

# **Product Information**

## Cheetah™ PEA-PLEX HotStart Taq

## Unit Size:

29138-100UL: 100 uL, 500 U 29138-1ML: 1 mL, 5 kU

## 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<sup>™</sup> PEA-PLEX HotStart Taq, 5 U/uL is a unique formulation of Biotium's chemically modified Cheetah<sup>™</sup> Taq polymerase validated by third parties specifically for NGS-based high-throughput proximity extension assays (PEA) for protein detection. Cheetah<sup>™</sup> PEA-PLEX is suitable for fast cycling qPCR, as well.

The proprietary chemical modification of Cheetah<sup>™</sup> Taq renders it inactive at room temperature for improved specificity and reduced primer-dimer formation in PCR. Cheetah<sup>™</sup> Taq also has a longer 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<sup>™</sup> Taq is only 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<sup>™</sup> Taq is relatively insensitive to pH, permitting use of reaction buffers between pH 6 and pH 10. Unlike antibody-based hot-start Taq polymerases, chemically modified Cheetah<sup>™</sup> Taq is free of animal DNA, and its activity is completely suppressed prior to heat activation. After heat activation, Cheetah<sup>™</sup> Taq has the same functional properties and fidelity as native Taq polymerase.

We also offer Cheetah<sup>™</sup> FLEX HotStart Taq for fast cycling qPCR as a standalone polymerase and in our ready-to-use Forget-Me-Not<sup>™</sup> qPCR Master Mixes (see Related Products). Cheetah<sup>™</sup> FLEX has also been validated by third parties for qPCR-based PEA assays. This product is available for licensing and bulk purchase.

## **Experimental Protocol**

Cheetah<sup>™</sup> PEA-PLEX HotStart Taq is supplied as standalone polymerase stock solution for custom assay development. We've provided an example PCR reaction protocol below as a starting point for optimization.

#### PCR reaction setup

| Reaction Component               | Volume for<br>50 uL Reaction | Final<br>Concentration |
|----------------------------------|------------------------------|------------------------|
| Molecular Biology Grade<br>Water | To 50 uL total               | N/A                    |
| 1 M Tris HCl pH 8.5              | 1.25 uL                      | 25 mM                  |
| 1 M KCI                          | 2.5 uL                       | 50 mM                  |
| 25 mM MgCl2                      | 5 uL                         | 2.5 mM                 |
| 10 mM dNTPs                      | 1 uL                         | 0.2 mM each<br>dNTP    |
| 10 uM forward primer             | 1 uL                         | 0.2 uM                 |
| 10 uM reverse primer             | 1 uL                         | 0.2 uM                 |
| Cheetah™ PEA-PLEX Taq            | 1 uL                         | 0.1 U/uL               |
| Template DNA                     | x uL                         | <1 ug total            |

## Three-step fast cycling protocol

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

| Related         | FIUUUCIS  |  |  |
|-----------------|---|--|--|
| Cat. No.        | Product   |  |  |
| 29079           | Cheetah™ FLEX HotStart Taq<br>(5 U/uL, Enzyme Only)                       |  |  |
| 29079-<br>KIT   | Cheetah™ FLEX HotStart Taq (500 U Taq with<br>Buffer Pack)                |  |  |
| 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-<br>31042 | Forget-Me-Not™ EvaGreen® qPCR Master Mix<br>(Separate ROX)                |  |  |
| 31043-<br>31044 | Forget-Me-Not™ Universal Probe Master Mix<br>(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<br>Activity Assay                      |  |  |
| 41041-<br>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<br>Quantitation Kit               |  |  |
| 31066           | AccuGreen <sup>™</sup> 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 <u>www.biotium.com</u> for information on our life science research products, including our environmentally friendly EvaGreen® qPCR master mixes, Cheetah<sup>™</sup> FLEX HotStart Taq, and Forget-Me-Not<sup>™</sup> qPCR Master Mixes featuring Cheetah<sup>™</sup> Taq.

AmpliTaq Gold is a registered trademark of Thermo Fisher Scientific; HotStarTaq is a registered trademark of Qiagen.

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