



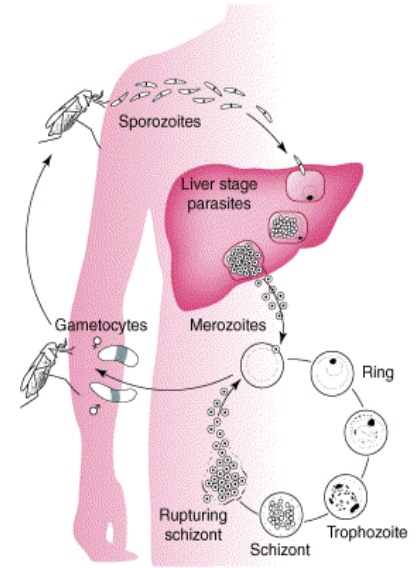
CBER science supports vaccine development and assays of vaccine safety and efficacy

Multiple Antigen Peptide Vaccines against *Plasmodium falciparum* Malaria

Babita Mahajan,¹ Jay A. Berzofsky,² Robert A. Boykins,¹ Victoria Majam,¹ Hong Zheng,¹ Rana Chattopadhyay,¹ Patricia de la Vega,³ J. Kathleen Moch,³ J. David Haynes,³ Igor M. Belyakov,^{2,5} Hira L. Nakhasi,¹ and Sanjai Kumar^{1*}

Division of Emerging and Transfusion Transmitted Diseases, Center for Biologics Evaluation and Research, Food and Drug Administration, Rockville, Maryland¹; Vaccine Branch, Center for Cancer Research, National Cancer Institute, National Institutes of Health, Bethesda, Maryland²; Walter Reed Army Institute of Research and Naval Medical Research Center, Silver Spring, Maryland³

Infection and Immunity
2010:70(11):4613-4624



Why We Need Malaria Vaccines and Blood Tests

- Malaria is endemic in more than 100 countries around the world. Each year, malaria infects 300-500 million people and results in more than 1 million deaths.
- Malaria threatens 28 million US travelers and thousands of military personnel each year.
- Approximately 1500 clinical cases of malaria occur in the US each year, generally among travelers and immigrants returning from countries where malaria transmission occurs, many from sub-Saharan Africa and Asia.
- Blood tests could identify potential donors who can safely provide blood despite having visited or lived in endemic countries, resulting in 100,000-150,000 fewer deferrals and an increased supply of blood and blood products.

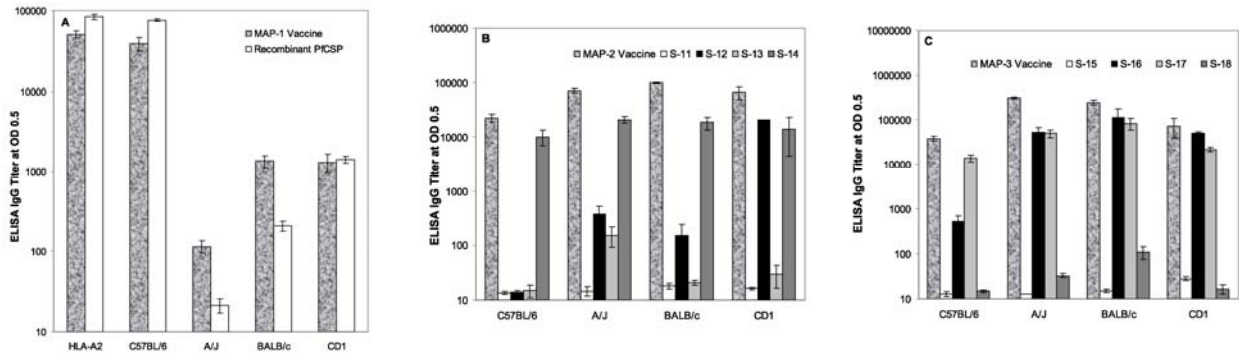
Multiple Antigen Peptide (MAP) Vaccines

Malaria parasites have a complex life that requires a vertebrate and a mosquito host. This complicates prevention, vaccination and treatment. Many malaria researchers believe that only a vaccine that targets multiple conserved antigens and multiple stages of the malarial parasite will induce effective immunity. A team of scientists led by researchers from CBER designed, synthesized, and tested in mice three multiple-antigen peptide (MAP) vaccines against multiple life cycle stages of the *P. falciparum* malaria.

- B-cell and T-cell epitopes from antigens of sporozoite, liver, and asexual blood stages
 - MAP-1: circumsporozoite (CSP) protein
 - MAP-2: circumsporozoite protein, liver stage antigen 1, merozoite surface protein-1₄₂ and merozoite surface protein-3
 - MAP-3: rhoptry associated protein-1 and 2, serine repeat antigen and merozoite surface protein-1₄₂

B-Cell (antibody) Responses in Various Strains of Mice

Immune responses in individual mouse strains to specific epitopes listed above bars.



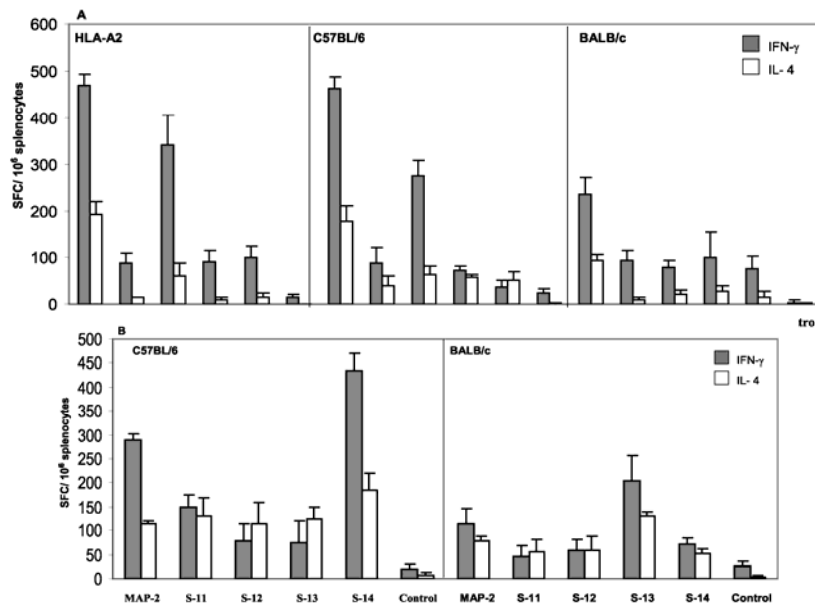
Mice were immunized by three subcutaneous injections of MAP vaccines; Enzyme-Linked Immunosorbent Assay (ELISA) antibody titers were determined for each immunization group.

A: Sera from MAP-1 immunized mice tested against MAP-1 or recombinant *Plasmodium falciparum* circumsporozoite protein (PfCSP).

B: Sera from MAP-2 immunized mice tested against MAP-2 or individual peptides.

C: Sera from MAP-3 immunized mice tested against MAP-3 or individual peptides.

T-Cell (IFN- γ & IL-4) Responses in Various Strains of Mice



Spleen cells pooled from 5 mice per group were cultured with MAP vaccines or individual peptides. Data presented as antigen-specific IFN- γ / IL-4 spot-forming cells (SFC) per million spleen cells; error bars are standard deviation of triplicate samples.

Selected Highlights of Findings

- All MAP vaccines were immunogenic & induced antibody and cellular responses.
- Antibodies induced by MAP-1 blocked invasion of liver cells by sporozoites.
- Antibodies induced by MAP-2 & MAP-3 reduced growth of blood stage parasites in erythrocyte cultures.

The data support further research and development of next -generation candidate MAP vaccines based on conserved protective epitopes from *Plasmodium* antigens.

Tests to Support Vaccine Development

A key part of supporting the development of malaria vaccines is the design and testing of quality control tests that determine the safety and efficacy of those vaccines. CBER is currently developing two electrochemiluminescence-based Western blot assays.

- Efficacy assay: Determine number of *P. falciparum* sporozoites in vaccine preparations.
 - Detects as few as one *P. falciparum* sporozoites
 - Quantifies number of parasites in unknown samples with 90% accuracy
- Safety assay : Determine amount of mosquito salivary gland protein in attenuated sporozoites produced by dissections of salivary glands of infected mosquitoes
 - Detects as little as 40 pg of mosquito salivary gland protein

These assays support the development of malaria vaccines that will protect the health of people globally, including the American travelers and military personnel returning to the US from endemic areas.