154. Severe Acute Respiratory Syndrome Coronavirus-Like Particles (SARS-CoVLPs) Stimulate Human Monocyte-Derived Dendritic Cell Activity and Induce Antibody Response in Mouse Model

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Severe acute respiratory syndrome (SARS) is a life-threatening disease caused by a novel SARS associated coronavirus (SARS-CoV). For public health measures, it is very important to develop an effective vaccine to prevent possible recurrence of a SARS epidemic. Dendritic cells (DCs) have been recognized as potent antigen presenting cells and the key regulators of innate and adaptive immune responses. In this investigation, we have studied the interaction between SARS-Co virus like particles (SARS-CoVLPs) and DCs as an initiation step of the immune response. Co-culturing of SARS-CoVLPs with DC at different time points in primary culture induced phenotypic maturation of human monocyte-derived DCs. Through ELISA, we also observed increased levels on the expressions of pro-inflammatory cytokines (e.g., TNF-a and IL-6) in the cell culture of SARS-CoVLPs-pulsed DCs. Furthermore, SARS-CoVLPs-pusled DCs induced allogeneic CD4 and CD8 T cell proliferations and increased IFN-g secretion. We then further investigated the *in vivo* immune response on the use of SARS-CoVLPs as an immunogen. SARS-CoVLPs and SAR-CoVLP-based DNA vaccine were either injected subcutaneously or delivered into skin of test mice via gene gun bombardment. The VLP- based ELISA indicated that IgG antibody responses to SARS-CoVLPs and SARS based DNA vaccine increased significantly after priming, boosting and a second boosting. We also observed an increased IFN-g production from splenocytes of vaccinated mice, as measured by ELISPOT assay. Taken these data together, our results indicated that the SARS-CoVLPs we developed can further induce a Th1-biased immune response and may represent a promising approach for SARS vaccine development.

155. Partial Protection Against H5N1 Influenza in Mice with a Single Dose of a Chimpanzee Adenovirus Vector Expressing Nucleoprotein

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The use of adenoviral vectors based on non-human serotypes such as the chimpanzee adenovirus AdC7 may allow for their utilization in populations harboring neutralizing antibodies to common human serotypes. Because adenoviral vectors can be used to generate potent T cell responses, they may be useful as vaccines against pandemic influenza such as may be caused by the H5N1 strains that are currently endemic in the avian population. The influenza nucleoprotein (NP) is known to provide MHC Class I restricted epitopes that are effective in evoking a cytolytic response. Because there is only low sequence variation in NP sequences between different influenza strains, a T cell vaccine may provide heterosubtypic protection against a spectrum of influenza A strains. An AdC7 vector expressing Influenza A/Puerto Rico/8/34 NP was tested for its efficacy in protecting BALB/c mice against the two H5N1 strains and compared to a conventional human adenovirus serotype 5 vaccine. The AdC7 NP vaccine elicited a strong anti-NP T cell response. When tested in a mouse challenge model, it showed improved survival to two strains of H5N1 that have caused human outbreaks, i.e., Vietnam/1203/04 and Hong Kong/483/97, although the improved survival achieved statistical significance only with the strain from Vietnam.

156. Protective Immune Responses Against Porcine Circovirus Type 2 Infection Generated by a Recombinant Modified Vaccinia Virus Expressing the Viral ORF Antigens as Vaccines

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Porcine circovirus type 2 (PCV2) is the primary causative agent of an emerging swine disease, the postweaning multisystemic wasting syndrome. In this study, we amplified three open reading frames (Orfs) of PCV2 from infected pigs which responsive to virus replication and pathogenesis. Transgenic expressions were confirmed after cloned into clinical and modified vaccinia virus ankara (MVA) vector. We then analyzed, using the PCV2 murine model, the effectiveness of prime-boost strategy in mice that were gene-gun vaccinated with a cDNA vector followed by two boosts with MVA expressing the same Orfs of PCV2. After vaccination, we challenged test mice with PCV2 virus, and then examined the PCV2 virus titer in the lungs and lymph nodes. As compared to individual orf vaccinated mice, the combination of Orf-2 and -3 vaccinated mice was observed to result in high antibody titer, virus neutralization activity, and reduce the PCV2 virus load in the lungs and lymph nodes of test mice. Three and 1.5 fold higher expression of MIP-1β and Rantes chemokines were observed in the lymph node of test mice. Vaccinated mice showed several folds increase in Th1 type cytokine (gamma interferon), and significant specific lysis of PCV2 infected cells, as compared to control mice, and a remarkable decrease of Th2 type cytokine interleukin-10 in vaccinated mice. These results demonstrate for the first time that the prime-boost vaccination strategy by using a recombinant modified vaccinia virus might be an attractive candidate swine vaccine for future prevention of the disease associated with PCV2 infection.

157. Enhancement of Antibody and Cellurar Responses to Malaria DNA Vaccines by In Vivo Electroporation

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Increasingly, electroporation-enhanced in vivo delivery of DNA has been shown to significantly enhance transgene expression as well as the immune response against transgenic antigens. We have evaluated the effectiveness of in vivo electroporation (EP) for the enhancement of immune responses induced by DNA plasmids encoding the pre-erythrocytic *Plasmodium yoelii* antigens PyCSP and PyHEP17 administered intramuscularly or intradermally into mice. Muscle EP was performed with a two-needle array electrode, while a non-invasive meander electrode was used for EP of skin. EP resulted in a 16-fold and 2-fold enhancement of antibody responses to PyCSP and PyHEP17, respectively. Immunization with 5μg of DNA via EP was equivalent to 50μg of DNA delivered via conventional needle