

**Plasmodium falciparum, Strain NF54-cg6-hsp86-PpyRE13**

**Catalog No. MRA-1325**

**Product Description:**

MRA-1325 is a *Plasmodium falciparum* (*P. falciparum*) parasite reporter strain produced by integrating the *hsp86-PpyRE13* luciferase reporter cassette into the *cg6* gene locus of *P. falciparum* parasite line NF54<sup>attB</sup>. MRA-1325 was produced by cultivation of deposited material in fresh human erythrocytes suspended in RPMI 1640 medium supplemented with 10% (v/v) heat-inactivated human serum (pooled Type A), 25 mM HEPES, 2 mM L-glutamine, 2 g/L D-glucose, 27 µg/mL hypoxanthine and 5 µg/mL gentamicin. The culture was incubated at 37°C in sealed flasks outgassed with a blood-gas atmosphere (90% N<sub>2</sub>, 5% CO<sub>2</sub>, 5% O<sub>2</sub>) and monitored for parasitemia for 14 days. Every 1 to 4 days, uninfected, leukocyte-filtered, Type O erythrocytes in complete culture medium were added dropwise to the culture as needed and monitored for hematocrit.

**Lot: 70056404**

**Manufacturing Date: 03MAR2023**

BEI Resources is committed to ensuring digital accessibility for people with disabilities. This Certificate of Analysis contains complex tables and may not be fully accessible. Please let us know if you encounter accessibility barriers and a fully accessible document will be provided: E-mail: [Contact@BEIResources.org](mailto:Contact@BEIResources.org). We try to respond to feedback within 24 hours.

TEST	SPECIFICATIONS	RESULTS
<b>Identification by Giemsa Stain Microscopy<sup>1</sup></b>	Blood-stage parasites present	Blood-stage parasites present
<b>Genotypic Analysis<sup>2</sup></b> Sequencing of Merozoite Surface Protein 2 (MSP2) gene (~ 750 base pairs)	Consistent with <i>P. falciparum</i>	Consistent with <i>P. falciparum</i> (Figure 1)
<b>Functional Activity of Luciferase Gene<sup>1,3</sup></b>	Positive	Positive
<b>Level of Parasitemia by Giemsa Stain Microscopy</b> Pre-freeze (14 days post-infection) <sup>2</sup> Ring-stage parasitemia Total parasitemia Post-freeze (2 days post-infection) <sup>1</sup> Ring-stage parasitemia Total parasitemia	Report results ≥ 2%  Report results ≥ 1%	3.4% 5.4%  10.4% 10.6%
<b>Viability (1 day 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>3</sup> 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
<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>Testing completed on bulk material prior to vialing and freezing

<sup>3</sup>Luciferase activity was determined using the Luciferase Assay System (Promega E1500). Parasites were lysed and incubated with luciferase assay reagent. Luciferase activity was measured using a luminometer with a bioluminescence emission spectra of ~ 620 nm.

<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-1325 MSP2 Sequence**

```
AACATTGTCTATTATAAAATTTCTTTATTTTTGTTACCTTTAATATTAATAATGAAAGTAAATATAGCAACACATTCATAAAACAATGC
TTATAATATGAGTATAAGGAGAAGTATGGCAGAAAAGTAAGCCTTCTACTGGTGCTGGTGGTAGTGCTGGTGGTAGTGCTGGTGGTAG
TGCTGGTGGTAGTGCTGGTGGTAGTGCTGGTGGTAGTGCTGGTGGTAGTGCTGGTGGTAGTGCTGGTGGTAGTGCTGGTGGTAGTGCTGGTGGTAG
TCCCGCTACTACCACAACACTACCAAACTACCACAACACTACCACAACACTACTAATGATGCAGAAGCATCTACCAGTACCTCTTCAGAAAA
TCCAAATCATAAAAATGCCGAAACAAATCCAAAAGGTAAAGGAGAAGTTCAAGAACCAAATCAAGCAAATAAAGAAACTCAAATAA
CTCAAATGTTCAACAAGACTCTCAAACCTAAATCAAATGTTCCACCCACTCAAGATGCAGACACTAAAAGTCCTACTGCACAACCTGA
ACAAGCTGAAAATTCTGCTCCAACAGCCGAACAAACTGAATCCCCCGAATTACAATCTGCACCAGAGAATAAAGGTACAGGACAACA
TGGACATATGCATGGTTCTAGAAATAATCATCCACAAAATACTTCTGATAGTCAAAAAGAATGTACCGATGGTAACAAAGAAAACCTG
TGGAGCAGCAACATCCCTCTTATAACTCTAGTAATATTGCTTCAATAAATAAATTTGTTGTTTTAATTTTCAGCAACACTTGTTTTAT
CTTTT
```

/Sonia Bjorum Brower/  
Sonia Bjorum Brower

14 MAR 2024

Technical Manager or designee, ATCC Federal Solutions

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

ATCC® is a trademark of the American Type Culture Collection.  
 You are authorized to use this product for research use only. It is not intended for human use.

