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The *Plasmodium falciparum* gp195 is a candidate malaria vaccine antigen. The C-terminal 42 kDa processing fragment of gp195 was expressed in baculovirus as a prototype vaccine. Analyses by ELISA and immunoblot showed that this recombinant polypeptide (BVp42) closely mimics the antigenicity of native gp195, and induced high antibody titers similar to levels induced by native gp195. These antibodies strongly inhibited parasite growth in vitro. Studies have shown that gp195 exists as several allelic forms and since BVp42 has both allelic and conserved sequences, the reactivity of anti-BVp42 antibodies with homologous vs heterologous gp195 was analyzed. Four parasite isolates, FUP, FVO, Hond-1, and FC27 were used and Southern blot analyses showed that FUP and FC27 belong to the MAD20, and FVO and Hond-1 belong to the K1 allele. Native gp195 was isolated from FUP and FVO parasites representing both alleles. In ELISAs, identical titers/binding curves were obtained with anti-BVp42 antibodies using either homologous or heterologous gp195 as antigens. Similar results were obtained by IFA. Moreover, anti-BVp42 antibodies inhibited the in vitro parasite growth of heterologous parasites/alleles (FVO & Hond-1) to similar degree as with homologous parasites (FUP & FC27). These data indicate that immunization with BVp42 induced high titers of biologically active antibodies that are extensively crossreactive with parasites carrying heterologous gp195 and may provide the basis for an effective vaccine against different parasite strains.

315 RELATIONSHIP BETWEEN H-2 HAPLOTYPE AND THE EFFECTIVENESS OF AN ADJUVANT IN INDUCING ANTIBODIES THAT INHIBIT THE GROWTH OF *PLASMODIUM FALCIPARUM*. Chang SP*, Hui GS, Nikaido C, Yokota B, and Hashimoto A. John A. Burns School of Medicine, Dept of Tropical Medicine and Medical Microbiology, Honolulu, HI.

In previous studies we have found that the induction of biologically-active antibodies to gp195, a major merozoite surface protein of P. falciparum, is dependent on the adjuvant used for immunization. While both Complete Freund's Adjuvant and Lipid A- 15-PH adjuvant induced gp195-specific antibodies in mice, only the LA-15-PH adjuvant formulation induced antibodies that affected parasite growth in this species. In the current study, the possible influence of H-2 linked immune response genes on the effectiveness of the LA-15-PH adjuvant in supporting the production of growth-inhibitory antibodies to gp195 was evaluated. Congenic mice of seven H-2 haplotypes were immunized with gp195 in LA-15-PH and their antibody responses were evaluated. While all seven strains produced gp195-specific antibodies, not all strains produced antibodies which inhibited parasite growth. These results suggest that the induction of inhibitory antibodies is dependent both on the adjuvant and on H-2 linked immune response genes. The degree of inhibition correlated with ELISA titers to purified, parasite gp195 and a C-terminal, baculovirus recombinant p42 polypeptide. There was no significant correlation of inhibition with antibody reactivity with an N-terminal, yeast recombinant polypeptide. Interestingly, the highest degree of correlation was between percent inhibition and ELISA titers against the heterologous FVO gp195, supporting the importance of antibodies against conserved epitopes in the growth inhibition assay.

316 T CELL REACTIVITY AGAINST MEROZOITE ANTIGENS OF *PLASMODIUM FALCIPARUM* AND HLA RESTRICTION. Dieye A*, Sarthou JL, Rogier C, Trape JF, Launois P, Holder AA, and Heidrich HG. Immunology Unit, Institut Pasteur de Dakar, B.P. 220 Senegal; Service d'Epidemiologie, Institut Pasteur de Dakar; ORSTOM, Dakar, Senegal; MRC, London, United Kingdom; and Max-Planck Institut fur Biochemie, Martinsried bei Munchen, Germany.

Cell proliferative response *in vitro* to merozoite antigens was analyzed from 99 individuals: 0-9 years (n=35); 10-19 years (n=27) and >20 years (n=37). Donors were selected in an holoendemic malaria area (Dielmo Village, Senegal, West Africa). Blood samples were collected at the end of the dry season,

when malaria transmission was low. Subjects had not acute malaria at the time of bleeding. No chemoprophylaxis was taken. Mononuclear cells from peripheral blood were cultured in the presence of 10 μg/ml of crude soluble merozoite antigens from *Plasmodium falciparum* during 6 days. Supernatants were harvested to IFN-γ determination by ELISA and the lymphoproliferation measured by ^{3}H-TdR incorporation. HLA-antigens were determined by microlymphocytotoxicity complement-dependant using Terasaki plates (One Lambda). Dead or viable cells were stained by AO/EB solution and the % estimated with a inverted fluorescence microscope. Cell proliferation was analyzed by the Stimulation Index (SI) and the Δ cpm.. Subjects were classified into two groups: High Responders (HR) and Low Responders (LR). HR were defined by a SI>4.25, Δ cpm>2263; LR had SI<4.25, Δ cpm<2263. The percentage of HR and LR was respectively 53.5 and 46.5%. T lymphocytes that were CD4-, CD8-expressing the TcR γδ was the proliferative subset found in HR group that had the Vγ9+ phenotype. The IFN-γ levels were significantly increased (p=0.001) in the HR group after 6 days of culture with merozoite antigens, 100 fold than LR. The HLA antigens: A26, HLA-B51, BW59, BW54, HLA-DR1 were present only in HR group. HLA-B51 and HLA-DR1 were found significantly associated ($p<0.05$; χ^2) with HR. HLA-BW53 and DRW13 were associated with LR but not significantly ($p>0.05$; χ^2). It would be interesting to have these evidences for conception of malaria vaccine.

317 *PLASMODIUM YOELII* CIRCUMSPOROZOITE PROTEIN SPECIFIC CD4+ T CELL CLONES ELIMINATE *P. YOELII* LIVER FORMS *IN VIVO* AND *IN VITRO*. Renia L*, Grillot D, Salone Marussig M, Corradin G, Miltgen F, Lambert PH, Del Giudice G, and Mazier D. INSERM U313, Hopital Pitie-Salpetriere, Paris, France; WHO-IRTC, University of Geneva, Geneva, Switzerland; Institut of Biochemistry, University of Lausanne, Epalinges sur Lausanne; and Museum d' Histoire Naturelle, Paris, France.

The efficacy of CD4+ T cell clones specific for a peptide of *Plasmodium yoelii* (amino acids 59-79) was evaluated *in vitro*. *In vitro* assays consisted of depositing BALB/c and C57BL/6 T cells cones on *P. yoelii* hepatic cultures. Most of the clones tested so far were found to significantly eliminate liver schizonts in a genetically restricted way. *In vivo* activity of CD4+ T cell clones were tested by transferring the cells in mice, 24 hours before challenging them with sporozoites. Protection was only observed in BALB/c mice for two clones. Previous studies have emphasized the role of CD8+ T cells in protection observed in mice immunized with irradiated sporozoites. Our results indicate that induction of CD4+ T cells could be important for pre-erythrocytic vaccine development.

318 INDUCTION OF I-A EXPRESSION IN PERITONEAL MACROPHAGES BY PARASITIZED ERYTHOCYTES OF LETHAL AND NON-LETHAL RODENT MALARIA *IN VITRO*. Shear HL*, Dunne J, and Remache Y. Division of Hematology, Montefiore Medical Center and Albert Einstein College of Medicine, Bronx, NY; and Department of Medicine and Molecular Parasitology, New York University School of Medicine, NY, NY.

The immune response to *Plasmodium chabaudi adami* is dependent on CD4+ T cells. Activation of these cells is via their interaction with peptides and Class II molecules expressed on antigen-presenting cells. In this study, we assayed the ability of parasitized erythrocytes to induce I-A expression on peritoneal macrophages. After incubation of macrophages from BALB/c mice with parasitized erythrocytes for 18 hrs, I-A expression was measured using the MK-D6 monoclonal antibody (anti I-Ad) followed by ^{125}I-labeled anti Ig Fab2 fragments. *P. chabaudi adami*-infected RBC induced greater I-A expression in peritoneal macrophages than did normal erythrocytes. We next compared I-A induction after incubation of macrophages with cloned parasites from a mild strain compared with a more virulent strain of *P. chabaudi*. The mild strain induced a higher level of I-A expression. In order to determine whether malarial peptides of *P. chabaudi* were expressed on the macrophage membranes, macrophages were incubated with moAbs against a *P. chabaudi adami*

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