

THERAPEUTIC VACCINATION OF CANCER PATIENTS WITH TUMOR-SPECIFIC ANTIGENS

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Tumor cells carry antigens such as MAGE antigens that are absent from normal tissues, and that can be targeted by cytolytic T lymphocytes (CTL) (1). While it is possible to make such CTL recognize and kill autologous tumor cells in vitro, the precise way to induce an effective CTL response against a MAGE antigen in cancer patients is not known yet. In clinical vaccination trials, patients with a MAGE expressing cancer, often melanoma, are treated repeatedly with a MAGE vaccine. These trials have two main objectives. First, the effectiveness of various vaccination modalities can be assessed by following the clinical evolution of the tumor, by analyzing whether a specific CTL response to the vaccine antigen occurred, and by determining whether immunological and clinical responses are correlated. Secondly, T lymphocytes and tumor samples collected at different timepoints during vaccination can be analyzed in detail, which improves our understanding on what happens in patients who experience regression of metastatic lesions, and which may explain why this does not happen in the majority of patients with overall disease progression. This knowledge can then be used to design new vaccination modalities.

Therapeutic vaccination with MAGE tumor antigens

We have set up small-scale clinical trials aimed at evaluating the toxicity, the clinical evolution and the immunological response in cancer patients immunized with MAGE vaccines involving either peptides, a recombinant protein or a recombinant viral vector. A total of about 330 patients have been included in these multicentric trials.

Current status of the trials

Twenty-one melanoma patients included in the ongoing study LUD 97-004 have received 9 immunizations with the MAGE-3.A1 peptide, injected intradermally (ID) and subcutaneously (SC) every 10-11 days. Tumor regression was observed in three patients, who had a mixed response. As compared with monthly immunizations with the same peptide, which were associated with 7 regressions among 26 evaluable patients (2), the increase in the vaccination

frequency does not seem to improve the clinical benefit. Using in vitro PBL stimulation in limiting dilution conditions followed by HLA/peptide tetramer staining, anti-MAGE-3.A1 CTL responses were detected in 1 of 2 patients with regression, and in none of 4 patients with disease progression. Seven additional patients have received the same peptide associated with the MAGE-3.DP4 peptide, in order to induce simultaneous CD4+ T lymphocyte responses. No regressions were observed, suggesting that no clinical benefit is obtained by the addition of this HLA class II restricted peptide. In a future clinical trial, the MAGE-3.A1 peptide will be mixed with an immunostimulatory CpG-containing oligonucleotide to increase its immunogenicity. In another trial, we will test whether a combination of 4 peptides (MAGE-3.A1, NA17.A2, tyrosinase.A2 and Melan-A.A2) that are individually associated with regression of melanoma metastases will improve the tumor response rate.

The clinical efficacy of the MAGE-3 protein injected ID and SC without adjuvant in non-visceral melanoma patients was tested in study LUD 99-003. Patients received 300 µg of MAGE-3 protein

on 6 occasions at 3-week intervals. To date, 5 out of 26 evaluable patients have shown regressions, including 1 complete response lasting for more than 1 year. Thus this vaccine does not seem to induce more regressions than the MAGE-3.A1 peptide, but it does not require that the patient carries a specific HLA type. We will now mix this recombinant protein with adjuvant AS15 containing an immunostimulatory CpG nucleotide, and combine these IM injections with the administration of selected class I or class II peptides by ID and SC routes, which may result in the simultaneous activation of both CD8+ and CD4+ specific T lymphocytes (Study LUD 02-002). Moreover, since a patient with metastatic bladder cancer experienced regression of lymph node metastases upon immunization with the MAGE-3 protein mixed with the adjuvant SB AS-2 (3), simultaneous administration of a MAGE recombinant protein and of some corresponding MAGE peptides will also be tested in the neo-adjuvant setting, in patients with bladder cancer (Study LUD 01-013).

In the LUD 97-005 trial, 40 patients with advanced cancer, including 37 with melanoma, were vaccinated with a recombinant canarypox (ALVAC) virus containing a minigene that encodes the MAGE-1.A1 and MAGE-3.A1 antigens, followed by booster vaccinations with the 2 corresponding peptides. The treatment comprised 4 ALVAC injections followed by 3 peptide injections, all ID and SC, separated by 3 weeks each. Local inflammatory reactions at the sites of ALVAC injection were common, but were moderate in intensity and transient in duration. Among the 30 melanoma patients who received at least 4 ALVAC vaccinations, six experienced regression of one or more melanoma metastases. Significant CTL responses were detected in 4 of the 6 patients with regressions, and in 2 of 11 patients with disease progression using our tetramer assay. We plan to investigate in a new trial whether increasing the dose of ALVAC would result in improved immunological and clinical responses.

In study LUD 01-006, patients with completely resected primary or regional metastatic melanoma with a high risk of relapse have been vaccinated with either the MAGE-3.A1 or MAGE-10.A2 peptide injected ID and SC every 2 weeks on 6 occasions. The objective of this ongoing trial is to analyze whether vaccination of melanoma patients with less advanced disease in the adjuvant setting improves the immunological response to a peptide vaccine. Up to now, no CTL response has been detected by our tetramer assay in the 13 patients who have received the complete treatment, including 7 patients with a resected tumor that did not express the appropriate antigen and who are assumed to be immunologically naive.

Relevant observations

Immunization with MAGE peptides, the MAGE-3 recombinant protein or the ALVAC recombinant viral vector, is devoid of significant toxicity.

A minority of melanoma patients (about 20 %) show regression of metastatic lesions following immunization, whatever the MAGE vaccine used. About 10% of the patients show complete or partial clinical responses. Some of them lasted for several years (4). This frequency is far beyond the reported incidence of spontaneous regressions of melanoma metastases, estimated at 0.2-0.3%, indicating that these regressions are linked to the vaccinations.

CTL responses can be detected in a minority of patients vaccinated either with peptide or ALVAC virus. The responses appear to be weak and are mainly monoclonal. The relative frequency of CTL responders versus non-responders is higher in patients who had tumor regressions (5). However, more patients need to be analyzed before statistically significant conclusions can be drawn.

No increased immunogenicity of peptide vaccines was observed in disease-free patients with less advanced melanoma, as compared with patients with active, antigen-bearing metastases.

Selected publications

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