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

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 10⁷ copies). Cq standard curve analysis for each targeted sequence must have a coefficient of determination (r²) ≥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 10⁷ copies) in a fixed background of 3 different plasmid targets at 1x10⁸ copies (each) must have an LOD of 10 copies. Cq standard curve analysis must have a coefficient of determination (r²) ≥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.

Limitations of Use
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This product was developed, manufactured, and sold for in vitro use only. The product is not suitable for administration to humans or animals. SDS sheets relevant to this product are available upon request.

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