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

portfolio. The growth of the specialty of palliative medicine represents both an opportunity and a challenge. Cancer as a progressive chronic disease requires management by a multidisciplinary team – the members of which may have different perspectives. The challenge is to provide patient-centered care, using all the interventions available to us.

462

Pain control for bone metastases

J.W.H. Leer, E. Steenland, H. van Houwelingen. *On behalf of the Dutch Bone Metastases Collaborative Group; University Hospital Nijmegen, 341 Institute of Radiotherapy, P.O. Box 9101, 6500 HB Nijmegen, Netherlands*

Purpose: to improve palliative care of patients with painful bone metastases by reducing the number of visits to the radiotherapy department. The effect on palliation of a single fraction of 8 Gy was compared to that of a total dose of 24 Gy in 6 fractions.

Patients: 1171 patients were randomized. The primary tumour was in the breast in 39%, in the prostate in 23%, in the lung in 25% and in other locations in 13% of the patients. Bone metastases were located in the spine (30%), the pelvis (36%), femur (10%), ribs (8%), humerus (6%) and other sites (10%).

Method: questionnaires were used to collect information on pain, analgesic consumption, side effects during treatment, quality of life and costs. Questionnaires were sent out every week up to 3 months, thereafter every 4 weeks up to 2 years. Pain was measured on a pain scale from 0 (no pain) to 10 (worst imaginable pain).

Results: on average patients participated in the study for 4 months. The median survival was 7 months. Different techniques were used to analyse the pain data. Overall no differences between the two treatment schedules were found. On average patients lowered their pain score from 7 to 4 and it was shown that this reduction occurred mainly in the first 4–6 weeks. The response rate was 89% defined as a decrease of at least 2 points as compared to the initial pain score. With regard to quality adjusted life years similar results were found. The number of retreatments was 188 (16%). This number was higher in the 1 × 8 irradiation group, namely 147 (25%) versus 41 (8%) in the 6 × 4 group. More pathological fractures occurred in the single dose group. The actual percentage however, was still below the percentages mentioned in other studies.

Conclusion: given the quality in the effects on palliation, we had to conclude that a single fraction is preferred in patients with painful bone metastases, even at the expense of a higher chance of retreatment. The results of the Dutch trial will be discussed with reference to other studies on this subject.

463

Abstract not received.

464

Radiation treatment for bone metastases from prostrate cancer

M. Russell[1]. [1]*Beatson Oncology Centre, Consultant Oncologist, Glasgow, United Kingdom*

Bone pain affects the majority of patients with metastatic prostate cancer. Following relapse from first-line hormonal treatment, palliation of such pain may include analgesia, further hormonal treatment, chemotherapy and bisphosphonates, but external beam radiotherapy (EBRT) or radio-isotope treatment is often the best option.

In randomised trials of palliative local EBRT, although in patients with a variety of cancers, not specifically prostate, a single radiation treatment of 8 Gy is comparable to fractionated regimens. Pain relief should be achieved in 70% to 80% of patients. Fractionated EBRT may still be appropriate where there is a low metastatic burden, for weight-bearing bones, and where there is a risk of impending pathological fracture or spinal cord compression. Hemibody radiotherapy has the attraction of treating multiple sites of metastases at one time provided they are confined to one "half" of the body, but while the response is rapid (50% within 48 hours, 80% within 7 days) many patients will experience nausea and vomiting, and diarrhoea is not uncommon in patients who receive "lower hemibody" treatment. Bone marrow suppression for a variable length of time may occur.

Early clinical trials with Sr89 demonstrated efficiency in end-stage patients, and two randomised controlled trials, from Canada and the UK, subsequently confirmed its benefit. In the Canadian trial, Sr89 showed a clear improvement in outcome when added to local EBRT, in terms of response rate, duration of effect, reduction in new sites of pain, and cost effectiveness. In the UK trial, two groups of patients were enrolled, one where painful metastases were felt suitable for local radiotherapy, and another where hemibody radiotherapy was more appropriate, and within each of these two groups patients were randomised to receive EBRT or Sr89. Sr89 was equivalent to EBRT in terms of response rate and duration, but Sr89 reduced the frequency with which new metastases developed. In both of these trials no survival benefit could be shown, and the main toxicities were an increase in bone pain for two or three days after the injection, and a reduction in platelets. Other isotopes, including Re186 and Sam153, have been less extensively researched, but appear to produce an earlier response than Sr89, but with a shorter duration of action. Sr89 has subsequently been combined with chemotherapy in an attempt to improve the response rates further, and currently the UK group is examining the role of Sr89 in patients with evidence of biochemical relapse from first line hormonal treatment.

465

Brain metastases – Radiosurgery or whole brain irradiation?

R.E. Wurm, L. Schlenger, A. Kaiser, M. Kömer, M. Fitzek, L. Röschel, D. Böhmer, G. Matnjani, M. Stuschke, V. Budach. *Radiation Oncology, Charité, Humboldt University, Berlin, Germany*

Brain metastases is an important cause of morbidity and mortality in cancer patients. The median survival for patients with symptomatic metastases to the brain is about 1 month if they remain untreated and about 3 to 6 months if they undergo conventional whole brain irradiation (WBRT). Patients treated with surgery and postoperative radiotherapy have a significantly longer survival, improved quality of life compared to those treated with WBRT alone. Following complete surgical resection no significant difference in overall length of survival or the time of functional independence has been noted. However, the frequency of recurrences and morality due to neurological cause is lower. A number of reports have shown that stereotactic, radiosurgery (SRS) is an effective, noninvasive therapeutic, approach that can produce substantial functional survival, especially in patients with good performance status and without extracranial metastasis when used alone or in combination with WBRT. The results of SRS are comparable to the aforementioned recent randomised trials of resection and WBRT. There is evidence that the efficacy of SRS is not increased by adding WBRT and there is a trend to withhold WBRT in as many cases as possible to avoid both the short- and long-term morbidity of that treatment. Furthermore the advantages of SRS over surgery in terms of cost, hospitalisation, morbidity, and wider applicability strongly suggest that a randomised trial to compare SRS with surgery in combination with and without WBRT is warranted. This would help to clarify should SRS be used instead of surgery and followed by WBRT, adjuvantly with WBRT, or on tumour progression or recurrence after WBRT.

466

Retreatment in head and neck squamous cell carcinoma

J. Bourhis, R. DeCrevoisier, F. Eschwege. *Institut Gustave Roussy, Villejuif, France*

Our experience of full dose re-irradiation (re-RT) in head and neck carcinoma (HNC) was reviewed in a series of 169 patients who presented with an inoperable HNC in a previously irradiated area (>50 Gy). The treatment consisted of a combination of radiotherapy (60 Gy) and chemotherapy (CT) (mainly 5FU-Hydroxyurea). Toxicity was markedly increased compared to the toxicity due to the first RT, but still remained acceptable. A 37% complete remission was observed at 6 months and the overall survival rate were 21 and 9% at 2 and 5 years, respectively. The median survival was 10 months which is higher that that usually reported in comparable studies using palliative CT. These findings are in agreement with few other studies showing that full-dose re-RT combined with CT is feasible in inoperable HNSCC and could lead to a small proportion of patients long-term free of disease.

These findings strongly suggest that re-RT + CT might constitute a standard in the difficult and frequent situation of HNSCC inoperable relapse in a previously irradiated area. However, in spite of the encouraging results, it remains to be proved that the combination of re-RT and CT is superior to palliative CT alone. It that aim, we have started a randomized trial within the GORTEC (French Group for Head and Neck Oncology), comparing re-RT + CT to CT alone. In addition, since there is strong evidence of added toxicity due to the re-RT, it is necessary to attempt to minimize this toxicity, which could be further tested by optimizing the re-irradiated volumes (3D conformal