>80% active after 3 months at room temperature

Ships at ambient temperature - No dry ice

Freeze/thaw up to 50 times

TUFF Taq™ Hot Start DNA Polymerase is engineered with a proprietary peptide tag that confers extreme stability at room temperature. Supplied in an inactive state with no polymerase activity at ambient temperatures due to a novel patent pending method. This prevents non specific amplification resulting from extension of nonspecifically annealed primers and primer–dimers formed at low temperatures during PCR setup and the initial PCR cycle. Provides high specificity, yield and sensitivity and is ideal for use in everyday PCR applications. Has 5' to 3' polymerase & exonuclease activity but no 3' to 5' exonuclease activity.

KIT CONTENTS

- TUFF Taq™ Hot Start DNA Polymerase
- 10X PCR buffer A: Tris-HCL + (NH4)2SO4 (contains no Mg2+ or detergent)
- 10X PCR buffer B: Tris-HCL + (NH4)2SO4 + detergent (contains no Mg2+)
- 10x Solution E (Use with difficult templates eg GC rich or when non specific amplifications occur). Determine optimal working concentration empirically)
- 25mM MgCl2

APPLICATIONS

- PCR assays requiring high specificity, yield or sensitivity
- Multiplex PCR
- Complex genomic or cDNA templates
- Very low-copy targets e.g. single-cell PCR

SHIPPING & STORAGE CONDITIONS

Shipping and temporary storage for up to 1 month at room temperature has no significant detrimental performance effects

Routine storage: -20ºC

Safety warnings: Products and components should be handled only by persons trained in laboratory techniques. Wear suitable protective clothing such as laboratory overalls, gloves and safety glasses. Avoid contact with eyes and skin and wash immediately with water should contact occur.

Usage restrictions: For Research Use Only. Not for any animal or human therapeutic or diagnostic use. Some potential applications may require a license which is not provided by purchase of this product and users should obtain the license if required. Rebel Bioscience products may not be re-sold, transferred, modified or used to manufacture commercial products without written approval from Rebel BioScience, LLC.

RECOMMENDED PCR REACTION MIX FOR 100 µl

Component	Volume	Final Conc
TUFF Taq™ Hot Start DNA Polymerase 5 U /µl	4µl	0.02 – 0.05 U /µl
10X PCR Buffer (A or B)	10 µl	1X
25mM MgCl2	6-10 µl	1.5-2.5 mM
20mM dNTP mix	1 µl	200 µM
Forward Primer (10 pmol / µl)	1-3 µl	0.1-0.3 µM
Reverse Primer (10 pmol / µl)	1-3 µl	0.1-0.3 µM
DNA Template	5-20 µl	5-100 ng / µl
10X Solution E (if required, not for standard PCR)	0, 10, 20 or 30 µl	1X, 2X or 3X
PCR Grade H2O	Up to 100 µl	
Total	**100 µl**	

RECOMMENDED PCR CYCLING CONDITIONS

Cycle Step	Temperature	Time	# Cycles
Initial denaturation	**95ºC**	**12-15 min**	**1**
Denaturation	95ºC	30-60 sec	26-35
Annealing	50-68ºC	30-60 sec	
Elongation	72ºC	1-4 min	
Final Elongation	72ºC	5-10 min	1