

Product Information	
<b>PerfeCTa® Multiplex qPCR SuperMix</b>	
Part Number	95063-01K
Number of Reactions	1000 reactions
Reaction Size	50 µL
Storage Temperature	-25°C to -15°C
Lot Number	66202163
Reference Number	071621, 060921, 100821
Expiration Date	06/30/2024

**Product Description:**

PerfeCTa Multiplex qPCR SuperMix is a 2X concentrated, ready-to-use reaction cocktail that contains all the necessary components except: primers, probe(s), and DNA template for highly-multiplexed, real-time quantitative PCR. This reagent formulation pushes the boundary of multiplex qPCR by enabling unbiased amplification of up to 5 targets in a single amplification reaction. Suppression of low abundance targets by high abundance reference targets during co-amplification is a common problem in multiplex PCR in which individual assay sensitivity can be significantly compromised. PerfeCTa Multiplex qPCR SuperMix, Low ROX delivers assay performance with exceptionally broad, linear detection and limit-of-detection (LOD) sensitivity to multiplexed qPCR that is comparable single-plex assay performance without the need for rigorous titration of individual primer assays. A key component of this SuperMix is ultra-pure AccuStart™ hot start Taq DNA polymerase that is completely arrested prior to the initial PCR denaturation step. Upon heat activation at 95°C, the antibodies are

rapidly and irreversibly denatured, releasing a fully active high-yielding Taq DNA polymerase mutant. This enables specific and efficient primer extension with the convenience of ambient room-temperature reaction assembly.

**Component Part Numbers:**

- 84164 Multiplex qPCR SuperMix, 25.0 mL
- 84165 ROX Reference Dye 50X, 1.25 mL
- 84166 Low ROX Reference Dye 50X, 1.25 mL

Product Specifications				
95063				
Assay	Multiplex qPCR Functional Assay	High Biased, Multiplex qPCR	DNase	RNase
Result	Pass	Pass	Pass	Pass

**Quality Control Analysis and Specifications:**

**Nuclease Assay:**

**DNase:** DNase activity must be below the detectable limits of 100 pg DNase I equivalent as assayed using a fluorogenic substrate following a 1 hour incubation at 37°C with each kit component at 1X concentration.

**RNase:** RNase activity must be below the detectable limits of 1 pg RNase A equivalent as assayed using a fluorogenic substrate following a 1 hour incubation at 37°C with each kit component at 1X concentration.

**Multiplex qPCR:** 4-color multiplex qPCR is performed in triplicate reactions on 10-fold serial dilutions of an equal copy mixture of four plasmid DNAs (10 copies to 1 x 10<sup>7</sup> copies). Cq standard curve analysis for each targeted sequence must have a coefficient of determination (r<sup>2</sup>) ≥0.990 with a slope between -3.25 and -3.65. Control reactions lacking template DNA (NTC) must remain below fluorescence threshold through 45 PCR cycles.

**High Biased, Multiplex qPCR:** 4-color multiplex qPCR of a 10-fold serial dilution of a FAM probe specific plasmid DNA (10 copies to 1 x 10<sup>7</sup> copies) in a fixed background of 3 different plasmid targets at 1x10<sup>8</sup> copies (each) must have an LOD of 10 copies. Cq standard curve analysis must have a coefficient of determination (r<sup>2</sup>) ≥0.990 with a slope between -3.25 and -3.65. No template controls (NTC) must be below threshold on at least 2 of the 4 copies. Each high copy target gene, ACTBD, IL1B, and TUBA is detected.

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