Patterns of Cytomegalovirus Reactivation Are Associated with Distinct Evolutive Profiles of Immune Reconstitution after Allogeneic Hematopoietic Stem Cell Transplantation

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T cell–mediated immunity is essential for the control of cytomegalovirus (CMV) infections in patients undergoing allogeneic hematopoietic stem cell transplantation (allo-HSCT). Our aims were to identify patterns of CMV-specific immune responses associated with multiple or prolonged reactivations. We analyzed findings in 116 recipients during the course of infection or reactivation and latency. CD8+ T cell responses were determined weekly, using HLA class I tetramers together with extended phenotypic analyses. Our results confirmed that recipients of allo-HSCT from unrelated donors were more susceptible to multiple reactivations and that the donor’s CMV serological status influenced the occurrence of prolonged reactivations. We found that a lack of CMV-specific T cells after the first episode of reactivation was associated with multiple subsequent reactivations. In patients with uncontrolled reactivations, CMV-specific T cells of the late differentiation phenotype CD45RA+CD27−CD28− did not develop. Longitudinal evaluation of CD27 and CD45RA expression within the tetramer-positive subset could help identify patients in whom a protective immune response is developing. Evaluation of CMV-specific immune responses during the first episode of reactivation, together with extended phenotypes, could thus improve immune monitoring, especially in recipients at risk of uncontrolled viral reactivation.

Evaluations of cytomegalovirus (CMV)–specific T cells in recipients of allogeneic hematopoietic stem cell transplantation (allo-HSCT), using tools that directly quantify specific immune responses, have already been reported. Major histocompatibility complex class I tetramer-peptide complexes and functional assays have been used to follow the rate of the specific antiviral response recovery, considering viral load, donor serological status, stem cell origin, the effect of T cell depletion, the mode of antiviral therapy, and the kinetics of global immune reconstitution [1–10]. CMV antigenemia has been associated with a more rapid development of CMV-specific immune responses, and the role played by specific CD8+ T cells as a marker of protection against CMV disease [11–14] has been widely confirmed [2, 3, 5, 7, 15]. Although these studies provided a better understanding of the kinetics and magnitude of CMV-specific immune responses during immune reconstitution, they were performed either in small cohorts or at relatively late time points. Therefore, some aspects of the dynamics between CMV reactivation and immune responses remain to be clarified, particularly in a larger cohort of patients with different patterns of reactivation. Indeed, prolonged reactivations, as well as multiple reactivations, may contribute to increased morbidity. Unex-
Expectedly, such reactivations occur despite the presence of CMV-specific T cells and may be due to qualitative disorders rather than quantitative defects of the immune response.

Recognition of viral antigens by the immune system induces a coordinate number of changes in lymphocyte subsets, including changes in cell surface molecules, lymphocyte migratory properties, the ability to proliferate, and T cell–mediated cytotoxicity [16, 17]. Persistent viruses elicit highly diverse functional responses. A linear differentiation model based on the cell surface [16, 17]. Persistent viruses elicit highly diverse functional properties, the ability to proliferate, and T cell–mediated cytotoxicity [16, 17]. The expression of the lymph node homing receptor CCR7 added to the complexity of the definitions of central memory and effector memory T cell subsets [20]. On stimulation, naive CD3+CD8+CCR7+CD45RA+ T cells give rise to distinct populations of central memory (CCR7+CD45RO−), effector memory (CCR7–CD45RO+), and differentiated memory (CCR7–CD45RA+) cells with distinct effector functions [21, 22]. During chronic infection, CMV-specific memory CD8+ T cells display a predominantly late phenotype [23]. We considered the possibility that the phenotype of CMV-specific T cells might differ between patients with controlled and those with uncontrolled reactivation. We benefited from the frequent monitoring of CMV reactivation after allo-HSCT and weekly determination of the viral load. We analyzed findings in 116 patients after allo-HSCT during the course of infection or reactivation and focused on the response against the tegument protein pp65, using HLA-class I tetramers and extended phenotypic analyses. Our aims were to identify biological markers that could be associated with multiple and/or prolonged reactivations.

**METHODS**

**Patient characteristics and blood samples.** We studied 116 HLA-A*0201 and/or HLA-B*0702 patients who underwent allo-HSCT from an HLA-identical sibling (n = 58) or a matched unrelated donor (n = 58). All recipients received non–T cell–depleted allo-HSCT (table 1) at the Hematology and Bone Marrow Transplantation Unit of Saint Louis Hospital, Paris. All donor–recipient pairs were typed at the allelic level for HLA class I and class II loci. Before transplantation, serum samples from recipients and donors were analyzed by ELISA for CMV-specific IgG antibodies. Heparin-treated blood (15 mL) was collected from recipients before and at intervals of up to 381 days after allo-HSCT. Samples were collected weekly from day 15 to day 90 and at subsequent visits, when CMV antigenemia was determined. Written informed consent was obtained from patients, and the Committee on Medical Ethics of Saint Louis Hospital approved the investigation.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>All recipients (n = 116)</th>
<th>Seropositive recipients (n = 66)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Donor’s relation to patient</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sibling</td>
<td>58 (50)</td>
<td>35 (53)</td>
</tr>
<tr>
<td>Unrelated</td>
<td>58 (50)</td>
<td>31 (47)</td>
</tr>
<tr>
<td><strong>Source of stem cells</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Peripheral blood</td>
<td>44 (38)</td>
<td>24 (36)</td>
</tr>
<tr>
<td>Bone marrow</td>
<td>69 (59)</td>
<td>41 (62)</td>
</tr>
<tr>
<td>Cord blood</td>
<td>3 (3)</td>
<td>1 (2)</td>
</tr>
<tr>
<td><strong>Donor sex/recipient sex</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female/male</td>
<td>25 (22)</td>
<td>11 (17)</td>
</tr>
<tr>
<td>Female/female</td>
<td>27 (23)</td>
<td>13 (20)</td>
</tr>
<tr>
<td>Male/female</td>
<td>20 (17)</td>
<td>13 (20)</td>
</tr>
<tr>
<td>Male/male</td>
<td>44 (38)</td>
<td>29 (44)</td>
</tr>
<tr>
<td><strong>Means age, years</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Recipients</td>
<td>30.8</td>
<td>28.2</td>
</tr>
<tr>
<td>Donors</td>
<td>32.6</td>
<td>28.9</td>
</tr>
<tr>
<td><strong>Diagnosis</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hemoglobinopathy</td>
<td>2 (2)</td>
<td>2 (3)</td>
</tr>
<tr>
<td>Aplasia</td>
<td>11 (9)</td>
<td>7 (11)</td>
</tr>
<tr>
<td>Acute lymphoblastic leukemia</td>
<td>24 (21)</td>
<td>15 (23)</td>
</tr>
<tr>
<td>Non-Hodgkin lymphoma</td>
<td>6 (5)</td>
<td>2 (3)</td>
</tr>
<tr>
<td>Hodgkin disease</td>
<td>7 (6)</td>
<td>4 (6)</td>
</tr>
<tr>
<td>Myeloma</td>
<td>3 (3)</td>
<td>1 (2)</td>
</tr>
<tr>
<td>Acute myeloid leukemia</td>
<td>33 (28)</td>
<td>19 (29)</td>
</tr>
<tr>
<td>Chronic myeloid leukemia</td>
<td>14 (12)</td>
<td>7 (11)</td>
</tr>
<tr>
<td>Myelodysplasia</td>
<td>12 (10)</td>
<td>6 (9)</td>
</tr>
<tr>
<td>Myeloproliferative syndrome (except CML)</td>
<td>3 (3)</td>
<td>2 (3)</td>
</tr>
<tr>
<td>Other</td>
<td>1 (1)</td>
<td>0 (0)</td>
</tr>
<tr>
<td><strong>CMV status</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D+/R+</td>
<td>30 (26)</td>
<td>...</td>
</tr>
<tr>
<td>D−/R+</td>
<td>36 (31)</td>
<td>...</td>
</tr>
<tr>
<td>D+/R−</td>
<td>14 (12)</td>
<td>...</td>
</tr>
<tr>
<td>D−/R−</td>
<td>36 (31)</td>
<td>...</td>
</tr>
</tbody>
</table>

*NOTE.* Data are no. (%) of subjects, unless otherwise specified. CML, chronic myeloid leukemia; CMV, cytomegalovirus; D+/R+, seropositive donor/seronegative recipient; D−/R+, seronegative donor/seropositive recipient; D+/R−, seropositive donor/seronegative recipient; D−/R−, seronegative donor/seronegative recipient.

**Monitoring for CMV infection and preemptive therapy.** Weekly monitoring for CMV infection started from engraftment and continued until day 100 after transplantation or longer. CMV infection was defined as pp65 antigenemia of ≥1/200,000 leukocytes [24] in 2 consecutive tests. CMV infection was treated with ganciclovir or foscarnet for 2–3 weeks or until a patient had 2 consecutive tests with negative results.

**Tetramers, monoclonal antibodies, and flow cytometry.** HLA-A*0201/NLVPMVATV– and HLA-B*0702/TPRVTTGGGAM–soluble molecules were biotinylated (BirA enzyme; Avidity) and tetramerized with either streptavidin-phycocerythrin (PE) or streptavidin–allophycocyanin (APC) (Molecular Probes), as
RPMI 1640 medium supplemented with 10% human serum, 50
Peripheral blood mononuclear cells were cultured 5 days in
Calibur flow cytometer (Becton Dickinson). Cytometry analysis was performed immediately using a FACS
CD27-FITC, CD28-APC, CD45RO-PE, CD45RA-FITC, and
The following antibodies were used for phenotypic analyses:

CD4 PE antibodies. Staining performed in healthy CMV-

dinin-chlorophyll– binding protein (PerCP), and CD8 antibody

One hundred microliters of whole blood was stained with a sat-

turating amount of tetramer, CD3 antibody conjugated to peri-

dinin–chlorophyll– binding protein (PerCP), and CD8 antibody

conjugated to fluorescein isothiocyanate (FITC). Absolute num-

The longitudinal evolution of these parameters was compared between the group of patients with no
reactivation, 42) or seropositive (day 41) or whether the donor
was a sibling (day 44) or unrelated (day 39). Sixteen patients had
prolonged reactivations (lasting >6 weeks). This status of long-
lasting viremia predominated in the group with seronegative
donors (P = .03) (figure 1A). Similarly, prolonged reactivations occurred more frequently in the unrelated donor group
(P = .044). Donor type was also analyzed in the context of mul-
tiple reactivations. Grafts from unrelated donors significantly
increased the risk of >1 episode of reactivation (P = .02) (fig-
ure 1B), whereas donor serological status had no observed effect.
The intensity of the first CMV reactivation, as indicated by the
maximal viral load, did not influence the occurrence of a subse-
quent reactivation.

The absolute numbers of CMV-specific T cells strongly corre-
lated with the total absolute counts of CD3+ CD8+ T cells
(R = .67; P < .0001), regardless of reactivation status. A cor-
relation between CMV-specific and CD3+ CD4+ T cell counts
(R = .41; P = .0001) was observed only in patients with reac-
tivation.

Relationships between T cell reconstitution and CMV
reactivation. Patients were divided into 2 groups, accord-
ing to CMV reactivation status. Absolute counts of CD4+ and
CD8+ T cell subsets were plotted each month (figure 1C and
1D, respectively). The longitudinal evolution of these param-
eters was compared between the group of patients with no
reactivation and the group with ≥1 reactivation. The results
showed that CD4+ T cell counts during the first 3 months
after transplantation provide a useful predictive marker, with
a threshold value of 100 × 106 cells/L delineating the risk of
reactivation. This was also the true for the CD8+ T cell subset,
but to a lesser extent.
CMV-specific immune responses were similarly detected in seropositive recipients with or without reactivation (59.5% and 41.7%, respectively). The onset of this response after transplantation was similar in both groups (days 42.5 and 52, respectively). However, recovery of tetramer-positive T cell counts was associated with the absence of reactivation in 77% of cases (P = .012). Twenty-nine of the 45 patients with reactivations had a CD8+ T cell response against CMV. A specific response was usually delayed after the onset of reactivation (mean interval, 22.4 days), but this interval was extremely variable between recipients and did not correlate with either serological status or donor type. Among the 21 patients in whom reactivation did not occur, 5 recipients from seronegative donor/seropositive recipient (D−/R+) pairs had a CMV-specific response. The lack of viral reactivation was confirmed in samples by PCR. The recipient-donor type of tetramer-positive cells was analyzed in 3 patients, and CMV-specific T cells were of recipient origin in all cases, despite a global donor-type chimerism (data not shown).

**Reconstitution of distinct CMV-specific T lymphocyte subsets in D−/R+ recipients.** The level of CMV-specific CD8+ T cells might not be the only marker of protective immune response, and the differentiation profile of these cells could provide an indirect insight into their function. We conducted longitudinal multicolor stainings for 19 recipients (8 seropositive donor/seropositive recipient [D+/R+] subjects and 11 D−/R+ subjects). Additional phenotypes of CMV-specific CD8+ T cell subsets were determined in 6 healthy CMV-seropositive
patients with multiple but limited reactivations (lasting <2 weeks), progressive enrichment of the differentiated memory CD45RA+/CCR7− and the late differentiation CD27−/CD28− subset in the CMV-specific CD8+ T cell pool was observed (figure 4C and 4D, respectively). Sixteen patients with prolonged reactivations were defined as noncontrollers. Most of these patients (12/16) received transplants from a seronegative donor. CMV-specific CD8+ T cells were detected in 12 patients at the time of R1, and the onset of the immune response was not delayed. This was not due to a weak expansion of these T cells, because similar levels of tetramer-positive T cells were found in single, multiple, and prolonged reactivations. Remarkably, CMV-specific CD8+ T cells remained CD45RA+/CD27+ (figure 4E), and the evolution to the CD27−/CD28− late phenotype was either absent or extremely delayed, compared with that during a controlled reactivation (figure 4F). CD4+ T cell counts for patients with prolonged reactivations were lower than those for patients with controlled reactivations, but the difference did not reach significance. Because of the limited availability of samples, interferon (IFN)–γ enzyme-linked immunospot assays with pp65-derived peptides were performed in 5 D+/R+ patients only. In 3 patients, the end of a prolonged CMV reactivation coincided with the development of CD45RA+CD27− CMV-specific T cells and the detection of IFN-γ-producing T cells (>1 cell/μL). This threshold was never reached in the other 2 patients with a single episode of reactivation and persistent CMV-specific CD45RO+/CD8+ T cells (data not shown). CD4+ T cell counts <100 × 10⁶ cells/L did not allow the analysis of CMV-specific CD4+ T cell reconstitution in our patients with reactivations.

**DISCUSSION**

Our main goal was to determine the extent to which the immune-response profile would predict protective immunity against CMV. We performed extensive longitudinal monitoring of a large cohort of recipients of allo-HSCT without T cell depletion. Our analyses focused on the seropositive recipients, who

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**Table 2. Risk factors for a second episode of reactivation.**

<table>
<thead>
<tr>
<th>Risk factor</th>
<th>P</th>
<th>OR (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tetramer-positive cells*</td>
<td>.017</td>
<td>6.401 (1.393–29.408)</td>
</tr>
<tr>
<td>Donor typeb</td>
<td>.036</td>
<td>6.698 (1.138–39.439)</td>
</tr>
<tr>
<td>Donor serological status for CMVc</td>
<td>.856</td>
<td>1.170 (0.214–6.380)</td>
</tr>
</tbody>
</table>

**NOTE.** Logistic regression analysis was performed to determine the odds of a second cytomegalovirus (CMV) reactivation in 45 patients with >1 reactivation. CI, confidence interval; OR, odds ratio.

* Tetramer-positive cells present vs. tetramer-positive cells absent during the first reactivation.

* Matched unrelated donors vs. sibling donors.

* Seropositive vs. seronegative donors.
Figure 3. Phenotypes of cytomegalovirus (CMV)–specific CD8\(^+\) T cells at the first episode of reactivation. A and B, Seropositive donors and seropositive recipients (D\(^-\)/R\(^+\)). C and D, Seronegative donors and sero-positive recipients (D\(^+\)/R\(^-\)). Dot plots are gated within the tetramer-positive CD3\(^+\)CD8\(^+\) fraction, and percentages of the different subsets are indicated at the top of each panel. Patterns for 3 representative patients are shown for each combination (for D\(^-\)/R\(^+\) recipients, patients 025, 030, and 124; for D\(^+\)/R\(^-\) recipients, patients 059, 053, and D01). Perforin and granzyme B expression was also studied in both tetramer-positive and tetramer-negative subsets, and no major differences were found between patients with different patterns of reactivation (data not shown). d, day.

are known to be more susceptible to CMV-related complications [26].

In accordance with previous data, we showed that CMV reactivation was a frequent event, occurring in >68% of seropositive recipients. Numerous studies have examined the role played by donor CMV serostatus in reactivation, CMV-related disease, or transplant outcome, with conflicting conclusions [15, 27–34]. Only one extensive study, which included multivariate analyses of >7000 seropositive patients, clearly demonstrated an adverse effect of seronegative donors on overall mortality in recipients of transplants from matched unrelated donors [35].

The preemptive therapy approach, which relies on strict virological monitoring [36], enabled almost complete prevention of CMV disease [26]. However, recurrent and prolonged reactivations and/or disease still developed in a subgroup of patients. Our results demonstrated that seropositive patients receiving transplants from seronegative or unrelated donors were at higher risk of prolonged reactivations. In addition, we showed that recipients from unrelated donors were more susceptible to multiple reactivations. These findings were consistent with those from a study with a more limited sample size, in which 4 of the 6 D\(^-\)/R\(^+\) recipients (3 unrelated and 1 sibling donor) required multiple courses of ganciclovir [15].

CMV-specific T cells were detected in 50% of recipients during the first 3 months after allo-HSCT, independent of donor serological status and origin. The precocity of this response in recipients with a graft from an unrelated and/or a seronegative donor was not observed by other investigators [1, 2, 33]. The difference between these studies and ours could be explained by the lack of T cell depletion in our study group. Recurrent CD4\(^+\) T cell counts <100 \(\times\) 10\(^6\) cells/L during the first 90 days were strongly associated with CMV reactivation, underscoring the adverse effect of impaired CD4\(^+\) T cell reconstitution on infectious morbidity [37] and late CMV disease [38]. Furthermore, the correlation between global CD4\(^+\) and CMV-specific CD8\(^+\) T cell counts, observed only in patients with reactivation, stressed the key role of the CD4\(^+\) T cell subset [3]. None of the seropositive patients experienced CMV-related disease, and we found that recovery to levels of >25 \(\times\) 10\(^6\) CMV-specific T cells/L was sufficient to prevent reactivation in most cases.

A puzzling observation was the occurrence of CMV-specific T cell responses in 5 D\(^-\)/R\(^+\) patients without viral reactivation. In 3 cases, analyses of global and virus-specific T cell chimerisms showed that all CMV-specific T cells were of recipient origin, whereas the global chimerism was of the donor type. This extends, into a setting without T cell depletion, findings already reported for cord blood transplantation [39] and T cell–depleted allo-HSCT [29].

Recurrent reactivations were diagnosed in 30 patients and correlated with the lack of CMV-specific T cells during R1. Using a different approach, one based on the simultaneous quantification of CMV-specific CD4\(^+\) and CD8\(^+\) T cells [40], Lilleri et al. suggested that repeated episodes of reactivation could occur in patients without prompt recovery of specific immunity on day 60 [6]. Moreover, multivariate analysis revealed that allo-HSCT with an unrelated donor and the absence of CMV-specific T cells during R1 were independent risk factors for recurrent reactivations.

Despite the development of CMV-specific T cells, prolonged reactivations were observed in 16 patients. As pointed out elsewhere, the cytotoxic activity of CMV tetramer–positive T cells could be impaired in the setting of immunodepression [5, 10] and could be related to changes in subset repartition of CMV-specific CD8\(^+\) T cells, as recently reported in HIV-infected individuals [41]. In the absence of T cell depletion, mature T cells are transferred and their population expands after transplantation. Accordingly, CMV-specific T cells detected in D\(^+\)/R\(^-\) recipients displayed phenotypic features similar to those observed in immunocompetent individuals [23, 42]. Because >85% of prolonged reactivations occurred in D\(^-\)/R\(^+\) recipients, we focused on this group. In such cases, the specific T cell responses observed during reactivation might mimic responses during pri-
mary infection, taking into account the specific restrictions conferred by the setting of allo-HSCT. When sufficient amounts of CMV-specific CD8\(^{+}\) T cells were detected, CD27\(^{-}\)CD28\(^{-}\) early/intermediate phenotypes predominated within the tetramer-positive fraction during the first wave of reactivation. This was rapidly followed by a progression through to the CD27\(^{-}\)CD28\(^{-}\) late phenotype. CD45RA and CD45RO expression did not correlate directly with the stages defined by CD27 and CD28, but enrichment of the CD45RA\(^{+}\)CD27\(^{-}\) subset could be associated with higher levels of IFN-\(\gamma\)-producing T cells. The development of CD45RA\(^{+}\)CD8\(^{+}\) T cells over a period of 70 days was observed only in patients with >=2 reactivations.

Because primary CMV infection is usually clinically silent, little is known about the longitudinal evolution of CMV-specific T cells in “healthy” individuals. In a cohort of 200 children, progressive expansion of the population of CMV-specific CD8\(^{+}\)CD45RA\(^{+}\)CD27\(^{-}\) T cells was observed during postprimary infection [43]. Analyses in infected individuals enrolled in clinical protocols enabled a more detailed longitudinal follow-up of CMV-specific responses, as observed in allo-HSCT [44] or kidney transplantation [45]. In the latter study, the number of CMV-specific CD27\(^{-}\) T cells was linearly related to the total number of CMV-specific T cells. In

Figure 4. Evolution of cytomegalovirus (CMV)–specific CD8\(^{+}\) T cell subsets according to the pattern of reactivation in recipients with seropositive donors. In a single episode of reactivation, CMV-specific CD8\(^{+}\) T cells remained CD45RO\(^{+}\) with evolution to the CD27\(^{-}\)CD28\(^{-}\) late phenotype (A and B). In multiple controlled reactivations, there was enrichment of CMV-specific CD45RA\(^{+}\) cells with less marked evolution to the late phenotype (C and D). In prolonged reactivations, CMV-specific CD8\(^{+}\) T cells remained CD45RO\(^{+}\) with early/intermediate phenotypes (E and F). Results are expressed as percentages within the tetramer (Tet)–positive CD3\(^{+}\)CD8\(^{+}\) subset. Data from 4 representative patients are shown in each graph.

Figure 5. Schematic representation of distinct profiles of cytomegalovirus (CMV)–specific CD8\(^{+}\) immune responses, according to diverse aspects of CMV reactivation. Uncontrolled reactivation was related to a defect of T cell differentiation more than to a quantitative defect of tetramer T cell subsets, because tetramer-positive T cell counts did not differ according to different patterns of reactivation. During the first 3 months after allogeneic hematopoietic stem cell transplantation (allo-HSCT), patients with multiple but controlled reactivations acquired late and differentiated phenotypes similar to the CMV-specific memory phenotypes found in healthy donors. In prolonged reactivations, CD4\(^{+}\) T cell counts were lower, and the process of differentiation was impaired. D\(^{-}\)/R\(^{-}\), seronegative donor/seropositive recipient.
such settings, the distribution of the differentiation subsets might reflect the profiles of preexisting memory T cells. The repeated phenotypic follow-up provided by our study clearly showed that, in D\(^{-}/\)R\(^{+}\) patients with uncontrolled reactivations, CMV-specific T cells of the late phenotype were unable to develop. The source of CMV-specific T cells was mainly T cells generated de novo, and the number of CMV-specific T cells of the late phenotype was not linearly related to the total number of CMV-specific CD8\(^{+}\) T cells.

These data confirmed the general consensus that most CMV-specific CD8\(^{+}\) T cells in infected individuals controlling CMV infection were IFN-\(\gamma\)–secreting CD27\(^{-}/\)CD28\(^{-}\) T cells [22]. In a quite different setting of immunodeficiency, higher numbers of early/intermediate CMV-specific T cells were associated with long-term recovery. These data in HIV-positive patients with marked CD4 T cell defects highlight the importance of CD4 in supporting the differentiation of CD8\(^{+}\) T cells from early to late effectors [41]. In allo-HSCT, the respective reconstitution of CD4\(^{+}\) and CD8\(^{+}\) T cell subsets is not synchronized, and the emergence of CD4\(^{+}\) T cells is delayed compared with CD8\(^{+}\) T cells [46]. In prolonged CMV reactivation in D\(^{-}/\)R\(^{+}\) recipients, differentiation of virus-specific CD8\(^{+}\) T cells may be impaired, probably because of the latency of the development of the CD4\(^{+}\) T cell subset. Figure 5 is a schematic representation summarizing the diverse aspects of CMV reactivation in allo-HSCT, according to distinct profiles of immune reconstitution.

Several studies have analyzed the relative contribution of immune responses against immediate early 1 protein [47] and pp65, with controversial conclusions [44, 48]. We are now investigating this question in the light of our present results. Our study reveals that immune monitoring in allo-HSCT provides a valuable extension of virologic monitoring, notably in the era of anti-CMV preemptive therapy. We suggest some guidelines for the management of CMV-related complications and recommend the determination of CD4\(^{+}\) T cell counts during the first 3 months after allo-HSCT as a predictive marker of reactivation, together with monitoring of tetramer-positive CD8\(^{+}\) T cells (risk of multiple reactivations) and the longitudinal evaluation of CD27 and CD45RA expression within the tetramer-positive subset (risk of prolonged reactivations).

References


