

**Plasmodium falciparum, Strain 3D7 KAHRP**

**Catalog No. MRA-554**

**Product Description:**

*Plasmodium falciparum* (*P. falciparum*), strain 3D7 KAHRP is a genetically modified version of strain 3D7 in which the gene for Knob-Associated Histidine Rich Protein (KAHRP) has been disrupted. MRA-554 lot 70011948 was produced by cultivation of the BEI Resources seed lot 3256722 in fresh human erythrocytes suspended in RPMI 1640 medium, adjusted to contain 10% (v/v) heat-inactivated human serum (pooled Type A), 25 mM HEPES, 2 mM L-glutamine, 4 g/L D-glucose, 0.005 µg/mL hypoxanthine and 2.5 µg/mL gentamicin. The culture was incubated at 37°C in sealed flasks outgassed with blood-gas atmosphere (90% N<sub>2</sub>, 5% CO<sub>2</sub>, 5% O<sub>2</sub>) and monitored for parasitemia every 1 to 2 days for 5 days. Every 1 to 2 days, uninfected, leukocyte filtered, Type O erythrocytes in complete culture medium were added dropwise to the culture as needed and monitored for hematocrit.

**Lot: 70011948**

**Manufacturing Date: 13FEB2018**

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TEST	SPECIFICATIONS	RESULTS
<b>Identification by Giemsa Stain Microscopy<sup>1</sup></b>	Blood-stage parasites present	Blood-stage parasites present
<b>Antimalarial Susceptibility Profile (<i>in vitro</i>)<sup>1</sup></b> Half-maximal Inhibitory Concentration (IC <sub>50</sub> ) by SYBR Green I <sup>®</sup> drug sensitivity assay <sup>2</sup>		
Chloroquine	Report results	7.0 ± 0.3 nM
Artemisinin	Report results	13.8 ± 1.0 nM
Quinine	Report results	33.9 ± 3.1 nM
Cycloguanil	Report results	328.1 ± 30.3 nM
Pyrimethamine	Report results	14840 ± 1025.9 nM
Sulfadoxine	Report results	483800 ± 33446.3 nM
<b>Genotypic Analysis<sup>1</sup></b> Sequencing of Merozoite Surface Protein 2 (MSP2) gene (~ 770 base pairs)	Consistent with <i>P. falciparum</i>	Consistent with <i>P. falciparum</i> (Figure 1)
<b>Level of Parasitemia by Giemsa Stain Microscopy</b>		
Pre-freeze (5 days post-infection) <sup>3</sup>		
Ring-stage parasitemia	Report results	5.93%
Total parasitemia	≥ 2%	10.27%
Post-freeze (4 days post-infection) <sup>1</sup>		
Ring-stage parasitemia	Report results	1.36%
Total parasitemia	≥ 1%	3.74%
<b>Viability (2 days post-infection)<sup>1</sup></b>	Growth in infected red blood cells	Growth in infected red blood cells
<b>Sterility (21-day incubation)<sup>1</sup></b>		
Harpo's HTYE broth, 37°C and 26°C, aerobic <sup>4</sup>	No growth	No growth
Trypticase soy broth, 37°C and 26°C, aerobic	No growth	No growth
Sabouraud broth, 37°C and 26°C, aerobic	No growth	No growth
DMEM with 10% FBS, 37°C, aerobic	No growth	No growth
Sheep blood agar, 37°C, aerobic	No growth	No growth
Sheep blood agar, 37°C, anaerobic	No growth	No growth
Thioglycollate broth, 37°C, anaerobic	No growth	No growth
<b>Mycoplasma Contamination<sup>1</sup></b> DNA detection by PCR	None detected	None detected

<sup>1</sup>Testing completed on vial, post-freeze material

<sup>2</sup>A SYBR Green I<sup>®</sup> anti-malarial drug sensitivity assay in 96-well plates was used to determine IC<sub>50</sub> values of an active (> 70% ring stage) parasite culture in the presence of each antimalarial drug [Hartwig, C. L., et al. "XI: I. SYBR Green I<sup>®</sup>-Based Parasite Growth Inhibition Assay for Measurement of Antimalarial Drug Susceptibility in *Plasmodium falciparum*." In *Methods in Malaria Research Sixth Edition*. (2013) Moll, K., et al. (Ed.), EVIMalaR, pp. 122-129. *Methods in Malaria Research Sixth Edition* is available on the [BEI Resources website](#).]

<sup>3</sup>Testing completed on bulk material prior to vialing and freezing

<sup>4</sup>Atlas, Ronald M. *Handbook of Microbiological Media*. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

**Figure 1: MRA-554 MSP2 Sequence**

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TTTAATATTA AAAATGAAAG TAAATATAGC AACACATTCA TAAACAATGC TTATAATATG AGTATAAGGA GAAGTATGGC AGAAAGTAAG
CCTTCTACTG GTGCTGGTGG TAGTGCTGGT GGTAGTGCTG GTGGTAGTGC TGGTGGTAGT GCTGGTGGTA GTGCTGGTGG TAGTGCTGGT
GGTAGTGCTG GTTCTGGTGA TGGTAATGGT GCAGATGCTG AGGGAAGTTC AAGTACTCCC GCTACTACCA CAACTACCAA AACTACCACA
ACTACCACAA CTAATAATGA TGCAGAAGCA TCTACCAGTA CCTCTTCAGA AAATCCAAAT CATAAAAATG CCGAAACAAA TCCAAAAGGT
AAAGGAGAAG TTCAAGAACC AAATCAAGCA AATAAAGAAA CTCAAAATAA CTCAAATGTT CAACAAGACT CTCAAACTAA ATCAAATGTT
CCACCCACTC AAGATGCAGA CACTAAAAGT CCTACTGCAC AACCTGAACA AGCTGAAAAT TCTGCTCCAA CAGCCGAACA AACTGAATCC
CCCGAATTAC AATCTGCACC AGAGAATAAA GGTACAGGAC AACATGGACA TATGCATGGT TCTAGAAATA ATCATCCACA AAATACTTCT
GATAGTCAA AAGAATGTAC CGATGGTAAC AAAGAAAAC GTGGAGCAGC AACATCCCTC TTAAATAACT CTAGTAATAT TGCTCAATA
AATAAATTG TTGTTTTAAT TTCAGCAACA CTTGTTTTAT CTTTTG
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27 OCT 2022

Technical Manager or designee, ATCC Federal Solutions

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