

# Fourth Annual Conference

## ABSTRACTS OF SUBMITTED POSTER PRESENTATIONS

### P33

Immune response to oral administered autovaccines.

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**BACKGROUND:** Autovaccines (AV) are therapeutic vaccines derived from a disease causing micro-organism. Prosperity of AV's was during the first decades of the twentieth century. In 1914 it was reported that AV's given orally do not yield beneficial effects for the patients (1). **OBJECTIVE:** To evaluate the immune response of human patients (pts) to a specific AV's after oral or oral/subcutaneous administration of AV's. **METHODS:** A number of 9 pts received specific AV's for the treatment of persistent or recurrent infections. AV's were given either oral only or oral and subcutaneous, depending on the pts choice. 3 blood samples were collected from each pt, one prior to autovaccination as well as 7 and 28 days after start of autovaccination treatment. PBMC were stimulated using serial dilution of the antigen. 3H-thymidine incorporation was measured and the amount of TGF $\beta$  was determined in cell culture supernatants. **RESULTS:** The immune response in terms of activation of PBMC revealed that those two pts who received the AV orally only showed no change in activation of PBMC throughout the vaccination schedule. PBMC from pts receiving the AV simultaneously (i.e. oral & subcutaneous) displayed either increasing responsiveness (n = 2) or a decreasing trend in responsiveness to the antigen throughout the vaccination schedule (n = 5). **DISCUSSION:** The results indicate that following autovaccination with a high dose of antigen the specific immune response to the antigen declined. This is in particular true for those pts who received the AV oral and subcutaneous whilst those pts receiving the AV only oral did not respond to the antigen. Although in veterinary medicine oral application seems to be useful (2) this is questionable for humans.

- (1) ALLEN RW (1914): Die Vakzintherapie. Ihre Theorie und praktische Anwendung. Dresden & Leipzig verlag Steinkopff.
- (2) Nolte O, et al (in press): Autovaccination of dairy cows to treat *post partum* metritis caused by *Actinomyces pyogenes*. Vaccine

### P35 The HIV-1 Vpu Protein and Generation of CD8<sup>+</sup> T cell Responses in Mice

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The HIV-1 Vpu protein has been shown to possess a number of biological activities *in vitro* including an induced blockade of MHC class I antigens in the endoplasmic reticulum (ER). By extension, these *in vitro* experiments predict that Vpu expression could potentially interfere with class I presentation *in vivo* thereby reducing the magnitude of CD8<sup>+</sup> T cell responses in mice. To test this hypothesis, we generated HIV envelope glycoproteins having the Vpu responsive element (CD4 transmembrane and cytoplasmic domains) and these chimeric HIV envelope glycoproteins were tested in mice for the generation Env-specific CD8<sup>+</sup> T cell responses. When injected intramuscularly, DNA vaccines encoding the chimeric envelope glycoprotein induced a CD8<sup>+</sup> T cell response, and this response is enhanced several folds in mice that received DNA plasmids expressing both the chimeric envelope glycoproteins and biologically active HIV-1 Vpu. In contrast, antibody responses to the HIV envelope glycoprotein are highly attenuated in the presence of Vpu plasmids suggesting that intracellular degradation of the envelope protein mediated perhaps by the ubiquitin-proteasome system is critical for the generation of CD8<sup>+</sup> T cell responses in the mouse. Although *in vitro* blockade of MHC class I molecules by HIV-1 Vpu in antigen presentation is not clearly understood, our *in vivo* data suggest that Vpu does not impede the generation Env-specific CD8<sup>+</sup> T cell responses in mice. These mouse studies are also consistent with the natural history of primary HIV infection where effective CD8<sup>+</sup> T cell responses are generated in HIV-1 infected individuals in the early phases of AIDS disease.

### P34

Directed Molecular Evolution of Vaccine Antigens  
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MolecularBreeding™ directed molecular evolution technologies include a powerful set of methods including DNA shuffling to rapidly improve the desired traits of biological molecules. These can be performed on genes, operons and entire genomes. We will discuss their application to the *Yersinia V* antigens and staphylococcal enterotoxins to develop improved vaccines.

*Yersinia pestis*, the causative agent of plague, is one of the most virulent bacteria known and can be lethal within days. Active or passive immunization with the V-antigen can provide protection from challenge with autologous yersinia species, yet protection against heterologous species is reported to be limited. We have cloned, produced, and evaluated parental and shuffled V-antigens from *Y. pestis*, *Y. enterocolitica*, and *Y. pseudotuberculosis* in a mouse challenge model for cross-protective immune responses.

*S. aureus* and group A streptococci are involved in a range of potentially lethal diseases including food poisoning, scarlet fever, and toxic shock syndrome. Many secrete enterotoxins (SE), also called superantigens. Attenuated SE's can form the basis of protective vaccines, but protection can be limited due to the high diversity of related, yet distinct SE's. We report results on using MolecularBreeding™ directed molecular evolution technologies to create chimeric attenuated toxins with improved immune responses. Chimeras were selected for high-level expression, solubility, and the presence of multiple epitopes and then evaluated in mouse immunization studies including lethal challenge.

### P36

WITHDRAWN