456

Antigens recognized by T lymphocytes on kidney cancer

B.J. Van den Eynde[1], M. Probst-Kepper[1], T. Boon[1]. [1]*Ludwig Institute for Cancer Research, Brussels, Belgium*

Shared tumor-specific antigens have been defined at the molecular level, and some of them are currently being tested in clinical trials as cancer vaccines. They are expressed mainly in melanomas, bladder carcinomas, non-small cell carcinomas and head and neck carcinomas. Although there is indirect evidence suggesting that renal cell carcinoma (RCC) is a tumor type that can be controlled by the immune system, very little is known about the antigens recognized by cytolytic T lymphocytes (CTL) on RCC. We previously identified two antigens on RCC. One is encoded by gene RAGE, a gene that is silent in most normal tissues but is expressed in RCC and in a few other tumors. However, although it is expressed in more than 30% of RCC cell lines, only a few percent of fresh RCC samples express RAGE. The other is a unique antigen resulting from a point mutation in the HLA-A2 gene of the RCC of a given patient.

We have pursued our effort aimed at identifying antigens recognized by CTL on RCC. We have identified a new antigen that results from the transcription of the reverse strand of a new housekeeping gene that we named RUR-1. This antisense transcript is expressed in a high proportion of tumors of many histological types. However it is also expressed at a low level in normal kidney, bladder and testis. Accordingly, we observed that cell lines derived from the proximal tubule epithelium of normal kidney were recognized by the CTL. We have also defined another RCC antigen that results from the translation of an alternative open reading frame of the M-CSF messenger RNA, which is strongly overexpressed in carcinomas of the kidney, bladder, prostate, breast, uterus and ovary. This antigen also appears to be expressed by the normal proximal tubule epithelium.

Because of their expression in normal kidney, these two new antigens are not tumor-specific, and the potential benefit of using them as targets for immunotherapy of RCC should be balanced against the risk of inducing autoimmunity against the proximal tubule.

457

Dendritic and tumour cell -derived exosomes as novel cancer vaccines

L. Zitvogel[1], A. Lozier, J. Wolfers, A. Regnault, C. Masurier, N. Fernandez, G. Raposo, S. Amigorena. [1]*Institut Gustave Roussy, Immunotherapy Depart. 12th floor, Villejuif, France*

We reported (Nat. Med. May 1998) that immature dendritic cells (DC) secrete antigen presenting vesicles of endosomal origin, namely "exosomes" that are potent cell free vaccines. Such DC derived-exosomes carry a discrete set of proteins compared with whole DC membranes that may be involved in antigen processing/presentation and targeting to effectors of the immune system. Here we show that not only DC but also tumor cells secrete such exosomes i.e 60–90 nm membrane vesicles that bear tetraspanins, lysosomal markers and MHC class I molecules, but also high levels of heat shock proteins hsc70, hsp80 (but no gp96). Interestingly, tumor derived-exosomes pulsed onto DC mediate potent CD8+ T cell dependant antitumor immunity in tumor beating mice in syngeneic models. Surprisingly, cross-protection between different histological types could be achieved using such immunization, i.e colon carcinoma derived-exosomes were efficient against mammary murine tumors of a different H2 background. We found that such tumor derived-exosomes (TEX) could contain cytosolic proteins (p53, MART1/MelanA, gfp) in addition to HSP that might account for their potent immunogenicity. Cross-presentation experimental setting was performed in vitro, using HLA A2−/MART1+ containing melanoma derived-exosomes loaded onto monocyte derived-DC (HLA-A2+) in order to stimulate a MART1 specific/A2 restricted CTL clone. While cross-presentation was not effective with native cytosolic MART1 proteins, TEX were dramatically capable of triggering CTL activation. These data support implementation of TEX for DC loading in cancer immunization protocols and suggest that TEX may contribute to the antigenic cross talk between rumors and APCs in vivo.

458

T-cell immunity against tumors, a delicate balancing act involving dendritic cells

C. Melief[1]. [1]*AZ Leiden, Department of Immunohematology & Blood Bank, Leiden, Netherlands*

T-cell immunity against tumors, a delicate balancing act involving dendritic cells. C.J.M. Melief Dept of Immunohematology and Blood Bank. University Hospital, Leiden, The Netherlands. T-cell immunity occurs naturally against tumors induced by viruses and other causes. In the latter case self antigens are increasingly found to be targets of tumor associated CTL. In all categories of tumors the T cell response usually falls short of the maximally possible response. This situation calls for vaccination, primarily in situations of low tumor burden and adoptive transfer with tumor specific T cells in case of higher tumor burden. We recently observed that the outcome of immunization with vaccines containing tumor virus CTL epitopes strongly depends on mode of epitope delivery. Surprisingly, vaccination with MHC class I binding peptides cause CTL tolerance associated with enhanced tumor outgrowth rather than immunity. Such specific CTL tolerance can be induced by a single infection of peptide in adjuvant. However, in vivo presentation of the same peptides on dendritic cells or in viral vector (adenovirus) causes strong antitumor protection. Thus tumors may escape from immune attack by specific tolerance induction. Tumor specificity of autoreactive CTL can be achieved by T cells directed against tumor associated self antigens of limited tissue distribution. Alternatively useful CTL can be directed against strongly overexpressed self antigens, as illustrated in our lab by the successful eradication of tumors overexpressing wild type p53 tumor suppressor protein, by the adoptive transfer of a wild p53-specific CTL clone. Apparently the low expression of p53 in many tissues does not cause the CTL clone to inflict tissue damage, while the p53 overexpressing tumor cells are specifically targeted and eradicated. Recently we showed that CD40 signalling can replace CD4+ T-cells in priming of helper dependent tumor-specific CD8+ responses. Blockade of CD40L results in profound inhibition of CTL priming that is overcome by CD40 signalling.

459

Abstract not received.

460

Vaccination with gene-modified tumor cells

Giorgio Parmiani. *Istituto Nazionale Tumori, 20133 Milan, Italy*

Previous studies in animal models showed that cytokine gene-modified cancer cells can be used to immunize mice against subsequent challenges of tumor cells and even to induce a therapeutic response in tumor-bearing animals. Based on this rationale, several studies have been planned and carried out in cancer patients, particularly in metastatic melanomas. Upon vaccination with either allogeneic or autologous melanoma lines transduced with IL-2, IL-4, GM-CSF or IL-12 genes, a limited clinical response was reported which is, however, accompanied with a T-cell reaction in a fraction of patients and, in the case of GM, with a strong local inflammatory-like reaction at tumor sites.

In an attempt to improve these results, we have constructed a new vaccine by transducing melanoma lines, selected for expression of several T and B cell-recognized antigens, with both the IL-2 and the B7.1 genes by a polycistronic retroviral vector. These lines were then characterized for the expression of transduced genes and used to stimulate both allogeneic HLA-A-2.1-matched and autologous patients' lymphocytes taken either from peripheral blood or from tumor-invaded lymph nodes. Double transduced melanoma lines (Me/B7/IL-2) were able to stimulate patients' lymphocytes more effectively than the lines transduced with either gene, with Me/B7 line still being more immunogenic than parental, untransduced line. This was shown to occur both in cytotoxic and cytokine-release assays. Thus, although cross-priming may substitute for direct tumor-mediated presentation of tumor antigens, gene-modified cells can still provide an advantage over parental cell vaccines due to induction of inflammatory-like reaction that, particularly in already antigen-primed patients, can favor the expansion of tumor-specific T cells.

461

Important issues in palliative oncology

E.J. Maher[1]. [1]*Mount Vernon Hospital, Regional Radiotherapy and Oncology Centre, Northwood Middlesex, United Kingdom*

There have been many changes in perspective of "palliative oncology" over the last 10 years: the movement from generalist to specialist; the recognition of a clear distinction between the palliative and terminal phase; the expansion of life quality instruments; and the patient's voice being more directly heard. The impact of specific interventions in the palliative phase make up an increasing part of an oncologists' research and development