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Vaccination with peptides derived from the extracellular domain of Her-2/neu elicits specific humoral and cellular immune responses in mice.

J.Jasinska[1], T.Brodowicz[1], S.Wagner[2], U.Wiedermann[2], C.Radauer[2], R Sedivy[3] M.Kubista[4], H.Breiteneder[2], Ch.Wiltschke[1], O.Scheiner[2,4],C.C.Zielinski[1,4]
1. Clinical Division of Oncology, Department of Medicine I, University Hospital, 2. Department of Pathophysiology, University of Vienna, 3. Department of Clinical Pathology, University of Vienna, and 4. BioLife Science GmbH, Vienna, Austria.

The humanized murine anti Her-2/neu specific antibody Trastuzumab is successfully used in the clinical routine for therapy of Her-2/neu overexpressing cancers. However, an active vaccination inducing a long-term immunity to Her-2/neu is still a desirable goal. The aim of this study was to elicit a Her-2/neu specific immune response in BALB/c mice by immunization with peptides derived from the extracellular domain of the human Her-2/neu. Seven different peptides were selected and coupled to keyhole limpet hemocyanin (KLH). Mice were immunized with combinations of each of two conjugated peptides, or for control purposes with KLH alone. Four subcutaneous immunizations were performed; seven days after the last immunization animals were sacrificed and antigen-specific antibody levels and T-cell proliferation were measured. Moreover, hearts, lungs, livers, and kidneys were histopathologically screened for inflammatory infiltrations. Spleen cells proliferated moderately upon stimulation with three of the peptides used for immunization. Anti-Her-2/neu antibodies - in particular of the IgG_1 isotype - were detected by immune precipitation and subsequent Western blot as well as by ELISA. These antibodies were able to precipitate human Her-2/neu from cell lysates of the human breast cancer line SKBR-3. Concerning the histopathological examinations no evidence of organ damages were detected. We conclude from our data that immunization with Her-2/neu peptide conjugates successfully induced anti-tumoural immune responses, which might contribute to the development of future peptide-based cancer vaccines.

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Her-2 expression is of independent prognostic significance in pNO prostate cancer undergoing curative radiotherapy. A.Fossa[1], W.Lilleby[2], S.D.Fossa[2], G.Gaudernack[1], G.Torlakovic[3], A.Berner[3]. Departments of [1]Immunology, [2]Medical Oncology and [3]Pathology, The Norwegian Radium Hospital, Oslo, Norway

Purpose: The prognostic relevance of Her-2 (c-erbB2) protein expression in localized prostate cancer (PC) undergoing curative radiotherapy (RT) was analyzed and compared to widely accepted prognostic factors such as prostate specific antigen levels, Gleason score and T-category.

Patients and methods: Pretreatment biopsies from 112 homogeneously treated patients (pts) with T1-4pN0M0 PC were examined by immunohistochemistry for Her-2 expression. At study end 25 pts had died of PC, with a median follow-up of surviving pts of 71 months (range 48-144).

Results: 37% of pts showed membrane bound Her-2 expression in more than 10% of cancer cells. No membrane staining was seen in normal prostate epithelium. In univariate analysis, Her-2 expression was associated with adverse outcome in terms of biochemical or clinical progression free survival (p=0.03), clinical progresion free survival (C-PFS, p=0.02) and disease specific survival (DSS, p=0.02). In multivariate analysis, Her-2 expression was independently associated with C-PFS (p=0.04) and DSS (p=0.03). By combining Her-2 expression data and Gleason score, pts with 5 year DSS of 95% could be discriminated from pts with a 5 year DSS of 79% (p<0.001).

Conclusion: Expression of Her-2 is of independent prognostic significance in localized PC undergoing definitive RT and may be helpful in risk stratification of PC pts. Studies investigating adjuvant therapy targetting Her-2 may be warranted in localized PC.

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VACCINATION WITH MODIFIED HER-2 MOLECULES CONTAINING POTENT HELPER T CELL EPITOPES INDUCES PROTECTIVE IMMUNE RESPONSES IN HER-2 TRANSGENIC MICE

D.R. Leach, L. Sonderbye, V. Renard, K. Ebbehøj, K. Gregorius, P. Birk, A. Gautam, S. Mouritsen. Pharmexa A/S, Hørsholm, Denmark

The HER-2 growth factor receptor is overexpressed in many human tumors and is an attractive target for immunotherapy. Since HER-2 is a self protein, tolerance may limit the effectiveness of vaccination. To overcome this, we constructed DNA and protein vaccines (AutoVac™) which utilize potent helper T cell epitopes derived from tetanus toxin. DNA vaccines encoding modified rat HER-2, or purified protein vaccines containing the helper epitopes were tested in two transgenic mouse models of rat HER-2 tolerance. DNA vaccination induced HER-2 specific antibody and CTL responses and significant protection to a transplantable HER-2 positive tumor line in one model, but had little effect in the other. In contrast, protein vaccination induced complete tumor protection in both transgenic models. T cell depletionexperiments indicated that the effect of protein vaccination was primarily mediated by antibodies. Additionally, treatment with a single vaccination of modified HER-2 protein could completely eliminate growing tumors, even when treatment was delayed up to 9 days after tumor implantation. Thus, AutoVac™ HER-2 vaccines can induce effective immune responses against tumors with potentially different susceptibilities to immune effector mechanisms.

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Intradermal ras peptide vaccination of colorectal cancer patients: A randomized trial of two different dose levels of mutant ras peptides with- or without GM-CSF

Marianne K. Gjertsen, Jan Petter Paulsen, Steinar Aamdal, Ingvil Sæterdal, Jon A. Eriksen, Møller Mona, Eva Vognild, Ragnhild Lothe and Gustav Gaudernack, *Section for Immunotherapy, Department of Immunology, The Norwegian Radium Hospital, University of Oslo, Norway.*

Following the observation that intradermal vaccination with single mutant ras 17-mer peptides can induce a positive immune response in approximately 50% patients, this study was designed to assess whether a higher dose of peptides could increase the percentage of responding patients. Patients with colorectal adenocarcinoma showing progression on 5FU-Leucovorin were randomized between two different doses of a five- peptide formulation. A total of 27 patients (13 male, 14 female, mean age 51.5 years) were recruited, split equally between two groups: (1) intradermal vaccination with 100 μg of each peptide together with GM-CSF and (2) intradermal vaccination with 400 μg of each peptide together with GM-CSF. In addition one group of 10 patients (7 male, 3 female, mean age 56.0 years) (3) received intradermal vaccination with 100 μg of each peptide without GM-CSF. The vaccination schedule consisted of four weekly administrations. The patients were monitored by measurement of DTH reaction and T cell responses were assayed in pre- and post vaccination PBMC. DTH responses were observed in 11 of 14 patients in group 1 (78%), and in 7 of 13 patients in group 2 (54%). In group 3 there were no DTH responses observed, indicating that peptide alone is poorly immunogenic. Only minimal adverse events were observed including local redness at the injection site and chills. T cell response data correlated well with the observed DTH responses. We conclude that GM-CSF is necessary to induce a positive immune response to the peptide vaccine and that the dose levels used are at the plateau. This observation is important in the future vaccine development in specific immunotherapy of cancer patients.