38

RECENT ADVANCES IN IDIOTYPE VACCINATION FOR THE TREATMENT OF NON-HODGKIN'S LYMPHOMA.

Hendrik Veelken, Stanford University Medical Center.

The hypervariable regions of the antigen receptors (idiotypes, Id) expressed by non-Hodgkin's lymphoma (NHL) cells constitute individual tumor-specific antigens and hence attractive target molecules for induction of specific anti-tumor immune responses. In two notable phase II trials, vaccination of follicular lymphoma patients in clinical remission with Id (produced from an individual lymphoma-derived hybridoma cell line) was associated with disappearance of minimal residual disease, and superior disease-free and overall survival in patients who developed a measurable anti-idiotype immune response.

However, the vast effort to produce an individual Id vaccine by hybridoma technology has hitherto limited large-scale clinical testing. Therefore, three recombinant approaches for Id vaccine production are currently being evaluated in early clinical trials: Transfection of a cell line with expression vectors encoding lymphoma-derived IgH and IgL chains, production of Id scFv fragments in tobacco plants by a hybrid TMV vector, and expression of functionally folded Id Fab fragments in the periplasma of *E. coli*. The current experience with the bacterial expression system indicates the potential to produce a vaccine within 3 weeks of obtaining a lymphoma biopsy, and yields of up to 20 mg highly purified Fab fragment in a 10 l fermentation system under GMP criteria. *In vitro* stimulation experiments with recombinant Id Fab proved the existence of Id-specific CTL in the blood of NHL patients. Immunizations with a mixture of Id Fab and a lipid-based adjuvant i.d., combined with s.c. GM-CSF, appear to be safe and well tolerated.

39

Naked DNA and viral vectors as tumor vaccines

Gabriele Pecher
Charité Humboldt-University Berlin, Dept. of Oncology/Hematology, AG Molecular Gene- and Immunotherapy, Hessische Str. 3-4, 10115 Berlin, Germany

The use of recombinant vaccines for active immunotherapy of cancer has long attracted attention. We have constructed adenoviral and vaccinia virus-based recombinant vaccines expressing the human tumor antigen mucin or carcinoembryonic antigen (CEA) or the model antigen beta-galactosidase (β-Gal). These vaccines were compared with mucin- or CEA- or β-Gal-DNA applicated as plasmid and mock-vaccines for their efficiency to protect mice against tumors expressing mucin, CEA or β-Gal epitopes, respectively. The quantification of induced cytotoxic lymphocytes was performed by an intracellular interferon gamma staining method. C57/BL6 mice were immunized twice on day 1 and day 10. Five days after the last immunization, tumor challenge experiments were performed. Tumor protection of all animals was observed by s.c. injection of recombinant adenovirus expressing the relevant antigens followed by an i.m. boost with naked DNA. In 85% of mice immunized with the mucin plasmid, tumor growth inhibition was observed, whereas all control mice developed tumors. Re-tumor challenge after three month revealed no tumor growth in mice immunized with the mucin plasmid showing long term protection against tumor growth. These encouraging results give raise to use naked DNA or adenoviral vectors as recombinant vaccines for clinical immunotherapy.

40

Results of a Phase II HPV Vaccine Trial with Life Recombinant Vaccinia Virus TA-HPV™

A M Kaufmann, E M Rankin, H Sommer, V. Nuessler, A Schneider, M Adams, H C Kitchener, T Bauknecht, U Wagner, K Kroon, J Hickling, C M Boswell, S Stacey, P Stern, J Gillard, J Wanders, J St J Roberts, H Zwierzina

Background: Human papillomavirus (HPV) positive tumour cells present antigens of the viral oncoproteins E6 and E7 in the context of HLA class I which can be recognised by cytotoxic T-lymphocytes (CTL). We conducted a study in patients with early cervical cancer, to assess the safety and immunological effects of vaccination with TA-HPV™, a live recombinant vaccinia virus expressing modified forms of the HPV 16 and 18, E6 and E7 proteins. Methods: Patients with clinical stage Ib or IIa cervical cancer were given two vaccinations with TA-HPV™ at least 4 weeks apart, starting 2 weeks before definitive surgery. Patients were controlled closely for side effects of the vaccination. Serial blood samples were examined for HPV specific CTL or changes in levels of antibodies to HPV 16 or 18, E6 and E7 proteins and to vaccinia virus. Results: Vaccination with recombinant vaccinia was well tolerated in all patients with only mild to moderate local toxicity and no serious adverse events were attributable to the vaccine. HPV-specific CTLs were found in 4 patients after only one vaccination, and HPV-specific serological responses developed in a further 8 patients. Conclusion: This study confirms the safety and immunogenicity of the vaccine. Statistically powered clinical trials to estimate the efficacy of TA-HPV™ in patients with minimum residual disease are warranted.

41

NOVEL GENE DELIVERY SYSTEMS

Jeffrey Ulmer, Vaccines Research, Chiron Corporation, Emeryville CA 94608 USA.

Although DNA vaccines have generated significant immune responses in laboratory animal studies, including protective immunity in infectious disease models, human clinical studies of DNA vaccines have yielded weak immune responses. In general, DNA vaccines are most effective at inducing CTL responses, while helper T cell and antibody responses are more modest. Therefore, for DNA vaccines to be fully effective in humans, enabling technologies must be developed in order to increase the potency of these responses. To increase the potency of DNA vaccines we have focused on two main areas: i) form of plasmid (including alphavirus replicon DNA), and ii) improved DNA delivery technologies (including physical and particle-mediated delivery). To improve DNA delivery in vivo, two approaches were taken. Electroporation in vivo was used to facilitate uptake of DNA by muscle cells, and a cationic microparticle (PLG) with adsorbed DNA was used as a carrier to target APCs. These systems were shown to be 100- to 1000-fold more potent for induction of both antibodies and T cell responses. Therefore, these DNA vaccine technologies, including improved DNA vector and delivery systems, can serve as potent inducers of immune responses, which can be increased and broadened by boosting with formulated recombinant protein or viral vector.