

Plasmodium falciparum, Strain D10
Catalog No. MRA-201
Product Description:

Plasmodium falciparum (*P. falciparum*), strain D10 was isolated in Papua New Guinea and is generally considered drug sensitive. MRA-201 was produced by cultivation of seed material 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% N₂, 5% CO₂, 5% O₂) and monitored for parasitemia for 24 days. Every 1 to 3 days, uninfected, leukocyte filtered, Type O erythrocytes in complete culture medium were added dropwise to the culture as needed and monitored for hematocrit.

Lot: 70037554
Manufacturing Date: 17AUG2020

TEST	SPECIFICATIONS	RESULTS
Identification by Giemsa Stain Microscopy¹	Blood-stage parasites present	Blood-stage parasites present
Antimalarial Susceptibility Profile (<i>in vitro</i>)¹ Half-maximal Inhibitory Concentration (IC ₅₀) by SYBR green I [®] drug sensitivity assay ²		
Chloroquine	Report results	14.1 ± 1.3 nM
Artemisinin	Report results	9.4 ± 0.4 nM
Quinine	Report results	34.9 ± 5.6 nM
Cycloguanil	Report results	6.9 ± 0.5 nM
Pyrimethamine	Report results	39.1 ± 4.5 nM
Sulfadoxine	Report results	342100 ± 23650 nM
Genotypic Analysis¹ Sequencing of Merozoite Surface Protein 2 (MSP2) gene (~ 790 base pairs)	Consistent with <i>P. falciparum</i>	Consistent with <i>P. falciparum</i> (Figure 1)
Functional Activity by PCR Amplification¹ MSP2 PCR amplicon analysis	~ 600-900 base pair amplicon	~ 800 base pair amplicon
Level of Parasitemia by Giemsa Stain Microscopy Pre-freeze (24 days post-infection) ³ Ring-stage parasitemia Total parasitemia Post-freeze (2 days post-infection) ¹ Ring-stage parasitemia Total parasitemia	Report results ≥ 2% Report results ≥ 1%	2.48% 3.73% 2.01% 2.55%
Viability (post-freeze; 2 days post-infection)¹	Growth in infected red blood cells	Growth in infected red blood cells
Sterility (21-day incubation)¹ Harpo's HTYE broth, 37°C and 26°C, aerobic ⁴ Trypticase soy broth, 37°C and 26°C, aerobic Sabouraud broth, 37°C and 26°C, aerobic DMEM with 10% FBS, 37°C, aerobic Sheep blood agar, 37°C, aerobic Sheep blood agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic	No growth No growth No growth No growth No growth No growth No growth	No growth No growth No growth No growth No growth No growth No growth
Mycoplasma Contamination¹ DNA detection by PCR	None detected	None detected

¹Testing completed on vial, post-freeze material

²A SYBR Green I[®] anti-malarial drug sensitivity assay in 96-well plates was used to determine IC₅₀ 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[®]-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. Available at: to <https://www.beiresources.org/Publications/MethodsInMalariaResearch.aspx>.]

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³Testing completed on bulk material prior to vialing and freezing

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

Figure 1: MRA-201 MSP2 Sequence

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GAAGGTAATT AAAACATTGT CTATTATAAA TTTCTTTATT TTTGTTACCT TTAATATTAA AAATGAAAGT AAAATATAGC
AACACATTCA TAAACAATGC TTATAATATG AGTATAAGGA GAAGTATGGC AAATGAAGGT TCTAATACTA ATAGTGTAGG
TGCAAATGCT CCAAATGCTG ATACTATTGC TAGTGGAAGT CAAAGGAGTA CAAATAGTGC AAGTACTAGT ACTACTAATA
ATGGAGAATC ACAAACACT ACTCCTACCG CTGCTGATAC TATTGCTAGT GGAAGTCAAA GGAGTACAAA TAGTGCAAGT
ACTAGTACTA CTAATAATGG AGAATCACAA ACTACTACTC CTACCGCTGC TGATACCCCT ACTGCTACAG AAAGTAATTC
ACCTTCACCA CCCATCACTA CTACAGAAAG TTCAAGTTCT GGCAATGCAC CAAATAAAAC AGACGGTAAA GGAGAAGAGA
GTGAAAAACA AAATGAATTA AATGAATCAA CTGAAGAAGG ACCCAAAGCT CCACAAGAAC CTCAAACGGC AGAAAATGAA
AATCCTGCTG CACCAGAGAA TAAAGGTACA GGACAACATG GACATATGCA TGGTTCTAGA AATAATCATC CACAAAATAC
TTCTGATAGT CAAAAAGAAT GTACCGATGG TAACAAAGAA AACTGTGGAG CAGCAACATC CCTCTTAAGT AACTCTAGTA
ATATTGCTTC AATAAATAAA TTTGTTGTTT TAATTTTCAGC AACACTTGTT TTATCTTTTG CCATA
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20 OCT 2020

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