



## Certificate of Analysis

Verified by:  
Jyothi Vishumangalam

*J. Vishumangalam*

Quality Release date:  
Jan 22, 2019

Chai Biotechnologies Inc.  
Santa Clara, CA 95050  
Phone: +1 (650) 779-5577

---

Product Name: **Sahara Multiplex qPCR Master Mix**  
Catalog Num: R02210  
Lot Num: 4888105  
Analysis Date: Jan 22, 2019  
Expiration Date: Jan 22, 2021  
Storage: Store at -20 °C

### Master Mix Tests

### Results

#### Hot Start Functional Test

Hot Start function of Taq polymerase was tested using bacteriophage lambda genomic DNA template and a gene fragment containing the mouse GAPDH gene. Cycling conditions of 2 min @ 95 °C, 40x (15 s @ 95 °C, 60 s @ 60 °C) were used. The products were resolved by agarose gel electrophoresis on a 4% gel. Decreased primer dimer formation and absence of non-specific amplification using Sahara Multiplex qPCR Master Mix compared to variant not containing hot start function was monitored.

PASS

#### Fourplex Functional Test

A fourplex assay with TaqMan probes was performed using a five-point standard curve with ten-fold serial dilutions of *Enterococcus* genomic DNA, bacteriophage lambda genomic DNA, and independent gene fragments containing a peanut gene and mouse Actin gene target. The TaqMan probes were labeled with FAM, HEX, Texas Red, or Quasar 705. The targets were amplified using cycling conditions of 2 min @ 95 °C, 40x (15 s @ 95 °C, 60 s @ 60 °C). The efficiency for each target is specified to be between 90 – 110% and  $R^2 \geq 0.99$ .

PASS

<b>ROX Reference Dye Tests</b>	<b>Results</b>
<b>ROX Emission Spectrum Test</b>	
<p>Fluorescence emission spectrum of ROX reference dye was measured at 1 nm intervals, at a scan speed of 6000 nm/min using 580 nm excitation wavelength. The peak emission wavelength is specified to be <math>600 \pm 5</math> nm.</p>	<p>PASS</p>
<b>ROX Performance Test</b>	
<p>ROX reference dye was incorporated into a duplex assay set up using a five-point standard curve with ten-fold serial dilutions of the template bacteriophage lambda genomic DNA and a gene fragment containing Arah2 target (from peanut) and amplified using cycling conditions of 2 min @ 95 °C, 40x (15 s @ 95 °C, 60 s @ 60 °C). The efficiency is specified to be between 90 – 110% and <math>R^2 \geq 0.99</math>.</p>	<p>PASS</p>