

Supplementary Material

Epitope-coated polymer particles elicit neutralizing antibodies against

Plasmodium falciparum sporozoites

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Supplementary Table 1. Sequences of proteins used in this study.

Fusion Protein	Sequence	Size
PhaC	MATGKGAAASTQEGKSQPFKVTGPFDPATWLEWSRQWQGTEG NGHAAASGIPGLDALAGVKIAPAQLGDIQQRYSALWQAMAEG KAEATGPLHRRRFAGDAWRTNLPYRFAAAFYLLNARALTELADAVE ADAKTRQIRFAISQWVDAMSPANFLATNPEAQRLLIESGGESLRA GVRNMMEDLTRGKISQTDESAFEVGRNVAVTEGAVVFENEYFQLL QYKPLTDKVVHARPLLMVPPCINKYYILDLPESLVRHVVEQGHTVF LWSWRNPDASMAGSTWDDYIEHAAIRAIEVARDISGQDKINVLGFCV GGTIVSTALAVLAARGEHPAASVTLLTLLDFADTGILDVDFVDEGHVQ LREATLGGGAGAPCALLRGLELANTFSFLRPNDLVWNYVVDNYLKG NTPVPFDLLFWNGDATNLPGPWYCWYLRHTYLQNELKVPKLTVC GVPVDLASIDVPTYIYGSREDHIVPWTAAAYASTALLANKLRFVLGAS GHIAGVINPPAKNKRSHWTNDALPESPQQWLAGAIEHHGSWWPD WTAWLAGQAGAKRAAPANYGNARYRAIEPAPGRYVKAKA	64.33 kDa
B cell epitope-PhaC	MNANPNANPNANPNANPNANPNANP-Linker-ATGKGAAASTQEG KSQPFKVTGPFDPATWLEWSRQWQGTEGNGHAAASGIPGLDAL AGVKIAPAQLGDIQQRYSALWQAMAEGKAEATGPLHRRRF GDAWRTNLPYRFAAAFYLLNARALTELADAVEADAKTRQIRFAISQ WVDAMSPANFLATNPEAQRLLIESGGESLRAGVRNMMEDLTRGKIS QTDESAFEVGRNVAVTEGAVVFENEYFQLLQYKPLTDKVVHARPLLM VPPCINKYYILDLPESLVRHVVEQGHTVFLVSWRNPDASMAGST WDDYIEHAAIRAIEVARDISGQDKINVLGFCVGGTIVSTALAVLAARG EHPAASVTLLTLLDFADTGILDVDFVDEGHVQLREATLGGGAGAPCA LLRGLELANTFSFLRPNDLVWNYVVDNYLKGNTVPFDLLFWNGDA TNLPGPWYCWYLRHTYLQNELKVPKLTVCGVPVDLASIDVPTYIY GSREDHIVPWTAAAYASTALLANKLRFVLGASGHIAGVINPPAKNKR SHWTNDALPESPQQWLAGAIEHHGSWWPDWTAWLAGQAGAKRAA PANYGNARYRAIEPAPGRYVKAKA	67.07 kDa
B/T cell epitope-PhaC	MDPNANPNVDPNANPNVNANPNANPNANPEYLNKIQNSLSTEWSP CSVTDPNANPNVDPNANPNVNANPNANPNANPEYLNKIQNSLSTE WSPCSVT-Linker-ATGKGAAASTQEGKSQPFKVTGPFDPATWLE WSRQWQGTEGNGHAAASGIPGLDALAGVKIAPAQLGDIQQRYSAL WQAMAEGKAEATGPLHRRRFAGDAWRTNLPYRFAAAFYLLN ARALTELADAVEADAKTRQIRFAISQWVDAMSPANFLATNPEAQR LLIESGGESLRAGVRNMMEDLTRGKISQTDESAFEVGRNVAVTEGA VVFENEYFQLLQYKPLTDKVVHARPLLMVPPCINKYYILDLPESLVR HVVEQGHTVFLVSWRNPDASMAGSTWDDYIEHAAIRAIEVARDIS GQDKINVLGFCVGGTIVSTALAVLAARGEHPAASVTLLTLLDFADT GILDVDFVDEGHVQLREATLGGGAGAPCALLRGLELANTFSFLRPND LVWNYVVDNYLKGNTVPFDL LFWNGDATNLPGPWYCWYLRHTYLQNELKVPKLTVCGVPVDLAS IDVPTYIYGSREDHIVPWTAAAYASTALLANKLRFVLGASGHIAGVINP PAKNKRSHWTNDALPESPQQWLAGAIEHHGSWWPDWTAWLAGQ AGAKRAAPANYGNARYRAIEPAPGRYVKAKA	74.92 kDa
B cell epitope peptide	MNANPNANPNANPNANPNANPNANP	2.53 kDa
B/T cell epitope peptide	MDPNANPNVDPNANPNVNANPNANPNANPEYLNKIQNSLSTEWSP CSVTDPNANPNVDPNANPNVNANPNANPNANPEYLNKIQNSLSTE WSPCSVT	10.25 kDa

Supplementary Table 2. Identification of proteins using tryptic peptide fingerprinting applying MALDI-TOF/MS.

Identified peptides are highlighted in green.

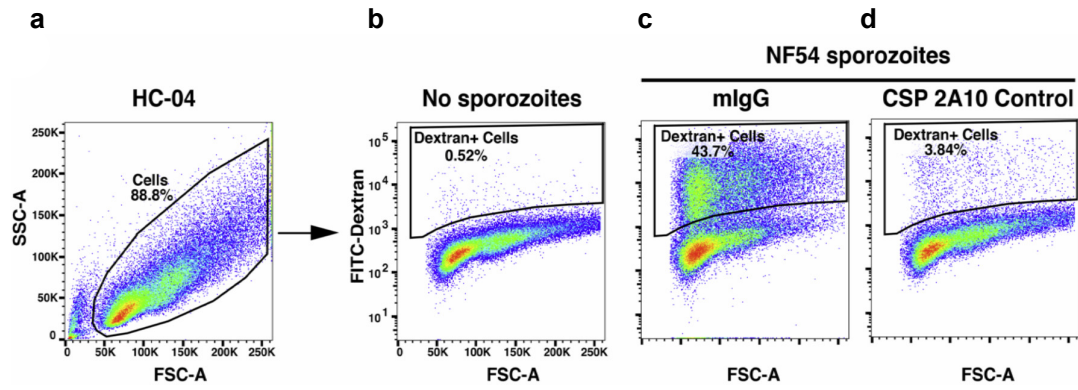
Sample Name	Identified Peptides
B cell epitope-PhaC fusion protein	<p>B cell epitope: MNANPNANPNANPGPGGNANPNANPNANP</p> <p>PhaC: ATGKGAAASTQEGKSQPFKVTGPFDPATWLEWSRQWQGTEGNHAAA SGIPGLDALAGVKAPAQLGDIQQRYMKDFSALWQAMAEGKAEATGPLHD RRFAGDAWRTNLPYRFAAAFYLLNARALTELADAVEADAKTRQRIRFAISQ WVDAMSPANFLATNPEAQRLLIESGGESLRAGVRNMMEDLTRGKISQTDE SAFEVGRNVAVTEGAVVFENEYFQLLQYKPLTDKVVHARPLLMVPPCINKYY ILDLPQESSLVRHVVEQGHTVFLVSWRNPDASMAGSTWDDYIEHAAIRAIE VARDISGQDKINVLGFCVGGTIVSTALAVLAARGEHPAASVTLTLLDFADT GILDVVFDEGHVQLREATLGGGAGAPCALLRGLELANTFSFLRPNDLVWN YVVDNYLKGNTVPFDLLFWNGDATNLPGPWYCWYLRHTYLQNELKVP KLTVCGVPVDLASIDVPTYIYGSREDHIVPWTAAASTALLANKLRFVLGAS GHIAGVINPPAKNKRSHWTNDALPESPQQWLAGAIEHHGSSWWPDWTAWL AGQAGAKRAAPANYGNARYRAIEPAPGRYVKAKA</p>
B/T cell epitope-PhaC fusion protein	<p>B/T cell epitope: MDPNANPNVDPNANPNVNANPNANPNANPEYLNKIQNSLSTEWSPCSVT GPGPGDPNANPNVDPNANPNVNANPNANPNANPEYLNKIQNSLSTEWSP CSVT</p> <p>PhaC: ATGKGAAASTQEGKSQPFKVTGPFDPATWLEWSRQWQGTEGNHAAA SGIPGLDALAGVKIAPAQLGDIQQRYMKDFSALWQAMAEGKAEATGPLHD RRFAGDAWRTNLPYRFAAAFYLLNARALTELADAVEADAKTRQRIRFAISQ WVDAMSPANFLATNPEAQRLLIESGGESLRAGVRNMMEDLTRGKISQTDE SAFEVGRNVAVTEGAVVFENEYFQLLQYKPLTDKVVHARPLLMVPPCINKYY ILDLPQESSLVRHVVEQGHTVFLVSWRNPDASMAGSTWDDYIEHAAIRAIE VARDISGQDKINVLGFCVGGTIVSTALAVLAARGEHPAASVTLTTLLDFAD TGILDVVFDEGHVQLREATLGGGAGAPCALLRGLELANTFSFLRPNDLVW NYVVDNYLKGNTVPFDLLFWNGDATNLPGPWYCWYYLRHTYLQNELKVP GKLTVCVGPVDLASIDVPTYIYGSREDHIVPWTAAASTALLANKLRFVLGA SGHIAGVINPPAKNKRSHWTNDALPESPQQWLAGAIEHHGSSWWPDWTAW LAGQAGAKRAAPANYGNARYRAIEPAPGRYVKAKA</p>

Supplementary Table 3. Strains used in this study.

Isolate	Use	Notable features	Reference
<i>E. coli</i> Top10	Plasmid propagation		Invitrogen, Carlsbad, CA, United States
<i>E. coli</i> Clearcoli	PHB bead production	Containing pMCS69 (Cm ^R). Induction via 1mM IPTG	1
<i>P. falciparum</i> NF54	Sporozoite staining and traversal assay		2
HC-04 hepatocytes	Cell traversal assay		3

Supplementary Table 4. Plasmids used in this study.

DNA	Use	Notable features	Reference
pET14b-phaC	Encodes PhaC for PHB production	phaC (64.33 kDa) under T7 promoter, Amp 100	4
pMCS69	Encodes PhaA and PhaB for PHB production	phaA, phaB under CAT promoter, Cm 50	4
pET14b B cell epitope-PhaC	Encodes PHB synthase fusion with B cell epitopes	B cell epitope-PhaC (64.33 kDa) under T7 promoter, Amp 100	This study
pET14b B/T cell epitope-PhaC	Encodes PHB synthase fusion with B/T cell epitopes	B/T cell epitope-PhaC (74.92 kDa) under T7 promoter, Amp 100	This study
pET14b PhaC-(GB1) ₃	Encodes PHB synthase fusion with protein G binding domain	PhaC-(GB1) ₃ under T7 promoter, Amp 100	5



Supplementary Figure 1. Gating strategy used in FACS analysis to generate traversal inhibition data shown in Figure 6. FACS dot plots measuring FITC-dextran uptake into human HC-04 hepatocytes following incubation with sporozoites (traversal). a, shows gating for HC-04 cells. b, shows no sporozoite control. c, shows addition of sporozoites in the presence of naïve mouse IgG (10 μ g/ml). d, shows addition of sporozoites in the presence of mouse anti-CSP 2A10 control antibodies (monoclonal anti-NANP3 antibodies). Shown is a single representative of three experiments. The same gating strategy was previously used to assess cell traversal by *P. falciparum* (see Figures 4 in Yang et al ^{6,7}).

References

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