Certificate of Analysis for MRA-711A

Monoclonal Antibody R217 Anti-*Plasmodium falciparum* Erythrocyte Binding Antigen-175 RII (produced *in vitro*)

Catalog No. MRA-711A

This reagent is the tangible property of the U.S. Government.

Product Description: Antibody Class: IgG1k

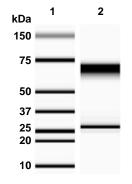
Monoclonal antibody R217 prepared against the erythrocyte binding antigen (EBA)-175 of *Plasmodium falciparum* (*P. falciparum*) 3D7 was purified from mouse hybridoma supernatant by protein G affinity chromatography.

Lot¹: 62011534 Manufacturing Date: 18NOV2013

TEST	SPECIFICATIONS	RESULTS
Antibody Class Determination	IgG1ĸ	lgG1κ
Experion Pro260 Analysis	Correct molecular weight (MW) for heavy and light chains Report results	Correct molecular weight (MW) for heavy and light chains (Figure 1) 97.1% pure
Concentration by Spectrophotometer at OD ₂₈₀	≥ 1.0 mg per mL	1.5 mg per mL
Functional Activity Western blot analysis with <i>P. falciparum</i> 3D7 (BEI Resources MRA-102) ² Immunofluorescence assay with <i>P. falciparum</i> 3D7 (BEI Resources MRA-102) ³ Indirect ELISA with recombinant EBA-175 RII (BEI Resources MRA-1162) ⁴	Reactive Reactive Reactive	Reactive (64 kDa non-glycosylated EBA-175 RII peptide; Figure 2) Reactive; fluorescence observed (Figure 3) Reactive (Figure 4)
Sterility	0.22 µm filter-sterilized	0.22 µm filter-sterilized

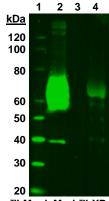
¹Produced from mouse hybridoma supernatant

Figure 1: Experion Pro260 Analysis



Lane 1: Pro260 Ladder Lane 2: MRA-711A

Figure 2: Western Blot Analysis



Lane 1: Invitrogen™ MagicMark™ XP Western Protein Standard

Lane 2: MRA-1162 (EBA-175 RII NG), non-reducing

Lane 3: MRA-102 P. falciparum 3D7 blood stage, whole cell extract 8 µL

Lane 4: MRA-1162 (EBA-175 RII NG), reducing

BEI Resources

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²Using a 1:1000 dilution of MRA-711A as a primary antibody and a 1:10000 dilution of goat anti-mouse IRDye conjugate as a secondary antibody. Monoclonal antibody R217 recognizes a conformational, disulfide-constrained epitope located within the F2 domain of region II (RII), under non-reducing conditions only.

³Specific labeling of *P. falciparum* smears with MRA-711A and FITC or DAPI was observed following paraformaldehyde fixation.

⁴Plates were coated with 100 μL of 1:1000 dilution of recombinant EBA-175 RII (BEI Resources MRA-1162). Secondary antibody was rabbit antimouse IgG-HRP conjugate.



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Figure 3: Immunofluorescence Assay on *P. falciparum* Blood Stage iRBC

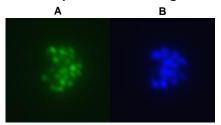
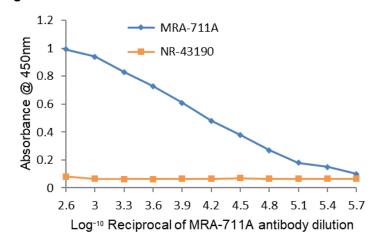


Figure 3A: MRA-711A (1:125) and fluorescein isothiocyanate (FITC; 1:500)

Figure 3B: 4' 6-diamiding-2-phenylindole (DAP

Figure 3B: 4',6-diamidino-2-phenylindole (DAPI)stained image

Figure 4: Indirect ELISA with Recombinant EBA-175 RII



/Heather Couch/

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Program Manager or designee, ATCC Federal Solutions

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