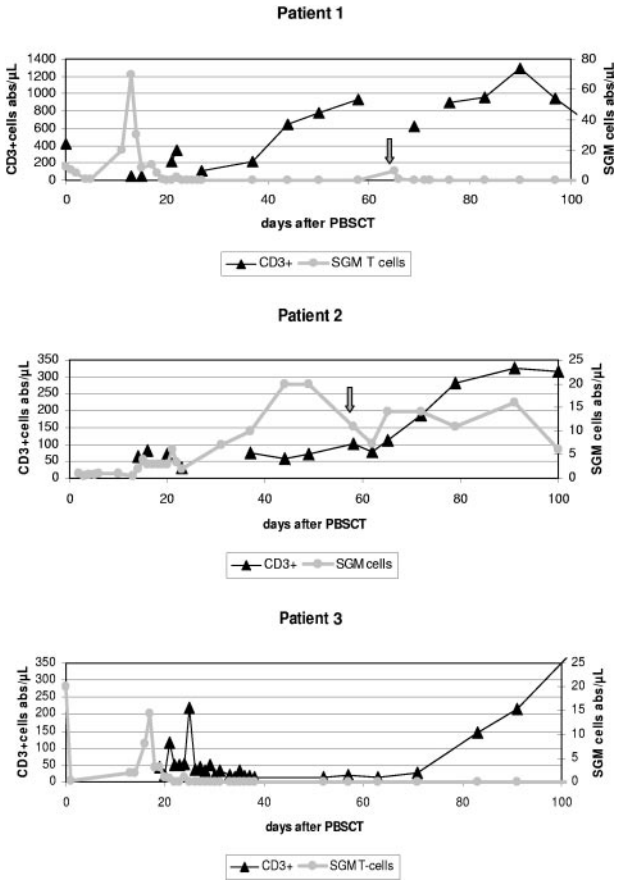

Figure 1. Absolute CD3$^+$ T-cell counts (including CD3$^+$CD56$^+$) and numbers of suicide gene–modified (SGM) donor T lymphocytes during the first 100 days after PBSCT in all 3 patients treated according to our protocol. Patient 2 showed stable numbers of SGM cells for about 3 months accompanied by increasing absolute CD3$^+$ counts and full donor chimerism. In contrast, in both patients 1 and 3 early in vivo depletion of SGM donor T lymphocytes was observed, mediated by ganciclovir applied to treat an acute GvHD grade II (patient 3) or most probably as the result of an anti–HSV-thymidine kinase (tk) immune reaction (patient 1). Both patients appeared to have higher absolute CD3$^+$ counts on day 100 compared with patient 2, but developed mixed chimerism (not shown) and eventually rejected their grafts at days 156 and 119. Arrows indicate a second donor SGM T lymphocyte infusion in patient 1 (day 65) and patient 2 (day 58). Note that different Y-axes should be applied to CD3$^+$ and SGM cells.

below the suggested threshold of 2×10^5/kg,[8] but 5×10^6 SGM T cells/kg was added. Obviously, complete loss of these SGM T lymphocytes about 3 weeks after transplantation in both patients (Figure 1) led to a situation comparable to full T-cell depletion. This, probably in concert with other factors (HLA mismatch + busulphan conditioning for CML, patient 1; slightly reduced busulphan dose, patient 3[8]), may have facilitated autologous T-cell recovery, as indicated by T-cell chimerism data from patient 3 (not shown).

In conclusion, we confirmed earlier reports[1,2] that the use of SGM T lymphocytes may allow control of acute GvHD. At the same time, we made the troubling observation that early total in vivo depletion of SGM donor T cells may be associated with an increased risk of transplant rejection. This suggests that minimum numbers of donor CD3$^+$ cells are required after transplantation, not only to facilitate engraftment[8] but also to prevent late rejection. To preclude the rejection risk associated with HSV-tk–mediated depletion of donor T cells, add-back of limited numbers of nonmodified T lymphocytes (eg, 2×10^5 CD3$^+$/kg) to the CD34-enriched PBSCs[8] may be a valuable approach.

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One of the authors (K.K.) is employed by a company (EUFETS) whose (potential) products may be related to the present work.

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To the editor:

Identification of immunodominant alloreactive T-cell epitopes on the Jk^a red blood cell protein inducing either Th1 or Th2 cytokine expression

Although antibodies implicated in red blood cell (RBC) alloimmunization have been studied for many years, little is known about helper T-cell responses that drive their production.[1-3] The aim of this work was to determine T-cell antigenic determinants involved in T-cell responses against RBC antigens, and the subsequent patterns of stimulatory cytokine production. The Kidd blood group antigen, Jk^a, was selected as a model. T lymphocytes from 11 donors whose anti-Jk^a alloimmunization was the result of previous pregnancies were stimulated during 6 hours by 4 overlapping peptides mimicking the Jk^a sequence 266 to 293 (Jk^{a}1, Jk^{a}2, Jk^{a}3, Jk^{a}4). The real-time reverse-transcriptase–polymerase chain reaction was chosen to quantify T helper 1 (Th1)– and Th2-type cytokines (respectively, interleukin-2 [IL-2] and IL-4). The results showed a clear Th1/Th2 dichotomy in

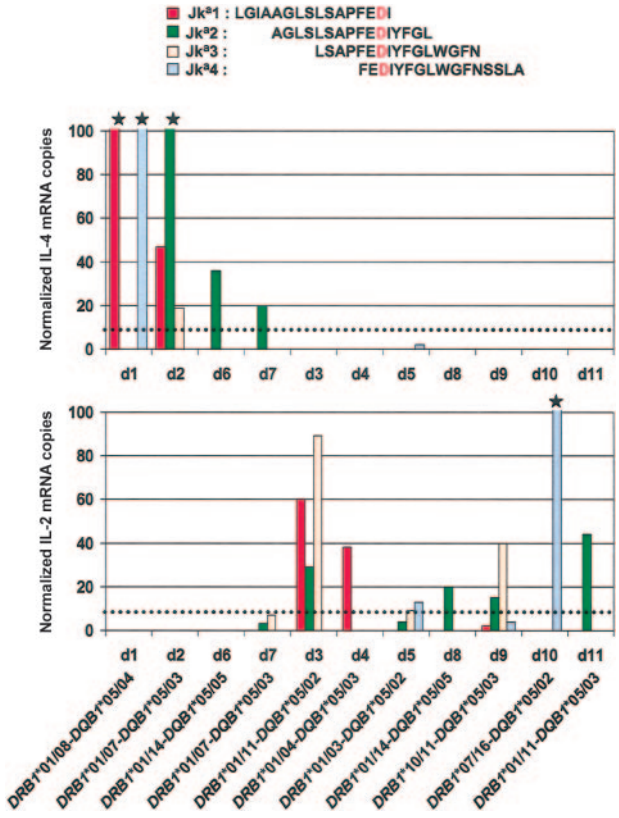

Figure 1. IL-2 and IL-4 mRNA quantifications among T cells stimulated by Jk^a synthetic peptides. The sequences of the 4 overlapping peptides tested are indicated at the top of the figure. The Asp280 (D) is indicated in red. The value of cytokine expression after stimulation was calculated after removing the background value of cytokine production by unstimulated peripheral blood mononuclear cells. The star indicates that the value obtained is more than 100. The dotted lines indicate the threshold for specific positive responses due to alloimmunization. Anti-Jk^a alloimmunized donors (d) are in abscissa. The donor *DRB1*/DQB1** low-resolution typing is indicated at the bottom of the figure.

cytokine responses induced by 2 immunodominant peptides: Jk^{a}1 and Jk^{a}2 (Figure 1). There were 2 populations of anti-Jk^a alloimmunized donors individualized: 4 donors with a Th1 (IL-2$^+$/IL-4$^-$) response, and 7 donors with a Th2 (IL-4$^+$/IL-2$^-$) response. In response to the same peptide, 2 different donors may produce different cytokines (IL-2 or IL-4). As a control of peptide-specific response, the nonalloimmunized population (7 donors) did not produce significant levels of cytokines (data not shown).

This Th1/Th2 dichotomy was not due to delays in kinetics of IL-2 and IL-4 productions, whatever the poststimulation time tested (3, 6, 20 hours; data not shown). Nor was it related to particular donor *DRB1** or *DQB1** molecules. On the contrary, the frequencies of *DRB1*01* (82%) and *DQB1*05* (100%) phenotypes observed in these alloimmunized donors producing anti-Jk^a, without other alloantibodies, were higher compared with the expected phenotypic frequencies in the white population, 18.1% and 20.8%, respectively.[4] This observation raises the question as to whether these molecules are associated with genetic susceptibility to Jk^a alloimmunization, as previously demonstrated for *DRB3*0101* and alloimmunization against platelet-specific antigen HPA-1a.[5] The Jk^{a}1/Jk^{a}2 immunodominance in this population may be explained by the high affinity of both peptides for *DRB1*01* as indicated by the HLA-peptide binding motif predictions (data not shown).[6] Because *DQB1*05* binding prediction data are not available, we cannot exclude that *DQB1*05* increased frequency is the consequence of the known linkage disequilibrium with *DRB1*01*.[7] Jk^{a}1 and Jk^{a}2 peptide presentation by HLA–class II molecules are currently being investigated.

In conclusion, our data suggest that (1) a clear dichotomy (IL-2/IL-4) exists among anti-Jk^a alloimmunized donors in response to specific Jk^a peptide stimulations; (2) the response induced by one peptide may vary (IL-2/IL-4) depending on the donor tested; and (3) as expected from peptide binding motif predictions, the Jk^a protein contains limited dominant T-helper epitopes, and the *DRB1*01* molecule could be implicated in the Jk^a peptide presentation.

A better knowledge of cellular alloimmunization against Jk^a antigen could be extended to other RBC antigens. Identification of such immunodominant peptides, the cytokine patterns induced, and the HLA class II molecules implicated in their presentation would facilitate the design of new therapeutic strategies including the specific control of alloimmunization with peptide antigen tolerogens or the ex vivo induction of regulatory T cells.

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To the editor:

Chondrogenic differentiation of mesenchymal stem cells is inhibited after magnetic labeling with ferumoxides

Magnetic resonance (MR) tracking of magnetically labeled cells is a relatively new technique to noninvasively determine the biodistribution and migration of transplanted or transfused stem cells in vivo.[1] The recent paper by Arbab et al[2] represents a significant step forward in bringing the prospect of magnetic resonance imaging cell tracking into the clinic. They describe the use of protamine