Product Information

Cheetah™ FLEX HotStart Taq

Unit Size (Enzyme Only):

29079-1ML: 1 mL, 5 kU
29079-10ML: 10 mL, 50 kU
29079-100ML: 100 mL, 500 kU

Kit Contents: Cheetah™ FLEX HotStart Taq (500 U Taq with Buffer Pack)

Component	Size
29079-100UL: Cheetah™ FLEX HotStart Taq, 5 U/uL	100 uL (500 U)
29050B: Cheetah™ Taq Dilution Buffer, 10X (without Mg^{2+})	1.5 mL
29050C: 25 mM $MgCl_2$	1.5 mL

Storage and Handling

Store at -20°C. Product is stable for at least 12 months from date of receipt when stored as recommended.

Product Description

Cheetah™ FLEX HotStart Taq is Biotium's proprietary chemically modified Taq polymerase designed for reducing non-specific DNA amplification due to primer-dimer formation in PCR. Cheetah™ Taq also has a better shelf life than AmpliTaq Gold® due to its unique chemical modification, which is less likely to form intramolecular cross-links.

The activation time for Cheetah™ Taq is only about 2 minutes at 95°C, which is 5- to 10-times faster than that for AmpliTaq Gold® or HotStarTaq®, making it especially advantageous for fast-cycling qPCR. Furthermore, unlike AmpliTaq Gold®, activation of Cheetah™ Taq is relatively insensitive to pH, permitting the use of reaction buffers between pH 6 and pH 10. Unlike antibody-based hot-start Taq polymerases, chemically modified Cheetah™ Taq is free of animal DNA, and its activity is completely suppressed prior to heat activation. After heat activation, Cheetah™ Taq has the same functional properties and fidelity as native Taq polymerase.

In addition to its applications in qPCR, Cheetah™ FLEX HotStart Taq has been validated by third parties in qPCR-based proximity extension assay (PEA) for protein detection.

Cheetah™ FLEX HotStart Taq is available as a ready-to-use kit, which includes a buffer pack, and as a standalone enzyme for assay development and bulk purchase. Our Forget-Me-Not™ qPCR Master Mixes featuring Cheetah™ Taq (see Related Products) may also be of interest.

We also offer Cheetah™ PEA-PLEX HotStart Taq, which is a specialized formulation of Cheetah™ Taq validated by third parties for NGS-based high-throughput PEA.

PCR Reaction Setup

The following reaction setup is provided as a general starting point for optimization:

Reaction Component	Volume for 50 uL Reaction	Final Concentration	Range for Optimization
Molecular Biology Grade Water	To 50 uL total	N/A	N/A
10X Cheetah Dilution Buffer	5 uL	1X	N/A
25 mM $MgCl_2$	5 uL	2.5 mM	1.5-3.5 mM
10 mM dNTPs	1 uL	0.2 mM each dNTP	N/A
10 uM forward primer	1 uL	0.2 uM	0.1-1 uM
10 uM reverse primer	1 uL	0.2 uM	0.1-1 uM
Cheetah™ FLEX Taq	1 uL	0.1 U/uL	0.02-0.1 U/uL
Template DNA	x uL	<1 ug total	N/A

Cycling Protocols

Choice of cycling protocol depends on your instrument capability and on the nature of your amplicon. If your instrument does not support fast cycling, use the parameters recommended in your instrument manual.

1. Two-step fast cycling protocol

This protocol should be applicable to most amplifications where the primer melting temperature (Tm) is designed to be 60°C.

Cycling Step	Temperature	Hold Time	Number of Cycles
Enzyme Activation	95°C	2 minutes	1
Denaturation	95°C	1-15 seconds	25-35
Annealing/ Extension	60°C	1 minute per kb	

2. Three-step fast cycling protocol

Use this protocol when optimal primer annealing and extension temperatures are desired.

Cycling Step	Temperature	Hold Time	Number of Cycles
Enzyme Activation	95°C	2 minutes	1
Denaturation	95°C	1-15 seconds	25-35
Annealing	5°C below primer Tm	5-30 seconds	
Extension	72°C	1 minute per kb	
Final Extension	72°C	5 minutes	1

Analysis

Analyze 10 uL of the PCR reaction by agarose gel electrophoresis to confirm successful amplification of the desired amplicon. PCR reactions may be stored at -20°C for downstream applications.

Related Products

Cat. No.	Product
29138	Cheetah™ PEA-PLEX HotStart Taq (5 U/uL, Enzyme Only)
31000	EvaGreen® Dye 20X in Water
31077	EvaGreen® Plus Dye 20X in Water
31079	EvaRuby™ Dye 20X in Water
29052	ROX Passive Reference Dye
29087	VeriFluor™ Far-Red Passive Reference Dye
31041-31042	Forget-Me-Not™ EvaGreen® qPCR Master Mix (Separate ROX)
31043-31044	Forget-Me-Not™ Universal Probe Master Mix (Separate ROX)
31078	N-Flux™ 5X Digital PCR Master Mix
40054	dNTP Mix, 10 mM each
41024	Water, Ultrapure Molecular Biology Grade
29051	EvaEZ™ Fluorometric DNA Polymerase Activity Assay
41041-41042	Precast GelRed® Agarose Gels, 1% Agarose/TAE
41043	EMBER™ Ultra DNA Gel Kit
41044	EMBER™ Ultra RNA Gel Kit
41011	GelRed® Prestain Plus 6X DNA Loading Dye
41003	GelRed® 10,000X in Water
41005	GelGreen® 10,000X in Water
41028	Agarose LE, Ultra-Pure Molecular Biology Grade
41029	GelRed® Agarose LE
41030	GelGreen® Agarose LE
22031	1X TAE (1L) Buffer Powder Packets
22032	1X TBE (1L) Buffer Powder Packets
41039	Go-Go™ Fast DNA Gel Running Buffer, 50X
31028	AccuClear® Ultra High Sensitivity dsDNA Quantitation Kit
31066	AccuGreen™ High Sensitivity dsDNA Quantitation Kit for Qubit®
31080	1 kb DNA Ladder in TE buffer
31081	100 bp DNA Ladder in TE buffer
31084	1 kb DNA Ladder, Ready-to-Load
31085	100 bp DNA Ladder, Ready-to-Load
E90005	Gel-Bright™ Laser Diode Gel Illuminator

Please visit our website at www.biotium.com for information on our life science research products, including environmentally friendly EvaGreen® qPCR master mixes, Cheetah™ PEA-PLEX HotStart Taq, and Forget-Me-Not™ qPCR Master Mixes featuring Cheetah™ Taq.